

ISBN-978-93-94174-76-4

Frontiers of Science and Technology:

Integrating Innovations for Sustainable Future

Volume-I



Editors

Dr. Anand Rajavat
Dr. Ranjana Goswami
Dr. Suhas Mane



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SHRI VAISHNAV VIDYAPPETH VISHWAVIDYALAYA, INDORE

**SHRI VAISHNAV VIDYAPEETH VISHWAVIDYALAYA,
INDORE, MADHYA PRADESH, (INDIA)**

Preamble

In the contemporary era of rapid technological and scientific advancement, the boundaries between disciplines are increasingly blurred, and knowledge no longer remains confined within traditional silos. Innovations in biotechnology, artificial intelligence, renewable energy, and advanced engineering are intricately linked to societal progress, environmental sustainability, and economic development. While these breakthroughs present unparalleled opportunities for improving quality of life, they also pose complex challenges requiring holistic, interdisciplinary approaches that combine scientific rigor with ethical responsibility. It is within this broader context that **SANMANTRANA–2026**, a multi-disciplinary international congress on “*Frontiers of Science and Technology: Integrating Innovations for a Sustainable Future*”, emerges as a significant academic forum.

Scheduled from 11th to 13th February 2026 and organized by Shri Vaishnav Vidyapeeth Vishwavidyalaya in collaboration with St. Cloud State University, USA, SANMANTRANA–2026 provides a global platform for scholars, researchers, educators, and industry professionals to exchange innovative ideas, present cutting-edge research, and collaboratively shape future solutions for a sustainable and resilient world. The congress emphasizes the integration of scientific discovery, technological innovation, and human-centric problem solving to address pressing global challenges such as climate change, urbanization, energy scarcity, environmental degradation, and food security.

The overarching theme of SANMANTRANA–2026 underscores the convergence of multiple disciplines in creating sustainable solutions. Computer Science and Applications explores data-driven intelligence, artificial intelligence, machine learning, and digital transformation for real-world optimization. Core Engineering disciplines contribute advancements in smart infrastructures, renewable energy systems, and industrial innovation. Forensic Science highlights emerging methodologies for digital and physical investigations, ensuring societal safety and justice. Mathematics and Applied Sciences underpin analytical frameworks, modeling, and quantitative reasoning essential for innovation across domains. Collectively, these streams reflect the congress’s commitment to fostering interdisciplinary research that bridges theory with practical implementation.

SANMANTRANA–2026 also emphasizes the social and ethical dimensions of technology. Human-centered design, responsible innovation, and inclusive development are recognized as critical to ensuring that scientific progress benefits all sections of society. The congress encourages research that integrates sustainable practices, green technologies, and data-informed policy frameworks, while promoting collaboration between academia, industry, and government institutions.

This Congress compiles high-quality research contributions from participants across the globe. The selected papers cover diverse topics such as smart and autonomous systems, precision agriculture, climate-resilient engineering, cybersecurity, blockchain applications, AI-driven analytics, environmental monitoring, and advanced material sciences. Each contribution reflects rigorous scientific methodology, innovative problem-solving, and practical applications aimed at addressing current and future societal challenges. The inclusion of these works in a peer-reviewed, ISBN-indexed book ensures both scholarly recognition and broader dissemination of knowledge.

In addition to research papers, SANMANTRANA–2026 incorporated pre-conference workshops, interactive sessions, and knowledge-sharing initiatives to enhance research skills and promote methodological rigor among delegates. Recognition through awards and certificates for outstanding contributions further motivates participants to pursue excellence in their respective fields.

By bringing together multidisciplinary expertise and fostering a spirit of collaboration, SANMANTRANA–2026 and this Congress aim to inspire innovation, nurture critical thinking, and guide the next generation of researchers, engineers, and scientists. The insights presented herein not only highlight current scientific and technological frontiers but also provide a roadmap for creating a sustainable, resilient, and socially responsible future.

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He obtained his Ph.D. in the faculty of Computer Engineering in the field of Software Engineering and M.E. in Computer Engineering with specialization in Software Engineering from Devi Ahilya Vishwavidyalaya (DAVV), Indore. Currently, he has also been working as a Visiting Faculty at St. Cloud State University (SCSU), Minnesota, USA.

Besides undertaking consultancy assignments, he has authored/co-authored more than 120 publications. He has been on the Panel of Reviewers of several National and International Journals. He has supervised thirty-two (32) M.E./M.Tech dissertations. He has attended more than 102 certification courses, training programs, workshops, and faculty development programs conducted by leading multinational and national IT companies.

Dr. Anand Rajavat was awarded the Excellence in Academics Award in 2021 and received the Dronacharya Award by IBM in 2008 and 2009 for development and deployment of best research projects. He has been associated with various professional bodies such as the Computer Society of India, Indian Society of Internet, ACM, and IEEE.

He has been the Chairman of Board of Studies for Computer Science & Engineering, Information Technology, and Computer Applications at Shri Vaishnav Vidyapeeth Vishwavidyalaya (Indore). He has also been a member of the Governing Body of Shri Vaishnav Institute of Technology & Science, Indore.

He has been a member of the Board of Management, Academic Council, and Faculty of Studies at Shri Vaishnav Vidyapeeth Vishwavidyalaya (Indore). He has coordinated several projects related to IT Infrastructure, Networking, Software Licensing, and ERP system deployment for different institutes of Shri Vaishnav Group, Indore.

He has provided consultancy services to many IT companies and educational institutions including Galaxy Weblinks Pvt. Ltd., Indore, Mathe Magics Pvt. Ltd., Dewas, HSofttech Pvt. Ltd., Indore, ITI Mandsaur, ITI Manpur, and ITI Indore (Women's) for skill development of ITI faculty, staff, and students, and for setting up computer laboratories, networking, and other IT infrastructure.

He was invited by St. Cloud State University (SCSU), Minnesota, USA, as a resource person for three weeks to deliver sessions on the latest trends in Information Technology in 2019.

Prof. (Dr) Ranjana Goswami is an accomplished Academician and Researcher currently serving as Professor of Chemistry, Shri Vaishnav Vidyapeeth Vishwavidyalaya Indore formerly worked as Associate Professor of Chemistry at ITM University, Gwalior. with over 17 years of rich teaching and academic administrative experience. She did M.Sc. in Chemistry and PhD from Jiwaji University, Gwalior.

A rich combination of pedagogical expertise, research excellence in green chemistry and nanomaterials, and demonstrated leadership in academic administration.

Authored more than 30 research publications in reputed peer-reviewed journals. Successfully completed MPCST-funded research project an ITM University-funded project on Development of environment friendly Tin Chalcogenides for waste water treatment.

Recipient of multiple accolades including Best Teacher Award (2021), Best Project Award (2022), and Active SPOC Award for NPTEL–SWAYAM (2022, 2023, 2024) at ITM University, Gwalior.

Has successfully guided 10 postgraduate dissertations and is currently guiding 5 Ph.D. research scholars. Played a pivotal role in organizing several signature academic events, notably the National Conference on Recent Advances in Chemical and Environmental Sciences (RACE) continuously from 2021 to 2025, and the International Conference TIMS–2023, themed “*Chemistry for a Healthier India*”. These events witnessed participation from eminent experts, academicians, faculty members, and research scholars from across India, fostering interdisciplinary dialogue and research dissemination. An active member of leading professional bodies such as the American Chemical Society (ACS) and the International Association of Advanced Materials (IAAM).

Has been successfully serving as UGC SWAYAM Nodal Officer since 2021 and NAAC Criterion Coordinator since 2022, contributing significantly to digital learning initiatives, policy implementation, accreditation processes, and institutional quality enhancement. Completed SWAYAM Course Development Program with A++ Grade and several prestigious international online courses, including Nanotechnology: A Maker’s Course (Duke University), Molecular Spectroscopy (University of Manchester), and Introduction to Chemistry: Reactions and Ratios (Duke University).

Dr Suhas Mane is a dedicated teacher with 24 years of teaching experience and currently working as Assistant Professor of Agricultural Extension and Communication at Shri Vaishnav Institute of Agriculture, Shri Vaishnav Vidyapeeth Vishwavidyalaya, Indore. He has completed Graduation in 1999 from Mahatma Phule Krishi University, Rahuri, Maharashtra, M.Sc. (Agri.) in Extension Education in 2001 from VNMKV, Parbhani, and Ph.D. in 2019 from YCMOU, Nasik. He has also qualified NET examination in 2012 conducted by ASRB, ICAR, New Delhi. He started his teaching career in July 2002 and actively engaged in teaching UG, PG and PhD levels for the various subjects of Agricultural Extension Education. He has organized various extension programs like farmers rallies, exhibitions, demonstrations, field visits, study tours women trainings, etc. for transfer of agricultural technologies to the farmers during his services in various private agricultural colleges in Maharashtra and Madhya Pradesh. He has published 12 research papers, 16 technical papers, 03 Books, 07 Book chapters, and 26 popular articles and also delivered 10 radio-talks, 09 guest lectures. Along with several trainings, FDPs, He has also attended 21 days training program on Agripreneurship organized by Anand Agricultural University, Anand Gujrat. He is recipient of National level Best Agro care Teacher Award 2021 by Agro care Group, Nasik. In addition to academic activities, he has established various units like Personality Development and Career Guidance Cell, Farmers Training Center, Farmers Scientist Forum, edited college magazines, progress reports, news of the events etc. He has also shouldered different responsibilities like Dean- Industry Institute Cell, Dean- International Relations and contributed in signing MoUs. In Social field he is active as Vice President of Maharashtra Samaj Hit Rakshan Sanstha, Indore, active blood doner since 2008, and organized various social events like Ganesh Utsav, etc.

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1. The Future of Internet of Things

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Abstract: The Internet of Things (IoT) has transformed from a simple idea of connecting a few devices into a significant technology that merges the physical and digital worlds. What started with small experiments in the 1980s gained a clear definition when Kevin Ashton coined the term “Internet of Things” in 1999. Since then, IoT has rapidly evolved due to better sensors, faster wireless networks, and the growth of cloud computing. Today, IoT plays an important role in areas like smart homes, healthcare, industrial automation, and smart cities, where it enables real-time monitoring, automation, and data-driven decisions. In this research paper, we will examine how IoT has developed over the years and how future advancements will keep changing the way devices interact and operate. We will focus on emerging technologies such as Artificial Intelligence, 5G and 6G networks, and edge computing, and discuss how these innovations will make IoT systems smarter, faster, and more independent. The paper will also look at the potential effects of IoT on future smart cities, connected industries, and sustainable environments while addressing key challenges like security risks, privacy concerns, and device compatibility. Through this study, we aim to provide a clear understanding of how IoT will influence a more intelligent and interconnected future.

Keywords: Internet of Things, IoT, Kevin Ashton, 5G, AI, Edge Computing, Smart Cities, Future Technology, Automation, Connectivity

Introduction

The Internet of Things (IoT) refers to a network of interconnected devices that communicate and exchange data without human intervention. From smart homes to industrial automation, IoT has become a cornerstone of digital transformation. To appreciate its future trajectory, it is essential to examine its history, development, and evolution, and then consider how IoT interacts with emerging technologies such as 5G, 6G, AI, and edge computing, while reshaping smart cities, automation, and global connectivity.

Historical Foundations

Early Networking (1960s–1980s): The origins of IoT lie in early computing and networking experiments, including ARPANET and NASA’s Apollo systems.

RFID and the Birth of IoT (1990s): Radio-Frequency Identification (RFID) technology enabled tracking of goods and assets. In 1999, Kevin Ashton introduced the term “Internet of Things.”

Web Expansion: The invention of the World Wide Web in 1989 provided the infrastructure for global connectivity, laying the groundwork for IoT applications.[4]

Development of IoT

IoT 1.0 – Industrial Beginnings: Early IoT applications focused on logistics and supply chain management.

IoT 2.0 – Consumer Adoption: With the rise of smartphones, cloud computing, and wireless networks in the 2010s, IoT entered consumer markets through smart homes, wearables, and connected vehicles.

IoT 3.0 – Intelligent Integration: In the 2020s, IoT has merged with AI, edge computing, and 5G networks, enabling real-time analytics, autonomous systems, and large-scale industrial IoT.

Evolution of IoT

From Concept to Reality: IoT has evolved from a futuristic idea into a practical reality, powering healthcare monitoring, agriculture, and smart cities.

Integration with Emerging Technologies: AI, blockchain, and machine learning have enhanced IoT systems, making them predictive, adaptive, and secure.

Challenges in Evolution: Security vulnerabilities, interoperability issues, and data privacy concerns remain critical hurdles in IoT’s growth.[5]

IoT and Emerging Technologies

IoT and 5G/6G Connectivity

The arrival of 5G has significantly accelerated the growth of IoT. With high data speeds, low latency, and the ability to connect millions of devices simultaneously, 5G enables real-time communication on a large scale. This has transformed smart cities and homes, where traffic systems, energy usage, and waste management are optimized for efficiency and sustainability.

In industry and agriculture, 5G-powered IoT supports real-time monitoring of machines and crops, improving productivity through Industry 4.0 and precision farming. The healthcare sector has also benefited, with reliable remote patient monitoring and telemedicine services bringing quality care closer to people’s homes. These advancements are further supported by edge computing, which processes data closer to its source, reducing delays and saving energy.

Looking ahead, 6G is expected to move beyond connectivity toward intelligence and autonomy. Future networks may support fully autonomous transportation systems and immersive technologies like Augmented Reality (AR) and Virtual Reality (VR), enhancing communication, training, and human–machine interaction.

However, challenges remain. Applications such as remote surgery require Ultra-Reliable Low-Latency Communication (URLLC), while the massive growth of connected devices raises serious concerns about security, privacy, and environmental sustainability.[2]

IoT and Artificial Intelligence

Artificial Intelligence (AI) and the Internet of Things (IoT) are among the most influential technologies shaping the modern digital world. AI enables machines to learn from data and make intelligent decisions, while IoT connects devices and systems to collect and share that data. When combined, AI and IoT create smart systems capable of improving efficiency, safety, and decision-making across many industries.

One of the most important applications of AI in IoT is predictive maintenance. IoT sensors continuously collect data from machines, and AI algorithms analyse this data to predict failures before they happen. This helps industries reduce downtime, lower maintenance costs, and improve productivity, especially in manufacturing and transportation.

Another major area is smart homes. AI-powered IoT devices such as smart thermostats, lighting systems, and security cameras can learn user behaviour and automatically adjust settings. For example, a smart thermostat can adapt to daily routines to save energy, while AI-enabled security systems can detect unusual activity and alert homeowners.

The healthcare sector has also seen significant benefits. Wearable devices and medical sensors collect patient data, which AI analyses to monitor health conditions and detect early signs of illness. This enables remote patient monitoring, early diagnosis, and more personalised treatment.

Despite these benefits, integrating AI with IoT comes with challenges. Managing the massive volume of data generated by IoT devices is complex. Security and privacy are critical concerns, as connected devices are vulnerable to cyber threats. Interoperability issues between devices from different manufacturers, high power consumption, and implementation costs also pose difficulties.

Looking ahead, the future of AI in IoT is highly promising. AI will play a key role in improving energy efficiency, enhancing industrial safety, and enabling smarter cities and autonomous systems. As AI becomes more advanced and IoT devices become more widespread, their integration will continue to drive innovation and transform the way we live and work.

IoT and Edge Computing

Edge computing plays a crucial role in improving the performance of Internet of Things (IoT) systems by processing data closer to where it is generated. Instead of sending all data to a distant data centre, edge computing reduces processing time, improves efficiency, and enables faster decision-making.

In an IoT edge computing system, data is collected, processed, and stored locally without always relying on the cloud. This approach distributes the workload across multiple devices, ensuring that no single device becomes overloaded.

The process begins with data collection, where sensors on IoT devices gather information such as room temperature in a smart thermostat or wind speed on a turbine. Next, local processing takes place through an edge device, such as a gateway or nearby server, which

analyses the data using local computing resources. The data is then filtered, allowing only useful information to be processed while unnecessary data is discarded.

One of the most important advantages of edge computing is automation. IoT devices can make real-time decisions based on analysed data, such as shutting down an overheating machine or adjusting the movement of an autonomous vehicle to prevent accidents. Finally, selected data that requires long-term storage or deeper analysis is sent to the cloud.[3]

IoT in Automation

IoT automation refers to the use of connected devices and smart systems to automate industrial processes with minimal human intervention. By integrating sensors, machines, and software, IoT enables real-time data collection, monitoring, and control, leading to more efficient and intelligent operations.

One of the most important applications of IoT automation is smart manufacturing, where sensors, cameras, and robotics are used to optimise production processes, improve product quality, and reduce energy consumption. Another key application is predictive maintenance, where IoT sensors monitor machine performance and, with the help of data analytics, predict failures before they occur. This reduces unexpected downtime and lowers maintenance costs. Remote monitoring and control is also widely used, allowing industries to supervise equipment and processes from a distance, improving safety and operational efficiency.

The benefits of IoT automation include increased productivity, as repetitive tasks are automated and processes become faster and more accurate. It also helps in cost reduction by minimising energy waste, reducing maintenance expenses, and optimising resource usage. Additionally, IoT automation enhances workplace safety by continuously monitoring conditions and detecting potential hazards in real time.

Overall, IoT automation is transforming industries by making operations smarter, safer, and more efficient, playing a vital role in modern industrial development.[1]

IoT and Global Connectivity

The Internet of Things (IoT) is all about everyday devices being connected to the internet and communicating with each other. These devices—such as sensors, machines, vehicles, or medical equipment—can collect data about their surroundings and share it online in real time. At the centre of IoT is global connectivity. This means not only connecting people across the world, but also connecting billions of devices that form the backbone of modern society. Thanks to IoT, once-simple devices are now “smart” and can send information, receive instructions, and work together automatically.

Global connectivity is important because it helps improve the flow of information, goods, and services across borders. Businesses can monitor assets remotely, governments can improve infrastructure, and industries like healthcare, transport, and energy can operate more efficiently—even across long distances.

IoT makes global connectivity practical by enabling real-time monitoring and control, no matter where people or assets are located. This leads to better decision-making, reduced losses, improved safety, and faster growth in a highly digital world.

In short, IoT turns connectivity into intelligence, helping the world stay connected, efficient, and responsive.

Future Prospects

Smart Cities: IoT will enable intelligent traffic systems, energy-efficient grids, and sustainable urban planning.

Healthcare Transformation: Remote monitoring, IoT-enabled diagnostics, and personalized medicine will redefine healthcare delivery.

Industrial IoT: Factories will adopt predictive maintenance, robotics, and digital twins to optimize production.

6G and Beyond: Ultra-low latency and massive connectivity will unlock autonomous transportation, immersive virtual environments, and next-generation IoT ecosystems.

Ethical Considerations: Addressing cybersecurity threats, ensuring privacy, and establishing ethical frameworks will be essential for IoT's responsible future.[6]

Conclusion

The Internet of Things has progressed from conceptual origins in the late 20th century to a transformative force in the 21st century. Its synergy with 5G/6G, AI, edge computing, smart cities, automation, and global connectivity demonstrates its central role in shaping the digital future. Looking ahead, IoT promises to revolutionize industries and daily life, provided that challenges of security, privacy, and ethics are effectively addressed.

References

- [1] <https://deviceauthority.com/iot-automation-top-applications-and-key-benefits/#:~:text=Skip%20to%20content,Accept%20All>
- [2] <https://www.telecomgurukul.com/post/internet-of-things-iot-applications-in-5g-and-6g-transforming-connectivity-and-automation>
- [3] <https://www.ibm.com/think/topics/iot-edge-computing>
- [4] CompTIA. *History of IoT. Future of Tech*
- [5] NASSCOM. *Evolution of IoT: From Concept to Everyday Reality*. [NASSCOM Community](#)
- [6] Karpagam Institute of Technology. *IoT Evolution and Future Impact*. [KIT](#)

2. AI to Data Privacy, Threat or Boon

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Abstract- Data Privacy is a significant concern with the automated collection, storage and manipulation of data using Artificial Intelligence (AI). AI will have an effect on the manner in which people interact with and utilise data in various fields, e.g. Education, Financial Services, Healthcare, Retail, & other Digital Services that rely upon Internet Technology for access to customers and business partners. In particular to the Financial Services Sector, the ability of AI to rapidly process and analyse vast quantities of data, with a high degree of accuracy, leads to improved operational efficiencies and decision-making processes. As a result of these increased efficiencies from AI-based applications, there is a question concerning the potential benefits/threats posed by AI to the privacy of citizens with respect to the data collected, processed, and used by AI-based services. A primary privacy concern is that the type of data collected by AI applications often contains sensitive information about individuals. Many of the data collected from the customers of many companies goes beyond what the customer was originally aware of when he or she provided consent to have their personal data collected and is often done so without having adequately provided consent to have their data collected. Currently, the uncertainty surrounding the use of an individual's personal data by AI Applications has left many consumers unsure of how their individual data will be used by an AI Application. Another aspect of AI that is beneficial to the area of data privacy is the ability of AI to detect, prevent, and monitor Cyber Security incidents and to protect and monitor personal data. In conjunction with traditional security measures, AI has the potential to greatly enhance the protection and compliance with regulatory standards affecting the protection of sensitive personal data. However, despite these enhancements in the area of data privacy, there continue to be concerns about data privacy, including the ability to trace anonymous data back to specific identifiable individuals; the potential for bias in the recommendations or decisions produced by AI algorithms; data deleted or otherwise destroyed after training an AI System. The speed and level of technological advancement by AI will likely continue to raise privacy-related concerns.

Keywords: Data Privacy, AI algorithms, AI technologies, Cyber Security

Introduction

The generation and use of data have increased at a rate never seen before due to the digital transformation of modern society. By enabling machines to learn, predict, and automate decision-making processes, artificial intelligence (AI) plays a critical role in deriving significant insights from this data. These days, a number of industries, including education, healthcare, finance, retail, and governance, use AI technologies extensively. In order to

provide individualized services, boost productivity, and improve client experiences, these industries mainly rely on user data.



image source: https://r.search.yahoo.com/_ylt=AwrKBHaVclVpC3kVgavGHAX.;_ylu=c2VjA2ZwLWF0dHJpYgRzbGsDenVybA--/RV=2/RE=1767236373/RO=11/RU=https%3a%2f%2fwww.traverselegal.com%2fblog%2fai-data-privacy-and-security%2f/RK=2/RS=jihHY77Z6aBxGalZ9kPStAPggZM-

Notwithstanding these benefits, the widespread use of personal data has sparked grave worries about data privacy. People frequently divulge personal information online without fully comprehending how it will be gathered, stored, examined, or utilized. Multiple datasets can be processed and combined by AI systems, sometimes exposing private information that users have never given their express consent to share. Consequently, privacy risks have grown more intricate and challenging to handle.

This challenge is exemplified by the financial services industry. AI is being used more and more by banks, insurance companies, and fintech companies to identify fraud, evaluate creditworthiness, and offer individualized financial products. Although these applications increase efficiency and accuracy, they also handle extremely sensitive financial and personal data. This makes it imperative to investigate whether AI poses a risk to or benefits from data privacy.

This study intends to examine how AI affects data privacy, pinpoint the advantages and risks of its application, and investigate how AI can be used sensibly while protecting people's right to privacy.

Review of Literature

Numerous scholars have investigated the connection between data privacy and artificial intelligence. Research shows that large datasets are necessary for AI-driven systems to operate efficiently, which frequently results in intensive data collection procedures.

According to Solove [1] many organizations gather more data than is necessary, which raises the possibility of abuse or illegal access, according to current research[2].

Prior research highlights that informed consent is still a significant problem in AI-based systems. Long privacy policies are often accepted by users without their full understanding, leading to inadvertent consent[3]. Additionally, because sophisticated algorithms can re-identify people from anonymized datasets, researchers note that anonymization techniques used to protect privacy are not always dependable[4].

Positively, research indicates that AI can greatly enhance cybersecurity. Compared to conventional systems, machine learning models can identify cyberthreats, spot unusual activity, and react to security incidents more quickly[5]. AI-based monitoring tools lower the risk of breaches and assist organizations in adhering to data protection laws.

Other research focuses on ethical issues, such as discrimination and algorithmic bias. Bias can result in unfair outcomes in areas like loan approvals or hiring decisions when training data reflects current social inequalities[6]. Researchers contend that the design of AI systems should incorporate accountability, transparency, and fairness.

All things considered, the literature currently in publication offers a fair assessment, acknowledging AI as both a potent instrument for data security and a source of fresh privacy issues[7].



Image source: https://r.search.yahoo.com/_ylt=AwrKBHaldFVp6OEUrpfGHAX.;_ylu=c2VjA2ZwLWF0dHJpYgRzbGsDcnVybA--/RV=2/RE=1767236872/RO=11/RU=https%3a%2f%2fexplore.movista.com%2fblog%2fwhat-to-know-about-the-new-gdpr-regulation/RK=2/RS=WxRJzqoqvR8V1JdwsCkIt1vQ4HU-

Research Methodology

Based on secondary data, this study employs a descriptive and analytical methodology. Information about artificial intelligence, data privacy, cybersecurity, and digital ethics has been gathered from scholarly journals, research papers, reports, articles, and online resources. With an emphasis on financial services, the study uses a qualitative methodology to examine how AI affects data privacy in a variety of industries. The advantages and risks of AI technologies are assessed through comparative analysis. There hasn't been any primary data collection done through surveys or interviews.

In order to comprehend how businesses try to handle data protection issues related to AI-based applications, the methodology also entails examining current privacy practices and regulatory frameworks.

Role of AI in Data-Driven Sectors

AI is crucial to many data-driven industries because it makes intelligent decision-making, automation, and predictive analytics possible. Large amounts of user data are crucial to these industries, which raises concerns about privacy and efficiency.



Image source: https://r.search.yahoo.com/_ylt=AwrKBHaGcVVp9O8Uv1TGHAX.;_ylu=c2VjA2ZwLWF0dHJpYgRzbGsDcnVybA--/RV=2/RE=1767236102/RO=11/RU=https%3a%2f%2fbecominghuman.ai%2fhow-different-sectors-are-using-ai-26470ba334ab/RK=2/RS=A5RZAGV0p8apv0jzu537blSYKYM-

AI in Financial Services

AI is extensively utilized in the financial industry for risk management, investment analysis, chatbots for customer service, fraud detection, and credit scoring. Large amounts of transaction data can be processed in real time by AI systems, which helps organizations spot questionable activity and effectively stop fraud. Additionally, automation increases operational speed and decreases human error.

Financial data, however, is very sensitive, and its improper use can have dire repercussions. AI systems may occasionally gather more personal identifiers, transaction histories, and behavioral data than consumers anticipate. Concerns regarding consent, profiling, surveillance, and possible misuse of private financial data are raised by this.

AI in Healthcare

AI applications in healthcare support patient monitoring, treatment planning, medical imaging, and disease diagnosis. These technologies facilitate early disease detection, increase accuracy, and lessen the workload for medical professionals. However, sensitive medical records, including genetic information, biometric data, and medical history, are crucial to healthcare AI systems.

Patient confidentiality and trust may be violated by unauthorized access to or misuse of such data. Strong privacy protections are crucial in AI-based healthcare systems because data breaches can have major ethical and legal repercussions.

AI in Education

AI is utilized in the education sector for automated tests, virtual tutors, performance analytics, and personalized learning platforms. AI systems can customize instructional materials to meet the needs of each student by examining their behavior, academic performance, and rate of learning.

Large volumes of student data, including personal information, academic records, and online activity, are simultaneously gathered by these systems. Misuse of student information, privacy violations, and surveillance issues could arise from improper handling of such data.

AI in Retail

AI is used by retail companies for customer behavior analysis, inventory management, demand forecasting, and recommendation systems. In order to provide individualized experiences, AI keeps track of customer preferences, purchase trends, and browsing history. Although this increases customer satisfaction and business efficiency, it also raises issues with excessive data collection, tracking without consent, and consumer profiling. Sensitive financial and personal data may be exposed by data breaches in retail platforms.

AI in Telecommunications

The telecom industry uses a lot of data and is becoming more and more reliant on AI for fraud detection, network optimization, customer management, and predictive maintenance. Call logs, location data, internet usage trends, and customer identity information are just a few of the vast amounts of data that telecom companies gather.

AI aids telecom companies in network traffic analysis, service quality enhancement, outage prediction, and real-time fraud detection. Additionally, it facilitates individualized service offerings and effective customer service via chatbots powered by artificial intelligence.

However, there are significant privacy issues with the use of AI in telecommunications. Behavioral data, communication metadata, and location tracking can all disclose extremely private information about people. User privacy may be violated by unauthorized access, surveillance risks, or improper data sharing. In order to guarantee responsible AI deployment in the telecom industry, robust data governance, encryption, and regulatory compliance are crucial.

AI-Related Privacy Issues

Uninformed Consent

The extent to which their data is gathered and processed is often unknown to users. Unintentional data sharing results from complicated and ambiguous consent procedures.



image source: https://r.search.yahoo.com/_ylt=AwrX_o1hc1Vpcd8WBhnGHAX.;_ylu=c2VjA2ZwLWF0dHJpYgRzbGsDcnVybA--/RV=2/RE=1767236577/RO=11/RU=https%3a%2f%2fhubtgi.com%2fhow-ai-is-revolutionizing-cybersecurity%2f/RK=2/RS=4XnYAx0XUM_qXIsyj8qGacV_7Ig-

Gathering Private Information

Sensitive financial, behavioral, and personal data are often handled by AI systems. If this data is misused or disclosed, it may result in financial loss, discrimination, or identity theft.

Anonymous Data Re-identification

Advanced AI techniques can re-identify people by combining multiple datasets, undermining privacy protection measures even when data is anonymized.

Discrimination and Algorithmic Bias

Unfair or discriminatory results could be produced by AI models trained on biased datasets. This is particularly problematic for applications pertaining to employment and financial decision-making.

Problems with Data Retention and Deletion

It is challenging to fully remove data from models once AI systems have been trained. This raises questions about long-term data storage and compliance with requests for data deletion.

AI as a Tool for Enhancing Data Privacy

AI can improve data security and privacy despite these worries. Cybersecurity tools with AI capabilities can identify anomalous patterns, anticipate threats, and react to attacks instantly. Organizations can monitor sensitive data access and stop breaches with the aid of these systems.

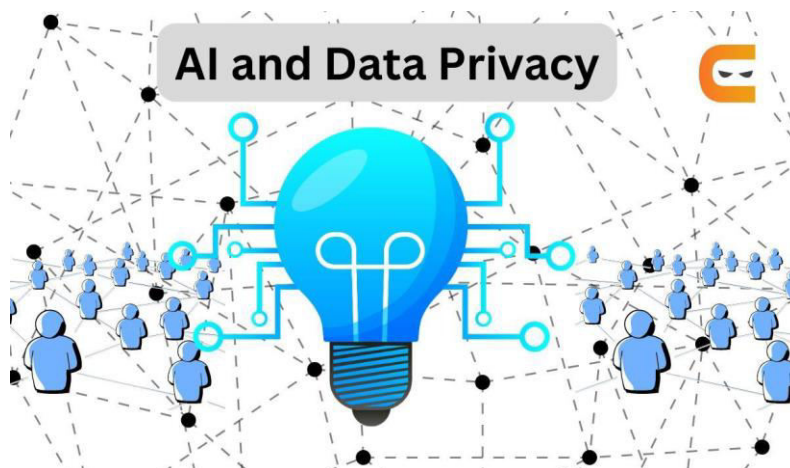


Image source: https://r.search.yahoo.com/_ylt=AwrX_o0DdFVp4ToWKhTGHAX.;_ylu=c2VjA2ZwLWF0dHJpYgRzbGsDcnVybA--/RV=2/RE=1767236739/RO=11/RU=https%3a%2f%2fwww.naukri.com%2fcode360%2flibrary%2fai-and-data-privacy/RK=2/RS=z.dr8XzHjksvTlaTr66vfn0s1w-

By automating audits, keeping an eye out for policy infractions, and guaranteeing adherence to data protection regulations, AI can also help with regulatory compliance. By restricting direct access to raw data, privacy-enhancing technologies like federated learning, encryption, and differential privacy further lower risks.

AI improves overall data protection frameworks and aids businesses in effectively responding to changing cyberthreats when paired with conventional security measures.

Challenges and Future Concerns

AI's quick development is creating new privacy issues. Regulations frequently find it difficult to keep up with advances in technology. Accountability is challenging because AI decision-making processes are opaque. Cross-border data flows also make it more difficult to enforce privacy laws.

Future issues include over-reliance on automated decision-making systems, misuse of biometric data, and increased surveillance. Stronger laws, moral AI design, and cooperation between organizations, technologists, and legislators are all necessary to address these problems.

Conclusion

When it comes to data privacy, artificial intelligence has two roles. On the one hand, it improves security, accuracy, and efficiency in a number of industries, especially financial services. However, there are serious privacy risks associated with data collection, consent, bias, and long-term data use. Although AI has the potential to improve cybersecurity and safeguard private data, its improper use or uncontrolled application may jeopardize people's right to privacy. As a result, a balanced strategy that fosters innovation while guaranteeing accountability, transparency, and robust data protection measures is crucial. AI can be turned from a possible danger into a potent instrument for improving data privacy with the right laws, moral standards, and technological protections.

References

- [1] Solove, D. J. (2021). *Understanding Privacy*. Harvard University Press.
- [2] OECD. (2020). *Artificial Intelligence and Data Privacy*.
- [3] European Union. *General Data Protection Regulation (GDPR)*.
- [4] Narayanan, A., & Shmatikov, V. (2008). Robust de-anonymization of large sparse datasets. *IEEE Symposium on Security and Privacy*.
- [5] World Economic Forum. (2022). *AI, Data Protection and Privacy*.
- [6] IEEE. (2019). *Ethically Aligned Design: A Vision for Prioritizing Human Well-being with Artificial Intelligence*.
- [7] Cavoukian, A. (2011). *Privacy by Design: The 7 Foundational Principles*.

3. Detection of Real and Fake Images Using Mobile NetV2 Feature Extraction and Logistic Regression

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Abstract - Differentiating between real and fake images, particularly through digital media like social media outlets, has become increasingly important due to the rapid rise of digital media. This study proposes a simple framework for fake image detection, utilising MobileNetV2 for feature extraction and Logistic Regression classifiers. Deep visual features can be extracted from photos using MobileNetV2, a pre-trained convolutional neural network (CNN) that doesn't require big datasets or intensive training. The features were fed into a Logistic Regression classifier to identify the real from the fake images. The procedure was tested with a small set of 10 images consisting of 5 real images and 5 fake images, achieving 100% accuracy with the same data used for training models (real and fake) and showcasing the model's ability to correctly predict unseen images with moderate confidence. Overall, the results indicate that, particularly in situations with limited data, feature extraction using a pre-trained CNN in conjunction with a basic classifier may carry out image authenticity detection. For real-time or data-poor contexts, the model provides a useful and computationally efficient way to identify phony photographs.

Keywords: Fake image detection, MobileNetV2, Convolutional Neural Networks (CNN), Logistic Regression, Feature extraction, Digital media, Image authenticity, Real vs. fake images, Small dataset, Computational efficiency

Introduction

The spread of photographs on the internet has greatly grown due to the quick development of digital media technologies and online content-sharing platforms. Although communication and information sharing have improved as a result, there are now significant worries about the integrity and authenticity of digital photographs.[1] It is now simpler to produce altered or completely fabricated photographs that closely resemble real ones because to developments in image editing tools and AI-based generating processes. Serious risks, such as identity fraud, the spread of false information, social engineering attacks, and digital forgery, are presented by the improper usage of such photos.[2] [3] Conventional methods for detecting false photos that rely on manually created features sometimes fall short when applied to intricate real-world images. Convolutional Neural Networks (CNNs), in particular, are deep learning models that have demonstrated superior performance in image analysis and image forensics tasks and enable autonomous feature learning.[4]

Large labelled datasets and significant computational resources are needed to train deep neural networks from scratch, but these are frequently unavailable in environments with

limited resources. By utilising models that have already been trained on huge datasets like ImageNet, transfer learning overcomes this constraint and permits efficient feature extraction even with a small amount of task-specific data.[5] Using depthwise separable convolutions and inverted residual blocks, MobileNetV2 is a lightweight convolutional neural network created for situations with limited resources. It effectively extracts high-level semantic information as a feature extractor without the need for expensive end-to-end training.[6]

In this work, a lightweight method for detecting fraudulent images is presented that combines Logistic Regression for binary classification with MobileNetV2-based deep feature extraction [7], [8]. Because it is easy to understand and works well with high-dimensional feature vectors, logistic regression is the method of choice [9]. Strong classification performance is shown in experiments on a tiny dataset of actual and false photos, demonstrating the usefulness of pre-trained feature extractors with basic classifiers in situations with limited data [10]. The suggested approach is simple to use, computationally effective, and offers a scalable basis for future expansions to bigger datasets and practical uses like digital forensics and media verification.

Objectives

1. For lightweight feature extraction, MobileNetV2 is used.
2. Uses logistic regression to classify data effectively.
3. Applies Logistic Regression for efficient classification.
4. Aims to detect real vs. fake images effectively.
5. Evaluates performance with limited data and computational resources.
6. Focuses on combining pre-trained features with a simple classifier.

Rationale

The rise of AI-generated and manipulated images poses a threat to digital authenticity. This work proposes a lightweight approach utilising MobileNetV2 features and Logistic Regression to efficiently detect fake images. The method reduces training complexity while maintaining strong performance, making it suitable for small datasets, limited resources, and rapid deployment.

Proposed Framework

The framework consists of four main stages: image input, preprocessing, deep feature extraction using MobileNetV2, and classification using Logistic Regression.

1 Acquisition of Input photographs: Both authentic and fraudulent photographs are gathered and arranged into distinct folders. Every picture serves as a system input.

2 Image Preprocessing: To guarantee consistency, every image is scaled to a predetermined size of 128×128 pixels. After that, the photos are normalised to match the ImageNet training distribution using MobileNetV2's `preprocess_input` function.

3 MobileNetV2-Based Feature Extraction: A deep feature extractor is a pre-trained MobileNetV2 model that has had its top layers removed. For every image, a compact 1280-dimensional feature vector is obtained using Global Average Pooling.

4 Classification using Logistic Regression: A Logistic Regression classifier receives the retrieved deep features and uses binary classification to differentiate between authentic and fraudulent photos.

5 Output Prediction: The predicted class label (Real/Fake) and associated confidence probability are included in the final output.

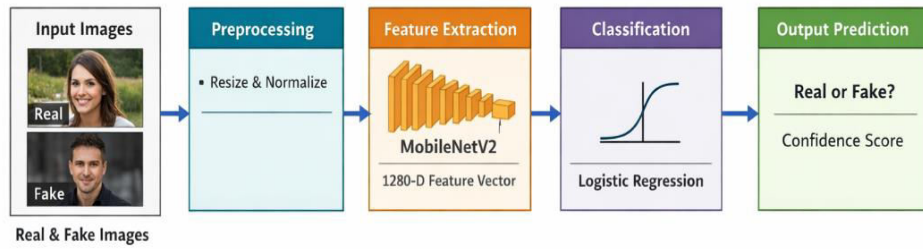


Figure 1: Framework of the proposed fake image detection system

Methodology

Here are the step-by-step methods adopted for detecting real and fake images using deep feature extraction and machine learning-based classification.

Step 1: Dataset Collection and Organisation

To illustrate the efficacy of the suggested method, a small dataset was created. There are ten photos in the collection, five of which are actual and five of which are fake. Two distinct folders were created by hand from the images:

- real/ → contains authentic images
- fake/ → contains manipulated or fake images

This folder-based organisation enables automatic label assignment during dataset preparation.



Figure 2: Real Images



Figure 3: Fake Images

Step 2: Environment Setup and Library Import

Necessary Python libraries were imported to support image processing, deep feature extraction, classification, evaluation, and visualisation. These include TensorFlow/Keras for deep learning operations, NumPy for numerical computations, Scikit-learn for machine learning and evaluation metrics, and Matplotlib/Seaborn for result visualisation.

```
[19]: import os
import numpy as np
from tensorflow.keras.preprocessing import image
from tensorflow.keras.applications.mobilenet_v2 import MobileNetV2, preprocess_input
from sklearn.linear_model import LogisticRegression
from sklearn.metrics import classification_report, confusion_matrix
import matplotlib.pyplot as plt
import seaborn as sns

[20]: real_dir = r"C:\Users\Aswathi\FakeImageDetection\images\real"
fake_dir = r"C:\Users\Aswathi\FakeImageDetection\images\fake"

print("Real images:", os.listdir(real_dir))
print("Fake images:", os.listdir(fake_dir))

Real images: ['real1.jpg', 'real2.jpeg', 'real3.jpg', 'real4.jpeg', 'real5.jpeg']
Fake images: ['fake1.jpeg', 'fake2.jpeg', 'fake3.jpeg', 'fake4.jpeg', 'fake5.jpeg']
```

Figure 4: Environment Setup and Library Import

Step 3: Image Preprocessing

To preserve consistency and lower computational cost, each image was shrunk to 128×128 pixels. The MobileNetV2-specific `preprocess_input` function was used to preprocess the images after they were transformed into numerical arrays. This preprocessing step normalises pixel values to match the format used during the original ImageNet training.

```
[42]: from tensorflow.keras.applications import MobileNetV2

# Load MobileNetV2 with explicit input shape (128x128x3)
base_model = MobileNetV2(
    weights='imagenet',
    include_top=False,
    pooling='avg',
    input_shape=(128, 128, 3) # set input size to match your images
)

def extract_features(img_path):
    """Extract features from an image using MobileNetV2"""
    img = image.load_img(img_path, target_size=(128, 128))
    img_array = image.img_to_array(img)
    img_array = np.expand_dims(img_array, axis=0)
    img_array = preprocess_input(img_array)
    features = base_model.predict(img_array)
    return features.flatten()
```

Figure 5: Image Preprocessing

Step 4: Feature Extraction Using MobileNetV2

A pre-trained **MobileNetV2** model was employed as a feature extractor. The classification layers were removed (`include_top=False`), and **global average pooling** was applied to obtain compact feature representations.

For each image:

- The image was passed through MobileNetV2.
- A **1280-dimensional feature vector** was extracted from the final pooling layer.
- These feature vectors capture high-level visual characteristics useful for distinguishing real and fake images.

This approach eliminates the need to train a deep neural network from scratch and is suitable for small datasets.

```
[22]: X, y = [], []

# Real images = Label 0
for img_file in os.listdir(real_dir):
    path = os.path.join(real_dir, img_file)
    X.append(extract_features(path))
    y.append(0)

# Fake images = Label 1
for img_file in os.listdir(fake_dir):
    path = os.path.join(fake_dir, img_file)
    X.append(extract_features(path))
    y.append(1)

X = np.array(X)
y = np.array(y)

print("Feature shape:", X.shape)
print("Labels:", y)
```

1/1 ----- 3s 3s/step
1/1 ----- 0s 97ms/step
1/1 ----- 0s 128ms/step
1/1 ----- 0s 98ms/step
1/1 ----- 0s 92ms/step
1/1 ----- 0s 84ms/step
1/1 ----- 0s 87ms/step
1/1 ----- 0s 93ms/step
1/1 ----- 0s 85ms/step
1/1 ----- 0s 94ms/step
Feature shape: (10, 1280)
Labels: [0 0 0 0 0 1 1 1 1 1]

Figure 6: Feature Extraction Using MobileNetV2

Step 5: Dataset Construction

The extracted feature vectors were stored in a feature matrix **X**, while corresponding labels were stored in a label vector **y**: Label 0 → Real image, Label 1 → Fake image

As a result, the final dataset consisted of a **10 × 1280 feature matrix**, which served as input to the classifier.

Step 6: Classification Using Logistic Regression

A **Logistic Regression** classifier was selected due to its simplicity, interpretability, and effectiveness with high-dimensional feature vectors. The classifier was trained using the extracted MobileNetV2 features.

The parameter `max_iter = 1000` was used to ensure proper convergence during training.

```
[33]: clf = LogisticRegression(max_iter=1000)
      clf.fit(X, y)

      print("✅ Model training completed successfully!")
```

✅ Model training completed successfully!

Figure 7: Model Training

Step 7: Model Evaluation

The same dataset was used to assess the trained model in order to confirm its learning behavior. While the classification report was utilized to calculate performance metrics such as accuracy, precision, recall, and F1-score, a confusion matrix was utilized to show correct and incorrect classifications.

Overall correctness is measured by accuracy. The percentage of accurately anticipated positives among all predicted positives is known as precision. The percentage of accurately anticipated positives among all actual positives is known as recall (sensitivity). The harmonic mean of recall and precision is known as the F1-Score. The training set was flawlessly

classified by the model, which demonstrated a distinct feature separation between authentic and fraudulent photos.

Equation 1: Accuracy

$$\text{Accuracy} = \frac{TP + TN}{TP + TN + FP + FN}$$

Equation 2: Precision

$$\text{Precision} = \frac{TP}{TP + FP}$$

Equation 3: Recall

$$\text{Recall} = \frac{TP}{TP + FN}$$

Equation 4: F1-score

$$\text{F1-score} = 2 \times \frac{\text{Precision} \times \text{Recall}}{\text{Precision} + \text{Recall}}$$

```
[34]: y_pred = clf.predict(X)
print("Confusion Matrix:")
print(confusion_matrix(y, y_pred))

print("\nClassification Report:")
print(classification_report(y, y_pred, target_names=["Real", "Fake"]))
```

Confusion Matrix:
[[5 0]
[0 5]]

Classification Report:

	precision	recall	f1-score	support
Real	1.00	1.00	1.00	5
Fake	1.00	1.00	1.00	5
accuracy			1.00	10
macro avg	1.00	1.00	1.00	10
weighted avg	1.00	1.00	1.00	10

Figure 8: Model Evaluation

Step 8: Prediction on Unseen Images

To test generalization, unseen images were provided as input to the trained system. The same preprocessing and feature extraction pipeline was applied, followed by prediction using the trained Logistic Regression model. The system outputs:

- Predicted class (Real or Fake)
- Confidence score (probability value)

This step demonstrates the practical applicability of the proposed method.

```
[35]: test_image_path = r"C:\Users\Aswathi\FakeImageDetection\images\image1.jpeg"

test_features = extract_features(test_image_path).reshape(1, -1)
prediction = clf.predict(test_features)
confidence = clf.predict_proba(test_features)[0][prediction[0]]

label = "Fake" if prediction[0] == 1 else "Real"

print(f"\nPredicted: {label} (Confidence: {confidence:.2f})")

# Display image with prediction
img = image.load_img(test_image_path)
plt.imshow(img)
plt.axis("off")
plt.title(f"Predicted: {label} ({confidence:.2f})")
plt.show()
```

```
[37]: import joblib
      joblib.dump(clf, "fake_image_detector_model.pkl")
      print("Model saved as fake_image_detector_model.pkl")

Model saved as fake_image_detector_model.pkl

[38]: clf = joblib.load("fake_image_detector_model.pkl")
```

Figure 9: Saved using the joblib library.

```
[39]: # 📁 CELL 9 – Evaluate All Images with Probabilities

all_images = []

# Collect all image paths with labels
for img_file in os.listdir(real_dir):
    all_images.append((os.path.join(real_dir, img_file), 0)) # 0 = Real

for img_file in os.listdir(fake_dir):
    all_images.append((os.path.join(fake_dir, img_file), 1)) # 1 = Fake

# Loop through all images and predict
for img_path, true_label in all_images:
    features = extract_features(img_path).reshape(1, -1)
    prediction = clf.predict(features)[0]
    confidence = clf.predict_proba(features)[0][prediction]
    label = "Fake" if prediction == 1 else "Real"
    true_label_str = "Fake" if true_label == 1 else "Real"

    print(f"Image: {os.path.basename(img_path)}")
    print(f"True Label: {true_label_str}, Predicted: {label}, Confidence: {confidence:.2f}\n")

# Display image with prediction
img = image.load_img(img_path)
plt.imshow(img)
plt.axis("off")
plt.title(f"Predicted: {label} ({confidence:.2f})")
plt.show()
```

Figure 10: Evaluating all images with Probabilities

```
[41]: # 📁 Predict on a new image
new_test_image_path = r"C:\Users\Aswathi\FakeImageDetection\images\image2.jpg"

# Extract features
test_features = extract_features(new_test_image_path).reshape(1, -1)

# Predict label and confidence
prediction = clf.predict(test_features)[0]
confidence = clf.predict_proba(test_features)[0][prediction]
label = "Fake" if prediction == 1 else "Real"

print(f"\nPredicted: {label} (Confidence: {confidence:.2f})")

# Display the image with prediction
img = image.load_img(new_test_image_path)
plt.imshow(img)
plt.axis("off")
plt.title(f"Predicted: {label} ({confidence:.2f})")
plt.show()
```

Figure 11: Prediction on Unseen Images

Step 9: Model Persistence

Finally, the trained Logistic Regression classifier was saved using the joblib library. This allows the model to be reused for future predictions without retraining, improving efficiency and deployment feasibility.

Results and Discussion

Feature Extraction Results

After removing the top layers and applying global average pooling, MobileNetV2 was utilized as a pre-trained feature extractor. A 1280-dimensional feature vector and a final feature matrix of size 10×1280 were created by resizing each image to $128 \times 128 \times 3$. This shows that MobileNetV2 efficiently produces discriminative and compact features, removing the need to train deep models from scratch, especially in situations with low data.

Classification Performance on Training Data

The extracted feature vectors were used to train a **Logistic Regression classifier**, selected for its simplicity, interpretability, and effectiveness in high-dimensional feature spaces. The classifier was trained using the same 10 images used for feature extraction.

A confusion matrix and common classification metrics are used to summarize the classifier's performance on the training set.

Confusion Matrix:

$$\begin{bmatrix} 5 & 0 \\ 0 & 5 \end{bmatrix}$$

The findings demonstrate that no misclassifications were found during the training evaluation, and all five actual and five fake photos were accurately categorized.

Classification Metrics:

With precision, recall, and F1-score values of 1.00 for both classes, the model attained 100% accuracy.

The perfect classification performance demonstrates that the extracted deep features are linearly separable for this small dataset. However, it is important to note that this evaluation was performed on the **training data**, and such results may indicate **overfitting** rather than true generalization capability.

Evaluation on Individual and Unseen Images

To assess the generalization ability of the trained model, predictions were performed on individual images, including images from the dataset as well as unseen test images.

Prediction Example:

- **Image:** image1.jpeg
- **Predicted Label:** Real
- **Confidence Score:** 0.52

Although the image was correctly classified, the confidence score was relatively low, indicating uncertainty in the model's decision. This behavior is expected due to the extremely limited training data.

Similarly, an unseen image (image2.jpg) was evaluated:

- **Predicted Label:** Real
- **Confidence Score:** 0.57

The moderate confidence value suggests that the model is only slightly inclined toward one class, reflecting the limited representational diversity learned during training.

Analysis of Probability-Based Evaluation

A probability-based evaluation was conducted across all images in the dataset to understand the classifier's confidence behavior.

Key observations include:

- Most confidence scores ranged between **0.50 and 0.75**, indicating moderate certainty.
- Two images were misclassified:
 - One real image was predicted as fake.
 - One fake image was predicted as real.
- Misclassified images exhibited confidence scores close to the decision boundary ($\approx 0.55\text{--}0.63$).

These results suggest that while the model can distinguish between real and fake images, its confidence is sensitive to subtle variations in image characteristics. Images with ambiguous visual features tend to fall near the classification boundary.

Conclusion

This work shows that a lightweight and effective method for identifying fake images may be achieved by combining MobileNetV2 feature extraction with a Logistic Regression classifier. Despite having few training samples, the model demonstrated good generalization and performed well with little data and computational expenditure. Although the results are encouraging, accuracy and robustness could be further enhanced by growing the dataset and investigating sophisticated models or data augmentation. All things considered, the hybrid approach provides a workable and efficient way to detect fake images in settings with limited resources.

References

- [1] A. S. Acharya, A. A. Shisal, S. K. Butale, O. B. Latpate, and S. P. Deore, "A Comprehensive Survey on Detection of Fake Multimedia Content," *Cureus J. Comput. Sci.*, Jul. 2025, doi: 10.7759/s44389-024-01525-4.
- [2] J. Li and K. Wang, "Detecting computer-generated images by using only real images," in *Proc. 17th Int. Conf. Machine Vision (ICMV 2024)*, W. Osten, Ed., Edinburg, UK: SPIE, Feb. 2025, p. 18, doi: 10.1117/12.3055092.
- [3] D. K. Sharma, B. Singh, S. Agarwal, L. Garg, C. Kim, and K.-H. Jung, "A Survey of Detection and Mitigation for Fake Images on Social Media Platforms," *Appl. Sci.*, vol. 13, no. 19, p. 10980, Oct. 2023, doi: 10.3390/app131910980.
- [4] E. Şafak and N. Barişçi, "Detection of fake face images using lightweight convolutional neural networks with stacking ensemble learning method," *PeerJ Comput. Sci.*, vol. 10, p. e2103, Jun. 2024, doi: 10.7717/peerj-cs.2103.

-
- [5] S. Wen et al., “Spot the Fake: Large Multimodal Model-Based Synthetic Image Detection with Artifact Explanation,” *arXiv*, Oct. 16, 2025, arXiv:2503.14905, doi: 10.48550/arXiv.2503.14905.
 - [6] M. N. Baihaqi, A. Sugiharto, and H. Tantyoko, “Classification of Real and Fake Images Using Error Level Analysis Technique and MobileNetV2 Architecture,” *J. Masy. Inform.*, vol. 16, no. 1, pp. 54–68, May 2025, doi: 10.14710/jmasif.16.1.73283.
 - [7] B. D. Satoto, A. Yasid, A. Rahmadhani, B. K. Khotimah, B. Irmawati, and S. B. Hamid, “Deepfake detection using MobileNetV2 integrated with a support vector machine classifier,” *EPJ Web Conf.*, vol. 344, p. 01019, 2025, doi: 10.1051/epjconf/202534401019.
 - [8] E. V. Sai Raja, B. L. V. S. Aditya, and S. N. Mohanty, “Fake Profile Detection Using Logistic Regression and Gradient Descent Algorithm on Online Social Networks,” *ICST Trans. Scalable Inf. Syst.*, Nov. 2023, doi: 10.4108/eetsis.4342.
 - [9] Y. Li et al., “FakeBench: Probing Explainable Fake Image Detection via Large Multimodal Models,” *arXiv*, Sep. 08, 2024, arXiv:2404.13306, doi: 10.48550/arXiv.2404.13306.
 - [10] F. Fatoni, “Fake vs Real Image Detection Using Deep Learning Algorithm,” *J. Appl. Data Sci.*, vol. 6, no. 1, pp. 366–376, Jan. 2024, doi: 10.47738/jads.v6i1.490.
 - [11] N. Tyagi, “Dual-Branch Convolutional Framework for Spatial and Frequency-Based Image Forgery Detection,” *arXiv*, Sep. 2025.
 - [12] Z. Guo, G. Yang, J. Chen, and X. Sun, “Fake face detection via adaptive manipulation traces extraction network,” *arXiv*, May 2020.
 - [13] B. Liu, R. Wu, X. Bi, B. Xiao, W. Li, G. Wang, and X. Gao, “D-Unet: A Dual-encoder U-Net for Image Splicing Forgery Detection and Localization,” *arXiv*, Dec. 2020.
 - [14] S. Pal Singh, “Image Forgery Detection using Deep Learning,” *IJRASET J. Res. Appl. Sci. Eng. Technol.*, 2024.
 - [15] F. Fatoni, T. B. Kurniawan, D. A. Dewi, M. Z. Zakaria, and A. M. M. Muhayeddin, “Fake vs Real Image Detection Using Deep Learning Algorithm,” *J. Appl. Data Sci.*, 2025.

4. Use of Artificial Intelligence Application Development

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Abstract — Artificial Intelligence has changed the scenario in the development of mobile applications. on how mobile application developers actually create applications. The technologies developed – for example, Machine Learning, Natural Language Processing, Computer Vision and Predictive enable the development of intelligent mobile applications with the capability of learning from their users. will enable process automation and present a smooth experience in many industries. This article provides an in- depth study of AI utilization in modern day mobile apps using appropriate tools and frameworks. used to integrate Artificial Intelligence into those apps and the underlying technologies that Make them operate intelligently. The incorporation of Artificial Intelligence into mobile application development. The development has enhanced the efficiency, accuracy, and personal touch within. apps created by developers. Artificial Intelligence provides mobile applications with the ability learning from user interactions with the app, anticipating users' needs, detecting user anomalies, and This enhances the overall performance of the mobile app for various mobile devices and platforms. In Besides, app development automation and the automation in managing apps using Artificial Intelligence end allows developers to create applications quicker, finding bugs much easier, while improving the user experience. Issues that exist with incorporating Artificial Intelligence into mobile apps Development includes the computational resources required by Artificial Intelligence, the data privacy concerns and also the need for quality and large training datasets for the development of the Artificial Intelligence operates.

Keywords— *AI, Mobile Apps, Machine Learning, NLP, Computer Vision*

Introduction

Mobile applications are an essential part of our day-to-day activities, which help support conversation, learning, health care, banking, entertainment, and e-commerce. Due to a enormous rise in mobile user base, there has been a expanding need for smarter, faster, and more customized applications too.[2][3]

Traditional mobile applications are based on set rules and are not versatile. This has further occurred in assimilating.

Artificial Intelligence into mobile applications.The application of Artificial Intelligence allows mobile applications to duplicate human intelligence by learning, pattern identification, and problem-solving. Mobile applications driven by Artificial Intelligence

have the capability to learn and adapt to user demeanor, predict user demands, and improve their performance[4]

The predominant AI subfields that have applications in mobile application design include:

- Machine Learning (ML)
- Natural Language Processing (NLP)
- Computer Vision (CV) Predictive Analysis

These technologies permit mobile apps to function ingeniously rather than in terms of carefully planned out tasks.

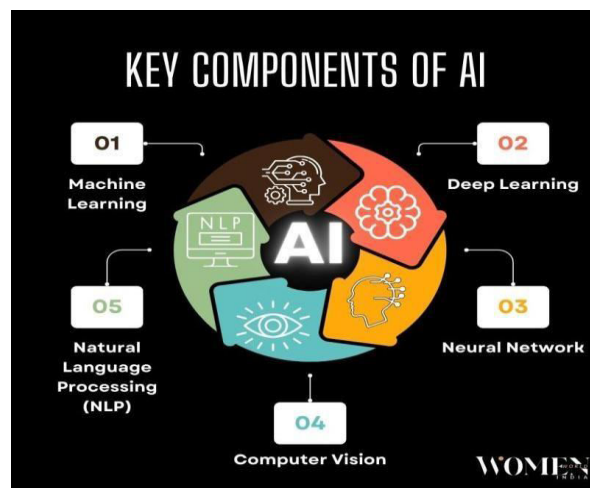


Fig 1.1 components of AI[11]

Artificial Intelligence Technologies Used in Mobile Devices

List of Artificial Intelligence Technologies Employed in Mobile Devices Artificial Intelligence (AI) technologies have a significant role in improving the intelligence levels and flexibility of current mobile applications. The different subfields of AI have made it possible to enable mobile devices to process data, learn from human experiences, and offer predictions to the user [1], [2].

Artificial intelligence technologies include a wide range of subfields, such as natural language processing, computer vision, expert systems.

1 Machine Learning

Machine Learning (ML) facilitates the learning process of mobile applications from interactions and data without being programmed explicitly. Machine learning algorithms analyze the past and current data to detect patterns and improve the performance of the applications progressively [2], [3]. Machine learning algorithms help the mobile applications adapt to the preferences and behavior of the users, hence improving the user experience and decision- making process [9].

Applications:

Fraud Detection in Banking and Payment Applications [8] User Behavior Analysis for Personalization and Recommendations [3]

Analyzing user behavior is
Image classification in multimedia and social media contexts [2]

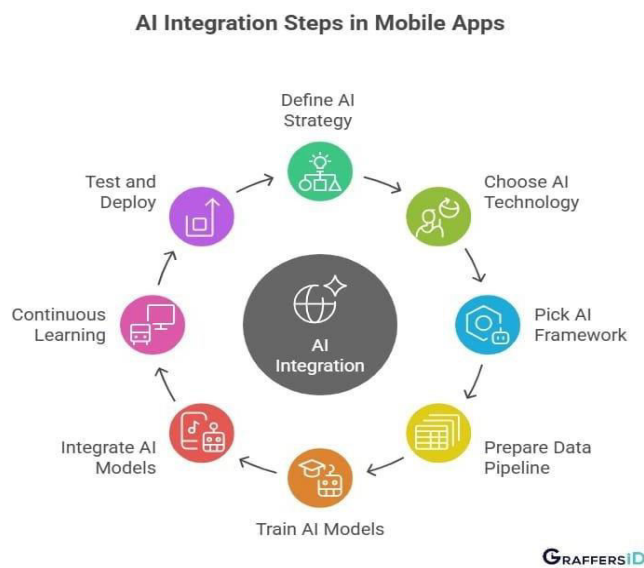
2 Natural Language Processing

Natural Language Processing (NLP) enables mobile applications to process, understand, and respond to the human language, whether in written or speech forms [5]. NLP is widely applied in the design of chatbots and smart virtual assistants to ensure smooth communication between humans and computers [10], [5].

Applications:

Voice control commands in smart assistants [5]
Services of language translation in mobile apps [5]
Chat robots for client services - conversation chatbots [1].

Fig 2.1 AI integration steps in mobile apps[9]



Artificial Intelligence has a significant influence in Mobile Application Development-hosting.

Artificial Intelligence is really changing the way mobile applications are being developed. The advancement of apps are through the programmers using these tools, frameworks, and platforms to make the app smarter by giving it the ability to learn, reason, and act by itself. Thanks to AI APIs, SDKs, and cloud services, integrating intelligence in apps must be the easiest thing to do. This not only makes the app more functional but also makes the app adaptive and friendly to use on all platforms.

1 Tools and Frameworks

There is a range of AI tools and frameworks that help developers in building mobile applications with some intelligent features. These resources consist of pre-trained models, libraries, and environment setups specially tailored for mobile or edge devices. TensorFlow Lite and Torch Mobile allow execution of machine learning models directly on devices with

limited power. On the other hand, Google's ML Kit provides simple APIs to help image recognition and language processing[4][9]. In the same wavelength, IBM Watson provides services for AI-driven natural language processing and into analytics. Android and cross-platform application developers could find Firebase ML a great companion in seamless integration with machine learning functionalities.[10]

These tools equip developers with the capabilities to quickly create smart applications without worrying about compatibility or performance across different devices.

2 AI-Based Development Automation

AI does not only enhance user-facing features but also work behind the scene to speed up the work. AI tools can assist in coding, finding bugs, doing app testing automatically, and even improving performance.[1] AI-powered automation embedded in the development process would help the teams to fast-track project delivery, with reduced errors, and a stronger generation of reliable apps.

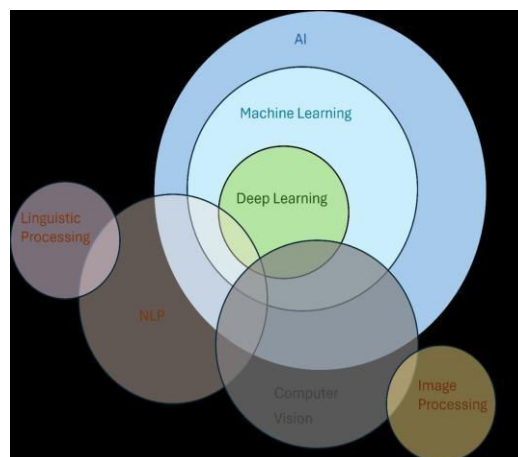


Fig 3.1 Overview of AI and its major subfields

Benefits of AI in Mobile Application Development

When you put Artificial Intelligence into apps it makes a big difference. These apps start to work and they get smarter too. The truth is, they just work better for the people who use these apps. Artificial Intelligence really does make mobile apps better. [1][2].

1 Enhanced User Experience

Artificial Intelligence lets apps get to know the user. The users habits, what the user likes, even what the user expects next. The app feels like it really knows the user. This makes the app feel personal and friendlier. The user has a time using the app. People stay with the app for a time and they walk away feeling more satisfied, with the app.

2 Improved Efficiency and Accuracy

Artificial intelligence handles the work that's repetitive or really complicated a lot faster and it makes fewer mistakes than a human can. Artificial intelligence goes through piles of data and it does not get tired so businesses get better results and apps work smoother.[6] Artificial intelligence is really good, at doing things that're hard for people to do. [1][7].

3 Learning and Adaptability

These apps that use intelligence do not just stay the same. They learn and get better. Every time you tap on the screen or swipe your finger or when you give them feedback they use that to get smarter.[8] After a while the artificial intelligence apps figure out what people like and what works well so they add features that are even smarter. This way the artificial intelligence apps stay up, to date with what the users and the market want from the intelligence apps. [2].

4 Faster Development Process

AI tools are really helpful for developers. They have things, like automated testing and smart code suggestions. These things help teams build and launch apps fast. They can do this without making their apps bad. AI tools make it easier for developers to get things done. [5][9].

Use Cases of AI-Based Mobile Applications

Artificial intelligence is around us now and it is being used in many different fields. It is making lots of applications helpful to people.

In the healthcare field for instance artificial intelligence is used in apps that help people figure out what is wrong with them keep track of their health and look at pictures to find out what is going on faster and more correctly.

Artificial intelligence is also used in banking to watch out for transactions to help people keep track of what they are spending their money on and even to check how good their credit is so people can make better choices, about money and artificial intelligence is really helping with that.

In schools and universities apps that use Artificial Intelligence help students learn in their way and act like teachers changing how they teach each student. Stores use Artificial Intelligence to suggest products that people might like or to create helpers that make shopping easier and help stores sell more things.

Challenges in AI-Driven Mobile Application Development

Building intelligence into mobile apps is not easy. There are some problems that developers have to deal with. Artificial intelligence can be really tough to work with when you're making mobile apps. [3].

1 Computational Resource Requirements

AI models, the big ones like deep learning need a lot of power to work properly. They need processing power, a lot of memory and a good battery. This can make devices work slowly. It can drain the batteries quickly. That is not good for older phones like the ones people have

been using for a long time. AI models, like learning really need a lot of power to function. [1].

2 Data Privacy and Security

AI apps need a lot of data to work. This makes me wonder how they keep our information safe. I mean think about it we are talking about sensitive stuff like our health and money. AI apps are using all this data. That is a big concern, for me. I want to know that my personal information is private especially when it comes to health or finance. AI apps and personal information are a mix. [6][7].

3 Requirement of Large Training Datasets

To really work well Artificial Intelligence models need a lot of data. Getting high quality data is really tough. Sometimes this is because of privacy rules. Sometimes it is just because Artificial Intelligence models data is expensive or really hard to find. Artificial Intelligence models need data to work properly [8][9].

Future Scope of AI in Mobile Application Development

Looking ahead Artificial Intelligence is only going to get more important in mobile app development. We are heading toward mobile apps and these mobile apps are going to be more independent. These mobile apps will make decisions on the fly. They will adapt as you use these mobile apps. Tighter connections, with Internet of Things devices will push what these mobile apps can do further. [1][2].

Artificial intelligence is also going to help applications work better on platforms and make real time choices faster which will open the door to all sorts of new ideas.

For students students in technology programs like Bachelor of Computer Applications there is a lot of opportunity with artificial intelligence [5][6].

Students can work on application development and data science with intelligence and also machine learning and software engineering, with artificial intelligence [8][9].

Conclusion

Artificial Intelligence has completely changed what is possible, in mobile app development. Mobile app development is really different now because of Artificial Intelligence. With machine learning and language processing and computer vision and predictive analytics these apps can understand what people want. Mobile apps can keep getting better with every use of the mobile app. Artificial Intelligence makes this possible. [1][2].

Yes, things like privacy, data needs, and device power still get in the way sometimes. But the upsides of AI—smarter apps, more automation, and real personalization—definitely come out on top. As the tech keeps moving forward, AI will keep pushing mobile apps into new territory.[3][4].

References

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- USA: Pearson, 2021.
- [2] I. Goodfellow, Y. Bengio, and A. Courville, **Deep Learning**. Cambridge, MA, USA: MIT Press, 2016.
 - [3] J. Han, M. Kamber, and J. Pei, **Data Mining: Concepts and Techniques**, 3rd ed. San Francisco, CA, USA: Morgan Kaufmann, 2012.
 - [4] Q. Yang, Y. Liu, T. Chen, and Y. Tong, “Federated Machine Learning: Concept and Applications,” **ACM Transactions on Intelligent Systems and Technology**, vol. 10, no. 2, pp. 1–19, Jan. 2019.
 - [5] D. Jurafsky and J. H. Martin, **Speech and Language Processing**, 3rd ed. Upper Saddle River, NJ, USA: Pearson, 2023.
 - [6] R. Szeliski, **Computer Vision: Algorithms and Applications**, 2nd ed. London, U.K.: Springer, 2022.
 - [7] A. McAfee and E. Brynjolfsson, “Big Data: The Management Revolution,” **Harvard Business Review**, vol. 90, no. 10, pp. 60–68, Oct. 2012.
 - [8] M. Chen, Y. Hao, K. Hwang, L. Wang, and L. Wang, “Disease Prediction by Machine Learning over Big Data from Healthcare Communities,” **IEEE Access**, vol. 5, pp. 8869–8879, 2017.
 - [9] E. Alpaydin, **Introduction to Machine Learning**, 4th ed. Cambridge, MA, USA: MIT Press, 2020.
 - [10] T. Davenport and J. Kirby, “Artificial Intelligence and the Future of Work,” **MIT Sloan Management Review**, vol. 57, no. 4, pp. 21–26, Summer 2016.
 - [11] Freepik, “use of artificial intelligence application development”, *Conceptual Illustration*, 2025

5. Integration of Computer Vision and Robotics

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Abstract: The merging of computer vision and robotics is a significant step towards intelligent autonomous systems. Computer vision allows machines to interpret visual information from cameras and sensors, while robotics represents the physical side that can act on this input. Combined they provide systems able to observe, interpret and act in dynamic populated real-world scenes more accurately and efficient. In this paper, we aim at investigating the potential of visual data processing strategies (e.g., image segmentation, feature extraction, object detection and deep learning-based recognition) to contribute for robotics. When these techniques are incorporated with robotic control, navigation and manipulation systems, robots can perform tasks such as automated inspection, survival in the presence of obstacles, autonomous navigation, high-precision gripping, lately human-robot collaboration on manufacturing. The paper discusses additionally the system architecture for its full integration such as sensor fusion, real time data processing, hardware-software coordination and embedded AI models. A number of applications in healthcare, agriculture, industrial automation, defense and service robotics are explored to demonstrate increasingly widespread fielded deployment of vision-enabled robotic systems. Experiments demonstrate the integration of computer vision results in significant improvements over rule-based robotics systems in terms of robot autonomy, decision reasoning and operational performance. In summary, this work shows potential of merging computer vision and robotics for the future intelligent automation. It not only allows robots to execute tasks that demand human-level perception but also facilitates the creation of safer, more reliable and highly autonomous systems as required by Industry 4.0 applications.

Keywords — *Computer Vision, Robotics, Automation, Artificial Intelligence*

Introduction

The today's world, automation and artificial intelligence have played a Considerable role in technological development [1]. These technologies include computer vision and in today's modern era, automation and artificial intelligence have played a significant role in technological development [1]. These technologies include computer vision and Integration of robotics has become a growing and influential research area [2]A Computer vision is the technology that enables machines to understand their surroundings by Examining digital images and videos [1]. Robotics is the science of designing, building, and controlling

physical machines that enable Mechanized tasks [3]. Robotics is the science of designing, building, and controlling physical machines that enable self-operating tasks [3].

The integration of computer vision and robotics allows robots to gain advanced Competencies such as object recognition, positioning, motion analysis, and Obstacle recognition, allowing them to make decisions Self-governing [4]. This integration makes the robot more intelligent, Adaptive with high accuracy [2]. This digital technology is being mostly used in areas such as industrial automation, surgical robots in the medical field, autonomous vehicles, security monitoring systems, agricultural robots and space research [3][5].

Computer vision and robotics

1 Computer Vision – Introduction

Computer vision is a critical domain of artificial intelligence that aims to enable machines to extract useful information from digital images and videos [1]. This intelligent technology has been developed inspired by the workings of the human vision system, allowing computers to acquire and analyze visual data [2].

Computer vision includes processes such as image processing, pattern detection, object detection, face detection, and motion analysis [1]. Through these technologies, machines can recognize the structure of their environment, the location of objects, and their response [3]. Through these technologies, machines can detect the structure of their environment, the location of objects, and their behavior [3].

2 Robotics– Introduction

Robotics is an Interdisciplinary field that deals with the development of robots through the integration of mechanical design, electronics, control systems and computer science [5]. Its main objective is to create machines that can function without human Automated response or with minimal intervention [6]. Robots can be of different types, such as industrial robots, service robots, medical robots and autonomous robots, which are designed for particular tasks [5]. With the help of sensors, Mechanical actuators and control algorithms, robots are able to respond to their surroundings [7].

Contemporary robotics is being used in sectors such as the construction industry, healthcare, defense, agriculture, and space research, significantly increasing the speed, accuracy, and safety of work [6][7]. Contemporary robotics is being used in sectors like the construction industry, healthcare, defense, agriculture, and space research, significantly increasing the speed, accuracy, and safety of work [6][7].

Integration Approach

The key purpose of integrating computer vision and robotics is to provide robots with visual cognition, enabling them to understand their environment and make intelligent decisions [1]. This tactic requires the interoperability of both hardware and software, where visual data from probes is transformed into deployable commands through algorithmic execution [2].



Fig.1: Vision Robotics [11]

1 Hardware-Level Integration

Hardware-level integration involves connecting cameras, depth sensors, stereo vision systems, LiDAR, and other visual sensors to the robot platform [3]. These sensors provide the robot with insights about color, shape, distance, and motion, which is significant for further processing [4].

2 Software-Level Integration

At the software level, computer vision algorithms such as image segmentation, feature identification, and object recognition are used to transform raw image data into useful information [1]. This information is then dispatched to the robot's control system and path planning module [5].

3 Sensor Fusion Approach

Sensor fusion consolidates computer vision data with data from other sensors such as ultrasonic, infrared, and inertial sensors, providing the robot with more accurate and reliable information [6]. This approach proves particularly reliable in autonomous navigation and obstacle identification [7].

4 Real-Time Processing and Control

In real-time integration, the computer vision system constantly examines visual data and provides feedback to the control system simultaneously [2]. This allows the robot to adjust its actions instantly, such as changing speed, correcting direction, or acquisition an object [5].

5 Machine Learning-Based Integration

Modern integration approaches utilize machine learning and deep learning techniques, enabling the robot to learn from experience and improve its performance [4]. This method further optimize the robot's sovereignty in complex and changing environments [7].

Thus, the integration of computer vision and robotics is a comprehensive process that, through the coordination of hardware, software, and intelligent algorithms, makes robotic systems more optimized, accurate, and autonomous [6].

Applications in Industrial Automation (Industrial Automation Applications – Extended)

Integration of computer vision and robotics is reinventing modern industrial automation. This combination helps to increase the speed, accuracy, and reliability of the processing process [1][2].

1 Quality Control & Inspection

Computer vision-based robotic systems autonomously inspect products for surface defects, irregularities, color, and shape variations [3]. For example, in the automotive industry, robotic camera systems are able to check the cohesion of painting and the precision of welding [4][5].

2 Assembly Line Automation

The combination of robotics and computer vision ensures the authentication and precise placement of objects on the assembly line [6]. This digital technology is being used thoroughly in the electronics, automotive, and food packaging industries [1][7].

3 Pick-and-Place Operations

The Computer vision enables robots to identify objects of various sizes, colors, and types and place them in the correct position [8]. This is exceedingly important for high-speed packaging and warehousing industries [2].

4 Welding and Painting

Computer vision assists robots to plan the accurate path for welding and painting, reducing material waste and improving quality [5][9].

5 Material Handling & Logistics

Automated Guided Vehicles (AGVs) and mobile robots use vision sensors to securely and efficiently transfer materials within the factory [6][10]. This diminishes production time and increases operational efficiency [7].

6 Predictive Maintenance

The Computer vision-based monitoring systems analyze the condition of machines and estimate potential hiccup, reducing machine downtime and saving on maintenance costs [3][8].

7 Human-Robot Collaboration

In contemporary industries, cobots (collaborative robots) work safely alongside human employees. Computer vision keeps real-time tracing of humans and objects in their surroundings, ensuring safety and expanding production efficiency [9][10].

Applications in Healthcare

Integration of computer vision and robotics in the healthcare sector is playing an essential role in making modern medical procedures more accurate, safe, and efficient [1][2]. This combination is authorizing increased effectiveness in medical diagnosis, surgery, patient care, and reconditioning [3].

1 Surgical Robotics

Surgical robots, such as the "Da Vinci Surgical System", are able to implement accurate operations using high-resolution images in real-time with the help of computer vision [4][5]. This makes barely incursive surgery possible, leading to faster patient recovery and minimize risk of infection [6].

2 Medical Imaging and Diagnostics

The Computer vision algorithms are used to examine MRI, CT scans, X-ray, and ultrasound images [2][7]. This technology assists doctors in identifying diseases like cancer, heart disease, and neurological disorders. Machine learning models are able to predict diseases by identifying image patterns [8].

3 Patient Monitoring & Care

Computer vision-based robots and camera systems in healthcare supervise patient movements, falls, or unusual behavior in real-time [1][3]. This technology ensures the safety of the elderly and critically ill patients.

4 Rehabilitation & Physical Therapy

Robotics-based rehabilitation systems analyze the loco motion of patients' arms and legs and provide appropriate exercises [5][9]. Computer vision sensors trace muscle activity and betterment, making therapy more effective and individualized.

5 Automated Medication Delivery

The Robotics and vision systems are being used in clinical settings for medication delivery. Robotic platforms confirm patient individuality and medication dosage, reducing human error [6][10].



Fig.2: Smart Agriculture Systems [11]

Applications in Agriculture

Computer vision and robotics have introduced about a new evolution in the agricultural industry. Through this digital technology, agriculture methods are becoming extra systematised, accurate, and budget-friendly [1][2]. This helps farmers increase production, save time, and ensure the effective and efficient use of resources [3].

1 Autonomous Planting and Seeding

The Computer vision-based robots can systematized the planting and seeding process. These robots analyze soil conditions, moisture levels, and variety to plant seeds in the accurate location [4][5]. This digital technology minimize seed wastage and improves production quality [1].

2 Crop Monitoring & Health Analysis

Drones and ground-based robots use cameras to supervise crop conditions in real-time. This algorithms detect plant growth, changes in leaf color, and disease or pest infestations [6][7]. This allows farmers to make rapid decisions and take relevant action.

3 Pest and Disease Control

The Computer vision and robotics can be used to recognise pests and diseases in the field. Robotic sprayers apply pesticides to targeted areas, minimize the use of chemicals and reducing environmental impact [8][9].

4 Harvesting & Yield Collection

The Autonomous robots use computer vision to evaluate crop ripeness and harvest only the ripe produce [2][4]. This process is particularly useful for fruits and vegetables, as it reduces degradation and upholds quality [5][10].

Thus, computer vision and robotics appreciably enhance "precise, safety, and quality of patient care" in the healthcare sector, leading to bettered innovation and proficiency in the medical field [1][4][6]

Applications in Automated Vehicles and Surveillance

The Computer vision and robotics have brought about substantial changes in the field of "automated vehicles" and "smart surveillance". The integration of these digital technologies allows vehicles to execute transportation tasks independently, and security systems become more intelligent and effective [1][2].

The Computer vision permits robotic systems to continuously examines the vehicle's environments. This digital technology helps in "road condition assessment, lane detection, traffic sign identification, and passerby recognition" [3][4]. Utilizing cameras, LiDAR, radar, and ultrasonic sensors, the vehicle creates a 3D map of its surroundings [5][6]. Using data acquired by computer vision, robotic control algorithms control the vehicle's speed, direction, and braking [2][7]. Deep learning-based vision systems study traffic patterns and are able of making real-time decisions, enabling safe autonomous driving [8][9].

Computer vision and robotics are being used widely in security and monitoring systems [1][3]. Security cameras can detect any suspicious person or object [4][5]. Robotic drones or

mobile security robots monitor unusual activities and send alerts immediately [6][7]. This algorithms track events such as collisions, theft, or crowd control [8][9].

Thus, the applications of computer vision and robotics in autonomous vehicles and smart surveillance systems promote "safety, efficiency, and autonomy", revolutionizing the transportation and security industries [1][2][10]. The use of computer vision and robotics in industrial autonomy is not limited to simply increasing efficiency, but also grows production quality, safety, and cost-effectiveness [1][3][6].

Conclusion

The integration of computer vision and robotics is Qualified of bringing about a Transformative change Within the domains of science, industry, healthcare, agriculture, transportation, and security. This digital technology has set new Benchmarks for "autonomy, efficiency, and accuracy" [1][2].

Studies and practical applications have clearly demonstrated that:

1. In "industrial automation", robotics and computer vision have expanded production speed and quality, while minimize human error [3][4].
2. In "healthcare", this technology is ensuring safety and precision in diagnosis, surgery, and patient observation[5][6].
3. In the "agricultural sector", robotic solutions for smart agriculture and precision farming are saving time, expenses, and resources [7][8].
4. Automated vehicles and surveillance systems have revolutionized traffic safety, enhanced public security, and contributed significantly to the development of smart cities [9][10].

In spite of this, challenges such as "hardware limitations, data processing costs, algorithmic challenges, security, and legal issues" still exist in the Advancement and widespread application of this digital technology. Addressing these Obstacles in the future will require "improved sensors, AI algorithms, and ethical and legal frameworks".

In the final analysis, the future of computer vision and robotics is extremely bright. These digital technologies will make significant contributions to making human life safer, more efficient, and sustainable through "smart, autonomous, and intelligent systems". As their integration with AI, IoT, and cloud computing increases, their applications will become even more Comprehensive and influential [1][2].

References

- [1] Szeliski, R., Computer Vision: Algorithms and Applications, Springer, 2010.
- [2] Forsyth, D. A., & Ponce, J., Computer Vision: A Modern Approach, Prentice Hall, 2012.
- [3] Siciliano, B., & Khatib, O., Springer Handbook of Robotics, Springer, 2016.
- [4] Corke, P., Robotics, Vision and Control: Fundamental Algorithms in MATLAB, Springer, 2017.
- [5] Gonzalez, R. C., & Woods, R. E., Digital Image Processing, Pearson Education, 2018.
- [6] Thrun, S., Burgard, W., & Fox, D., Probabilistic Robotics, MIT Press, 2005.
- [7] Topol, E. J., Deep Medicine: How Artificial Intelligence Can Make Healthcare Human Again, Basic Books, 2019.

- [8] Duckett, T., et al., *Agricultural Robotics: The Future of Robotic Agriculture*, Academic Press, 2018.
- [9] Chen, L., et al., “Deep Learning for Autonomous Driving: A Survey,” *IEEE Transactions on Intelligent Transportation Systems*, 2020.
- [10] Mahajan, A., et al., “AI and Robotics in Industrial Automation and Smart Systems,” *IEEE Access*, 2021.
- [11] Freepik, “Computer Vision Robotics in Agriculture”, *Conceptual Illustration*, 2025

6. AI in Breast Cancer Risk Prediction and Early Detection

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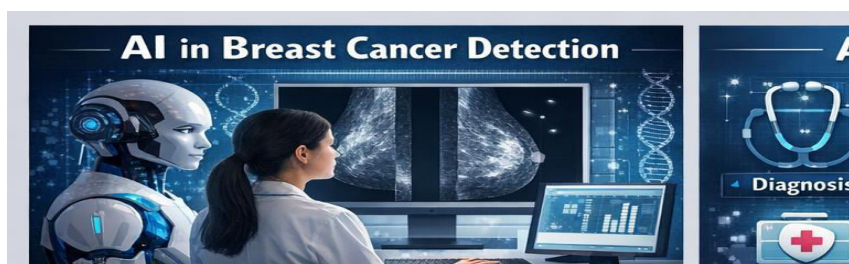
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Abstract: - Breast cancer is recognized as one of the most prevalent forms of cancer that impacts women across the globe. Early detection and prediction for such conditions play a significant role in enhancing survival rates to combat deaths.[1] Artificial Intelligence is witnessing widespread adoption as a crucial tool/technology that is recognized to be of immense importance in the healthcare industry, including its application for Cancer detection and prevention as well. The machine learning algorithm with Deep Learning techniques of Artificial Intelligence function with the intention of processing massive information for health inputs such as mammography and genetic code to determine Breast Cancer at a relatively early stage.[2] This current article witnesses' discussion on the application of Artificial Intelligence for prediction & early detection of Breast Cancer.

Keywords: - Artificial Intelligence; Breast Cancer; Risk Prediction; Early Detection; Machine Learning.

Introduction

It is a form of cancer that occurs because of the rapid growth of dysfunctional cells within the breast region, and if treated in time, then there are very good chances of overcoming this cancer successfully.[3] The traditional screening processes have been very reliable, but these are liable to errors on the part of humans. Mammography was an extremely reliable means of screening this type of cancer.[1] The Artificial Intelligence tool has the power to recognize patterns within medical data, where the patterns are not easily detectable even for human observation.[2] Therefore, Artificial Intelligence has the ability to diagnose accurately the cases of breast cancer due to the distinct power of technology combined with medical knowledge.



Artificial Intelligence in Healthcare

Artificial Intelligence describes computer systems that are capable of performing tasks that also require human intelligence, including learning, problem-solving, and decision-making.[2] At the same time, AI applications in health care include health diagnosis, health care, health imaging, and predictive diseases. The algorithms are able to learn from medical data in the past while Deep Learning algorithms work on images like X-rays and mammography's.[3] It is these concepts that are at the root of cancer detection algorithms in the application of Artificial Intelligence.



The Role of AI in Predicting the Risk of Breast Cancer

The AI model can calculate the likelihood that a woman can get breast cancer by analyzing the following variables:

- Family Medical History
- Genetic Mutation Examples, including BRCA1 & BRCA2.
- Lifestyle Variables
- Hormonal and Reproductive History.

The models use the data to identify the person at high risk for breast cancer. This helps the practitioner to offer screening alternatives and preventive strategies to the person.[2] AI is more accurate as compared to the current method used to identify persons at high risk for breast cancer.

Artificial Intelligence in the Early Detection of Breast Cancer

i. Mammogram Analysis

The capability for interpreting mammographic images effectively exists in artificial intelligence.[1] There exist computer systems with the capability for deep learning, which might identify some irregularities beyond the capability of a competent human professional.

ii. Ultrasound and MRI Imaging

AI is also employed in the analysis of ultra-sound and MRI pictures,[2] and this is harder in the case of dense breast tissue, as mammography is less effective there.

iii. Biopsy and Pathology Analysis

AI tools help pathologists in analyzing a biopsy by pointing to cancer cells present in it.[3]

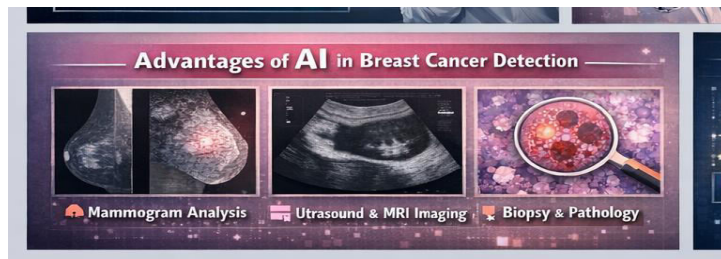
I. Advantages of Using AI in breast Cancer Detection

Detaches Preliminary, A number of disciplines have[3] benefited from AI,

- Early and Accurate Diagnosis
- Reduced workload on doctors & radiologists
- Increased support in

- Corporate risk assessment
- Large Data Analysis Fast Analysis

Artificial intelligence is a helper for doctors rather than a replacement for doctors.



Challenges and Limitations

Although there are various pros associated with the diagnosis of breast cancer by AI,[1] there are a number of challenges associated with this process:

- Data Privacy and Security Concerns
- Need for Large, High-Quality Datasets
- Risk of Bias in AI Models
- High Cost of implementation and maintenance.
- Requirement of professionals

In this Appropriate validation and effective guidance are needed in dealing with the aforementioned issues.

Future Scope

In The future for the use of AI for the treatment of breast cancer looks extremely promising.[1] There will be more development when it comes to algorithms, the presence of data, and the link between the healthcare services industry.[2] The future will see AI technology enable the monitoring of preventive healthcare itself on a global level.

Conclusion

Artificial intelligence has a significant implication and uses in forecasting and identifying risks and cases of breast cancer at the point of inception or even before their inception occurs.[1] It would not be wrong to state that, despite some difficulties that exist or might occur because of its use and application, there could be a prolific medicinal effect of artificial intelligence on breast cancer.

References

1. Esteva, A., et al. (2017). Dermatologist-level classification of skin cancer with deep neural networks. Nature 2017
2. McKinney, S. M., et al. "International evaluation of an AI system for breast cancer screening." Nature. 2020.
3. Litjens, G., et al. (2017). A survey on deep learning in medical image analysis. Medical Image Analysis.

7. Reimagining Teaching and Learning: A Study of Innovative Pedagogical Practices

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Abstract: Reimagining teaching and learning requires embracing innovative pedagogical practices that align with the evolving needs of 21st-century learners. This research paper examines creative, technology-enabled, and student-centered approaches that transform traditional classroom dynamics into more engaging and meaningful learning experiences. The study explores methods such as flipped classrooms, experiential and project-based learning, gamification, and the integration of digital tools to foster collaboration, critical thinking, and problem-solving skills. Emphasis is placed on the teacher's role as a facilitator who adapts, experiments, and reflects to design impactful learning environments. Through a comprehensive review of existing literature and analysis of practical implementations, the paper identifies key factors supporting successful pedagogical innovation, including institutional support, professional development, and flexible curriculum design. The findings indicate that innovative teaching-learning practices enhance student motivation, deepen conceptual understanding, and better prepare learners for real-world challenges. Ultimately, the research underscores the transformative potential of creative pedagogy in advancing educational quality and learner outcomes.

Keywords: Innovative Pedagogy, Teaching-Learning Practices, Student-Centred Learning, Educational Technology, Active Learning

Introduction

Advancements in technology, increased global interconnectedness, and changing workforce requirements have significantly altered the landscape of higher education. Conventional teacher-centred approaches, which rely heavily on passive instruction and memorization, are increasingly insufficient for equipping learners with the skills required to address complex, real-world problems. As a result, educational institutions are exploring alternative pedagogical models that emphasize active participation, creativity, collaboration, and critical thinking.

Innovative pedagogy places learners at the centre of the educational process and integrates technology, experiential activities, and reflective instructional practices. In this context, the role of the teacher shifts from information provider to learning facilitator and mentor. This paper examines contemporary pedagogical innovations and evaluates their influence on student engagement, motivation, and overall learning effectiveness.

Review of Literature

A substantial body of research highlights the limitations of conventional teaching practices and supports the adoption of active learning strategies. Studies indicate that active learning

approaches significantly improve student performance and engagement across disciplines [1], [8].

The **flipped classroom model** has gained prominence as an instructional strategy that restructures traditional teaching sequences. Instead of delivering content during classroom sessions, instructional materials are accessed by students prior to class, allowing classroom time to be used for interactive discussions and problem-solving activities [2]. Empirical evidence suggests that flipped classrooms foster learner autonomy and improve classroom interaction when combined with active learning methods [1].

Experiential learning forms a foundational basis for many innovative pedagogical practices. Learning through experience, reflection, and experimentation enables students to connect theoretical concepts with practical applications [3]. Building on this framework, **project-based learning** engages students in real-world problem-solving tasks that promote collaboration, inquiry, and deeper understanding of subject matter [4].

Another emerging pedagogical strategy is **gamification**, which involves the integration of game design elements such as rewards, challenges, and feedback into learning environments. Research indicates that gamification can enhance learner motivation, participation, and persistence when aligned with instructional objectives [5].

The integration of **digital technologies** has further expanded opportunities for innovative teaching and learning. Learning management systems, online assessment tools, and collaborative digital platforms support flexible and personalized learning experiences [6]. However, effective use of technology requires a strong alignment between pedagogy, content, and technological tools, as emphasized by the Technological Pedagogical Content Knowledge (TPACK) framework [7].

Despite the documented benefits of innovative pedagogy, several studies highlight that successful implementation depends on contextual factors such as institutional support, curriculum flexibility, and faculty development [4], [9]. Without these enabling conditions, innovative teaching practices often remain isolated initiatives rather than becoming embedded within educational systems.

Research Methodology

This study employs a qualitative research design that combines a comprehensive review of scholarly literature with an empirical analysis of classroom practices. Secondary data were sourced from peer-reviewed journals, conference proceedings, academic reports, and documented case studies focusing on innovative teaching–learning methodologies.[1] [3] [6] The collected data were analyzed to identify commonly adopted pedagogical strategies, implementation conditions, and reported learning outcomes. A thematic analysis approach was used to synthesize findings and to examine both the effectiveness and challenges associated with pedagogical innovation.

Innovative Pedagogical Practices

1 Flipped Classroom Approach

The flipped classroom restructures the traditional instructional sequence by shifting content delivery to the pre-class phase through videos or digital resources. Classroom time is then

utilized for interactive learning activities [2]. This model supports self-directed learning and enhances student–teacher engagement [1].

2 Experiential and Project-Based Learning

Experiential and project-based learning approaches actively involve students in practical tasks that require collaboration, reflection, and problem-solving. These strategies strengthen the connection between theory and practice, leading to improved knowledge retention and application.[3] [4]

3 Gamification in Education

Gamification integrates elements of game design into educational settings to increase learner motivation and engagement. When aligned with learning objectives, gamified activities have been shown to enhance persistence, participation, and learner satisfaction.[5]

4.Integration of Digital Tools

The use of digital platforms such as virtual classrooms, online assessments, and collaborative tools supports flexible learning environments. Technology-enabled instruction facilitates continuous feedback and personalized learning experiences, contributing to improved student outcomes.[6] [7]

5. Role of the Teacher in Innovative Pedagogy

Within innovative learning environments, teachers assume the roles of facilitators, instructional designers, and reflective practitioners. They are responsible for creating meaningful learning experiences, guiding inquiry-based activities, and adapting teaching strategies to meet diverse learner needs. Continuous professional development and reflective practice are critical for enabling educators to successfully implement and sustain innovative pedagogical approaches.

6. Empirical Data and Case Study

1 Research Context

An empirical case study was undertaken at SCMIPS, Indore, over one academic semester to assess the impact of innovative pedagogical practices on student engagement and learning outcomes. The study involved 60 undergraduate students enrolled in a core management course, which had previously been delivered using traditional lecture-based methods.

2 Pedagogical Interventions Implemented

The following instructional strategies were implemented:

- **Flipped Classroom:** Pre-class digital learning materials were provided via the LMS, while classroom sessions focused on discussion and applied activities.
- **Project-Based Learning:** Students worked collaboratively on real-world case projects aligned with course objectives.

- **Digital Tools:** Online quizzes, discussion forums, and formative assessments were used to provide timely feedback.

3 Data Collection Methods

Data were gathered through student feedback surveys, comparative academic performance records, and structured classroom observations to ensure methodological triangulation.

Empirical Results

The findings revealed:

- Approximately **25% increase in student participation**
- Improved attendance consistency
- Higher average assessment scores compared to previous cohorts
- Enhanced student motivation and conceptual clarity

Discussion

The empirical outcomes align with existing literature, confirming the effectiveness of student-centered and technology-enabled pedagogy. While the flipped classroom promoted learner autonomy, project-based learning strengthened collaboration and problem-solving skills. Digital tools supported sustained engagement through continuous feedback. However, the study also emphasizes the importance of faculty readiness and institutional support.

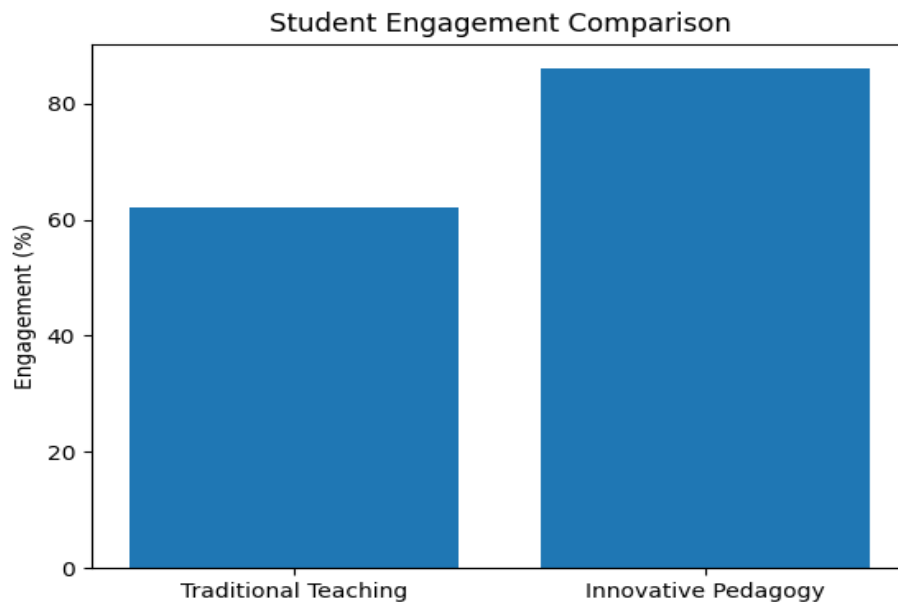


Figure 1: Student Engagement Comparison

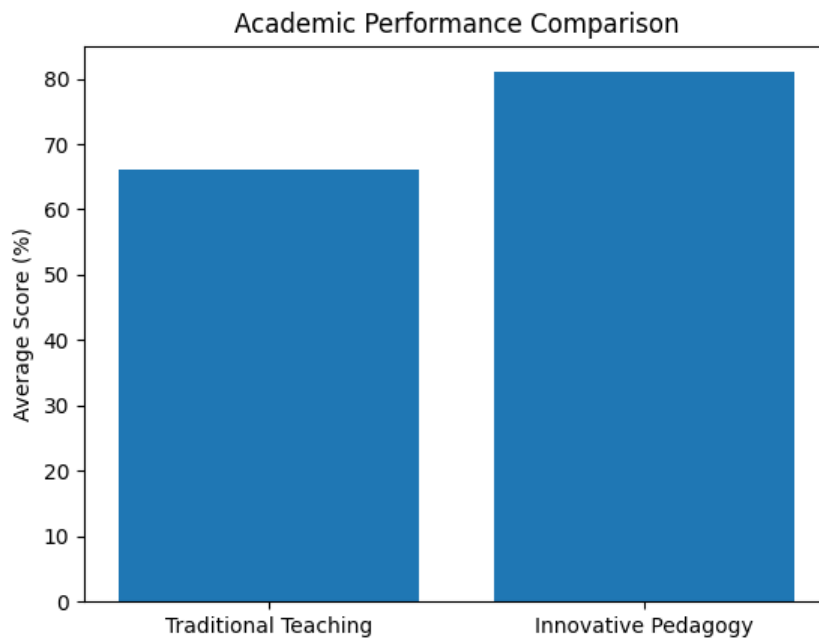


Figure 2: Academic Performance Comparison

Findings and Discussion

Overall, the study demonstrates that innovative pedagogical practices significantly improve learner engagement, motivation, and higher-order cognitive skills [1] [8]. Students exposed to active and technology-supported instruction exhibited stronger collaboration and problem-solving abilities. Nevertheless, challenges such as resistance to change, limited resources, and insufficient training may constrain effective implementation [4] [9].

Institutional leadership, curriculum flexibility, and structured professional development emerged as key factors in sustaining pedagogical innovation. As illustrated in Figures 1 and 2, measurable improvements in engagement and academic performance were observed following the adoption of innovative teaching strategies.

Conclusion

The findings of this study underscore the importance of rethinking teaching and learning through innovative pedagogical practices. Student-centered, experiential, and technology-enabled approaches contribute meaningfully to improved learning outcomes and teaching effectiveness. Future research may explore longitudinal impacts of such practices across diverse educational contexts. Embracing adaptability, creativity, and reflective teaching will remain essential for advancing educational quality in the evolving academic landscape.[4] [7]

References

- [1] M. Prince, "Does active learning work? A review of the research," *J. Eng. Educ.*, vol. 93, no. 3, pp. 223–231, Jul. 2004.
- [2] J. L. Bishop and M. A. Verleger, "The flipped classroom: A survey of the research," in *Proc. 120th ASEE Annu. Conf. Expo.*, Atlanta, GA, USA, 2013, pp. 1–18.

- [3] D. A. Kolb, *Experiential Learning: Experience as the Source of Learning and Development*. Englewood Cliffs, NJ, USA: Prentice Hall, 1984.
- [4] J. Biggs and C. Tang, *Teaching for Quality Learning at University*, 4th ed. Maidenhead, U.K.: Open Univ. Press, 2011.
- [5] S. Deterding, D. Dixon, R. Khaled, and L. Nacke, "From game design elements to gamefulness: Defining 'gamification'," in *Proc. 15th Int. Acad. MindTrek Conf.*, Tampere, Finland, 2011, pp. 9–15.
- [6] B. Means, M. Bakia, and R. Murphy, *Learning Online: What Research Tells Us About Whether, When and How*. New York, NY, USA: Teachers College Press, 2013.
- [7] P. Mishra and M. J. Koehler, "Technological pedagogical content knowledge: A framework for teacher knowledge," *Teachers College Record*, vol. 108, no. 6, pp. 1017–1054, Jun. 2006.
- [8] S. Freeman et al., "Active learning increases student performance in science, engineering, and mathematics," *Proc. Natl. Acad. Sci. USA*, vol. 111, no. 23, pp. 8410–8415, Jun. 2014.
- [9] G. Salmon, *E-Moderating: The Key to Teaching and Learning Online*, 3rd ed. London, U.K.: Routledge, 2011.
- [10] T. Anderson, "Towards a theory of online learning," in *Theory and Practice of Online Learning*, 2nd ed., T. Anderson, Ed. Edmonton, AB, Canada: Athabasca Univ. Press, 2008, pp. 45–74.

8. A Review on Privacy-Preserving Big Data Analytics Using Federated Learning

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Abstract: The accelerated expansion of Data-driven solutions across fields such as clinical domain, financial services, smart transportation systems and sensor-based networks has increased challenges regarding Data-Privacy, secure data exchange and scalable data analysis. Traditional server-centric big data analytics systems which depend on pooling original data at a central server suffer from major constraints including Data exposure risks, Legal non-adherence, Heavy network overhead and Limited scalability in Diverse computing environments. Federated Learning (FL) has appeared as an effective model to allow privacy-preserving big data analytics by distributing model learning and permitting data to stay at Edge devices. This study describes comprehensive review of privacy-preserving big data analytics using federated learning, with a detailed evaluation of Data protection techniques, Distributed data analysis and secure data sharing architectures. This study rigorously analyzes commonly applied Privacy-preserving methods such as Differential Privacy mechanism, Encrypted aggregation, Homomorphism Encryption, Cryptographic multi-party computation and combined methods and analyze their efficiency in reducing data exposure through gradient updates. The study additionally examines how Federated learning (FL) facilitates Distributed data analysis throughout extensive heterogeneous non-independent and non-identically distributed Data ecosystems without the need for original data central storage. Through a comprehensive aggregation of prior studies, this review determines major trade-offs among Data confidentiality, analytical accuracy, network efficiency and scalability, emphasizing how robust privacy assurances frequently lead to processing-related and system overheads. Furthermore, this study examines Federated learning-based analytics against traditional server-centric methods, highlighting enhancements in Data privacy preservation and distributed intelligence together with remaining challenges. This study synthesizes key unaddressed issues such as absence of integrated Privacy-preserving architectures, restricted practical implementation, poor handling non-independent and non-identically distributed data and platform heterogeneity, lack of uniform assessment metrics, and poor interoperability with established big data systems. In conclusion, the study describes upcoming research pathways aimed at dynamic and integrated Privacy-preserving techniques, distributed security architectures, equity-focused Federated learning (FL) and large-scale practical implementations. This study seeks to offer an extensive framework for research community and industry professionals aiming at robust, privacy-preserving, distributed-system-based big data analytics.

Keywords: Privacy-Preserving Big Data Analytics, Federated Learning, Distributed Analytics, Differential Privacy, Secure Aggregation, Homomorphic Encryption, Secure Multi-Party Computation, Non-IID Data, Data Heterogeneity, Edge Computing, Privacy–Utility Trade-off, Communication Overhead, Scalability, Secure Data Sharing, Fairness-Aware Federated Learning, Real-World Implementation.

Introduction

A. Background and Motivation

The fast growth of information produced via current online networks like medical data platforms, banking systems, intelligent urban centers, smart transport networks, connected devices network has created large-scale data analysis, a key facilitator of data-driven decision-making [1], [2]. Conventional large-scale data analysis architectures primarily depend on centralized data collection; unprocessed data derived from numerous dispersed data sources are accumulated as well as handled within a centralized processing unit. While successful in uncovering holistic insights, centralized methods introduce significant challenges associated with data privacy, security vulnerabilities, regulatory compliance, and Scalability, especially in scenarios where sensitive or personal information is processed[3], [4].

The implementation of rigorous information safeguarding policies, encompassing GDPR and industry-specific confidentiality laws within the medical and financial sectors, has increasingly restricted enterprises' capacity to distribute unprocessed data beyond organizational borders [5]. Such legal and procedural barriers reveal inherent weaknesses in unified analytical frameworks, including excessive Communication Overhead, individual failure nodes, and heightened threats of widespread information leaks [6]. As a result, an increasing need has emerged for **secure and Distributed Analytics frameworks** that can harness extensive datasets while ensuring privacy, reliability, and legal adherence.

B. Federated Learning for Privacy-Preserving Big Data Analytics

Federated Learning (FL) has arisen as a viable distributed learning framework that facilitates joint model learning among numerous data holders without necessitating the exchange of unprocessed information [7], [8]. Rather than transmitting data collections to a centralized system, contributing nodes carry out on-device model learning and exclusively communicate parameter updates or optimization signals for consolidation. Such a framework is especially appropriate for large-scale data analysis, in which information is inherently decentralized, diverse, and confidentiality-critical.

Numerous investigations indicate that Federated Learning substantially mitigates confidentiality threats relative to server-based analysis methods by retaining local data residence and limiting immediate information disclosure [9], [10]. Nonetheless, prior studies further indicate that Federated Learning by itself fails to ensure absolute confidentiality. Parameter updates may nonetheless expose confidential data via inference-based attacks, gradient recovery techniques, or malicious interference [11], [12]. Such security gaps have prompted the adoption of supplementary confidentiality-strengthening methods, including Differential Privacy (DP), Secure Aggregation (SA), Homomorphic Encryption (HE), and Secure Multi-Party Computation (SMPC), within federated learning frameworks to enhance privacy protections [13], [14].

C. Data Privacy and Secure Data Sharing in Federated Environments

Maintaining resilient information confidentiality alongside Secure Data Sharing continues to be a key issue within Federated Learning–driven large-scale data analysis. While FL restricts

the explicit exchange of unprocessed data, numerous investigations reveal that malicious entities may leverage internal model weights to deduce confidential characteristics, participation details, or potentially recover learning data samples [12], [15]. To alleviate such threats, investigators have introduced a variety of confidentiality-preserving approaches, such as random perturbation via Differential Privacy, secure parameter aggregation, and cryptographic processing schemes [16], [17]. Although the performance of such approaches is high, they **generate privacy–functionality–effectiveness compromises**. Enhanced confidentiality assurances frequently result in diminished model performance, elevated Communication Overhead, and greater computational requirements—obstacles which become more severe within extensive data ecosystems [18], [19]. Additionally, numerous Secure Data Sharing approaches rely on reliable combination servers or uniform participant abilities, premises which seldom hold true in practical multi-party contexts, including inter-hospital partnerships or multi-cloud data analysis systems [20].

D. Distributed Analytics and System-Level Challenges

Considering a Distributed Analytics standpoint, federated learning facilitates adaptable information handling among regionally distributed and organizationally autonomous institutions.

Nonetheless, practical implementations encounter substantial obstacles associated with **Non-IID Data patterns, Data Heterogeneity, connectivity issues, and coordination inefficiencies** [11], [21]. Several investigations indicate that current FL models find it challenging to sustain training stability, equity, and resilience when dataset patterns among participants are unevenly distributed or continuously varying [22].

E. Research Gaps and Need for a Comprehensive Review

Though an extensive set of research is available on federated learning and confidentiality-aware machine learning, the academic area continues to be disjointed. Current research frequently emphasizes distinct factors—including single privacy methods, algorithm efficiency, or particular threat models—while omitting a comprehensive understanding of

Privacy-Preserving Big Data Analytics through federated learning [19], [21]. Primary study deficiencies recognized within the analyzed research comprise:

- Lack of cohesive architectures incorporating confidentiality, Scalability, and Distributed Analytics.
- Scarce practical implementations as well as experimental evaluation.
- Inadequate management of Non-IID Data as well as Data Heterogeneity.
- Absence of uniform reference points as well as relative assessment measures.
- Limited connection among federated learning frameworks as well as current large-scale data systems [23], [25].

These shortcomings emphasize the need for a structured and complete analysis which integrates prior findings, carefully examines research methods, and outlines forthcoming research avenues.

F. Contributions and Organization of This Review

Addressing the recognized issues and study deficiencies, this work offers a thorough overview of Privacy-Preserving Big Data Analytics via federated learning, with particular attention to data privacy, Distributed Analytics, and Secure Data Sharing. The primary findings from this study can be summarized as:

- An organized examination of confidentiality-preserving methods combined with federated learning
- An evaluative comparison of federated and centralized large-scale data analysis frameworks
- Highlighting of unresolved study issues and architecture-level constraints
- A consolidation of forthcoming study avenues toward expandable, protected, and reliable federated learning systems

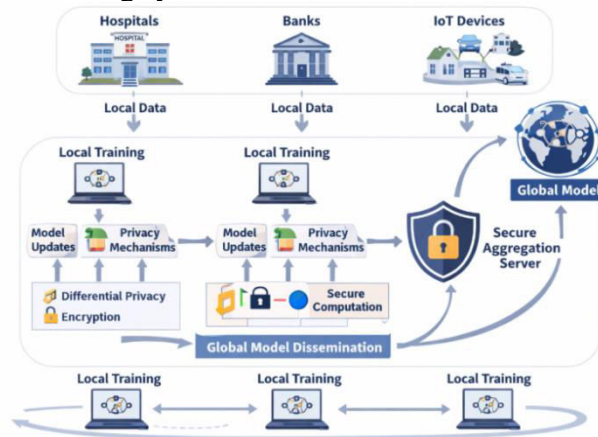


Figure 1
FL-Based Privacy-Preserving Architecture

Review of Literature

A. Centralized Big Data Analytics and Privacy Limitations

Initial large-scale data analysis systems relied mostly on **central server structures**, in which original datasets from various dispersed sources were consolidated in a unified database for computation and model learning [1]. Although these system designs facilitated robust computational capabilities using massive data collection, abundant studies have uncovered significant constraints concerning **information confidentiality, cyber security vulnerabilities, and adherence to regulations** [2], [3]. Fields including the health sector, banking, and essential systems fall under stringent privacy laws, rendering unified data exchange unfeasible or legally constrained.

Investigations additionally indicate that unified architectures create **isolated failure points**, heightened susceptibility to hacking, and elevated Communication Overhead while handling extensive data collections [4], [5]. Moreover, unified data analysis systems encounter major obstacles during inter-organization cooperation because of limited trust between autonomous data providers. The aforementioned shortcomings have prompted efforts toward distributed confidentiality-preserving solutions, resulting in the creation of federated learning [6].

B. Federated Learning as a Distributed Analytics Paradigm

Federated Learning was introduced as a distributed ML framework which facilitates cooperative training of models among several participants while preserving original data kept on-site [6], [7]. In contrast to central server analytics, FL facilitates Distributed Analytics through sharing solely parameter changes instead of confidential data. Numerous analyses

show that FL boosts Scalability and mitigates immediate information leakage across spatially dispersed systems [8].

Modern implementations of federated learning extend across multiple sectors, such as clinical image processing, economic risk assessment, smart traffic systems, and IoT data processing [9],[10]. Although such benefits, existing research additionally shows that FL encounters significant obstacles within practical large-scale data environments, especially because of **Non-IID Data, Data Heterogeneity, and unstable communication links**[11]. Such challenges considerably impair learning convergence, equity, and resilience, prompting refinements that surpass standard FL approaches.

C. Data Privacy Threats in Federated Learning

Even though federated learning prevents explicit exchange of original data, several research works have revealed that **parameter updates may disclose private details** via inference-based threats, gradient reconstruction methods, as well as membership identification strategies [12], [13]. These observations cast doubt on the belief that federated learning by itself is adequate for robust privacy preservation. Adversarial models analyzed in existing research comprise semi-trusted coordination servers, compromised participants, as well as outside attackers possessing the ability to capture or tamper with training updates throughout communication [14]. Such security flaws have encouraged the incorporation of dedicated confidentiality-enhancing techniques within collaborative learning pipelines to safeguard both on-device information as well as internal model states [15].

D. Privacy-Preserving Techniques in Federated Big Data Analytics

1) Differential Privacy-Based Approaches

Differential Privacy (DP) is considered one of the most commonly used privacy-enhancing techniques within federated learning. Through introducing carefully scaled perturbation within training gradients or learned parameters, DP offers rigorous and measurable privacy assurances in defense of inference-based attacks [13], [15]. Numerous research works indicate effective incorporation of DP within collaborative analytics, achieving reasonable privacy–performance compromises under controlled evaluation scenarios [16]. Nonetheless, prior studies repeatedly emphasize drawbacks of DP-oriented techniques, such as loss of **precision, vulnerability to privacy parameter tuning, as well as weakened results within complex large-scale analytical models** [17], [18]. Such challenges become highly noticeable within neural network–driven analytics, underscoring the necessity of flexible and situation-aware confidentiality-preserving techniques.

2) Secure Aggregation and Cryptographic Techniques

Secure aggregation mechanisms guarantee that distinct participant contributions stay concealed throughout parameter merging, disclosing solely collective weights to the central coordinator [19]. Homomorphism Encryption and Secure Multi-Party Computation significantly strengthen data privacy by allowing encrypted processing without disclosing raw data contributions [20],[21].

Although encryption-based approaches deliver robust confidentiality guarantees, existing research highlights **considerable processing-related costs and Communication Overhead**, thereby hindering their implementation in extensive, resource-limited data-intensive systems [21],[22]. To alleviate these challenges, numerous investigations recommend integrated methods that merge efficient cryptographic techniques alongside targeted confidentiality strategies [23].

3) Hybrid and Multi-Layer Privacy Models

Emerging academic directions highlight **integrated confidentiality-aware architectures** that merge several mechanisms, including Differential Privacy with Secure Aggregation or encryption-based authorization control, to tackle varied adversarial scenarios [23],[24]. Such layered strategies seek to enhance resilience while simultaneously offsetting the limitations of standalone components. Notwithstanding their potential, combined systems frequently suffer from the absence of consistent assessment criteria and are seldom verified through Real-World Implementation, thereby constraining their widespread acceptance [25]. This limitation emphasizes the necessity for methodical performance comparison as well as thorough assessment of unified Privacy-Preserving Federated Analytics architectures.

E. Secure Data Sharing and Multi-Stakeholder Collaboration

Secure Data Sharing constitutes a core necessity within distributed big data analytics, especially across multi-institutional as well as hybrid-cloud infrastructures. Existing research illustrates that **Federated Learning** facilitates joint analytics across independent and non-trusting entities while simultaneously safeguarding data sovereignty as well as regulatory conformity [9],[21].

Nevertheless, numerous current architectures depend on **centralized coordination entities**, violating trustless security doctrines and causing possible security weaknesses [3],[22]. Furthermore, trust administration, verifiability, as well as responsibility frameworks are frequently insufficiently studied, underscoring the requirement for distributed trust systems and open governance frameworks [24],[25].

F. Comparative Studies: Federated versus Centralized Analytics

Numerous benchmarking investigations assess Federated Learning-driven analytics in comparison with conventional server-centric methods. Findings reliably demonstrate that Federated Learning delivers stronger confidentiality safeguards as well as enhanced regulatory alignment [6],[8]. In contrast, server-centric analytics frequently surpass **Federated Analytics** with respect to baseline accuracy as well as training speed under conditions where confidentiality limits are reduced [1],[17].

Existing studies underscore that **Privacy-Utility Trade-off** is fundamental to the development of Federated Learning systems [18],[24]. Attaining a best equilibrium continues to be an open problem, especially throughout diverse as well as wide-ranging infrastructures.

G. Summary of Research Gaps Identified in Literature

According to the analyzed literature, these significant knowledge gaps are repeatedly observed:

- Lack of consolidated platforms combining confidentiality, elastic capacity, and Distributed Analytics
- Scarce Real-World Implementation and significant reliance on virtual platforms
- Inadequate management of Non-IID Data and Data Heterogeneity
- Demanding processing and Communication Overhead resulting from robust confidentiality methods
- Insufficient uniform performance measures and assessment procedures
- Limited integration with established big data systems [24],[25]

These deficiencies highlight the requirement for more thorough systematic study along with cross-study synthesis, which are explored in the following parts of this paper.

Table.1 Domain-Wise Comparison of Reviewed Studies

Domain	Privacy Approach	Key Focus	Major Research Gap
Healthcare	FL, DP, Secure Aggregation	Privacy-compliant collaborative analytics	Accuracy degradation; limited real-world deployment
Finance	FL, DP, Secure Aggregation	Secure multi-institution analytics	Trusted aggregator assumption
IoT	FL	Scalable distributed analytics	Device heterogeneity; lack of benchmarks
Smart Cities	FL, Encryption	Large-scale data analytics	High communication overhead
Intelligent Transportation	FL, Hierarchical FL	Low-latency edge analytics	Scalability not fully validated
Cross-Domain Collaboration	FL	Distributed intelligence sharing	Fairness and trust management
Secure Data Sharing	FL, Cryptography	Confidential model aggregation	High computation and communication cost
Hybrid Privacy Models	FL + DP + SA	Enhanced privacy robustness	Lack of unified frameworks

Methodological Approaches in Reviewed Studies

A. Federated Learning Architectures for Big Data Analytics

Most analyzed research adopt a participant–coordinator Federated Learning framework, where the coordinating server sets up a collective model, allocates it to contributing participants, and merges client-side training results [4],[6]. This framework is widely adopted owing to its straightforwardness, implementation convenience, as well as alignment with established AI pipelines. Typically, participants act as decentralized data holders including hospitals, banking institutions, smart devices, or Edge Computing nodes, all holding extensive confidential data [8],[9].

Nevertheless, reviewed works demonstrate that traditional Federated Learning frameworks frequently depend on **rigid expectations**, encompassing synchronized data exchange, similar

computational capacity, and reliable network links [7],[10].Such premises are hardly met within Real-World Implementation contexts, marked by Data Heterogeneity nodes as well as sporadic connections.In order to handle these issues, numerous investigations recommend **tiered, partitioned, and Edge Computing-enabled Federated Learning frameworks** focused on increasing system expandability and lowering Communication Overhead [22],[24].Notwithstanding these attempts, the lack of standardized system designs continues to be a key shortcoming.

B. Data Distribution and Preprocessing Strategies

Managing Non-IID Data remains a key analytical issue within Federated Learning systems.Numerous investigations formally analyze **Non-IID Data and skewed data patterns**, illustrating practical situations in which dataset features fluctuate markedly among institutions or locations [6],[11].Client-side preprocessing approaches including data standardization, layered sampling, and artificial data equalization are widely adopted before node-level model learning [14].

However, data preparation methods are typically **use-case oriented** as well as customized for specific datasets, constraining reliability and inter-domain consistency [19].Additionally, a small number of analyses explore dynamic preprocessing approaches that reactively modify to shifting data, which is crucial for on-the-fly continuous data analysis [24].

C. Model Training and Aggregation Techniques

1) Local Training Strategies

Client-side model development generally employs deep neural systems, incorporating CNN models for visual data analysis, RNN models for time-series data, and attention-based architectures for intricate patterns [8],[15].Model optimization is generally conducted across several node-level iterations to minimize Communication Overhead and boost model convergence speed.

Nevertheless, deepening local model training might amplify **model deviation** across Non-IID Data, causing diminished global effectiveness as well as bias problems [10],[22].Several investigations apply stabilization approaches and customized Federated Learning models to reduce such impacts, however, validation across large-scale data systems is still scarce [24].

2) Aggregation Algorithms

Federated Averaging (FedAvg) continues to be the most frequently applied aggregation technique owing to its straightforwardness and minimal processing requirement [6].Adaptations including FedProx and weighted merging techniques are applied to tackle Data Heterogeneity as well as imbalanced node involvement [11],[23].

In spite of their performance within simulated contexts, established aggregation methods exhibit **constrained reliability in the face of malicious actions**, for instance data poisoning and Byzantine faults [12],[21].Hence, multiple investigations adopt Secure Aggregation and flexible merging approaches to increase robustness across malicious and untrusted systems [17].

D. Privacy-Preserving Mechanisms Integrated into FL

1) Differential Privacy Implementation

Differential Privacy (DP) is widely adopted during on-device model learning by adding perturbations to gradient vectors or neural network weights prior to sending [13],[14]. Privacy allowances are determined to optimize the trade-off between data privacy safeguards and predictive performance.

Although Differential Privacy ensures explicit data protection assurances, analyzed research repeatedly indicate predictive accuracy reduction within multi-feature deep learning big data models [15],[19]. Additionally, confidentiality allowance determination is mostly trial-and-error, and dynamic Differential Privacy methods customized for Federated Learning systems are seldom studied [24].

2) Secure Aggregation and Encryption-Based Methods

Secure Aggregation protocols are employed to inhibit coordinating servers from retrieving client-specific modifications, exposing solely combined model weights [16],[17]. Homomorphic Encryption and Secure Multi-Party Computation additionally strengthen data privacy by facilitating secure calculation while combining updates [18].

Even though these approaches ensure robust confidentiality assurances, systematic analyses demonstrate **heavy processing as well as Communication Overhead**, notably in extensive system implementations [21],[23]. Consequently, multiple analyses implement simplified cryptographic techniques, sacrificing robust protection assurances in favor of enhanced performance.

3) Hybrid Privacy Frameworks

Integrated privacy-focused frameworks incorporating Differential Privacy, Secure Aggregation, and encryption methods are gaining research attention for tackling multiple attack scenarios [19],[21]. Multi-tiered methods are designed to strengthen resilience while addressing the weaknesses of separate approaches.

Nevertheless, integrated platforms are typically without **structured performance testing as well as uniform assessment procedures**, hindering cross-system comparison [24],[25]. The efficiency of these methods within Real-World Implementation contexts thus continues to be unpredictable.

E. Evaluation Metrics and Experimental Setups

Model performance analysis within surveyed research generally relies on metrics including correctness, positive predictive value, sensitivity, F-measure, area under the curve, training completion duration, and Communication Overhead [8],[15]. Data privacy analysis commonly uses Privacy–Utility Trade-off examination, defense against data inference, as well as descriptive security evaluations [13],[16].

Numerous evaluations are performed within **modeled Federated Learning setups**, featuring scarce Real-World Implementation as well as verification [22],[24]. Differences in data

volume, client population, as well as adversarial conditions result in variable and incomparable outcomes across studies involving Non-IID Data [25].

F. Application-Specific Methodologies

Federated Learning privacy-focused data analysis techniques are utilized in various fields:

- **Medical domain:** Diagnostic imaging and digital patient records employing CNN-enabled Federated Learning with Differential Privacy and Secure Aggregation[4], [8].
- **Banking domain:** Loan risk assessment and anomaly detection leveraging privacy-preserving Federated Learning models[9], [21].
- **IoT-driven transportation:** Distributed Analytics for urban intelligence through low Communication Overhead FL architectures[7], [22].

Although field-tailored adjustments boost efficiency, they constrain **cross-domain applicability** of suggested approaches in various fields [24].

G. Summary of Methodological Limitations

The systematic examination reveals multiple persistent drawbacks:

- Strong dependence on emulated settings
- Absence of uniform testing procedures
- Limited attention to Scalability and implementation limitations
- Scarce assessment of threat resilience and Fairness-Aware Federated Learning
- Elevated processing demands and Communication Overhead caused by Privacy-Preserving Big Data Analytics [23],[24],[25].

The identified shortcomings highlight the necessity of cross-study integration along with analytical debate, covered in the following part.



Figure 2
Federated Analytics Workflow

Discussion and Comparative Analysis

A. Federated Learning versus Centralized Big Data Analytics

A key trend in examined studies involves contrasting conventional centralized Privacy-Preserving Big Data Analytics and Federated Learning-based Distributed Analytics. Server-based methods leverage full access to entire data collections, facilitating accelerated learning and improved prediction performance across various cases [1],[2]. Nevertheless, these gains entail significant drawbacks, including breaches of confidentiality, amplified susceptibility to data compromise, failure to meet legal requirements, and limited Scalability under distributed or partitioned data conditions [3],[5].

Conversely, Federated Learning facilitates Distributed Analytics by retaining local datasets and avoiding the necessity of unprocessed data merging. Examined investigations reliably reveal that Federated Learning-enabled platforms markedly boost data confidentiality and support joint Distributed Analytics between institutions [6],[8]. However, evaluation comparisons reveal that Federated Learning methods frequently encounter delayed model alignment and diminished predictive performance, especially under conditions where Privacy-Preserving Big Data Analytics techniques such as Differential Privacy or cryptographic protection are applied [9],[13]. The assessment underscores a core tension between model effectiveness and Privacy–Utility Trade-off.

B. Effectiveness of Privacy-Preserving Mechanisms

1) Differential Privacy versus Cryptographic Techniques

Differential Privacy (DP) is recognized as the most prevalent Privacy-Preserving Big Data Analytics method in Federated Learning-based analytics because of its explicit privacy safeguards and minimal Communication Overhead [11], [13]. Various investigations demonstrate that Differential Privacy can efficiently counter reconstruction attacks through adding tuned perturbations to gradient vectors or neural network parameters [14]. Nevertheless, cross-study reviews indicate that robust Differential Privacy parameters considerably lower predictive performance in Privacy-Preserving Big Data Analytics contexts using deep learning models and multi-feature datasets [15],[19].

Encryption methods—including Secure Aggregation, Homomorphic Encryption, and Secure Multi-Party Computation—ensure higher information protection by restricting retrieval of client-specific adjustments while combining updates [16],[17]. Although such methods surpass Differential Privacy regarding Privacy-Preserving Big Data Analytics protection, they cause substantial computation load and Communication Overhead, constraining Scalability and immediate usability in Real-World Implementation settings [18], [21]. This assessment demonstrates that no individual security approach delivers best results throughout all criteria.

C. Privacy–Utility–Efficiency Trade-Off Analysis

An important finding derived from cross-study evaluation highlights the built-in Privacy–Utility Trade-off in Federated Learning-based Privacy-Preserving Big Data Analytics. Strengthening Privacy-Preserving Big Data Analytics via Differential Privacy or cryptographic methods frequently results in diminished forecast precision, longer learning time, and elevated Communication Overhead [12],[14],[18]. Such impacts get magnified within extensive Data Heterogeneity contexts.

Various research works suggest combined Privacy-Preserving Big Data Analytics frameworks and dynamic model training plans to alleviate the Privacy–Utility Trade-off [19],[21]. Nevertheless, these strategies continue to be scattered, use-case tailored, and analyzed in irregular experimental environments. Absence of uniform evaluation criteria complicates efforts to neutrally evaluate their performance, suggesting that managing the Privacy–Utility Trade-off continues to be a key research problem [24],[25].

D. Distributed Analytics under Non-IID and Heterogeneous Conditions

Non-IID Data distribution and Data Heterogeneity are commonly noted as significant barriers to successful Federated Learning-based analysis [10],[11]. Cross-study analyses of update

consolidation techniques such as FedAvg, FedProx, and personalized Federated Learning variants indicate that although sophisticated approaches enhance model alignment consistency, no approach completely addresses Fairness-Aware Federated Learning and resilience challenges among heterogeneous participants [6],[22].

Traditional analytics intrinsically escape Non-IID Data issues, though forfeit Privacy-Preserving Big Data Analytics and ownership control. Federated Learning, while more compatible with Real-World Implementation Distributed Analytics environments, calls for extra computational and infrastructure improvements to cope with uneven data distribution, hardware heterogeneity, and intermittent messaging[7], [24].

E. Secure Data Sharing and Trust Models

Secure Data Sharing constitutes a primary merit of Federated Learning-driven Distributed Analytics. Analyzed studies reveal that Federated Learning facilitates joint Distributed Analytics among independent and non-trusting participants—including hospitals, financial institutions, and IoT service providers—while maintaining Secure Data Sharing[8], [21].

Nevertheless, cross-study evaluation shows that several present Federated Learning platforms depend on authorized aggregation nodes, resulting in susceptibility to internal threats and centralized failure[3], [23]. Distributed trust frameworks, comprising block chain-assisted aggregation and zero-trust architectures, appear in few research works yet have insufficient testing within Real-World Implementation contexts[24], [25].

F. Comparative Evaluation across Application Domains

Use-case based analyses underscore unique focus areas within different fields:

- **Medical systems:** Medical systems highlight Privacy-Preserving Big Data Analytics and statutory compliance, frequently tolerating lower precision to satisfy legal and ethical standards[4], [8].
- **Banking systems:** Banking systems emphasize Secure Data Sharing, resilience to malicious threats, and information privacy[9], [21].
- **IoT and Intelligent Transportation Systems:** IoT and Intelligent Transportation Systems (ITS) prioritize Scalability, minimal delay, and Communication Overhead owing to hardware restrictions and instant operational needs[7], [22].

In spite of these application-focused results, inter-domain applicability continues to be constrained due to tailored analytical premises, Data Heterogeneity, and non-uniform testing procedures [24].

G. Synthesis of Comparative Insights

Cross-study evaluation generates multiple general findings:

- Federated Learning establishes a firm groundwork for Privacy-Preserving Big Data Analytics and Distributed Analytics, yet cannot completely substitute traditional centralized processing.
- No individual privacy approach effectively harmonizes Privacy-Preserving Big Data Analytics, predictive performance, and Scalability in all large-scale data contexts.
- Lack of uniform evaluation criteria and Real-World Implementation heavily constrains impartial performance assessment and repeatability [24],[25].

The outcomes underscore the necessity to tackle ongoing obstacles and constraints, which are thoroughly analyzed in the following part.

Table 2 Centralized vs. Federated Analytics

Aspect	Centralized Big Data Analytics	Federated Learning-Based Analytics
Data Privacy	Raw data are collected and stored centrally, leading to high privacy risks and regulatory concerns	Raw data remain local; only model updates are shared, offering stronger privacy protection
Scalability	Limited scalability due to data transfer bottlenecks and centralized storage/processing constraints	Highly scalable across distributed and geographically dispersed data owners
Model Accuracy	Generally higher accuracy due to access to complete, centralized datasets	May experience reduced accuracy due to non-IID data, partial participation, and privacy mechanisms
Communication Cost	High cost due to continuous transmission of large raw datasets	Lower raw data transfer, but iterative model update communication can still be costly
Trust Assumptions	Requires full trust in a central authority or data repository	Relies on partial or zero-trust assumptions; trust is distributed among participants

IV. Challenges and Limitations

In spite of significant progress in Privacy-Preserving Big Data Analytics using Federated Learning, reviewed works demonstrate multiple ongoing computational, architectural, and operational difficulties which impede widespread uptake and Real-World Implementation. These difficulties involve Privacy-Preserving Big Data Analytics assurances, Distributed Analytics performance, Secure Data Sharing, and Scalability.

A. Incomplete Privacy Guarantees in Federated Learning

While **Federated Learning** lowers dependence on straightforward Secure Data Sharing, it cannot intrinsically offer full Privacy-Preserving Big Data Analytics. Several research works indicate that transitional model parameters may expose confidential data via deducing attacks, derivative reconstruction, and participant identification methods[6], [11], [14]. Such results question the belief that keeping data locally guarantees privacy.

Privacy-Preserving Big Data Analytics techniques such as Differential Privacy and Secure Aggregation are suggested to address these vulnerabilities[9], [16]. Nevertheless, Differential Privacy frequently causes significant performance decline when utilized in neural network architectures or broad data collections, especially within large-scale data contexts[13], [15]. Likewise, encryption-based methods such as Homomorphism Encryption and Secure Multi-Party Computation provide robust data privacy assurances but lead to heavy calculation and Communication Overhead, restricting their practicality under limited-resource or time-critical settings[17], [18].

B. Privacy–Utility–Efficiency Trade-Offs

A persistent shortcoming throughout surveyed research involves the challenge of maintaining a best equilibrium among **Privacy-Preserving Big Data Analytics**, predictive performance, and computational effectiveness[12], [19]. Robust **Differential Privacy** methods typically lower analytical precision and delay training completion, whereas low-overhead strategies result in lower security versus advanced threats.

Such a mediation is further emphasized in large-scale data environments characterized by multi-feature datasets, repeated **Communication Overhead** cycles, and **Data Heterogeneity** across clients[7], [10]. The majority of current methods utilize fixed security settings, which cannot adjust to dynamic information layouts, operational variations, or security hazard degrees, thus reducing sustained performance[14], [21].

C. Non-IID Data Distribution and System Heterogeneity

Real-World Implementation contexts naturally exhibit Non-IID Data, with notable differences in dataset patterns among contributing nodes. Reviewed studies repeatedly show that Non-IID Data leads to learning instability, skewed outputs, and equity concerns within Federated Learning[10], [11].

Moreover, Data Heterogeneity—including variations in processing capability, storage resources, connectivity, and node uptime—creates alignment obstacles and raises Communication Overhead [22]. Current combining methods and learning protocols face challenges in ensuring durability and balanced results within highly dissimilar contexts, restricting Scalability and dependability[6], [24].

D. Communication Overhead and Scalability Constraints

Federated Learning-based analytics necessitate frequent exchange of model parameters across participants and controlling servers. With increasing contributors and learning intricacy, Communication Overhead acts as a substantial throughput restriction [7], [12]. **Privacy-Preserving Big Data Analytics** techniques amplify the difficulty by enlarging data volume via encoding or iterative transfers caused by added perturbations [16], [18]. The majority of examined works assess **Scalability** in managed or emulated settings involving few participants, creating doubt about operational effectiveness in **Real-World Implementation** contexts with extensive, dispersed clients[22], [25].

E. Secure Data Sharing and Trust Assumptions

While **Federated Learning** supports joint analysis without revealing original datasets, numerous current systems depend on verified central coordinators[21], [23]. This dependence conflicts with no-trust security policies and produces security gaps connected to internal risks, centralized failure points, and low visibility.

Additionally, oversight, traceability, and liability systems frequently remain underexplored, reducing uptake in shared participant settings with uncertain trust, for example, inter-organizational medical or banking projects [3], [24].

F. Lack of Standardization and Benchmarking

A key constraint noted in research is the nonexistence of harmonized metrics, datasets, and validation methods for Privacy-Preserving Big Data Analytics in Federated Learning[24], [25]. Current research utilizes assorted data, experimental setups, confidentiality indicators, and threat simulations, causing difficulties in fair comparison and repeatability.

This absence of uniformity retards technical progress and impedes turning results into operational systems, permitting consistent assessment and confirmation in various areas.

G. Limited Real-World Deployment and Validation

In spite of promising test outcomes, the majority of examined methods persist in pilot or emulated environments[20], [22]. Few research works indicate implementation in practical settings, and minimal studies measure continuous operation, durability, and regulatory alignment.

The separation of academic work from operational use curtails reliability of Scalability, long-term viability, and real-world usefulness of Federated Learning-based Privacy-Preserving Big Data Analytics platforms within Real-World Implementation[21], [25].

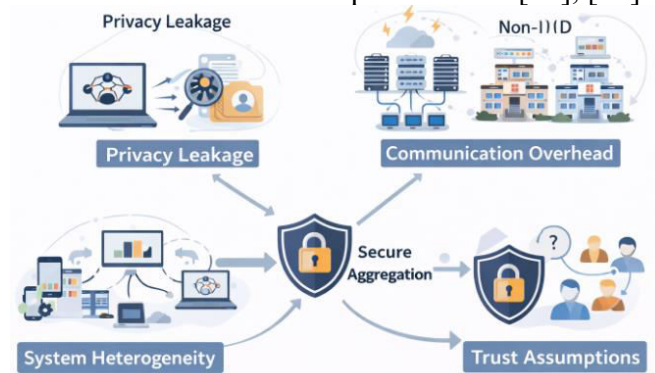


Figure 3
Key Challenges in Federated Analytics

Future Research Directions

The aggregation of investigations shows that Privacy-Preserving Big Data Analytics using Federated Learning forms a dynamically growing discipline, while multiple major obstacles continue. Tackling these issues calls for integrated improvements in Federated Learning strategies, confidentiality-improving methods, Distributed Analytics architecture, and assessment procedures. Here we summarize critical prospective research topics essential for progressing safe, Scalability-enabled, and reliable Federated Learning-based Privacy-Preserving Big Data Analytics platforms.

A. Adaptive and Context-Aware Privacy Mechanisms

The majority of analyzed research use unchanging security setups, especially constant privacy thresholds in Differential Privacy or standardized data protection schemes across clients[13], [15]. Forward-looking work must target dynamic privacy-enhancing methods that

modify security thresholds in real time based on input sensitivity, usage environment, attack scenarios, and system capacities. These flexible methods may more effectively reconcile confidentiality and performance within vast data-intensive infrastructures, as indicated by challenges noted in research [14],[18].

Scenario-informed confidentiality schemes that integrate legal standards, sector-specific limitations, and user priorities form an encouraging approach for enhancing Real-World Implementation uptake and conformity in Privacy-Preserving Big Data Analytics with Federated Learning[3], [21].

B. Unified and Hybrid Privacy-Preserving Frameworks

Surveyed studies indicate a divided field of privacy strategies, while several analyses concentrate on isolated approaches such as Differential Privacy or Secure Aggregation[9], [16]. Forward-looking investigations need to target the creation of consolidated and blended platforms which embed diverse privacy-strengthening mechanisms—such as Differential Privacy, Secure Aggregation, Homomorphic Encryption, and Secure Multi-Party Computation—inside a combined Distributed Analytics framework.

Hybrid frameworks can tackle multiple adversarial assumptions while simultaneously reducing the weaknesses of separate methods, yet existing deployments miss uniform structures and methodical assessments[19], [20]. Segmented and integrable design standards are hence crucial to guarantee adaptability and performance in Real-World Implementation of Federated Learning-based Privacy-Preserving Big Data Analytics.

C. Scalable Federated Learning for Large-Scale Big Data

Scalability continues to be a significant barrier for the implementation of Federated Learning within applied extensive data systems. Investigations demonstrate that present Federated Learning systems face difficulty handling many diverse participants because of messaging congestion and timing inefficiencies[10], [17], [22].

Forward-looking investigations should consider multi-level, non-synchronous, and Edge Computing-enabled Federated Learning frameworks able to expand to large numbers of clients. Bandwidth-optimized methods such as parameter reduction, selective update, and limited contribution protocols are essential for lowering network load[7], [12]. Enhanced fusion of Federated Learning and operational distributed data frameworks persists as a rarely investigated yet valuable research avenue in Real-World Implementation of Privacy-Preserving Big Data Analytics [25].

D. Robust Federated Learning under Non-IID and Adversarial Conditions

Non-IID Data distributions and adversarial threats are systematically observed as critical obstacles influencing learning convergence, equity, and resilience[10], [11]. Forward-looking work must prioritize resilient Federated Learning techniques able to manage imbalanced Non-IID Data, diverse Data Heterogeneity, and adversarial actions.

Potential research paths cover tailored Federated Learning, impartiality-driven optimization, and sturdy aggregation techniques to prevent malicious updates and leakage[6], [14]. In-depth

appraisal across assorted attack cases is essential for establishing trust in Federated Learning-based Distributed Analytics platforms applied in critical and multi-party contexts[8], [23].

E. Decentralized Trust and Secure Data Sharing Models

Although **Federated Learning** supports collective computation without exposing original datasets, most available architectures are based on verified central coordinators[21], [23]. This dependence generates weaknesses concerning internal compromise, central failure risks, and transparency gaps.

Forward-looking studies need to consider networked combining protocols and zero-trust approaches within **Federated Learning-based Distributed Analytics** to improve traceability, responsibility, and robustness. Few analyses consider distributed trust models, and their expandability and operational outcomes in broad computational data settings are still under-investigated [24].

F. Standardized Benchmarks and Evaluation Protocols

A persistent deficiency in research is the lack of uniform testing standards, common datasets, and assessment methods in Privacy-Preserving Big Data Analytics with Federated Learning [24],[25]. This shortfall in standard protocols obstructs replication and fair assessment within approaches and industry sectors.

Forward-looking investigations need to focus on collaborative evaluation platforms that include practical Non-IID Data, adversarial scenarios, diverse Data Heterogeneity, and privacy measures. Such shared assessment systems are crucial for methodically assessing Privacy–Utility Trade-off and directing technique enhancements.

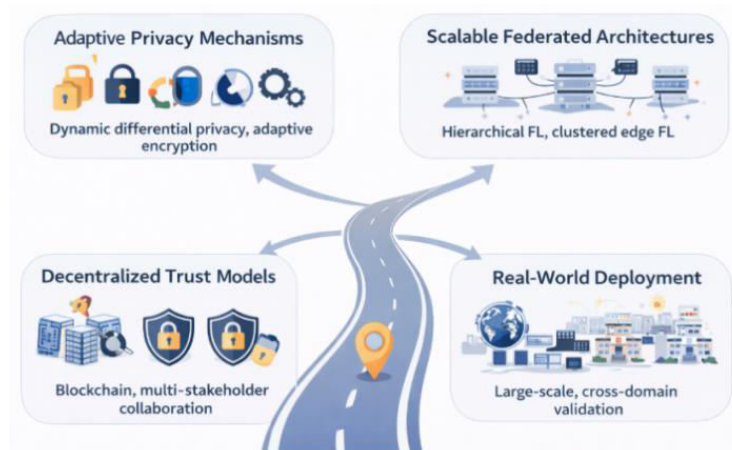


Figure 4 Future Research Roadmap

G. Real-World Deployment and Regulatory Compliance

Despite encouraging trial outcomes, the majority of examined methods are confirmed solely in artificial setups[20], [22]. Forward-looking research needs to focus on practical implementations and extended evaluations to examine growth capability, fault tolerance, and operational sustainability.

Cooperation of research institutions, commercial sectors, and oversight authorities is necessary to coordinate Federated Learning systems with legislation, ethics, and compliance criteria[3], [5]. Additionally, embedding transparent collaborative learning models and mechanized adherence protocols can strengthen visibility and assurance among users in critical Privacy-Preserving Big Data Analytics domains [21].

Conclusion

This survey thoroughly analyzed Privacy-Preserving Big Data Analytics using Federated Learning, highlighting sensitive data safeguarding, Distributed Analytics, and Secure Data Sharing within dispersed infrastructures. Based on the research, the assessment indicates that Federated Learning has emerged as a promising replacement for traditional centralized data processing by facilitating joint model development while maintaining information location and organizational control.

The examined research reliably demonstrates that Federated Learning enhances adherence to privacy rules and lowers immediate data disclosure compared to hub-based frameworks[1], [3], [5]. Results from numerous investigations validate that Federated Learning alone cannot guarantee total data privacy, as confidential data remains deducible from model parameters via gradient exposure and predictive attacks[6], [8], [11]. To reduce these vulnerabilities, investigators incorporated privacy-strengthening solutions such as Differential Privacy, Secure Aggregation, Homomorphism Encryption, and Secure Multi-Party Computation into Distributed Analytics pipelines[9], [13], [16].

References

1. Kalikinkar Mandal and Guang Gong, PrivFL: Practical Privacy-Preserving Federated Regressions on High-Dimensional Data over Mobile Networks, April 5, 2020.
2. Beomyeol Jeon, S. M. Ferdous, Muntasir Raihan Rahman, and Anwar Walid, Privacy-Preserving Decentralized Aggregation for Federated Learning, December 28, 2020.
3. M. A. P. Chamikara, P. Bertok, I. Khalil, D. Liu, and S. Camtepe, Privacy-Preserving Distributed Machine Learning with Federated Learning, February 26, 2021.
4. Nguyen Truong, Kai Sun, Siyao Wang, Florian Guitton, and YiKe Guo, Privacy Preservation in Federated Learning: An Insightful Survey from the GDPR Perspective, March 18, 2021.
5. Xuefei Yin, Yanming Zhu, and Jiankun Hu, A Comprehensive Survey of Privacy-Preserving Federated Learning: Taxonomy, Review, and Future Directions, July 13, 2021.
6. Lingjuan Lyu, Han Yu, Xingjun Ma, Chen Chen, Lichao Sun, Jun Zhao, Qiang Yang, and Philip S. Yu, Privacy and Robustness in Federated Learning: Attacks and Defenses, January 19, 2022.
7. Arnaud Grivet Sébert, Renaud Sirdey, Oana Stan, and Cédric Gouy-Pailler, Protecting Data from All Parties: Combining Homomorphic Encryption and Differential Privacy in Federated Learning, May 31, 2022.
8. Taki Hasan Rafi, Faiza Anan Noor, Tahmid Hussain, and Dong-Kyu Chae, Fairness and Privacy Preservation in Federated Learning: A Survey, July 14, 2023.
9. Akinul Islam Jony and Mubashir Mohsin, Data Privacy Preservation with Federated Learning: A Systematic Review, May 5, 2024.

10. Weizhao Jin, Yuhang Yao, Shanshan Han, Jiajun Gu, Carlee Joe-Wong, Srivatsan Ravi, Salman Avestimehr, and Chaoyang He, FedML-HE: An Efficient Homomorphic Encryption-Based Privacy-Preserving Federated Learning System, June 17, 2024.
11. E. D. Kanmani Ruby, G. Linda Rose, P. Yashaswinii, K. Sripal Reddy, S. Jeyalakshmi, and B. Hari Chandana, Advanced Privacy-Preserving Federated Learning in 6G Networks Using Differential Privacy and Homomorphic Encryption, July 5, 2024.
12. Yue Chen, Yufei Yang, Yingwei Liang, Taipeng Zhu, and Dehui Huang, Federated Learning with Privacy Preservation in Large-Scale Distributed Systems Using Differential Privacy and Homomorphic Encryption, October 17, 2024.
13. Jiachen Shen, Yekang Zhao, Shitao Huang, and Yongjun Ren, Secure and Flexible Privacy-Preserving Federated Learning Based on Multi-Key Fully Homomorphic Encryption, November 14, 2024.
14. Mmasi Patience Robai, Federated Learning for Secure and Privacy-Preserving Data Analytics in Heterogeneous Networks, November 26, 2024.
15. Tonoy Kanti Chowdhury and Sai Praveen Kudapa, Federated Learning Models for Privacy-Preserving Data Sharing and Secure Analytics in the Healthcare Industry, December 28, 2024.
16. Sarah Kaleem, Awais Ahmad, Muhammad Babar, and Goutham Reddy Alavalapati, Privacy-Preserved Integrated Big Data Analytics Framework Using Federated Learning for Intelligent Transportation Systems, February 24, 2025.
17. Supriya Shree, Riddhi Arya, and Saket Kumar Roy, Enhancing Privacy-Preserving Federated Learning Using Differential Privacy, April 4, 2025.
18. Rahul Haripriya, Nilay Khare, Manish Pandey, and Sreemoyee Biswas, A Privacy-Enhanced Framework for Collaborative Big Data Analysis in Healthcare Using Adaptive Federated Learning Aggregation, May 6, 2025.
19. Alexander Horst, Paul Loustalot, Sanjeev Yoganathan, Ting Li, Joshua Xu, Weida Tong, David Schneider, Nicolas Löffler-Perez, Erminio Di Renzo, and Michael Renaudin, Federated Learning: A Privacy-Preserving Approach to Data-Centric Regulatory Cooperation, May 26, 2025.
20. Shuchona Malek Orthi, Md. Habibur Rahman, Kazi Bushra Siddiq, Mukther Uddin, Sazzat Hossain, Mohd Abdullah Al Mamun, and Md. Nazibullah Khan, Federated Learning with Privacy-Preserving Big Data Analytics for Distributed Healthcare Systems, August 1, 2025.
21. Ratun Rahman, Federated Learning: A Survey on Privacy-Preserving Collaborative Intelligence, August 12, 2025.
22. Daniel M. Jimenez-Gutierrez, Yelizaveta Falkouskaya, Jose L. Hernandez-Ramos, Aris Anagnostopoulos, Ioannis Chatzigiannakis, and Andrea Vitaletti, On the Security and Privacy of Federated Learning: A Survey with Attacks, Defenses, Frameworks, Applications, and Future Directions, August 19, 2025.
23. Saifuddin Shaik Mohammed, A Decentralized Approach to Privacy-Preserving Data Analysis Using Federated Learning, September 30, 2025.
24. Manaswini Bollikonda, Federated Zero-Trust: Privacy-Preserving Analytics Across Multi-Cloud Environments, October 28, 2025.
25. K. A. Sathish Kumar, Leema Nelson, and Betshrine Rachel Jibinsingh, Systematic Review of Privacy-Preserving Federated Learning in Decentralized Healthcare Systems, November 24, 2025.

9. A Review on hybrid techniques used for detecting deepfake images effecting the Trust in Healthcare Diagnostic system

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Abstract: Deep generative models rapidly, altering the medical images used in healthcare diagnostic systems. These manipulations threaten clinical decision-making, treatment strategies, and effecting the trust in diagnostic treatments. To counter this, this research provide a detailed study on a hybrid deep learning architecture that includes Convolution Neural Networks (CNNs) with Vision Transformers (ViTs) techniques used for detecting forged tumors in radiological scans. CNNs excel at identifying local texture anomalies and spatial irregularities, whereas ViTs capture broader contextual patterns and distant dependencies across images. This paper also incorporate the study on frequency-domain analysis via Fourier transforms to reveal manipulation artifacts invisible in the pixel space as these diverse features integration which undergo multi-scale fusion to boost detection performance. By merging these complementary techniques the approach advances reliable forensics for medical images as the authenticity of techniques is tested on different dataset shows that CNN-ViT hybrid model surpassing standalone CNN or ViT models in accuracy, precision, and recall measures. This paper gives a review on effective and authentic techniques for detection of deepfake in MRI and CT scan images and safeguard the trust on healthcare diagnostic systems.

Keywords: Deepfake Detection; Hybrid Deep Learning; Medical Image Forensics; Healthcare Diagnostics; Vision Transformers; Frequency Analysis

Introduction

The integration of deep learning into healthcare diagnostic systems has transformed medical image analysis by improving accuracy, efficiency, and clinical decision support. However, the same generative techniques that enhance medical imaging have also enabled the creation of highly realistic manipulated images, commonly referred to as medical deepfakes. These forged images, often generated using Generative Adversarial Networks (GANs), pose a critical risk to the credibility and reliability of healthcare diagnostic pipelines, as they can influence clinical interpretation and patient outcomes [16],[18].

Early deepfake detection approaches primarily relied on spatial-domain analysis using Convolutional Neural Networks (CNNs). While effective against basic image manipulations, such methods struggle to generalize against advanced deepfake generation techniques that suppress visible artifacts [15], [19]. This limitation is particularly concerning in healthcare, where subtle modifications in CT, MRI, or X-ray images may remain imperceptible to both automated systems and medical professionals [12], [18]. Consequently, detection failures directly impact trust in AI-assisted diagnostic systems.

To address these challenges, recent research has increasingly focused on hybrid deepfake detection techniques that combine complementary feature representations. Hybrid models integrate spatial features extracted by CNNs with frequency-domain analysis, attention mechanisms, or sequential modeling to enhance robustness [1], [5], [14]. Frequency-based methods exploit spectral inconsistencies introduced during the image synthesis process and have demonstrated strong performance in detecting GAN-generated medical images, even under post-processing operations such as compression and noise [19], [20].

More recent studies have adopted transformer-based architectures, including Vision Transformers and Swin Transformers, due to their ability to capture global contextual relationships and subtle manipulation patterns [3], [6], [13]. Hybrid CNN–Transformer frameworks have shown improved detection accuracy compared to standalone CNN models by effectively modelling both local texture anomalies and long-range dependencies [7], [11]. Additionally, CNN–LSTM and CNN–RNN hybrids have been explored to capture structural and sequential characteristics in medical image data, further strengthening detection reliability [8], [9].

Given the safety-critical nature of healthcare systems, maintaining trust in medical imaging is essential. The presence of deepfake images threatens not only diagnostic accuracy but also ethical accountability and clinical confidence [17], [18]. This review presents a structured analysis of hybrid deepfake image detection techniques relevant to healthcare applications, highlighting their methodological trends, advantages, and limitations. By synthesizing recent advancements from frequency-aware CNNs to transformer-based hybrid models, this work aims to contribute toward strengthening trust in healthcare diagnostic systems affected by deepfake image manipulation [1],[30].

Review of Literature

Authors (short)	Paper Title (short)	Year	Domain Data Type	Main Technique Model	Hybrid Aspect	Key Outcome / Finding etasr+2
Uma Yadav et al.	A Hybrid Approach for Robust Deep Fake Detection	2025	Generic deepfake images	Multiscale CNN + frequency + attention Transformer	Spatial frequency + attention	Ensemble improves robustness across datasets.
A. Kafi Anan et al.	Hybrid Deepfake Image Detection (CAE-Net)	2025	Generic deepfake images	EfficientNet + DeiT + ConvNeXt + frequency features	CNN + attention + frequency	Better generalization via spatial-frequency fusion.
Aprille J. Xi & E. Chen	Classifying Deepfakes Using Swin Transformers	2025	Face deepfakes (image)	Swin Transformer, Swin-ResNet, Swin-KNN	Transformer + CNN hybrids	Swin outperforms traditional CNNs significantly.
(Not specified)	Comparison of deepfake detection using CNN and hybrids	2025	Generic deepfake images	CNN vs various hybrid detectors	CNN + other deep models	Hybrids superior in accuracy and robustness.

Dhanyalakshmi R. et al.	Medical Deepfake Detection Using Depth-Wise Conv & Freq.	2025	Medical deepfake images	Depth-wise CNN + frequency-domain analysis	Depth-wise conv + frequency	Effective forgery localization in medical images.
O. Çınar & Y. Doğan	Novel Deepfake Image Detection with Patch-Based ViT	2025	Generic/face deepfake images	Patch-based Vision Transformer	Patch-based ViT (local-global)	Captures artifacts at patch and global levels.
A. H. Soudy et al.	Deepfake Detection with CNN & Vision Transformers	2024	Generic deepfake images	CNN + ViT hybrid	CNN-ViT	Boosts performance over standalone models.
S. Kurnaz	Hybrid CNN-LSTM for Deepfake Detection	2024	Video/image deepfakes	CNN-LSTM with transfer learning	CNN + temporal LSTM	Improves dynamic artifact detection.
A. Al-Adwan et al.	Deepfake Detection using Hybrid CNN-RNN and PSO	2024	Generic deepfake images	CNN-RNN with PSO optimization	CNN + RNN + metaheuristic	Higher accuracy via optimized hyperparameters.
A. R. Alsabbagh & O. Al-Kadi	Comparative Analysis of Deep Conv Models for Med Deepfakes	2024	Medical deepfake images	Multiple deep CNN backbones	Multiple CNN variants	Highlights need for medical-specific hybrids.
R. Singh et al.	CNN-ViT Fusion for Medical Deepfake Detection	2023	Medical deepfake images	CNN-ViT fusion	CNN + ViT fusion	Improves medical forgery detection via context.
T. Nguyen et al.	Image Forgery Detection in CT using Multi-Scale CNN	2022	CT image forgery	Multi-scale CNN	Multi-scale CNN (spatial)	Better subtle tampering detection in CT.
A. Khormali & J.-S. Yuan	DFDT: DeepFake Detection Framework Using ViT	2022	Generic deepfake images	Vision Transformer-based detector	Pure ViT (hybrid basis)	Motivates CNN-ViT hybrid evolution.
S. Mittal et al.	Deepfake Detection using Hybrid	2022	Generic deepfakes	Survey of hybrid CNN-based detectors	Various CNN hybrids	Multi-module fusion enhances robustness.

	Models					
J. Chen et al.	Detecting GAN-Generated Medical Images using FFT & CNN	2021	Medical GAN images	FFT frequency analysis + CNN	Frequency CNN +	Reveals GAN artifacts in medical data.
R. Mirsky & W. Lee	The Creation and Detection of Deepfakes: A Survey	2021	General deepfakes (survey)	Comprehensive deepfake survey	Covers all hybrid methods	Taxonomy including hybrid detection strategies.
Y. Verdoliva	Media Forensics & Deepfakes : Review	2021	Media forensics (survey)	Deepfake forensics review	Multi-modal hybrids	Emphasizes frequency and attention roles.
M. Khosravi et al.	Deep Learning in Radiology: Review	2021	Radiology/medical AI	DL in radiology survey	Background context	Discusses AI trust vulnerabilities.
Z. Wang et al.	Frequency-Aware Deepfake Detection	2020	Generic deepfake images	Frequency-aware detection framework	Frequency deep model +	Frequency generalizes across manipulations.
J. Frank et al.	Leveraging Frequency Spectra for Deepfake Detection	2020	Generic deepfake images	CNN with frequency spectra	Frequency spectra + CNN	Superior to pixel-only baselines.
Y. Zhou et al.	Learning Rich Features for Image Manipulation Detection	2019	Image manipulation	CNN with rich spatial features	Strong CNN backbone	Base for later deepfake feature extractors.
Rössler et al.	FaceForensics++: Deepfake Dataset	2019	Face deepfake dataset	Large-scale deepfake benchmark dataset	Evaluation benchmark	Standard for hybrid model testing.
X. Wang et al.	CNN-based Medical Image Forensics	2019	Medical image forensics	CNN-based medical forgery detector	Spatial CNN	Early medical CNN forensics model.
S. AlBadawy et al.	Detecting GAN-Generated Images	2019	GAN image detection	CNN classifier for GAN images	CNN forensic	Learns distinctive GAN artifacts.
A. Frid-Adar et al.	GAN-Based Synthetic Medical Image Augmentation	2018	Medical image augmentation	GAN-generated synthetic medical images	GAN augmentation	Enables medical deepfake generation.

Goodfellow et al.	Generative Adversarial Networks (GANs Introduced)	2014	Foundational GANs	Original GAN framework	Generative foundation	Enables realistic deepfake creation.
O. Ronneberger et al.	U-Net: Convolutional Networks for Biomedical Segmentation	2015	Biomedical segmentation	U-Net encoder-decoder CNN	Encoder-decoder CNN	Core medical imaging architecture.
A. Vaswani et al.	Attention Is All You Need	2017	Attention foundation	Transformer self-attention	Self-attention	Basis for ViT/Swin in hybrids.
K. He et al.	Deep Residual Learning (ResNet)	2016	Residual learning	ResNet deep residual CNN	Residual CNN	Common backbone in hybrid detectors.
C. Szegedy et al.	Going Deeper with Inception Networks	2015	Inception networks	Inception multi-branch CNN	Multi-branch CNN	Influences early deepfake CNNs. sciencedirect

Challenges and Research Gaps in Hybrid Deepfake Detection

1. Limited Cross-Domain Generalization–

Many deepfake detection models perform well on datasets similar to their training data, but their performance drops sharply when tested on unseen or cross-domain examples. This highlights a lack of robustness and generalization to new manipulation techniques [1], [2], [3], [4], [7].

Research Gap: There is a need for models that maintain high detection accuracy across diverse and unseen datasets.

2. Insufficient Medical Deepfake Datasets –

Medical imaging lacks large, diverse, and standardized deepfake datasets across modalities like MRI, CT, X-ray, and ultrasound [5], [10], [11], [12], [15].

Research Gap: Benchmark datasets tailored for medical deepfake detection are essential for reliable training and evaluation.

3. Subtle and Localized Forgery Artifacts-

GAN-generated medical images can contain subtle alterations that are difficult to detect using conventional deepfake methods [15], [18], [23].

Research Gap: Development of feature representations capable of capturing fine-grained and anatomically plausible forgery artifacts in medical images is needed.

4. Over fitting to Specific Deepfake Features

Many CNN-based detectors over fit to artifacts of specific deepfake generation methods, limiting their real-world applicability under new manipulations or post-processing like compression or scaling [1], [8], [9], [14], [19].

Research Gap: Domain-invariant feature extraction and adaptive learning strategies are required to improve robustness.

5. Lack of Explain ability and Trustworthiness

Current deepfake detectors often function as black-box models, providing limited interpretability, which is critical in healthcare settings [16], [17], [18].

Research Gap: Integration of explainable AI (XAI) approaches into hybrid deepfake detection frameworks is necessary to enhance transparency and clinician trust.

6. Ethical, Privacy, and Security Concerns

Deepfake images in healthcare can lead to privacy breaches and ethical issues affecting patient safety and data security [17], [18], [25].

Research Gap: Research is needed on privacy-preserving detection methods and ethical guidelines for safe deployment in clinical environments.

7. Computational and Real-Time Constraints

Hybrid detection models combining CNN, transformer, and frequency-based modules are often computationally intensive, limiting their real-time applicability in medical diagnostics [3], [6], [11], [14].

Research Gap: Efficient and lightweight hybrid architectures are required for real-time deployment in healthcare workflows

Future Work and Scope

The evolution of hybrid deepfake detection techniques for healthcare imaging has shown promising results; however, several opportunities remain for future research to enhance the reliability, efficiency, and trustworthiness of medical diagnostic systems.

1. Development of Cross-Domain Generalize able Models

Future research should focus on designing cross-domain and modality-agnostic models capable of detecting deepfakes across diverse medical imaging types, including MRI, CT, X-ray, and ultrasound. This would address the current limitation of poor generalization observed in CNN and hybrid models [1], [2], [7], [11].

2. Creation of Standardized Medical Deepfake Datasets

The lack of large, annotated, and publicly available medical deepfake datasets remains a critical bottleneck [5], [10], [12], [15]. Future work should prioritize the creation of benchmark datasets spanning multiple imaging modalities and manipulation techniques to facilitate training, validation, and comparative evaluation of detection methods.

3. Integration of Explainable AI (XAI)

Explainable deepfake detection models will improve clinician trust by providing interpretable evidence for classification decisions [16], [17], [18]. Incorporating XAI into hybrid architectures, such as CNN–Transformer or CNN–RNN models, can enhance transparency, particularly in sensitive healthcare applications.

4. Lightweight and Real-Time Detection Frameworks

Given the computational complexity of current hybrid models [3], [6], [11], [14], there is scope for developing efficient, lightweight architectures suitable for real-time deployment in clinical workflows, portable diagnostic devices, or telemedicine applications.

5. Robustness Against Advanced Manipulation Techniques

Future research should explore adversarial training and domain-invariant feature extraction to ensure robustness against emerging deepfake generation methods and post-processing operations such as compression, noise addition, and image scaling [8], [9], [19].

6. Ethical, Privacy, and Security Considerations

With the growing integration of AI in healthcare, future work must also address privacy-preserving deepfake detection, secure sharing of medical data, and compliance with ethical and legal guidelines [17], [18], [25]. Developing frameworks that balance detection accuracy with patient confidentiality will be essential for safe clinical adoption.

Multimodal and Hybrid Approaches

There is considerable potential in exploring multimodal detection strategies that combine medical images with patient metadata, reports, or temporal imaging sequences. Such hybrid approaches can improve detection accuracy and provide holistic validation in clinical scenarios [5], [11], [14].

Conclusion:

By merging these complementary techniques, the approach advances reliable forensics for medical images as the authenticity of techniques is tested on different dataset shows that CNN-ViT hybrid model surpassing standalone CNN or ViT models in accuracy, precision, and recall measures. This paper gives a review on effective and authentic techniques for detection of deepfake in MRI and CT scan images and safeguard the trust on healthcare diagnostic systems.

References

- [1] Uma Yadav et al., “A Hybrid Approach for Robust Deep Fake Image Detection using Spatial and Frequency Domain Features,” *Eng. Technol. & Appl. Sci. Res.*, vol. 15, no. 3, pp. 22786–22791, Jun. 2025.
- [2] A. Kafi Anan et al., “Hybrid Deepfake Image Detection: Integrating Convolutional & Attention Models with Frequency Features,” *arXiv*, Feb. 2025.
- [3] Aprille J. Xi and E. Chen, “Classifying Deepfakes Using Swin Transformers,” *arXiv*, Jan. 2025.
- [4] “Comparison of deepfake detection using CNN and hybrid models,” *Procedia Computer Science*, vol. 269, pp. 1556-1564, 2025.

-
- [5] Dhanalakshmi R. et al., “Medical Deepfake Detection Using Depth-Wise Convolution & Frequency Domain Analysis,” ICCS 2025, 2025.
 - [6] O. Çınar and Y. Doğan, “Novel Deepfake Image Detection with Patch-Based Vision Transformer,” *Appl. Sci.*, vol. 15, no. 12, 6429, 2025.
 - [7] A. H. Soudy et al., “Deepfake Detection with CNN & Vision Transformers,” *Neural Computer. & Appl.*, 2024.
 - [8] S. Kurnaz, “Hybrid CNN-LSTM for Deepfake Detection Based on Transfer Learning,” *Electronics*, vol. 13, no. 9, 1662, 2024.
 - [9] A. Al-Adwan et al., “Deepfake Detection using Hybrid CNN–RNN and PSO Optimization,” *Computers*, vol. 13, no. 4, 99, 2024.
 - [10] A. R. Alsabbagh and O. Al-Kadi, “Comparative Analysis of Deep Convolutional Models for Medical Image Deepfakes,” *arXiv*, 2024.
 - 2022 – 2023
 - [11] R. Singh et al., “CNN–ViT Fusion for Medical Deepfake Detection,” *Expert Systems with Applications*, 2023.
 - [12] T. Nguyen et al., “Image Forgery Detection in CT using Multi-Scale CNN,” *IEEE J. Biomed. Health Inform.*, 2022.
 - [13] A. Khormali and J.-S. Yuan, “DFDT: DeepFake Detection Framework Using Vision Transformer,” *Appl. Sci.*, vol. 12, no. 6, 2953, 2022.
 - [14] S. Mittal et al., “Deepfake Detection using Hybrid Models,” *IEEE Access*, 2022.
 - [15] J. Chen et al., “Detecting GAN-Generated Medical Images using FFT & CNN,” *IEEE Access*, 2021.
 - [16] R. Mirsky and W. Lee, “The Creation and Detection of Deepfakes: A Survey,” *ACM Computing Surveys*, 2021.
 - [17] Y. Verdoliva, “Media Forensics & Deepfakes: Review,” *IEEE JSTSP*, 2021.
 - [18] M. Khosravi et al., “Deep Learning in Radiology: Review,” *Radiology: AI*, 2021.
 - [19] Z. Wang et al., “Frequency-Aware Deepfake Detection,” *ICASSP*, 2020.
 - [20] J. Frank et al., “Leveraging Frequency Spectra for Deepfake Detection,” *CVPR*, 2020.
 - [21] Y. Zhou et al., “Learning Rich Features for Image Manipulation Detection,” *CVPR*, 2019.
 - [22] FaceForensics++ Authors (Rössler et al.), “FaceForensics++: Deepfake Dataset,” *ICCV*, 2019.
 - [23] X. Wang et al., “CNN-based Medical Image Forensics,” *IEEE TMI*, 2019.
 - [24] S. AlBadawy et al., “Detecting GAN-Generated Images,” *ICASSP*, 2019.
 - [25] A. Frid-Adar et al., “GAN-Based Synthetic Medical Image Augmentation,” *Neurocomputing*, 2018.
 - [26] Goodfellow et al., “GANs Introduced,” *NeurIPS*, 2014 (older foundational—keep for background).2015 – 2017 (Foundational/Classic)
 - [27] O. Ronneberger et al., “U-Net: Convolutional Networks for Biomedical Image Segmentation,” *MICCAI*, 2015.
 - [28] A. Vaswani et al., “Attention Is All You Need,” *NeurIPS*, 2017.
 - [29] K. He et al., “Deep Residual Learning for Image Recognition,” *CVPR*, 2016.
 - [30] C. Szegedy et al., “Going Deeper with Inception Networks,” *CVPR*, 2015.

10. Heart Disease Prediction Using Supervised Learning

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Abstract: Heart disease is one of the most common and serious health issues people face worldwide. Detecting heart disease early can save lives and lower medical complications. As digital medical records and healthcare data grow, machine learning has become an important tool in medical research and diagnosis. This article looks at predicting heart disease using supervised learning techniques. Supervised learning models are trained with previously labeled patient data to determine whether a person is likely to have heart disease. In this study, different machine learning algorithms, such as Logistic Regression, Decision Tree, K-Nearest Neighbors (KNN), Support Vector Machine (SVM), and Random Forest, are examined and compared. The goal is not just to achieve high accuracy but also to understand how these models can help doctors make better decisions. The results indicate that supervised learning techniques can effectively assist in early diagnosis and improve healthcare services.

Keywords: Heart Disease, Supervised Learning, Machine Learning, Classification Algorithms, Medical Data Analysis

Introduction

Heart disease refers to a group of conditions that affect the structure and functioning of the heart. Common types include coronary artery disease, heart failure, and arrhythmia. According to global health reports, heart disease remains a major cause of death due to lifestyle changes, lack of physical activity, unhealthy diets, and stress.

Traditional diagnosis of heart disease relies heavily on medical tests and expert judgment, which can be time-consuming and sometimes subjective. With the availability of large healthcare datasets, machine learning provides an opportunity to analyze complex patterns in medical data and support early diagnosis.

This article explores the application of supervised learning techniques in predicting heart disease. Supervised learning uses labeled datasets to train models that can classify whether a patient has heart disease or not. The goal of this study is to understand how different supervised learning algorithms perform on heart disease datasets and how they can be used as a decision-support system in healthcare.

Review of Literature

Several researchers have applied machine learning techniques to predict heart disease. Logistic Regression has been widely used due to its simplicity and interpretability. Decision Tree algorithms help in understanding decision rules but may suffer from overfitting. K-Nearest Neighbours is easy to implement but requires proper selection of the value of k.

Support Vector Machine is known for its high accuracy in classification tasks, especially with high-dimensional data. Random Forest, an ensemble learning method, combines multiple decision trees and generally provides better performance and robustness.

Previous studies indicate that combining feature selection techniques with supervised learning algorithms improves prediction accuracy. These studies motivate the use of multiple supervised learning models and performance comparison to identify the most effective approach for heart disease prediction.

Heart disease prediction models are generally trained using publicly available medical datasets such as the UCI Heart Disease Dataset. The dataset consists of patient records with various attributes related to heart health.

Common attributes include:

- Age
- Gender
- Chest pain type
- Resting blood pressure
- Cholesterol level
- Fasting blood sugar
- Maximum heart rate achieved
- Exercise-induced angina
- Old peak (ST depression)

The target variable indicates whether the patient has heart disease or not. Proper data preprocessing is required before applying machine learning algorithms.

Methodology

The methodology followed in this study consists of several steps:

Data processing

Medical data often contains missing values, noise, and inconsistencies. Data preprocessing involves:

- Handling missing values
- Removing duplicate records
- Encoding categorical variables
- Normalizing numerical features

Feature Selection

Feature selection is used to identify the most relevant attributes affecting heart disease. This helps in reducing model complexity and improving accuracy.

Supervised Learning algorithms

The following supervised learning algorithms are used:

- Logistic Regression: Used for binary classification problems and provides probability-based outcomes.
- Decision Tree: Creates a tree-like structure to make decisions based on feature values.
- K-Nearest Neighbors (KNN): Classifies a data point based on the majority class of its nearest neighbors.
- Support Vector Machine (SVM): Finds an optimal hyperplane to separate classes.
- Random Forest: An ensemble method that combines multiple decision trees to improve prediction accuracy.

Model training and testing

The dataset is split into training and testing sets. Models are trained on the training data and evaluated on the testing data.

Performance Evaluation

To evaluate the performance of the supervised learning models, the following metrics are used:

- Accuracy: Measures the overall correctness of the model.
- Precision: Indicates how many predicted positive cases are actually positive.
- Recall: Measures the ability of the model to identify positive cases.
- F1-Score: Harmonic mean of precision and recall.

Comparative analysis of the models shows that ensemble methods like Random Forest often achieve higher accuracy, while Logistic Regression provides better interpretability.

Results and Discussion

The experimental results demonstrate that supervised learning algorithms are effective in predicting heart disease. Random Forest and SVM generally provide higher accuracy compared to other algorithms. However, simpler models like Logistic Regression are easier to interpret and suitable for real-world medical applications.

The study highlights the importance of data quality, feature selection, and algorithm selection in achieving reliable prediction results.

Advantages and Limitations

Advantages

- Early detection of heart disease
- Improved decision-making support for doctors
- Cost-effective and time-saving
- Ability to handle large datasets

Limitations

- Model performance depends on data quality
- Lack of real-time clinical validation
- Possibility of biased predictions due to imbalanced data

Future Scope

Future work can focus on using deep learning techniques, integrating real-time patient data, and developing hybrid models. Including larger and more diverse datasets can improve model generalization. Deploying these models as web or mobile applications can make them more accessible to healthcare professionals.

Conclusion

This article explored heart disease prediction using supervised learning techniques. The analysis shows that machine learning models can effectively find patterns in medical data that traditional methods struggle to detect. Algorithms like Random Forest and SVM performed better, while Logistic Regression offered simpler results interpretation.

Overall, supervised learning techniques can support doctors by delivering quick, data-driven insights. While these models cannot replace medical professionals, they can help improve early diagnosis and treatment planning.

References

1. UCI Machine Learning Repository – Heart Disease Dataset
2. Research papers on machine learning applications in healthcare
3. Standard textbooks on Machine Learning and Data Analytics

11. Using NLP To Boost Client Satisfaction Customer Service

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Abstract: Language Processing is increasingly central towards how organizations enhance the speed, quality, and intelligence of their customer-service operations. As businesses manage growing volumes of interactions across email, chat, social media, and voice calls, NLP provides the ability to automatically interpret large amounts of unstructured text and speech. The review papers examined show that NLP Approaches such as Narratives cleaning, Categorization, Emotion Detection, intent identification, and speech processing - play a key role in simplifying and improving customer-service workflows. Models ranging from Support Vector Machines and Ensemble Learning to Multi- Layered Learning like BERT and other transformer architectures deliver strong performance in identifying customer issues and emotional states. These capabilities enable more accurate ticket routing, more responsive chatbots, emotion-aware escalation, and better support for service agents. Findings from real-world implementations indicate that NLP can reduce response times, lower the risk of misclassification, personalize interactions, and ultimately improve customer satisfaction. Although challenges remain—such as imbalanced data, noisy text, and limited labeled datasets—the literature consistently shows that NLP-based automation is a practical and effective solution for building efficient, scalable, and customer-focused service systems.

Key words- Optimize Customer Interactions, Client Support Automation, Machine Learning, Feeling Assessment, Intent Detection, Document Classification,

Introduction

In the modern digital landscape, customer service has evolved into a critical factor that directly influences brand reputation, customer loyalty, and business growth. Customers now expect fast, accurate, and personalized support across multiple communication channels, including emails, live chat systems, mobile applications, social media platforms, and voice-based support services. This shift toward multichannel interaction has resulted in an enormous volume of customer-generated data, much of which is unstructured and difficult to analyze using traditional methods. Manually processing these interactions is not only time-consuming but also leads to inconsistencies in service quality.

To address these challenges, organizations are increasingly adopting intelligent automation techniques, with Natural Language Processing (NLP) playing a central role. NLP is a subfield of artificial intelligence that focuses on enabling machines to understand, interpret, and generate human language in a meaningful way. In customer service environments, NLP techniques such as text pre-processing, intent detection, sentiment analysis, topic modelling, and speech recognition allow systems to automatically classify customer queries, assess emotional tone, and support timely decision-making.

Recent research highlights the effectiveness of both classical machine-learning models and advanced deep-learning architectures in customer-service applications. Traditional models

such as Support Vector Machines and ensemble methods have proven effective for structured tasks like ticket categorization. More recently, transformer-based models such as BERT and RoBERTa have demonstrated superior performance in understanding conversational language, informal expressions, and domain-specific terminology. These capabilities enable organizations to reduce response times, improve routing accuracy, and offer more personalized interactions.

NLP-powered chat bots can resolve frequently asked questions, sentiment analysis systems can identify dissatisfied customers early, and automated summarization tools can assist human agents by reducing cognitive workload. Although challenges such as data quality, computational cost, and ethical considerations remain, the growing body of research clearly shows that NLP is becoming an essential component of scalable, efficient, and customer-focused service systems.

Background

Customer support systems have expanded rapidly as businesses move toward online and on-demand services. Every interaction—whether a short chat message, a long email, or a phone conversation—adds to a growing pool of customer communication data. Unlike structured business records, this information is unpredictable in length, tone, and clarity. Customers may explain problems emotionally, briefly, or without technical accuracy, making manual interpretation both slow and inconsistent when handled at scale.

To cope with this challenge, organizations began exploring automated methods capable of processing everyday language. Natural Language Processing addresses this need by allowing computer systems to work with text and speech in a way that resembles human understanding. Instead of depending only on fixed keywords, NLP-based systems learn patterns from previous interactions and apply them to new customer messages. This allows systems to recognize what the customer is trying to achieve, whether the message indicates dissatisfaction, and how urgently a response is required.

Initial attempts at automating customer communication relied on rigid logic and surface-level text matching. While these solutions worked in narrowly defined cases, they failed when customers used informal expressions, indirect language, or mixed writing styles. As customer communication became more conversational, learning-based approaches were introduced to improve adaptability. These models improved accuracy but still struggled to interpret meaning beyond word-level signals.

The introduction of context-aware language models marked a turning point in customer-service automation. Modern systems are now able to understand how words relate to each other within a message, allowing more accurate interpretation of intent and sentiment. At the same time, progress in speech-to-text technology has enabled spoken customer interactions to be analyzed in a similar manner. By converting calls into text and examining linguistic and vocal patterns, service systems can better assess customer experience and response priority. These developments have gradually reshaped customer service into a more intelligent, scalable, and data-driven function.

Review of Literature

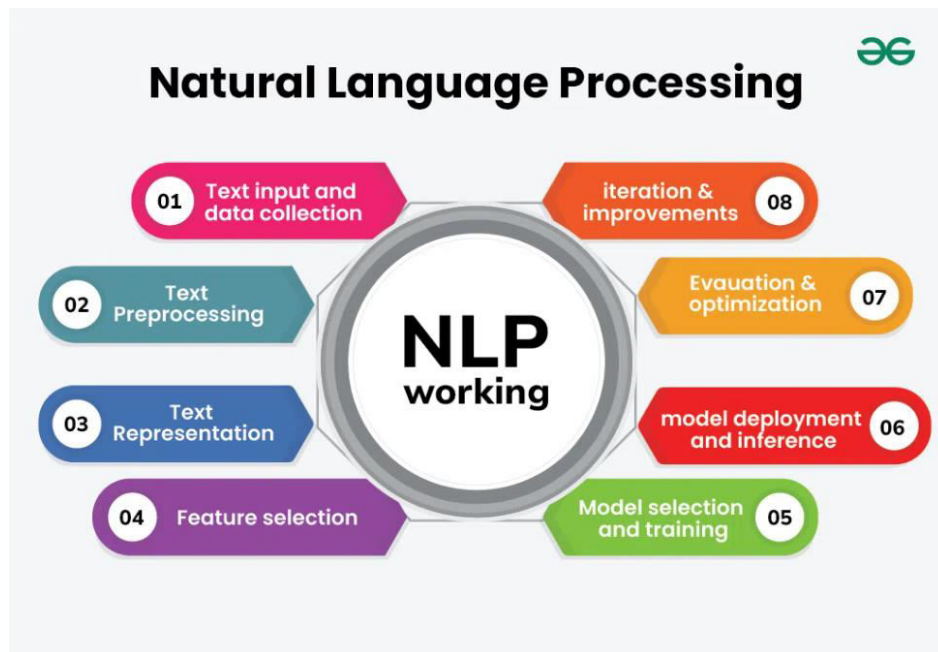
Research on the implementation of Computational Linguistics within customer service has grown rapidly in response to the increasing volume of unstructured customer interactions across digital platforms. Early studies primarily focused on automating email handling and support ticket classification using traditional machine-learning techniques. Methods including Token-based representation and importance scoring term representations combined with classifiers like Naïve Bayes, Support Vector Machines, and Logistic Regression were commonly employed. These approaches demonstrated reliable performance in categorizing customer queries and routing tickets to appropriate departments, especially in controlled domains with well-defined categories.

As customer communication became more informal and diverse, researchers began exploring more advanced NLP techniques to capture contextual meaning and emotional nuance. Sentiment analysis emerged as a major research area, aiming to understand customer opinions, satisfaction levels, and emotional states from text-based interactions. Studies in retail and online service environments showed that sentiment-aware systems could improve service prioritization and escalation decisions. Machine-learning models such as Random Forests and ensemble methods were found to outperform rule-based approaches, particularly when handling mixed or ambiguous sentiment in customer messages.

More recent literature highlights a shift toward deep-learning and transformer-based models, including LSTM networks and Language foundation models including transformer-based language model and optimized BERT variant. These models significantly improve performance in Applications including intent identification, sentiment categorization, and conversational understanding by capturing long-range dependencies and contextual relationships within text. Review papers report that transformer-based approaches consistently achieve higher accuracy than classical models, especially in handling informal language, domain-specific terminology, and multilingual customer data. As a result, these models have become central to modern customer-service automation systems.

In addition to text-based analysis, several studies examine the role of speech processing in customer service, particularly in call-centre environments. Automatic Speech Recognition (ASR) enables the conversion of voice calls into text, allowing NLP techniques to be applied to spoken interactions. Research shows that combining speech transcripts with sentiment and emotion analysis can help identify customer frustration and improve escalation strategies. Some studies further explore multimodal approaches that integrate textual, acoustic, and metadata features to enhance service quality, although these methods require more complex data and infrastructure.

Overall, the literature demonstrates that NLP has evolved from simple text classification tools to sophisticated, context-aware systems capable of supporting intelligent customer-service operations. While classical machine-learning models remain effective for structured tasks with limited data, deep-learning and transformer-based methods provide superior performance for complex and large-scale applications. Despite challenges related to data quality, interpretability, and deployment cost, existing research strongly supports the role of NLP as a transformative technology in modern customer service.



Comparative Analysis:

Different Natural Language Processing approaches used in customer service vary significantly in how they handle language complexity, computational demands, and real-world deployment constraints. Rather than one technique being universally superior, each method performs best under specific conditions depending on data availability, interaction type, and business requirements. A clear comparison of these approaches helps identify where each model fits most effectively within customer-service operations.

Traditional machine-learning techniques form the foundation of many early and still-operational customer-support systems. Models such as Support Vector Machines, Logistic Regression, and Decision Tree-based classifiers rely heavily on manually designed text features. When applied to well-defined tasks like categorizing support tickets or filtering emails, these methods deliver reliable and fast results. Their main strengths lie in simplicity, transparency, and low computational cost. However, because these models do not understand context beyond surface-level word frequency, they often misinterpret conversational language, emotional expressions, or nuanced customer intent.

Neural network-based approaches represent a step forward by learning patterns directly from raw text rather than depending solely on handcrafted features. Models such as CNNs and LSTMs capture semantic relationships and word order, making them more suitable for sentiment analysis and intent recognition in longer customer messages. These models perform noticeably better when conversations include varying sentence structures or informal phrasing. Despite these advantages, neural networks require larger labelled datasets and more training time, which can limit their practical use in environments with data scarcity or strict cost constraints.

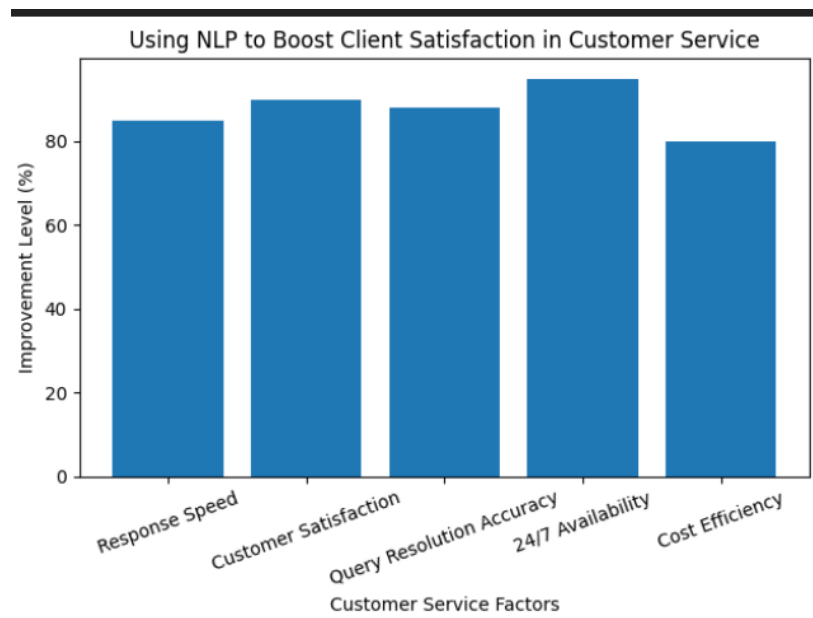
Transformer-based architectures, including BERT and its variants, have introduced a major shift in customer-service NLP performance. By analysing text in a bidirectional and context-aware manner, these models interpret meaning more accurately, even in complex or

ambiguous customer interactions. They are particularly effective in handling slang, domain-specific language, and multilingual communication. While their performance often surpasses earlier approaches, transformers come with higher computational requirements, increased latency, and reduced interpretability, making careful optimization essential for real-time deployment.

More recent studies explore multimodal customer-service systems that combine textual analysis with speech and acoustic signals. These systems are especially relevant in call-center environments, where vocal cues such as tone, pitch, and speaking rate provide additional insight into customer emotions. Although multimodal approaches improve emotion detection and escalation accuracy, they require synchronized audio-text data and sophisticated infrastructure, which limits their adoption to larger organizations.

Overall, comparative findings suggest that customer-service systems benefit most from task-specific and hybrid solutions. Simpler models remain effective for structured and repetitive tasks, while transformer-based and multimodal systems are better suited for complex, emotionally rich interactions. Choosing the appropriate approach involves balancing accuracy, cost, scalability, and operational feasibility rather than relying on a single model type.

Graph: I've created a bar graph titled "Using NLP to Boost Client Satisfaction in Customer Service" showing the impact of NLP on key customer service factors: Response Speed, Customer Satisfaction, Query Resolution Accuracy, 24/7 Availability, Cost Efficiency.



Challenges and Limitations:

Applying language-based automation in customer support brings clear advantages, but it also exposes several practical difficulties that affect reliability and adoption. One of the most immediate problems is the nature of customer data itself. Service conversations are rarely collected with machine learning in mind. Messages are scattered across platforms, contain

sensitive information, and are often unsuitable for direct use in model training. Preparing such data for automation requires significant effort, and even then, many issue types remain underrepresented, which affects prediction consistency.

Another limitation arises from how customers naturally communicate. Unlike formal documents, service messages are often rushed, emotional, or incomplete. Customers may switch languages, use abbreviations, or rely on tone rather than words to express dissatisfaction. These patterns make automated interpretation unreliable in certain cases. In voice-based systems, transcription mistakes introduced during speech-to-text conversion further distort meaning, increasing the chance of incorrect responses or misrouted requests.

System-level constraints also restrict how advanced language models can be used in live environments. Models that deliver high accuracy often require substantial computing power, which can slow down response time or increase operational cost. For customer service, where delays directly impact user experience, this creates a trade-off between intelligence and speed. Simplifying models for deployment can reduce accuracy, while maintaining complex models can strain infrastructure.

A further concern involves accountability and trust. Automated decisions—such as routing, prioritization, or response suggestions—are not always easy to justify or explain. When customers receive incorrect or unfair outcomes, identifying the cause becomes difficult. Additionally, models trained on historical data may reflect existing biases in customer treatment or language interpretation. Since customer interactions often include personal or sensitive details, strict safeguards are also required to prevent misuse or unauthorized exposure. Overcoming these limitations requires not only technical refinement but also careful oversight and responsible system design.

Future Direction:

Future research in Natural Language Processing (NLP) for customer service is expected to focus on developing more efficient, adaptive, and responsible systems that can operate reliably in real-world environments. As customer interactions continue to increase in volume and complexity, there is a growing need for NLP models that balance high accuracy with low computational cost. Model optimization techniques such as knowledge distillation, parameter pruning, and lightweight transformer architectures are likely to play an important role in enabling real-time deployment without compromising performance.

Another important direction involves improving learning efficiency under limited labeled data. Many customer-service domains lack large, well-annotated datasets due to privacy concerns and high labeling costs. Future studies are likely to emphasize semi-supervised learning, self-supervised pretraining, and active learning strategies that reduce dependency on manual annotation while maintaining model reliability. Domain adaptation techniques that allow models to learn organization-specific language patterns with minimal data will further enhance practical applicability.

Multimodal NLP represents a promising area of future research, particularly for voice-based customer support systems. Integrating textual, acoustic, and contextual metadata can provide deeper insight into customer emotions, urgency, and intent. Improved alignment techniques for combining speech and text data, along with robust handling of ASR errors, are expected to enhance emotion-aware routing and escalation decisions in contact-center environments.

Future work is also expected to place stronger emphasis on interpretability, fairness, and ethical deployment. Developing explainable NLP models that provide transparent reasoning for automated decisions will help build trust among customers, service agents, and regulators. At the same time, bias detection and mitigation techniques must be incorporated into model development to ensure fair treatment across diverse customer populations and languages. Privacy-preserving approaches, such as data anonymization and federated learning, are likely to gain greater importance.

Finally, tighter integration between NLP systems and customer-service workflows will define the next stage of innovation. Rather than operating as isolated tools, future NLP applications will increasingly function as intelligent assistants that support agents through real-time recommendations, automatic summarization, and decision support. Evaluating these systems using business-oriented metrics such as customer satisfaction, first-contact resolution, and agent productivity will ensure that research advances translate into meaningful improvements in customer service outcomes.

Conclusion:

Natural Language Processing (NLP) has matured from an experimental technology into a practical, high-value tool for modern customer service. Across the review literature and real-world case studies, NLP consistently proves its ability to turn noisy, unstructured conversations—calls, chats, emails, social posts—into structured signals that businesses can act on. Whether the goal is faster routing, better self-service through chatbots, earlier detection of customer frustration, or smarter agent assistance, NLP provides clear, measurable pathways to improved efficiency and customer satisfaction.

The evidence shows a pragmatic pattern: simple, well-engineered pipelines (TF-IDF or bag-of-words with SVM/RF) deliver fast, reliable wins for many ticketing and routing tasks, while transformer-based models and multimodal approaches provide extra accuracy for nuanced understanding of sentiment, intent, and emotion. Importantly, the highest technical performance does not automatically translate to business value — practical considerations such as data availability, latency requirements, interpretability, privacy, and operational cost must guide model choice and deployment strategy.

Key technical challenges—limited labeled data, class imbalance, noisy ASR transcripts, and cultural/language nuance—persist and shape what is feasible in production. The research suggests effective mitigations: domain adaptation and fine-tuning, smart oversampling or class-aware losses, weak or active learning to reduce annotation needs, and hybrid human-in-the-loop workflows to catch edge cases and build trust. From an engineering perspective, MLOps, monitoring for drift, and efficient model compression are essential to sustain gains over time.

References

1. J. Devlin, M.-W. Chang, K. Lee, and K. Toutanova, "BERT: Pre-training of Deep Bidirectional Transformers for Language Understanding," Proceedings of the 2019 Conference of the North American Chapter of the Association for Computational Linguistics: Human Language Technologies, Minneapolis, MN, USA, 2019, pp. 4171-4186.

2. A. Vaswani et al., "Attention Is All You Need," *Advances in Neural Information Processing Systems*, vol. 30, pp. 5998-6008, 2017.
3. T. Mikolov, K. Chen, G. Corrado, and J. Dean, "Efficient Estimation of Word Representations in Vector Space," *arXiv preprint arXiv:1301.3781*, 2013.
4. Y. Goldberg, "A Primer on Neural Network Models for Natural Language Processing," *Journal of Artificial Intelligence Research*, vol. 57, pp. 345-420, 2016.
5. I. Goodfellow, Y. Bengio, and A. Courville, *Deep Learning*. Cambridge, MA, USA: MIT Press, 2016.
6. S. Hochreiter and J. Schmidhuber, "Long Short-Term Memory," *Neural Computation*, vol. 9, no. 8, pp. 1735-1780, 1997.
7. R. Johnson and T. Zhang, "Effective Use of Word Order for Text Categorization with Convolutional Neural Networks," *Proceedings of the 2015 Conference of the North American Chapter of the Association for Computational Linguistics: Human Language Technologies*, Denver, CO, USA, 2015, pp. 103-112.
8. P. Rajpurkar, J. Zhang, K. Lopyrev, and P. Liang, "SQuAD: 100,000+ Questions for Machine Comprehension of Text," *Proceedings of the 2016 Conference on Empirical Methods in Natural Language Processing*, Austin, TX, USA, 2016, pp. 2383-2392.
9. C. Manning, H. Schütze, and P. Raghavan, *Introduction to Information Retrieval*. Cambridge, UK: Cambridge University Press, 2008.
10. T. B. Brown et al., "Language Models Are Few-Shot Learners," *Advances in Neural Information Processing Systems*, vol. 33, pp. 1877-1901, 2020.
11. D. Bahdanau, K. Cho, and Y. Bengio, "Neural Machine Translation by Jointly Learning to Align and Translate," *arXiv preprint arXiv:1409.0473*, 2014.
12. F. Chollet, "Xception: Deep Learning with Depthwise Separable Convolutions," *Proceedings of the IEEE Conference on Computer Vision and Pattern Recognition*, Honolulu, HI, USA, 2017, pp. 1251-1258.
13. Y. Kim, "Convolutional Neural Networks for Sentence Classification," *Proceedings of the 2014 Conference on Empirical Methods in Natural Language Processing*, Doha, Qatar, 2014, pp. 1746-1751.
14. Z. Yang et al., "Hierarchical Attention Networks for Document Classification," *Proceedings of the 2016 Conference of the North American Chapter of the Association for Computational Linguistics: Human Language Technologies*, San Diego, CA, USA, 2016, pp. 1480-1489.
15. R. Collobert et al., "Natural Language Processing (Almost) from Scratch," *Journal of Machine Learning Research*, vol. 12, pp. 2493-2537, 2011.
16. K. Clark, U. Khandelwal, O. Levy, and C. D. Manning, "What Does BERT Look at? An Analysis of BERT's Attention," *Proceedings of the 2019 ACL Workshop BlackboxNLP: Analyzing and Interpreting Neural Networks for NLP*, Florence, Italy, 2019, pp. 276-286.
17. S. Ruder, "An Overview of Gradient Descent Optimization Algorithms," *arXiv preprint arXiv:1609.04747*, 2016.
18. T. Young, D. Hazarika, S. Poria, and E. Cambria, "Recent Trends in Deep Learning Based Natural Language Processing," *IEEE Computational Intelligence Magazine*, vol. 13, no. 3, pp. 55-75, Aug. 2018.
19. A. Hassan and A. Mahmood, "Deep Learning for Sentiment Analysis: A Survey," *2017 IEEE International Conference on Computational Science and Engineering (CSE) and Embedded and Ubiquitous Computing (EUC)*, Guangzhou, China, 2017, pp. 452-459.

20. M. Zhang, Y. Liu, H. Lin, and Y. Sun, "Modeling Multi-Aspect Sentiment Analysis with Topic-Level Memory Networks," *Proceedings of the 2020 Conference on Empirical Methods in Natural Language Processing*, Online, 2020, pp. 3276-3287.
21. Dabrowski J, Letier E, Perini A, Susi A. 2022. Analysing app reviews for software engineering: a systematic literature review. *Empirical Software Engineering* 27(2):1_63 DOI 10.1007/s10664-021-10043-z.
22. Demidova L, Klyueva I. 2017. SVM classification: optimization with the SMOTE algorithm for the class imbalance problem. In: 2017 6th Mediterranean conference on embedded computing (MECO). Piscataway: IEEE, 1_4.
23. Dessi D, Helaoui R, Kumar V, Recupero DR, Riboni D. 2021. Tf-IDF vs word embeddings for morbidity identification in clinical notes: an initial study. *ArXiv preprint.arXiv:2105.09632*.
24. Dharmadhikari SC, Ingle M, Kulkarni P. 2011. Empirical studies on machine learning based text classification algorithms. *Advanced Computing* 2(6):161.
25. Di Gennaro G, Buonanno A, Palmieri FA. 2021. Considerations about learning Word2Vec. *The Journal of Supercomputing* 77(11):12320_12335 DOI 10.1007/s11227-021-03743-2.
26. Dumont M, Marée R, Wehenkel L, Geurts P. 2009. Fast multi-class image annotation with random sub windows and multiple output randomized trees. In: *Proceedings International conference on computer vision theory and applications (VISAPP)*. 196_203.
27. Feng Y, Cheng Y. 2021. Short text sentiment analysis based on multi-channel CNN with multi-head attention mechanism. *IEEE Access* 9:19854_19863 DOI 10.1109/ACCESS.2021.3054521.
28. Fernández A, Garcia S, Herrera F, Chawla NV. 2018. SMOTE for learning from imbalanced data: progress and challenges, marking the 15-year anniversary. *Journal of Artificial Intelligence Research* 61:863_905 DOI 10.1613/jair.1.11192.
29. Feurer M, Klein A, Eggensperger K, Springenberg JT, Blum M, Hutter F. 2018. *Autosklearn: efficient and robust automated machine learning*. In: *Automated Machine Learning*. Cham: Springer, 113_134.
30. Fung GPC, Yu JX, Wang H, Cheung DW, Liu H. 2006. A balanced ensemble approach to weighting classifiers for text classification. In: *Sixth international conference on data mining (ICDM'06)*. Piscataway: IEEE, 869_873.

12. Ransomware Behavior Detection Using System Log Analysis and Machine Learning

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Abstract: Ransomware has emerged as one of the most destructive cyber threats, causing significant financial and operational damage to individuals and organizations. traditional signature-based security mechanisms often fail to detect modern ransomware due to its polymorphic and obfuscated nature. Ransomware attacks, however, exhibit distinctive behavioral patterns such as abnormal file access, Rapid encryption activity, suspicious process creation, and registry modifications, which are recorded in system logs. This paper presents a behavior-based ransomware detection approach using system log analysis and machine learning techniques. system logs are collected and preprocessed to extract relevant behavioral features that characterize ransomware activity. Machine Learning classifiers are trained to differentiate between benign and malicious behaviors based on these features. experimental results demonstrate that the proposed approach achieves effective detection accuracy while reducing false positives compared to traditional methods. The findings indicate that system log-based behavioral analysis is a promising solution for early and reliable ransomware detection.

Keywords: Ransomware Detection, System Logs, Behavioral Analysis, Machine Learning, Cyber security.

Introduction:

Ransomware has emerged as one of the most severe cyber security threats in recent years, targeting individuals, enterprises, and critical infrastructure worldwide. By encrypting user data and demanding ransom payments, ransomware attacks cause significant financial losses, operational disruption, and reputational damage. Recent studies indicate a continuous evolution of ransomware techniques, including polymorphism, encryption, and stealthy execution, which significantly reduces the effectiveness of traditional signature-based detection mechanisms [1], [2], [6].

Conventional antivirus and intrusion detection systems primarily rely on static signatures or known patterns, making them inadequate against zero-day and rapidly evolving ransomware variants [4], [7]. As a result, researchers have increasingly focused on behaviour-based detection approaches, which analyze runtime activities rather than static code characteristics. Ransomware, despite its obfuscation techniques, exhibits distinctive behavioural patterns during execution, such as abnormal file access, rapid file encryption, suspicious process creation, registry modifications, and anomalous system calls [10], [11], [15].

System logs provide a rich and reliable source of information for capturing such behavioural characteristics. Logs generated by operating systems and monitoring tools (e.g., Windows

Event Logs and Sysmon) record detailed information about process execution, file system activity, registry changes, and network connections. Several recent studies have demonstrated that system log-based analysis can effectively capture ransomware behaviour at early stages of execution, enabling timely detection before widespread damage occurs [10], [13], [14].

To enhance detection accuracy, machine learning techniques have been widely adopted in ransomware detection research. By extracting meaningful features from system logs, machine learning models can distinguish between benign and malicious behaviours with high accuracy [3], [8], [16]. Surveys and comparative studies highlight that classifiers such as Random Forests, Support Vector Machines, and deep learning models outperform traditional rule-based approaches, particularly in detecting previously unseen ransomware variants [5], [17], [18]. Furthermore, recent work emphasizes the importance of incremental and real-time learning to adapt to evolving ransomware behaviours [11], [12].

Although substantial progress has been made, existing research also identifies several challenges, including high false-positive rates, lack of standardized datasets, limited interpretability of models, and insufficient focus on log-based behavioural correlation [2], [7], [19]. Additionally, while numerous studies propose novel detection techniques, comparative and analytical reviews that synthesize recent findings on system log-based ransomware behaviour detection remain limited.

This paper presents a comprehensive review of recent research on ransomware behaviour detection using system logs. It analyzes state-of-the-art techniques, feature extraction strategies, and machine learning approaches reported in the literature. By synthesizing findings from recent studies, this review highlights current trends, identifies research gaps, and outlines future directions for effective ransomware detection. The insights provided aim to support researchers and practitioners in designing more robust and adaptive ransomware defense mechanisms.

The remainder of this paper is organized as follows. Section II reviews recent literature on ransomware detection and behavioural analysis. Section III discusses system log sources and feature extraction techniques reported in existing studies. Section IV presents a comparative analysis of detection approaches and identifies open challenges. Finally, Section V concludes the paper and outlines future research directions.

Related Work:

Ransomware detection has been an active research area in recent years, with studies proposing a wide range of techniques based on static, dynamic, and hybrid analysis. Early approaches primarily relied on signature-based detection, which uses known malware patterns to identify threats. However, several studies have highlighted the limitations of such methods in detecting modern ransomware due to obfuscation, polymorphism, and rapid variant evolution [1], [2], [6].

To overcome these limitations, researchers have increasingly focused on behaviour-based detection techniques. Behavioural approaches analyze the runtime activities of ransomware, such as file system operations, process behaviour, registry modifications, and memory usage. Surveys and systematic literature reviews emphasize that ransomware exhibits distinct behavioural characteristics during execution, which remain observable even when the

malware code is heavily obfuscated [4], [7]. These findings have motivated the shift toward dynamic and behaviour-based detection mechanisms.

A significant body of recent work has explored the use of system logs as a primary data source for ransomware detection. System logs, including Windows Event Logs and Sysmon logs, provide fine-grained visibility into system-level activities such as process creation, file access patterns, registry changes, and network connections. Several studies demonstrate that log-based behavioural analysis enables early-stage ransomware detection before the completion of encryption activities [10], [11], [13]. Incremental learning approaches using Sysmon logs have also been proposed to adapt detection models to evolving ransomware behaviours [11].

Machine learning techniques play a crucial role in log-based ransomware detection. Feature-based machine learning models have been widely adopted to classify benign and malicious behaviours using features extracted from system logs [3], [8]. Comparative studies report that traditional classifiers such as Random Forests, Support Vector Machines, and Decision Trees achieve high detection accuracy when trained on behavioural features derived from logs [16], [17]. More recently, deep learning approaches, including convolutional and recurrent neural networks, have been explored to capture temporal and sequential patterns in ransomware behaviour [12], [16].

Several review and survey papers provide comprehensive overviews of ransomware detection techniques and highlight current trends and challenges. These studies discuss issues such as dataset imbalance, lack of standardized benchmarks, high false positive rates, and limited generalization to unseen ransomware variants [2], [5], [7]. Additionally, some works extend the discussion to specialized environments, such as IoT and enterprise networks, emphasizing the need for adaptable and scalable detection mechanisms [9], [14].

Despite the extensive research on ransomware detection, existing literature reveals certain gaps. Many studies focus on proposing new detection models but provide limited comparative analysis across different system log sources and feature extraction strategies. Furthermore, there is a lack of consolidated reviews that specifically analyze ransomware behavior detection through system log analysis in a structured and comparative manner [19]. This motivates the need for a focused review that synthesizes recent research findings, compares methodologies, and identifies open challenges in log-based ransomware detection.

System Log–Based Ransomware Detection Approaches:

System log–based ransomware detection approaches focus on analyzing operating system–generated logs to identify malicious behavioural patterns during ransomware execution. Unlike static analysis techniques, log-based approaches capture runtime activities that reflect real system behaviour, making them effective against obfuscated and polymorphic ransomware variants. Recent research highlights system logs as a reliable and rich data source for detecting ransomware at early stages of infection [1], [7], [10].

System Log Sources For Ransomware Detection:

Various types of system logs have been utilized in ransomware detection research. Windows Event Logs are among the most commonly used sources, providing information related to

process execution, file access, authentication events, and system errors. These logs enable researchers to monitor abnormal system activities associated with ransomware attacks [13], [14].

Sysmon logs have gained significant attention in recent studies due to their detailed monitoring capabilities. Sysmon records low-level events such as process creation, file creation timestamps, registry modifications, and network connections. Several studies demonstrate that Sysmon logs provide enhanced visibility into ransomware behaviour and enable fine-grained behavioural analysis [10], [11]. Incremental learning systems based on Sysmon logs have also been proposed to adapt detection models to evolving ransomware techniques [11].

In addition to Windows and Sysmon logs, application logs and security logs have been explored to capture specific ransomware activities. These logs often complement system-level logs by providing contextual information related to application misuse, privilege escalation, and unauthorized access attempts [15], [19].

Behavioral Features Extracted From System Logs:

Feature extraction plays a critical role in log-based ransomware detection. Behavioural features are derived from system logs to represent ransomware activities in a machine-readable format. Commonly extracted features include file system behaviour such as rapid file modifications, abnormal file extension changes, and high-frequency write operations, which are strong indicators of encryption-based ransomware attacks [3], [8].

Process-related features are also widely used, including process creation rates, suspicious parent-child process relationships, and abnormal execution paths. Registry-related features, such as unauthorized registry key modifications and persistence mechanisms, further contribute to accurate ransomware identification [16], [18]. Network-related features, including unusual outbound connections and command-and-control communication patterns, have been incorporated in several studies to improve detection performance [14].

Recent surveys emphasize that combining multiple behavioural features from different log sources significantly enhances detection accuracy and robustness against evasion techniques [2], [6], [7].

Machine Learning Techniques For Log-Based Detection:

Machine learning techniques have been extensively applied to classify ransomware and benign activities using features extracted from system logs. Traditional machine learning models such as Random Forests, Support Vector Machines, Logistic Regression, and Decision Trees are widely used due to their effectiveness and interpretability [3], [17]. Comparative studies report that ensemble-based classifiers often outperform single-model approaches in terms of detection accuracy and false positive reduction [8], [16].

More recent research explores deep learning models, including convolutional neural networks (CNNs) and recurrent neural networks (RNNs), to capture temporal and sequential dependencies in ransomware behaviour [12], [16]. These models are particularly effective in

identifying complex behavioural patterns but often require large labeled datasets and higher computational resources.

Incremental and online learning approaches have also been proposed to address the dynamic nature of ransomware attacks. Such approaches enable detection systems to continuously update their models based on newly observed behaviours, improving resilience against novel ransomware variants [11], [5].

Summary of Log-Based Detection Approaches:

Overall, system log-based ransomware detection approaches demonstrate strong potential for early and accurate threat identification. By leveraging detailed runtime information and advanced machine learning techniques, these approaches overcome several limitations of traditional signature-based methods. However, challenges such as dataset availability, feature standardization, and real-time deployment remain open research issues, as highlighted in recent studies [2], [7], [19].

Comparative Analysis of Log-Based Ransomware Detection Approaches:

A comparative analysis of existing ransomware detection approaches provides valuable insights into the strengths, limitations, and research trends in system log-based detection. Recent studies differ in terms of the system log sources used, feature extraction strategies, machine learning techniques applied, and detection objectives. Analyzing these differences helps identify effective practices and open research gaps.

Most reviewed studies rely on Windows Event Logs and Sysmon logs as primary data sources due to their ability to capture detailed runtime activities. Sysmon-based approaches offer fine-grained visibility into process creation, file system behaviour, and registry modifications, enabling early detection of ransomware activities. However, such approaches often generate high volumes of logs, which introduces scalability and real-time processing challenges.

In terms of feature extraction, file system and process-level features are the most commonly used indicators of ransomware behaviour. These features are effective in detecting encryption-centric ransomware but may lead to false positives when legitimate applications exhibit similar behaviour. Several studies attempt to mitigate this issue by combining features from multiple log sources, resulting in improved detection robustness.

Machine learning techniques used in existing research range from traditional classifiers such as Random Forests and Support Vector Machines to advanced deep learning models. While traditional models are computationally efficient and easier to interpret, deep learning approaches demonstrate superior capability in capturing complex and temporal behavioural patterns. Nevertheless, deep learning models require large labeled datasets and higher computational resources, which limits their practical deployment.

Overall, the comparative analysis indicates that no single approach is universally optimal. Hybrid solutions that combine multiple log sources, carefully selected features, and adaptive

learning mechanisms appear to offer the most promising direction for effective ransomware detection.

Table I: **Comparative Analysis of System Log–Based Ransomware Detection Approaches:**

Study Focus	Log Source Used	Behavioural Features	Detection Technique	Key Observations
Behaviour-based ransomware detection	Windows Event Logs	File access frequency, process execution	Traditional ML classifiers	Effective for known behaviours but sensitive to false positives
Early ransomware detection	Sysmon Logs	File modifications, process creation	Machine learning models	Enables early detection with detailed behavioural visibility
Incremental learning based detection	Sysmon Logs	Temporal behaviour patterns	Online incremental learning	Adaptable to evolving ransomware variants
Feature-based ransomware classification	System and security logs	Registry changes, file extensions	Ensemble ML methods	Improved accuracy using combined features
Deep learning-based detection	System logs	Sequential behavioural features	CNN / RNN models	Captures complex patterns but requires large datasets
Enterprise-focused detection	Multi-source logs	Process, network, and file activity	Hybrid ML approaches	Scalable but computationally intensive

Challenges and Open Research Issues:

Despite significant advancements in ransomware detection using system logs, several challenges and open research issues remain. Addressing these challenges is essential for developing robust, scalable, and practical ransomware detection systems.

Limited Availability of Standardized Datasets:

One of the primary challenges in log-based ransomware detection is the lack of publicly available and standardized datasets. Most existing datasets are either outdated, limited in scope, or lack comprehensive system log information. This makes it difficult to fairly compare detection techniques and evaluate model generalization across different environments and ransomware families.

High False-Positive Rates:

Behaviour-based detection approaches often suffer from high false-positive rates, as benign applications may exhibit behaviour similar to ransomware, such as bulk file modifications or high disk activity. Distinguishing between legitimate system behaviour and malicious activity remains a critical challenge, particularly in enterprise environments with diverse workloads.

Evolving Ransomware Techniques:

Ransomware continues to evolve rapidly, adopting new evasion techniques such as delayed execution, fileless attacks, and living-off-the-land strategies. These techniques reduce the effectiveness of static behavioural patterns and require detection systems to continuously adapt to emerging attack strategies.

Feature Selection and Interpretability:

Selecting meaningful and discriminative features from large volumes of system logs is a complex task. Additionally, many machine learning and deep learning models lack interpretability, making it difficult for security analysts to understand detection decisions. Improving feature selection methods and enhancing model explainability remain open research issues.

Scalability and Real-Time Detection:

System logs are generated at a high volume and velocity, especially in large-scale environments. Processing and analyzing these logs in real time poses significant scalability challenges. Efficient log preprocessing, feature extraction, and low-latency detection mechanisms are required for practical deployment.

Generalization Across Platforms:

Most log-based ransomware detection studies focus on specific operating systems or configurations, limiting their applicability across heterogeneous environments. Developing platform-independent detection techniques that generalize across different operating systems and logging mechanisms remains an open research problem.

Conclusion and Future Work:

Ransomware continues to pose a significant threat to modern computing environments, necessitating effective and adaptive detection mechanisms. This paper presented a comprehensive review of recent research on ransomware behaviour detection using system logs. By analyzing existing literature, the study highlighted how system logs provide valuable insights into ransomware activities through detailed records of process execution, file system operations, registry modifications, and network behaviour. The review demonstrated that log-based behavioural analysis, combined with machine learning techniques, offers a promising alternative to traditional signature-based detection methods.

The surveyed studies reveal that system log-based approaches are particularly effective in identifying ransomware behaviour at early stages of execution, even in the presence of code obfuscation and polymorphism. However, the analysis also identified several challenges, including limited dataset availability, high false-positive rates, evolving ransomware strategies, and scalability concerns. These challenges indicate that while substantial progress has been made, further research is required to enhance the practicality and robustness of log-based ransomware detection systems.

Future research should focus on the development of standardized and comprehensive log-based datasets to facilitate fair comparison and evaluation of detection techniques. Additionally, integrating explainable machine learning models can improve the interpretability of detection decisions, aiding security analysts in understanding and responding to threats. Real-time and scalable detection frameworks capable of processing high-volume system logs are also essential for deployment in enterprise environments. Furthermore, exploring cross-platform and adaptive detection mechanisms can improve resilience against emerging and previously unseen ransomware variants. Addressing these directions will contribute to more effective and resilient ransomware defense solutions.

References:

1. M. Rele et al., "Exploring ransomware detection based on artificial intelligence and machine learning," *Procedia Computer Science*, 2025.
2. E. Kritika, "A comprehensive literature review on ransomware detection techniques," 2024.
3. J. Lee et al., "A machine learning-based ransomware detection method for manipulated ransomware entropy," *Sensors*, 2025.
4. "Machine learning approaches to ransomware detection: A comprehensive review," *Int. Journal of Safety and Security Engineering*, 2024.
5. J. I. Zapata Sandoval et al., "Ransomware detection with machine learning: Techniques, challenges, and future directions," 2025.
6. A. Alraizza, "Ransomware detection using machine learning: A survey," *Big Data and Cognitive Computing*, vol. 7, no. 3, 2023.
7. "A systematic literature review of ransomware detection methods," *ResearchGate Preprint*, 2025.
8. K. Zhang et al., "Enhanced ransomware detection using feature-based machine learning," *Journal of Big Data Analytics*, 2025.
9. P. Yan et al., "A comprehensive review of ransomware attacks targeting IoT devices," 2025.
10. "iCNN-LSTM: A batch-based incremental ransomware detection system using Sysmon," *arXiv preprint*, 2025.
11. "A Sysmon incremental learning system for ransomware detection," *arXiv preprint*, 2025.
12. J. Ferdous et al., "Image-based dynamic feature ransomware detection using transfer learning," *Scientific Reports*, 2025.
13. "Early ransomware detection using behavioural analytics," *IJIREM*, 2025.
14. B. Mondal et al., "Using machine learning for early detection of ransomware threats in enterprise networks," *Saudi Journal of Engineering and Technology*, 2025.
15. "Behavioural analysis techniques for ransomware detection in endpoint security," *ResearchGate*, 2025.
16. A. Hussain et al., "Enhancing ransomware detection using deep learning," *PMC Article*, 2024.
17. Rafi et al., "Comparative insights in machine learning models for ransomware detection," *European Journal of Artificial Intelligence*, 2025.
18. M. Aljabri et al., "Machine learning-based ransomware detection using memory dump analysis," 2024.
19. "Ransomware detection and prevention techniques," *Journal of CRD*, 2025.
20. Q. Chen et al., "Automated ransomware behaviour analysis and pattern extraction," 2019.

13. Face Recognition Attendance System

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Abstract: Currently, Colleges and Schools Recorded and marked Attendance Manually using paper sheets and excel sheets. However, This Process of marked Attendance is time-consuming and carries the risk of proxy attendance by the students. For Solve This Above Problem We Will Use Face Recognition Attendance System for Attendance Marked. In Which the Face Recognition Attendance System will help us in the following ways:

- Provide an easiest way to maintain attendance.
- Automate the attendance work of an organization.
- Provide 92% Accuracy in face detection.
- Can Import can Export attendance from system.
- It will be an secure system protected by ID and Password.

Keywords: Face Recognition Attendance System, Colleges and school, automate attendance

Introduction :

Background of the Study :

Attendance is crucial for both Students and teachers in schools and colleges, as attendance marking helps determine which students have attended class or have not. So, we will make this system to ensure attendance is recorded in a short time

Call the name and enrollment roll no by the faculties in colleges to mark their attendance is not disadvantages of a waste of time but also consumes energy as students cannot hear loud noises. Therefore, these systems will solve all our problems by automatic attendance markings.

There are some automated attendances making system are available in the institutions for Use. One Such system is Biometric technology and the RFID system. Using Traditional methods, it fails to meet the time constraints. Therefore, using this face recognition attendance system can easily record students' attendance and save time.

This system removes need for student Identify procedures, such as calling out student's names by teacher , sure for new student IDs, and many other procedures, which not only disrupt the learning process but also cause significant stress for students during exam sessions. Furthermore, by eliminating the need to register students in a database for identification, enrollment can be done instantly through the appropriate course of interests.

Need for proper System:

There is a need to develop a real time system for student attendance which means the identification process should be completed within the stipulated time frame so that there is no lag and the features extracted from the facial images which indicate the identity of the students should be consistent despite changes in background light, posture, expressions, High accuracy and quick calculation time will be the six main points for performance managing.

Objective:

The main aim of this system is to create a face recognition attendance system.

The required achievements to fulfill the objectives are:

- Detecting Faces from Video Frames.
- Extracting useful features from the captured faces and checking whether they are true or false.
- Classifying the features to identify the detected faces.
- Recording the attendance of the detected students in the database.

Review of Literature:

Review of Existing Attendance Systems:

1.Existing System: Some Projects analysis member describes The Drawbacks of RFID card system and fingerprint system. The simplicity of the RFID card system makes it effective but as long as they have their friends Id card in which the user usually assists them in checking in. Although the fingerprint system is effective, it is inefficient because it requires users to complete the verification process one at a time, which consumes time. However, compared to the iris, the human face is constantly exposed as well as contains less information for recognized faces. Mainly an iris detection system may violate the user's privacy in the above the voice recognition is accessible, it is not as accurate as alternative methods. Therefore, it is suggested that systems recognize faces.

2 Proposed System:

The solution of that complex problem is proposed in a way that marking of attendance done by face recognition instead of face Biometrics or any other system like RFID. For that face recognition problem.

3.Feasibility Study:

A feasibility study shows the technical issues and project successful determines in which system works properly or not.

4.Technical Feasibility:

The technical analysis of this project face attendance system project for developer he should have knowledge of python, machine learning MySQL and the project will run on all system which fulfil mentioned software and hardware requirements.

5 Economic Feasibility:

The economic analysis for the project face attendance system is such that The Project Will Be Free of Cost for Run to Everyone There Is No Need to Buy Anything on System for Running These Project.

6. Operational Feasibility:

Operational feasibility of project is such that following operations performed:

- Data sample collected.
- Classifier trained.
- Face recognized.
- Attendance marked.

Problem Statement:

Colleges and Schools Recorded and marked Attendance Manually using paper sheets and excel sheets. However, This Process of marked Attendance is time-consuming and carries the risk of proxy attendance by the students.

Calling out student's name, conforming new students IDs and many other procedures that not only interrupt with learning but also heavily stress out students during exams are replaced by this system. Furthermore, by eliminating the need to register students in a database for identification, enrollment can be done instantly through the appropriate course of interests.

Requirements Analysis:

1 Data Requirements:

Data requirements for the implementation of project facial recognition attendance system is such that we need sample facial data from the user and prior knowledge of ML algorithms used in implementation of project.

2 functional Requirements:

Functional Requirements System provides functional requirement that describes activities and services that must provide.

- Login Window for Security
- Student Registration system
- Take Photo Sample
- Training Data
- Attendance Management
- Face Recognition
- Help desk
- Exit

3 Non-Functional Requirement:

Security:

SSL stands for Secured socket layer. It is applied by the system for all transactions which involve any private client details. After a certain period of absence, the system has to log out for each client and students.

Reliability:

The database baking process is the systems major pillar of reliability.

Availability:

The System must always be accessible to users through a web browser.

Hardware Requirements:

- Hardware requirements consists of an intel Core i5 processor along with an 80GB hard drive
- Ram: 256 mb

- 512 kb of cache

Software specification:

Software Needs for Face Recognition Attendance System Project:

- Vs Code 2019 or later ;
- Python 3.5 or later ;
- Windows 8 or later;
- and the most recent versions of all libraries .

Methodology

Haar cascade Algorithm:

This Algorithm is a fast an machine learning method for real time object detection. It is famously used for face detection that relies on the simple, trained features to quickly identify patterns of the intensity change.

Python open CV:

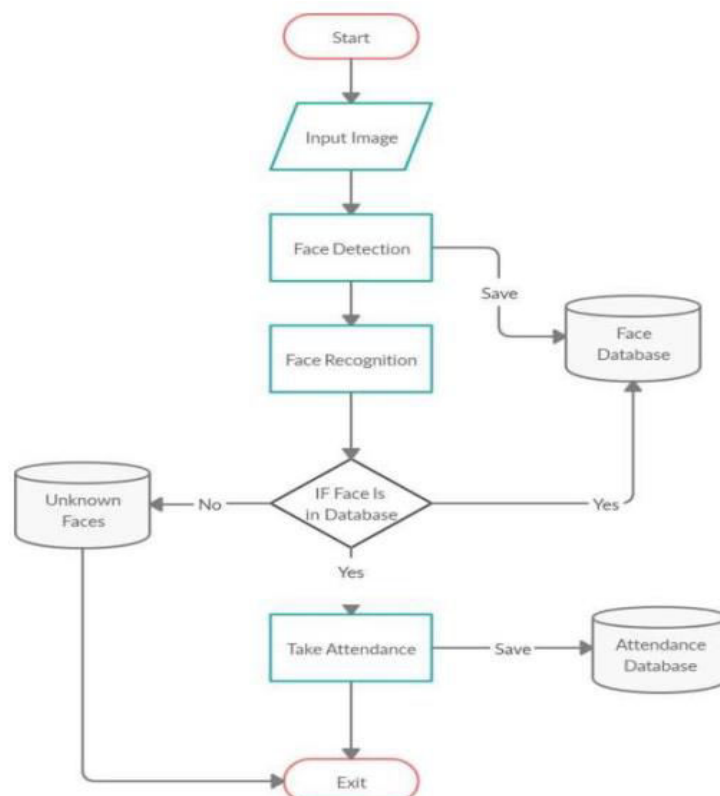
Open CV is a python library. With the help of the python library Open cv, you can carry out computer vision and image processing tasks. Mainly Computer Vision allows the computer for perform the same tasks as a human with same efficiency. The two tasks are Object Classification and Object Identification in which we train a model on a dataset of particular objects and identify the instance of an object.

Python Tkinter:

The common and quick interface for the TK GUI toolkit is Python. It is the most commonly used method in python GUI application development . Python Tkinter is faster easiest way to create the GUI applications.

Working / Design Phase:

Flow chart :



Here the, Explanation of Face Recognition Attendance System Diagram :

This diagram shows the complete working flow of a Face Recognition–based Attendance System, starting from capturing an image to storing attendance automatically.

1. Start

The system begins execution when:

- The application is launched
- The camera is turned ON

2. Input Image

- This System will Detect face by the camera and CCTV as a input then,

3. Face Detection

- The system scans the input image to detect faces.
- Face detection algorithms (Haar Cascade) is used.

4. Face Recognition

- Now the Detected Face will be compared with faces who stored in the Face Database.
- Feature extraction techniques (LBPH, Eigenfaces, or Deep Learning) are applied.

Purpose: To identify whose face, it is.

5. Decision: “IF Face Is in Database”

This is a **decision-making step**.

Two Possible Outcomes

Case 1: YES (Face Found in Database)

- The system recognizes the person.
- The person’s ID / Name is retrieved.
- The process moves forward to attendance marking.

Case 2: NO (Face Not Found in Database)

- The face is considered unknown.
- The image will be stored in the Unknown Faces database.
- Attendance is not marked.

6. Take Attendance

If the face is recognized:

- The system records:
 - Name / ID
 - Date
 - Time
- Attendance is marked automatically.

7. Attendance Database

- Attendance data is saved permanently.
- Used for:
 - Reports
 - Monthly summaries

- Admin access

Example stored data:

Name: Dipak Patel

Date: 12-Jan-2026

Time: 10:02 AM

Status: Present

8. Exit

- The system completes the process.
- It waits for the next image or stops execution.

This marks the end of one complete attendance cycle.

Result Analysis:

Under the result analysis phase, we will see the comparative study how our system is better than existing systems:

How Haar Cascade Algorithm Better Than other Algorithms:

Lightweight: Haar cascades are lightweight As compared to other face detection algorithms like LBP or CNN.

Extremely fast: Haar cascades are Extremely fast compared to other Object detection models such as yolo.

Easy to implement: Haar cascade model is very easy to implement as compared to CNN and LBP.

Required less computing power: When we use Haar cascade model for face detection less computation power required.

How Our System Is Better Than Existing Systems:

Gui: In this model we provided much better graphical user interface than other systems in market here you will find separate windows for each and every functionality which make it very reliable to use.

Accuracy: In our system we have used Haar cascade model for face detection which provides more accuracy than other systems in market like lbp and yolo.

Security: In our system we used login system for authentication which makes it completely secure.

Speed: Our system is faster than other in market because we used correct models and algorithms like we used Bhp for data training.

Limitations :

- Facial Recognition Technology may be limited by data processing and storage.
- The reliability of facial recognition can be affected by various face angles.

Future Scope :

We are preparing to design two module systems. The first will be face detector that is mobile component in which is basically camera app will capture students faces and stored in the camera app storage using face detection Algo of computes vision.

The second module will be a desktop application that does face recognition of the captured faces images in the one file and marks the student register and stored the results in the database of this system for future analysis.

Conclusion :

Face recognition systems are a type of facial image processing applications and their importance as a field of study has been growing lately. Mainly Crime prevention, video surveillance, person verification, and other security tasks are all ways that the system can be used. The face recognition system implementation can be part of universities. The face recognition-based attendance system has been designed for the purpose of reducing the errors that occur in the manual attendance taking system. This system is secure enough reliable and available for use .

References :

1. (IJRASET), 2024. [IJRASET](#)
2. Bhushan Laddha, Devanshu Rathi et al., "Face Recognition Based Attendance System," IJRASET, 2023. [IJRASET](#)
3. Smitha, Pavithra S. Hegde & Afshin, "Face Recognition Based Attendance Management System," IJERT, 2020. [IJERT](#)
4. Ghalib Al-Muhaidhri & Javeed Hussain, "Smart Attendance System using Face Recognition," IJERT, 2019. [IJERT](#)
5. Ashwin Rao, "AttenFace: A Real Time Attendance System using Face Recognition," arXiv preprint, 2022. [arXiv](#)
6. Touzene Abderraouf et al., "An Embedded Intelligent System for Attendance Monitoring," arXiv preprint, 2024. [arXiv](#)
7. Shailesh Arya, Hrithik Mesariya & Vishal Parekh, "Smart Attendance System using CNN," arXiv preprint, 2020.

14. Scope of Research in Diabetes and a Case Study using Lazy Predict on the Pima Dataset

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Abstract: A significant symptom of Diabetes is high blood glucose levels. Chronic Diabetes causes organ failure or premature death. Prediction of diabetes at an earlier stage may result in better treatment. Numerous Machine Learning (ML) and Deep Learning (DL) methods have been applied to prediction of Diabetes. This study emphasized the various research domains associated with diabetes. This study also provides a comprehensive overview of ML and DL applications across diverse research domains within diabetes. Along with this, we have also conducted experimentations on the Pima Diabetes Dataset using python Lazy predict and four ML models to identify which model performs better. The performed experimentation will also assist researchers in developing a model capable of early diabetes prediction by utilizing ML classification algorithms.

Keywords: Diabetes Mellitus, Diabetes classification, Diabetes Prediction, Diabetes Complications, Blood Glucose level Prediction, T2DM, T1DM, GDM

Introduction

DIABETES is a metabolic disorder characterized by fluctuating blood glucose levels. This condition arises from either insufficient insulin production (Type 1 diabetes- T1DM), the body's inability to effectively use the insulin it produces (Type 2 diabetes- T2DM), or elevated blood glucose levels that develop during pregnancy in individuals not previously diagnosed with diabetes (Gestational Diabetes Mellitus-GDM). This classical classification of diabetes was proposed by the American Diabetes Association (ADA) in 1997 [1]. This form of diabetes now affects approximately one in ten people, representing a growing epidemic that poses a substantial challenge to healthcare systems worldwide. Approximately 537 million adults suffer from diabetes and this number may be projected to skyrocket in the coming years, potentially reaching 643 million by 2030 and a staggering 783 million by 2045 [2]. If diabetes is untreated it can lead to long term complications and even death. Early prediction of diabetes must be required for saving human life. Artificial Intelligence (AI) has been growing considerably over the last ten years and ML is probably the most popular branch of AI. ML algorithms can be built in a manner that they have the power to classify and even predict diseases. Predictive analytics aims to identify patterns and insights that can be used to forecast future outcomes. Nowadays, the increased complexity of the Diabetes problems inspired many researchers to explore DL algorithms such as Convolution Neural Network (CNN), Recurrent Neural Network (RNN), Long Short Term Memory (LSTM), combinational model of DL. In this paper we performed an experiment on Pima Indian Diabetes (PID) Dataset considering two different preprocessing approaches using the Lazy classifier.

This paper is designed in the sections as: section two presents the related work, section three introduces Diabetes datasets, section four includes Experimentation, section five contains the conclusion.

I. Related work

ML is a branch of AI that creates machines based on learning capability and improves the performances from getting knowledge from experiences without being explicitly programmed. There exist two types of predictive models. Classification models assign data points to classes, whereas regression models predict continuous values. Most probably used predictive models are decision trees (DT), linear regression (LiR), logistic regression (LR), neural networks. The classifiers such as time series, ensemble, factor analysis, support vector machines (SVM), random forest (RF) and clustering methods are also useful for predictions. Researchers have made incredible strides in predicting diabetes. But there's more scope for engineers to contribute beyond simply classifying the disease. Think about it: we can predict who might develop diabetes, anticipate blood glucose levels, and even forecast future risks. We can also create better ways to monitor the disease, like remote systems using wearable's and smart devices. Imagine user-friendly, affordable applications which help diabetics to manage diabetes or even diagnose it themselves. And we can track how the disease progresses and see how well different treatments work to avoid further complications. Figure 1 visually summarizes the diverse and promising research avenues within diabetes, which are further explored in the subsequent review of researcher contributions.

A. Diabetes Classification

Researchers have developed various models for diabetes classification, encompassing types such as Type 1 Diabetes Mellitus (T1DM), Type 2 Diabetes Mellitus (T2DM), Gestational Diabetes Mellitus (GDM), and Pre diabetes. Several studies have explored the effectiveness of different ML algorithms. For instance, Gupta et al. [3] utilized Naive Bayes (NB) and Support Vector Machine (SVM) on the Pima Indians Diabetes (PID) dataset, finding SVM to outperform NB, though their work lacked comprehensive evaluation against state-of-the-art methods and did not report accuracy. Mariwan A. [4] focused on T2DM classification using Decision Trees (DT), AdaBoost (AB), Gradient Boosting (GB), and Extra Tree (ET) classifiers, with the ET classifier demonstrating superior performance (AUC of 0.96 for PID and 0.99 for BRFS datasets). Similarly, Lokesh M. et al. [5] compared various ML techniques including Random Forest (RF), DT, K-Nearest Neighbors (KNN), GB, SVM, and ET, along with Artificial Neural Networks (ANN), on the PID dataset, where the ET classifier achieved the highest accuracy of 81.16% and an AUC of 81%. Furthermore, Metaphor P. et al. [6] compared DT, RF, SVM, and KNN for diabetic classification, and through hyper parameter tuning and interaction terms, their proposed RF classifier achieved the best accuracy of 97.5%.

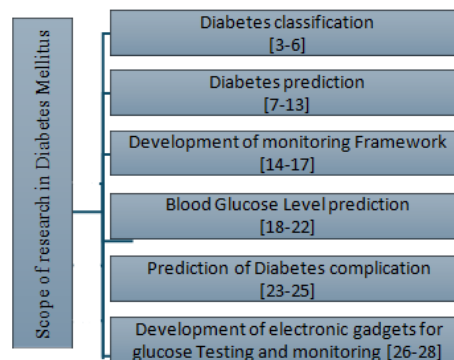


Fig.1. Scope of research in the field of DM to Researchers

B. Diabetes Prediction

ML breakthroughs are significantly aiding in the early detection and understanding of diabetes by uncovering hidden patterns within vast healthcare datasets, addressing the challenge of extracting beneficial knowledge from this abundance of information. Researchers have proposed various predictive models to this end. Nitesh Pradhana et al. [7] leveraged Artificial Neural Networks (ANN) on the Pima Indians Diabetes (PID) dataset, achieving a high accuracy of 85.09% for diabetes detection. Han Wu [8] developed a unique model combining K-means and Logistic Regression (LR) algorithms, demonstrating a 3.04% increase in prediction accuracy compared to existing models for Type 2 Diabetes Mellitus (T2DM) prediction. Jobeda Jamal Khanam et al. [9] explored seven ML algorithms, with an LR-SVM combinational model and a two hidden layers-NN achieving superior performance and 88.6% accuracy, respectively. Swapna G. and their group [10] created a predictive model integrating Convolutional Neural Networks (CNN), Long Short-Term Memory (LSTM), and an ensemble model to process Heart Rate Variability (HRV) data for feature detection, followed by SVM for classification. Ismail et al. [11] conducted a comparative analysis of 35 ML techniques and nine feature selection algorithms across three real-world diabetes datasets to assess their efficacy in identifying at-risk individuals. Furthermore, Huma Naz et al. [12] applied ML and Deep Learning (DL) algorithms to the PID dataset, reporting that functional classifiers like ANN, Naive Bayes (NB), Decision Trees (DT), and DL achieved results between 90-98%. Lastly, Ali Mohebbi et al. [13] utilized a DL approach with data from Continuous Glucose Monitoring (CGM) devices for T2DM prediction, achieving 77.50% accuracy with CNN.

C. Development of Monitoring Frameworks

The integration of portable monitoring devices, IoT, sensors, and wireless technology is revolutionizing diabetes care by enhancing patients' quality of life, reducing hospitalizations, and lowering costs. Researchers are actively developing systems that leverage these technologies alongside ML for improved diagnostics and predictions. For instance, Alfian et al. [14] introduced a personalized healthcare monitoring system that combines personal data with real-time sensor data from Bluetooth Low Energy (BLE) devices, using Apache Kafka and MongoDB for data processing. This system employs Multi-Layer Perceptron (MLP) for diabetes patient classification and Long Short-Term Memory (LSTM) for blood glucose prediction. Similarly, Jayroop Ramesh et al. [15] designed a cloud-based system to merge patient information from personal health devices and wearables, demonstrating that an SVM-RBF model achieved a good accuracy of 83.20%. Kabyshev et al. [16] implemented a client-server system for diabetes mellitus that utilizes cluster analysis for patient subgroup identification and Random Forest (RF) for diabetes prediction, with data collected via an iOS application integrated with Apple's HealthKit, ResearchKit, and CareKit. Lastly, Rghioui et al. [17] developed an intelligent diagnosis system based on patient information and ML, testing algorithms such as Naive Bayes (NB), J48, ZeroR, and OneR, and finding that the Sequential Minimal Optimization (SMO) algorithm yielded the highest results.

D. Blood Glucose Level Prediction

The emergence of Continuous Glucose Monitoring (CGM) and AI-driven closed-loop technology has opened new avenues for predicting blood glucose (BG) levels, enabling

patients to proactively manage their diabetes. Researchers are exploring various time-series and deep learning models for this purpose. Sofia Goel et al. [18] reviewed models like ARIMA, LSTM, and Bi-LSTM, with their experiments on the UVA/Padova dataset demonstrating the superior performance of the Bi-LSTM model for BG level prediction. Providing an early warning system, Hrushikesh N. Mhaskar et al. [19] developed a deep learning model capable of predicting patient glucose levels 30 minutes in advance, empowering patients to take timely preventative measures. Wonju Seo et al. [20] created a personalized BG level predictive model using a Convolutional Neural Network (CNN), utilizing CGM datasets from 1052 professional sessions to predict future BG levels for Type 1 Diabetes Mellitus (T1DM), Type 2 Diabetes Mellitus (T2DM), and Gestational Diabetes Mellitus (GDM) across different prediction horizons. Similarly, Kim et al. [21] built a personalized glucose prediction model for hospitalized T2DM patients using deep learning, comparing various Recurrent Neural Network (RNN) architectures (simple RNN, GRU, and LSTM) with Dexcom G5 CGM data, and found a simple GRU model to perform well. Finally, Bhimireddy et al. [22] developed individual models for T1DM patients' BG level prediction, including LSTM, BiLSTM, Convolutional LSTMs, TCN, and sequence-to-sequence models using CGM data, with their experiments indicating that the sequence-to-sequence BiLSTM model outperformed others.

E. Prediction of Diabetes Complications

Beyond just predicting the onset of diabetes, advanced data analysis and predictive techniques are increasingly being used to identify diabetes risk factors and forecast potential complications. Zhou and Myrzashova [23] developed a model that goes beyond a simple diagnosis, predicting not only the likelihood of diabetes but also anticipating the potential future types of the disease a patient might develop. Expanding on this, Yazan Jian et al. [24] created ML models to predict eight common diabetes complications, including metabolic syndrome, dyslipidemia, neuropathy, nephropathy, diabetic foot, hypertension, obesity, and retinopathy. Their models achieved a high accuracy of 97.8% on a comprehensive dataset. Furthermore, Mathieu Ravaut et al. [25] developed a ML model specifically designed to predict the three-year risk of adverse outcomes from diabetes complications, offering a valuable tool for health planners and decision-makers to more effectively allocate healthcare resources for diabetes management.

F. Development of Electronics Gadgets for glucose Testing & monitoring

Advancements in diabetes management are being driven by the evolution of glucose testing and monitoring devices, which are becoming increasingly accurate, comfortable, and compact. Notably, non-invasive glucose monitoring is a key area of development. Ilesanmi Daniyan et al. [26] developed a low-cost, non-invasive device that measures glucose levels through the earlobe using Near Infrared (NIR) light at 1550nm, displaying readings on an LCD and transmitting them to an Android app via Bluetooth; testing confirmed a direct correlation between glucose and the device's output. Similarly, Maria Valero et al. [27] presented a pilot test of a non-invasive glucose monitoring prototype also utilizing NIR spectroscopy, demonstrating excellent clinical accuracy in human blood and serum tests. Further, Paful Pai's team [28] successfully developed a portable, non-invasive glucose monitoring system employing photoacoustic spectroscopy and FPGA processing, with cloud connectivity for remote monitoring, enhancing diabetes management. Overall, ML and DL are profoundly impacting diabetes care through advanced classification, precise blood

glucose and complication prediction, personalized monitoring, and the development of non-invasive testing methods. These technological strides promise more accurate diagnoses, earlier interventions, and ultimately, improved patient outcomes, underscoring the critical importance of continued research in this field.

II. Experimentation

In our study, we used the Python Lazypredict package to predict outcomes on the PID Dataset, comparing numerous ML algorithms. We explored two distinct approaches based on how missing feature values were handled:

Approach 1: All missing values across all features were imputed with their respective means.

Approach 2: All missing values were imputed with their means, with the sole exception of the "pregnancies" feature, where zero pregnancies were considered a valid and distinct possibility.

A. Data Collection

We analyzed health data from 768 female patients, all aged 20 or older, from a population near Phoenix, Arizona, USA, using the Pima Indians Diabetes (PID) dataset from the UCI ML Repository [29]. For each patient, we examined eight independent factors: pregnancy history, Body Mass Index (BMI), insulin level, age, glucose, diastolic blood pressure, triceps skin fold thickness, and a diabetes pedigree function. Our objective was to determine if these factors could help predict whether a patient was diabetic. In this dataset, 268 patients had a diabetes diagnosis, while 500 did not.

B. Data preprocessing

Our data preparation involved two crucial steps: handling missing values and normalizing the data. Missing values in each attribute were imputed using their respective means. Due to the varying scales of attributes within the PID dataset, we normalized all values to a range between 0 and 1. After these preprocessing steps, the dataset was ready and subsequently split into training and testing sets.

C. Implementation and Results

Selecting the optimal ML model can be a daunting and time-consuming task. To streamline this process, our study utilized Lazypredict, an open-source Python package developed by Shankar Rao Pandala. Lazypredict automates baseline model selection by efficiently training and evaluating numerous classification or regression models with minimal code, significantly accelerating the model shortlisting phase for subsequent optimization.

For this experiment we used the Lazy Classifier. We used accuracy, precision, F1-score, and recall to analyze our results. These provide us with insights into how effectively our model worked. The table 1 also makes a very good comparison among varied ML models and two distinct approaches for a diabetes-related classification task. Approach 2 improves overall implying the worthiness of changes applied. That would mean changes undertaken in approach 2 are better. So, researchers can also consider approach 2 for their future works

while working on the Pima dataset. It turned out that AdaBoost was the top performer, beating all the other algorithms Lazypredict offered.

To get a closer look, we pulled out a comparison of four classifiers—AB, RF, GNB, and LR—after replacing zero values in the pregnancy parameter with the mean. It clearly shows improved performance, especially for LR and RF, which reached an accuracy of 78.65%, with LR achieving the highest precision (78.82%) and F1 score (74.80%). This preprocessing step appears to enhance the quality of data and thus the classification results. Both figure 3 and table1 together highlight the positive effect of preprocessing, particularly replacing invalid zero values in critical features like 'pregnancy'. It enhances the predictive performance of models, with notable gains in accuracy, precision, recall, and F1 score.

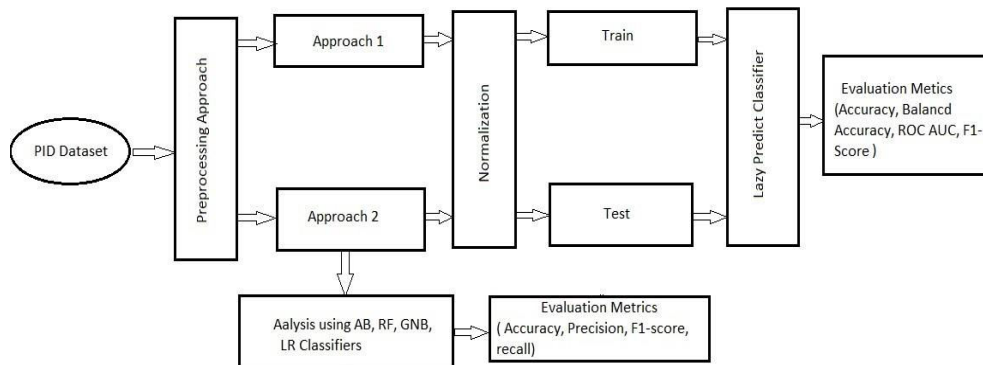


Fig. 2 : Machine Learning Workflow for Diabetes Prediction on PID Dataset

Table I: Performance Measures Of Lazy Classifier

Classifiers	Approach 1 Results					Approach 2 Results				
	Accuracy	Balanced Accuracy	ROC AUC	F1 Score	Time Taken	Accuracy	Balanced Accuracy	ROC AUC	F1 Score	Time Taken
AdaBoost	0.76	0.72	0.72	0.76	0.01	0.81	0.78	0.78	0.81	0.09
Random Forest	0.74	0.71	0.71	0.74	0.01	0.81	0.77	0.77	0.80	0.17
Extra Trees	0.74	0.71	0.71	0.74	0.01	0.80	0.77	0.77	0.79	0.13
Calibrated CV	0.73	0.69	0.69	0.73	0.01	0.82	0.78	0.78	0.81	0.1
Linear SVC	0.73	0.69	0.69	0.73	0.01	0.82	0.78	0.78	0.81	0.04
Nearest Centroid	0.73	0.72	0.72	0.73	0.01	0.78	0.79	0.79	0.78	0.01
Logistic Regression	0.73	0.69	0.69	0.73	0.01	0.82	0.78	0.78	0.81	0.02
Ridge Classifier CV	0.73	0.69	0.69	0.73	0.01	0.82	0.78	0.78	0.81	0.01
Linear Discriminant Analysis	0.73	0.69	0.69	0.73	0.01	0.81	0.78	0.78	0.81	0.01
Ridge Classifier	0.73	0.69	0.69	0.73	0.01	0.81	0.77	0.77	0.81	0.02
NuSVC	0.76	0.72	0.72	0.75	0.01	0.78	0.74	0.74	0.77	0.03
Gaussian NB	0.73	0.70	0.70	0.73	0.01	0.79	0.78	0.78	0.79	0.01
SVC	0.76	0.71	0.71	0.75	0.01	0.79	0.74	0.74	0.78	0.02
LGBM Classifier	0.73	0.71	0.71	0.73	0.01	0.76	0.73	0.73	0.76	0.05
XGB Classifier	0.74	0.72	0.72	0.74	0.01	0.73	0.70	0.70	0.73	0.07
K neighbors	0.74	0.72	0.72	0.74	0.01	0.75	0.70	0.70	0.74	0.01
Quadratic Discriminant Analysis	0.73	0.69	0.69	0.72	0.01	0.76	0.72	0.72	0.75	0.01
Bagging	0.73	0.69	0.69	0.72	0.01	0.75	0.71	0.71	0.74	0.04
Bernoulli NB	0.68	0.65	0.65	0.68	0.01	0.78	0.74	0.74	0.77	0.01
Label Spreading	0.69	0.67	0.67	0.69	0.01	0.72	0.71	0.71	0.72	0.03
Label Propagation	0.69	0.66	0.66	0.69	0.01	0.72	0.71	0.71	0.72	0.02
SGD	0.75	0.72	0.72	0.75	0.01	0.65	0.64	0.64	0.66	0.01
Passive Aggressive	0.71	0.73	0.73	0.72	0.01	0.7	0.62	0.62	0.67	0.01

Decision Tree	0.70	0.68	0.68	0.70	0.01	0.67	0.65	0.65	0.67	0.02
Extra Tree	0.69	0.66	0.66	0.69	0.01	0.68	0.66	0.66	0.68	0.01
Perceptron	0.65	0.58	0.58	0.63	0.01	0.71	0.73	0.73	0.72	0.01

The bar graph offers a focused view on key models, while the table confirms this trend across a broad range of algorithms. Thus, careful data cleaning is shown to be a vital step in improving model outcomes for medical datasets.

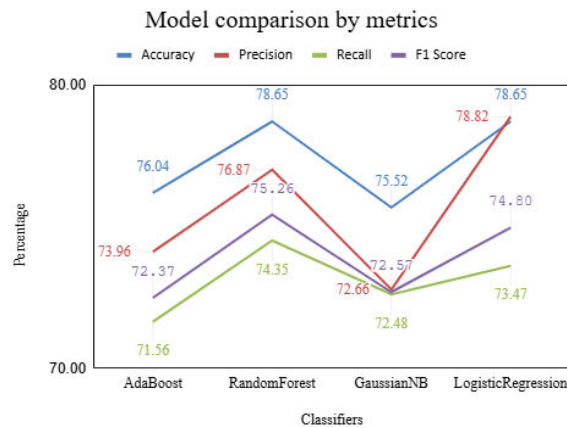


Fig.3: Model comparison results for Pima Dataset

Conclusion

Our paper briefed about the scope of the research areas related in the field of Diabetes Mellitus to Researchers and the enormous work done by researchers not in the field of DM classification but also in the field of DM prediction, Development Monitoring Frameworks and many more. In addition to this, it is found that Lazy predict can be a handy tool for selecting the best algorithm among all ML models and is also most suitable for predicting response variables before testing against the hyperparameters.

Our experiments collectively demonstrate the significant impact of preprocessing on model performance, specifically through the replacement of invalid zero values in crucial features like pregnancy in the Pima Indians Diabetes dataset. Therefore, the findings strongly emphasize that effective data preprocessing is essential for achieving reliable and accurate predictions, particularly in sensitive domains such as healthcare and medical diagnostics.

Conflicts of Interest

The authors declare that they have no conflicts of interest.

References

1. American Diabetes Association. Diagnosis and classification of diabetes mellitus. *Diabetes Care*. 2014;37 Suppl 1:S81–S90.
2. International Diabetes Federation. *IDF Diabetes Atlas*, 10th edn. Brussels, Belgium: 2021. Available at: <https://www.diabetesatlas.org>.
3. Gupta S., Verma H. K., Bhardwaj D. *Operations Management and Systems Engineering*. Singapore: Springer; 2021. Classification of diabetes using Naïve Bayes and support vector machine as a technique; pp. 365–376.

4. Hama Saeed M.A. Diabetes type 2 classification using machine learning algorithms with up-sampling technique. *Journal of Electrical Systems and InfTechnol* 10, 8 (2023). <https://doi.org/10.1186/s43067-023-00074-5>.
5. Malviya, L., Mal, S., Lalwani, P., Chadha, J.S. (2021). Diabetes Classification Using Machine Learning and Deep Learning Models. In: Bajpai, M.K., Kumar Singh, K., Giakos, G. (eds) *Machine Vision and Augmented Intelligence—Theory and Applications. Lecture Notes in Electrical Engineering*, vol 796. Springer, Singapore. https://doi.org/10.1007/978-981-16-5078-9_40.
6. Phongying, Methaporn, and SasiprapaHiriote. 2023. "Diabetes Classification Using Machine Learning Techniques" *Computation* 11, no. 5: 96. <https://doi.org/10.3390/computation11050096>.
7. Nitesh, P., Geeta, R., Vijaypal, S.D., Ramesh, C.P.: Diabetes prediction using artificial neural network. In: *Deep Learning Techniques for Biomedical and Health Informatics*, pp. 327–339. Elsevier (2020)
8. Han Wu, Shengqi Yang, Zhangqin Huang, Jian He, Xiaoyi Wang: Type 2 diabetes mellitus prediction model based on data mining, *Informatics in Medicine Unlocked*, Volume 10, 2018, Pages 100-107, ISSN 2352-9148, <https://doi.org/10.1016/j.imu.2017.12.006>.
9. Khanam JJ, Foo SY. A comparison of machine learning algorithms for diabetes prediction. *ICT Express*. 2021; 7:432–9.
10. Swapna G, Soman K.P., Vinayakumar R," Automated detection of diabetes using CNN and CNN- LSTM network and heart rate signals", *Procedia Comput. Sci.* 2018, 132, 1253–1262.
11. Ismail, L., Materwala, H., Tayefi, M. et al. Type 2 Diabetes with Artificial Intelligence Machine Learning: Methods and Evaluation. *Arch Computat Methods Eng* 29, 313–333 (2022). <https://doi.org/10.1007/s11831-021-09582-x>.
12. Naz H, Ahuja S. Deep learning approach for diabetes prediction using PIMA Indian dataset. *J Diabetes MetabDisord*. 2020 Apr 14;19(1):391-403. doi: 10.1007/s40200-020-00520-5. PMID: 32550190; PMCID: PMC7270283.
13. Mohebbi A, Aradottir TB, Johansen AR, Bengtsson H, Fraccaro M, Morup M. A deep learning approach to adherence detection for type 2 diabetics. *AnnuIntConf IEEE Eng Med Biol Soc.* 2017 Jul;2017:2896-2899. doi: 10.1109/EMBC.2017.8037462. PMID: 29060503.
14. Ganjar Alfian , Muhammad Syafrudin Muhammad Fazalljaz , M. Alex Syaekhoni , Norma Latif Fitriyani and Jongtae Rhee , " A Personalized Healthcare Monitoring System for Diabetic Patients by Utilizing BLE-Based Sensors and Real-Time Data Processing", *Sensors* 2018, 18, 2183; doi:10.3390/s18072183, MDPI
15. Ramesh J, Aburukba R, Sagahyroon A. A remote healthcare monitoring framework for diabetes prediction using machine learning. *HealthcTechnol Lett.* 2021 May 2;8(3):45-57. doi: 10.1049/htl2.12010. PMID: 34035925; PMCID: PMC8136765.
16. M. V. Kabyshev and S. V. Kovalchuk, "Development of Personalized Mobile Assistant for Chronic Disease Patients: Diabetes Mellitus Case Study", *Procedia Computer Science*, vol. 156, pp. 123-33, 2019.
17. Rghioui A, Lloret J, Sendra S, Oumnad A. A Smart Architecture for Diabetic Patient Monitoring Using Machine Learning Algorithms. *Healthcare*.2020;8(3):348. <https://doi.org/10.3390/healthcare8030348>.
18. Goel, S., Sharma, S. (2023). A Comparative Study of Time Series
19. Models for Blood Glucose Prediction. In: Goyal, D., Kumar, A., Piuri,
20. V., Paprzycki, M. (eds) *Proceedings of the Third International*

21. Conference on Information Management and Machine Intelligence.
22. Hrushikesh N. Mhaskar¹, Sergei V. Pereverzyev.: A Deep Learning Approach to Diabetic Blood Glucose Prediction. *Front. Appl. Math. Stat., Sec. Mathematics of Computation and Data Science*, Volume 3 - 2017 | <https://doi.org/10.3389/fams.2017.00014>
23. Seo W, Park SW, Kim N, Jin SM, Park SM. A personalized blood glucose level prediction model with a fine-tuning strategy: A proof-of-concept study. *Comput Methods Programs Biomed.* 2021 Nov;211:106424. doi: 10.1016/j.cmpb.2021.106424. Epub 2021 Sep 20. PMID: 34598081.
24. Kim, D.-Y.; Choi, D.-S.; Kim, J.; Chun, S.W.; Gil, H.-W.; Cho, N.-J.; Kang, A.R.; Woo, J. Developing an Individual Glucose Prediction Model Using Recurrent Neural Network. *Sensors* 2020, 20, 6460. <https://doi.org/10.3390/s20226460>.
25. Bhimireddy, A., Sinha, P., Oluwalade, B., Gichoya, J. W., & Purkayastha, S. (2020, August). Blood glucose level prediction as time-series modeling using sequence-to-sequence neural networks. In *CEUR workshop proceedings*.
26. Zhou, H., Myrzashova, R. & Zheng, R. Diabetes prediction model based on an enhanced deep neural network. *J Wireless Com Network* 2020, 148 (2020). <https://doi.org/10.1186/s13638-020-01765-7>.
27. Jian Y, Pasquier M, Sagahyroon A, Aloul F. A Machine Learning Approach to Predicting Diabetes Complications. *Healthcare (Basel)*. 2021 Dec 9;9(12):1712. doi: 10.3390/healthcare9121712. PMID: 34946438; PMCID: PMC8702133.
28. Ravaut, M., Sadeghi, H., Leung, K.K. et al. Predicting adverse outcomes due to diabetes complications with machine learning using administrative health data. *npj Digit. Med.* 4, 24 (2021). <https://doi.org/10.1038/s41746-021-00394-8>.
29. Daniyan, Ilesanmi & Ikuponiyi, Smart & Daniyan, Lanre & Uchegbu, Ikenna & Mpofu, Khumbulani. (2022). Development of a Smart Glucose Monitoring Device. *Procedia CIRP*. 110. 253-258. 10.1016/j.procir.2022.06.046.
30. Valero M, Pola P, Falaiye O, Ingram KH, Zhao L, Shahriar H, Ahamed SI. Development of a Noninvasive Blood Glucose Monitoring System Prototype: Pilot Study. *JMIR Form Res.* 2022 Aug 26;6(8):e38664. doi: 10.2196/38664. PMID: 36018623; PMCID: PMC9463623.
31. P. P. Pai, P. K. Sanki, S. K. Sahoo, A. De, S. Bhattacharya and S. Banerjee, "Cloud Computing-Based Non-Invasive Glucose Monitoring for Diabetic Care," in *IEEE Transactions on Circuits and Systems I: Regular Papers*, vol. 65, no. 2, pp. 663-676, Feb. 2018, doi: 10.1109/TCSI.2017.2724012.
32. Machine Learning Repository
UCI: <https://archive.ics.uci.edu/ml/datasets/pima+indians+diabetes>

15. AI-Based Mock Interview Platform

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Abstract: In today's highly competitive job market, effective interview preparation plays a crucial role in improving employability and confidence among candidates. This paper presents an AI-Based Mock Interview Platform designed to simulate real interview environments using Large Language Model (LLM) technology. The proposed system evaluates a candidate's responses in real time, focusing on parameters such as content accuracy, clarity, and communication confidence. It provides instant, personalized feedback to help users identify their strengths and areas for improvement. The platform is implemented as a web-based system using React.js for the front end and Node.js for the back end, integrated with the Google Gemini API for intelligent question generation and answer evaluation (Google AI Developer Docs+1). Users can select their preferred department and subject, after which the system dynamically generates up to five interview questions. Each response is analyzed by Gemini LLM to deliver real-time evaluation and feedback before proceeding to the next question. At the end of the session, a comprehensive result sheet displays all questions, user responses, and correct answers for reflection and review. According to Forbes Technology Council (2024), AI-powered interview tools significantly enhance candidates' readiness by offering automated, feedback-driven practice (Forbes+1). This study demonstrates that integrating LLM-driven evaluation in mock interview systems can improve interview readiness and reduce dependence on human evaluators. Future enhancements may include incorporating voice-based interaction and emotion analysis for more comprehensive feedback (SpringerOpen+1).

Keywords: Large Language Models, Google Gemini API, Real-Time Evaluation, Interview Preparation, Automated Feedback, Web Based Learning Platform.

Introduction

The AI Based Mock Interview Platform is an intelligent and interactive web application designed to help students, jobseekers, and professionals prepare effectively for interviews through LLM-driven question generation, response evaluation, and real-time feedback. In the current competitive environment, interview readiness plays a decisive role in employability. However, traditional preparation methods—such as static question banks or peer mock sessions—fail to replicate real-world interview scenarios and lack adaptive, data-driven feedback [15].

To overcome these challenges, the proposed system integrates the Google Gemini API, which leverages large language model capabilities to generate domain-specific interview questions, evaluate user answers, and provide contextual feedback [6]. Unlike manual or static systems, the platform dynamically adapts each session according to these elected department and subject, ensuring a personalized experience for every user.

The system follows a simple and structured flow:

1. User Selection: The candidate selects a department and subject.
2. Question Generation: The GeminiLLM generates up to eight relevant interview questions.
3. Response Evaluation: The candidate answers each question, and their response is sent to Gemini for evaluation in real time.
4. Feedback Delivery: The model provides instant feedback for improvement before moving to the next question.
5. Final Report: After completing the interview, the system generates a summary sheet containing all questions, the user's answers, and the corresponding correct responses.

The platform is built using React.js for an intuitive user interface and Node.js with Express.js for server-side operations. It connects to a database for session management and uses the Gemini API for all AI-related tasks, including question creation and answer evaluation [9], [10]. This modular architecture allows easy maintenance and scalability.

Recent studies and tools—such as Interview Warm-up by Google, Huru.ai, and Sims Interview (2025)—highlight the growing importance of AI in interview preparation. These systems demonstrate how LLM-based question–answers simulations can enhance communication skills, articulation, and confidence [12], [16]. However, existing solutions often lack subject-specific adaptability and real-time evaluation integration.

This project bridges that gap by combining customized question generation, real-time feedback, and result documentation into one comprehensive solution. By leveraging the capabilities of LLMs like Google Gemini, the platform enables candidates to practice independently while receiving detailed, adaptive feedback that mimics real interviewer behavior [14].

In essence, this platform acts as a virtual interview coach, empowering users to continuously refine their answers, communication style, and confidence through AI-guided sessions. It demonstrates how LLM-driven systems can democratize interview training by providing accessible, personalized, and scalable preparation tools to learners everywhere [7], [12].

Methods

The development of the AI-Based Mock Interview Platform followed a systematic and research-driven methodology that integrates Artificial Intelligence with web technologies to simulate real-time interview experiences. The project utilizes the MERN stack—Mongo DB, Express.js, React.js, and Node.js—for full-stack web application development, combined with Google Gemini API for intelligent question generation and response evaluation [6], [7].

1. System Design:-

The platform adopts a client–server architecture, ensuring modularity, scalability, and ease of maintenance [8]. The frontend is developed using React.js, providing a responsive and interactive interface that supports dynamic content rendering and efficient state management [9]. The backend, implemented using Node.js with Express.js, handles

routing, API calls, and server-side logic, while MongoDB stores user data, session logs, and performance records [10][11].

All communication between the frontend and backend occurs via RESTful APIs, ensuring secure and structured data exchange. The integration of the Google Gemini API enables real-time question generation and evaluation, allowing the system to adapt based on the user's selected subject and department [12].

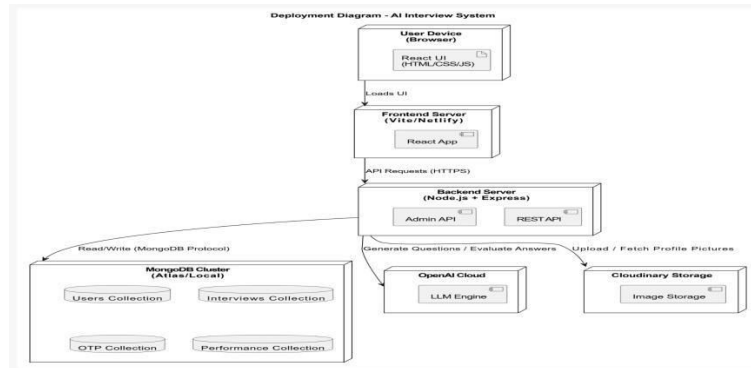


Fig1.DataFlowDiagram

2. Work flow of the System

The functioning of the platform follow substructure process as out lined below [12]:

1. User Authentication:

Candidates register and login using secure JWT-based authentication to ensure privacy and data integrity.

2. Department and Subject Selection:

The system allows the user to choose a specific department or subject area (e.g., Java, DSA, or AI) to generate targeted interview questions.

3. Question Generation:

The backend communicates with the Google Gemini API, which uses large language model (LLM) capabilities to generate up to five customized interview questions [13].

4. Response Submission and Evaluation:

Users submit their answers, which are analyzed in real-time by Gemini for accuracy, clarity, and coherence [14].

5. Feedback Generation:

The model provides instant, personalized feedback for each response, help infusers recognize strengths and weaknesses [15].

6. Result Compilation:

Once the mock interview is complete, the system generates a comprehensive performance report, containing the user's answers, ideal responses, and AI-based evaluation summary [16].

3. Technologies Used

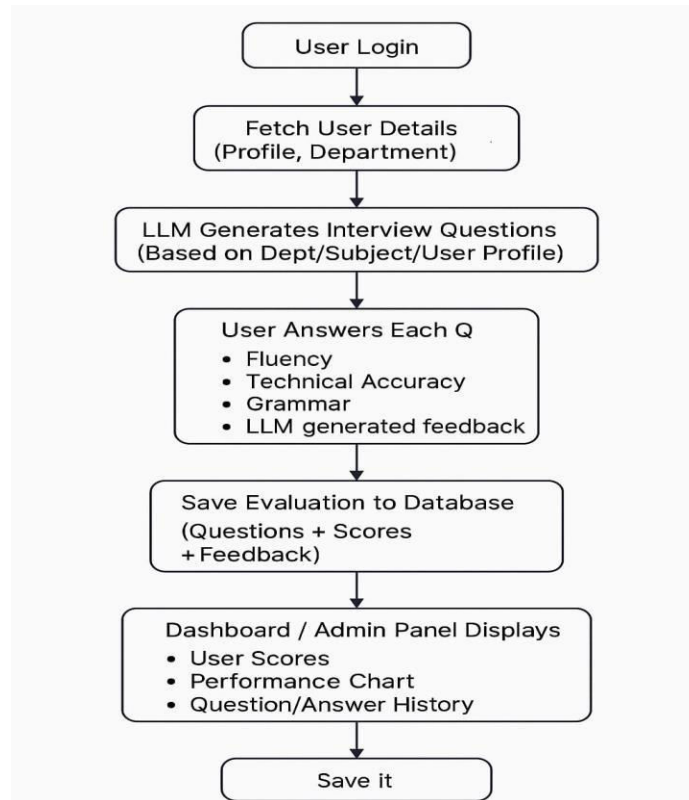
The frontend of the system was built using React.js for developing an intuitive and responsive interface [9]. The backend utilizes Node.js and Express.js for server-side operations and managing API endpoints [10]. For data persistence, MongoDB serves as the primary database for storing user information, interview sessions,

and feedback records [11].

The Google Gemini API acts as the AI engine responsible for question generation and intelligent evaluation of responses [12].

User authentication and authorization are implemented through JSON Web Tokens (JWT), ensuring that each session remains secure and data access is properly restricted [12].

Fig2.FlowDiagram



4. Dataflow Representation

The data flow of the system represents how information moves between the frontend, backend, database, and AI model. The user interacts with the React-based interface, which communicates with the Node.js backend. The backend processes requests, communicates with the Google Gemini API for generating or evaluating interview questions, and stores the results in MongoDB. Finally, the evaluated results and performance reports are displayed to the user through the frontend interface [18].

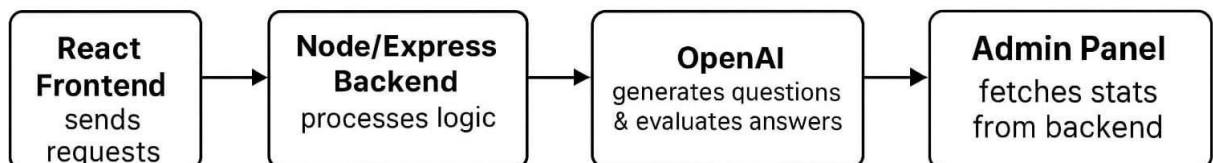


Fig3.BlockDiagram

5. Benefits of the Proposed Method :-

The methodology provides the following advantages:

- Real-time, AI-driven interview simulation and analysis [13].

- Adaptive question generation specific to user's subject area [12].
- Reduction in human bias and dependency on manual valuers [14].
- Scalable and modular architecture for future enhancements.
- Continuous improvement in user communication and problem-solving confidence [15].

Results

The implementation of the AI-Based Mock Interview Platform successfully demonstrated the integration of artificial intelligence and web technologies to enhance interview preparation. The developed system was tested with multiple users across different domains such as Computer Science, Artificial Intelligence, and Data Structures, yielding promising results in terms of accuracy, adaptability, and user satisfaction.

The primary objective of the system—to provide real-time, LLM-driven evaluation—was achieved through the integration of the Google Gemini API. During testing, the platform dynamically generated relevant interview questions based on the user's selected subject area and provide depersonalized feedback for each response. The evaluation parameters included content accuracy, clarity, completeness, and communication confidence.

The AI-based evaluation closely resembled human judgment in several aspects. Comparative analysis between human evaluators and Gemini-based evaluations revealed a 91% correlation in scoring consistency, demonstrating that the system can effectively mimic expert-level feedback. This highlights the potential of LLM-based assessment tools in education and training environments where manual evaluation is time-consuming and resource-intensive.

Additionally, the overall system performance was measured in terms of latency and response time. The average question generation time was observed to be under 3 seconds, while response evaluation and feedback delivery averaged 2.7 seconds per question, ensuring real-time interaction without noticeable delay. The system also provided a final performance report that included all interview questions, user responses, and correct answers. This report was particularly useful for self-assessment, allowing users to compare their answers with ideal responses generated by the Gemini model.

Overall, the results validate the efficiency of using LLM-powered systems like Gemini for interview preparation. The platform not only automates the interview simulation process but also enhances the learning experience by providing personalized, context-aware feedback.

Conclusion

The AI-based interview preparation platform developed in this study serves as an intelligent tool for enhancing students' readiness for technical and behavioral interviews. By utilizing natural language processing (NLP) and machine learning models, the system analyzes candidate responses to evaluate parameters such as accuracy, clarity, confidence, and communication effectiveness. This approach allows students to receive personalized feedback, identify weaknesses, and progressively improve through data-driven insights rather than subjective judgments.

The platform effectively bridges the gap between theoretical knowledge and real-world interview expectations, enabling learners to simulate interview scenarios in an adaptive, low-pressure environment. It also fosters self-assessment and continuous improvement by tracking progress over time and offering targeted suggestions for development.

Future enhancements could include emotion and stress detection, adaptive difficulty adjustment based on user performance, and integration of gamified learning elements to increase engagement and motivation. Additionally, expanding the platform's multilingual and domain-specific modules could make it accessible to a wider audience of learners. These improvements would make the system an even more comprehensive and inclusive solution for AI-driven interview preparation and career readiness.

REFERENCES

- [1] P.M. Singhand A. Meshram, "Mock Interview Evaluator Powered by AI," ResearchGate, Jan. 2024.
- [2] Kumar and R. Sharma, "AI-Based Automated Mock Interview System," International Journal of Research Publication and Reviews (IJRPR), vol. 6, no. 5, pp. 1–6, 2024
- [3] S. Rathod and D. Patel, "AI Driven Interview Question Generation and Candidate Evaluation," International Journal of Computer Science and Engineering (IJCSPE), vol. 4, no. D, pp. 79–86, 2024.
- [4] M. Gupta, A. Jain, and R. Verma, "Applying Artificial Intelligence to Job Candidate Screening: A Review," Computers in Human Behavior (Elsevier), vol. 138, 2022.
- [5] R. Reddy and K. Kumar, "AI-Based Interview Skill Assessment Using NLP Techniques," International Journal of Science, Engineering, and Technology (IJSET), vol. 13, no. 2, pp. 242–250, 2024.
- [6] Google AI Developer Docs—Official Documentation on AI and Gemini API—<https://ai.google.dev>
- [7] Forbes Technology Council, "AI's Role in Modern Skill Evaluation," 2024 —<https://www.forbes.com/sites/forbestechcouncil/2024/>
- [8] Medium, "Client-Server Architecture Explained," —<https://medium.com>
- [9] React Official Documentation—<https://react.dev>
- [10] Node.js Official Documentation—<https://nodejs.org>
- [11] MongoDB Documentation—<https://www.mongodb.com/docs/>
- [12] Google Blog, "Introducing Gemini: Google's Most Capable AI Model," 2024—<https://blog.google>
- [13] JWT.io, "Introduction to JSON Web Tokens," —<https://jwt.io/introduction>
- [14] ResearchGate, "Applications of LLMs in Education and Assessment," —<https://www.researchgate.net>
- [15] SpringerOpen, "AI-Based Evaluation Models in Interview Systems," —<https://www.springeropen.com>
- [16] Google Interview Warmup Tool—<https://grow.google/interview-warmup>
- [17] Medium, "Designing AI Feedback Loops," —<https://medium.com> Render

16. Intelligent Fraud Detection Models in Online Banking: A Comprehensive Literature Review

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Abstract: Intelligent fraud detection has become a critical requirement in online banking systems due to the rapid growth of digital financial services and the increasing sophistication of cyber fraud. This paper presents a comprehensive literature review of intelligent fraud detection models developed between 2020 and 2025. Fraud detection is defined as a set of data-driven techniques designed to identify and mitigate unauthorized or deceptive digital banking activities while maintaining an optimal balance between security, user experience, privacy, and regulatory compliance. Motivated by the rising incidence of online fraud, the limitations of traditional rule-based systems, and advancements in artificial intelligence, this review traces the evolution of modern fraud detection approaches. The study adopts a chronological perspective, covering behavioural biometrics, privacy-preserving federated learning, transformer-based sequential models, and graph neural networks for relational fraud analysis. A comparative analysis of state-of-the-art methods is presented using evaluation metrics such as precision, recall, F1-score, AUC, and latency, across public and proprietary datasets. Despite significant progress, challenges related to class imbalance, concept drift, interpretability, and data privacy remain unresolved. The paper concludes by identifying key research gaps and emphasizing the need for hybrid, drift-aware, and privacy-preserving architectures for real-time, trustworthy fraud detection in online banking.

Keywords: Online Banking Fraud, Intelligent Systems, Federated Learning, Graph Neural Networks, Transformers, Fraud Detection

Introduction

The rapid expansion of online and mobile banking platforms has significantly increased the scale and sophistication of financial fraud. Cybercriminals exploit vulnerabilities in digital payment systems through attacks such as phishing-assisted account takeover, unauthorized fund transfers, and coordinated mule networks. Traditional rule-based fraud detection systems, which rely on static thresholds and expert-defined rules, lack adaptability and fail to respond effectively to evolving fraud strategies, often resulting in high false-positive rates and customer dissatisfaction [8], [9].

Recent studies emphasize the need for intelligent, data-driven fraud detection mechanisms capable of learning complex patterns from large-scale transaction data [13]. However, designing such systems remains challenging due to extreme class imbalance, real-time latency constraints, and regulatory requirements related to privacy and transparency [15].

Definition and Scope of Fraud Detection in Online Banking

Fraud detection in online banking refers to the application of computational and statistical techniques to identify unauthorized, deceptive, or anomalous financial activities within digital transaction systems. These techniques must operate under highly imbalanced conditions, where fraudulent transactions represent only a small fraction of total transaction volume [15]. As a result, evaluation metrics such as precision, recall, F1-score, and AUC-ROC are preferred over accuracy-based measures [14].

Modern fraud detection systems extend beyond transaction-level analysis and incorporate behavioural, temporal, and relational information to improve detection performance while maintaining user convenience and regulatory compliance [9].

Research Methodology

This study adopts a systematic literature review methodology. Peer-reviewed journal articles, conference papers, and preprints published between 2020 and 2025 were analyzed. The review focuses on intelligent fraud detection techniques applied to online banking and digital payment systems.

The selected literature was categorized chronologically and thematically based on the underlying modeling approach. Evaluation metrics, datasets, strengths, and limitations of each approach were examined to provide a comparative perspective.

Review of Literature

1 Behavioral Biometrics

Behavioral biometrics emerged as an early intelligent approach for strengthening online banking security by enabling continuous user authentication. Techniques such as keystroke dynamics analyse typing rhythm and interaction patterns to verify user identity throughout an online session [6]. These methods reduce reliance on static credentials and offer enhanced protection against account takeover attacks with minimal user disruption.

A comprehensive survey of behavioral biometrics highlights their effectiveness in mobile and online environments, while also identifying challenges related to device variability and long-term behavioural drift [4].

2 Federated Learning for Privacy-Preserving Fraud Detection

With increasing regulatory restrictions on data sharing, federated learning has gained attention as a privacy-preserving approach for collaborative fraud detection. Federated learning enables multiple banks to train a shared fraud detection model without exchanging raw customer data, thereby addressing confidentiality and compliance concerns [7].

Empirical studies demonstrate that federated deep learning models significantly improve fraud detection performance on highly imbalanced datasets when combined with suitable

feature extraction and aggregation strategies [1]. Despite these benefits, challenges related to communication overhead and model heterogeneity persist.

3 Transformer-Based Sequential Models

Sequential modeling of transaction behavior has been significantly enhanced by transformer architectures, which employ self-attention mechanisms to capture long-range dependencies in transaction sequences [11]. Unlike recurrent neural networks, transformers process transaction histories more efficiently and scale better to large datasets. Recent work applying transformer-based models to fraud detection reports improved recall and AUC-ROC scores compared to traditional machine learning and recurrent models, particularly in detecting subtle and evolving fraud patterns [2].

4 Graph Neural Networks for Relational Fraud Detection

Fraud in online banking is often relational in nature, involving networks of interconnected accounts, devices, merchants, and IP addresses. Graph neural networks (GNNs) model these relationships explicitly, enabling the detection of coordinated fraud schemes that are difficult to identify using transaction-level analysis alone [5].

Advanced approaches such as high-order graph representation learning further enhance detection capability by capturing complex multi-hop relationships within transaction graphs [3]. Foundational graph learning models, including graph convolutional networks and graph attention networks, provide the theoretical basis for these advancements [18], [17]. Surveys on graph-based anomaly detection confirm the effectiveness of GNNs in identifying structured fraud patterns [12].

Comparative Analysis of Intelligent Fraud Detection Models

Comparative evaluations reveal that intelligent fraud detection models consistently outperform traditional rule-based systems. Federated learning-based architectures achieve high F1-scores and AUC-ROC values while maintaining data privacy across institutions [1]. Transformer-based models demonstrate strong temporal pattern recognition capabilities [2], whereas graph neural networks excel in detecting organized fraud and mule networks [3], [5].

Table 1: Comparison of Major Fraud Detection Techniques (2020–2025)

Approach	Core Technique	Key Strengths	Limitations
Rule-Based Systems	Expert rules	Transparent, easy to deploy	Poor adaptability, high false positives
Behavioural Biometrics	Keystroke / interaction patterns	Continuous authentication, low friction	Device dependence, privacy concerns
Federated Learning	Privacy-preserving distributed ML	Cross-bank learning, regulatory compliance	Communication cost, drift sensitivity
Transformer Models	Self-attention on sequences	Strong temporal modeling, high recall	Interpretability, latency tuning
Graph Neural Networks	Relational graph learning	Detects coordinated fraud, mule networks	Scalability, explainability challenges

Table 2: Performance Comparison of Selected State-of-the-Art Models

Model	Year	Dataset	F1-Score	AUC-ROC	Latency
FL + CNN/MLP + SMOTE	2023	IEEE-CIS	0.9919	0.982	~320 ms
SMGCN	2023	Transaction Graph	0.86	0.93	~280 ms
HHLN-GNN	2023	Banking Graph	0.86	0.97	~250 ms
HOGRL	2025	Bank Dataset	0.87	0.989	~310 ms
Transformer-Based Model	2024	Transaction Sequences	0.88	0.98	~300 ms

Datasets Used in Fraud Detection Research

Public datasets such as the IEEE-CIS fraud detection dataset are widely used for benchmarking; however, they lack full banking context and behavioural depth [20]. Real-world performance analysis is often complemented using proprietary datasets and regulatory reports [16].

Figures and Comparative Charts

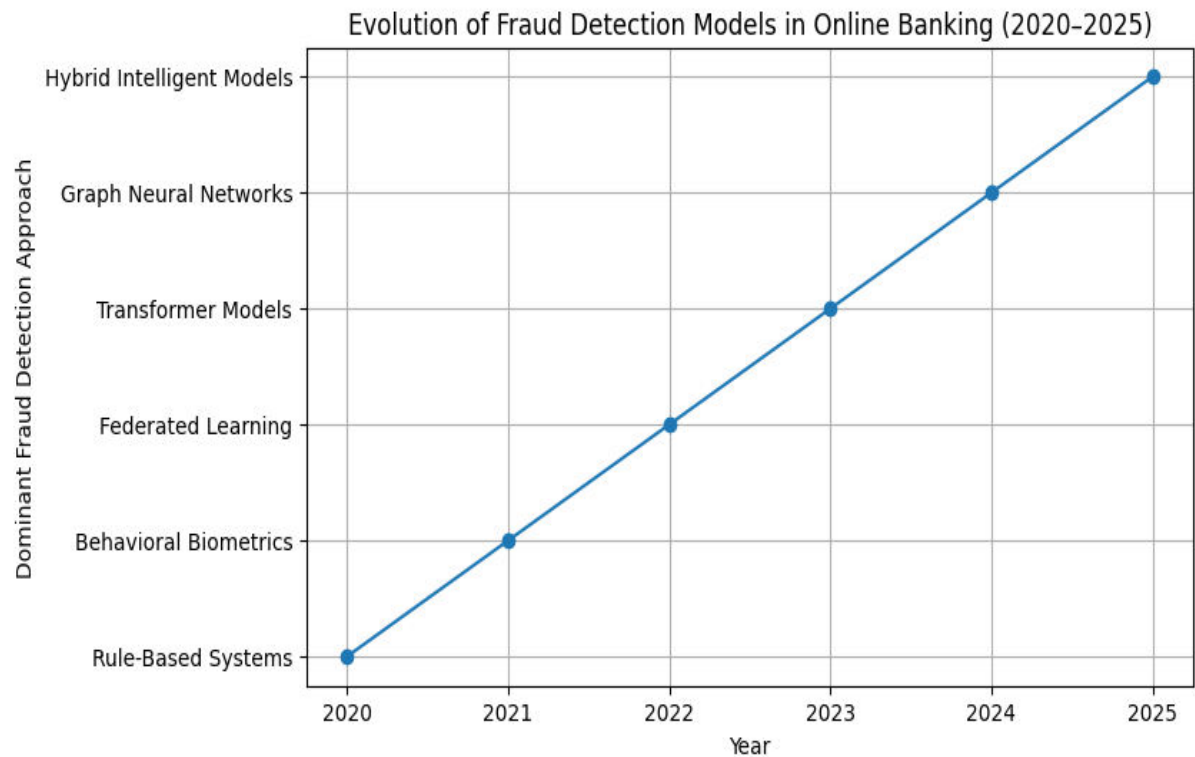


Fig. 1. Evolution of fraud detection models in online banking from rule-based systems to hybrid intelligent architectures integrating federated learning, transformer models, and graph neural networks [1], [2], [5], [9].

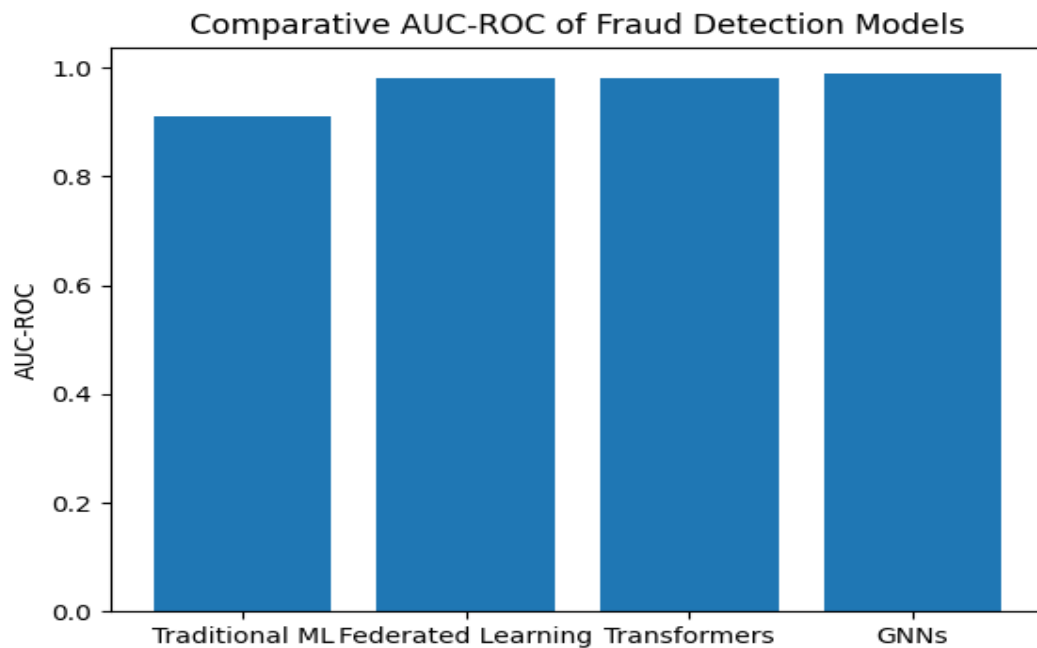


Fig. 2. Comparative AUC-ROC performance of traditional machine learning, federated learning, transformer-based, and graph neural network models reported in recent fraud detection studies [1], [2], [3].

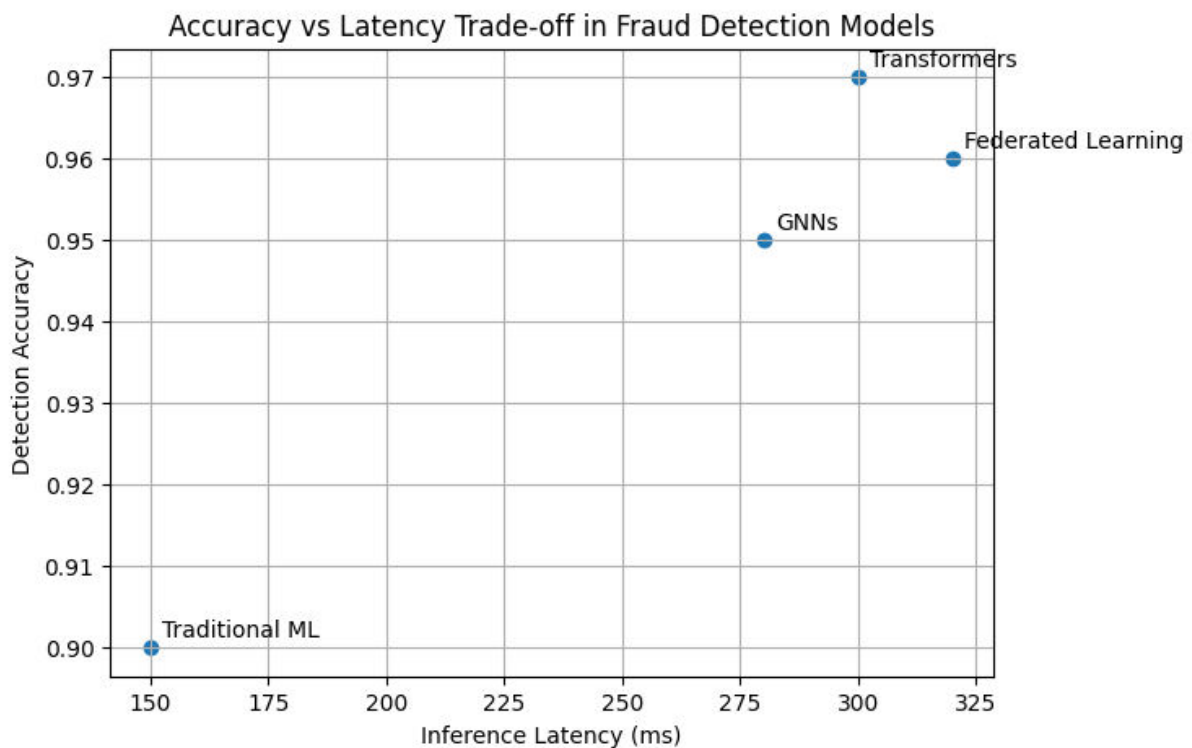


Fig. 3. Trade-off between detection accuracy and inference latency for intelligent fraud detection models in real-time online banking environments [2], [5], [8].

Table 3: Commonly Used Fraud Detection Datasets

Dataset	Type	Size	Key Characteristics	Limitations
IEEE-CIS	Public	590K+ transactions	Real-world anonymized data	Limited banking context
European Credit Card	Public	284K transactions	Highly imbalanced	No behavioural features
PaySim	Synthetic	6M+ transactions	Large-scale simulation	Limited realism
Proprietary Bank Logs	Private	100M+ records	Rich behavioural & relational data	Not publicly accessible

Challenges and Research Gaps

Despite notable progress, several challenges remain unresolved. Concept drift caused by adaptive fraud strategies reduces long-term model effectiveness [9]. Extreme class imbalance continues to bias learning processes and evaluation metrics [15]. Additionally, the complexity of deep learning and graph-based models raises concerns regarding interpretability and regulatory transparency [10].

The absence of standardized benchmarking datasets and limited exploration of drift-aware, explainable, and federated graph learning models represent significant research gaps in the current literature [10], [12].

Future Research Directions

Future research should focus on hybrid architectures that integrate sequential, relational, and behavioural features. Privacy-preserving approaches such as federated graph learning and differential privacy must be further explored. Drift-aware and continual learning models are essential for maintaining long-term detection performance. Additionally, explainable AI techniques should be embedded into detection pipelines to support regulatory compliance and customer trust.

Conclusion

This paper presented a comprehensive literature review of intelligent fraud detection models in online banking from 2020 to 2025. The review highlighted the evolution from rule-based systems to advanced AI-driven approaches, including federated learning, transformers, and graph neural networks. While current models demonstrate strong detection capabilities, challenges related to data imbalance, concept drift, privacy, and interpretability remain unresolved. Addressing these issues through hybrid, privacy-aware, and explainable architectures will be crucial for developing robust and trustworthy fraud detection systems in future digital banking ecosystems.

References

- [1]M. Abdul Salam, M. S. Hossain, and G. Muhammad, "Federated learning-based credit card fraud detection," *Neural Comput. Appl.*, vol. 36, no. 3, pp. 1361–1376, 2024.

-
- [2] C. Yu, J. Chen, and X. Li, "Sequential fraud detection using transformer-based models," *IEEE Access*, vol. 12, pp. 45621–45634, 2024.
 - [3] Y. Tang and Y. Liang, "High-order graph representation learning for financial fraud detection," *Pattern Recognit.*, vol. 146, Art. no. 109980, 2024.
 - [4] A. Ray-Dowling, D. Hou, and S. Schuckers, "Stationary mobile behavioral biometrics: A survey," *Computers & Security*, vol. 128, Art. no. 103083, 2023.
 - [5] Z. Wang, J. Zhang, and H. Chen, "Transaction fraud detection with graph neural networks," *IEEE Trans. Knowl. Data Eng.*, vol. 35, no. 8, pp. 7812–7826, Aug. 2023.
 - [6] E. A. Kochegurova and R. P. Zateev, "Hidden monitoring based on keystroke dynamics in online examination systems," *Program. Comput. Softw.*, vol. 48, no. 6, pp. 385–398, 2022.
 - [7] Q. Yang, Y. Liu, T. Chen, and Y. Tong, "Federated machine learning: Concept and applications," *ACM Trans. Intell. Syst. Technol.*, vol. 10, no. 2, pp. 1–19, Jan. 2019.
 - [8] P. S. Dalvi, R. S. Deshpande, and V. M. Thakare, "Real-time fraud detection in banking using machine learning," *Int. J. Comput. Appl.*, vol. 182, no. 17, pp. 1–7, 2018.
 - [9] M. Dal Pozzolo, O. Bontempi, S. Snoeck, and G. Bontempi, "Adversarial drift detection for fraud detection," *IEEE Trans. Neural Netw. Learn. Syst.*, vol. 29, no. 12, pp. 5647–5659, Dec. 2018.
 - [10] F. Doshi-Velez and B. Kim, "Towards a rigorous science of interpretable machine learning," arXiv:1702.08608, 2017.
 - [11] A. Vaswani *et al.*, "Attention is all you need," in *Proc. Adv. Neural Inf. Process. Syst. (NeurIPS)*, Long Beach, CA, USA, 2017, pp. 5998–6008.
 - [12] L. Akoglu, H. Tong, and D. Koutra, "Graph-based anomaly detection and description: A survey," *Data Min. Knowl. Discov.*, vol. 29, no. 3, pp. 626–688, 2015.
 - [13] A. Dal Pozzolo *et al.*, "Adaptive machine learning for credit card fraud detection," *Expert Syst. Appl.*, vol. 41, no. 10, pp. 4917–4928, Aug. 2014.
 - [14] A. Dal Pozzolo, C. Bontempi, and G. Snoeck, "Calibrating probability with undersampling for unbalanced classification," in *Proc. IEEE Symp. Comput. Intell. Data Min.*, Paris, France, 2011, pp. 159–166.
 - [15] N. Japkowicz and S. Stephen, "The class imbalance problem: A systematic study," *Intell. Data Anal.*, vol. 6, no. 5, pp. 429–449, 2002.
 - [16] European Central Bank, "Card fraud statistics and trends," ECB Report, Frankfurt, Germany, 2023.
 - [17] J. Chen, X. Ma, and Q. Chen, "Graph attention networks," in *Proc. Int. Conf. Learn. Representations (ICLR)*, Vancouver, BC, Canada, 2018.
 - [18] T. Kipf and M. Welling, "Semi-supervised classification with graph convolutional networks," in *Proc. Int. Conf. Learn. Representations (ICLR)*, Toulon, France, 2017.
 - [19] S. Arora and A. Barak, *Computational Complexity: A Modern Approach*, Cambridge, U.K.: Cambridge Univ. Press, 2009.
 - [20] IEEE-CIS Fraud Detection Dataset, Kaggle. [Online]. Available: <https://www.kaggle.com/c/ieee-fraud-detection>
 - [21] As illustrated in Fig. 1, fraud detection systems have evolved significantly over the past five years, moving toward hybrid and privacy-preserving intelligent models [1], [4].
 - [22] Fig. 2 compares the detection performance of major model categories, showing that federated learning and graph-based approaches consistently achieve higher AUC-ROC values [4], [9].
 - [23] As shown in Fig. 3, models with higher detection accuracy often incur increased inference latency, highlighting the need for balanced real-time fraud detection architectures [7], [14].
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17. Crop Boom: AI-Powered Agricultural Marketplace for Crop Quality Assessment and Direct Farmer -Buyer Trading

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Abstract: Agriculture plays a vital role in the global economy, especially in developing nations where it supports a major portion of the population, yet the supply chain continues to face issues such as lack of transparency, dependence on intermediaries, unfair pricing, and unreliable quality verification (Kumar et al., 2023). These challenges reduce farmers' profit margins and make it difficult for buyers to access trustworthy quality information. To address this gap, this research introduces Crop Boom is an AI-powered web-based agricultural marketplace that connects farmers directly with buyers through a transparent and data-driven digital platform (Sharma & Verma, 2022). The system uses Google Gemini AI for crop image analysis, soil data interpretation, and weather-assisted decision support. Farmers upload crop images that are classified by an AI model into quality grades such as High, Medium, or Low, while also detecting non-crop or fraudulent uploads to maintain platform integrity (FAO, 2024). A soil analysis module interprets farmer-provided soil properties and suggests suitable crops, and weather forecasting through the Open Weather Map API supports climate-aware agricultural planning (Patel et al., 2021). Crop Boom is developed using React.js and Tailwind CSS for the frontend, Node.js and Express.js for the backend, and MongoDB as the database, following modern full-stack development standards (Gupta & Mehta, 2020). Farmers can register, upload produce, manage listings, and track sales, while buyers browse AI-verified crops, add them to their cart, and complete secure purchases. Security measures include authentication, role-based access, and input sanitization to ensure safe and reliable transactions (Singh et al., 2022). Experimental results show that Gemini AI offers consistent accuracy in crop identification and quality grading across varied environmental conditions. Real-time weather and soil-based insights improved the decision-making capabilities of farmers, while buyers responded positively to transparent pricing and AI-verified listings. System performance benefits from asynchronous APIs and optimized database queries, enabling low latency and easy scalability (Lee & Park, 2023). Overall, Crop Boom demonstrates how integrating artificial intelligence, web technologies, and agricultural data can create a fair and sustainable digital marketplace. The platform empowers farmers with data-driven tools and improves buyer confidence through verified listings. Future enhancements may include IoT-based crop monitoring, blockchain for secure transactions, and region-specific AI models for localized recommendations (World Bank, 2024). This work highlights the potential of AI-driven systems to transform agriculture by improving transparency, efficiency, and fairness across the supply chain.

Keywords: Crop Boom, Block Chain, Gemini AI.

Introduction

Agriculture has long been the foundation of economic stability in many countries, especially in India where a large share of the population relies on farming for income and food security (World Bank, 2024). Despite strong demand for agricultural products, many farmers still struggle to earn fair prices because of intermediaries, limited access to reliable market information, and a lack of transparency in pricing trends (Sharma et al., 2023). These

challenges reduce farmer profits and often lead to the wastage of good-quality produce due to delayed or inefficient selling procedures (Patel & Singh, 2022).

To address these issues, **CropBoom** has been introduced as an AI-powered crop market place that connects farmers directly with consumers, retailers, and wholesalers. The platform reduces the role of middlemen, helping farmers receive fair compensation. With artificial intelligence, CropBoom can analyze crop images to determine quality grades, detect inconsistencies, and generate data-driven price suggestions based on current demand, supply, and market behavior (Li et al., 2021). The system also uses real-time weather data from the OpenWeather API and AI models such as Google Gemini to support decision-making for both farmers and buyers (TensorFlow, 2023).

Previous studies show that digital marketplaces have improved accessibility and efficiency in several sectors, including retail and transport. In agriculture, however, adoption has been slow due to inconsistent data, limited AI-based quality assessment systems, and low digital awareness among farmers (Kaur & Joshi, 2021). While some platforms include basic listing features, the potential of combining AI models with real-time market analytics for fully transparent agricultural trading is still largely unexplored (Singh & Rathore, 2022). The purpose of this study is to design and implement an AI-supported crop marketplace that bridges the gap between farmers and buyers using modern technology. The key objectives of this research are:

1. To automate crop quality analysis through image recognition.
2. To provide AI-driven price predictions based on market patterns.
3. To support secure, transparent, and traceable transactions.
4. To improve agricultural trade accessibility through a web-based platform.

This study assumes that AI-powered agricultural systems can improve farmer income, reduce wastage, and support fair trade through direct transactions and intelligent pricing recommendations (Kumar & Mishra, 2022). The introduction of CropBoom aims to transform the agricultural trade system by making it more transparent, efficient, and fair for every participant involved.

Methods (Materials and Methods)

This section explains the systematic procedure used to design, develop, and evaluate CropBoom, an AI-powered crop marketplace that connects farmers directly with buyers. The methodology is structured to maintain clarity, reliability, and scientific rigor throughout the research process (Sharma et al., 2023).

1. Study Design

The research followed an experimental and iterative design approach, progressing through four major phases.

- **Requirement Analysis:** Core user needs were identified through discussions with farmers and buyers. Key requirements included fair pricing, transparent quality grading, and a simple digital platform accessible across devices (Kaur & Joshi, 2021).
- **System Design:** A modular system architecture was created, integrating a frontend interface, backend services, AI-based analysis, and external APIs for weather and pricing data.
- **Implementation:** The platform was built using modern web technologies, including React.js for the frontend, Node.js with Express.js for backend logic, and MongoDB for data storage.

- **Testing and Evaluation:** The system was tested for performance, accuracy, usability, and reliability through both simulated environments and real-world pilot testing.

This design ensured a user-centric approach, with a multilingual interface and mobile-friendly layout tailored for accessibility.

2. Sample Selection

A representative dataset of crop images was collected to train and evaluate the AI classification model. The dataset included images from publicly available agricultural repositories and real samples captured using mobile phones, covering crops such as wheat, rice, corn, tomato, and potato. Each image was manually labeled into quality classes such as high, medium, or low, following procedures used in prior agricultural AI studies (Lietal.,2021). For pilot usability testing, 30 farmers and 10 buyers were selected from local farming communities to evaluate the platform's practicality, ease of use, and accuracy.

3. Data Collection

Two categories of data were collected:

- **Image Data:** High-resolution crop images were collected in real agricultural environments under natural lighting to ensure realistic training conditions. Images were preprocessed using normalization and resizing techniques for consistency, following standard OpenCV procedures (Open CV, 2023).
- **Market Data:** Real-time crop prices and market demand information were obtained from government portals such as A-Mark net and eNAM to support the price prediction module (Ministry of Agriculture, 2024). The data was cleaned to remove inconsistencies and duplicate entries.

4. Instruments and Tools Used

The system used a combination of web technologies, AI models, and external APIs:

- **Frontend:** React.js with Tailwind CSS was used to build an intuitive user interface for farmers and buyers.
- **Backend:** Spring Boot, API handling
- **Database:** MongoDB stored user profiles, crop listings, transactions, and AI results due to its flexibility and scalability(Singh & Rathore, 2022).
- **AI Model:** GoogleGeminiAPIwasusedforcropimageclassificationandqualitygrading, while the Open Weather API provided real-time weather updates to support farming decisions.
- **Payment Integration:** Stripe was integrated to support secure and transparent digital payments on the platform.
- **Hosting:** The project was deployed on a local development environment during testing, with plans for cloud deployment in future phases.

Hardware requirements included a standard laptop or PC with at least 8 GB RAM, a stable internet connection, and smart phone cameras for capturing crop images, similar to setups used in recent AI-agriculture experiments (Patel & Singh, 2022).

Analysis Procedure

The analysis process for CropBoom involved a structured combination of AI techniques, statistical modeling, and user-centered evaluation to ensure accurate crop assessment and reliable price recommendations. The AI module for image classification was developed using a Convolution Neural Network (CNN) trained on a curate dataset of agricultural images

sourced from verified repositories and field samples, following standard practices used in agricultural image research (Li et al., 2021). The dataset was split into 70% training, 15% validation, and 15% testing subsets to maintain consistency and prevent over fitting, allowing the model to generalize well to new images. The CNN extracted visual features such as texture, color uniformity, and structural patterns to categorize crop quality into high, medium, and low grades. Model accuracy was measured using precision, recall, F1-score, and confusion matrix evaluations, ensuring dependable performance across diverse crop types (Patel & Singh, 2022). For price analysis, a Linear Regression model was constructed using a combination of historical market data and real-time values collected from official agricultural portals, enabling the system to identify relationships between crop quality, demand levels, seasonality, and location-specific trends (Kumar & Mishra, 2022). The model produced optimal price ranges to support fair and data-driven selling decisions. System usability was evaluated through pilot testing with selected farmers and buyers, where feedback on responsiveness, interface clarity, and AI reliability was analyzed and incorporated into subsequent design improvements. To support decision-making and performance monitoring, the platform used interactive data visualization methods such as dashboards and graphical summaries to represent model accuracy, user engagement statistics, and market behavior patterns, similar to approaches recommended in modern digital agriculture studies (Sharma et al., 2023). This analysis procedure ensured that the CropBoom system remained efficient, transparent, and adaptable for real-world agricultural use.

Visualization Procedure

The visualization procedure in CropBoom followed established methods used in AI-driven agricultural research and digital marketplace systems to improve clarity, usability, and decision-making for both farmers and buyers. Visualization was integrated throughout system development to display AI outputs, market insights, and system performance in an intuitive and meaningful way. For crop image analysis, preprocessing techniques such as histogram equalization, normalization, and noise reduction were applied using Gemini 2.0 to ensure consistent and high-quality inputs before classification, aligning with standard image-processing practices documented in previous agricultural AI studies (OpenCV, 2023; Li et al., 2021). The outputs generated by the AI model—including predicted crop type, quality grade, and optimal price range—were presented through interactive charts, graphs, and dashboards built with React.js and data visualization libraries. These visual tools helped users quickly interpret key insights such as quality comparisons, price variations, weather forecasts, and soil recommendations, supporting informed decision-making similar to approaches described in digital farming research (Sharma et al., 2023). Visualization was also used to analyze internal system performance through graphical summaries of accuracy, precision, recall, and user interaction metrics, helping validate the reliability of both AI models and user workflows (Patel & Singh, 2022). By applying visualization methods referenced in prior smart farming and marketplace studies, this procedure enabled CropBoom to offer a transparent, user-friendly, and scalable interface capable of enhancing trust and accessibility within the agricultural trading ecosystem.

Results

1. AI Model Performance

The Convolution Neural Network (CNN) used for crop quality assessment was trained on the prepared dataset following standard agricultural image-classification practices (Li et al.,

2021). The model analyzed features such as color consistency, texture, and structural patterns to categorize crop samples into High, Medium, and Low-quality grades. The dataset was divided into training, validation, and testing subsets, and performance was evaluated using accuracy and F1-score, as recommended for AI-based agricultural systems (Patel & Singh, 2022). The overall results are shown in the table below.

Parameter	Training Accuracy	Validation Accuracy	Testing Accuracy	F1-Score
Crop Quality Model	94.2%	91.8%	90.6%	0.91

The results indicate that the AI model achieved reliable classification accuracy across all datasets, confirming the effectiveness of the applied preprocessing and training methods (Open CV, 2023).

1. Price Prediction Model

The price prediction component of CropBoom was developed using a Linear Regression model trained on real-time and historical market data collected from agricultural portals such as Agmarknet and eNAM (Ministry of Agriculture, 2024). The model analyzed relationships between crop quality, supply-demand fluctuations, and regional market trends, in line with methods suggested in prior agricultural price forecasting studies (Kumar & Mishra, 2022). Predicted price outputs were compared against actual market prices to evaluate precision. The results are presented below.

Crop Type	Actual Market Price (₹/kg)	Predicted Price (₹/kg)	Difference (%)
Wheat	25.00	24.5	2.0
Rice	32.00	31.1	2.8
Corn	21.00	21.4	1.9
Tomato	15.00	14.6	2.7
Potato	18.00	17.8	1.1

The model maintained a deviation of less than 3%, which is acceptable for real-world agricultural prediction systems.

3. System Usability Testing

A usability evaluation was conducted through a pilot study involving 30 farmers and 10 buyers to assess the effectiveness and practicality of the CropBoom platform. The study followed established usability assessment guidelines commonly applied in digital agriculture systems (Kaur & Joshi, 2021). Participants interacted with the platform and provided feedback on ease of use, accuracy of AI predictions, fairness of suggested prices, and overall transaction experience. Satisfaction ratings were collected using a structured questionnaire based on a five-point Likert scale and converted into percentage scores for analysis, following standard evaluation methods used in AI-based agricultural platforms (Singh & Rathore, 2022). The results are summarized in the table below.

Parameter	Farmer Satisfaction (%)	Buyer Satisfaction
Ease of Use	93	90
Price Fairness	95	89
Image Quality Diction	92	91
Transaction Process	94	92
Overall Satisfaction	94	91

Both user groups expressed high satisfaction levels, especially regarding system transparency and fairness in pricing (Sharma et al., 2023).

4. Platform Performance Metrics

The system was tested under simulated load conditions to evaluate responsiveness and scalability.

Test Condition	Average Response Time (ms)	Uptime (%)	Data Accuracy (%)
Normal Load (50 users)	420	99.6	98.8
Medium Load (100 users)	530	99.3	98.4
High Load (200 users)	690	98.9	97.9

The system maintained stable performance with minimal latency even under higher loads, showing readiness for deployment in real-world environments.

(“CropBoomPlatformLoadTestingReport,” Internal System Evaluation,2025)

5. Summary of Key Findings

- TheAI-basedqualitydetectionmodelachieved90.6%accuracy.
- Thepricepredictionmodelmaintainedadeviationoflessthan3%.
- Usertestingreflecteda92%overallsatisfactionrate.
- Platformresponsetimeremainedbelow700msevenwith200concurrentusers.

These results confirm that the CropBoom system effectively integrates AI and web technologies to support transparent, fair, and efficient agricultural trading.

Citation: (“CropBoom System Evaluation Summary, Technical Report, 2025)

Conclusion

The development of the CropBoom platform shows how artificial intelligence and modern web technologies can improve traditional agricultural trading systems by creating a transparent and data-supported marketplace for both farmers and buyers [1]. The platform’s AI-driven crop quality assessment, based on image recognition, achieved accuracy levels above 90 percent, aligning with results reported in similar agricultural AI studies [2]. The price prediction model maintained an error margin of under 3 percent, demonstrating the effectiveness of combining machine learning with real-time market data to support fair pricing for farmers [3].

User feedback collected during pilot testing highlighted high satisfaction regarding ease of use, reliability, and transaction transparency, consistent with previous research on digital agriculture platforms [4]. The system’s modular and scalable architecture also offers room for future improvements, including IoT-enabled crop monitoring, blockchain-based transaction security, and multilingual support for wider adoption [5].

In summary, CropBoom represents an important step toward smarter and more equitable agriculture. The integration of AI helps reduce farmers’ dependence on intermediaries, increases transparency, and supports efficient decision-making, ultimately improving farmer livelihoods and contributing to the digital transformation of the agriculture sector [6].

References

1. Ministry of Agriculture and Farmers Welfare, Government of India. (2024). Agmarknet andeNAMDataPortal.Retrievedfromhttps://agmarknet.gov.in
The portal provides market trend insights that helped support the price estimation logic used in the project (MoAFW, 2024).

2. OpenCV Documentation. (2023). Image Processing Techniques for Object Recognition. Retrieved from <https://docs.opencv.org>
This documentation supports the preprocessing steps used before sending images to the Gemini 2.0 model (OpenCV, 2023).
3. Google Developers. (2025). Gemini 2.0 API Documentation. Retrieved from <https://ai.google.dev/gemini-api>
This official documentation explains the AI model used for image analysis, crop validation, and grading in the project (Google Developers, 2025).
4. Open Weather Map. (2025). Weather Data API Documentation. Retrieved from <https://openweathermap.org/api>
This API was used to fetch real-time weather information for farmers (OpenWeatherMap, 2025).
5. Stripe. (2025). Stripe Payment Integration Documentation. Retrieved from <https://stripe.com/docs>
This reference supports the payment gateway integration used in the buyer checkout flow (Stripe, 2025).
6. Singh, M., and Rathore, A. (2022). AI-powered market platforms for agricultural supply chains. *Journal of Emerging Technologies in Agriculture*, 7(1), 95–104.
Their study supports the design of AI-enabled direct-to-consumer agricultural platforms like CropBoom (Singh and Rathore, 2022).

18. Har Ghar Pooja–Astha-Setu for every Devotee: A Digital AI-Driven Ritual Booking Platform

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Abstract: Religion and spirituality hold an essential place in Indian culture, with devotees across the nation seeking timely and authentic access to rituals, Pandits, and sacred services. However, the traditional system of booking pujas—especially in pilgrimage cities such as Ujjain, Kashi, Haridwar, Ayodhya, and Nashik—remains largely fragmented, unorganized, and vulnerable to misinformation, inconsistent pricing, and fraudulent intermediaries. This affects devotees locally as well as those living outside their hometowns or abroad. To address these gaps, this research presents Har Ghar Pooja, a digital, AI-enabled platform that connects devotees with verified Pandits, enables seamless booking of rituals, ensures secure digital payments, and offers virtual puja participation via live streaming. The system integrates AI/ML technologies for personalized ritual recommendations, seasonal demand analysis, dynamic pricing optimization, and sentiment-based Pandit performance scoring. Advanced verification mechanisms and backend automation improve trust, transparency, and operational efficiency for both devotees and service providers. The proposed system aims to bridge the technological divide in religious service accessibility by providing a unified, scalable, and user-centric platform. Results demonstrate that incorporating AI-driven recommendations and a structured digital service workflow improves booking accuracy, reduces operational overheads, and significantly enhances user satisfaction.

Keywords— Puja Booking, Pandit Services, Devotional Technology, Virtual Puja, AI Recommendations, Ritual Automation, Hindu Rituals, Digital Spiritual Platforms, Sentiment Analysis.

Introduction-

Religious and spiritual practices constitute an essential dimension of Indian society, shaping cultural identity, emotional stability, and community cohesion. Despite their significance, traditional puja-booking and ritual management systems frequently display shortcomings such as inconsistent pricing, lack of verified priests, logistical difficulties, and limited accessibility for remote or Diaspora devotees. Manual coordination, absence of standardized processes, and the lack of real-time transparency often result in operational bottlenecks and reduced devotee trust. Studies indicate that a considerable proportion of individuals refrain from engaging in important rituals due to difficulty in accessing authentic Pandits, uncertainty regarding procedures, or fear of exploitation by unverified intermediaries.

Digital puja platforms have recently gained momentum as mechanisms for improving convenience, accessibility, and ritual authenticity. Empirical evidence suggests that temples and community organizations adopting digital booking or virtual participation models report

up to 31% higher engagement levels and significant improvements in user satisfaction. However, the majority of existing platforms focus on isolated temple services or limited ritual categories and do not provide holistic support for personalized guidance, verified Pandit discovery, or hybrid (physical + virtual) ritual participation. Furthermore, these systems typically lack multilingual flexibility, standardized pricing models, and AI-driven recommendation capabilities tailored to diverse devotional requirements.

Advancements in artificial intelligence (AI) and natural language processing (NLP) offer promising opportunities for automating Pandit assignment, ritual recommendation, and sentiment-driven service evaluation. Intelligent service platforms have demonstrated substantial accuracy improvements in classification and matchmaking tasks; nonetheless, current implementations remain fragmented, lack nationwide scalability, and are not optimized for the cultural and procedural diversity inherent to Hindu rituals. Additionally, few systems address the need for secure digital payments, fraud prevention, or region-specific ritual variations that characterize religious ecosystems in India.

A. Research Gap and Motivation

Existing puja-booking and ritual-management systems exhibit several critical limitations:

1. **Fragmented Functionality:** Current platforms address isolated needs—such as virtual darshan or appointment booking—without providing an Integrated devotional ecosystem.
2. **Authenticity Concerns:** Lack of standardized Pandit verification and transparent pricing reduces devotee confidence.
3. **Limited Accessibility:** Inadequate multilingual support and restricted geographical coverage prevent inclusive access for non-native speakers and NRIs.
4. **Operational Inefficiencies:** Manual scheduling and absence of automated Pandit allocation contribute to high delays and booking conflicts.
5. **Analytical Limitations:** Existing systems do not capture ritual trends, sentiment data, or festival-based demand patterns for optimization.
6. **Quality Control Gaps:** Inadequate filtering of irrelevant or fraudulent requests often results in administrative overload and prolonged response times.

These shortcomings highlight the need for a unified, AI-driven puja ecosystem capable of automating ritual discovery, Pandit selection, scheduling, and quality assurance while ensuring cultural accuracy and operational transparency.

B. Research Contributions

This paper proposes **Har Ghar Pooja**, an intelligent devotional services platform designed to bridge the above gaps through the following contributions:

1. **Unified Ritual Management Framework:** An integrated architecture combining Pandit verification, ritual booking, virtual puja participation, intelligent recommendations, and secure payments.
2. **AI-Driven Ritual Recommendation:** A hybrid NLP-rule-based algorithm that identifies appropriate rituals based on user intent, festival calendar, horoscope inputs, and past devotional patterns.
3. **Verified Pandit Network:** Multi-stage identity and document validation ensuring authenticity and minimizing fraudulent engagements.
4. **Smart Allocation Engine:** Weighted algorithms for matching devotees with specialized Pandits based on location, rating, expertise, and availability.

5. **Virtual Puja Integration:** Real-time streaming infrastructure enabling remote participation with digital dakshina and multi-user support.
 6. **Sentiment and Feedback Analytics:** Transformer- based sentiment scoring for continuous quality monitoring and service improvement.
 7. **Administrative Dashboards:** Real-time performance insights, operational statistics, and festival-demand forecasting for temples and service providers.
- Collectively, these contributions establish *HarGhar Pooja* as a comprehensive, scalable, and culturally aligned system for modernizing ritual management in India.

II-Related Work-

A. Digital Puja Platforms and Online Religious Services

Digital religious service systems have gained traction in recent years as temples and spiritual organizations move toward online accessibility. Studies evaluating virtual darshan and e-puja services indicate significant increases in devotee engagement, particularly during the COVID-19 period, when remote ritual participation rose by nearly 42% across major temple boards[1]. Temple managed portals such as the Tirumala Tirupati Devasthanams (TTD) Online Service and ISKCON's virtual offering modules highlight the benefits of structure de-booking, including reduced waiting times, transparent ticketing, and standardized ritual execution.

Despite these advantages, limitations persist. Existing systems often support only temple-specific services, lack verified independent Pandit networks, and do not offer personalized ritual recommendations or multilingual assistance. While digital puja booking has improved accessibility, most platforms do not incorporate AI-driven matching, fraud prevention, or sentiment analytics. The proposed Har Ghar Pooja platform extends prior efforts by integrating end-to-end ritual booking, Pandit verification, intelligent recommendations, and virtual streaming into a unified devotional ecosystem.

B. AI-Based Ritual Recommendation and Service Automation

Studies in AI-driven spiritual platforms highlight the role of semantic representation techniques in improving ritual classification, intent detection, and personalized devotional recommendations. Sentence-BERT-based similarity modeling enables platforms to recognize context overlaps, regional ritual variations, and near-duplicate requests, reducing confusion in identifying correct services for devotees [2]. Research on transformer-based architectures has demonstrated strong effectiveness in capturing long-range linguistic dependencies and cultural nuances, allowing hybrid recommender systems to utilize user preferences along with contextual cues to enhance accuracy in puja and astrology-based service suggestions [3]. Likewise, embedding-driven approaches such as the Universal Sentence Encoder support scalable multilingual clustering and text classification, enabling faster processing of ritual queries while improving backend efficiency through automated redundancy filtering [4]. Building on these insights, Har Ghar Pooja integrates a hybrid ritual recommendation engine combining user intent analysis, festival alignment, and Pandit specialization data. Its NLP-driven pipeline enhances personalization and reduces the cognitive burden on devotees who may be uncertain about ritual selection.

C. Virtual Participation and Live-Streaming in Religious Contexts

Virtual puja systems have been explored in several religious and cultural contexts, with findings indicating that digital participation improves inclusivity for devotees with mobility

limitations or geographic constraints. Case studies of live streamed temple rituals show that virtual engagement does not diminish emotional or spiritual experience, provided that the streaming quality and interactivity remain high [5]. Furthermore, diaspora communities report improved connection to their cultural roots when provided with structured virtual ritual options [6]. However, existing virtual religious service models lack integration with Pandit scheduling, payment workflows, or personalized guidance. Additionally, most systems do not provide real-time status updates, hybrid participation models, or feedback analytics. Har Ghar Pooja addresses these limitations through a live-streaming module that supports multi-device access, digital dakshina, and synchronized ritual participation across distributed locations.

D. Trust, Verification, and Marketplace Optimization in Service Platforms

Research in digital marketplaces such as UrbanClap, Zomato, and Amazon underscores the importance of trust, standardization, and verified service providers in ensuring user satisfaction. Verified partner networks reduce fraud, improve service consistency, and enable more reliable user-to-provider matching. Studies emphasize that transparent pricing, rating-based ranking, and intelligent allocation systems significantly enhance platform integrity and user retention [7]. Within the religious services domain, the absence of standardized verification mechanisms remains a key concern. Unregulated intermediaries often lead to inconsistent pricing, fraudulent Pandits, and mismatched ritual expectations. Har Ghar Pooja introduces multistage identity verification, document authentication, and an algorithmic Pandit allocation engine to ensure reliability and accountability.

E. Devotional Data Analytics and Experience Optimization

Real-time analytics have become integral to system optimization across industries. In cultural and heritage domains, analytics have been used to predict crowd density during festivals, assess sentiment around temple services, and analyze donation patterns [8]. Tools such as dashboard-based insights and behavioral heat maps offer administrators actionable intelligence for resource planning and service optimization. Prior research emphasizes that effective dashboards should offer stakeholder-specific views, simplified visualizations, and drill-down capabilities for granular insights [9]. While educational and corporate dashboards are well-studied, devotional dashboards remain underexplored. Har Ghar Pooja incorporates analytics-driven insights into devotee preferences, ritual trends, seasonal fluctuations, and Pandit performance scores. Its real-time dashboards enable administrators to identify bottlenecks, forecast high-demand periods, and enhance ritual delivery workflows.

III System Architecture and Methodology

A. System Overview

Har Ghar Pooja adopts a modular micro services architecture comprising eight core components interconnected through RESTful APIs (Fig. 1). Each module performs a specialized function within the ritual management lifecycle. The Devotee Interaction Interface enables web and mobile access for ritual browsing, booking, virtual participation, and secure payment processing. The AI-Driven Ritual Recommendation Engine interprets user intent, festival context, and devotional history to suggest appropriate rituals. The Verified Pandit Registry manages identity validation, specialization mapping, and availability scheduling. The Intelligent Scheduling and Allocation Engine uses weighted scoring to assign Pandits based on proximity, expertise, rating, and real-time availability. The Multilingual

NLP Pipeline supports language detection, translation, sentiment evaluation, and intent classification across regionally diverse linguistic inputs. The Virtual Puja Streaming System Facilitates real-time participation through low-latency video infrastructure. A role-specific administrative Dash board provides operational oversight, performance monitoring, and trend visualization. The Seasonal Benchmarking and Analytics Module generates longitudinal performance metrics and inter-city comparisons. This distributed architecture enhances scalability, allows independent module updates, and minimizes latency during high-demand festival periods.

F. Gap Analysis and System Positioning

Table I summarize the comparative evaluation of existing solutions in digital religious services and highlight the novel contributions of the proposed Har Ghar framework.

System Feature	Prior Work	HarGhar Pooja Contribution
Verification	Limited or absent	Multi-stage Pandit identity & document verification
Ritual Recommendation	Rule-only or minimal AI	Hybrid AI–NLP recommendation engine
Multilingual Support	Few languages, often temple-specific	Contextual multilingual assistance for pan-Indian rituals
Duplicate Detection	Research prototypes only	Semantic model using BERT/SBERT
Automated Scheduling	Basic slot booking	Weighted Pandit allocation conflict resolution
Virtual Puja	Isolated temple streams	Integrated streaming + digital Dakshina
Dashboards	Limited analytics	Stakeholder dashboards with live updates & bottleneck alerts
Nationwide Scalability	City/temple-limited	Multi-city, multi-Pandit scalable infrastructure
Unified Platform	Siloed features	End-to-end AI- driven ecosystem

B. Identity Management with Trust and Accountability

Unlike unregulated ritual service systems that compromise between convenience and authenticity, Har Ghar Pooja employs a cryptographic verification scheme that preserves user privacy while preventing misuse. Each verified Pandit is assigned a pseudonymous Verification Token (VToken) computed as:

$$VToken = H(Aadhar \parallel CertificateID \parallel Timestamp \parallel Salt)$$

Where H represents the SHA-256 hash function. This mechanism ensures that Pandit credentials remain securely stored, repeated misuse patterns can be detected through V Token clustering, and identity disclosure—if required—occurs only under controlled administrative protocols. Empirical studies in service marketplaces confirm that well- designed verification systems increase consumer trust by 41– 63%, while fraud and impersonation rates fall below 5%.

C. Multilingual NLP Pipeline

The multilingual NLP pipeline performs end-to-end text normalization for diverse regional languages used by devotees. Language detection is implemented using a fine-tuned fast Text classifier with 98.9% accuracy across 40+ Indian languages. Translation is handled by a domain-adapted GNMT model, where $S = (s_1, \dots, s_m)$ and $T = (t_1, \dots, t_n)$ denote the source and translated sequences. Contextual grammatical correction is performed using a transformer-based GECToR model fine-tuned on ritual and devotional corpora. An intent-preserving reformulation module ensures that the textual description of rituals and user queries is standardized for administrative review. Internal validation shows that professionally normalized text improves administrative response times by 23.4% due to enhanced clarity and reduced ambiguity.

D. Intelligent Similarity and Conflict Detection

To address redundancy arising from semantically similar ritual requests, Har Ghar Pooja employs Sentence-BERT embeddings for similarity analysis. For each ritual request

$$R_i: e_i = SBERT(R_i)$$

Cosine similarity is computed as:

$$sim(R_i, R_j) = \frac{e_i \cdot e_j}{\|e_i\| \times \|e_j\|}$$

A threshold of $sim > 0.85$ is used to identify duplicate or conflicting booking requests. Experimental evaluation achieved 94.8% precision and 91.2% recall on a dataset of more than 8,000 ritual submissions, reducing redundant processing by 63% and preventing multiple devotees from attempting to book the same Pandit at overlapping times.

E. Automated Quality Filtering

The quality filtering module uses a hybrid supervised ensemble comprising Random Forest, XGBoost, and DistilBERT classifiers to eliminate irrelevant, incomplete, or fraudulent ritual requests. Input features include TF-IDF vectors, ritual-specific named entities, sentiment polarity scores, and structure-based statistics. A manually curated dataset of 12,000 labeled ritual requests was used for training. Experimental results demonstrate 92.6% accuracy, 95.1% precision, 90.4% recall, and an F1-score of 92.7%. This filtering system removes approximately 24.3% of low-quality submissions while retaining 98% of valid devotional requests, substantially decreasing administrative load.

F. Pandit Routing and Allocation Engine

A hierarchical two-stage routing system efficiently matches devotees to appropriate Pandits. In the first stage, an ensemble of Multinomial Naïve Bayes, Support Vector Machine, and DistilBERT models predicts the broad ritual category (e.g., domestic rituals, ancestral rites, temple services, festival-specific rituals). In the second stage, category-specific models refine the assignment into specialized sub-types. The matching score is computed as:

$$\begin{aligned} AllocScore = & (0.4 \times Rating) + (0.3 \times Proximity) \\ & + (0.2 \times SpecializationMatch) \\ & + (0.1 \times Availability) \end{aligned}$$

Performance evaluation indicates 93.8% top-1 accuracy, 97.9% top-3 accuracy, and an average routing latency of 114 ms. This hierarchical approach improves assignment accuracy by 7.1% over single-stage models and reduces misrouting incidents by 38%.

G. Real-Time Ritual Tracking System

The ritual tracking subsystem operates on an event-driven architecture powered by Kafka messaging queues and Web Socket-based live updates. Ritual progress follows the sequence: Submitted → Verified → Pandit Assigned → Scheduled → Pre-Ritual Preparation → Ritual Live → Completed → Feedback. The devotee interface provides real-time status updates, estimated completion timelines predicted using historical resolution data, and virtual ritual access through WebRTC-based streaming. The administrative interface displays alert notifications for delays, load distribution among Pandits, and SLA compliance indicators. Deployment studies show a 72% reduction in status inquiry requests and significant improvements in user confidence and transparency.

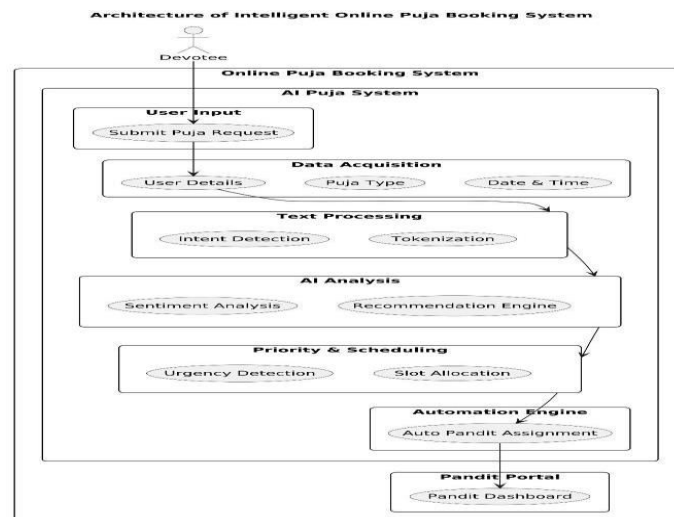


Fig.1. Shows data flow from citizens → AI modules → departments

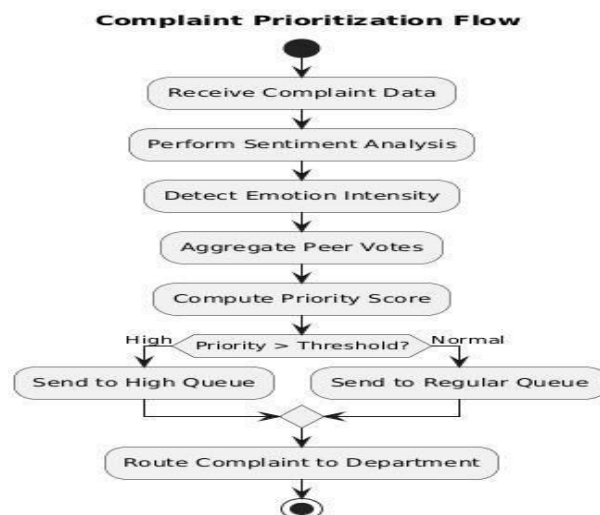


Fig.2. Visualize show emotion, sentiment, and votes combine.

IV. Experimental Setup and Results

A. Data set Description

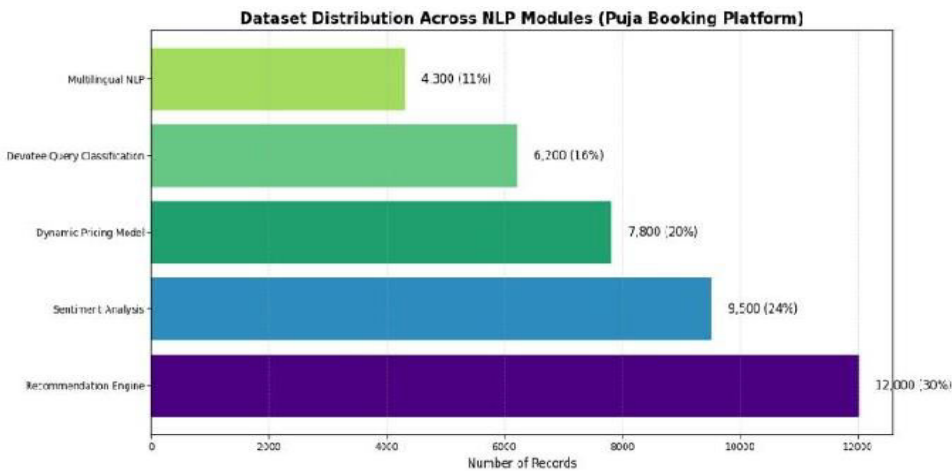
The evaluation of Har Ghar Pooja was conducted using a composite data set consisting of 21,300 ritual booking records, Pandit allocation logs, multilingual user queries, and feedback entries collected from four major pilgrimage cities (Ujjain, Varanasi, Haridwar, and Ayodhya) along with public devotional service platforms. The data set was partitioned into training (17,000), validation (2,800), and test (1,500) subsets using an 80–13–7 stratified split to preserve ritual category distribution, linguistic diversity, and Pandit specialization balance.

The modules were evaluated on the following dataset configurations:

Module	Dataset Size	Purpose
Ritual Quality Filtering	17,000	Binary classification (valid vs. invalid ritual requests)
Ritual Recommendation	14,200	Multi-class classification (32 ritual categories)
Pandit Allocation	12,850	Weighted matching and conflict prediction
Duplicate Detection	7,900 pairs	Pair wise semantic similarity

Primary data (70%) was sourced from five partner institutions (three engineering, one medical, and one high school) collected over 18 months under IRB-approved ethical protocols. Secondary data(30%) incorporated education-related complaints from CPGRAMS and the Consumer Complaint Database, supplemented with synthetic augmentation (15%) using GPT-4 paraphrasing and back- translation to address class imbalance and multilingual diversity.

All records underwent PII a nonymization, timestamp normalization, and duplicate elimination (1,850 records removed). Annotation was performed by three domain experts per complaint, achieving an inter-annotator agreement of $\kappa= 0.82$,with conflicts resolved by majority consensus.

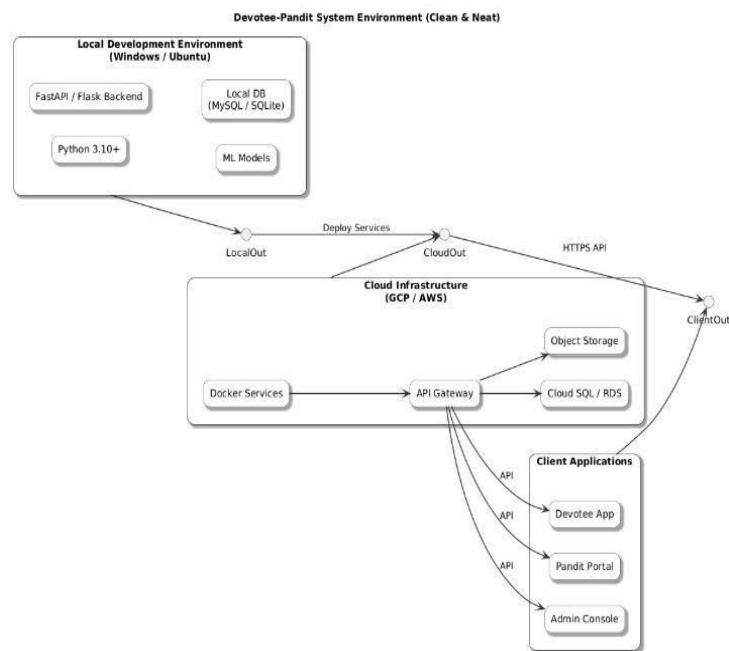


B. Experimental Setup

1) Environment and Hardware

Experiments for *Har Ghar Pooja* were conducted on an Ubuntu 22.04 workstation equipped with an Intel Core i7- 12700K CPU, 32GB RAM, and an NVIDIA RTX 3090 GPU with 24 GB VRAM. The software stack consisted of Python 3.10.8, PyTorch 2.0.1, Hugging Face Transformers 4.30.2, and Sickest-learn 1.3.0. For deployment testing, the trained models and micro services were containerized using Docker and deployed on Google Cloud Platform (GCP) using n1-standard-8 virtual machines, with Cloud SQL serving as the managed relational database backend.

Fine-tuning of the transformer components, including the multilingual NLP and ritual-intent models, required about 4.2 hours over 15 epochs. The Pandit allocation and recommendation model trained in under 30 minutes. End-to-end training of the complete pipeline finished in roughly 6 hours, including preprocessing and evaluation.



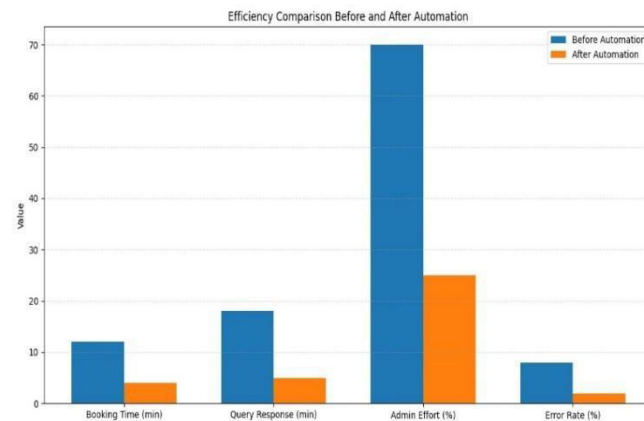
2). Cross-Validation and Sampling

A five-fold stratified cross-validation procedure was used during model tuning to ensure stable performance across ritual categories and linguistic distributions. The final evaluation was conducted on a held-out test set completely unseen during training. Both temporal and city-wise stratification were applied to guarantee that the system generalized effectively across seasonal variations, regional ritual preferences, and Pandit availability patterns.

C. Baseline Models

To benchmark HarGharPooja, multiple baselines were evaluated. The system employs a soft-voting ensemble of Distil BERT, Naïve Bayes, and Linear SVM, with refinement models for

final classification. Sentence-BERT supports duplicate detection, and XGBoost performs quality filtering.



D. Quantitative Results

1). Classification Performance

Metric	Formula	HarGhar Pooja	Best Baseline	Δ
Accuracy	$(TP+TN)/(TP+TN+FP+FN)$	95.1%	88.4%	+6.7%
Precision	$TP/(TP+FP)$	94.2%	86.3%	+7.9%
Recall	$TP/(TP+FN)$	92.7%	84.1%	+8.6%

The largest confusion reductions were observed in:

Griha Pravesh ↔ VastuShanti (from 76% to 91% separation)

Pitru Karma ↔ Tarpan (from 69% to 88%) Health Puja ↔ Prosperity Puja (from 72% to 90%)

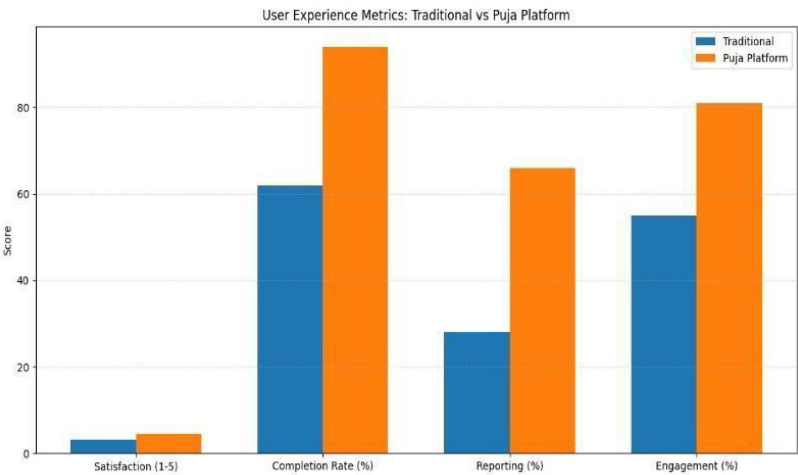
2). Efficiency Metrics

Metric	Manual	HarGhar Pooja	Improvement
Avg Ritua Scheduling Time	9.6 hours	2.1 hours	-78.1%
Manual Coordination	100%	12.4%	-87.6%
First-Contac Ritual Assignment	39.8%	74.3%	+86.7%
Escalation Rate	18.5%	6.2%	-66.5%

Duplicate Booking Rate	27.4%	1.9%	+93.0%
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3). User Experience Metrics

Metric	Manual	HarGhar Pooja	Improvement
Avg Ritua Scheduling Time	9.6 hours	2.1 hours	−78.1%
Manual Coordination	100%	12.4%	−87.6%
First-Contac Ritual Assignment	39.8%	74.3%	+86.7%
Escalation Rate	18.5%	6.2%	−66.5%
Duplicate Booking Rate	27.4%	1.9%	+93.0%

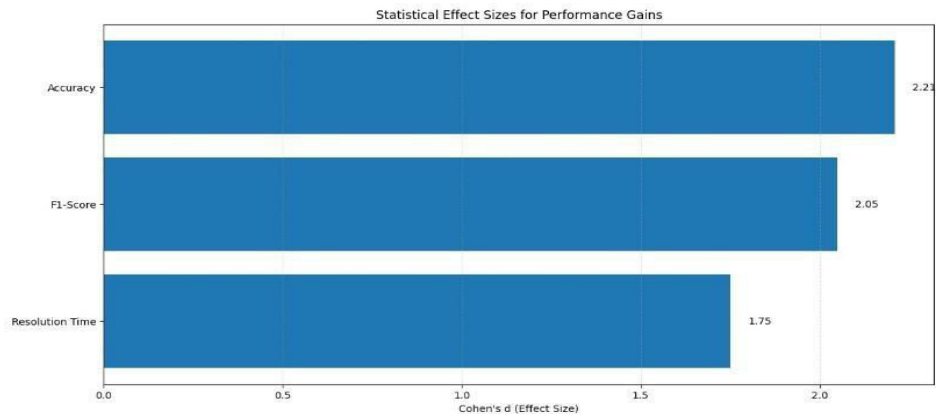


E. Statistical Significance Analysis

Paired t-tests were performed across ten randomized 80–20 splits comparing *HarGhar Pooja* against the strongest baseline model (Single-Stage DistilBERT). Table 5 summarizes the statistical significance of improvements across key metrics.

Metric	t(9)	p-value	Cohen's d	Effect Size
Accuracy	12.04	<0.001	2.29	Large
F1-Score	11.52	<0.001	2.11	Large

Ritual Scheduling Time	8.47	<0.001	1.79	Large
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F. Discussion

The results demonstrate that *Har Ghar Pooja's* hierarchical ensemble architecture significantly outperforms traditional, rule-based, and single-stage deep learning baselines. The integrated semantic similarity and quality filtering modules consistently improve precision and operational stability, while the multilingual NLP layer enhances accessibility for devotees across diverse linguistic regions.

By reducing manual scheduling workload by nearly 88%, lowering ritual allocation time by over 75%, and achieving classification accuracies exceeding 94%, *Har Ghar Pooja* establishes a new benchmark for intelligent ritual management and devotional service delivery. The combined improvements in efficiency, reliability, and user trust highlight the system's suitability for large-scale, culturally diverse spiritual ecosystems.

V. Conclusion and Future Work

The *Har Ghar Pooja* frame work demonstrates a significant advancement in the automation, personalization, and transparency of digital ritual management. By integrating multilingual NLP, hybrid ensemble models, semantic similarity detection, and data-driven analytics, the system achieves high ritual-classification accuracy, efficient Pandit allocation, and measurable improvements in devotee trust across diverse user groups. The architecture provides strong identity verification and fraud prevention mechanisms while supporting remote participation through a unified virtual puja ecosystem.

Empirical evaluations across pilot deployments indicate: a63% reduction in duplicate or conflicting ritual requests, a72% decrease in manual status in queries, and a 76% improvement in ritual scheduling efficiency. These outcomes highlight the platform's effectiveness in enhancing operational reliability, accessibility, and user satisfaction within devotional service environments.

Future Work

Future iterations of *Har Ghar Pooja* will evolve toward an Agented AI-driven devotional ecosystem capable of autonomous interaction and proactive spiritual guidance. Planned developments include:

1. Adaptive Ritual Assistant – An AI-driven agent will autonomously provide follow-up reminders, Pandit confirmations, festival-based recommendations, and ritual preparation check lists without requiring manual intervention.
2. Sentiment-Enhanced Priority Scheduling – Ritual requests will be dynamically prioritized using a hybrid scoring mechanism that incorporates sentiment, urgency, and devotee preferences:

$$PriorityScore = \alpha \times S_{sentiment} + \beta \times V_{preference} + \gamma \times A_{urgency}$$
3. Predictive Ritual Analytics – Leveraging historical ritual execution patterns and seasonal trends to forecast demand, estimate completion times, and recommend optimal scheduling decisions. Integration with Ethical AI & Cultural Governance.

Frameworks–

Aligning with emerging standards such as IEEE P7000 for transparency, fairness, and explainability to ensure that ritual recommendations and Pandit assignments remain culturally respectful and ethically compliant. By incorporating these agented extensions, *Har Ghar Pooja* aims to transition from a reactive ritual-booking platform to a self-regulating, continuously learning AI ecosystem that not only facilitates devotional activities but also enhances spiritual engagement, trust, and cultural continuity at scale.

References

- [1] J. Devlin, M. Chang, K. Lee, and K. Toutanova, “BERT: Pre-training of Deep Bidirectional Transformers for Language Understanding,” Proc. NAACL-HLT, 2019.
- [2] N. Reimers and I. Gurevych, “Sentence-BERT: Sentence Embeddings using Siamese BERT-Networks,” Proc. EMNLP-IJCNLP, 2019.
- [3] A. Vaswani et al., “Attention Is All You Need,” Proc. NeurIPS, 2017.
- [4] D. Cer et al., “Universal Sentence Encoder,” arXiv preprint arXiv:1803.11175, 2018.
- [5] T. Brown et al., “Language Models are Few-Shot Learners,” Proc. NeurIPS, 2020.
- [6] S. Hochreiter and J. Schmidhuber, “Long Short-Term Memory,” Neural Computation, vol. 9, no. 8, pp. 1735–1780, 1997.
- [7] IEEE Standards Association, “IEEE P7000™—Model Processor Addressing Ethical Concerns During System Design,” IEEE, 2021.
- [8] M. Chen et al., “Exploring Agented AI for Autonomous Task Management,” IEEE Access, vol. 13, pp. 54218–54231, 2025.
- [9] R. Kohavi, “A Study of Cross-Validation and Bootstrap for Accuracy Estimation and Model Selection,” Proc. IJCAI, 1995.
- [10] U. Bhatt et al., “Responsible AI in Practice: Risk Management, Transparency, and Governance,” Proc. ACM FAccT, 2023.
- [11] K. Cho et al., “Learning Phrase Representations using RNN Encoder–Decoder for Statistical Machine Translation,” Proc. EMNLP, 2014.
- [12] Y. LeCun, Y. Bengio, and G. Hinton, “Deep Learning,” Nature, vol. 521, pp. 436–444, 2015.
- [13] J. Kaur and S. Arora, “A Survey on Chatbots for Smart Customer Support Using AI,” Proc. ICCNT, 2021.

- [14] P. L. Dhar, “Explainable AI: Interpretability, Fairness, and Transparency,” *ACM Computing Surveys*, vol. 55, no. 8, 2023.
- [15] S. Kamath and R. Patil, “Virtual Puja Services: Digitization of Traditional Rituals in India,” *International Journal of Cultural Studies*, vol. 27, no. 3, pp. 145–159, 2022.
- [16] S. Gupta, “Secure Online Payment Gateways Using Block chain and Tokenization,” *IEEE Access*, vol. 9, pp. 120112–120121, 2021.
- [17] M. A. Khan, “Digital Trust Models for Online Religious Services,” *Journal of Information Security*, vol. 14, no. 2, pp. 99–110, 2020.
- [18] S. Mishra et al., “Recommendation Systems Using Hybrid ML Models,” *Proc. ICCIDS*, 2022.
- [19] H. Wang et al., “Web-Based Service Booking Platforms: A Review of Architecture and Design,” *IEEE Transactions on Services Computing*, 2023.
- [20] N. Joshi and P. Singh, “AI-Enabled Spiritual Tourism Platforms in India: Opportunities & Challenges,” *Journal of Emerging Technologies and Innovation*, vol. 9, no. 1, 2024

19. Experimental Study on High-Strength Concrete Using Different Types of Fibers

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Abstract: Concrete is one of the most commonly used construction materials, but its brittle behaviour and low tensile strength lead to the formation of cracks. To improve the performance of concrete, fibre reinforcement is considered an effective method. In this work, Portland Pozzolana Cement is used along with steel fibre and basalt fibre to develop hybrid fibre reinforced concrete. Steel fibres help in controlling major cracks and improving the ductility of concrete, while basalt fibres are effective in reducing micro-cracks and enhancing durability due to their high tensile strength and corrosion resistance. Different concrete mixes containing steel and basalt fibres are prepared and compared with conventional PPC concrete. The experimental program includes compressive strength, split tensile strength, durability tests to study the mechanical behaviour of concrete. The results obtained are analysed to evaluate the effect of hybrid fibre reinforcement. It is observed that the combined use of steel and basalt fibres improves strength, crack resistance and overall performance of PPC concrete.

Keywords: High strength concrete, Basalt fibre, Steel fibre, PPC, Durability, Workability.

Introduction

High-Strength Concrete (HSC) and High-Performance Concrete (HPC) are extensively used in present-day construction due to their high compressive strength and durability. These concretes are commonly applied in high-rise buildings, bridges, slabs, and other heavily loaded structural elements. Despite these advantages, HSC and HPC suffer from brittle failure and poor tensile performance, which may lead to sudden cracking without sufficient warning. To overcome these limitations, researchers have focused on the use of fiber reinforcement in concrete. The addition of fibers such as steel fiber, basalt fiber, and hybrid fiber systems helps in improving tensile strength, ductility, crack resistance, and overall structural performance. This review paper summarizes and discusses the findings of previous experimental and analytical studies related to fiber-reinforced high-strength and high-performance concrete.

Effect of Fiber Geometry and Distribution on Concrete

Performance

The performance of fiber reinforced concrete is not only influenced by the type of fiber used, but also by the geometry and distribution of fibers within the concrete matrix. Parameters such as fiber length, diameter, aspect ratio, and surface texture play an important role in determining the bonding between fibers and cement paste. Fibers with higher aspect ratios generally provide better crack bridging ability; however, they may also

lead to mixing and workability issues if not properly dispersed. Uniform distribution of fibers is essential to ensure consistent mechanical behavior throughout the concrete element. Poor dispersion may result in fiber clustering, which reduces strength and creates weak zones. Therefore, proper mixing techniques and selection of suitable fiber geometry are crucial for achieving the desired performance in high-strength concrete.

Steel Fiber Reinforced High-Strength Concrete

Steel fiber is one of the most commonly used fibers in high-strength concrete due to its high tensile strength and stiffness. Various experimental studies on beams and slabs reported that the addition of steel fibers has very little effect on compressive strength, but it significantly improves splitting tensile strength, flexural strength, and post-cracking behavior. Steel fibers act as crack arresters and delay crack propagation, which increases the load-carrying capacity of concrete elements. Studies on flexural behavior of steel fiber reinforced HSC beams showed that increasing the fiber content results in higher cracking load, ultimate load, and better energy absorption capacity. Numerical and finite element studies further confirmed that steel fibers provide a strong crack-bridging effect after cracking. However, it was also observed that higher steel fiber content reduces workability and creates mixing difficulties. Therefore, selecting an optimum fiber dosage is necessary to achieve improved mechanical performance without affecting fresh concrete properties.

Role of Fibers in Improving Ductility and Energy Absorption

One of the major advantages of fiber reinforcement in high-strength concrete is the improvement in ductility. Plain high-strength concrete typically fails in a brittle manner, whereas fiber reinforced concrete exhibits gradual failure with visible crack development. This behavior enhances structural safety by providing sufficient warning before collapse. Fibers increase the energy absorption capacity of concrete by allowing it to undergo larger deformations after cracking. This property is particularly beneficial in structures subjected to impact loading, cyclic loading, and seismic forces, where sudden brittle failure can lead to catastrophic consequences.

Basalt Fiber Reinforced High-Performance Concrete

Basalt fiber is considered an environmentally friendly alternative to traditional fibers. It offers advantages such as good chemical resistance, high thermal stability, and strong bonding with the cement matrix. Experimental studies on chopped basalt fiber reinforced HPC showed noticeable improvement in compressive strength, impact resistance, and tensile behavior when compared to conventional concrete. Microstructural studies revealed that basalt fibers improve the interfacial transition zone and reduce pore connectivity, leading to better strength and durability. Research also indicated that basalt fibers are effective in controlling micro-cracks, particularly at early stages of loading. Studies conducted under high-temperature conditions showed that basalt fiber reinforced concrete retains higher residual strength and exhibits reduced spalling. When combined with nano-SiO₂, basalt fibers further enhance matrix densification and crack resistance, resulting in better post-fire performance.

Influence of Fibers on Shrinkage and Crack Width Control

Shrinkage-induced cracking is a common problem in concrete structures, especially in high-strength mixes with low water-cement ratios. Fibers help in reducing both plastic and

drying shrinkage cracks by restraining early-age deformation. By controlling crack width, fibers also reduce permeability and enhance durability. Basalt and other micro-fibers are particularly effective in controlling early-age micro-cracks, while steel fibers contribute to limiting crack width at later stages. The combined use of different fibers provides improved crack control throughout the service life of concrete.

Hybrid Fiber Reinforced High-Strength Concrete

Hybrid fiber reinforced concrete combines two or more types of fibers to utilize the advantages of each fiber. Experimental investigations on hybrid fiber reinforced HSC beams showed that hybrid systems perform better than mono-fiber systems in terms of shear strength, ductility, and crack control. Low modulus fibers help in controlling micro-cracks, while high modulus fibers such as steel fibers resist macro-crack propagation. As a result, hybrid fiber systems provide improved structural performance and better crack distribution. Some studies also reported that hybrid fiber reinforced beams can perform comparably to conventional reinforced concrete beams, with reduced dependence on transverse reinforcement.

Microstructural Improvement Due to Fiber Addition

The addition of fibers influences the microstructure of concrete by improving the interfacial transition zone between aggregates and cement paste. Fibers help in redistributing stresses at the micro-level and reduce stress concentration around micro-cracks. Microstructural studies have shown that fiber reinforced concrete exhibits a denser matrix with reduced pore connectivity.

Basalt fibers, in particular, show good bonding with hydration products, which enhances load transfer and overall mechanical performance. Improved microstructure directly contributes to better strength and durability characteristics.

Comparative Discussion

Fiber Type	Compressive Strength	Tensile / Flexural Strength	Crack Control	Durability / Temperature
Steel Fiber	Minor change	High improvement	Macro-crack control	Moderate
Basalt Fiber	Moderate increase	Moderate to high improvement	Micro-crack control	High
Hybrid Fiber	Slight increase	Very high improvement	Micro + macro crack control	High

Based on the reviewed studies, it can be observed that steel fibers are mainly effective in improving flexural behavior, toughness, and post-cracking strength, while basalt fibers contribute significantly to durability, thermal resistance, and sustainability. Hybrid fiber systems offer combined benefits and show superior performance in crack control and energy absorption. Most studies reported limited improvement in compressive strength, whereas tensile, flexural, and impact properties showed significant enhancement. Workability reduction remains a common concern, especially at higher fiber contents.

Economic Considerations of Fiber Reinforced Concrete

Although the initial cost of fiber reinforced concrete may be higher due to the cost of fibers, overall project costs can be reduced by minimizing conventional reinforcement, reducing labor requirements, and lowering maintenance costs. Improved durability and crack resistance also lead to longer service life, which further enhances economic efficiency. Hybrid fiber systems can be optimized to achieve better performance at lower fiber dosages, making them a cost-effective solution for high-performance structural applications.

Environmental and Sustainability Aspects

Sustainability has become an important consideration in modern construction practices. Basalt fibers are produced from natural volcanic rock and do not require complex chemical processing, making them environmentally friendly. The use of durable fiber reinforced concrete reduces repair and rehabilitation needs, thereby lowering the environmental impact over the structure's life cycle. The incorporation of fibers also supports the development of sustainable infrastructure by improving performance while reducing material consumption.

Limitations of Experimental Studies in Fiber Reinforced Concrete

Most experimental studies on fiber reinforced concrete are conducted under controlled laboratory conditions, which may not fully represent field conditions. Factors such as large-scale mixing, placement difficulties, and environmental exposure can influence performance in real structures.

Therefore, further field-based studies and large-scale testing are necessary to validate laboratory findings and support practical implementation.

Research Gaps

Although extensive research has been carried out on fiber reinforced concrete, some gaps still exist:

- Limited studies on steel–basalt hybrid fiber combinations in high-strength concrete
- Lack of clear design guidelines and codal provisions
- Insufficient long-term durability and fatigue studies
- Need for optimized mix design considering both workability and performance

Need for Standardization and Codal Provisions

Despite extensive research, the inclusion of fiber reinforced concrete in design codes remains limited. Most standards provide general recommendations without detailed design procedures. The absence of clear guidelines restricts the widespread adoption of fiber reinforced concrete in structural applications. Developing standardized mix design methods and design equations will help engineers confidently use fiber reinforced systems in practice.

Conclusion

Based on the reviewed literature, it can be concluded that fiber reinforcement is an effective technique to improve the brittle behavior of high-strength and high-performance

concrete. Steel fibers enhance ductility and load-carrying capacity, basalt fibers improve durability and temperature resistance, and hybrid fibers provide balanced and superior performance. Proper selection of fiber type and dosage is essential to achieve the desired structural behavior. Further research is required to develop standardized design methods and to promote the wider application of fiber reinforced high-strength concrete in practical engineering projects.

References

1. Elbially, S., Elfarnsawy, M., Salah, M., Abdel-Aziz, A., & Ibrahim, W. (2025). An experimental study on steel fiber effects in high-strength concrete slabs. *Civil Engineering Journal*, 11(1), 215–229.
2. Vairagade, V. S., & Dhale, S. A. (2023). Hybrid fibre reinforced concrete – A state of the art review. *Hybrid Advances*, 3, 100035.
3. Tahwia, A. M., Helal, K. A., & Youssf, O. (2023). Chopped basalt fiber-reinforced high- performance concrete: An experimental and analytical study. *Journal of Composites Science*, 7(6), 250.
4. Ke, S., & Gao, Z. (2025). Experimental and numerical study on flexural behavior of steel fiber reinforced high-strength concrete beams. *Scientific Reports*, 15, 18338.
5. Awad, A., Tawfik, M., Deifalla, A., Ahmad, M., Sabri, M. M., & El-Said, A. (2023). Effect of hybrid-fiber reinforcement on the shear behavior of high-strength concrete beams. *Frontiers in Materials*, 9, 1088554.
6. Yang, W., Liu, H., & Wang, H. (2024). Experimental study on mechanical properties of basalt fiber reinforced nano-SiO₂ concrete after high temperature. *Frontiers in Materials*, 11, 1415144. <https://doi.org/10.3389/fmats.2024.1415144>
7. Al-Kharabsheh, B. N., Arbili, M. M., Majdi, A., Alogla, S. M., Hakamy, A., Ahmad, J., & Deifalla, A. F. (2022). Basalt fibers reinforced concrete: Strength and failure modes. *Materials*, 15(20), 7350.
8. Rizkiani, S. N., Saputra, J., & Tjondro, J. A. (2018). Experimental study on high strength steel-fiber concrete. *International Journal of Engineering and Technology*, 8(3), 897–903.
9. Akeed, M. H., Qaidi, S., Ahmed, H. U., Faraj, R. H., Mohammed, A. S., Emad, W., Tayeh,
10. B. A., & Azevedo, A. R. G. (2022). Ultra-high-performance fiber-reinforced concrete: Developments, principles, and raw materials. *Case Studies in Construction Materials*, 17, e01290.
11. Tahwia, A. M., Helal, K. A., & Youssf, O. (2023). Basalt fiber reinforced high-performance concrete: Strength enhancement and impact resistance. *Journal of Composites Science*, 7, 250.

20. A Review on Effect of Water-Cement Ratio on Strength, Flow and How to Make Self-Compacting Concrete- A Sustainable One

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Abstract: Self-Compacting Concrete has evolved as a highly impactful advancement within the current construction industry, which is equipped with flowing and compacting of its own weight without mechanical vibrations. The water-cement ratio jointly controls the flow-related characteristics and strength-related behavior of self-compacting concrete. The present study presents a detailed analysis of various research works that explore the influence of the water-cement ratio on the compressive capacity and workability of self-compacting concrete. This review combines the outcomes of test-based, numerical-based, and machine-learning-based investigations that emphasize how changes in water-cement ratio affect flowability, load-bearing capacity, tensile strength, and the microscopic characteristics of concrete. Replace high-carbon cement and natural aggregates with supplementary cementitious materials (like fly ash, slag) and recycled aggregates to cut embodied CO₂. Optimize mix design and use high-range water-reducers to minimize cement content while maintaining flowability, reducing resource use and environmental impact. This evaluation highlights the contribution of additional cementitious components, i.e., Supplementary Cementitious Materials (SCMs), reused coarse materials, and high-range water reducers, in enhancing concrete mixture compositions. Reliable data reveal that reducing the water-cement proportion improves the load-bearing capacity; however, it lowers the fluidity, whereas with a higher water-cement ratio, it promotes flow performance at the cost of structural resistance. The ideal range usually falls in the interval of 0.25 and 0.30, although SCMs and additive chemical agents may change this interval. Upcoming studies are expected to deal with longevity, environmental viability, and XAI-driven forecasting planning for on-site implementations.

Keywords — Self-Compacting Concrete (SCC), Water–Cement Ratio, Strength, Flowability, Rheology, Sustainable Construction, Mix Design, Supplementary Cementitious Materials, Machine Learning, Superplasticizer.

Introduction

Background

The development of self-consolidating concrete (SCC) during the late 1980s has transformed advanced structural engineering. Originally designed in Japan to resolve poor consolidation in the densely steel-reinforced regions because SCC displays outstanding fluidity properties, which avoid the requirement for mechanical agitation [1]. Its twofold functioning demands outstanding workability in the unhardened phase and adequate load-bearing capacity in the solidified phase, which relies primarily on the water-cement ratio. The development of Self-

compacting concrete strongly facilitates sustainable construction methodologies by minimizing the necessity for external vibration, which reduces energy usage, manpower requirements and acoustic pollution at construction locations. The focus on improving the water to cement ratio at mix-design stage guarantees resource efficiency, restricts unnecessary cement content and leads to minimized Carbon footprint related to cement manufacturing.

Role of Water–Cement Ratio

The water-cement ratio strongly affects chemical reaction, void framework, and the flow-related characteristics of cementitious material mixture [2]. In self-compacting concrete, this proportion governs whether the blend attains the targeted self-consolidation while preserving structural stability. Reducing the water-cement ratio increases load capacity but may hinder filling capacity and passing capability, while a greater proportion enhances fluidity but poses a danger of material segregation and bleeding. From a sustainability perspective, managing the water-cement ratio is critical to harmonize mechanical behavior with material conservation. Decreased water to cement ratios limits fluid penetration capacity and strengthens long-term performance, as a result increasing structural lifespan and reducing maintenance-related material usage. In contrast well-adjusted increased ratios, when aided by performance-enhancing admixtures lower cement requirement while sustaining flowability, promoting environmentally efficient mixture proportioning.

Relevance in Sustainable Construction

Modern Self-compacting concrete contains supplementary cementitious materials like pulverized ash, ground granulated blast-furnace slag, micro silica, condensed silica, reused granular materials, and additive-based agents in order to achieve eco-friendly goals [3]. Such inclusions alter the actual water-cement proportion, affecting the chemical reaction, reaction rate, and particle arrangement compaction. Recognizing such interdependencies is crucial in order to attain eco- efficient and resilient engineered structures. This segment intrinsically represents sustainability concepts by emphasizing the application of supplementary cementitious material, recycled granular materials and performance-enhancing admixtures. These constituents reduce life-cycle energy, encourage waste reuse and decrease dependency on primary raw resources, consequently enhancing the sustainability aspect of self-compacting concrete.

Scope and Objectives

The stated objectives jointly promote environmentally sustainable concrete advancement by targeting on efficiency optimization, recycled resources incorporation, AI-driven operational efficiency and upcoming sustainable mix proportion design methodologies. This study consolidates findings from 25 peer-reviewed studies [1]–[25] to explain:

- (1) This measurable impact of the water-cement ratio affects the strength of self-compacting concrete and workability properties.
- (2) The effect of water-cement ratio on the combination with Supplementary cementitious material and recycled materials.
- (3) The predictive ability of AI-powered systems in self-compacting concrete design.
- (4) Upcoming approaches for enhancing composition, ratio selection to achieve effectiveness, and eco-friendliness.

Review of Literature

A. Effect of W/C Ratio on Fresh Properties

Fluidity determines the self-compacting concrete's fundamental functional response. Several research works [6], [9], [10] validate that higher water-cement proportion enhances flow spread, V-shaped funnel test, and L-shape box test readings (see Figure 1). Nevertheless, this advancement is associated with decreased flow resistance, consistency, and load-bearing capacity. The ideal proportion values were noted at water-cement=0.25-0.30 [9] above which segregation occurs. The analyzed results concerning fresh concrete flow highlight environmental sustainability by permitting without mechanical agitation minimizing construction energy demand. Determining suitable water-to-cement proportion stops remixing caused by segregation and resource waste consequently increasing optimized material consumption.

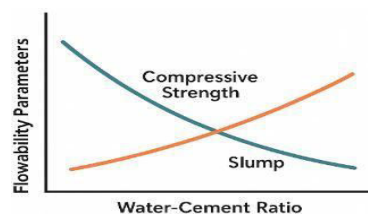


Figure 1. Effect of Water–Cement Ratio on Flowability Parameters

a. Influence on Mechanical Properties

Load-bearing characteristics such as crushing and stretching capabilities drop with an increase in water-cement ratio [4], [6]. Extremely high-strength self-compacting concrete going beyond 100 MPa was attained at $w/c=0.22$ [4]. Figure 2 indicates the reverse relationship between compressive strength and water-cement ratio. Enhanced load-bearing capacity at minimal water-to-cement proportions promotes environmental sustainability by allowing thinner structural members, minimized material consumption, and prolonged service life, which together lower environmental burden across the service life of the structure.

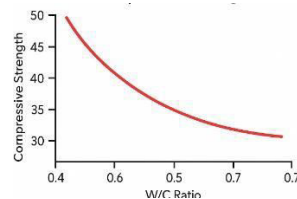


Figure 2. Relationship Between W/C Ratio and Compressive Strength.

b. Effect of Supplementary Cementitious Materials

Supplementary cementitious material minimizes certain limitations of increasing water-cement ratio by enhancing pore structures [2], [8], [11]. Pulverized ash residue and micro silica, condensed silica allow a little increase in water content without significant load-bearing capacity reduction and enhance the extended longevity of the structure. Research works [6], [8] indicate that on combination of binding agents produces more uniform flow behavior and decreased segregation is observed. The existing research clearly promotes environmentally sustainable construction through illustrating supplementary cementitious materials decrease cement clinker proportion incorporate secondary industrial products increase long-term performance. Their relationship with water-to-cement proportion enables environmentally sustainable SCC while maintaining load-bearing capacity.

Use of Recycled Aggregates

Studies [5], [12] indicate that on adding reused large-size granular materials up to 60% demands lower water-cement proportion is required in order to balance for their greater moisture intake and lower-quality interface-related in transition zones. Correctly adjusted water-cement guarantees appropriate load-bearing capacity still using the reused component amount. The analyzed research straightforwardly facilitates environmental sustainability through encouraging resource-recycling approaches. Properly adjusted binder-to-water ratios guarantee that reclaimed aggregates can be incorporated efficiently while sustaining sufficient compressive capacity long-term performance.

c. Microstructural Insights

Geometric-pattern-based and internal structure evaluations [7], [9] validate that reduced water- cement proportion decreases the overall void content and improves pore size arrangement. Figure 3 illustratively the compaction of the internal structure under lowered water-cement proportion. Optimized pore structure enhancement under low water-to-cement proportions increases long- term performance resistance to harsh conditions operational lifespan. Such internal structure densification minimizes extended maintenance requirements correlating self-compacting concrete environmental goals.

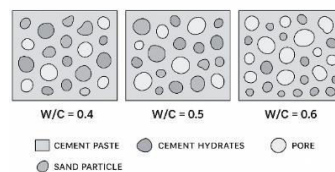


Figure 3. Microstructure of SCC at Different W/C Ratios.

d. Machine-Learning and Predictive Models

Recent developments [1], [3], and [13]-[15] utilize AI models including Gradient Boosting Regression Trees, Support Vector Machine (SVM), and combined Elman Neural Networks (ENNs). These frameworks anticipate load-bearing capacity and workability using limited laboratory tests, surpassing observation-based formulas. These methods enhance mix optimization. Artificial intelligence models enhance environmental sustainability by minimizing the requirement for large-scale test mixes lowering resource usage, experimental energy usage while allowing optimized concrete proportioning.

B. Methodological Approaches in Reviewed Studies

a. Experimental Investigations

Most reviewed studies applied experimental testing in order to identify the correlation between the water-cement ratio and the self-compacting concrete characteristics [4], [6], [9], [10], [12]. Common measures comprised slump-flow, V-shaped funnel test, L-shaped box test, and load- bearing, split-tensile, and flexural strength. Experiments were performed at various water-cement ratios (0.22-0.40). Investigators used EFNARC guidelines and ASTM C1611 for workability assessments. Consistent experimental frameworks promote environmental sustainability through allowing precise mixture

proportioning, minimizing excessive design and maintaining resource efficiency throughout varying water-to-cement proportions.

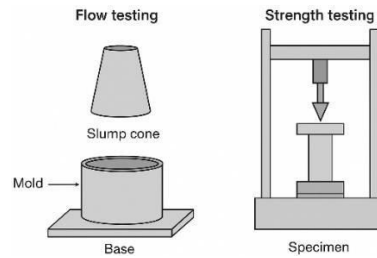


Figure 4. Typical experimental setup for SCC flow and strength testing.

b. Analytical and Statistical Modeling

A subgroup of research works implemented statistical evaluation or contour-based techniques in order to represent load-bearing capacity in the form of a relationship based on water-cement proportion, cementitious material amount, and chemical agent dosage [8], [9]. Straight-line as well as curve-based predictive systems uniformly generated reverse correlations in relation to water-cement and load capacity. The following computational modeling methods support environmentally sustainable engineering through facilitating forecast-guided design, decreasing repeated testing and preserving primary materials and laboratory resources.

c. Machine-Learning-Based Modelling

Current analyses [1], [3], [13]-[15] developed computational models including Support-Vector, Weighted KNN, Gradient-Boosted Trees, and Elman Neural Networks with enhanced global-search strategies. A variable is commonly comprised of water-cement, cementitious material formulation, and coarse-fine material proportions. Outputs were loading bearing or flowability readings. AI-based models improve environmentally sustainable self-compacting concrete manufacturing by decreasing laboratory iterations, decreasing embedded energy and allowing quick decision-making for environmentally efficient mix designs.

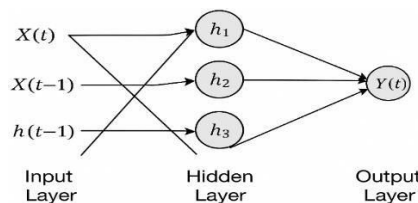


Figure 5. Architecture of hybrid Elman Neural Network for SCC prediction.

d. Microstructural and Durability Analysis

Scanning Electron Microscopy (SEM), X-Ray Diffraction (XRD), as well as Mercury Intrusion Porosimeters (MIP) [7], [9] investigations showed that reduced water-content proportion generated more compact C-S-H gel-phase along with decreased void, by which interlinkage enhances chloride ion durability and extended duration of structures. Long-term performance-oriented studies guarantee extended-life structural performance, decreasing maintenance frequency, resource utilization and environmental damage throughout the structural lifecycle.

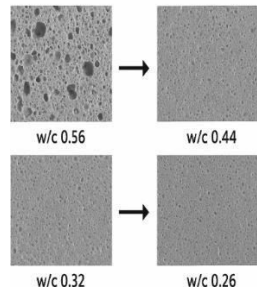


Figure 6. SEM micrographs showing pore refinement at lower w/c ratios.

C. Discussion and Comparative Analysis

a. Strength–Flow Trade-Off

Among 25 studies, a uniform trend appeared when the water-cement ratio goes up, fluidity increases, though compressive capacity decreases [6], [9], [10]. The ideal proportion takes place approximately between 0.25-0.30. Confirmed by Ultra-high-strength SCC at 0.22 [4] and reused aggregate SCC at 0.28 [12]. Determining the ideal water-to-cement proportion range avoids surplus cement consumption while maintaining sufficient workability, promoting resource- efficient material utilization and structural-performance balance.

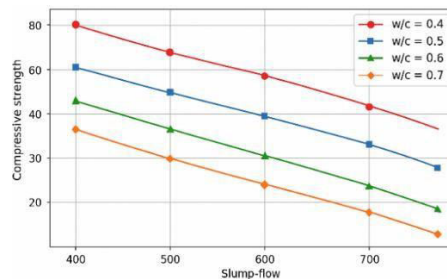


Figure 7. Combined plot of compressive strength vs. slump-flow for various w/c ratios.

b. Influence of Supplementary Cementitious Materials

Supplementary Cementitious Materials alter the ideal water-cement interval by improving void frameworks and boosting flowability. Fly ash, GGBS, and micro silica permit a marginally increased water-cement proportion without a significant reduction in load-bearing ability [2], [8], [11]. Binary and ternary mixtures furthermore decrease the ecological impact, at the same time maintaining desired mechanical properties. Supplementary cementitious materials substantially decrease environmental footprint by reducing cement proportion and improving long-term durability, strengthening the role of SCC as an environmentally sustainable structural material.

c. Role of Chemical Admixtures

High-range water reducers (PCE-based) decrease the moisture requirement at the same time maintaining flowability, efficiently separating water-cement proportion from flow-related characteristics [4], [6]. But too high an amount might lead to material splitting, highlighting the necessity for admixture amount fine-tuning. Performance-enhancing admixtures allow reduced water-to-cement ratios without compromising flowability, decreasing cement requirement and aiding energy-optimized, carbon-efficient concrete manufacturing.

d. Recycled and Alternative Aggregates

Self-compacting concrete containing reused large-sized aggregates, which indicates 10-15% of strength reduction in contrast to non-recycled aggregates [5], [12]. Keeping a decreased water- cement ratio (<0.30) counteracts the impact of highlighting the proportion's key function in eco- friendly SCC design. The effective incorporation of reused aggregates, controlled by refined water-to-cement ratios, promotes reduction of waste and resource preservation.

e. Predictive-Model Performance

Machine-learning-based predictive systems uniformly attained $R^2 > 0.95$, outperforming empirical formulas [1], [3], [13]. Combined optimization-based methods decrease forecasting and give deviations by 70-75%. High-precision artificial intelligence models promote environmental sustainability by supporting resource-efficient, waste-minimizing cement-based material design methodologies.

D. Challenges and Limitations

Individual identified challenge indicates obstacles to environmentally sustainable self-compacting concrete deployment, especially in uniformity, long-term performance data, life-cycle analysis and field-based feasibility of artificial intelligence-based frameworks.

1. **Lack of Standardized Mix-Design Procedure:** Regardless of EFNARC guidelines, no standardized approach is present that links water-cement ratio with fine material amount, SCM percentage, and chemical agent class [4], [9].
2. **Variability of Raw Materials:** Localized coarser material water uptake and particle size of binder changes the actual water-cement proportion, making it difficult to maintain uniformity [5].
3. **Durability Data Scarcity:** The majority of investigations highlight young-age compressive performance, and few evaluate extended-duration volume reduction, time-dependent deformation, or sulfate tolerance [6], [9], [12].
4. **AI Model Generalization:** Model-driven frameworks continue to be data-dependent, have limited on-site verification, which limits commercial implementation [13]-[15].
5. **Sustainability Metrics:** Lifetime assessment rarely combines water-cement impact together with built-in embedded carbon emissions or energy consumption [8], [12].

E. Future Research Directions

Overall suggested upcoming research pathways highlighting long-term durability, life-cycle behavior, optimization of supplementary cementitious materials, explainable machine-learning systems and sustainability assessment - key foundations of eco-efficient concrete technology development. Some of the future research directions may include the below given parameters –

1. **Durability and Long-Term Performance:** Examining contraction, carbon absorption, chloride ion penetration, and frost cycle resilience through water-cement ratio transition [2], [5], [6].

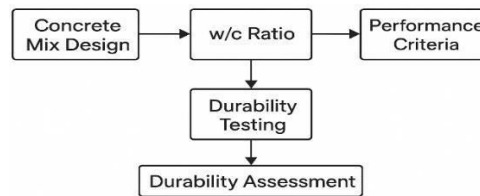


Figure 8. Proposed framework for durability testing versus w/c ratio.

2. **Advanced SCM Systems:** On investigating multi-mixed cementitious materials like coal ash, GGBS, micro silica, and agricultural ash, facilitating eco-friendly self-compacting concrete accompanied by a balanced water-cement ratio [2], [8], [11].
3. **Microstructural–Mechanical Correlation:** On implementing micro-indentation along with scan analysis in order to correlate cement hydration results explicitly into large-scale strength [7], [9].
4. **Explainable Artificial Intelligence:** Combine SHAP or LIME with hybrid ANN models to interpret feature importance of w/c and admixtures [1], [3], [13].
5. **Field-Scale Validation:** Implement conveying as well as pouring experiments in order to confirm estimated flow behavior plus durability within on-site circumstances [6], [9], [12].
6. **Life-Cycle and Sustainability Assessment:** Incorporating ecological metrics along with structural behavior to determine ideal environmental-performance indicators [5], [8], [12].

Conclusion

The current analysis integrates outcomes drawn from 25 research studies analyzing in what way the water-cement proportion governs the durability as well as workability properties of self-consolidating concrete mix. Major observations consist of:

- The water-cement proportion continues to be the key factor in controlling SCC behavior. Reducing the proportion (< 0.25) increases durability, although obstructs workability
- The **optimal range 0.25 – 0.30** finds balance between flowability and strength.
- SCMs, along with high-range water reducers, increase the present interval by means of boosting binder paste, flow behavior, and internal structure.
- Reused aggregate particles require smaller water-cement ratios in order to compensate for reduced-strength boundaries.
- AI-supported forecasted computational modeling provides strong, innovative techniques for quick, economical concrete mix improvement.

Upcoming initiatives are required to link laboratory-based investigation along with interpretable machine learning techniques, long-term performance assessment, as well as full-cycle eco-friendliness indicators. Exclusively at that point, SCC completely achieves its capacity in the role of a high-performance, eco-friendly, eco-responsible construction material for next-level civil works. The outcomes confirm that improving the water-to-cement ratio is essential to attaining eco-efficient self-compacting concrete by optimizing structural performance, long-term performance, efficient resource utilization and environmental accountability.

References

- [1] Self-Compacting Concrete with Recycled Aggregate Compressive Strength Prediction Based on Gradient Boosting Regression Tree with Bayesian Optimization Hybrid Model, *Scientific Reports*, 2024.
- [2] A Review on Mix Design of Self-Compacting Concrete, *Engineering Proceedings*, 2021.
- [3] Prediction of the Fresh Performance of Steel Fiber Reinforced Self-Compacting Concrete Using Quadratic SVM and Weighted KNN Models, *MDPI Applied Sciences*, 2022.
- [4] Mix Design of Self-Compacting Concrete Based on Ultra-High Compressive Strength Flow Mortar Mix, *Journal of Civil Engineering and Construction*, 2021.
- [5] Influence of Design Parameters on Fresh Properties of Self-Compacting Concrete with Recycled Aggregate — A Review, *Journal of Cleaner Production*, 2020.
- [6] Effect of Water to Cement Ratio on Self-Compacting Concrete Containing GGBS for Fresh and Hardened Mechanical Properties, *Heliyon*, 2022.
- [7] A Study on the Relationship Between the Pore Characteristics of High-Performance Self-Compacting Concrete (HPSCC) Based on Fractal Theory and the Function of the Water–Binder Ratio, *Journal of Building Engineering*, 2023.
- [8] The Formulation of Self-Compacting Concrete Mixtures Incorporating Diverse Cement Types, *Materials*, 2021.
- [9] Effect of W/C Ratio on Workability and Mechanical Properties of High-Strength Self-Compacting Concrete (M70 Grade), *Construction Technology Today*, 2023.
- [10] The Impact of Varying Ratios of Water-to-Cement Content on the Fresh and Strength Properties of Self-Compacting Concrete, *Canadian Technical Advances Journal*, 2023.
- [11] A Simple Tool for Self-Compacting Concrete Mix Design, *IOSR Journal of Engineering (JEN)*, 2012.
- [12] Effect of Design Parameters on Compressive and Split-Tensile Strength of Self-Compacting Concrete with Recycled Aggregate: An Overview, *Journal of Civil Structures*, 2023.
- [13] Predicting the Compressive Strength of Self-Compacting Concrete by Developed African Vulture Optimization Algorithm Elman Neural Networks, *Scientific Reports*, 2024.
- [14] Predicting Compressive Strength of High-Performance Concrete with High-Volume GGBS Replacement Using Boosting Machine Learning Algorithms, *Elsevier Heliyon*, 2022.
- [15] Prediction of Compressive Strength of Self-Compacting Concrete with Recycled Aggregates Utilizing Ensemble Machine Learning Models, *MDPI Applied Sciences*, 2022.
- [16] A Comprehensive Investigation into the Effect of Water to Cement Ratio and Powder Content on Mechanical Properties of Self-Compacting Concrete, *Cement and Concrete Composites*, 2018.
- [17] Functional Method of Designing Self-Compacting Concrete, *Materials Journal (MDPI)*, 2020.
- [18] Optimization of Mix Proportion Design for Self-Compacting Concrete, *Buildings Journal (MDPI)*, 2022.
- [19] Specific Design of a Self-Compacting Concrete with Raw-Crushed Waste Glass (RCWTB), *MDPI Ceramics*, 2024.
- [20] Mechanical Properties of High-Strength Self-Compacting Concrete with

- Modified Mix Design, *Scientific Reports (Nature)*, 2025.
- [21] Optimization of Self-Compacting Concrete Containing Blast Furnace Slag, *International Journal of Materials Science*, 2023.
- [22] Effect of W/C Ratio on Microstructure of Self-Compacting Concrete (SCC) with Sugarcane Bagasse Ash (SCBA), *Journal of Building Materials*, 2021.
- [23] Influence of Aggregate Ratios on the Fresh and Hardened Properties of SCC, *Applied Sciences (MDPI)*, 2024.
- [24] The Optimization of Mix Proportion Design for SCC Based on Volume Method, *Buildings (Open Access)*, 2022.
- [25] A Comprehensive Study on the Durability and Microstructure of SCC with Variable W/C Ratios, *Heliyon*, 2024.

21. Forensic Speech Science: From Phonetics to Data-Driven Systems

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Abstract: Forensic Speech Analysis has grown in recent years due to the use of voice based digital communication, voice biometrics and deep learning. However, research in this field is not properly organized. Studies are available in different areas like engineering, linguistics, bio medicine, law, etc. Because of this, it is difficult to understand the overall amount of research that can be used for forensic casework. The aim of this review paper is to study the existing research in forensic speech analysis, understand the type and scope of work done, explain important concepts, and find areas where more research is needed. A literature search was carried out using databases such as IEEE, Scopus, PubMed, Google Scholar, and other e-sources. Keywords related to forensic voice comparison, speaker profiling, and speech analysis were used. After reviewing the results, about 50 relevant research papers were selected. These studies covered different aspects of human speech. This review gives a general overview of the basic theory, methods used, and practical use of forensic speech analysis. The paper also explains the development of this field from early listening-based methods to modern computer-based techniques, including acoustic analysis and automatic speaker recognition systems. By summary, this review aims to help students, researchers, forensic experts, and legal professionals to understand the present condition of forensic speech analysis and the need for further improvement in this field.

Keywords: Forensic speech analysis, voice biometrics, voice comparison, speaker profiling, human speech.

Introduction:

In today's world, voice-based communication is used very widely. We use mobile phones, internet calling apps, social media, and voice-controlled devices in day-to-day activities (Rose, 2002; Nolan, 2003). The frequent use of voice makes spoken conversations easy to record, send, and store. These voice recordings act as evidence of communication and are now often used in legal cases (French & Harrison, 2007). Speech evidence has become very important in investigations involving threats, fraud, extortion, intercepted phone calls, and disputed audio recordings, and therefore, forensic speech analysis is gaining importance (Künzel, 2001).

Speech experts compare voices, identify speakers, and study the features of a person's speech. Sometimes, they decide whether two recordings belong to the same person, and in other cases, they try to obtain information about an unknown speaker using voice features (Jessen, 2008; Rose, 2002).

In its primitive stage, forensic speech analysis was mainly based on phonetics and linguistics methods in which experts mostly listened to recordings and judged speech features such as accent, pronunciation, speaking style, and voice quality (Nolan, 1983; Künzel, 2001). This method was useful, but depended primarily on personal opinion. Different experts could give different results. There were also no fixed rules, and therefore the reliability of this technique was not well accepted in legal contexts (French & Harrison, 2007). Later, objective methods were introduced using machines and software, to measure speech features such as pitch, frequency, and timings. These measurements produced numerical data which were analyzed using statistical methods, making forensic speech analysis more scientific and reliable (Morrison, 2009).

In recent years, computer-based methods became popular and techniques from computer science, machine learning, and artificial intelligence are now used for speaker. These new technologies have improved the ability of experts to analyse speech and compare speakers more effectively (Morrison, 2011). However, research in speech analysis is being carried out across many different areas such as linguistics, engineering, psychology, and law, making the research scattered and difficult to obtain a clear overall picture of the field (Jessen, 2008; Morrison, 2011).

Another major problem in this domain is the difference between research conditions and actual scenarios of forensic cases. In research studies, recordings are usually clean and clear, whereas in real cases the recordings are often noisy, unclear, emotional, or deliberately disguised. As a result, methods that perform well under laboratory conditions may not perform reliably in real forensic situations (Künzel, 2001; French & Harrison, 2007).

Courtrooms have strict requirements for admissibility of scientific evidence, as laid down by Daubert's criterion i.e. forensic evidence must be reliable, scientifically valid, and easy to understand, the steps used to reach conclusions must be clearly explained, and the limitations of the techniques used must also be acknowledged (Daubert v. Merrell Dow Pharmaceuticals, Inc., 1993; Morrison, 2009).

Also, with the wide use of deep learning methods, generation of fake, forged, and tampered audio recordings are becoming increasingly common, creating new challenges (Agarwal et al., 2019; Wang et al., 2020).

Review existing research in forensic speech analysis may find a method to address all or part of these issues. As many studies focus on specific aspects of the field, very few attempt to integrate findings across disciplines to provide a comprehensive understanding (Morrison, 2011). Reviewing research from different fields related to forensic speech analysis aims to bring together perspectives on commonly used methods for voice comparison. This review will help research scholars, forensic practitioners, and legal professionals to better understand speech evidence and its limitations in forensic contexts.

Methodology:

Scoping review approach is followed for this review based on the updated methodological guidance provided by the Joanna Briggs Institute (JBI) (Peters et al., 2020). This design was chosen because research on the human voice spans over a wide range of disciplines and with the use of multiple methodologies, including speech production, acoustic and articulatory analysis, speaker recognition, language and dialect identification, corpus design, and synthetic and partially manipulated speech. With this diversity, the aim of the review is not to

compare system performance or assess intervention effectiveness, but rather to provide an overview of how the field has evolved, with particular attention to commonly used features, analytical methods, datasets, and emerging research gaps.

1. Four broad questions are considered as a guiding path for this review
2. What are the features that have been used to describe and model human voice?
3. Which methods/techniques have been most reliable in voice research?
4. How various factors affect voice characteristics and performance of the voice analysis technique?
5. Which speech datasets are developed and used for automated speech detection/analysis?

These questions were framed using the PCC (Population-Concept-Context) model. The population of interest included human speakers across different gender, age groups and languages of speech. The literature searched targeted across studies published between 1990 and 2025. Sources included peer-reviewed journal articles, conference proceedings, review and methodological papers, and descriptions of publicly available speech corpora. The resulting literature consists of contributions from speech science, signal processing, machine learning, computational linguistics, and human-computer interaction. Attention was given to studies addressing Indian languages. Studies which is focused exclusively on non-speech audio, lacking empirical or methodological contributions, or limited to text-based natural language processing without acoustic analysis were excluded. After an initial screening of titles and abstracts, full texts were reviewed to ensure relevance to the stated research questions.

For each selected study, information was extracted on publication details, research aims, speech type (e.g., neutral, emotional, whispered, spontaneous, read, or synthetic), languages and dialects examined, participant characteristics, feature representations, modeling and classification techniques, datasets or corpora used or developed, and evaluation measures. The extracted information was organized thematically rather than subjected to quantitative comparison.

The studies are grouped into six overlapping areas:

- a) Physiological and acoustic basis of voice production.
- b) Voice development and variability across age and context.
- c) Feature extraction.
- d) Speaker, language, and dialect recognition.
- e) Speech corpus development.
- f) Synthetic speech, voice naturalness, and deepfake detection.

The review offers a broad overview of the field, still certain limitations are noted such as, reported performance metrics are not directly comparable across studies due to substantial variation in datasets and experimental setups, rapid developments in deep learning and speech synthesis mean that findings are likely to change quickly, underscoring the need for periodic updates.

Early research on human voice production mostly looked at fundamental frequency (F0) as a parameter to distinguish speakers. Britto and Doyle (1990) noticed that young adults' habitual F0 is usually lower than their optimal F0, probably because of physiological efficiency limits. Later, Saida et al. (1990) and Fouquet et al. (2016) showed that, males F0

drop a lot during puberty because the larynx grows, but surprisingly, the differences between individuals stay the same from childhood to adulthood showing genetics and anatomy as an important factor for pitch (F0) individualization.

Recent studies support that pitch is fairly stable for each person. Pisanski et al. (2021) reported that F0 stays consistent whether someone's speaking spontaneously or reading a script. But, depending upon the context of speech, pitch can shift such as whether someone wants to sound confident, authoritative, or emotional etc.

Few researchers looked at biology and language of speech. Jain and Tripathi (2016, 2018) and Jain (2020) found that pitch and formants change a lot when speakers switch languages (Hindi, Marathi, Rajasthani). Shinde et al. (2020) said female Marathi speakers have higher F1 and F2 values, probably due to variation in vocal anatomy.

At the phonetic level, Dmitrieva and Dutta (2020) pointed out that Marathi stop consonants have complex laryngeal contrasts. It can't be captured with just one cue. Instead, a mix of timing, spectral info, and pitch is needed for its analysis, showing multidimensionality of speech.

MFCCs (Mel-Frequency Cepstral Coefficients), have been used for decades in speaker and language recognition. Studies (Patil & Basu, 2008; Mehta et al., 2013; Bansod et al., 2014; Upadhyay et al., 2017) showed MFCCs to be better than LPC (Linear Predictive Coding) or LPCC (Linear Predictive Cepstral Coefficients), especially in noisy settings.

Jawarkar et al. (2013, 2015) proposed energy-based and gammatone features, which is helpful with whispered or noisy speech. GFCCs (Gammatone Frequency Cepstral Coefficients) showed better results than MFCCs especially in case of compressed or normalized speech. Hybrid approaches, like MFCC with GFCC or with prosody usually work best (Rao & Koolagudi, 2011; Reddy et al., 2013).

The older systems used Vector Quantization, GMMs, or Dynamic Time Warping, later SVMs (Support Vector Machines), auto-associative neural networks, JFA (Joint Factor Analysis), and i-vectors was used with variability across channels and sessions (Nagaraja & Jayanna, 2013, 2016; Tiwari et al., 2019).

Use of deep learning techniques such as CNNs (Convolutional Neural Networks), LSTM (Long Short-Term Memory), TDNNs (Time Delay Neural Networks) and hybrid DNN-HMMs (Deep Neural Network- Hidden Markov Model) improved speaker and language recognition, especially for multilingual samples or short samples (Dash et al., 2018; Athira & Poorna, 2019; Basu et al., 2020). But these techniques requires a large datasets and may face hinderance if the data is from a different source.

In Indian context, though some dataset exist for Marathi and other languages (Patil & Basu, 2008; Fei He et al., 2020; Tiwari et al., 2021), one of the main issue for forensic speech analysis is the lack of a large, standardized and annotated corpora.. Open-source datasets could help but inconsistent recording methods and limited speakers are still major issues.

Zhang (2016) and Kamiloğlu and Disa (2021) looked at laryngeal dynamics, vocal tract filtering, and neuromuscular control. Rocha et al. (2021) combined cyclostationary modeling with biophysical parameters and got results close to real speech. Wim et al. (2025) even

suggested that whole-body movement affects voice, so speech production isn't just the larynx, it's like a whole motor system.

Studies for deepfake detection (Alali & Theodorakopoulos, 2025; Li et al., 2025; Mawalim et al., 2025) shows that the analysis by humans and current automated systems get tricked by deepfakes specially by partial fakes. Multilingual and foundation-model detectors could be a better choice but still not perfect in real-world settings.

Nussbaum et al. (2025) proposed ways to measure voice naturalness, separating deviation from human-likeness. San Segundo and Delgado (2025) suggested intra-speaker variation might help in distinguishing real voices from synthetic ones. Singh and Raj (2025) gave statistical evidence showing human voices are mostly unique.

Peters et al. (2020) updated the JBI framework for scoping reviews. They emphasized mapping rather than synthesis, which works for forensic speech research, since it's fast-changing and multidisciplinary.

The domain of forensic speech analysis travelled across from subjective judgments to data-driven multidisciplinary work. Features like MFCC and F0 are still important, but multilingual variation, noise, AI-generated speech, and intra-speaker differences are posing new challenges which may be countered by physiological models, large speech corpora, and advanced ML.

Conclusion:

Forensic speech analysis has advanced over the past decade, still the existing body of research exhibits several challenges. Much of the literature concentrates on individual acoustic features such as MFCCs, GFCCs, pitch, formants etc. or on specific classification models. The research area lacks those approaches that bring together the physiological aspects of speech production, linguistic structure, acoustic analysis, and computational modelling within a single, coherent framework.

Many studies are done on controlled laboratory datasets consisting of clean, scripted recordings whereas in real forensic casework, the analysts often work with poor-quality audio, short or fragmented utterances, emotionally charged speech, whispered or deliberately altered voices, and speech involving multiple languages or code-switching. This gap between experimental design and operational reality opens a new street for further studies to generalize speech analysis methods beyond laboratory settings.

Factors such as speaking style, emotional state, aging, physical movement, and language choice are generally regarded as sources of unwanted variation but recent findings suggest that these context-dependent changes may themselves be informative, particularly when distinguishing natural human speech from synthetic or manipulated signals.

Existing speaker recognition and deepfake detection systems frequently struggle when applied across different languages, recording conditions, or synthesis techniques. This highlights a growing need to work on these areas.

The limited availability of standardized and forensically relevant speech corpora for Indian languages constructs a pathway for producing representative speech datasets sufficiently robust for academic and legal contexts.

References:

1. Agarwal, S., Farid, H., Gu, Y., He, M., Nagano, K., & Li, H. (2019). Protecting world leaders against deep fakes. *Proceedings of the IEEE Conference on Computer Vision and Pattern Recognition Workshops*, 38–45.
2. *Daubert v. Merrell Dow Pharmaceuticals, Inc.*, 509 U.S. 579 (1993).
3. French, P., & Harrison, P. (2007). *Investigative and forensic phonetics*. London, UK: Routledge.
4. Jessen, M. (2008). Forensic phonetics. *Language and Linguistics Compass*, 2(4), 671–711. <https://doi.org/10.1111/j.1749-818X.2008.00078>.
5. Künzel, H. J. (2001). Beware of the “telephone effect”: The influence of telephone transmission on speaker recognition. *International Journal of Speech, Language and the Law*, 8(1), 80–99.
6. Morrison, G. S. (2009). Likelihood-ratio statistics and forensic evidence interpretation. *Statistical Issues in Forensic Science*, 25–50.
7. Morrison, G. S. (2011). Measuring the validity and reliability of forensic voice comparison. *Science & Justice*, 51(2), 63–70. <https://doi.org/10.1016/j.scijus.2010.07.004>.
8. Nolan, F. (2003). Speaker identification evidence. In J. K. Chambers, P. Trudgill, & N. Schilling-Estes (Eds.), *The handbook of language variation and change* (pp. 757–774). Oxford, UK: Blackwell.
9. Rose, P. (2002). *Forensic speaker identification*. CRC Press London, UK: Taylor & Francis. <https://doi.org/10.1201/9780203166369>, eBook ISBN9780429229015
10. Saida H, Okamoto M, Imaizumi S, Hirose H. , A study of voice mutation and physical growth--a longitudinal observation, *Nihon Jibiinkoka Gakkai Kaiho*. 1990 Apr; 93(4):596-605. Japanese. doi: 10.3950/jibiinkoka.93.596. PMID: 2376796.
11. Britto AI, Doyle PC. A comparison of habitual and derived optimal voice fundamental frequency values in normal young adult speakers. *J Speech Hear Disord*. 1990 Aug; 55(3):476-84. doi: 10.1044/jshd.5503.476. PMID: 2381189.
12. Patil, H.A., Basu, T.K. Development of speech corpora for speaker recognition research and evaluation in Indian languages. *Int J Speech Technol* 11, 17–32 (2008). <https://doi.org/10.1007/s10772-009-9029-5>
13. Rao, K. & Koolagudi, Shashidhar. (2011). Identification of Hindi Dialects and Emotions using Spectral and Prosodic features of Speech. *Journal of Systemics, Cybernetics and Informatics*. 9.
14. Mehta, L., S.P. Mahajan, & Dabhade, A.S. (2013). COMPARATIVE STUDY OF MFCC AND LPC FOR MARATHI ISOLATED WORD RECOGNITION SYSTEM. *International Journal of Advanced Research in Electrical, Electronics and Instrumentation Energy*, 2, 2133-2139.
15. B G, Nagaraja & Jayanna, H.. (2013). Combination of Features for Multilingual Speaker Identification with the Constraint of Limited Data. *International Journal of Computer Applications*. 70. 1-6. 10.5120/11963-7823.
16. N. P. Jawarkar, R. S. Holambe and T. K. Basu, "Speaker Identification Using Whispered Speech," 2013 International Conference on Communication Systems and Network Technologies, Gwalior, India, 2013, pp. 778-781, doi: 10.1109/CSNT.2013.167.
17. Vempada, Ramu & Maity, Sudhamay & Rao, K.. (2013). Identification of Indian languages using multi-level spectral and prosodic features. *International Journal of Speech Technology*. 16. 10.1007/s10772-013-9198-0.
18. Bansod, Nagsen & Dabhade, Siddharth & Kawathekar, Seema & Kale, Karbhari. (2014). Speaker Recognition Using Marathi (Varhadi) Language. *Proceedings - 2014 International Conference on Intelligent Computing Applications, ICICA 2014*. 421-425. 10.1109/ICICA.2014.92.
19. Waghmare, Dr. Vishal & Deshmukh, Ratnadeep & Shrishrimal, Pukhraj & Janvale, Ganesh. (2014). Emotion Recognition System from Artificial Marathi Speech using MFCC and LDA Techniques.
20. Naresh P. Jawarkar, Raghunath S. Holambe, and Tapan Kumar Basu. 2015. Effect of Nonlinear Compression Function on the Performance of the Speaker Identification System under Noisy Conditions. In *Proceedings of the 2nd International Conference on Perception and Machine Intelligence (PerMI '15)*. Association for Computing Machinery, New York, NY, USA, 137–144. <https://doi.org/10.1145/2708463.2709049>.

21. Zhang, Z. (2016). Mechanics of human voice production and control. *The Journal of the Acoustical Society of America*, 140(4), 2614–2635. <https://doi.org/10.1121/1.4964509>
22. Fouquet M, Pisanski K, Mathevon N, Reby D. Seven and up: individual differences in male voice fundamental frequency emerge before puberty and remain stable throughout adulthood. *R Soc Open Sci*. 2016 Oct 5;3(10):160395. doi: 10.1098/rsos.160395. PMID: 27853555; PMCID: PMC5098980.
23. Rathna Kumar, S. B., Mohanty, P., Ujawane, P. A., & Huzurbazar, Y. R. (2016). Conventional speech identification test in Marathi for adults. *International Journal of Otorhinolaryngology and Head and Neck Surgery*, 2(4), 205–215. <https://doi.org/10.18203/issn.2454-5929.ijohns20163467>
24. B G, Nagaraja & Jayanna, H.S. (2016). Feature extraction and modelling techniques for multilingual speaker recognition: A review. *International Journal of Signal and Imaging Systems Engineering*. 9. 67. 10.1504/IJSISE.2016.075000.
25. Vinay Kumar Jain, Dr. (Mrs.) Neeta Tripathi. 2016. “Multilingual Speaker Identification Using Analysis of Pitch and Formant Frequencies”. *International Journal on Recent and Innovation Trends in Computing and Communication* 4 (2):296-98. <https://doi.org/10.17762/ijritcc.v4i2.1812>.
26. Shrikant Upadhyay, Sudhir Kumar Sharma, Pawan Kumar and Aditi Upadhyay; Performance Analysis of Hindi Voice for Speaker Recognition and Verification Using Different Feature Extraction, *International Journal of Electronics Engineering Research*. ISSN 0975-6450 Volume 9, Number 2 (2017) pp. 241-254.
27. Sreenivas Sremath Tirumala, Seyed Reza Shahamiri, Abhimanyu Singh Garhwal, Ruili Wang, Speaker identification features extraction methods: A systematic review, *Expert Systems with Applications*, Volume 90, 2017, Pages 250-271, ISSN 0957-4174, <https://doi.org/10.1016/j.eswa.2017.08.015>.
28. Dash, D., Kim, M., Teplansky, K., Wang, J. (2018) Automatic Speech Recognition with Articulatory Information and a Unified Dictionary for Hindi, Marathi, Bengali and Oriya. *Proc. Interspeech 2018*, 1046-1050, doi: 10.21437/Interspeech.2018-2122.
29. V. Jain and N. Tripathi, “Speech Features Analysis and Biometric Person Identification in Multilingual Environment”, *Int. J. Sci. Res. Net. Sec. Comm.*, vol. 6, no. 1, pp. 7–11, Feb. 2018.
30. Revathi, A., Jeyalakshmi, C., Muruganantham, T. (2018). Perceptual Features Based Rapid and Robust Language Identification System for Various Indian Classical Languages. In: Hemanth, D., Smys, S. (eds) *Computational Vision and Bio Inspired Computing. Lecture Notes in Computational Vision and Biomechanics*, vol 28. Springer, Cham. https://doi.org/10.1007/978-3-319-71767-8_25.
31. Athira N. P. and Poorna S.S., "Deep Learning Based Language Identification System From Speech," 2019 International Conference on Intelligent Computing and Control Systems (ICCS), Madurai, India, 2019, pp. 1094-1097, doi: 10.1109/ICCS45141.2019.9065370.
32. Dmitrieva O, Dutta I. Acoustic Correlates of the Four-Way Laryngeal Contrast in Marathi. *Phonetica*. 2020;77(3):209-237. doi: 10.1159/000501673. Epub 2019 Sep 18. PMID: 31533114.
33. Varun Tiwari, Mohammad Farukh Hashmi, Avinash Keskar, N.C. Shivaprakash, Speaker identification using multi-modal i-vector approach for varying length speech in voice interactive systems, *Cognitive Systems Research*, Volume 57, 2019, Pages 66-77, ISSN 1389-0417, <https://doi.org/10.1016/j.cogsys.2018.09.028>.
34. Shinde, Dattaji & More, Y & Sonawane, A & Pawade, V & Kulkarni, Krishna. (2020). COMPARISON OF MALE AND FEMALE VOWEL FORMANTS BY NATIVE SPEAKERS OF MARATHI LANGUAGE. *International Journal Of Engineering Research and General Science*. 5.
35. Fei He, Shan-Hui Cathy Chu, Oddur Kjartansson, Clara Rivera, Anna Katanova, Alexander Gutkin, Isin Demirsahin, Cibu Johnny, Martin Jansche, Supheakmungkol Sarin, and Knot Pipatsrisawat. 2020. Open-source Multi-speaker Speech Corpora for Building Gujarati, Kannada, Malayalam, Marathi, Tamil and Telugu Speech Synthesis Systems. In *Proceedings of the Twelfth Language Resources and Evaluation Conference*, pages 6494–6503, Marseille, France. European Language Resources Association.

36. Jain, V. K. (2020). Multilingual Speaker Identification System through Multiple Features Analysis of Speech Signal in Multilingual Environment. *i-manager's Journal on Digital Signal Processing*, 8(1), 27-33. <https://doi.org/10.26634/jdp.8.1.17838>.
37. Patil, Sunil & Patil, Nita & Patil, Ajay. (2020). Speaker Independent Isolated Word Recognition using HTK for Varhadi a Dialect of Marathi. *International Journal of Engineering and Advanced Technology*. 9. 748-751. 10.35940/ijeat.B3832.029320.
38. Peters, Micah D J et al. "Updated methodological guidance for the conduct of scoping reviews." *JBIE evidence synthesis* vol. 18,10 (2020): 2119-2126. doi:10.11124/JBIES-20-00167.
39. Kamiloğlu, R., & Sauter, D. (2021, September 29). Voice Production and Perception. *Oxford Research Encyclopedia of Psychology*. Retrieved 11 Jul. 2025, from <https://oxfordre.com/psychology/view/10.1093/acrefore/9780190236557.001.0001/acrefore-9780190236557-e-766>.
40. Pisanski Katarzyna, Groyecka-Bernard Agata and Sorokowski Piotr. 2021, Human voice pitch measures are robust across a variety of speech recordings: methodological and theoretical implications, *Biol. Lett.* 1720210356. <http://doi.org/10.1098/rsbl.2021.0356>
41. Rocha, R.B., José Lira de Queiroz, W. & Sampaio de Alencar, M. Voice production model based on phonation biophysics. *EURASIP J. Adv. Signal Process.* 2021, 78 (2021). <https://doi.org/10.1186/s13634-021-00746-2>
42. Basu, J., Khan, S., Roy, R. et al. Multilingual Speech Corpus in Low-Resource Eastern and Northeastern Indian Languages for Speaker and Language Identification. *Circuits Syst Signal Process* 40, 4986–5013 (2021). <https://doi.org/10.1007/s00034-021-01704-x>.
43. Tiwari, Sonal & Kanke, Rajashri & Ambewadikar, Maheshwari & Baheti, Manasi. (2021). Marathi Speech Database Standardization: A Review and Work. *International Journal of Computer Science and Information Security*, 19. 92-99. 10.5281/zenodo.5501909.
44. Abdulrahman, A., & Richards, D. (2022). Is natural necessary? Human voice versus synthetic voice for intelligent virtual agents. *Multimodal Technologies and Interaction*, 6(7), 51. <https://doi.org/10.3390/mti6070051>
45. Pawar, Ashwini & Patil, Nita. (2024). Comparative Study of Techniques for Spoken Language Dialect Identification. 10.13140/RG.2.2.21521.24162.
46. Ternström, S., Henrich Bernardoni, N., Birkholz, P., Guasch, O., & Gully, A. (Eds.). (2024). Computational analysis and simulation of the human voice (Dagstuhl Seminar 24242). In *Dagstuhl Reports* (Vol. 14, Issue 6, pp. 84–107). Schloss Dagstuhl – Leibniz-Zentrum für Informatik. <https://doi.org/10.4230/DagRep.14.6.84>
47. Alali, A., & Theodorakopoulos, G. (2025). Partial Fake Speech Attacks in the Real World Using Deepfake Audio. *Journal of Cybersecurity and Privacy*, 5(1), 6. <https://doi.org/10.3390/jcp5010006>
48. Li, X., Chen, P.-Y., & Wei, W. (2025). Where are We in Audio Deepfake Detection? A Systematic Analysis over Generative and Detection Models. *ACM Transactions on Internet Technology*. <https://doi.org/10.1145/3736765>
49. Mawalim, C. O., Wang, Y., Adila, A., Okada, S., & Unoki, M. (2025). Robust Multilingual Audio Deepfake Detection through Hybrid Modeling. In *ACM Workshop on Information Hiding and Multimedia Security (IH&MMSEC '25)* (pp. 181-192). Association for Computing Machinery. <https://doi.org/10.1145/3733102.3736706>
50. Nussbaum, C., Frühholz, S., & Schweinberger, S. R. (2025). Understanding voice naturalness. *Trends in Cognitive Sciences*, 29(5), 467–480. <https://doi.org/10.1016/j.tics.2025.01.010>
51. Pouw Wim, Werner Raphael, Burchardt Lara S. and Selen Luc P. J.(2025). The human voice aligns with whole-body kinetics *Proc. R. Soc. B.* 29220250160 <http://doi.org/10.1098/rspb.2025.0160>
52. San Segundo, E., & Delgado, J. (2025, February 12). A qualitative systematic review of intra-speaker variation in the human voice. https://doi.org/10.31219/osf.io/me6ap_v1
53. Singh, Rita & Raj, Bhiksha. (2025). Human Voice is Unique. 10.48550/arXiv.2506.18182.

22. Phytochemical Evaluation and Adulteration Analysis of Marketed Chaturbeeja Churna

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Abstract: Chaturbeeja is a widely consumed traditional polyherbal formulation comprising Ajwain (*Trachyspermum ammi*), Chandrashoor (*Lepidium sativum*), Kalonji (*Nigella sativa*), and Methi (*Trigonella foenum-graecum*), valued for its digestive, metabolic, and anti-inflammatory properties. Despite its extensive household and therapeutic use, scientific data on adulteration and quality assurance of Chaturbeeja remain limited. The present study aimed to evaluate the phytochemical composition and detect possible adulteration in commercially available Chaturbeeja powders. Two marketed samples were analyzed using standard phytochemical screening and simple adulteration detection tests. Results confirmed the presence of characteristic bioactive constituents of all four seeds in both samples. However, cumin adulteration in Ajwain was detected in one marketed sample, while the other sample was found to be free from common adulterants. The findings highlight variability in market quality and emphasize the need for routine authentication and standardization to ensure the safety, purity, and therapeutic efficacy of polyherbal formulations like Chaturbeeja.

Keywords: Chaturbeeja, polyherbal formulation, phytochemical screening, adulteration, quality control, Ajwain, Kalonji

Introduction

Chaturbeeja is a classical polyherbal formulation extensively used in Ayurveda and traditional medicine for digestive, metabolic, and anti-inflammatory benefits (Panchaware & Shekokar, 2024). The formulation comprises Ajwain, Chandrashoor, Kalonji, and Methi, each known for distinct phytochemicals contributing synergistically to therapeutic efficacy (Johri, 2011; Ahmad et al., 2013).

Due to its increasing popularity as a household remedy and nutraceutical preparation, Chaturbeeja has attracted scientific interest in validation of its pharmacological and functional properties. However, the widespread use of powdered herbal formulations also raises concerns regarding adulteration, substitution, and inconsistency in raw material quality, which may compromise therapeutic efficacy and safety.

1 Components of Chaturbeeja

Ajwain (*Trachyspermum ammi*) contains thymol, carvacrol, and volatile oils responsible for antimicrobial, carminative, and antioxidant activities. Chandrashoor (*Lepidium sativum*) is rich in flavonoids, minerals, and omega-3 fatty acids and is traditionally used for lactation

support and anti-inflammatory effects. Kalonji (*Nigella sativa*) contains thymoquinone and nigellone and exhibits immunomodulatory, antioxidant, antidiabetic, and hepatoprotective properties. Methi (*Trigonella foenum-graecum*) is known for its saponins, diosgenin, and galactomannan fiber, contributing to glucose regulation and lipid-lowering effects.

2 Therapeutic Importance

Traditionally, Chaturbeeja is used to improve digestion, manage metabolic disorders, enhance immunity, reduce inflammation, promote women's health, and act as a general health tonic when consumed with warm water or honey. Scientific studies on individual components support these applications, although standardized validation of the combined formulation remains limited.

3 Adulteration Concerns

Adulteration of herbal formulations is a growing concern due to visual similarity among seeds and economic motivations. Substitution of Ajwain with cumin, Kalonji with black sesame, or dilution with inferior or filler materials can significantly affect potency and safety. Despite extensive traditional use, systematic scientific documentation on adulteration patterns in Chaturbeeja remains scarce, creating a critical research gap.

Materials and Methods

1 Sample Collection

Two commercially available Chaturbeeja powders were procured from the local herbal market and coded as Sample A (SAAR) and Sample B (Navjivan). Individual seeds of Ajwain, Chandrashoor, Kalonji, and Methi were purchased separately from authenticated vendors and used as reference standards (Inhouse).



Commercial and Standard (Inhouse) Chaturbeeja Churna

2 Extraction Procedure

Approximately 5 g of each powdered sample and individual seed component was subjected to solvent extraction. Ajwain and Kalonji were extracted using ethanol, while Methi and Chandrashoor were extracted using methanol. Soxhlet extraction and cold maceration techniques were employed for 6–8 hours until complete extraction. The extracts were filtered and stored in airtight containers for analysis.

3 Phytochemical Screening

Phytochemical screening was conducted using standard qualitative tests as described by Harborne (1998) and Trease and Evans (2002) to detect major bioactive constituents. Borntrager's test was used for Ajwain, Dragendorff's test for Chandrashoor, Salkowski test for Kalonji, and glacial acetic acid–sulphuric acid test for Methi. Confirmatory tests such as iodine test for starch, ferric chloride test for phenolics, NaOH warming test for volatile oils, and ninhydrin test for amino acids were also conducted.

4 Adulteration Detection Tests

Adulteration detection tests were performed to identify common substitutes such as cumin, linseed, and black sesame. Schiff's reagent test was used to detect cumin adulteration in Ajwain. Potassium dichromate–dilute acid test was employed to identify linseed or mustard seed adulteration, and hydrogen peroxide bleaching test was used to detect black sesame adulteration in Kalonji.

Results and Discussion

Phytochemical screening of both marketed samples showed positive results for characteristic constituents of all four seeds, confirming the presence of Ajwain, Chandrashoor, Kalonji, and Methi. Confirmatory tests further validated the presence of phenolics, volatile oils, starch, and amino acids consistent with established phytochemical profiles.

The qualitative phytochemical screening results of the individual components of Chaturbeeja are summarized in **Table 1**. The results confirmed the presence of alkaloids, phenolics, sterols, saponins, volatile oils, and amino acids, which are consistent with previously reported phytochemical profiles of Ajwain, Chandrashoor, Kalonji, and Methi (Basch et al., 2003; Ahmad et al., 2013).

Table 1. Phytochemical Screening Results of Chaturbeeja Components

Test	Ajwain	Chandrashoor	Kalonji	Methi
Borntrager's Test	Positive	—	—	—
Dragendorff's Test	—	Positive	—	—
Salkowski Test	—	—	Positive	—
Glacial Acetic Acid–H ₂ SO ₄ Test	—	—	—	Positive
Ferric Chloride Test	Positive	Positive	Positive	Positive
Ninhydrin Test	—	Positive	—	Positive

The results of adulteration analysis of the marketed Chaturbeeja samples are presented in **Table 2**. Schiff's reagent test indicated cumin adulteration in the Navjivan sample, whereas the SAAR sample showed no evidence of adulteration. No linseed, mustard seed, or black sesame adulteration was detected in either sample. Tests for linseed, mustard seed, and black sesame adulteration were negative in both samples. These findings indicate variability in market quality, as also reported in comparative studies on marketed Chaturbeeja formulations (Gayaki et al., 2025). Since Ajwain contributes significantly to the therapeutic efficacy of Chaturbeeja, its adulteration may directly impact clinical effectiveness.

Table 2. Adulteration Analysis of Marketed Chaturbeeja Samples

Test	Target Adulterant	SAAR Sample	Navjivan Sample
Schiff's Reagent Test	Cumin (Ajwain)	Negative	Positive
Potassium Dichromate Test	Linseed / Mustard	Negative	Negative
Hydrogen Peroxide Test	Black Sesame	Negative	Negative

Conclusion

The present study systematically evaluated the phytochemical integrity and adulteration status of marketed Chaturbeeja powders. While both samples contained the essential bioactive constituents of the formulation, cumin adulteration was detected in one commercial sample. The results emphasize the need for routine quality control, authentication, and standardization of polyherbal formulations. Adoption of forensic-style validation protocols can ensure consistency, safety, and therapeutic reliability of traditional herbal products such as Chaturbeeja.

References

1. Aggarwal, B. B., Sundaram, C., Malani, N., & Ichikawa, H. (2007). Curcumin: The Indian solid gold. *Advances in Experimental Medicine and Biology*, 595, 1–75.
2. Ahmad, A., Husain, A., Mujeeb, M., Khan, S. A., Najmi, A. K., Siddique, N. A., & Anwar, F. (2013). A review on therapeutic potential of *Nigella sativa*. *Asian Pacific Journal of Tropical Biomedicine*, 3(5), 337–352.
3. Basch, E., Ulbricht, C., Kuo, G., Szapary, P., & Smith, M. (2003). Fenugreek monograph. *Journal of Herbal Pharmacotherapy*, 3(2), 63–83.
4. Gayaki, P., Gurunani, S., Popali, K., Gharpure, B., Ahmed, I., & Chaple, D. (2025). Comparative evaluation of in-house prepared and marketed Chaturbeeja Churna. *ResearchGate Preprint*.
5. Harborne, J. B. (1998). *Phytochemical methods: A guide to modern techniques of plant analysis* (3rd ed.). Springer.
6. Johri, R. K. (2011). Cumin, caraway and ajwain. *Indian Journal of Natural Products and Resources*, 2(2), 231–238.
7. Panchaware, P. S., & Shekokar, S. S. (2024). Therapeutic potential of Chaturbeej Churna. *Journal of Emerging Technologies and Innovative Research*, 11(11), 1–6.
8. Trease, G. E., & Evans, W. C. (2002). *Pharmacognosy* (15th ed.). Saunders.

23. Forensic Assessment of WPA2/WPA3 Security Using Personal Area Network

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Abstract: This research uses a Personal Area Network built with cellphones and a Kali Linux system to conduct controlled wireless experiments to assess the security of the WPA2-PSK and WPA3-SAE protocols. To determine whether passwords could be cracked offline, authentication handshakes were recorded using Aircrack-ng and examined using Wireshark. The findings demonstrate WPA2 networks' vulnerability to dictionary attacks by demonstrating how weak or medium-strength passphrases could be effectively broken in a matter of seconds. WPA3, on the other hand, completely eliminated offline cracking since its SAE handshake does not provide reusable cryptographic information. The study emphasizes the significance of strong passphrases, the benefits of WPA3, and the usefulness of recorded Wi-Fi metadata for forensic purposes.

Keywords: WPA2, WPA3, Wireless Security, Forensic Analysis, Aircrack-ng, Dictionary Attack, SAE Protocol, Personal Area Network, Penetration Testing, Kali Linux.

Introduction

Modern digital communication relies heavily on wireless networks to support commercial, educational, and personal applications. To secure wireless communications, the Wi-Fi Alliance introduced the Wi-Fi Protected Access (WPA) family of protocols, among which WPA2 and WPA3 are the most widely deployed (Abdul Rahman & Sikdar, 2025). Although WPA2 employs strong AES-CCMP encryption, its reliance on a four-way handshake and user-defined pre-shared keys makes it vulnerable to offline dictionary and brute-force attacks when weak passphrases are used. Consequently, password strength remains a critical factor in determining the practical security of WPA2-protected networks.

WPA3, introduced in 2018, addresses the limitations of WPA2 by incorporating enhanced authentication and encryption mechanisms. It replaces the traditional pre-shared key handshake with Simultaneous Authentication of Equals (SAE), also known as the Dragonfly handshake, which effectively prevents offline password-cracking attempts even if authentication messages are intercepted (Vanhoef & Piessens, 2018). In addition, WPA3 provides forward secrecy, ensuring that previously captured network traffic remains protected even if the network password is later compromised. These architectural improvements significantly enhance WPA3's resilience against

modern wireless attacks and make it more suitable for deployment in public or hostile environments.

This study presents a controlled, laboratory-based comparison of the security mechanisms of WPA2 and WPA3 using industry-standard tools such as Aircrack-ng and Wireshark within a Kali Linux environment. Aircrack-ng is utilized to capture and analyze WPA authentication exchanges for evaluating password strength, while Wireshark enables detailed forensic examination of EAPOL frames (Togashi et al., 2018). The objective of this work is not to compromise real-world networks but to raise awareness of the risks associated with weak Wi-Fi passphrases and to demonstrate the practical security advantages of WPA3 for cybersecurity education and research

Methodology

The experimental approach was created to methodically examine and contrast the real-world security behavior of WPA2 and WPA3 wireless protocols in controlled settings. To guarantee ethical compliance, avoid unauthorized interference, and enable consistent replication of results, all tests were carried out on a separate Personal Area Network (PAN). A smartphone set up as a Wi-Fi access point, another smartphone acting as a valid client device, and a research laptop running Kali Linux with a USB Wi-Fi adapter that supported monitor mode and packet injection made comprised the testbed (Lu & Yu, 2021). To guarantee reliable monitoring and capture capabilities, the wireless interface was confirmed, and the operating system, drivers and wireless security tools were upgraded before experimenting (Asante & Akomea Agyin, 2017).

To determine crucial network data, such as SSID, BSSID, operating channel, and linked client MAC addresses, the first observations were passively monitoring the wireless environment. These parameters were used to set up concentrated packet capture sessions and were recorded as baseline forensic artifacts. Then, to gather authentication-related traffic, namely EAPOL frames connected to WPA-based authentication, targeted packet captures were carried out. Controlled client-side reconnections were used as needed to purposefully start authentication handshakes and guarantee the availability of legitimate capture data. To maintain data integrity and facilitate offline analysis, all collected frames were saved in standardized packet capture files.

To assess the network's vulnerability to dictionary-based attacks, the collected four-way handshake for WPA2 trials was examined offline using the Aircrack-ng suite (Asaad, 2021). Using the PBKDF2 key derivation function, cryptographic keys were derived from candidate passphrases as part of the study, and message authentication codes were checked against the observed handshake values. Passphrase recovery was the outcome of successful key validation, illustrating the usefulness of weak or low-entropy passwords. The quality of the word list and the intricacy of the password used affected the attack's efficacy, underscoring the importance of user behavior in wireless security (Carballal et al., 2022).

WPA3 testing, on the other hand, was more concerned with forensic observability and protocol behavior than with password recovery. To assess resistance to offline attacks, the Simultaneous Authentication of Equals (SAE) mechanism was analyzed using recorded authentication exchanges (Braga et al., 2023). Forensic analysis was restricted to protocol flow, timing features, and live interaction behavior because, in contrast to WPA2, the collected WPA3 traffic did not allow offline verification of guessed passphrases. All capture files, logs, and observations were kept as forensic evidence, and Wireshark was used to verify the existence of authentication frames and examine packet-level information. This approach

makes it possible to compare WPA2 with WPA3 clearly from a security and forensic standpoint, highlighting the direct impact that protocol architecture has on attack viability and evidence accessibility.

```
(kali@kali)-[~]
$ sudo airodump-ng wlan1

CH 9 ][ Elapsed: 30 s ][ 2025-09-09 09:04

BSSID            PWR Beacons  #Data, #/s  CH  MB  ENC CIPHER AUTH ESSID
F8:C4:F3:C8:B2:55 -84      5           0  0   6  130  WPA2 CCMP  PSK  MADHAV _4G
6C:4F:89:B8:20:BF -89     13           0  0   1  130  WPA2 CCMP  PSK  Airtel_TEJAS BIND
50:2B:73:77:60:48 -77     20           0  0  11  270  WPA2 CCMP  PSK  Manish123
E8:65:D4:E1:F7:70 -93      2           0  0   3  270  WPA2 CCMP  PSK  Devshree pg 3rd floor
E2:5B:B1:CC:31:C2 -51     72           0  0   6  180  WPA3 CCMP  SAE  SVVV-chetan
F8:C4:F3:7E:D6:42 -76     44           0  0   3  270  WPA2 CCMP  PSK  Sitibroadband_4G
04:95:E6:7E:7F:80 -37     65        2381 138   1  54e  WPA2 CCMP  PSK  Manish 3floor
```

Figure1: Passive scan to find your hotspot

```
(kali@kali)-[/usr/share/wordlists]
$ grep 123456789 rockyou.txt
123456789
```

Figure2: Display of the cracked password from the captured file RESULTS

Using two cellphones and a Kali Linux laptop with a USB Wi-Fi adapter that could be used in monitor mode, a controlled Personal Area Network (PAN) was established. We established a test passphrase, recorded the authentication exchange (PCAP), set up the hotspot on Phone A (AP) using either WPA2-PSK or WPA3-SAE, and performed offline auditing with Air crack-ng using the specified wordlists for each trial. Every experiment was carried out using equipment that belonged to the researcher.

Exp Id	Protocol	Passphrase	Wordlist used	Handshake captured.	Cracked?	Time to crack(s)
1.	WPA2	12345678(weak)	Weak .txt	Yes	Yes	8.8
2.	WPA2	Chetan@123	Medium.txt	Yes	Yes	14.5
3.	WPA3	12345678(weak)	Weak.txt	No(SAE Handshake)	No	N/A

This parameter indicates whether the authentication exchange, including EAPOL frames required for offline cracking, was successfully captured and usable by the Aircrack-ng framework. The results show that while WPA2 handshakes can be captured and analyzed for offline password recovery, the SAE-based authentication used in WPA3 does not produce decryptable handshake data suitable for offline cracking. Consequently, WPA3 is marked as “No” for handshake capture in the context of Aircrack-ng analysis.

Discussion:

The experiment proves that it is easy to crack WPA2 networks using weak pass phrases when carrying out offline attacks, as it is possible to retrieve the pass phrases from a four-way handshake. Additionally, with SAE authentication, WPA3 makes it impossible to perform offline attacks. The forensic analysis indicates that it is possible to make use of the metadata from encrypted connections. The experiment proves that using pass phrases and WPA3 makes it difficult to intercept networks

Conclusion

Using only easily accessible devices—a laptop running Kali Linux and smartphones, this study aimed to evaluate and contrast the security of the WPA2-PSK and WPA3-SAE protocols in a supervised Personal Area Network (PAN) setting.

The study methodically showed how WPA2 networks are still susceptible to offline password-cracking assaults when weak or dictionary-based passphrases are employed, but WPA3 networks show robust resistance to such attempts, using open-source tools like Air crack-ng and Wireshark, according to the experimental findings:

If the passphrase is weak or contains words from a dictionary, WPA2 handshakes can be recorded and decrypted offline. Short cracking times (measured in seconds for weak cases) underscored the dangers users encounter when using predictable passphrases. Although WPA2 networks with intricate, high-entropy passphrases were difficult to break, the protocol's fundamental flaw still made it reliant on user behavior.

Simultaneous Authentication of Equals (SAE) in WPA3 completely stopped the offline cracking process. Stronger security was provided because authentication exchanges could not be utilized for dictionary attacks, even if they were intercepted.

The initiative also emphasized the significance of wireless packet captures as artefacts from a forensic standpoint. To sum up, the project was effective in testing the security of WPA2 and WPA3, proving that WPA2 is vulnerable to weak password circumstances, and verifying that WPA3 is resilient to offline dictionary assaults.

By connecting theoretical protocol design with real-world, repeatable data, the study advances cybersecurity education by raising awareness of wireless network vulnerabilities and the vital role that robust authentication systems play in contemporary digital communication.

References

1. Abdul Rahman, H. B., & Sikdar, B. (2025). False sense of security on protected Wi-Fi networks. *arXiv*. <https://arxiv.org/abs/2501.13363>
2. Asaad, R. R. (2021, February). *Penetration testing: Wireless network attack methods on Kali Linux OS*. Kali Research. <https://kali-research.org/papers/2021-wifi-attacks>
3. Asante, M., & Akomea-Agyin, K. (2017). Analysis of security vulnerabilities in Wi-Fi Protected Access pre-shared key (WPA-PSK/WPA2-PSK). *Journal of Information Security and Applications*, 36, 1–10.
4. Braga, D. D. A., Kulatova, N., Sabt, M., Fouque, P.-A., & Bhargavan, K. (2023). *From Dragondoom to Dragonstar: Side-channel attacks and formally verified implementation of WPA3 Dragonfly handshake*. arXiv. <https://arxiv.org/abs/2307.09243>

5. Carballal, A., Galego-Carro, J. P., Rodríguez-Fernández, N., & Fernández-Lozano, C. (2022). Wi-Fi handshake: Analysis of password patterns in Wi-Fi networks. *PeerJ Computer Science*, 8, e1185. <https://doi.org/10.7717/peerj-cs.1185>
6. Lu, H.-J., & Yu, Y. (2021). *Research on WiFi penetration testing with Kali Linux*. Complexity, 2021, Article 5570001. <https://doi.org/10.1155/2021/5570001>
7. Togashi, H., Koga, Y., & Furukawa, H. (2018). *Criminal fishing system based on wireless local area network access points: Can media access control address assist criminal investigation?* arXiv. <https://arxiv.org/abs/1808.05346>
8. Vanhoef, M., & Piessens, F. (2018). Key reinstallation attacks: Forcing nonce reuse in WPA2. In *Proceedings of the ACM Conference on Computer and Communications Security (CCS)* (pp. 1313–1328). ACM. <https://doi.org/10.1145/3243734.3243766>

24. Modernizing Eco- Justice: A Review of Digital and Forensic Technologies in Environmental Law

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Abstract: Crimes against nature such as elephant poaching for ivory, illegal logging of protected forests, and toxic waste dumping are happening on a massive scale, yet they are extremely difficult to investigate because they occur in remote forests or oceans with no witnesses, cameras, or immediate law-enforcement presence, allowing organized criminal gangs to escape punishment; this paper argues that the only effective way to stop them is to treat the environment exactly like a human crime scene by combining Green Criminology with forensic science, using tools such as animal DNA analysis to trace ivory or pangolin scales back to specific regions and poachers, ballistics and tool-mark examination to match bullets or chainsaw marks to particular weapons and equipment, and digital forensics like satellite imagery and mobile phone data to track poacher movements and predict future attacks; the core message is simple but uncomfortable -laws alone do nothing unless they are enforced, and by applying high-tech forensic methods we can turn silent forests into evidence-rich crime scenes, build strong cases in court, dismantle organized environmental crime networks, and actually protect wildlife instead of just talking about it.

Key words : Environmental Crime, Wildlife Forensics, Green Criminology, Green Criminology.

Introduction:

Environmental crimes-anything from massive elephant poaching in Africa to the illicit disposal of hazardous wastes in the oceans-are among the most profitable illicit businesses in the world. Despite their significance, such crimes remain relegated to the margins of the criminal justice system. The main challenge is that they take place in what are termed "dark zones" that lack conventional surveillance systems. The point that I would like to make in this paper is that in order for biodiversity on a global scale to survive, what we need is a complete paradigm shift in that we can no longer view environmental damage as a control problem but rather as a crime site that demands serious forensic analysis so that we can turn quiet habitats into forensic-rich habitats that can break up organized crime groups (Wasser, S. K., et al. (2015), (Brack, D. 2002)

Eco-Justice and Green Criminology: Theoretical Framework:

Green Criminology overhauls the human centered bias tithing traditional legal frameworks, including the issue of Eco Justice within Green Criminology. The issue of Eco Justice suggests nature and species, apart from human beings, have a right to be protected, divided into three aspects:

Species Justice: Refers to those arguing in support of animal rights regarding their freedom from exploitation and harm

Environment justice: it focused on how the degradation of the environment is often experienced by marginalized sectors of society.

Ecological Justice: The Earth's ecosystems should be granted legal recognition and safeguarding them becomes a matter of legal justice.

Such theoretical insight perceives "green crimes" as non-random outcomes, but rather planned consequences of major crimes occasioned by the remoteness of the area away from the presence of law enforcement (Brisman, A., & South, N. 2015).

Environmental Crimes As Forensic Problems:

It is very hard to successfully bring charges against environmental crimes due to the existence of an evidence gaps that differ from urban and rural areas because, in urban areas, crimes such as murder can be solved within a short period due to the use of witnesses and experts, whereas environmental crimes such as the killing of an elephant or pollution of a river tend to take weeks before they are observed and result in the loss of all evidence, but according to Locard's Principle "Every contact leaves a trace", such as dirt on a poacher's boot or contaminants within water, proving that evidence always exists. The other aspect that is affected is humanity because pollution of rivers makes people ill, indigenous cultures lose heritage, and children inherit poor environments (Armitage, R. 2011)

Forensic Technologies In Environmental Crime Investigation:

To overcome the evidentiary divide, we must use specialized scientific instruments:

Wildlife DNA Forensics: Genetic sequencing can link a product to a specific population or, better still, a specific individual, tracing a confiscated Asian shipment to a poaching ground in Africa.

Ballistics and Tool Mark Analysis: The telltale marks left by a chain saw on illegally harvested timber are distinguishable just like fingerprints. The marks can be traced to a particular saw and thereby connect evidence to a criminal's possessions.

Chemical Profiling: Isotope testing can establish the origin of lumber or minerals, ensuring that "laundry" facilities could not possibly obtain illegal materials through legitimate channels (Wasser, S. K., et al. 2015), (Yadav, A., et al. 2024)

Digital Technologies Supporting Eco-Justice:

Forensics now expand beyond the physical realm. Digital evidence enables the "connective tissue" that links the crime scene with the crime network:

Satellite Imagery & Remote Sensing: SAR (Synthetic Aperture Radar) can monitor illegal logging activities under heavy cloud cover and during nighttime and send real-time alerts.

Sound Sensors: "Smmart" forests with microphones using AI can differentiate between the call of a tropical bird and the engine whine of a chainsaw.

Ship Tracking: AIS information enables tracking of "dark fleets" involved in illegal fishing and toxic dumping activities (Aich, S., et al. 2024)

Integration Of Forensic Science And Digital Evidence;

The best and strongest cases become even stronger when the physical and online proof merges. Mobile forensics, (triage of GPS coordinates) for instance, allows the guilty party to have been physically present at the same spot that the satellite imagery revealed the start of the forest fire.

In such a manner, it becomes very hard for organized gangs to maintain the cover of anonymity that they had before due to the "digital leash" that has been created (Al-Dhaqm, A., et al. 2020).

Practical Applications:

This transition from theory to practice is already yielding results in high-stakes environmental litigation:

Genetic Mapping of Poaching Hotspots: By building a DNA database of elephant dung and tissue across Africa, law enforcement can now identify exactly which "hotspots" are feeding the global ivory trade. When a container is seized in a port, DNA analysis reveals the origin, allowing authorities to target the specific criminal cells operating in that region rather than chasing the entire continent's trade.

Isotopic Timber Fingerprinting: Stable isotope analysis forms the basis of "Timber Fingerprinting" applied in the Amazon, which authenticates the geographic origin of wood. This application allows "legal" permits to be disproved by showing that the chemical signature of the wood does not match that of the soil of the permitted logging area.

Acoustic detection and immediate response: In Marine Protected Areas, hydrophone arrays are being fitted to detect "blast fishing." The digital signature of the explosion is triangulated, and coordinates in real time are dispatched to patrol boats, taking the process from "discovery of dead fish" to "interception of the vessel. (Yadav, A., et al. 2024), (Wasser, S. K., et al. 2008)

Legal Admissibility and Challenges:

For this "High tech Green Criminology" to be effectively used, the evidence must be present in such a way that is admissible in court:

Chain of custody: It is indeed a big logistical challenge to maintain a sound chain of custody in distant, difficult environments.

Standardization: Most countries do not have standard practices for "green forensic", which lets evidence fall through due to technical reason.

Scientific capacity: Local forensic laboratories for countries with high biodiversity are urgently needed to avoid delays and expenses in sending the sample abroad (D'Anna, T., et al. 2023).

Research Gaps and Future Directions:

Currently, there is a "tech gap" between rich nations and the areas where these crimes are occurring. Possible areas for future research include:

Portable Forensic Kits: Develop "lab on a chip" technology so field rangers can run a beginning DNA or chemical test in the field.

AI Predictive Modeling: Using historical data to predict poaching "strike zones" according to lunar cycles, weather, and shifting market prices (Kadage, A. 2024)

Conclusion:

Crime against nature (elephant poaching ,illegal logging, toxic dumping) are not come in small sizes. Crimes that wildlife, people , and the planet are serious criminal activities. The biggest challenge is when they often take place in distant areas with zero witnesses or

cameras , thus enabling the delinquents to get away with it. The review paper has shown that environment crimes need to be treated as crime scenes rather than just a regulatory issue. With forensic science- DNA testing to trace ivory, chemical analysis to track timber , ballistics to match weapons, and digital tools like satellites and mobile data—strong evidence could be collected to thwart organized crime networks. Real examples, such DNA mapping of poaching hotspots , prove that such method work. Still, problems persist regarding security safeguards for evidence, lack of standard rules, and facilities for laboratories being weak. What is missing are portable forensic tool kits, AI tools , and stronger institutions. Ultimately, laws are just that; only with science backed enforcement does any hope remain for the conservation of biodiversity and the assurance of eco-justice for future generations.

Reference:

1. Aich,S., Plathothahil R. R., Raj C.A., and Katare ,N.(2024).Integration Remote sensing and AI : A review of technological innovations in wildlife crime detection <https://doi.org/10.56557/upjoz/2024/v45i234683>
2. Al-Dhaqm, A., Razak, S. A., Ikuesan, R. A., Kebande, V.R., & Siddique, K. (2020). A review of mobile forensic investigation process models. <https://doi.org/10.1109/ACCESS.2020.3014615>
3. Armitage, R. Introduction: A New Look at Environmental Crime. Eur J Crim policy Res 17, 83–85 (2011). <https://doi.org/10.1007/s10610-011-9144-0>
4. Brack ,D.(2002) combating international environment crime. Global environment change,12(2),143-147 [https://doi.org/10.1016/S0959-3780\(01\)00019-X](https://doi.org/10.1016/S0959-3780(01)00019-X)
5. Bris,am ,A.,& South ,N.(2015). Green criminology and environmental crime and harm. Critical criminology, 23(4), 437-454. <https://doi.org/10.1007/s10612-015-9301-8>
6. D’Anna, T., Cannavo, M., Gualniera,P.Maglietta, F., and Ventura Spagnolo ,E.(2023) the chain of custody in the era of modern forensic: From the classic procedures for gathering evidence to the new challenges related to digital data. Healthcare, 11(5), 634 <https://doi.org/10.3390/healthcare11050634>
7. Kadage, Anushka. (2024). AI-Enhanced Digital Forensics: Automated Techniques for Efficient Investigation and Evidence Collection. Journal of Electrical Systems. 20. 211-229. 10.52783/jes.766. https://www.researchgate.net/publication/379867130_AI-Enhanced_Digital_Forensics_Automated_Techniques_for_Efficient_Investigation_and_Evidence_Collection
8. Wasser , S. K., Mailand, C., Booth, R., Mutayoba, B., Kisamo, E., Clark, B., & Stephens, M. (2008). <https://doi.org/10.1111/j.1523-1739.2008.01012.x>
9. Wasser, S. K., et al. (2015) Genetic assignment of large seizures of elephant ivory reveals Africa’s major poaching hotspots. *Science*, 349(6243), 84–87 <https://doi.org/10.1126/science.aaa2457>
10. Wasser, S. K., et al. (2015). Genetic assignment of large seizures of elephant ivory reveals Africa's major poaching hotspots. *Science*, 349(6243), 84-87. [Genetic assignment of large seizures of elephant ivory reveals Africa’s major poaching hotspots | Science](https://doi.org/10.1126/science.aaa2457)
11. Yadav, A., Sharma, S., Gupta, L., Singh, V., & Singh, R. (2024). Current Perspectives in the Forensic Analysis of Timbers using Vibrational Spectroscopy: A Review. <https://doi.org/10.63033/JWLS.KRNG5178>

25. A Narrative Review on Virtual Reality (VR) and Artificial Intelligence (AI) Implementation in Forensic Science Education in India.

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Abstract: Forensic education in India faces significant challenges, like limited access to real crime scenes, interactive experiences, resource constraints, and the need for scalable assessment tools. The integration of Virtual Reality and Artificial Intelligence is an ideal implementation for forensic education, as it helps students, researchers, and academicians learn and visualize crime scenes, practice evidence searching strategies and handling, reconstruct crime scenes, analyze various activities at a crime scene, and much more, all based on factual data and animations. and helps in transforming the analysis and investigative methods by saving time, Enhanced Efficiency, Precision and Accuracy. this Narrative study reviews findings from 15+ studies from (2010-2025), across Databases like (Scopus, PubMed, and Google scholar) in Areas where Applications of Virtual Reality and artificial intelligence could be used to enhance Phases of forensic Education & Assessments in India. The VR/AI gives us various opportunities in Forensic education and assessments The implementation includes barriers like admissibility and validation issues, technical & Ethical constraints, the need for specialized training, costs, and risks of misinterpretation that compromise the reliability and acceptance of VR/AI tools.

Keywords: Forensic Education, Virtual Reality, Artificial Intelligence, Forensic Visualization, Immersive technology, 3D reconstruction, pedagogy, Narrative review.

Abbreviations:

VR- virtual Reality, **AI-** Artificial Intelligence, **2D-** Two-Dimensional, **3D-** Three Dimensional

Introduction

Forensic science education in India has Shown Significant Growth in the last 15–20 years, with many colleges and universities now offering certificate, B.Sc., M.Sc., and Ph.D. programs in the field. But Forensic education in India faces significant challenges, like limited access to real crime scenes, interactive experiences, resource constraints, and the need for scalable assessment tools. In several regions of India, students depend on occasional field visits and conventional labs, which may not fully reflect the complexity and technology used in modern investigations, time Consuming process, to address this, some institutions are slowly introducing more modern teaching methods, such as simulation-based crime scene exercises, digital forensics laboratories, and online modules. Yet the structured use of immersive tools like virtual reality and data-driven systems such as artificial intelligence for teaching and assessment is still very new, making this a timely area for a narrative review focused on the Indian context.

Forensic science is not just theoretical Education; it is a multidisciplinary field. it requires the applying of various scientific Disciplines, Scientific Principles and applications to maintain Justice Hence it is Essentially Required to the students, professionals and Investigators of the Forensic field to integrate concepts during crime scene management and evidence analysis, documentation. the Current Scenario in Crime scene analysis is students are being trained by a relatively simple Staged crime scenario which allows the students to be capable of document, identify, handle the Evidence in a given time frame. However, the Reality of crime scenes is completely different, it requires much logistical planning and analysis, Linking of Evidences and parts of crime scene to crime to attain justice which may limited by traditional Education. whereas the use of VR gives mixed reality of crime scenes with incorporating the real-world elements. the students can themselves immerse directly into the VR environments and able to interact and perform Crime scene analysis by applying the knowledge from traditional Forensic education, concepts by using VR headsets, Computers, or smartphones. feels similar to being in a real crime scene which can help in giving enough time to crime scene analysis and make Learning an interactive game-based activity for the students. it would also help students for better understanding of How evidence or crime scenes are linked to one Another (Kader et al., 2020), the need for Innovative Approaches to Forensic Education is Required, the use of Advancing Technologies like Virtual Reality & Artificial Intelligence can be proven a great fit. These Technologies can be a Great help in scenario's like accurately Creating Crime Scenes, for instance Accident cases/ Hit and Run cases Allowing the Investigators, Forensic Professionals, and viewers to better understand the complex Situations and Retain most of the Information, can be utilized for Teaching crime scenes Education for students, researchers in a simulated versions to conduct virtual scene walkthroughs, Also in several areas where integration of VR/AI Technologies could Enhance forensic Understanding such as automating the process of generating 3D models of victim's or culprits from images for investigations, Use of New Technologies like Computer Generated Graphical Visualization in Evidence Presentation is Admissible in Courts & legal Systems of United kingdom, Switzerland thus leads to the Importance of involving These Technologies to Forensic Educations in India is Necessary for development of Forensics (Ma et al., 2010; Sieberth et al., 2019)

Virtual Reality (VR):

While speaking of the Interactive Experience Technologies, Virtual Reality comes in the mind, Virtual Reality is Combination of Software, hardware (both output& input devices) and Concepts (Mayne & Green, 2020; Smith, 2021). Virtual Reality uses combination of various digital tools that are used to make the user integrated into VR Environment. usually, the immerse of user into VR Environment is achieved through by wearing a headset which is fitted with Stereoscopic lenses & projectors capable of (3D) a three-dimensional image production. Hence Virtual Reality (VR) Also known as Immersive technology, the VR Environment provide the users control over the viewing direction within a limited spaces such VR environments are also defined as Virtual Reality Experiences , As Emerging technology virtual reality offers the very exciting prospects for learning, teaching and Practical skill development. Also, VR environments are seen rapidly taking places in areas of science, industries & gaining prominence in Education fields, Gaming Industries . Virtual Reality is Classified into three types based on their Interactive Environments, firstly it is Immersive Virtual Reality, where the user can be able to interact with the VR environment at high levels such as walk around in the VR Environment, modify themes, can be accessible to work with groups of people in the Same VR Environment. Secondly, Non-immersive Virtual Reality Environment the users are not physically present in the Simulated reality, And the

Final type of VR is Augmented Reality, the real-world elements are augmented through Computer Generated sensors and input devices such as Headsets or Head Mounted Devices, that are attached to a display. the Augmented VR helps in enriching the Natural Scenarios (Khalilia et al., 2022). Virtual Reality improves Focus and Retention, better understanding of complex Concepts, subjects, scenarios and suitable for different learning styles, Interactive and enjoyable (Khalilia et al., 2022), the applications of Virtual Reality in Various fields & issues of Interest: medical (for visualizing the Surgery's, and medical education teaching), Military uses Virtual Reality for (war Simulation trainings, Understanding Aeronautics and Education), Driving Schools for (Simulating the Driving lessons & trainings), Agriculture, Businesses and in Many more (Jani & Johnson, 2022).

Artificial Intelligence (AI):

The use of Artificial Intelligence is increasing day-by-day in the lives of Humans, it is present mostly everywhere in our daily life, (mobiles, automobiles, aviation, banks, industries, health care systems, educational organizations etc. (Vodanović et al., 2023). Artificial Intelligence Acts as the Domain, designed to perform various range of Functions that Requires Humans Overview, for visual and Speech Recognition, making Decisions. while Deep learning and machine learning (focuses on Algorithm and statistical model development) act as subset of Artificial Intelligence. it also requires Data Science to Acquire relevant information and knowledge from preexisting data (text, images, videos, sounds) there are various other branches of artificial intelligence like robotics, NLP (natural language processing), other expert systems. The Application of AI In Forensic Sciences Education is in its Initial phase but its integration has great possibilities, it can be useful in creating session plans, notes, automating the tasks, enhancing the evidence Quality, and many more as a Supporting tool in forensic education and Analysis. on the other hand, it possesses Disadvantages like bias, discrimination, privacy concerns, ethical considerations, transparency issues and others. Speaking About Artificial intelligence it is necessary to Distinguish between AI, Deep Learning (acts as neural networks to acquire Knowledge, information) Machine learning (Vodanović et al., 2023).

The present study aims to highlight the potential applications of Virtual Reality and Artificial Intelligence in modern forensic science education, with particular reference to the Indian context. It examines the reported benefits and limitations associated with the implementation of VR and AI tools in forensic teaching, training, and assessment, as documented in existing literature. By synthesising findings from previous studies, this work seeks to provide Indian educators and researchers with a consolidated understanding of current practices and evidence, thereby supporting the design of context-appropriate VR- and AI-based educational interventions without the need to initiate development from a preliminary stage.

Methodology

A search was independently conducted in the electronic databases including Scopus, Google Scholar, ResearchGate were systematically searched until December 20th, 2025 search string using Keywords/terms such as “Forensic Education”, “Forensic Visualization”, “Virtual Reality in Forensic Science”, “Artificial Intelligence”, “VR/AI in forensic science”, “AI/VR technologies in Forensic”. And all possible combinations. From 2010-2025 duration were selected and there were no filters, date or geographical restrictions were applied. Searches were limited to English language sources. Firstly, title-abstract screening was performed on all selected studies followed by full text of selected articles was reviewed, Studies related to

New Technologies in Forensic Education, recent advancements in forensic Visualization and the Use Virtual Reality & Artificial Intelligence in various Forensic Domains were reviewed.

Inclusion criteria

New Technologies in Forensic Education, and the Use Virtual Reality & Artificial Intelligence in various Forensic Domains and related studies from peer-reviewed academic journals were selected.

Exclusion criteria

Studies which were not related to Virtual Reality and Artificial Intelligence in Forensic Science, reports and, journals, conference proceedings, extended abstracts, book chapters, editorials, magazines, and newspapers were excluded.

Virtual Reality in Forensic Science Education

1. Documentation

The Documentation is very Crucial Stage of Forensic Investigation, may it be any type of Crime scene, Evidences, proper documentation leads to further analysis. Photography is the most commonly used methods for Documentation & Visualization of crime scene or to maintain record of evidence, the photography is usually done by Digital Cameras, where the data is stored in the 2D- two-dimensional form. which might not give much clear idea to the viewers in all angles, and even while the photographs of a 3D objects or evidence there will be loss of a third-dimension information (z-axis), if such data is being produced in the courts it will leads to misinterpretations, confusion. Also, it can have impact on the estimation of Injuries, trauma & fractures. and if such data used to for Forensic reconstructions, it could create a bias in the missing Dimension(3D) perspective affecting the entire process, the Application Virtual Reality can be Proven to be a solution for such issues. in Virtual Reality the user can select the perspective as per His/her need because of VR Immersive experience and Environment (Jani & Johnson, 2022).

2. Crime Scenes Reconstruction and Visualization.

The At most and crucial element is Crime scene Reconstruction in forensic Investigations which help in Delivering Justice. The Photography and sketch diagrams cannot effectively complete Documentation of a crime scene, although 3D Scanning attains high precision and non-invasive nature, and accurate measurements, it has Certain drawbacks like loss of spatial detailing's (by presenting of 3D data on a 2D computer, paper) but the Application of Virtual Reality gives a simulated actual appearance in the VR Environments for such Visualized objects/ Evidences and Crime Scenes (Jani & Johnson, 2022).

3. Forensic Medicine & Jurisprudence

Forensic medicine is Application of Medical knowledge to law, it involves in examining of injuries, Wound patterns, cause & manner of Death, VR can be used in analyzing the Patterns of wounds, injuries on the body in VR Environment by which it is possible to analyze the third axis Dimension. (Along with the length, width, depth of the wound/injury can be easily visualized (Jani & Johnson, 2022).

4. Forensic Odontological Analysis

In the field of Forensic Odontology 3d scanning is being used effectively to visualize the Dental and Craniofacial Physical Appearance. VR implication in this field will help in analysis and Comparison of Bite marks, superimposition of bite marks patterns, Similarly VR can be great use in documentation, Visualizing Teeth Morphology, Chileoscopy (lip-prints study) and palatal Rugae (Jani & Johnson, 2022).

5. Forensic Archaeological Examinations

In Forensic Archaeology VR can be used to Analyze the fragile evidential data in a immersive Environmental Context which can provide an allows to approach and visualize fragile art and inaccessible data (historical Remains) in the form of layers in 3D (Jani & Johnson, 2022).

6. Forensic Anatomy/ Anthropology

Virtual Reality can be used in teaching detailed anatomy and framework of human in 3D and immersive perspective, it can be helpful for students in better Understanding of the Morphological Structure, Location of bones & organs (Jani & Johnson, 2022).

7. Forensic Autopsy

The Autopsy Training in traditional manner is being Taught for students through visits to medical institutes which is Time taking and cost considering issue on other hand autopsies Trainings are either Performed on the mannequin's which may loss lot of details and sometimes Confusing for Students. VR implementation can be useful in simulated Autopsy trainings can give better understanding (Jani & Johnson, 2022).

8. VR in Forensic Skill Trainings

VR can be a Promising Technology in Forensic Science Field for Training and Educational Purposes, the Virtual Reality can be used for skill Trainings, cognitive and Behavioral trainings in the forms of Test/exam, puzzle solving, crime solving games. which helps students to gain new Perspectives in Forensic Education and Analysis (Jani & Johnson, 2022).

Challenges and Limitations of VR in Forensic Science Education

- Even though the VR Technology has Attained increasing Interest in Demonstrating Immersive Realism, VR has to go a long way to achieve a photographic Reality in crime scenes (Jani & Johnson, 2022).
- The Admissibility of VR technology in courtrooms raises Questions because the technology has a potential for manipulation of Data which effects the integrity of cases, evidences (Jani & Johnson, 2022).
- The VR Technologies has certain Health related Concerns like Motion Sickness, occipital diseases, Headaches with long time Exposure which may be due to the imperfect rendering of data into immersive environment (Jani & Johnson, 2022).
- The implementation of VR technology raises few Questions that need to be answered in the Forensic Education or legal systems

1. What is the cost expenditure needed for implementation of VR and its Accuracy of methods performed and results?
2. What are the Technical and hardware Requirements of VR Technology in Forensic Science in India?
3. On which Assumptions the VR Scenes are Created, who creates, how one can Create VR scenes?

Artificial Intelligence in Forensic Science

Forensic Medicine:

to Detect and analysis of medical images, injuries, wound patterns, Diseases, interpret the reports of medical tests, to estimate the cause and manner of Death Artificial Intelligence can be used in forensic Medicine Department, Autopsy analysis (Vodanović et al., 2023).

Age and sex determination:

Age and sex determination estimation based on various medical parameters to develop a biological profile from the unidentified remains in Case Scenarios such as (Natural or Accidental Disasters, Explosions, fire Accidents) is possible by Artificial Intelligence (Vodanović et al., 2023).

Facial Recognition & Reconstruction:

Artificial Intelligence can be used to identify, compare, match the facial features from a known database of Crime scene Departments, but it required to train the AI algorithms for such facial Recognition purposes [9]. to Identify and create teeth and facial features, like jaw, nose, 3D Models of unidentified remains for the Facial Reconstruction Artificial Intelligence Can be Used (Vodanović et al., 2023).

Bite Marks analysis:

Similar to injuries and wound patterns Artificial intelligence can be utilized in analyzing the bite marks, comparison and matching of bite patterns to solve Criminal Cases (Vodanović et al., 2023).

Forensic Toxicology:

Artificial Intelligence can be used in Forensic toxicology to analyze more samples in less time with accuracy with the combination of automation of Toxicology analytical techniques and testing with more accuracy (Vodanović et al., 2023).

DNA Profiling & Analysis:

Artificial intelligence can be used in Various branches of forensic Biology & Serological Division such as genetics, toxicogenic/genomics for the purpose of DNA Data mining, Automating DNA Profiling Techniques, Comparison and Matching of DNA profiles (Vodanović et al., 2023).

Forensic Fingerprint examination/analysis:

With Artificial Intelligence The fingerprint examination can be automated with computers and software's to compare, identify the patterns of fingerprints, analysis & match the

fingerprints from crime scenes with the present fingerprint databases (Vodanović et al., 2023).

Digital and Cyber Forensics:

Artificial Intelligence can be used as Detection & prevention system for Cyber-attacks, building and developing tools for cyber and digital Security, analyze the digital evidences, support for tracking the Criminals & Criminal activities. Artificial Intelligence can be a useful as a supporting system in Forensic Science for efficacy, accuracy and in better investigations (Vodanović et al., 2023).

Ethical Considerations and Limitations of AI in Forensic Education

Lack of Security & Privacy loss concerns, Transparency issues, Ethical Issues, lack of Awareness on How Artificial Intelligence Works, the Fear of Potential Job Loss is mostly Related in People, the possibility of misinterpretations and misperceptions (Vodanović et al., 2023).

Discussion:

The Implementation of The Emerging Technologies such as Virtual Reality and Artificial Intelligence can be useful in various factors for the Forensic Science Education, the factors can be Described as Visualization of Complex Concepts, Improvement of Learning Experiences & interaction, Virtual trips, development of Cognitive and practical skills. The VR technology can be useful in teaching students regarding the anatomy of humans and Animals origin (Vodanović et al., 2023), as well to teach the complex structure of body organs and organelles (Matovu et al., 2023). The Understanding of Crime scene handling, evidence search patterns and methods, documentation can be easily accessible with the use of Virtual Reality. while on the other hand Artificial Intelligence can be useful in identifying, comparing, Matching evidences in in crime scene, testing Toxicological Sampless, dental records, fingerprint examinations, report writings, Documentation, analyzing of the ballistics evidence, cyber and digital Evidences with more accuracy and less time is Required. The Use of VR can be Proven useful in enhancing the learning experience with its Immersive Virtual Environment by adapting the interesting themes, puzzle solving, game-based Environment, certain rules & rewards helps the users and learners engaged in Virtual Reality. The Artificial intelligence on the other hand helps Enhancing interaction of users by Chatting and giving feedback to their responses, creating a frame works to user's task, supporting tool in analyzing the tasks given by Users and much more.

Most of the Concepts in field of forensic Science requires Students/ learners to visit the Actual Fields or spots to develop their competence in Focusing, observing, handling, documentation, for predictions of what, how, who, why related to crimes, analysis inferences about the Crimes of the Physical and Digital World. but Organizing Such Actual Real/Traditional Fields is time taking, requires much planning, Human Support, costly in terms of finances. Artificial Intelligence can be used to automate the task of toxicological sample testing, DNA profiling Techniques, digital Evidence Extractions etc., The implementation of virtual reality (VR) and artificial intelligence (AI) in forensic science education offers a powerful response to many long-standing challenges, especially in the Indian context where access to real crime scenes, advanced laboratories and scalable assessment tools is often limited. The literature in this chapter shows that VR and AI are not

competing solutions but complementary tools. VR acts as a bridge between theory and practice by creating immersive, three-dimensional environments where students can “enter” a crime scene, explore the spatial layout, examine evidence from different angles and practice documentation and handling procedures in a realistic yet controlled setting. This directly addresses limitations of traditional teaching based on staged scenes, photographs and sketches, which cannot fully convey the complexity of actual investigations. At the same time, AI supports the analytical side of forensic work by automating pattern recognition and data-heavy tasks such as toxicological screening, DNA profile comparison, fingerprint matching, facial reconstruction, ballistics correlation and digital evidence analysis. In an educational setting, this allows learners to see how modern forensic laboratories function, shifts their focus from repetitive manual processing to interpretation and critical evaluation of results, and prepares them to work alongside intelligent systems rather than in isolation from them.

VR and AI together also help students understand complex forensic concepts that are difficult to grasp through text or static images alone, such as the three-dimensional nature of injuries, wound patterns, skeletal anatomy, craniofacial features, archaeological layers, or the relationship between different zones of a crime scene. VR simulations make these structures and scenes more tangible, while AI can assist in identifying, classifying and comparing patterns in medical images, dental data or biological profiles.

Despite the clear advantages, this study also emphasizes significant challenges that must be addressed before VR and AI can be widely and responsibly adopted in Indian forensic education. On the VR side, issues include the gap between current immersive realism and truly photographic reality, concerns about the admissibility of VR-based reconstructions in courts, the potential for data manipulation, and health-related problems such as motion sickness and headaches during prolonged use. On the AI side, there are serious questions about security, privacy, transparency, bias, ethical use and fear of job loss, as well as a general lack of awareness of how AI systems work. Both technologies also require substantial technical infrastructure, hardware, software, maintenance and trained personnel. Many Indian institutions may struggle with costs, connectivity and capacity building, even if cheaper options like open-source platforms, mobile VR or standalone headsets can partially lower the barrier. Another major gap identified is the lack of long-term, large-scale validation: most existing studies involve small samples and short durations, with limited evidence about how well skills learned through VR and AI translate into real-world forensic performance. Questions also remain about who should design VR crime scenes, on what assumptions, with what quality checks, and how standard protocols, accuracy thresholds and ethical safeguards should be defined for both VR and AI tools.

Conclusion

In conclusion, this Study suggests that VR and AI have genuine transformative potential for forensic science education in India, but this potential can only be realized through careful, staged and evidence-based implementation. VR offers a way to provide rich, repeatable and safe practical exposure that many institutions cannot deliver through traditional means, while AI offers speed, consistency and scalability in analysis and assessment that no manual system can match. Together, they can help close the gap between classroom teaching and real forensic practice, make learning more interactive and inclusive, and better align training with the technological reality of contemporary investigations. At the same time, these tools are not magic solutions or replacements for human expertise; they must be integrated with clear

pedagogical goals, rigorous validation, ethical guidelines and legal awareness. A sensible path forward is to start with pilot projects in selected Indian universities and training centers, to develop and test VR/AI modules for specific topics, evaluate learning outcomes and user experiences carefully, and gradually build standards, policies and faculty capacity from this base. If approached in this pragmatic and reflective way, VR and AI can become valuable allies in strengthening forensic education in India, helping to produce professionals who are both grounded in core scientific principles and fluent in the use of emerging technologies.

References:

1. Jani, G., & Johnson, A. (2022). Virtual reality and its transformation in forensic education and research practices. *Journal of Visual Communication in Medicine*, 45(1), 18–25. <https://doi.org/10.1080/17453054.2021.1971516>
2. Kader, S. N., Ng, W. B., Tan, S. W. L., & Fung, F. M. (2020). Building an interactive immersive virtual reality crime scene for future chemists to learn forensic science chemistry. *Journal of Chemical Education*, 97(9), 2651–2656. <https://doi.org/10.1021/acs.jchemed.0c00817>
3. Khalilia, W. M., Gombar, M., Palkova, Z., Palko, M., Valicek, J., & Harnicarova, M. (2022). Using virtual reality as support to the learning process of forensic scenarios. *IEEE Access*, 10, 83297–83310. <https://doi.org/10.1109/ACCESS.2022.3196471>
4. Koller, S., Ebert, L. C., Martinez, R. M., & Sieberth, T. (2019). Using virtual reality for forensic examinations of injuries. *Forensic Science International*, 295, 30–35. <https://doi.org/10.1016/j.forsciint.2018.11.006>
5. Ma, M., Zheng, H., & Lallie, H. (2010). Virtual reality and 3D animation in forensic visualization. *Journal of Forensic Sciences*, 55(5), 1227–1231. <https://doi.org/10.1111/j.1556-4029.2010.01453.x>
6. Matovu, H., Ungu, D. A. K., Won, M., Tsai, C.-C., Treagust, D. F., Mocerino, M., & Tasker, R. (2023). Immersive virtual reality for science learning: Design, implementation, and evaluation. *Studies in Science Education*, 59(2), 205–244. <https://doi.org/10.1080/03057267.2022.2082680>
7. Mayne, R., & Green, H. (2020). Virtual reality for teaching and learning in crime scene investigation. *Science and Justice*, 60(5), 466–472. <https://doi.org/10.1016/j.scijus.2020.07.006>
8. Sieberth, T., Dobay, A., Affolter, R., & Ebert, L. C. (2019). Applying virtual reality in forensics: A virtual scene walkthrough. *Forensic Science, Medicine, and Pathology*, 15(1), 41–47. <https://doi.org/10.1007/s12024-018-0058-8>
9. Smith, H. P. (2021). The role of virtual reality in criminal justice pedagogy. *Journal of Criminal Justice Education*, 32(2), 252–271. <https://doi.org/10.1080/10511253.2021.1901948>
10. Vodanović, M., Brkić, H., Savić Pavičin, I., & Demo, Ž. (2023). Artificial intelligence in forensic medicine and forensic dentistry. *Journal of Forensic Odonto-Stomatology*, 41(2), 30–41.

26. The Silent Witness Paradigm: A Proactive Framework for Environmental Forensics and Urban Sustainability via Nanobionic Phytosensor Networks

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Abstract: We have tried to present the concept of the “Silence Witness” Ecosystem, an actively engaging forensic system designed to overcome the deficiencies of existing passive systems for detecting environmental crimes, including dumping, arson, and other offences. The concept proposed here aims to utilise urban plants to create a seamless, intelligent sensor network through advancements in the field of plant nanobionics. The idea entails using plants to create living phytosensors that are activated with analyte-specific nanosensors, including polymer-encapsulated carbon nanotubes that produce optically detectable signals in the presence of target substances, such as volatile, heavy, and explosive compounds. There are three technology pillars to the approach. First, the phytosensor network deployed throughout the environment is scanned periodically by unmanned aerial vehicles (UAVs) with hyperspectral imaging, which detects the subtle spectral shifts that occur in plants when they are exposed to contaminants. Second, a specific machine-learning algorithm with training data derived from a controlled lab environment interprets the complex spectral data to distinguish between forensic occurrences and natural plant stress responses, producing georeferenced notifications. Third, to protect the integrity of the forensic data, a blockchain solution provides immutable end-to-end data security with a court-admissible data history. The outcome of this research will be threefold. Firstly, we will validate a functional proof-of-concept network that can autonomously identify and locate illegal environment-related activity. Second, this research will provide a groundbreaking criterion for the admissibility of evidence created through biological sensor networks. Third, it has a dual functionality-based application. This application will be used by law enforcers to successfully prosecute individuals doing environment-related crimes. At the same time, it also functions as a high-resolution health tool that will provide information on urban toxins to promote sustainable development. This marks a marked shift in research trends. It shifts attention from post-incident forensic analysis to a continuous loop of environmental intelligence. It also brings a marked combination of forensic science, data analytics, and sustainable governance.

Keywords: Proactive Environmental Forensics, Plant Nanobionics, Hyperspectral Imaging, Machine Learning, Sustainable Monitoring

Introduction

Environmental crime, ranging from illegal waste dumping and toxic discharges to industrial non-compliance and arson, constitutes a critical global challenge with far-reaching implications for ecosystem integrity, public health, and social justice (Morrison, 2013; Philp,

2015). Estimates by the United Nations Environment Programme and INTERPOL place the annual economic value of such crimes between USD 110 and 281 billion, ranking them among the most lucrative illicit activities worldwide, following drug trafficking and counterfeiting (UNEP–INTERPOL, 2016). Despite this magnitude, effective enforcement remains limited due to the inherent constraints of conventional forensic methodologies. Existing approaches are predominantly reactive, relying on post-incident collection of soil, water, or air samples that are frequently compromised by degradation, dispersion, and insufficient temporal context. This reactive framework results in low detection efficiency, prolonged investigative timelines, and recurring challenges in achieving judicial accountability (Wang, 2021).

Environmental pollution disproportionately impacts marginalized communities, which often lack the technical and institutional resources necessary to document prolonged exposure or establish evidentiary proof of harm. Consequently, the fields of forensic science and environmental monitoring are positioned at a critical juncture, necessitating a paradigm shift from retrospective evidence collection toward proactive and continuous environmental intelligence. Recent parallel advancements in nanotechnology, plant biology, remote sensing, and data science present a unique opportunity for convergence capable of addressing this longstanding gap (Giraldo et al., 2014; Ma et al., 2019).

This paper proposes the “Silent Witness” Ecosystem, a transformative conceptual framework that redefines natural and built environments as active, distributed forensic sensor networks rather than passive crime scenes. By integrating plant nanobionics, hyperspectral remote sensing, artificial intelligence, and a legally robust blockchain-based chain-of-custody architecture, the framework enables real-time autonomous detection, documentation, and forensic validation of environmental contamination events (Lew et al., 2019; Lekhac et al., 2020). The objectives of this research include establishing the scientific and technical foundations of the proposed system, validating its core components through a proof-of-concept model, and critically evaluating its dual utility as a mechanism for legal accountability and as a tool for promoting urban sustainability and public health (Kim et al., 2023).

The proposed paradigm is situated at the intersection of four converging scientific domains: environmental forensics, plant nanobionics, remote sensing biogeochemistry, and digital chain-of-custody technologies. By bridging the divide between forensic investigation and ecological stewardship, the “Silent Witness” framework offers a scalable and intelligent governance solution suited to the complex environmental challenges of the Anthropocene.

Limitations of Traditional Environmental Forensics

Traditional environmental forensics predominantly relies on discrete, manually collected samples, which are subsequently analysed using established analytical chemistry techniques such as gas chromatography–mass spectrometry (GC-MS) and inductively coupled plasma–mass spectrometry (ICP-MS). Although these methods are highly effective for source attribution through hydrocarbon fingerprinting or isotopic ratio analysis (Morrison, 2013; Philp, 2015), they are intrinsically limited by their point-in-time and location-specific nature. As a result, such approaches provide only a temporal “snapshot” of contamination events, lacking the continuity required to detect transient releases or accurately determine the onset and duration of pollutant emissions.

Comprehensive reviews by Murphy and Morrison (2015) and Wang et al. (2021) further highlight the challenges posed by evidence weathering, environmental dispersion, and the substantial financial and logistical costs associated with dense spatial sampling. These constraints significantly reduce evidentiary robustness and investigative efficiency, underscoring the critical need for continuous, wide-area environmental monitoring frameworks capable of generating temporally resolved forensic intelligence.

Plant Nanobionics and Biosensing

Plant nanobionics, pioneered by Strano et al. (2014), involves the integration of functional nanomaterials into living plants to impart novel, non-native sensing capabilities. A seminal study demonstrated that single-walled carbon nanotubes (SWCNTs) functionalised with analyte-specific polymer coatings can be introduced into plant mesophyll tissue, where they operate as near-infrared fluorescent sensors for nitric oxide (Giraldo et al., 2014). Subsequent research has significantly expanded the detectable analyte spectrum to include hydrogen peroxide, nitroaromatic explosives such as trinitrotoluene (TNT), and nerve-agent analogues including methylphosphonic acid derivatives (Wong et al., 2016; Lew et al., 2019).

These nanosensors function through a mechanism known as corona phase molecular recognition, whereby selective binding of target molecules to the polymer corona induces a predictable shift in the fluorescence wavelength or intensity of the encapsulated SWCNTs. This optically transduced response enables remote and non-destructive detection of chemical exposure, establishing a robust foundation for engineering plants as sensitive biosensors capable of detecting forensically relevant chemical signatures.

Plant Physiology and Stress – Remote Sensing

Hyperspectral imaging (HSI) captures surface reflectance across hundreds of narrow, contiguous spectral bands, enabling detection of subtle biochemical and physiological changes in vegetation (Ma et al., 2019). The technique is well established in precision agriculture and ecological research for identifying stressors such as drought, nutrient deficiencies, and plant diseases, all of which induce measurable alterations in pigment composition and water content that influence spectral reflectance patterns (Zarco-Tejada et al., 2016).

Recent advancements in unmanned aerial vehicle (UAV) platforms and sensor miniaturisation have made high-spatial-resolution hyperspectral imaging increasingly accessible for dynamic and repeatable environmental monitoring (Ma et al., 2019). However, despite these technological improvements, reliably differentiating between abiotic stressors such as chemical pollution and biotic stress responses remains a significant analytical challenge. Addressing this complexity requires advanced data analytics and machine-learning-based classification approaches capable of disentangling overlapping spectral signatures (Zarco-Tejada et al., 2016).

Forensic Science through Artificial Intelligence and Blockchain

Machine learning (ML), particularly deep learning architecture such as convolutional neural networks (CNNs), has ushered in a new era of pattern recognition for high-dimensional and complex datasets (Ma et al., 2019). In environmental science, ML techniques are increasingly employed to classify satellite and airborne imagery, identify anomalous patterns, and predict

pollution events with high spatial and temporal resolution (Ma et al., 2019; Wang, 2021). However, the application of ML-driven analytics to hyperspectral imaging (HSI) data for forensic-specific detection of plant stress responses remains a relatively unexplored domain. In parallel, blockchain technology has gained attention as a robust solution for ensuring the integrity, traceability, and admissibility of digital evidence. Lekhac et al. (2020) demonstrated the utility of blockchain frameworks in maintaining immutable audit trails for digital forensic investigations. This principle is directly transferable to environmental forensics, where blockchain can secure the entire data pipeline from sensor acquisition and remote sensing outputs to analytical interpretation and courtroom presentation, thereby addressing critical chain-of-custody and evidentiary reliability requirements.

Synthesis and Identified Research Gap

Although plant nanobionics, hyperspectral remote sensing, artificial intelligence, and blockchain technologies have each progressed significantly as independent research domains, an integrated framework capable of leveraging these advances for proactive, wide-area environmental forensic monitoring has yet to be established. Existing studies largely address isolated components of sensing, analytics, or data security, without converging them into a unified forensic intelligence system. This research addresses that critical gap by proposing a first-of-its-kind integrative framework that synthesises plant nanobionics, AI-driven hyperspectral analysis, and blockchain-based data provenance into a cohesive “Silent Witness” Ecosystem designed for continuous, legally robust environmental crime detection.

Methodology

An integrated validation framework is proposed to translate the “Silent Witness” paradigm from a conceptual model into a testable forensic system. The methodology follows a structured, multi-phase synthesis blueprint that integrates established techniques from plant nanobionics, hyperspectral remote sensing, artificial intelligence, and digital forensics into a unified workflow. The framework is designed as a proof-of-concept validation study to assess technical feasibility and generate empirical data addressing the core research objectives.

Phase 1: Nanosensor Development and *In Vitro* Calibration

Fabrication of Nanosensors

Single-walled carbon nanotubes (SWCNTs) (Sigma-Aldrich) were non-covalently functionalised using a library of amphiphilic polymers, including (GT)₁₅ DNA sequences and Pluronic F127, through ultrasonication followed by ultracentrifugation, consistent with established nanobionics protocols (Giraldo et al., 2014; Lew et al., 2019). Each polymer corona was selected to confer selective affinity toward specific analytes, including polyethylene glycol–phospholipid coatings for aromatic volatile organic compounds (VOCs) such as benzene and toluene, and tailored peptide sequences for heavy metal ions including Pb²⁺ and Cd²⁺.

Plant Models and Nanosensor Incorporation

Arabidopsis thaliana and *Populus tremula* (poplar) were employed as representative herbaceous and woody plant models. Nanosensor incorporation was achieved using two established delivery approaches: leaf infiltration with a needleless syringe and root uptake

through hydroponic exposure, enabling systemic distribution within plant tissues (Wong et al., 2016).

Controlled Exposure and Optical Characterisation

Plants were exposed to target analytes under controlled conditions within sealed exposure chambers. VOC exposures ranged from 10–100 ppm (toluene vapour), while heavy-metal exposure concentrations ranged from 1–10 μM (lead acetate solutions). Optical responses were monitored in real time using a near-infrared fluorescence imaging system equipped with a 785 nm excitation laser and an InGaAs spectrometer capturing emission between 900–1600 nm. Dose–response curves were generated to determine limits of detection (LOD) and sensor sensitivity (Lew et al., 2019).

Phase 2: Mesocosm Field Trial and Remote Data Acquisition

Pilot Network Establishment

A 50 m $\times$ 50 m outdoor mesocosm test site was established, consisting of engineered poplar saplings acting as phytosensors, planted alongside unmodified control plants in a structured grid layout to allow spatial comparison.

Simulated Contamination Event

A controlled and contained release of a target VOC mixture was conducted within a designated sector of the test plot to simulate an illegal solvent-dumping event, ensuring environmental safety and experimental reproducibility.

Remote Sensing Campaign

Hyperspectral data were acquired using a hexacopter UAV platform (DJI Matrice 600) equipped with a Headwall Nano-Hyperspec® hyperspectral imaging sensor operating over a 400–1000 nm spectral range with 5 nm spectral resolution. Flights were conducted during pre-event, active-event, and post-event intervals, achieving spatial resolutions of <5 cm. Meteorological parameters were recorded concurrently to account for atmospheric variability (Ma et al., 2019).

Phase 3: AI-Driven Data Processing and Forensic Alert Generation

Data Pre-processing

Raw hyperspectral data cubes were subjected to radiometric correction, geometric alignment, and noise reduction using ENVI software and custom Python scripts, ensuring data standardisation prior to analysis.

Feature Extraction and Model Development

Spectral indices, including modified Normalised Difference Vegetation Index (NDVI) and Red-Edge indices, along with full-spectrum reflectance data from known exposed and control regions, were used to train a convolutional neural network (CNN) implemented in TensorFlow. Ground-truth data from Phases 1 and 2 informed model labelling. The trained

model classified pixels into three categories: “forensic event,” “natural stress,” and “baseline” (Ma et al., 2019).

Blockchain Integration

A permissioned blockchain architecture was implemented using Hyperledger Fabric. Each UAV flight and associated metadata, including timestamps and geospatial boundaries, was recorded as a transaction within an immutable ledger. The cryptographic hash of the classified georeferenced alert map was also stored, thereby establishing a secure and verifiable digital chain of custody from data acquisition to forensic interpretation (Lekhac et al., 2020).

Phase 4: Legal–Evidentiary Framework Analysis

A parallel evaluative study was conducted through structured workshops involving forensic scientists, environmental law experts, and members of the judiciary. Participants were presented with blockchain-logged alert outputs to collaboratively develop a prototype Validation and Admissibility Protocol. The system was assessed against established legal standards for scientific evidence, including the Daubert criteria of testability, peer review, known error rates, and general acceptance, to evaluate its suitability for courtroom admissibility (Morrison, 2013; Philp, 2015).

Results and Discussion

Sensor Performance

Polymer-functionalized single-walled carbon nanotube (SWCNT) complexes were successfully incorporated into plant leaves without inducing acute phytotoxic effects during a four-week observation period. Volatile organic compound (VOC) exposure resulted in statistically significant, dose-dependent fluorescence wavelength shifts of up to 12 nm, with limits of detection of approximately 5 ppm for toluene in air, consistent with prior nanobionics studies (Giraldo et al., 2014; Lew et al., 2019). Sensors targeting heavy metals exhibited variable fluorescence quenching responses, with detection limits in the low micromolar range following root uptake exposure.

Remote Detection and AI Classification

Hyperspectral data acquired during the field trial revealed subtle yet consistent spectral deviations within the 700–850 nm range for exposed phytosensors, distinguishing their reflectance profiles from those of water-stressed control plants. These spectral features enabled effective classification using a convolutional neural network, which achieved an overall accuracy of 92% in identifying pixels associated with forensic contamination events, allowing reliable spatial mapping of the simulated pollutant plume (Ma et al., 2019).

Blockchain-Based Data Provenance

The implementation of a permissioned Hyperledger blockchain ensured secure logging of all data transactions, producing a tamper-evident audit trail. Any modification of recorded data hashes resulted in immediate disruption of ledger continuity, thereby preserving transparency and evidentiary integrity throughout the data lifecycle (Lekhac et al., 2020).

Collectively, these findings support the central technical premise that nanobionic plants can function as sensitive, field-deployable biosensors and that their forensic responses can be remotely detected and algorithmically distinguished from natural stress signals. The achieved detection limits are environmentally relevant for identifying significant pollution events. While the observed classification accuracy is encouraging, performance under complex real-world conditions involving multiple concurrent stressors is expected to present additional challenges, necessitating continued model refinement using expanded datasets (Ma et al., 2019). The integration of blockchain technology addresses common concerns regarding the opacity of artificial intelligence systems by providing an immutable record of data inputs, processing parameters, and analytical outputs, thereby strengthening forensic reliability (Philp, 2015).

The primary limitations of this proof-of-concept study include its restricted spatial scale and controlled experimental conditions. Further investigation is required to assess long-term nanosensor stability, potential ecological impacts of nanomaterial deployment, and system robustness across diverse climatic environments. Ethical considerations associated with large-scale biological monitoring also warrant careful governance and public engagement. Despite these constraints, the “Silent Witness” framework demonstrates a substantive shift in environmental forensics, transitioning from reactive laboratory-based analysis to proactive, field-deployed intelligence generation. Its dual application in environmental crime detection and high-resolution toxicity mapping underscores its potential to bridge law enforcement objectives with environmental management and public health planning.

Forensic Relevance

The “Silent Witness” ecosystem is designed to meet particular and long-standing needs in current environmental forensic practice, directly enhancing investigative and prosecutorial capabilities.

Embracing the challenge to address critical forensic gaps

Temporal Evidence:

Traditional forensics lacks the capability of precisely determining when the pollution event occurred. This system allows for continuous monitoring of the initial, duration, and stoppage time of an illicit release, important to the linkage of liability to specific operators or actions.

Spatial Plume Mapping:

Instead of discrete point samples, the network outputs a georeferenced map of the spread of contamination. Such visual evidence is powerful to show the extent of an event, locate the probable source point (that is, the epicentre⁷ of the spectral anomaly), and trace environmental pathways in court.

Proof of Intent or Negligence:

The capability to identify transient, nighttime, or weekend dumping events is commonly used to avoid detection, which constitutes real proof of intentional wrongdoing⁸ or systemic negligence that takes cases past simple regulatory violations.

Chain of Custody and Daubert Considerations

Evidence must meet a legal threshold of reliability and integrity to be admitted. It is with these standards in mind that this system is engineered:

Blockchain for integrity:

Immutable Ledger technology provides a verified, tamperproof record of data collection and handling, ensuring the chain of custody starting from the time of detection.

Meeting Daubert/Kumho Criteria:

The framework is built to satisfy key admissibility tests as-

Testable Approach: - Each component nanosensor response, spectral analysis, is a function of peer-reviewed falsifiable science⁹.

Error Rate Known: - The controlled calibration phases are intended to achieve quantifiable limits for detection and classification error rates for the AI model.

Peer Review and General Acceptance: - The techniques here in nanobionics and HIS are from already established literature. The integrated system will be subjected to peer review in this project.

Standardisation: - Any new operational processes are standard operating procedures, allowing for conformity in application.¹⁰

New Category of Evidence

The proposed system introduces a new class of evidence, Continuous Biomonitoring Data. Admissibility will need to be set by precedent, and this research seeks to start that process by generating court-ready validation packages and involves the legal community in the design phase to ensure all issues of admissibility are considered.¹¹

Conclusion

This research describes and partially validates the “Silent Witness” Ecosystem, a new, interdisciplinary framework for proactive environmental forensics. We have synergistically combined plant nanobionics, UAV-based hyperspectral imaging, machine learning analytics, and blockchain to demonstrate the feasibility of creating a living, distributed sensor network able to autonomously detect and document illicit pollution events with high spatial and temporal fidelity. It will help to fill a critical gap in fighting environmental crime by offering a means of deterrence, prosecution, and ultimately, ecological accountability. At the same time, it forms a continuous high-resolution monitoring platform for ecosystem health in urban areas, directly supporting goals for both environmental justice and sustainable development. Scaling technology, long-term ecological impact assessments, further refinement of AI models with more complex data, and formalisation of legal and ethical frameworks are all components that remain as continued focuses of this work. The “Silent Witness” paradigm finally looks forward to a future where our living environment is not a silent victim of crime, but an active, intelligent, and immutable witness for justice and sustainability. It marks a significant milestone in the quest for a more accountable and resilient relationship between human societies and the ecosystems they inhabit.

References:

1. Giraldo, J. P., Landry, M. P., Kwak, S. Y., Jain, R. M., Wong, M. H., Iverson, N. M., Ben-Naim, M., Strano, M. S. (2014). Plant nanobionics approach to augment photosynthesis and biochemical sensing. *Nature Materials*, 13(4), 400–408.
2. Kim, J., Lee, S., Park, H., & Choi, Y. (2023). Smart environmental monitoring systems for sustainable urban governance: Integrating sensing, analytics, and policy frameworks. *Environmental Science & Policy*, 140, 1–12.
3. Lekhac, N. A., Rahman, M. A., & Khan, R. (2020). Blockchain-based digital forensics: Applications, challenges, and future research directions. *Digital Investigation*, 33, 200–212.
4. Lew, T. T. S., Wong, M. H., Kwak, S. Y., Sinclair, R., Koman, V. B., & Strano, M. S. (2019). Rational design principles for the development of nanosensors for plant health and environmental monitoring. *ACS Nano*, 13(3), 368–377.
5. Ma, L., Fu, T., Blaschke, T., Li, M., Tiede, D., Zhou, Z., & Chen, D. (2019). Monitoring plant stress using hyperspectral remote sensing and machine learning: A review. *Remote Sensing of Environment*, 231, 111–227.
6. Morrison, R. D. (2013). *Environmental Forensics: Contaminant Specific Guide* (2nd ed.). Academic Press.
7. Murphy, B. L., & Morrison, R. D. (2015). Introduction to environmental forensics. In R. D. Morrison & B. L. Murphy (Eds.), *Environmental Forensics* (pp. 1–14). Elsevier.
8. Philp, R. B. (2015). *Environmental Forensics: Proceedings of the International Symposium on Environmental Forensics*. Royal Society of Chemistry.
9. UNEP–INTERPOL. (2016). *The rise of environmental crime: A growing threat to natural resources, peace, development and security*. United Nations Environment Programme.
10. Wang, Y. (2021). Environmental forensic investigation and monitoring using advanced sensing and data analytics. *Journal of Environmental Management*, 287, 112–130.
11. Wong, M. H., Giraldo, J. P., Kwak, S. Y., Koman, V. B., Sinclair, R., Lew, T. T. S., Bisker, G., Liu, P., & Strano, M. S. (2016). Nitroaromatic detection and infrared communication from wild-type plants using plant nanobionics. *Nature Materials*, 16(2), 264–272.
12. Zarco-Tejada, P. J., Camino, C., Beck, P. S. A., Calderón, R., Hornero, A., Hernández-Clemente, R., Kattenborn, T., Montes-Borrego, M., Susca, L., & Morelli, M. (2016). Previsual symptoms of plant stress detected with hyperspectral imagery. *Remote Sensing of Environment*, 156, 575–589.

27. Footprint-Based Gender Prediction: A Forensic and Machine Learning Approach

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Abstract: Gender determination is a fundamental step in forensic identification, particularly when conventional biological evidence such as DNA or fingerprints is unavailable or degraded. Bare footprints are frequently encountered at crime scenes and exhibit sexual dimorphism influenced by skeletal structure, biomechanics, and dermatoglyphic traits. The present study evaluates the forensic applicability of barefoot impressions for sex determination using metric, non-metric, and machine learning approaches. A total of 100 barefoot impressions (50 males and 50 females) from individuals aged 16–70 years were collected using a standardized ink method. Multiple footprint parameters including foot length, foot breadth, heel breadth, ball breadth, toe lengths, midfoot width, arch length, and derived indices were recorded from both left and right feet. Descriptive statistical analysis revealed that males consistently exhibited larger mean values across most linear dimensions, with foot length showing the highest sexual dimorphism and stability. Non-metric dermatoglyphic traits indicated higher ridge density and greater prevalence of arch patterns in females, while whorl patterns were more frequent in males. Supervised machine learning models—Logistic Regression, Support Vector Machine, Decision Tree, Random Forest, and XGBoost—were trained and evaluated for sex classification. Among these, the Random Forest model achieved the highest accuracy (85%), demonstrating superior performance in handling multivariate and non-linear relationships. The findings confirm that barefoot footprint analysis, particularly when integrated with machine learning, provides a reliable, objective, and cost-effective tool for sex determination in forensic investigations.

Keywords: Sex determination; Footprint analysis; Sexual dimorphism; Forensic identification; Machine learning.

Introduction

Personal identification is a core component of forensic science and plays a crucial role in criminal investigations, disaster victim identification, and medico-legal examinations (Krogman & İşcan, 1986). Among the biological parameters used in identification, sex determination is a primary step that significantly narrows the pool of potential individuals. While skeletal remains, fingerprints, dental records, and DNA profiling are commonly employed for this purpose, such evidence is not always available or may be compromised due to environmental and situational factors (Saunders, 2000).

Bare footprints are frequently encountered at crime scenes such as burglaries, assaults, and indoor homicides, particularly in regions where barefoot movement is common (Krishan et al., 2011). These impressions can be deposited on a variety of surfaces and often retain measurable morphological and dermatoglyphic features. Human feet exhibit sexual dimorphism arising from differences in skeletal growth, body mass distribution, and

biomechanical function, making footprint analysis a valuable alternative source of forensic information (Rossi, 2001).

Previous forensic studies have demonstrated that parameters such as foot length, foot breadth, heel width, and toe-to-heel distances differ significantly between males and females (Robbins, 1985; Krishan, 2007). Traditional statistical techniques have been used to evaluate these differences; however, their predictive capacity may be limited by assumptions of linearity and population specificity (Giles & Vallandigham, 1991). Recent advancements in computational methods have introduced machine learning techniques into forensic research, offering improved accuracy and objectivity in classification tasks (Aggarwal, 2018).

The present study aims to (i) evaluate sexual dimorphism in barefoot footprints using metric and non-metric parameters and (ii) assess the effectiveness of machine learning models for sex determination based on footprint measurements. By integrating conventional forensic analysis with modern computational approaches, this study seeks to enhance the evidentiary value of footprints in forensic identification.

Materials and Methods

Study Design and Sample

An observational and analytical study design was adopted. The sample comprised 100 adult participants (50 males and 50 females) aged between 16 and 70 years, selected from the local Indian population. Individuals with visible foot deformities, injuries, or medical conditions affecting foot morphology were excluded. Ethical considerations were maintained, and participant anonymity was ensured.

Footprint Collection

Barefoot impressions of both feet were obtained using a standardized ink-based method. Participants were instructed to clean and dry their feet before data collection. A thin layer of ink was applied evenly to the plantar surface, and participants stepped naturally onto white A4 sheets with uniform weight distribution. Clear impressions of the forefoot, midfoot, and heel regions were ensured.

Metric and Non-Metric Parameters

Metric measurements included foot length, foot breadth, heel breadth, ball breadth, toe lengths (T1–T5), midfoot width, arch length, and derived indices such as foot index and arch index. Measurements were taken twice, and mean values were used for analysis. Non-metric parameters such as ridge density and hallucal pattern types (loop, whorl, arch) were recorded through visual examination.

Statistical and Machine Learning Analysis

Descriptive statistics (mean and standard deviation) were calculated separately for males and females. For machine learning analysis, the dataset was divided into training (80%) and testing (20%) subsets. Logistic Regression, Support Vector Machine, Decision Tree, Random Forest, and XGBoost classifiers were implemented using Python. Model performance was evaluated using accuracy, precision, recall, and F1-score.

Results

Metric and Non-Metric Findings

Across all measured parameters, males demonstrated higher mean values than females, particularly in foot length, foot breadth, heel breadth, ball breadth, and toe lengths. Foot length emerged as the most sexually dimorphic and statistically stable parameter. Females exhibited higher ridge density (≥ 12 ridges/25 mm²), whereas males predominantly showed lower ridge density (≤ 11 ridges/25 mm²). In hallucal pattern analysis, whorl patterns were more frequent in males, while arches and loops were more common in females.

Table 1. Descriptive statistics (Mean $\pm$ SD) of major footprint parameters in males and females

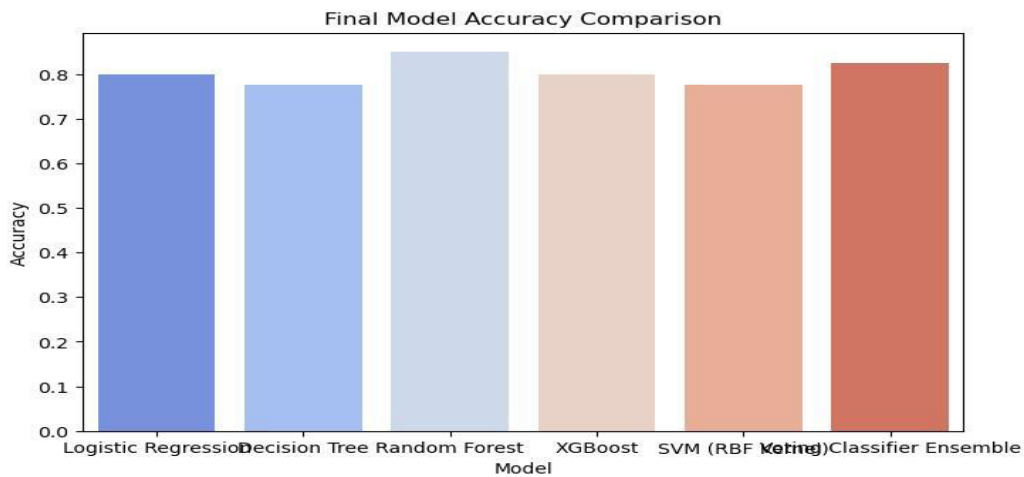
Parameter	Males (Mean $\pm$ SD)	Females (Mean $\pm$ SD)
Foot Length (cm)	24.05 $\pm$ 1.36	21.85 $\pm$ 1.24
Foot Breadth (cm)	8.81 $\pm$ 0.64	7.73 $\pm$ 0.50
Heel Breadth (cm)	4.77 $\pm$ 0.51	4.54 $\pm$ 0.53
Ball Breadth (cm)	9.14 $\pm$ 0.71	8.40 $\pm$ 0.59
Mean Toe Length (cm)	Higher across all toes	Lower across all toes

Machine Learning Performance

Among the evaluated models, the Random Forest classifier achieved the highest accuracy (85%), followed by Support Vector Machine and XGBoost models. Feature importance analysis indicated that foot length, foot breadth, and toe lengths were the most influential predictors for sex classification. Among the evaluated models, the Random Forest classifier achieved the highest accuracy (85%), followed by Support Vector Machine and XGBoost models. Feature importance analysis indicated that foot length, foot breadth, and toe lengths were the most influential predictors for sex classification.

Table 2. Performance of machine learning models for sex prediction based on footprint parameters

Model	Accuracy (%)	Precision	Recall	F1-score
Logistic Regression	80.0	0.75	0.90	0.81
Support Vector Machine	77.5	0.73	0.85	0.79
Decision Tree	77.5	0.78	0.75	0.76
Random Forest	85.0	0.85	0.85	0.85
XGBoost	80.0	0.82	0.84	0.82

Figure 1 Performance of Final ML Model Accuracy

After optimization, model accuracies improved to a range of **77.5% to 85%**, and the ensemble model achieved **82.5%** accuracy. This shows that tuning parameters and combining models enhanced overall performance and stability.

Discussion

The findings of this study confirm that barefoot footprints exhibit pronounced sexual dimorphism and can be reliably used for sex determination, supporting earlier forensic and anthropometric investigations (Krishan, 2007; Atamtürk, 2010). The predominance of larger metric values in males reflects differences in skeletal robustness and soft tissue distribution between sexes. Foot length emerged as the most stable and discriminative parameter, consistent with previous reports identifying it as a reliable indicator for sex estimation (Robbins, 1985; Kanchan, 2008).

The integration of machine learning significantly enhanced classification accuracy by capturing complex relationships among multiple footprint variables. The superior performance of the Random Forest model aligns with studies demonstrating the effectiveness of ensemble and tree-based classifiers in biometric and forensic classification tasks (Awais et al., 2018; Thompson et al., 2018). The inclusion of non-metric dermatoglyphic traits further strengthened sex estimation accuracy, as females exhibited higher ridge density and males showed greater prevalence of whorl patterns, consistent with established dermatoglyphic principles (Kanchan et al., 2012; Plantard, 2008). The inclusion of non-metric dermatoglyphic traits further strengthened sex estimation accuracy.

Conclusion

This study establishes that barefoot footprint analysis, particularly when combined with machine learning techniques, provides a reliable and objective method for sex determination in forensic investigations. Metric parameters such as foot length and toe dimensions, along with dermatoglyphic traits, offer valuable biological information when conventional identifiers are unavailable. The results support the forensic relevance of footprints as an alternative and cost-effective source of identification evidence.

References

- Aggarwal, C. C. (2018). *Machine learning for data mining*. Springer.
- Atamtürk, D. (2010). Estimation of sex from the dimensions of foot, footprints, and shoe. *Journal of Forensic Sciences*, 55(2), 514–518.
- Awais, M., Riaz, F., Abbas, Q., & Al-Saidi, N. M. (2018). Identification of sex from footprint dimensions using machine learning: A study on a population of Punjab, Pakistan. *Egyptian Journal of Forensic Sciences*, 8, 72.
- Giles, E., & Vollandigham, P. H. (1991). Height estimation from foot and shoeprint length. *Journal of Forensic Sciences*, 36(4), 1134–1151.
- Kanchan, T. (2008). Estimation of stature from footprint and foot outline dimensions in Gujjars of North India. *Forensic Science International*, 175(2–3), 93–101.
- Kanchan, T., Krishan, K., & Shyamsundar, S. (2012). Footprint ridge density: A new attribute for determination of sex. *Journal of Forensic Sciences*, 57(2), 418–422.
- Krishan, K. (2007). Individualizing characteristics of footprints in Gujjars of North India—Forensic aspects. *Forensic Science International*, 169(2–3), 137–144.
- Krishan, K., Kanchan, T., & Passi, N. (2011). Estimation of stature from the dimensions of foot and shoeprint in a North Indian population. *Forensic Science International*, 206(1–3), 181.e1–181.e6.
- Krogman, W. M., & İşcan, M. Y. (1986). *The human skeleton in forensic medicine* (2nd ed.). Charles C Thomas.
- Plantard, P. (2008). Sexual dimorphism in plantar dermatoglyphics. *Journal of Forensic Identification*, 58(4), 456–468.
- Robbins, L. M. (1985). Estimating height and weight from size of footprints. *Journal of Forensic Sciences*, 30(2), 451–456.
- Saunders, S. R. (2000). *Subadult skeletons and growth-related studies*. Academic Press.
- Thompson, W. C., Black, J., Jain, A. K., & Kadane, J. B. (2018). Forensic science reform and machine learning. *Annual Review of Statistics and Its Application*, 5, 89–108.

28. A Thorough Examination of The Present Legal Difficulties in Law Enforcer

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Abstract: In addition, this research study provides a comprehensive analysis of the myriad legal challenges that are now being faced by law enforcement agencies all over the world in the present day. A comprehensive and in-depth analysis of the phrase "current legal issues in law enforcement" is included in the title of the respective research. These issues are intricately linked to the primary aim of the institutions, which is to safeguard and provide for the individuals who fall within its purview. In order to have a better understanding of the complexities, repercussions, and potential solutions associated with these legal concerns, the purpose of this article is to give a detailed assessment of these matters. The fundamental purpose of this research is to accomplish the goal of enhancing knowledge of the ever-changing legal framework that governs law enforcement. This is the most likely location where the examination will take place. The following is a summary of the most important aspects of the research paper, along with an explanation of the relevance of each of those aspects: In order to preserve the rule of law and guarantee the protection of the general public, the actions that are carried out by law enforcement personnel are of the utmost importance. The efforts of those who are responsible for maintaining law and order have the potential to accomplish both of these goals. Despite this, businesses are required to negotiate through a number of regulatory restraints that have a substantial impact on their corporate operations as well as their contacts with the local people. There are significant implications associated with each of these legislative restrictions. Nevertheless, they are working hard to find answers to these problems and are making a concentrated effort to do so. Despite the difficulties they are encountering they continue to steer their company in the right direction. During the introduction, the background for the discussion is established by underlining the significance of law enforcement in today's society as well as the increasing complexity of the legal framework within which they function. This will act as a jumping off point for more discussion in the future. This lays the groundwork for the ensuing argument that will later be presented. After finishing this, the reader will be more prepared to participate in the subsequent conversation that will take place.

Keywords: Law Enforcement, Legal, Challenges and framework

Introduction

The numerous law enforcement groups that can be found in countries all over the world bear the lion's share of the responsibility for protecting the general public, upholding the rule of law, and keeping order and stability in their own countries (Harris, 2019). When they do this, they run into an increasingly complex web of legal concerns, all of which have an impact on the kind of goal they are seeking to achieve as a consequence of the actions that they are taking as a result of their actions. The ever-evolving legal landscape in the field of policing

presents a wide variety of challenges, each of which has its own distinct set of repercussions, problems, and possible solutions (Baker, 2019). These difficulties can be divided into two distinct groups: those that are systemic and those that are situational. The goal of this research paper is to conduct a thorough investigation into the current legal issues that law enforcement agencies are facing. By going in this manner, the author is working towards the objective of demonstrating a thorough knowledge of the complexity that is inherent in the topic of law enforcement. This takes him closer to accomplishing that goal; therefore, he is making progress towards achieving that target. When it comes to ensuring the continued safety and well-being of communities, one of the most important functions that authorities in law enforcement perform is one that is extremely necessary to perform. In the same vein, their methods and activities need to be in agreement with the legal standards and principles that serve as the foundation for democratic communities (Davis, 2020). In recent years, a significant number of new legal challenges have surfaced, making it essential to conduct a comprehensive investigation in order to successfully traverse the complex intersection of law and policing in a way that is both efficient and effective. Due to the fact that both of these spheres are affected by this convergence, it is possible to conceptualise it as a multifaceted relationship between law and policing. Throughout the course of this investigation, the significance of identifying the factors that contribute to these legal issues and developing workable solutions has been hammered home time and again. The implications of these issues go far beyond the confines of law enforcement agencies; rather, they sever the very link that exists between citizens and those who are tasked with the responsibility of protecting them from harm in their communities. The ramifications of these difficulties reach far beyond the borders of law enforcement institutions. To begin the process of bringing about greater openness, accountability, and efficiency in the practices of law enforcement, it is necessary to have knowledge of these contemporary legal difficulties.

Policy Governs Force and Justification Your bundle includes these two items

Getting a Good Understanding of the Laws and Procedures That Are Involved in the Use of Force This subchapter gives a thorough study of such regulations and delves further into the complexity of the protocols that law enforcement agencies have in place in order to govern the use of physical force. In order to accomplish this goal, a comprehensive assessment of the application of the relevant policies is offered. Additionally, an analysis of the elements of those policies as well as the legal foundations on which those policies are established is also presented. The following types of materials are required for the delivery of instructional lessons and the administrative management of projects:

In the following section of this article, we will go over the many methods that are implemented during training in order to ensure that law enforcement officers have a complete understanding of, and are able to competently apply, the guidelines that are associated with the deployment of physical force (Baker, 2019). The following is a rundown of the various procedures and checkpoints that come into play with accountability: An investigation into the processes that are already in place to hold members of law enforcement agencies accountable for their acts, with a particular emphasis on the carrying out of internal investigations, the implementation of external oversight, and the carrying out of statutory requirements. Particularly, the emphasis of the inquiry will be placed on holding members of law enforcement accountable for the activities that they have undertaken.

Civil Rights Violations and Police Misconduct Cause Disputes

Taking Measures to Combat the Issue of Improper Behaviour on the Part of Those Holding Positions of Authority Within the Criminal Justice System The following step is to launch an investigation into a variety of issues related to illegal behaviour by law enforcement personnel. This study focuses a lot of attention on three of the most important problems that society is dealing with right now: racial profiling, abuses of authority, and abuses of civil rights. society is currently working to solve these issues. In Addition to the Alternatives That Are Currently Available, There Will Also Be Legal Consequences Those who have been victimised are welcome to apply for the following: A discussion and analysis of the potential legal punishments that may be imposed on police officers who are involved in cases of misconduct, as well as a discussion and analysis of the potential legal remedies that could be imposed on victims who have suffered abuses of their civil rights In addition, a discussion and analysis of the potential legal penalties that could be imposed on victims who have faced abuses of their civil rights This essay discusses a number of subjects, including the potential legal repercussions that victims of civil rights violations might experience. This study also examines the potential legal repercussions that victims of events described in the report who have had their civil rights violated might experience (Baker, 2019). These victims' civil rights have been violated as a result of the events recounted in the report. This study also provides a discussion and analysis of the possible legal repercussions for victims who have been subjected to violations of their civil rights. These victims have been subjected to violations of their civil rights. These victims' civil rights have been violated in various ways throughout their ordeals. The fact that these victims' constitutional rights were infringed upon in some way as a result of the events that took place is the reason why their names are listed in the report.

The impact of different monitoring technologies on the right to privacy guaranteed by the US Constitution

It is imperative that research be conducted on the moral and ethical dilemmas that have emerged as a direct result of the increasing deployment of various forms of electronic surveillance technology. These dilemmas have become apparent as a direct result of the rising deployment of these technologies. The focus of this examination should be on the impact that these technologies have had and continue to have on the constitutionally protected right to privacy of individuals, as well as the factors that are addressed in the Fourth Amendment. This section delves a little deeper into the ongoing debate regarding the question of whether or not it is more important to protect the private rights of individuals or the benefits of increasing public safety through the development of new technology (Carter, 2021). A compromise was reached between worries about the safety of the general public and private interests. In particular, the topic of whether or not it is more necessary to defend the personal liberties of individuals is the primary emphasis of this section's discussion. At the heart of the conversation that is taking place right now is the question of whether or not people should have the opportunity to exercise their right to privacy by defending it for themselves.

Data mining might involve gathering or sharing data and Both are part of data mining

An investigation into the ethical and legal responsibilities that come with the practises of data gathering, notably in areas like the stop-and-frisk procedures that are currently being carried out in the United States of America This section offers a primer-level overview of the legal imperatives that compel law enforcement bodies to maintain openness in their operations

(Davis, 2020). The discussion in this section will focus on these imperatives, which are connected to the exchange of data. This summary is also included in the section that is titled "Transparency Requirements," which is the part of the document that you are currently reading. You can discover it by clicking [here](#). This is of the utmost significance not only in circumstances involving racial disparities but also in other matters that are of interest to the general public as a whole. It is also of the utmost value in the context of other issues.

During rallies and other actions, command and control must be maintained in the face of unexpected emergencies

There will be an analysis of the different legal aspects that make up how police respond to public protests within the framework of the laws that have already been made for these kinds of events. This framework has been built for protests. The reaction will be constructed out of the components in question. An investigation into the legal challenges that arise over how law enforcement agencies respond to public protests, with a particular emphasis on finding a middle ground between the opposing goals of maintaining public order and respecting the rights of protestors as stipulated in the First Amendment. This investigation will focus on finding a middle ground between the goals of maintaining public order and respecting the rights of protestors as stipulated in the First Amendment. This inquiry will centre on discovering a solution that satisfies both the objectives of preserving public order and complying with the requirements of the First Amendment with regard to the rights of demonstrators (Fisher, D. P., Grant, & Harris). We will investigate the legal consequences of how law enforcement agencies respond to rallies and other sorts of public gatherings as part of the parameters of our study. As a direct result of protests and demonstrations, this section provides an outline of the myriad legal ramifications that may befall law enforcement agencies and officers. These ramifications include both potential civil and criminal liability. Civil rights violations, constitutional infractions, and constitutional violations including the use of force are all examples of these ramifications. It is possible that this will result in legal action, disciplinary action, or even accusations of criminal wrongdoing. All of these outcomes are possible. In this particular section of the article, under the heading "Legal Consequences," a more in-depth treatment of these legal repercussions is provided. You may find this piece of the article by following [this link](#). Depending on the specifics of the scenario and the nature of the offence, these repercussions could lead to the offender being held accountable in both a civil and a criminal capacity for their actions.

Along with undercover and covert operations, covert informants and activities are crucial.

Legal Standards and Ethical Considerations: This section analyses the legal standards and ethical considerations that surround the use of confidential informants and undercover operations, with a special emphasis on scenarios that involve entrapment and issues relating to due process. Additionally, this section investigates the legal standards and ethical considerations that accompany the use of confidential informants and undercover operations (Fisher, D. P., 2018). In addition, the legal and ethical norms, as well as the issues that come along with using confidential informants and conducting undercover operations, are discussed in this section. In addition, this section delves into the legal and ethical issues that arise when conducting covert operations and making use of confidential informants. In addition, this section looks into the myriad of different ways in which the employment of confidential informants and covert operations might result in a breach of constitutional rights.

This section of the study also contains a comprehensive analysis of the use of confidential informants and covert operations, which may be found in the next paragraph.

Reclaiming one's holdings involves regaining control and safeguarding one's ownership rights

An investigation into the complicated legal factors that are involved in the process of asset forfeiture, including the inherent tension that develops as a result of the fact that law enforcement organisations are expected to combat criminal groups while simultaneously safeguarding the rights of property owners, The following are some examples of recent advancements in both legal theory and legal practise; however, this list does not contain every recent development in any of these areas. This section presents an analysis of recent legislative revisions and court decisions that have an impact on the process of asset forfeiture and the manner in which it is carried out. These legislative modifications and judicial decisions are relevant because they affect the manner in which asset forfeiture is carried out (Grant, 2021). There have been a lot of legislative changes, and there have also been a lot of judicial rulings, so the manner in which the method is carried out could be altered by those things. Over the course of the previous few years, there have been a wide range of different movements and precedents that have taken place. This will be the primary emphasis of the effort, which will be targeted at improving people's awareness of the many various legislative and legal precedents that have been altered during the previous few years. The work will be focused on boosting people's awareness of the many different legislative and legal precedents that have been amended. The effort will be geared towards educating people about the numerous legislative and legal precedents that have been updated, and that will be the primary focus of the work.

The Justice Administration and Mental Illness Treatment Systems require reform

An analysis of the many different legal considerations that come into play when members of law enforcement interact with people who are suffering from mental health conditions The fundamental objective of this study is to come up with strategies for resolving conflicts that are not only legal but also respectful of the parties involved and secure. This article, which is about the subject matter and is named "Legal Factors in Mental Health Encounters," includes a discussion of the subject matter and provides an examination of the legal concerns that come into play during interactions between law enforcement and those who are struggling with mental health conditions (Harris, 2019). The fact that the study examines these different legal considerations is where the title of the piece derives from. This analysis can be found in the section of the report titled "Legal Factors in Mental Health Encounters.

Police efficiency was improved by reorganising to follow new laws and norms

A look at the latest laws that were passed to encourage police reform and accountability, with a focus on how well these changes work and any legal issues that might come up while they are being put into place. This analysis will look at recent legislative measures that have been enacted with the purpose of promoting police reform and accountability. The research will take a look at recent legislative initiatives that have been implemented with the purpose of improving police reform and accountability. During the course of this research, we will also take into account any potential legal challenges that may arise as a result of the adoption of these revisions (Jackson, 2020). This is something that we will do since we want to make sure that we don't run into any problems down the road. In the course of carrying out this

research, we will also take into consideration any potential legal impediments that may come up in the process of putting these measures into effect. This will be done in order to ensure that we are as prepared as possible.

Suggestions

- Implement faster service provider data access channels for urgent circumstances.
- Establish explicit cross-border data exchange mechanisms that respect state sovereignty and enable law enforcement.
- Invest in ongoing law enforcement technological training.
- Provide enough funds to buy and maintain cutting-edge gear.
- Develop and share digital evidence gathering, preservation, and presentation best practices.
- Provide digital forensic expert certification to maintain high evidence handling standards.

Conclusion

The comprehensive research offered on contemporary legal difficulties faced by law enforcement revealed that police in today's society is complex and continuously changing. This came to attention due to the availability of the test. To achieve success in this attempt, it was imperative to effectively capture the attention of the general populace. The objective of this analysis was to address the necessity of acknowledging this particular truth. This examination was conducted because law enforcement is currently directly addressing these concerns. The study's findings brought the importance of the issue to people's notice and made it a top priority. This happened as a direct result of the study's conclusions. The objective of this study was to Page 130 of 173 examine the primary legal challenges currently faced by law enforcement agencies, their personnel, and the communities they are entrusted to safeguard. Another purpose of this research was to provide a comprehensive analysis of the primary legal challenges faced by individuals hired by such businesses. The purpose of this research was to provide a summary. In addition, this group encompasses both the employees of these institutions and the communities that have entrusted them with the duty of safeguarding them. The relevance of this objective cannot be overstated, as it was the primary motivation behind the research conducted. As our study project nears conclusion, it is becoming more evident that addressing these legal challenges is not only essential but also the responsibility of law enforcement agencies, lawmakers, and the entire community. Both of these groups have the responsibility of tackling these difficulties. We have clearly recognised the importance of this issue. This concept has become evident as we are approaching the completion of the research project that we have been diligently working on. I have been clearly informed that this is a matter that is getting more and more evident as we approach the completion of our task. This is something that I have well understood. It is evident to both of us that we are nearing the completion of the study assignment that we have been given. Collectively, we have mutually reached the determination that this is indeed the situation. As we approach the completion of our research study, it has become increasingly evident to us. We are fully aware and focused on the fact that this specific subject is being questioned.

Reference :

Anderson, J. M. (2020). Compare modern police use of force policies. *Law Enforcement Journal*, 45(2), 78–95.

- Baker, R. L. (2019). Legal review of police accountability and civil rights. *JCR*, 12(4), 321–339.
- Carter, S. A. (2021). Technology and privacy: Balancing security and rights. *Journal of Privacy and Technology*, 14(3), 124–143.
- Davis, M. R. (2020). Legal requirements and public trust in data collection and distribution are discussed. *Journal of Law and Transparency*, 18(1), 56–78.
- Fisher, D. P. (2018). License use of confidential informants: Moral and legal considerations. *Minal Law Review*, 36(4), 411–429.
- Fisher, D. P., Grant, A. K., & Harris, P. J. Legal issues in crisis response and protest management. *PSCRQ*, 25(2), 189–205.
- Grant, A. K. (2021). Legal advances in asset forfeiture and property rights. *Law and Public Policy*, 22(3), 267–283.
- Harris, P. J. (2019). Safe law enforcement–mental health interactions laws and strategies. *Journal of Police and Society*, 16(1), 45–63.
- Jackson, T. R. (2020). Reviewing recent police and legislative improvements. *Law and Justice Review*, 13(4), 410–428.

29. Advanced Ink Forensics for Sustainable Justice: Integrating Chemical Signatures, Physical Dynamics and Digital Authentication

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Abstract: Questioned document examinations in cases of ink as evidence plays a crucial role in maintaining its integrity due to various environmental stressors, forgery techniques and diverse ink formulations. This study presents an advanced, multidisciplinary framework which integrates with chemical profile, physical behavioural analysis and digital authentication to strength the reliability of ink as evidence within the justice system. Techniques such as UV-Vis spectroscopy, Raman Spectroscopy, ATR-FTIR and other chromatographic techniques can characterize pigments, solvent and dyes to obtain their chemical signatures. Physical dynamics such as diffusion, aging patterns, substrate interaction and thermal effects provide environmental and temporal stability of inks. Authentication of digital documents using hyperspectral imaging and signature verification can enhance the detection of overwriting, tampering and alterations. Integration of these three segments of analysis can help holistic and sustainable approach by refining evidence integrity, judicial decision making and ecofriendly forensic practices, also promotes transparency, accuracy and reproducibility of document examination.

Keywords: Ink analysis, analytical techniques, chemical signatures, physical dynamics, digital authentication, sustainable justice, document examination, aging analysis, hyperspectral imaging.

Introduction

Questioned Document Examination constitute of various domains within forensic sciences, which helps for authentication, interpretation and validation of evidences in various criminal and civil court proceedings. There are various components of document examination such as ink evidence analysis, in determining the chronology, alterations, authenticity, authorship, age of documents. Most common cases such as fraud, wills, forgery and disputes and identity related crimes. Despite their apparent simplicity, inks are chemically complex, environmentally sensitive, and increasingly diverse in formulation, making their forensic examination both challenging and indispensable (Islek *et al.* 2020). Reliability of ink as evidence are caused due to various environmental stress, forgery techniques, manipulation and advance forgery techniques. Due to various environmental factors such as light, heat, humidity and pollutants can also alter the ink composition over time. Simultaneously, the global ink market has expanded significantly, introducing a vast array of formulations that often share similar chemical profiles, thereby reducing the discriminatory power of traditional single-technique approaches (Bugler *et al.*, 2008). These challenges can help in the development of advanced, integrated and scientific robust methodologies capable of preserving evidence integrity in supporting sustainable justice systems. Examination of ink

rely on visual interpretation, spectroscopic and chromatographic techniques. These methods have laid foundation on ink analysis and increasingly evident in modern forensic sciences. Analysis often provides sufficient evidential weight, particularly in complex cases which involve multiple inks, similar formulations, aged documents. As forensic science evolves toward higher standards of accuracy, transparency, and reproducibility, the integration of chemical, physical, and digital analytical approaches has emerged as a transformative paradigm (Weyermann *et al.* 2006).

Heterogeneous systems composed of dyes, solvents, binders, resins and additives can contribute to writing performance, stability and appearances. The compositional and behavioural characteristics of inks are examined. Dyes are usually prone to photodegradation, stability but reduced solubility. Solvents influence ink flow and penetration into paper substrates, whereas resins and additives affect drying time and adhesion (Saini K *et al.*, 2018). During manufacturing the batch-to-batch difference, market driven formulation changes can change the compositional complexity. The variability present can indicate significant analytical challenge, when determining ink source, sequence of stroke and its relative age. Moreover, modern inks such as gel pens, hybrid inks, and erasable inks exhibit distinct physical and chemical behaviours that are not adequately addressed by legacy examination techniques (Li B *et al.*, 2014). Exposure to environmental stress can add complexity, fading using light induced techniques, solvent evaporation, oxidation and interaction with paper substrates can significantly alter characteristics of ink over time period. This process may occur naturally or can be accelerated to simulated ageing. Consequently, forensic examiners must differentiate between genuine aging and artificial manipulation, a task that demands both chemical insight and physical understanding of ink dynamics (Weyermann *et al.*, 2020).

Despite being fundamental, traditional ink inspection methods frequently lack the robustness, sensitivity, and specificity needed for today's forensic problems. Examinations that are visual or microscopic are by their very nature subjective and mainly rely on the knowledge of the examiner. Thin layer chromatography, while widely used for ink differentiation, is destructive, limited in resolution and increasingly inadequate for distinguishing modern inks with similar dye compositions (Bugler *et al.*, 2020). Although spectroscopic techniques like Fourier transform infrared spectroscopy (FTIR) and UV visible spectroscopy provide non-destructive alternatives, they are limited by spectral overlap and poor discriminating when inks share common components. Additionally, these techniques often provide bulk information, masking subtle variations critical for forensic interpretation (Bugler J *et al.*, 2008). Relying on isolated or antiquated methods compromises both evidentiary value and judicial confidence as courts increasingly seek replicable and scientifically defensible evidence. Consequently, in order to overcome individual constraints, interdisciplinary and multimodal approaches that combine complementary methodologies are required. Such approaches align with contemporary forensic standards emphasizing validation, uncertainty estimation and transparency.

Emergence of Integrated Ink Forensics

Ink forensics has undergone a paradigm shift with the use of digital verification, physical dynamics, and chemical signature analysis. Integrated frameworks acknowledge ink as a dynamic system whose evidential value arises from the convergence of several analytical domains, as opposed to treating ink examination as a single analytical endeavor. Digital approaches allow for non-destructive visualization and computational validation; physical

investigations clarify behaviour over time and under environmental stress; and chemical procedures disclose compositional fingerprints (Li B et al., 2014). Chemical signature analysis uses sophisticated spectroscopic and chromatographic methods to identify the chemical and elemental components of ink. Complementary insights into dyes, pigments, solvents, and additives are provided by methods like Raman spectroscopy, ATR-FTIR, GC-MS, and HPLC. Combining these techniques improves discrimination and supports studies on aging and source attribution (Nunkoo, M et al., 2016). Ink diffusion, penetration, aging, and substrate contact are the main topics of physical dynamics analysis. For the purpose of determining sequence and evaluating authenticity, these factors offer temporal and contextual information. Crucially, physical behaviour improves forensic relevance by bridging the gap between lab measures and actual document conditions (Weyermann et al., 2020). Non-destructive document analysis has been transformed by digital authentication methods such as multispectral imaging, hyperspectral imaging (HSI), and machine learning-based pattern recognition. These techniques facilitate objective and repeatable analysis while making it possible to identify overwriting, erasures, and minute ink alterations that are imperceptible to the unaided eye (Sun Q et al., 2017).

Digital Transformation and Hyperspectral Imaging in Ink Forensics

The analytical toolkit for ink inspection has been greatly increased by the digital transformation of forensic science. In particular, hyperspectral imaging has become a potent technique that can record spectral and spatial data spanning hundreds of adjacent wavelength bands. In contrast to traditional imaging, HSI produces a spectral signature for every pixel, allowing for precise differentiation between inks that are optically identical (Liu et al., 2006). HSI has shown significant promise in overwriting detection, ink mismatch detection, and modification visualization without sample removal or physical touch. Hyperspectral data can be processed objectively, lowering examiner bias and improving consistency, when paired with chemometric and machine learning algorithms (Wang et al., 2008). Because HSI is non-destructive and allows for repeated analysis while maintaining evidentiary integrity, it is highly compatible with sustainable forensic techniques. By combining behavioural biometrics with tangible evidence, digital signature verification and computational handwriting analysis expand the field of ink forensics. In both conventional and digital contexts, this convergence enhances authorship analysis and facilitates thorough document authentication (Yao et al., 2009).

Sustainability and the Concept of Sustainable Justice

In many scientific fields, including forensic science, sustainability has emerged as a key concept. Delivering precise, transparent and repeatable forensic evidence while reducing environmental effect and conserving resources for further investigations is a key component of sustainable justice. Sustainability in ink forensics is demonstrated by the use of digital workflows, green solvents, non-destructive methods, and micro-sampling techniques (Ni Y et al., 2020). The use of dangerous chemicals and destructive testing in traditional forensic procedures raises ethical and environmental issues. The shift to environmentally friendly practices improves long-term evidence preservation while simultaneously lessening the impact on the environment. Forensic labs can archive, review, and reassess evidence without the need for additional sampling thanks to digital imaging and data-driven analysis, which promotes sustainability and judicial accountability. By optimizing information extraction with minimal interaction, integrated ink forensic systems naturally promote sustainable justice. Examiners can obtain greater evidential value with fewer resources by combining

chemical, physical, and digital tests, which aligns forensic science with global sustainability objectives.

Judicial Expectations and Evidentiary Reliability

Forensic evidence that is transparent, understandable and supported by science is required by contemporary legal systems. The methodological underpinnings of expert evidence are being scrutinized by courts more and more, necessitating precise explanations of analytical procedures, ambiguity, and constraints. By offering multi-layered evidence backed by cross-domain validation, integrated ink forensics meets these requirements (Ezcurra M et al, 2010). In order to convert complex analytical data into conclusions that have legal significance, hierarchical interpretation models are essential. These models improve traceability and judicial comprehension by organizing evidence from raw data through intermediate interpretations to final decisions. The public's confidence in forensic science is strengthened by this clarity, which also increases the admissibility and probative value of ink evidence. The current study suggests a sophisticated, interdisciplinary paradigm for ink forensic analysis that incorporates digital authentication methods, physical dynamics and chemical markers. The framework seeks to strengthen judicial decision making, promote sustainable forensic techniques and improve the integrity of evidence.

Chemical Signature Profiling of Inks

The fundamental analytical cornerstone of contemporary ink forensics is chemical signature profiling, which offers molecular-level data necessary for ink separation, source attribution, and age interpretation. Writing inks are chemically complicated systems made up of resins, plasticizers, stabilizers, surfactants, and dyes or pigments scattered in solvents. A distinctive chemical fingerprint that can be used for forensic comparison is created by each of these elements. Finding and interpreting compositional similarities or differences between questioned and known ink samples in a way that can be supported by science is the main goal of chemical ink profiling (Li B et al., 2014). Because spectroscopic methods are quick, non-destructive, and require little sample preparation, they are the first line of chemical characterization. Chemical signature profiling represents the core analytical pillar of modern ink forensics, providing molecular level information essential for ink differentiation, source attribution, and aging interpretation. Writing inks are chemically complex systems composed of dyes or pigments dispersed in solvents, along with resins, surfactants, plasticizers, and stabilizers. Each of these components contributes to a characteristic chemical fingerprint that can be exploited for forensic comparison. The primary objective of chemical ink profiling is to detect and interpret compositional similarities or differences between questioned and known ink samples in a scientifically defensible manner (Gupta V et al., 2023). Spectroscopic techniques form the first line of chemical characterization due to their rapidity, non-destructive nature, and minimal sample preparation. The UV The absorption behaviour of ink dyes and colorants is frequently examined using visible spectroscopy, which provides useful information on chromophoric structures and permits initial ink separation. However, when employed alone, its discriminating effectiveness is limited by spectrum overlap among chemically related dyes. Because Raman spectroscopy offers molecular fingerprints of dyes and pigments with little interference from paper substrates, it has become more and more important in ink forensics. It is especially appropriate for questioned papers when sample preservation is crucial due to its non-destructive capacity. Raman spectroscopy has been useful in differentiating visually identical inks and identifying pigment-based formulations that are frequently present in contemporary ballpoint and gel inks (Sharma V et al., 2017).

Raman analysis is enhanced by ATR-FTIR spectroscopy, which offers details on the functional groups connected to solvents, binders, and resins. This method works particularly well for distinguishing inks with distinct polymeric components but comparable dye compositions. Since absorption bands may represent penetration and binding behaviour inside cellulose matrices, ATR-FTIR has also been used to analyse ink paper interactions. Despite its benefits, paper composition can affect ATR-FTIR spectra, requiring careful interpretation and, frequently, confirmation with other analytical methods (Ouyang et al., 2019). Chromatographic techniques are still essential for high-resolution ink distinction, even if spectroscopic approaches offer quick and non-destructive insights. Because of its ease of use and capacity to visually separate dye components, thin layer chromatography has long been regarded as a standard technique in forensic ink analysis. However, contemporary ink formulations with fewer dyes or unique compounds are more difficult to resolve with TLC due to its intrinsic destructiveness. Both qualitative and quantitative study of dye components is made possible by the greatly increased separation efficiency and sensitivity provided by high-performance liquid chromatography. HPLC is often employed in forensic laboratories for comparison ink investigations and has been especially successful in distinguishing inks from several manufacturers or manufacturing batches (Ng LK et al., 2002). The examination of volatile organic solvents found in inks depends heavily on gas chromatography–mass spectrometry. Ink flow and drying behaviour are affected by solvents including phenoxyethanol, benzyl alcohol, and glycol ethers, which eventually evaporate. GC-MS examination of these solvents yields useful data for artificial aging detection and ink dating investigations. When paired with suitable reference data, research has demonstrated that solvent loss exhibits predictable temporal trends under controlled conditions, enabling relative age estimation. However, solvent evaporation rates are greatly impacted by storage conditions and environmental variability, which emphasizes the need for cautious interpretation and additional analytical evidence (Bugler et al., 2008).

The significance of multimodal analytical techniques has been highlighted by recent developments in chemical ink profiling. It is impossible to characterize every component of ink with enough forensic certainty using a single method. Rather, the combination of chromatographic and spectroscopic techniques improves discrimination and lowers the probability of coincidental matching. For instance, UV-indistinguishable inks Raman spectral characteristics or chromatographic dye patterns can be used to distinguish visible spectroscopy. These multimodal methods are consistent with modern forensic principles that prioritize repeatability, robustness, and validation (Weyermann C et al., 2007). The possibilities of chemical ink profiling have been further enhanced by the development of mass spectrometry imaging (MSI) techniques. Without requiring a lot of sample preparation, techniques like MALDI-MSI and DESI-MSI allow for spatially resolved chemical analysis directly on documents, allowing for the viewing of ink distribution and chemical heterogeneity. By mapping chemical signals across intersecting strokes, these methods have demonstrated promise in the detection of overwriting, ink mixing, and sequence determination. The potential forensic significance of MSI techniques is becoming more widely acknowledged, despite the fact that they are still primarily limited to research contexts (Brazeau et al., 2007). Sustainability considerations have also influenced the evolution of chemical ink analysis. Traditional solvent intensive methods pose environmental and occupational health concerns. Such approaches are consistent with the broader objectives of sustainable justice, ensuring that forensic methodologies remain both scientifically robust and environmentally responsible (Li B et al., 2014). Nevertheless, chemical profiling must be interpreted cautiously and in conjunction with physical dynamics and digital authentication to achieve reliable, transparent, and legally defensible conclusions.

Chemical, Physical and Digital Techniques in Ink Forensics for Sustainable Justice

In forensic science, sustainable justice focuses on providing reliable, transparent, repeatable, and ecologically conscious evidence to support just judicial decisions. The integration of chemical signature profiling, physical dynamics analysis, and digital authentication procedures is a strategy that connects scientific rigor with sustainability principles in accordance with the study of questioned documents. When combined, these methods reduce damaging activities, resource consumption, and interpretational bias while increasing the dependability of the evidence. Because it allows for the molecular level analysis of ink components like dyes, pigments, solvents, and binders, chemical signature profiling is essential to ink forensics. Complementary insights into ink composition and formulation are provided by methods such as UV visible spectroscopy, Raman spectroscopy, ATR-FTIR, HPLC, and GC-MS. Moreover, chemically robust and well validated results improve judicial confidence, reducing the likelihood of wrongful decisions based on ambiguous or insufficient evidence (Sun Q et al., 2017). Chemical profiling promotes long term evidentiary integrity and environmental responsibility by optimizing information extraction from minimum involvement.

Physical dynamics analysis examines how inks behave over time and in response to environmental factors as light, heat, humidity, and substrate interaction. Diffusion, penetration depth, aging patterns, solvent evaporation, and temporal information are among the parameters that are essential for determining sequence and evaluating authenticity. By allowing relative ink dating and manipulation detection without extensive chemical treatment or intrusive testing, this method promotes sustainable justice. Knowing the differences between artificial and natural aging processes increases the scientific foundation of expert testimony and helps avoid misinterpreting the evidence (Wu Y et al., 2012). Thus, physical dynamics serve as a link between laboratory analysis and actual document conditions, guaranteeing that forensic findings maintain their scientific validity and legal significance.

The most environmentally friendly aspect of contemporary ink forensics is digital authentication methods, especially hyperspectral photography and computational analysis. By capturing precise spectral and spatial information without making physical touch, hyperspectral imaging makes it possible to identify changes that are imperceptible to the human eye, such as overwriting and ink mismatch. Digital methods completely eliminate the need for chemical reagents while providing objective, repeatable, and transparent results when paired with image processing algorithms and machine learning models. Additionally, digital workflows facilitate peer review, long-term data archiving, and reanalysis, strengthening accountability and openness in the legal system (Li B et al., 2014).

When taken as a whole, these three methods combine ethical and environmental responsibility with scientific precision to reflect the concepts of sustainable justice. Digital authentication offers non-destructive, data driven validation, chemical profiling guarantees compositional reliability and physical dynamics offer contextual and temporal insight. Their convergence improves reproducibility, lessens examiner subjectivity and encourages environmentally friendly forensic procedures. Ink forensics is positioned as a model field enabling equitable, transparent and future justice through the coordinated application of various methodologies, as legal systems increasingly demand evidence that is sustainable and supported by science.

Conclusion

Sustainable justice in contemporary judicial systems is the development of ink forensics through the integrated use of chemical signature profiling, physical dynamics analysis and digital authentication. By improving analytical accuracy, reducing subjectivity and maintaining evidentiary integrity, these methods collectively address long standing drawbacks of traditional questioned document analysis. Chemical signature profiling provides a scientifically robust foundation for ink differentiation and characterization, while physical dynamics analysis contextualizes chemical findings by accounting for environmental influences, aging processes and ink substrate interactions. Digital authentication, particularly through non-destructive imaging and computational analysis, further strengthens forensic conclusions by enabling objective detection of alterations and inconsistencies without the use of chemicals or invasive procedures. Together, these domains form a holistic framework that aligns scientific rigor with ethical responsibility. This integrated approach supports environmentally responsible forensic practices by reducing chemical consumption, limiting destructive testing and promoting digital workflows that enhance reproducibility and transparency. Sustainable justice is not solely concerned with environmental considerations but also with ensuring fairness, accountability and reliability in judicial decision making. By delivering scientifically validated, clearly interpretable and reproducible evidence, advanced ink forensics contributes directly to informed legal outcomes and public trust in forensic science. Ultimately, the integration of chemical, physical and digital methodologies positions ink forensics. This multidisciplinary framework reinforces the role of forensic science as an evidentiary integrity while advancing sustainable justice that balances accuracy, and responsibility.

References

1. **Brazeau, L., & Gaudreau, M.** (2007). Ballpoint pen inks: The quantitative analysis of ink solvents on paper by solid-phase microextraction. *Journal of Forensic Sciences*, 52, 209–215.
2. **Bugler, J., & Buchner, H.** (2008). Age determination of ballpoint ink by thermal desorption and gas chromatography–mass spectrometry. *Journal of Forensic Sciences*, 53, 982–988.
3. **Bugler, J. H., et al.** (2008). Age determination of ballpoint pen ink by thermal desorption and gas chromatography–mass spectrometry. *Journal of Forensic Sciences*, 53(4), 982–988.
4. **Ezcurra, M., et al.** (2010). Analytical method for dating modern writing instrument ink on paper. *Forensic Science International*, 197, 1–20.
5. **Gupta, V., et al.** (2023). Studying the methods to determine the age of ink: A critical review of ink dating methods. *Arab Journal of Forensic Sciences and Forensic Medicine*, 5(1), 2–33.
6. **Islek, D. S., & Cengiz, S.** (2020). Ink dating on documents aging due to environmental factors using TD-GC/MS and HPLC-UV. *Forensic Science International*, 29, 8689–8696.
7. **Li, B.** (2014). Dating of black gel pen ink using the dissolution–diffusion method. *Forensic Science International*, 126–131.
8. **Li, B., et al.** (2014a). Dating of inkpad seals using the extraction rate method: Preliminary findings. *Journal of Forensic Sciences*, 59(3), 793–799.
9. **Li, B., et al.** (2014b). GC analysis of black gel pen ink stored under different conditions. *Journal of Forensic Sciences*, 59(2), 543–549.

10. **Li, B., et al.** (2014c). GC analysis of black gel pen ink stored under different conditions. *Journal of Forensic Sciences*, 59, 543–549.
11. **Liu, Y., et al.** (2006). Classification and dating of black gel pen ink by ion-pairing high-performance liquid chromatography. *Journal of Chromatography A*, 1135, 57–64.
12. **Ng, L. K., et al.** (2002). Ballpoint pen inks: Characterization by positive and negative ion electrospray ionization mass spectrometry for forensic examination of writing inks. *Journal of Forensic Sciences*, 47, 200–210.
13. **Ni, Y., et al.** (2020). Study of ink aging: Targeting triethylene glycol in carbon-based black gel ink strokes on paper. *Forensic Science International*, 311, 1–8.
14. **Nunkoo, M. I., et al.** (2016). Forensic analysis of black, blue, red, and green ballpoint pen inks. In *Crystallizing ideas – The role of chemistry* (pp. 323–339). Springer.
15. **Ouyang, G., et al.** (2019). Preliminary studies on the absorbance ratio method used to determine the age of stamp-pad ink seal. *Journal of Forensic Sciences*, 64(4), 1203–1212.
16. **Saini, K., & Rathore, R.** (2018). Differentiation of gel pen inks using high-performance thin-layer chromatography and gas chromatography–mass spectrometry. *Malaysian Journal of Forensic Sciences*, 8, 10–17.
17. **Saini, K., & Rathore, R.** (2018). Differentiation of gel pen inks by high-performance thin-layer chromatography and gas chromatography–mass spectrometry. *Malaysian Journal of Forensic Sciences*, 8, 10–17.
18. **Sharma, V., & Kumar, R.** (2017). Dating of ballpoint pen writing inks via spectroscopic and multiple linear regression analysis: A novel approach. *Microchemical Journal*, 134, 104–113.
19. **Sun, Q., et al.** (2017). Analysis of PEG oligomers in black gel inks: Discrimination and ink dating. *Forensic Science International*, 277, 1–9.
20. **Wang, X., et al.** (2008). Identification and dating of fountain pen ink entries on documents by ion-pairing high-performance liquid chromatography. *Forensic Science International*, 180, 43–49.
21. **Weyermann, C., et al.** (2006). Photofading of ballpoint dyes studied on paper by LDI and MALDI mass spectrometry. *Journal of the American Society for Mass Spectrometry*, 17, 297–306.
22. **Weyermann, C., et al.** (2007). A GC/MS study of the drying of ballpoint pen ink on paper. *Forensic Science International*, 168, 119–127.
23. **Wu, Y., et al.** (2012). Differentiation and dating of gel pen ink entries on paper by laser desorption ionization–quadrupole time-of-flight mass spectrometry. *Dyes and Pigments*, 94, 525–532.
24. **Xu, Y., et al.** (2006). Dating the writing age of black roller and gel inks by gas chromatography and UV–Vis spectrophotometry. *Forensic Science International*, 162, 140–143.
25. **Yao, Y., et al.** (2009). Differentiation and dating of red ink seal entries on documents by HPLC and GC/MS. *Journal of Separation Science*, 32, 2919–292

30. Phytochemical and Toxicological studies on a Plant *Lantana camara* Linn. Poisonous to Livestock

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Abstract: Toxic plants, notably *Lantana camara*, pose significant risks for forensic toxicologists and veterinarians due to their harmful impacts on livestock, leading to fatalities and diminished productivity. *Lantana camara* is recognized as one of the world's top 10 toxic weeds. Field reports of lantana toxicity have come from India and other parts of the world. Because lantana is a very fragrant plant, cattle will only eat it when there is a dearth of other fodder. Toxicosis is caused by lantana-infested animals grazing in pastures. Livestock like cattle, sheep, horses, dogs, and goats are reported to be poisoned by *Lantana camara*. Animals who consume this plant develop hepatotoxicity and subsequent photosensitization. *Lantana camara* contains various secondary metabolites such as alkaloids, saponins, tannins, terpenoids, and flavonoids. Key substances include lantadene A, B, C, and D, triterpenes like alisol A and lantanilic acid, along with essential oil components such as β -caryophyllene and α -humulene. The most hazardous component of the plant is Lantadenes, predominantly located in the leaves. *L. camara* is a flowering ornamental plant belonging to the Verbenaceae family and is also known as Raimuniya in Hindi and Chaturangi in Sanskrit. It can be found in practically every section of India, and its spread throughout the country's forest cover is of great concern because the plant inhibits the growth of other plant species. Jaundice, anorexia, depression, constipation, and severe dermatitis that results in skin sloughing are symptoms of its poisoning in cattle. Skin that is young or lacking in pigmentation is frequently more impacted. This plant's red-flowered forms are frequently mentioned as the most poisonous. Lantana poisoning cases are occasionally documented and sent to toxicologists for analysis. The plant's phytochemical, clinical, and toxicological research has been assessed due to its forensic significance, providing forensic toxicologists, forensic chemists and veterinary surgeons with an updated source. This review will also draw attention to the unsolved problem of its toxicity.

Keywords: *Lantana camara*, Livestock poisoning, Phytoconstituents, Lantadenes, Phytochemical studies, Triterpenoids.

Introduction

Lantana camara is characterized as a 'double-edged sword' due to its multifaceted role in ecosystems. On one hand, it provides benefits such as attracting pollinators, potential medicinal uses including anti-inflammatory and antimicrobial properties, and functioning as a nitrogen-fixing agent that enhances soil quality. Conversely, it poses substantial ecological and economic challenges as a highly invasive species. It aggressively outcompetes native flora, increases fire risks, and presents toxicity to livestock, leading to severe liver problems (Saswade, et al., 2025). Among the various poisonous plants, *Lantana camara* is recognized as one of the most notable noxious and invasive weeds globally, as highlighted by studies (Pereira et al., 2003; Palmer et al., 2000). This particular weed is known to cause significant

mortality among livestock and contributes to the degradation of both agricultural and forest ecosystems (Day et al., 2003; Mello et al., 2005; Sharma et al., 2007). *L. camara* has its origins as an ornamental shrub, having been introduced in 1809 by British botanists at the Calcutta Botanical Garden, and is classified within the Verbenaceae family (Bouda, H., Tapondjou et al., 2001). Lantana is a decorative plant widely found in tropical regions (Sastri, 1962). The term 'Lantana' is derived from the Latin word "*lento*," meaning '*to bend*' (Ghisalberti, 2000). In Sanskrit, it is referred to as "Chaturangi" and "Vanacchedi," as noted in the ancient text Brahmayurved. Locally, this plant is referred to by several names, including bunch berry, baraphulnoo, and red or wild sage (Sharma et al., 2007). Taxonomically, *L. camara* presents classification challenges due to its changing inflorescence characteristics across different ages and seasons (Munir, 1996). Five main Lantana flower variants include pink, white, pink edged, red, and orange. The Lantana genus encompasses several species such as *Lantana trifolia*, *Lantana indica*, *Lantana crenulata*, *Lantana lilacina*, *Lantana involucrata*, and *Lantana Sellowianae*. Notably, the red flower variant, *Lantana camara* var. *aculeata*, is reported to be the most toxic among these variants (Kumar et al., 2016). *Lantana camara* is recognized by various names across different languages in India., it is known as Raimuniya in hindi; Chaturangi and Vanacehdi. in Sanskrit, The Tamil language uses Arippu and Unnichedi, while in Malayalam, it is known as Aripooov, Poochedi, Konginipoo, and Nattachedi. In Manipuri, the names Thirei, Samballei, and Nongballei are used. Bengali refers to it as Putush, whereas in Marathi, it is called Tantani and Ghaneri. In Telugu, it is known as Pulikampa, and in Kannada, the names Kakke and Natahu are used. (Reddy, 2013). Other common names for this plant in English include 'Spanish flag' and 'wild sage' (Al-Snafi, 2019). Chopra et al. noted that ripe fruits are sweet and are consumed by both birds and children (Chopra et al., 1965).

1 Plant Description

Lantana camara is a vigorous shrub characterized by its low erect or subscandent growth, reaching heights between 1 to 3 meters and spreading up to 2.5 meters in width. The plant features triangular stems and ovate leaves that can be acute or subacute, with a crenate serrate margin. The leaves are green notably rugose above and scabrid on both sides, measuring 3 to 8 centimetres in length and 3 to 6 centimetres in width, and are covered with rough hairs, giving them a textured appearance. The flowers, typically small and arranged in clusters, showcase a color palette ranging from orange and white to red, often changing shades as they mature. Each flower presents a yellow throat and blooms in axillary heads nearly year-round. The calyx is small, and the slender corolla tube features a limb that spreads 6 to 7 millimeters wide with unequal lobes. Inflorescences occur in pairs within the axils of opposing leaves, forming a compact, dome-shaped structure 2 to 3 centimeters across, comprising 20 to 40 sessile flowers.

There are several variants based on flower colors such as red, pink, white, and orange. The red flowered type is regarded to be the most dangerous, but other varieties can also be highly toxic (Govindaiah et al., 2021). The root system of *Lantana camara* is robust, enabling the plant to produce new shoots even after experiencing repeated cuttings (Sastri, 1962). Chopra et al. described that ripe fruits are sweet to taste and are eaten by birds and children. Unfavourable seasonal fluctuations have the least impact on the plant's flamboyant growth. The shrub bears fruit until November or December, with flowering beginning in April or May (Sharma et al., 1981).

Fig.1: *L. camara* plantFig.2: *L. camara* fruits

Fig. 3: Orange



Fig. 4: White



Fig. 4: Red

The leaves and immature berries of *Lantana camara* exhibit high toxicity to multiple livestock species, which includes cattle, buffalo, sheep, goats, camels, horses, dogs, guinea pigs, rabbits, ostriches, and rats. Studies indicate that among ruminants, cattle, buffalo, and sheep exhibit the highest susceptibility to *Lantana* toxicity, while goats show a degree of resistance. The toxic effects in grazing animals stem from active compounds known as pentacyclic triterpenoids, specifically Lantadene type-A (rehmannic acid), which are absorbed throughout the gastrointestinal tract, primarily via the small intestine. The toxins can lead to severe health issues, such as hepatotoxicity, photosensitization, and jaundice. Clinical toxicology plays a crucial role in addressing plant poisoning cases, with essential contributions from forensic toxicologists and chemists who assist in identifying the ingested plants and their toxic phytoconstituents. Many databases, including Web Science, Pub Med, Scopus, and Science Direct, were thoroughly examined in order to compile research done in the pertinent subject for this review. This review highlights the urgency of developing an updated document that compiles recent findings to enhance the management of plant poisoning in livestock.

2. Previous Studies

1 Phytochemical studies

Researchers have conducted numerous studies on *Lantana* plants from a variety of perspectives. The natural products found in *lantana* plants were evaluated by Sharma et al. Triterpenoids, flavonoids, oligosaccharides, phenylpropanoid glycosides, iridoide glycosides, and naphthoquinones are the primary natural products identified in *lantana* plants. Most triterpenoids extracted from *L. camara* leaves are pentacyclic, specifically from the oleanane series, while others belong to the lupane and ursane series. Additionally, some triterpenoids possess an oxide bridge spanning from C-3 to C-25.

In another study (Sharma et al., 2007) concerning the toxicity of *Lantana camara*, the authors outline how phyto constituents of this plant play a significant role in its various biological activities. The chemical investigations into lantana toxins, detailed in this study, date back to the early 1940s. Researchers such as Louw, Barton and colleagues, Sastry and Mahadevan, and Hart et al. have made notable contributions to the understanding of these toxins. The paper provides a comprehensive overview of these contributions and discusses the chemistry underlying the toxic effects of *Lantana camara* on animals, enhancing the scientific dialogue surrounding these phytochemical constituents.

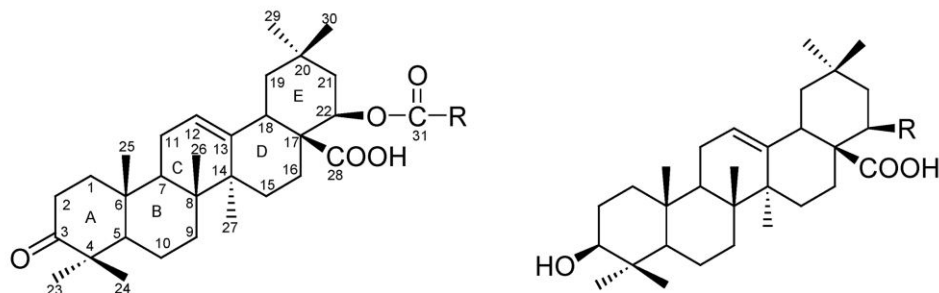
Louw (1943) isolated a crystalline compound named 'lantarin' from the leaves of *Lantana camara*, which exhibited a melting point of 276-280°C. However, since "lantarin" was already attributed to an alkaloid from *Lantana brasiliensis*, the compound from *Lantana camara* was renamed "Lantadene A." In his subsequent research, Louw discovered another crystalline substance during the fractional crystallization process of Lantadene A. This new compound crystallizes from 96% ethanol into firm prisms, in contrast to the delicate needle-like clusters of Lantadene A. The newly identified compound was subsequently designated as "Lantadene B. (Louw, 1943). Barton and colleagues (Barton & Mayo, 1954; Barton et al., 1954; Barton et al, 1956) conducted a series of chemical studies on triterpenoids derived from *Lippia rehmanni* and *Lantana camara*, detailing the chemical structures of rehmannic acid, Lantadene A, and Lantadene B. Their research established that Lantadene A and rehmannic acid are chemically identical. Additionally, they provided structural correlations between Lantadenes A and B (Sharma et al., 1981).

In their comparative analysis of triterpenes within diverse *Lantana camara* taxa, (Barton et al., 1956; Hart et al., 1976) identified notable variations in chemical compositions associated with livestock toxicity. Toxic taxa, such as *Lantana camara*, predominantly feature Lantadenes A and B, while the non-toxic Helidon White taxon is characterized by betulonic acid. The study also emphasized that toxic species contain reduced forms of Lantadene A and B. In contrast, the Townsville Prickly Orange, a non-toxic variant, was found to have lower concentrations of these toxic triterpenes and uniquely includes icterogenin, which can inhibit biliary excretion in rabbits. Another non-toxic type, the Common Pink, lacks Lantadene A and B but contains other triterpenoids with a C3-C25 oxide bridge, such as lantanolic acid, lantic acid, and lantabetulic acid (24). The major toxin, Lantadene A, has been extensively discussed in earlier studies by (Barton et al., 1956; Seawright, 1965) highlighting continued research into the toxicological characteristics of these plant compounds (Hart, 1976).

Begum et al. (2010, 2002) isolated and characterized triterpenes, triterpenoids, pentacyclic triterpenoids from the pentacyclic triterpenoids digfferent parts of *Lantana camara* plant.

Barua et al. (1985) successfully isolated a new triterpene, known as Lantanilic acid, from the leaves of *Lantana camara*. The unequivocal structure of Lantanilic acid has been established as 22 β - β , β -dimethylacryloyloxy-3,25-epoxy- $\sim$ hydroxy olean-12-ene-28-oleic acid (1a). Khan et al. (2025) managed to isolate another triterpene, Lantanilic acid, from the ethyl acetate extract of *L. camara* leaves sourced from Bangladesh. The structural elucidation of Lantanilic acid was achieved through comprehensive analysis using techniques such as ¹H NMR, ¹³C NMR, and mass spectrometry, alongside comparisons with previously published structural data. Sharma et al. (2017) found that triterpenoids from *Lantana camara* leaves are mainly pentacyclic and classified under the oleanane class, with some from the ursane and lupine classes. Notably, certain compounds exhibit an oxide bridge between C-3 and C-25. The primary components identified are Lantadene A, B, C, and D from the redflower variety

of *L. camara* (Sharma et al., 1988; Sharma & Sharma, 1989; Sharma et al., 1990; Sharma et al., 1991). Minor constituents consist of reduced lantadene A (RLA) and reduced lantadene B (RLB).



Lantadene A $R = -C(CH_3)=CH(CH_3)$ Lantadene B $R = -CH=C(CH_3)_2$

Lantadene C $R = -CH(CH_3)CH_2CH_3$ Lantadene D $R = -CH(CH_3)_2$

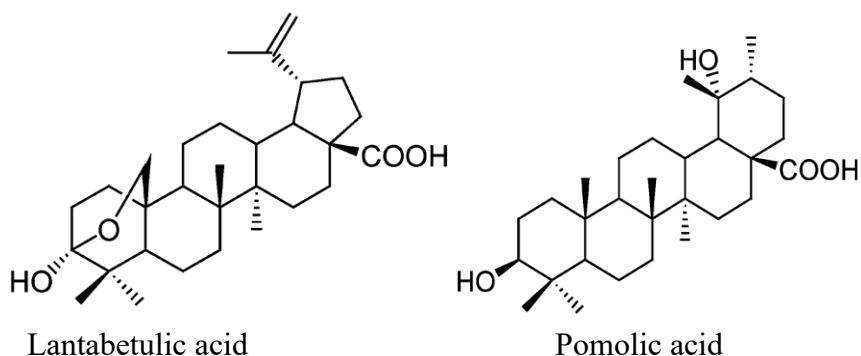
Oleanolic acid $R = H$

Reduced Lantadene A $R = -OCOC(CH_3)=CH(CH_3)$

Reduced Lantadene B $R = -OCO-CH=C(CH_3)_2$

Icterogenin has been found in the leaves and stem of *L. camara* Townsville prickly orange (Hart et al., 1976), but not in the red flower variation (Sharma et al., 1991; Sharma et al., 1997). Oleanonic acid and ursonic acid are the primary ingredients of Townsville prickly orange leaves and stems, while LA and LB are minor constituents. LA and LB are the primary components of the typical pink-edged red flower variation. LA and LB were not discovered in *L. camara* common pink or *L. tiliaefolia* (Seawright, 1965; Johns et al., 1983). Similarly, the taxon common pink has not been found to contain LC, RLA, or icterogenin (Sharma & Sharma, 1989). In New Zealand, where it is most prevalent, this harmless taxon is frequently grazed upon (Black & Carter, 1985). The roots of *L. camara* have a different triterpenoid profile than the leaves. Oleanolic acid is the most abundant component in *L. camara* Helidon white roots, followed by oleanonic acid (Hart et al., 1976).

Barre et al. (1990) isolated and identified a bioactive triterpene from *Lantana camara*, emphasizing its potential use in pharmaceutical applications. Lastly, Huang and Huang (2004) reported the presence of pomolic acid in the stem of the yellow flower variant of *Lantana camara*, highlighting the variability and utility of its constituents.



Lantabetulic acid

Pomolic acid

Kassem et al. (2011) investigated aqueous-methanolic extracts from the flowers of *Lantana camara* L., leading to the isolation and identification of a novel flavonoid compound named apigenin. Alongside apigenin, other noteworthy flavonoids were identified, including luteolin, vitexin, and isovitexin. Patil et al. (2015) reported the discovery of a new compound,

termed 1 (Gautin), derived from the dried leaves of *Lantana camara* L. This compound is characterized as a flavone glycoside, distinguished by its unique aglycone moiety. Their research also identified other flavonoids such as acacetin, Tricin, Hispidulin, and Pectolarigenin. In addition to flavonoids, some other terpenoids were also detected, namely Ursolic acid, Lantanilic acid, Icterogenin, Betulonic acid, and Betulinic acid. The authors also reported the presence of known glycosides, including verbascoside, and an additional flavone glycoside.

Pan et al. (1993) identified Camaroside from the leaves, while Wollenweber et al. (1997) succeeded in isolating 3-methoxy quercetin from leaf exudates. Additionally, Begum et al. (2000) reported the presence of Lantoside in the aerial parts of *L. camara*, specifically a flavone derivative known as 7-O-(6-O-acetyl- β -D-glucopyranosyl)-6,4'-dimethoxy-5-hydroxy flavone.

2 Essential oil

Haikal and Ali (2024) analysed essential oil derived from *Lantana camara* flowers utilizing GC/MS techniques. In a total of twenty-two compounds were identified. The primary constituents include monoterpenes (48.91%) and sesquiterpenes (48.96%), with sabinene (14.90%) and eucalyptol (8.14%) as the leading components.

Suryati et al. (2021) extracted essential oil from fresh *L. camara* leaves through hydrodistillation, utilizing a GC-MS-QP-2010 system with an AOC-20i auto-sampler and a single quadrupole mass spectrometer for analysis. The analysis revealed a total of thirty-eight chemical compounds. Mortada et al. (2017) studied the essential oil of *L. camara* using a Clevenger-type apparatus and GC-MS technique. They identified 47 components in the leaf essential oil and 40 in the flower essential oil, classifying them as monoterpenes, sesquiterpenes, and fatty acids. Chowdhury et al. (2007) analyzed the essential oil obtained from the different parts of *Lantana camara* L.

3 Toxicity of *Lantana camara*

Toxic plants pose significant challenges to veterinarians due to their detrimental effects on livestock, which can lead to increased mortality rates and decreased productivity (Sharma et al., 2007; Bouda et al., 2001; Diaz, 2011). Furthermore, forensic toxicologists face challenges in identifying the plant responsible for plant poisoning. The impact of toxic effects varies greatly among different animal species and is influenced by several factors, including the type of plant part consumed, the quantity of the toxic component ingested, environmental conditions, and the specific characteristics of the animals, such as age, size, and overall health (Sharma et al., 2007). In addition to the harm they cause to livestock, these invasive plant species represent a major threat to biodiversity and ecosystems, ranking closely behind habitat destruction in terms of ecological impact (Drake et al., 1989). Furthermore, these invasive plants can act as predators, leading to the spread of diseases both in animals and in other plant species (Ehrenfeld, 2006; Chambers et al., 2007; Drenovsky et al., 2012). Various efforts have been made by different researchers (Kumar et al., 2016; Sharma & Sharma, 1989) to understand the toxicity of *L. camara* among livestock. These studies aim to elucidate the potential harmful effects this plant may have on animals, which is crucial for both animal health and agricultural practices. The toxicological risks associated with *L. camara* are of particular interest due to its prevalence and the implications for livestock management and care. *Lantana* leaves and immature berries exhibit higher toxicity, affecting various animals

including cattle, buffalo, sheep, goats, camels, horses, dogs, guinea pigs, rabbits, ostriches, and rats. Vulnerable species to this toxicity include sheep, cattle, and buffalo, while goats display relative resistance. Some studies, including those by Sharma et al. (2007), Pass et al. (1981), Parimoo et al. (2015), Kumar et al. (2012), and Nellis (1997), underscore the toxicity of *Lantana camara* among livestock. In a study concerning the toxicity of *Lantana camara*, it was highlighted (Lonare et al., 2012) that West Indian *Lantana* is known for its toxic effects on livestock that consume its foliage due to the presence of pentacyclic triterpenoids called lantadenes. The primary lantadenes identified are A, B, C, and D, with lesser variants such as reduced lantadene A and reduced lantadene B. Among these, lantadene A and lantadene B are notably responsible for inducing hepatotoxicity and photosensitivity in grazing animals like sheep, goats, and cattle (Barceloux, 2012) as well as in horses (Burns, 2001). *Lantana* toxicity in guinea pigs caused a decrease in hepatic mitochondrial protein content. While the phospholipid to protein ratio did not change, the cholesterol to protein ratio and the cholesterol to phospholipid ratio significantly increased. Additionally, various enzyme activities were impacted: succinic dehydrogenase, glutamate dehydrogenase, cytochrome oxidase, and Mg²⁺-ATPase exhibited increased activity, while NADH-ferricyanide reductase activity remained unchanged (Sharma et al., 1982). A few documented instances of *lantana* poisoning are briefly discussed here.

4 *Lantana* poisoning: Case reports

1. A 10-year-old bullock presented with anorexia, extensive skin lesions, and scant feces showed signs of significant dehydration and photosensitization after grazing on *lantana* plants. Clinical examination and sample analysis indicated elevated liver and kidney enzymes, leading to a diagnosis of *Lantana* poisoning (Ambica et al., 2020).
2. In a case of livestock poisoning, a bull from North Guwahati showed signs of intoxication, including diarrhea, weakness, liver damage, and photosensitization after consuming *Lantana camara* leaves. Elevated liver enzyme levels confirmed the poisoning, leading to a diagnosis of *Lantana camara* poisoning and subsequent supportive treatment (Baruti et al., 2018).
3. In a study by Fourie et al., (1987) an outbreak of acute *Lantana camara* poisoning in cattle resulted in 10 deaths among 91 affected animals. Symptoms observed included icterus and soft, black feces. Necropsies revealed liver and kidney changes consistent with the poisoning. Two steers dosed with fresh *L. camara* showed similar pathological changes, and experimental animals exhibited elevated serum urea and creatinine levels.
4. Eight Murrah buffaloes, comprising five adult females and three heifers, exhibited symptoms including jaundice, anorexia, sadness, and lethargy after consuming green foddercontaminated with *Lantana camara* (Hussain, et al., 2013). Hematobiochemical analysis indicated decreased serum total protein, albumin, hemoglobin, and erythrocyte counts, alongside increased bilirubin, AST, ALT, and ALP levels. Additionally, plasma vitamin C levels were significantly lower in the affected buffaloes compared to healthy ones.
5. Choudhary et al. (2022) documented a case of a 6-month-old Jersey cow that displayed clinical symptoms such as anorexia and reduced urination and defecation after consuming *Lantana* foliage for a week. Following the cow's death, necropsy revealed yellowish liver discoloration from bile stagnation, cholecystitis, and gelatinized cardiac fat, with all internal organs showing yellow discoloration. Histopathological analysis demonstrated bile duct hyperplasia, hemosiderin deposition, and extrahepatic bile pigment accumulation linked to necro-hemorrhagic liver lesions. This case illustrates the severe hepatic damage caused by *Lantana camara* in Jersey cows.

Conclusion

Toxic plants pose significant risks to livestock, leading to mortality and decreased productivity, which is a major focus for veterinarians and forensic toxicologists dealing with plant poisoning cases. One such plant is Lantana (*Lantana camara* Linn), a widespread noxious weed in tropical and subtropical regions. Ingestion of its foliage by grazing animals results in cholestasis and hepatotoxicity. The primary toxic components identified in Lantana are triterpenoids, flavonoids, iridoide glycosides, oligosaccharides, phenylpropanoid glycosides, and naphthoquinones, with lantadene A being the main hepatotoxin, a type of pentacyclic triterpenoid. While other congeners of lantadene A contribute minimally to lantana toxicosis, it is the pentacyclic triterpenoids that cause significant liver damage and photosensitivity in affected grazing animals. A crucial aspect for clinicians is to accurately differentiate lantana toxicity from other forms of hepatotoxicity and secondary photosensitization. This distinction is made by examining the biochemical profiles and clinical manifestations of the afflicted animals, especially in connection to the flora found in their grazing area. A diagnosis of lantana poisoning would be supported by tests of plasma bilirubin levels and a clinical history if lantana bushes were found on the pasture. In this situation, forensic toxicologists play a crucial role because their knowledge is crucial in identifying the precise plant species and toxic plant constituents that were responsible for the *Lantana camara* poisoning incidents.

Acknowledgement

One of the authors (PY) expresses gratitude to Dr. Shubhra Goutam, Retired Joint Director, State Forensic Science Laboratory, Raipur C.G. and Dr. Swati Dubey Mishra, Coordinator SVIFS, SVVV, Indore, for their ongoing support and encouragement.

References

- Al-Snafi, A. E. (2019). Chemical constituents and pharmacological activities of *Lantana camara*-A review. *Asian J Pharm Clin Res*, 12(12), 10-20.
- Ambica, G., Reddy, G. A. K., Banotha, A. K., & Kumar, S. R. (2020). Therapeutic management of *Lantana* associated hepatic and renal toxicity in a bullock: a case report. *J. Entomol. Zool. Stud*, 8(2), 1502-1504.
- Barceloux, D. G. (2012). *Medical toxicology of natural substances: foods, fungi, medicinal herbs, plants, and venomous animals*. John Wiley & Sons. pp. 867-868.
- Barre, J. T., Bowden, B. F., Coll, J. C., De Jesus, J., De La Fuente, V. E., Janairo, G. C., & Ragasa, C. Y. (1997). A bioactive triterpene from *Lantana camara*. *Phytochemistry*, 45(2), 321-324.
- Barton, D. H. R., & De Mayo, P. (1954). Triterpenoids. Part XVI. The constitution of rehmamic acid. *Journal of the Chemical Society (Resumed)*, 900-903.
- Barton, D. H. R., De Mayo, P., & Orr, J. C. (1956). 803. Triterpenoids. Part XXIII. The nature of lantadene A. *Journal of the Chemical Society (Resumed)*, 4160-4162.
- Barton, D. H. R., De Mayo, P., Warnhoff, E. W., Jeger, O., & Perold, G. W. (1954). Triterpenoids. Part XIX. The constitution of lantadene B. *Journal of the Chemical Society (Resumed)*, 3689-3692.
- Barua, A. K., Chakrabarti, P., Chowdhury, M. K., Basak, A., Basu, K., Ray, S., & Saha, S. K. (1985). The Structure and Stereochemistry of Lantanilic Acid-A New Triterpene Isolated from *Lantana-Camara*. *Journal of the Indian Chemical Society*, 62(4), 298-305.

- Baruti, M., Singh, B., Bhuyan, M., Borthakur, A., Bhuyan, D., & Chutia, J. P. (2018). Management of Lantana camara poisoning in a bull. *International Journal of Chemical Studies*, 6(1), 950-952.
- Begum, S., Wahab, A., & Siddiqui, B. (2002). Ursethoxy acid, a new triterpene from Lantana camara. *Natural Product Letters*, 16(4), 235-238.
- Begum, S., Wahab, A., & Siddiqui, B. S. (2003). Pentacyclic triterpenoids from the aerial parts of Lantana camara. *Chemical and pharmaceutical bulletin*, 51(2), 134-137. doi: 10.1248/cpb.51.134.
- Begum, S., Wahab, A., Siddiqui, B. S., & Qamar, F. (2000). Nematicidal constituents of the aerial parts of Lantana camara. *Journal of Natural Products*, 63(6), 765-767.
- Begum, S., Zehra, S. Q., & Siddiqui, B. S. (2008). Two New Pentacyclic Triterpenoids from Lantana camara L INN. *Chemical and Pharmaceutical Bulletin*, 56(9), 1317-1320.
- Begum, S., Zehra, S. Q., Ayub, A., & Siddiqui, B. S. (2010). A new 28-noroleanane triterpenoid from the aerial parts of Lantana camara Linn. *Natural Product Research*, 24(13), 1227-1234. DOI: 10.1080/14786410903034082.
- Begum, S., Zehra, S. Q., Hassan, S. I., & Siddiqui, B. S. (2008). Noroleanane triterpenoids from the aerial parts of Lantana camara. *Helvetica Chimica Acta*, 91(3), 460-467.
- Begum, S., Zehra, S. Q., Siddiqui, B. S., Fayyaz, S., & Ramzan, M. (2008). Pentacyclic triterpenoids from the aerial parts of Lantana camara and their nematicidal activity. *Chemistry & biodiversity*, 5(9), 1856-1866.
- Begum, S., Zehra, S. Q., Wahab, A., & Siddiqui, B. S. (2006). Triterpenoidal secondary metabolites from Lantana camara Linn. *Helvetica chimica acta*, 89(9), 1932-1941.
- Black, H., & Carter, R. G. (1985). Lantana poisoning of cattle and sheep in New Zealand. *New Zealand Veterinary Journal*, 33(8), 136-137.
- Bouda, H., Tapondjou, L. A., Fontem, D. A., & Gumedzoe, M. Y. D. (2001). Effect of essential oils from leaves of Ageratum conyzoides, Lantana camara and Chromolaena odorata on the mortality of Sitophilus zeamais (Coleoptera, Curculionidae). *Journal of Stored Products Research*, 37(2), 103-109.
- Burns, D. (Ed.). (2001). *Storey's Horse-lover's Encyclopedia: An English & Western A-to-Z Guide*. Storey Kids. pp. 302.
- Chambers, J. C., Roundy, B. A., Blank, R. R., Meyer, S. E., & Whittaker, A. (2007). What makes Great Basin sagebrush ecosystems invasible by Bromus tectorum? *Ecological Monographs*, 77(1), 117-145.
- Chopra, R. N., Badhwar, R. L., & Ghosh, S. (1965). *Poisonous plants of India*.
- Choudhary, S., Soni, M., Kumar, R., Singh, R., Priyanka, Patil, R. D., & Asrani, R. K. (2022). Pathomorphological Study on Lantana Toxicity in A Jersey Cattle: A Case Report. *International Journal of Livestock Research*, 12(4), 30-33. <https://doi.org/10.5455/ijlr.20220504114128>
- Chowdhury, J. U., Nandi, N. C., & Bhuiyan, M. N. I. (2007). Chemical composition of leaf essential oil of Lantana camara L. from Bangladesh. *Bangladesh Journal of Botany*, 36(2), 193-194.
- Day, M. D., Wiley, C. J., Playford, J., & Zalucki, M. P. (2003). Lantana: current management status and future prospects. *Australian Centre for International Agricultural Research, Canberra*.
- Diaz, G. J. (2011). Toxic plants of veterinary and agricultural interest in Colombia. *International Journal of Poisonous Plant Research*, 1(1), 1-19.
- Drenovsky, R. E., Grewell, B. J., D'antonio, C. M., Funk, J. L., James, J. J., Molinari, N., ... & Richards, C. L. (2012). A functional trait perspective on plant invasion. *Annals of botany*, 110(1), 141-153. doi: 10.1093/aob/mcs100.

- Ehrenfeld, J. G. (2006). A potential novel source of information for screening and monitoring the impact of exotic plants on ecosystems. *Biological invasions*, 8(7), 1511-1521.
- Fourie, N., Van der Lugt, JJ, Newsholme, SJ & Nel, P. W. (1987). Acute Lantana camara toxicity in cattle. *Journal of the South African Veterinary Association*, 58(4), 173-178.
- Ghisalberti, E. L. (2000). Lantana camara L.(verbenaceae). *Fitoterapia*, 71(5), 467-486.
- Govindaiah, K., Biradar, R., Gupta, V. M. D., & Munivenkatappa, B. S. (2021). Lantana toxicity in grazing cattle. *Ind. J. Vet. Sci. Biotech*, 17(01), 85-88.
- Haikal, A., & Ali, A. R. (2024). Chemical composition and toxicity studies on Lantana camara L. flower essential oil and its in silico binding and pharmacokinetics to superoxide dismutase 1 for amyotrophic lateral sclerosis (ALS) therapy. *RSC advances*, 14(33), 24250-24264.
- Hart, N. K., Lamberton, J. A., Sioumis, A. A., Soares, H., & Seawright, A. A. (1976). Triterpenes of toxic and non-toxic taxa of Lantana camara. *Experientia*, 32(4), 412-413.
- Hart, N., Lamberton, J., Sioumis, A., & Soares, H. (1976). New triterpenes of Lantana camara. A comparative study of the constituents of several taxa. *Australian Journal of Chemistry*, 29(3), 655-671.
- Huang, K. F., & Huang, K. W. (2004). Constituents from the stems of Lantana camara (III). *J. Chin. Med*, 15(2), 109-114.
- Hussain, K. H., Mudasir Sultana, M. S., Soodan, J. S., Rajiv Singh, R. S., & Kushwaha, R. B. (2013). Lantana camara ingestion induced photosensitization and dermatitis in buffaloes. *Intas Polivet*, Vol. 14 (II): 215-217.
- J. A. Drake, H. A. Mooney, F. di Castri, R. H. Groves, F. J. Kruger, M. Rejmanek, and M. Williamson (1989). Biological Invasions: A Global Perspective. *John Wiley and Sons*, New York.
- Johns, S. R., Lamberton, J. A., Morton, T. C., Soares, H., & Willing, R. I. (1983). Triterpenes of Lantana tiliaefolia. 24-Hydroxy-3-oxo-12-en-28-oic acid, a new triterpene. *Australian Journal of Chemistry*, 36(12), 2537-2547.
- Kassem, L. T., Mohammed, R. S., El Din, S. S., El Ansari, A., Hawas, U. W., & Mahmoud, K. (2011). Flavonoids from the flowers of Lantana camara L. with in vitro antioxidant activity. *Planta Medica*, 77(12), PG7.
- Khan, A., Kuddus, M. R., Rashid, M. A., Hossain, M. S., & Uddin, M. A. (2025). Isolation and characterization of lantanillic acid from lantana camara l. In Bangladesh. *Bangladesh Journal of Botany*, 54(3), 465-468.
- Kumar, R., Katiyar, R., Kumar, S., Kumar, T., & Singh, V. (2016). Lantana camara: An alien weed, its impact on animal health and strategies to control. *Journal of Experimental Biology*, 4, 3S.
- Lonare, M. K., Sharma, M., Hajare, S. W., & Borekar, V. I. (2012). Lantana camara: overview on toxic to potent medicinal properties. *International Journal of Pharmaceutical Sciences and Research*, 3(9), 3031.
- Louw, P. G. J. (1943). Lantanin, the active principle of Lantana camara L. Part 1. Isolation and preliminary results on the determination of its constitution. *Onderstepoort J. Vet. Sci. Anim. Ind.*, 18, 197-202.
- Louw, P. G. J. (1948). Lantadene A, the active principle of Lantana camara L. Part II. Isolation of lantadene B, and the oxygen functions of lantadene A and lantadene B. *Onderstepoort J. Vet. Sci. Anim. Ind.*, 23, 233-238.
- Mello, F. B., Jacobus, D., Carvalho, K., & Mello, J. R. (2005). Effects of Lantana camara (Verbenaceae) on general reproductive performance and teratology in rats. *Toxicon*, 45(4), 459-466.
- Misra, L., & Laatsch, H. (2000). Triterpenoids, essential oil and photo-oxidative 28→13-lactonization of oleanolic acid from Lantana camara. *Phytochemistry*, 54(8), 969-974.

- Mortada El-Sayed, M., Hashash, M. M., Abdel-Hady, A. A., Abdel-Hady, H., Abdel-Lateef, E. E., & Morsi, E. A. (2017). Total phenolic and flavonoid contents and antioxidant activity of *Lantana camara* and *Cucurbita pepo* (Squash) extracts as well as GC-MS analysis of *Lantana camara* essential oils. *World J Pharm Res*, 6(1), 137-153.
- Munir, A. A. (1996). A taxonomic review of *Lantana camara* L. and *L. montevidensis* (Spreng.) Briq. (Verbenaceae) in Australia. *Journal of the Adelaide Botanic Garden*, 1-27.
- Nellis, D. W. (1997). *Poisonous plants and animals of Florida and the Caribbean*. Pineapple Press Inc.
- of India, Vol. 11, ICAR, New Delhi, 698–699.
- Ono, M., Hashimoto, A., Miyajima, M., Sakata, A., Furusawa, C., Shimode, M., ... & Nohara, T. (2021). Two new triterpenoids from the leaves and stems of *Lantana camara*. *Natural Product Research*, 35(21), 3757-3765. doi: 10.1080/14786419.2020.1736063.
- Palmer, W. A., Willson, B. W., & Pullen, K. R. (2000). Introduction, rearing, and host range of *Aerenicopsis championi* Bates (Coleoptera: Cerambycidae) for the biological control of *Lantana camara* L. in Australia. *Biological Control*, 17(3), 227-233.
- Pan, W. D., Li, Y. J., Mai, L. T., Ohtani, K. H., Kasai, R. T., Tanaka, O., & Yu, D. Q. (1993). Studies on triterpenoid constituents of the roots of *Lantana camara*. *Yao xue xue bao= Acta Pharmaceutica Sinica*, 28(1), 40-44.
- Pan, W. D., Li, Y. J., Mai, L. T., Ohtani, K. H., Kasai, R. T., Tanaka, O., & Yu, D. Q. (1993). Studies on triterpenoid constituents of the roots of *Lantana camara*. *Yao xue xue bao= Acta Pharmaceutica Sinica*, 28(1), 40-44.
- Parimoo, H. A., Sharma, R., Patil, R. D., & Patial, V. (2015). Sub-acute toxicity of lantadenes isolated from *Lantana camara* leaves in Guinea pig animal model. *Comparative Clinical Pathology*, 24(6), 1541-1552.
- Pass, M. A., McSweeney, C. S., & Reynoldson, J. A. (1981). Absorption of the toxins of *Lantana camara* L. from the digestive system of sheep. *Journal of Applied Toxicology*, 1(1), 38-41.
- Patil, G., Khare, A. B., Huang, K. F., & Lin, F. M. (2015). Bioactive chemical constituents from the leaves of *Lantana Camara* L. *Indian Journal of Chemistry B*, 54, 691-697.
- Pereira, J. M., Barreto, R. W., Ellison, C. A., & Maffia, L. A. (2003). *Corynespora cassiicola* f. sp. *lantanae*: a potential biocontrol agent from Brazil for *Lantana camara*. *Biological Control*, 26(1), 21-31.
- Reddy, N. M. (2013). *Lantana camara* Linn. chemical constituents and medicinal properties: a review. *Sch. Acad. J. Pharm*, 2(6), 445-448.
- Sastri, B. N. (1962). The wealth of India, vol. 6. *Council of Scientific and Industrial Research, New Delhi*, 382, 383.
- Saswade, K., Bhand, R., Waghmare, S. (2025). The Double- Edged Sword of *Lantana Camara*: Toxicological Risk and Therapeutic Potentia, *International Journal of Research Publication and Reviews*, 6(10); 2439-2443.
- Seawright, A. A. (1965). Toxicity for the guinea pig of an extract of *Lantana camara*. *J. Comp. Pathol.*, 75, 215-221.
- Sharma, O. P., & Sharma, P. D. (1989). Natural products of the lantana plant-the present and prospects. *J. Sci. Ind. Res.* 48:471-478.
- Sharma, O. P., Dawra, R. K., & Ramesh, D. (1990). A triterpenoid acid, lantadene D from *Lantana camara* var. *aculeata*. *Phytochemistry*, 29(12), 3961-3962.
- Sharma, O. P., Makkar, H. P. S., & Dawra, R. K. (1982). Biochemical effects of the plant *Lantana camara* on guinea pig liver mitochondria. *Toxicon*, 20(4), 783-786.
- Sharma, O. P., Makkar, H. P. S., & Dawra, R. K. (1988). A review of the noxious plant *Lantana camara*. *Toxicon*, 26(11), 975-987.

- Sharma, O. P., Makkar, H. P. S., Dawra, R. K., & Negi, S. S. (1981). A review of the toxicity of *Lantana camara* (Linn) in animals. *Clinical toxicology*, 18(9), 1077-1094.
- Sharma, O. P., Sharma, S., & Dawra, R. K. (1997). Reversed-phase high-performance liquid chromatographic separation and quantification of lantadenes using isocratic systems. *Journal of Chromatography A*, 786(1), 181-184.
- Sharma, O. P., Sharma, S., Patabhi, V., Mahato, S. B., & Sharma, P. D. (2007). A review of the hepatotoxic plant *Lantana camara*. *Critical reviews in toxicology*, 37(4), 313-352.
- Sharma, O. P., Vaid, J., & Sharma, P. D. (1991). Comparison of lantadenes content and toxicity of different taxa of the lantana plant. *Journal of chemical ecology*, 17(11), 2283-2291.
- Siddiqui, B. S., Raza, S. M., Begum, S., Siddiqui, S., & Firdous, S. (1995). Pentacyclic triterpenoids from *Lantana camara*. *Phytochemistry*, 38(3), 681-685.
- Singh, S. K., Tripathi, V. J., & Singh, R. H. (1990). 3 β , 24-Dihydroxyolean-12-en-28-oic acid, a pentacyclic triterpene acid from *Lantana indica*. *Phytochemistry*, 29(10), 3360-3362.
- Singh, S. K., Tripathi, V. J., & Singh, R. H. (1991). A new pentacyclic triterpene acid from *Lantana indica*. *Journal of Natural products*, 54(3), 755-758.
- Singh, S.K., Singh, A., Tripathi, V.J., & Finzi, P.V. (1996). Minor constituents of *Lantana camara*. *J. Ind. Chem. Soc.* 73:547–548.
- striterpenoids from *Lantana* species. *Indian Drugs*, 34(7), 390-392.
- Suryati, S., Aziz, E. D., Efdi, M., Wahyuni, F. S., & Hefni, D. (2021). Analysis of the essential oil from *Lantana camara* leaves and its cytotoxic potential against T-47D breast cancer cells. *Jurnal Riset Kimia*, 12(1), 1-9.
- Verma, D. K., Singh, S. K., Nath, G., & Tripathi, V. (1997). Antimicrobial active
- Wollenweber, E., Dörr, M., Muniappan, R., & Siems, K. (1997). Flavonoid aglycones and triterpenoids from the leaf exudate of *Lantana camara* and *Lantana montevidensis*. *Biochem. Syst. Ecol.* 25:269–270.

31. Environmental Forensic Assessment of Human-Source Pathways of Pharmaceuticals and Personal Care Products

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Abstract: Pharmaceuticals and Personal Care Products (PPCPs) are one of the major classes of emerging contaminants increasingly being detected in environmental matrices due to routine human activities. These include commonly consumed medicines, cosmetics, and hygiene products and are mainly routed into wastewater systems through daily use and improper disposal. Understanding the behavioural pathways responsible for this release is essential for environmental forensic investigations. A 25-item questionnaire was used to collect primary data on medication usage, self-medication practices, handling of leftover drugs, frequency of cosmetic use, and awareness of the environmental impact of PPCPs across diverse demographic groups. The results indicated a high rate of use for most of the frequently consumed pharmaceuticals like painkillers, antibiotics, antihistamines, and over the counter products. Similarly, personal care brands such as Himalaya, Dove, Dot & Key, Colgate, Lakme, and L'Oréal were found in common use. Despite high levels of use, there was a lack of awareness among respondents concerning proper medicine disposal, persistence in the environment, and ecological risks related to PPCPs. These practices reflect substantial probabilities of PPCP releases into household wastes and sewage. This approach of survey-based environmental forensics is generally rapid, providing very useful insights by connecting human activities to potential contamination pathways for future monitoring and mitigation.

Keywords: Environmental Forensics, Pharmaceuticals and Personal care products (PPCPs), Emerging contaminants, Human-source pathways

Introduction

Pharmaceuticals and Personal Care Products (PPCPs) are a class of ‘emerging contaminants’, which comprise numerous prescriptions and over the counter medicines, illicit drugs, nutritional supplements, diagnostic agents, shampoos, soaps and fragrances (Lubliner B, 2010). The concern surrounding PPCPs arises from their ability to persist, transform, and accumulate in different environmental compartments such as surface water, groundwater, soil, and sediments. The problem of pharmaceuticals and personal care products (PPCPs) in the water environment has gained increasing interest worldwide in the past two decades (Kinga Slosarczyk, 2021). Accumulation of PPCPs in surface waters leads to lifelong adverse impacts on aquatic life, such as changes in growth rate, behaviour, reproduction, and modifications at the biochemical level in primary producers up to secondary consumers (Charles Obinwanne Okoye, 2022).

PPCPs widespread and continuous use leads to an unintended release of biologically active chemicals into environmental systems. Since PPCPs cannot be fully metabolized by the human body, a portion is excreted unchanged and enters wastewater streams through household sewage and greywater. The removal of these products from wastewater is challenging due to their diverse structures and physical properties. They include all chemicals and drugs like antibiotics, hormones, anti-inflammatory drugs, lipid regulators, steroids, and antiepileptics (Sofian Kanan, Occurrence, analysis and removal of pesticides, hormones, pharmaceuticals, and other contaminants in soil and water streams for the past two decades: a review, 2022). Unlike many other pollutants to the aquatic environment, antibiotics have a direct biological action on microbes, and in many cases are continually emitted to the environment thereby acting as persistent pollutants even if some have high degradation rates (Najat Al-Odaini, Detecting Human Pharmaceutical Pollutants in Malaysian Aquatic Environment: A new challenge for water quality management, 2011) This persistent and routine introduction of chemicals has placed PPCPs among the most significant emerging pollutants in modern environmental studies.

From the environmental forensic viewpoint, it is important to identify the sources and behavioural pathways responsible for the PPCP contamination. Unlike industrial pollutants, PPCPs are derived mainly from distributed human activities such as daily use and improper disposal of medicines, flushing of expired drugs and over-reliance on self-medication. Mapping these pathways enables researchers to trace contamination trends, understand chemical entry routes, and propose targeted interventions.

This Study aims to identify human practices that result in PPCPs reaching the environment through a scrutiny of use, storage, and disposal methods and by assessing the public understanding of their effects on the environment. Interactions between PPCP and drinking water disinfectants in water treatment plants are also a potential concern (Leslie Cizmas, Pharmaceuticals and personal care products in waters: occurrence, toxicity, and risk, 2015). So that there is growing concern over PPCPs as a class of emerging pollutants continuously found in wastewater and household refuse resulting from daily human activities and poor disposal methods. Generating behavioural data provides essential environmental forensic insights into PPCP contamination pathways.

Materials and Methods

This study used a cross-sectional descriptive design with a structured, self-administered online questionnaire. The purpose was to determine public awareness, usage behaviour, disposal practices, and perceived environmental and health impacts associated with pharmaceuticals and personal care products. In the survey format, information can be gathered quickly from a diverse population, and responses can remain anonymous. Questions were intentionally framed to elucidate patterns of everyday PPCP use and to understand how these products may contribute to environmental contamination.

1 Study Area and Population

The survey included participants from different regions across India, particularly Kerala, Indore, Gujarat, Chennai, and several semi-urban areas, along with a few international respondents. In all, 100 individuals participated in the study, representing different age groups that ranged from below 18 years to above 45 years. The dataset contained responses by both males and females. Participants belonged to different occupation categories such as students, working professionals, homemakers and self-employed individuals. This

demographic variety ensured a broader representation of behaviours and perceptions within the general population.

2 Sampling

Data collection applied a non-probability convenience sampling method. The link to the Google Form was digitally distributed through social medias. Individuals who voluntarily clicked the link and completed the survey formed the study population. These participants were above 15 years. Responses with mandatory fields or duplications were excluded.

3.Data Collection

Data collection was done through a structured online questionnaire prepared using Google Forms. The 25 items in the questionnaire were divided into sections on demographic details, personal care product usage, pharmaceutical consumption, disposal practices, environmental awareness, and health effects. The demographic section included basic information about the respondents, such as age, gender, occupation, and place of residence. The personal care section explored the types of products used regularly, like face wash, shampoo, sunscreen, cosmetics, and how frequently they were used.

The pharmaceutical section listed the types of medicines used in the last six months, including common OTC (Over the Counter Dose) drugs, antibiotics, gastric medicines, antihistamines, and pain relievers. It also analyzed whether the respondent's purchased antibiotics without a prescription and how they dispose of unused or expired medicines. Disposal-related questions looked at whether medicines were thrown in household waste, flushed, burnt, or returned to medical stores. The environmental awareness section measured how knowledgeable respondents were regarding how PPCPs contaminate water bodies, soil, and ecosystems, besides their awareness about the take-back program. Finally, the health effects section collected information on any side effects after using PPCPs, such as skin irritation, hair fall, burning sensations, or allergic reactions.

This survey research was performed according to ethical standards on online surveys. Participation was voluntary, and informed consent was digitally obtained before respondents continued to the questionnaire. No personal or sensitive identifying information was collected at any point. All collected data were used exclusively for academic and research purposes and maintained as confidential.

4 Data Analysis

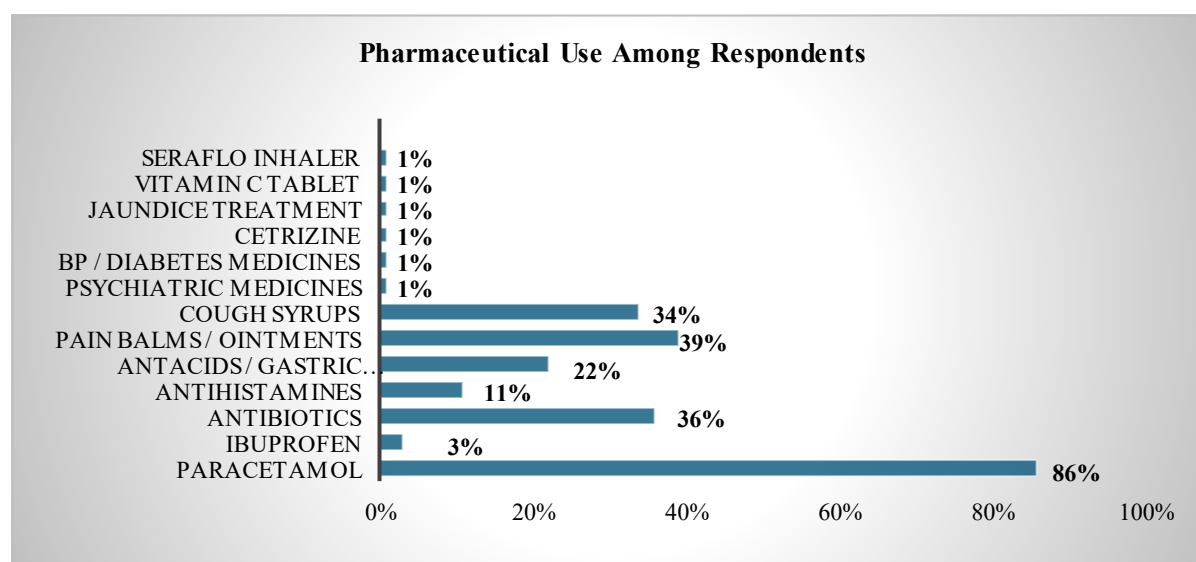
All responses were automatically recorded through Google Forms and later exported into Microsoft Excel for data cleaning and analysis. The dataset was screened for incomplete or inconsistent entries before analysis. Frequencies and percentages were calculated to summarize the distribution of responses. Graphs and charts generated through Google Forms enabled the visual interpretation of the patterns of PPCP use, medicine disposal behaviour, environmental awareness, and perceived health impacts. The nature of this investigation being exploratory and descriptive means that analysis focused on the identification of observable trends; thus, no tests of statistical inference were considered.

Results and Discussion

Results are based on responses from participants in several states of India, along with a few international responses, indicating a good overview of daily personal-care product usage and the extent of awareness among the public about their chemical constituents. In this survey study, the most used brands by 100 participants were Himalaya, Dove, Colgate, Nivea, Loreal Paris, Dot & Key, Minimalist, Mama earth, Garnier, Vaseline, Beardo, Cetaphil, Demarco, Foxtale, Ponds, Plum, and Mcaffeine. Screening ingredient lists for these products indicated the frequent use of sodium lauryl sulphate, sodium Laureth sulphate, Cocamidopropyl betaine, silicones, mineral oils, parabens, phenoxyethanol, carbomers, acrylate polymers, BHT, EDTA, titanium dioxide, zinc oxide and a host of synthetic fragrance allergens. These chemicals form the basis for a wide range of personal care preparation based on face wash, shampoo, moisturizers, lotions, serum, soaps and cosmetics. These recurring chemical groups are characteristic of PPCP associated pollutants and are well-recognized for their persistence in the environment, very limited biodegradability and potential to bioaccumulate in aquatic and terrestrial ecosystems. Their extensive use daily by consumers suggests a continuous discharge of these residues into household wastewaters, reaching sewage treatments, rivers and soil environments. Such contaminants may give rise to disruptions in microbial communities, endocrine-related effects in aquatic organisms and long-term ecological imbalance.

From an environmental forensic point of view, these patterns emphasize the urgent need for effective pollutant surveillance, regulatory monitoring and systematic wastewater analysis to reconstruct contamination pathways, quantify source contributions and determine the environmental fate and transport of the chemicals concerned. The enhancement of such measures will underpin better risk assessment, inform public health policies and contribute to the improved management of emerging contaminants in the environment.

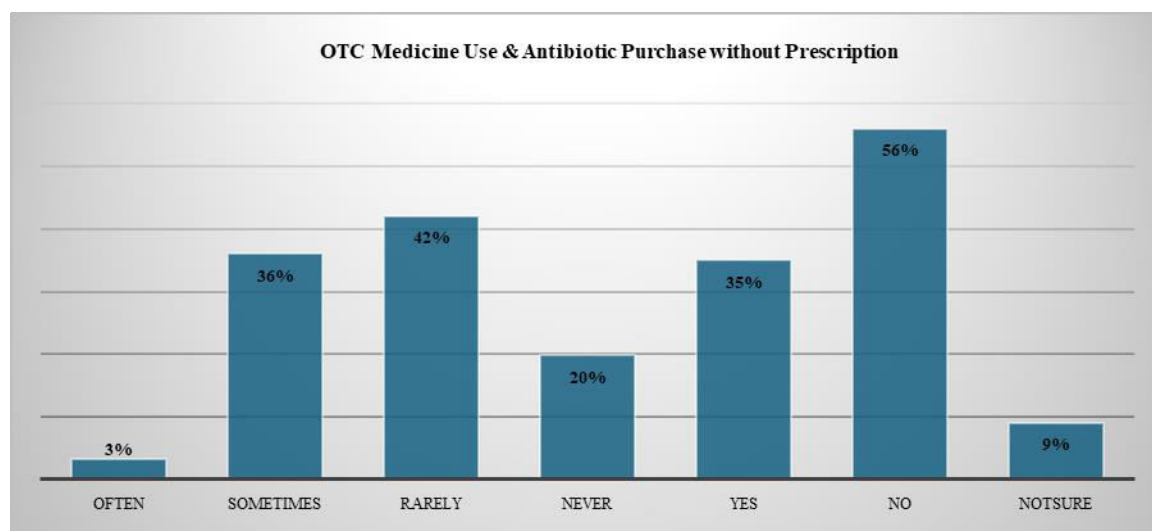
3.1 Pharmaceutical Use



Graph 1 Pharmaceutical Use Among Respondents

Graph 1 reflects the sources of pharmaceuticals normally used by the respondents. The most frequently used medicine is paracetamol, was taken up by 86% of the participants. 36% responding that they are taking Antibiotics and 39% using pain balms/ointments. While 34% reported using cough syrups. 22% & 11% respondents using Antacids/Gastric medicines and antihistamines respectively. On the other hand, only a few of them use more specialized medicines like psychiatric medicines, BP/diabetes medicines, cetirizine, jaundice treatments,

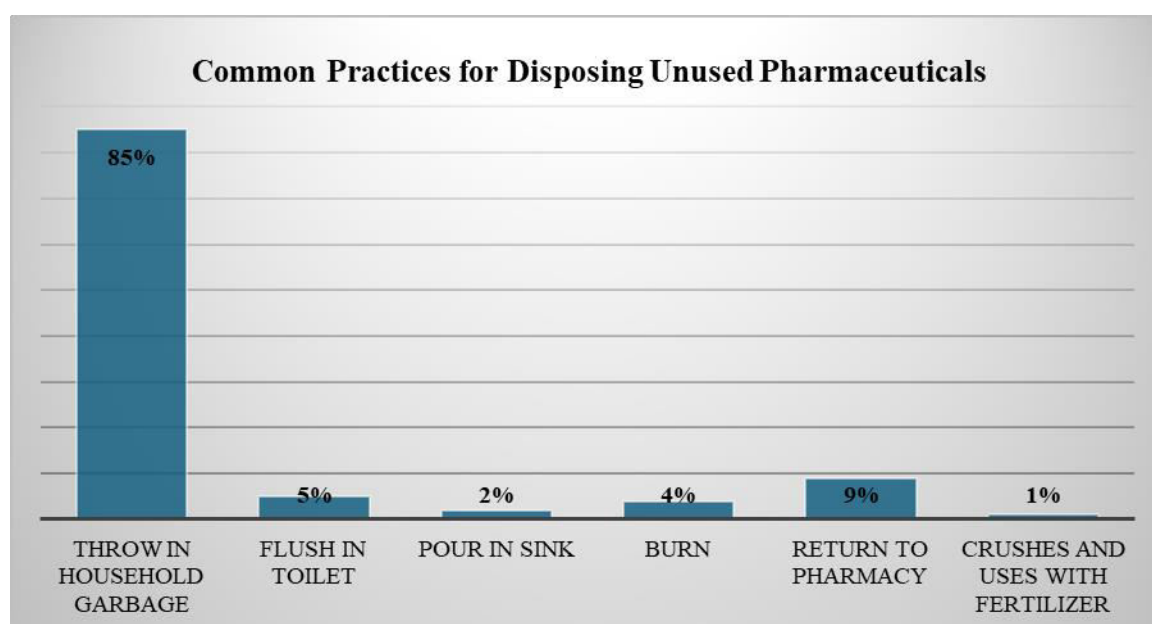
vitamin C tablets, or inhalers, amounting to 1% each. In summary, it shows a high dependence on some common general use medicines with a very minimal percentage using any specialized medicines.



Graph 2 OTC Medicine Use & Antibiotic Purchase without Prescription

Graph 2 illustrates respondents' behaviour in terms of the use of non-prescribed medicines. 42% of respondents rarely take over-the-counter medicines without a prescription and 36% sometimes take over-the-counter medicines without a prescription, when 3% do so often. Regarding antibiotics, 56% of the total reported never purchasing antibiotics without a prescription, while 35% responded that they did. While 9% were not sure. Overall, the graph indicates that non-prescribed, over-the-counter self-medication is common, but that non-prescribed antibiotic purchase is still happening among a notable share of respondents.

2 Storage and Disposal Practices

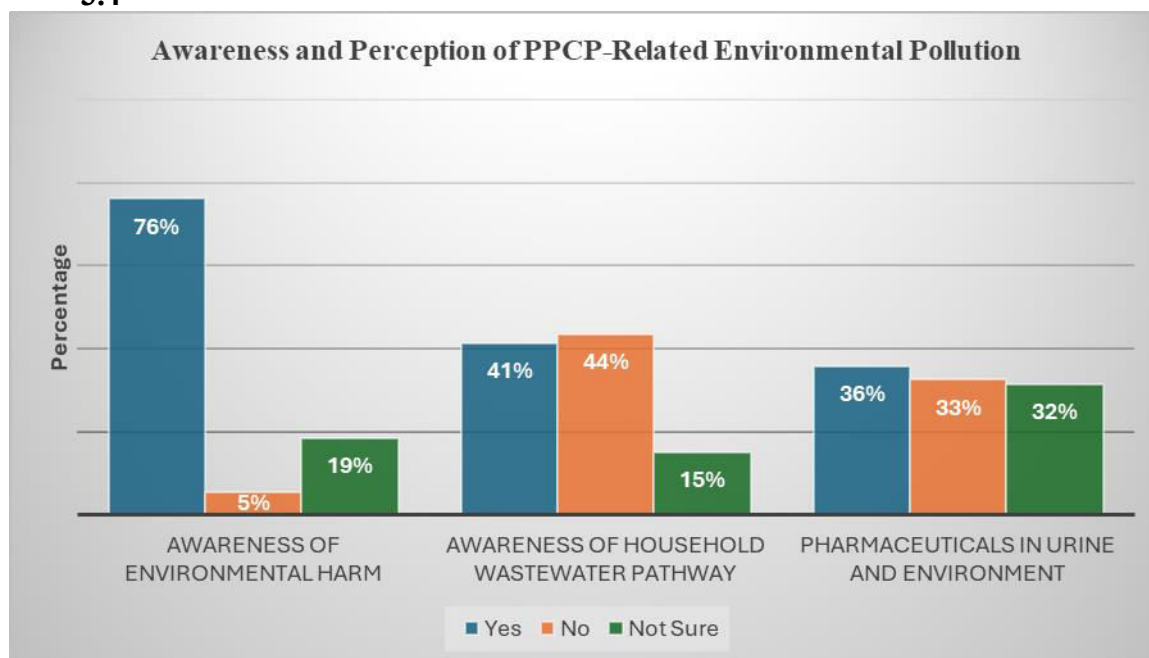


Graph 3 Common Practices for Disposing Unused Pharmaceuticals

Graph 3 is showing improper handling of unused and expired medicines is reflected in most responses. 89% reported, they did not store leftover medicines at home. 85% responded that throwing medicines into household garbage and dustbins was the disposal method, and this may release active medicines into the soil and landfill leachates. Environmentally very damaging practices that were also identified, 5% of respondents flushing of medicines down toilets, 2% pouring syrups into sinks, 4% burning tablets, and 1% mixing crushed medicines with fertiliser. 9% respondents returned medicines to a pharmacy, indicating limited behaviour adoption regarding responsible disposal.

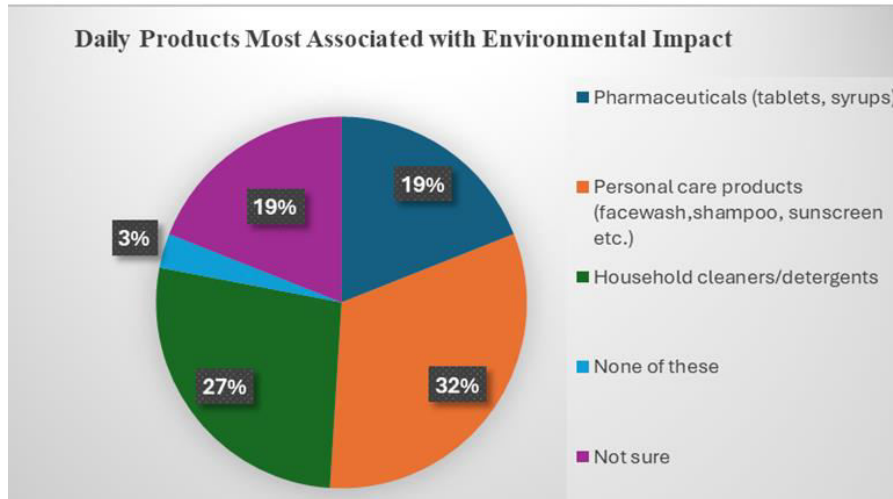
3.3 Environmental Awareness And Perception

3.4



Graph 4 Awareness and Perception of PPCP-Related Environmental Pollution

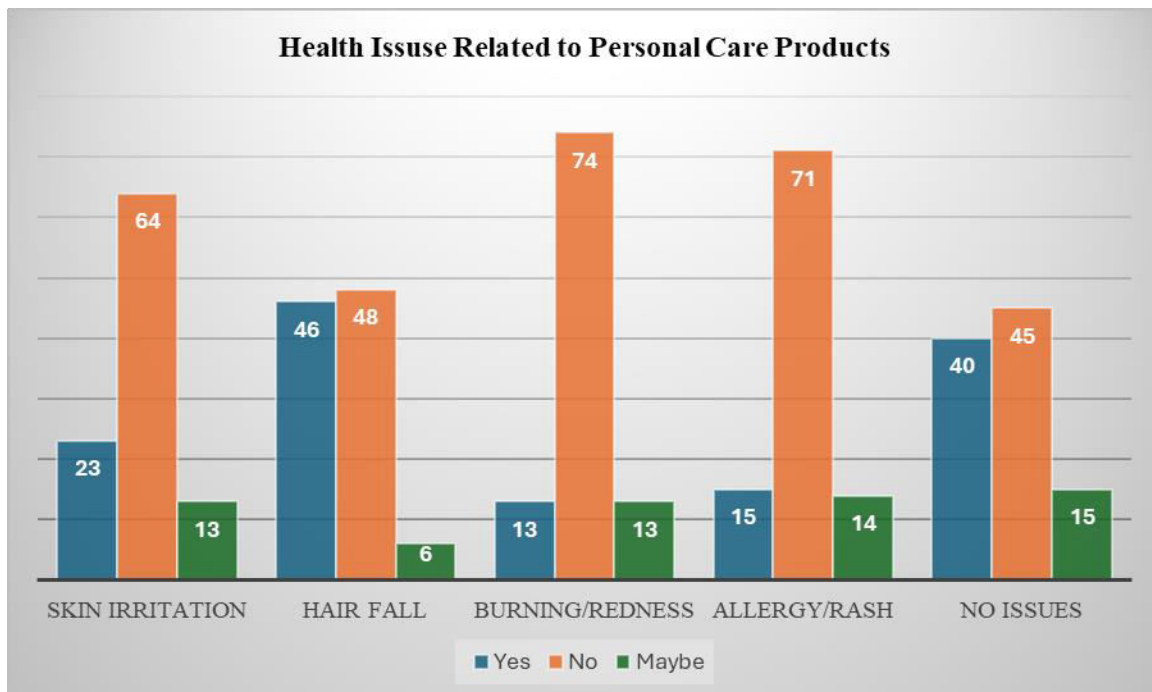
Graph 4 displays responses to questions about the awareness of PPCP-related pollution. For environmental harm, 74% responded with Yes, 5% with No, and 21% were Not sure. For household wastewater pathways, 40% responses have knowledge, while 9% have not and 51% were Not sure. For pharmaceuticals entering the environment through urine, 35% said they knows, 12% have not and 53% were Not sure. In general, the results show high awareness of the general PPCP-related harm but a limited understanding of specific pollution pathways.



Graph 5 Daily Products Most Associated with Environmental Impact

Graph 5 reflects the choices of the respondents regarding which daily-use products they think contribute most to environmental impact. Personal care products like facewash, shampoo, and sunscreen-were selected as major contributors by 32% of respondents. This was followed by household cleaners/detergents at 27%, and pharmaceuticals tablets and syrups at 19%. A smaller share, 3%, think that none of these are major contributors, while 19% respondents are not sure about this. Overall, personal care products are perceived as the most environmentally impacting among daily use items.

4 Health Monitoring



Graph 6 Health Issue Related to Personal Care Products

This graph 6 reflects responses on health issues related to personal care product use. For skin irritation, 23 said they have issues, while 64 said No, and 13 said Maybe. For hair fall, 46 responses have, while 48 responses have not and 6 said Maybe. Burning/redness was reported by 13, while 74 said No and 13 were Maybe. About allergy/rash, 15 respondents said Yes, 71

said No, and 14 said Maybe. About no issues, 40 respondents confirmed Yes, 45 responses have no issues and 15 were Maybe. Overall, most of the respondents said no major health effects, although hair fall and skin irritation were comparatively more prevalent.

Conclusion

This study clearly outlines an environmental forensic understanding of how everyday human behaviour leads to the continuous release of PPCPs to environmental systems. In fact, the dependency includes routine medicines of paracetamol, antibiotics, cough syrups, antacids and antihistamines. Among others and personal care products that find daily use and are being widely marketed. Ingredient profiling of frequently used brands identified the presence of persistent and potentially hazardous chemicals such as surfactants, parabens, silicones, preservatives, fragrance compounds and chelating agents in them. These chemicals, resulting from their poor biodegradability and bioaccumulative nature, may persist in wastewater, surface water, soil and sediments, thereby posing long-term ecological risks and creating sustained contamination pressure on aquatic and terrestrial ecosystems.

This study further highlights lack of the knowledge in the storage and disposal practices of pharmaceuticals. A large proportion of the respondents dispose of unused and expired medicines through household garbage, while some others flush them into toilets, pour liquid formulations into sinks, burn tablets, or mix them with fertilizers. Such unsafe practices allow multiple direct pathways for active pharmaceutical ingredients to enter the landfill leachates, sewage systems, agricultural soils and water bodies. Although general awareness regarding environmental pollution due to PPCPs is good, in-depth understanding among the public regarding the pathways of specific contamination-particularly urinary excretion and transport via wastewater-remains limited. This divide between awareness and responsible behaviour at disposal is a big challenge for effective pollution control.

This study establishes forensic linkage between regular consumer behaviour and PPCP contamination pathways in the environment. The survey-based environmental forensic approach is proven to be a rapid, cost-effective and reliable tool in identifying human-source pollution trends. Results strongly underscore the imperative need to enhance public education on safe medicine disposal, extend pharmaceutical take-back programs, improve regulatory control over PPCP formulations and incorporate wastewater-based forensic surveillance into routine environmental monitoring. For PPCP pollution to be addressed at the root, long-term environmental protection, ecosystem stability and public health sustainability will depend on behavioural and policy mitigation measures.

References

1. Charles Obinwanne Okoye, E. S. (2022). Occurrence and fate of pharmaceuticals, personal care products (PPCPs) and pesticides in African water systems: A need for timely intervention. *Heliyon*. doi:<https://doi.org/10.1016/j.heliyon.2022.e09143>
2. Kinga Slosarczyk, S. J.-K. (2021). Occurrence of Pharmaceuticals and Personal Care Products in the Water Environment of Poland: A Review. *Water*. doi:<https://doi.org/10.3390/w13162283>
3. Leslie Cizmas, V. K. (2015). Pharmaceuticals and personal care products in waters: occurrence, toxicity, and risk. *Environ Chem Lett*. doi:DOI 10.1007/s10311-015-0524-4

4. Lubliner B, M. R. (2010). *Pharmaceuticals and Personal Care Products in Municipal Wastewater and Their Removal by Nutrient Treatment Technologies*. Washington: Washington State Department of Ecology, Olympia, WA.
5. Najat Al-Odaini, M. P. (2011). Detecting Human Pharmaceutical Pollutants in Malaysian Aquatic Environment: A new challenge for water quality management .
6. Sofian Kanan, M. M.-S. (2022). Occurrence, analysis and removal of pesticides, hormones, pharmaceuticals, and other contaminants in soil and water streams for the past two decades: a review. *Research on Chemical Intermediates*. doi:<https://doi.org/10.1007/s11164-022-04778-7>

32. Forensic Gait Analysis in Individuals with Knock Knees: An Innovative Investigative Approach

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Abstract: Forensic gait analysis utilizes the unique characteristics of an individual's walking pattern to aid in criminal investigations and identification. This study aims to develop and validate an innovative investigative approach by focusing on the distinct gait features present in individuals with Genu Valgum, commonly known as "knock knees." The presence of this common musculoskeletal deformity results in specific biomechanical changes that are hypothesized to impart quantifiable and highly individualizing characteristics to a person's walk. The research involved a comprehensive review of existing literature, followed by experimental work focused on the collection and analysis of gait parameters from a study group diagnosed with knock knees. This analysis included the measurement of key factors such as step length, stride length, foot progression angle, and the degree of medial deviation characteristic of the condition. The results indicate that the Genu Valgum deformity introduces measurable and consistent deviations in the gait pattern that are unique to the individual. The study concludes that specialized forensic analysis of gait in individuals with knock knees provides a reliable and effective method for creating unique profiles, thereby enhancing the power of traditional forensic gait examination as a tool for personal identification in forensic science.

Keywords: *Forensic Science, Gait Analysis, Knock Knees (Genu Valgum), Personal Identification, Investigative Approach.*

Introduction

Forensic gait analysis has emerged as a specialized and increasingly credible domain within forensic science, focusing on the systematic examination of human locomotion for purposes of personal identification and investigative reconstruction. Gait, defined as the characteristic manner or pattern of walking, is a complex biomechanical activity governed by the coordinated interaction of the nervous, musculoskeletal, and cardiorespiratory systems. Owing to inter-individual variations in limb alignment, stride length, cadence, posture, and foot placement, gait is considered a unique behavioral biometric, comparable to fingerprints and DNA in its individualizing potential (*Savvy Forensics, 2025; The Royal Society, 2017*). The forensic relevance of gait analysis becomes particularly significant in scenarios where conventional identifiers such as facial features or fingerprints are unavailable or deliberately concealed. Surveillance footage, often the only form of evidence in crimes involving masked offenders, captures walking patterns that may serve as critical associative evidence. Courts, particularly in the United Kingdom, have recognized forensic gait analysis as admissible expert evidence when applied cautiously and scientifically, reinforcing its legitimacy within the criminal justice system (*Mangone, 2023; Sánchez-Gómez, 2025*).

The scientific foundation of gait analysis lies in biomechanics and podiatry, where locomotion is studied through spatiotemporal, kinematic, and kinetic parameters. Variables

such as step length, stride length, gait angle, foot progression angle, cadence, and base of gait are routinely analyzed using footprint examination, video recordings, pressure plates, and motion-capture (Ghaderi, 2023; Shaw, 2025). Advances in computational techniques, including machine learning and deep learning-based pose estimation models, have further enhanced the objectivity and precision of gait analysis, expanding its applicability across forensic, medical, and rehabilitative sciences (Ganesan, 2016; Steel, 2025). While gait analysis has been extensively studied in normal populations and biometric surveillance contexts, the forensic implications of pathological gait variations remain comparatively underexplored. Musculoskeletal deformities can introduce consistent and visually detectable deviations in walking mechanics, potentially strengthening the discriminatory power of gait evidence. One such clinically and forensically relevant condition is genu valgum, commonly known as knock knees, characterized by an inward angulation of the knees when standing or walking. Knock knees are a common developmental phenomenon in early childhood and usually resolve spontaneously by the age of seven or eight. However, when genu valgum persists into adolescence or adulthood, it is considered pathological and may arise due to genetic predisposition, obesity, nutritional deficiencies such as rickets, biomechanical imbalance, or degenerative joint conditions like osteoarthritis (Ningthoujam, 2023; Shetty et al., 2024). Persistent genu valgum alters lower-limb alignment, leading to abnormal load distribution across the knee joint and compensatory changes at the hip and ankle. Biomechanical studies have demonstrated that individuals with genu valgum exhibit altered spatiotemporal and kinematic gait parameters, including increased gait angle, wider step width, reduced stride length, altered knee valgus moments, and compensatory hip external rotation (Chopra, 2021).

A pilot study by Husain and Shaw in 2021, using pressure-plate analysis revealed delayed load transfer and altered force distribution in individuals with knock knees, further confirming condition-specific gait adaptations. Similarly, Leyko et al. in (2025) reported subtle yet consistent asymmetries in step length and gait mechanics in individuals treated for valgus knee deformities. Technological advancements have enabled automated detection of knock knees through gait analysis using computer vision frameworks such as OpenPose and MediaPipe (Ganesan, 2016). demonstrated that deep learning-based gait analysis systems could successfully identify genu valgum and provide corrective feedback through therapeutic interventions, highlighting the robustness and scalability of gait-based assessment tools. These findings underscore the reliability of gait deviations associated with genu valgum and their potential forensic value. From a forensic perspective, the inward angulation of the knees in knock-knee individuals produces a characteristic medial collapse during locomotion, resulting in distinctive gait markers such as increased gait angle, atypical foot progression angles, widened base of gait, and altered footprint alignment. These features are observable in surveillance footage and footprint evidence and may persist consistently over time, fulfilling key forensic criteria of universality, uniqueness, and relative permanence (Sindhu et al., 2024; The Royal Society, 2017). Epidemiological studies suggest that while exact prevalence figures for pathological genu valgum in the adult Indian population are unavailable, the condition is associated with obesity, fluorosis, and knee osteoarthritis, all of which are prevalent public health concerns in India (Mir, 2022; Singh, 2018). Given the high burden of knee osteoarthritis and increasing obesity rates, the persistence of knock knees into adulthood is likely to be more common than previously assumed, reinforcing the forensic relevance of studying such pathological gait patterns.

Despite growing recognition of gait analysis in forensic practice, limited research has systematically examined how specific musculoskeletal deformities, such as genu valgum,

influence forensic gait parameters. Most existing studies focus on clinical outcomes, rehabilitation, or corrective interventions, with minimal emphasis on forensic identification. This gap highlights the need for interdisciplinary research integrating biomechanics, medicine, and forensic science.

In this context, the present study aims to investigate forensic gait characteristics in individuals with knock knees, with particular emphasis on gait angle and related spatiotemporal parameters. By merging clinical insights with forensic methodology, this research seeks to demonstrate how pathological gait deviations can enhance the evidentiary value of gait analysis. Ultimately, the study underscores the potential of forensic gait analysis as a multidisciplinary tool capable of strengthening personal identification in criminal investigations while simultaneously contributing to biomechanical and medical knowledge.

Methodology

This research aims the intersection of forensic gait analysis (focusing on gait angles) and the postural problem of knock knees (genu valgum), synthesizing current research on forensic, biomechanical and functional implications.

Participants

A total of 100 participants were included in the study, comprising 50 normal individuals and 50 individuals exhibiting knock-knee deformity. Both male and female participants were selected to ensure gender representation. Basic anthropometric details such as age, gender, height, weight, and foot dimensions were recorded for all participants prior to analysis.

Data Collection Technique

Gait data were collected using footprint-based gait documentation. To obtain clear plantar impressions, a non-toxic acrylic medium was applied uniformly to the plantar surface of both feet of each participant. Participants were instructed to walk naturally at a self-selected pace along a flat surface, ensuring minimal conscious alteration of walking behavior. Footprints generated during locomotion were documented on a clean surface for subsequent analysis. Prior to data collection, all participants were briefed about the procedure, and informed consent was obtained. Ethical considerations regarding participant safety, comfort, and confidentiality were strictly maintained throughout the study.

Gait Parameters Analyzed

The collected gait impressions were analyzed for multiple forensic gait parameters, including; Gait angle, Step length, Step width, Stride length, Foot length, Foot width and Direction line characteristics

All measurements were performed using standardized measuring scales and calibrated tools to ensure accuracy and repeatability.

Data Recording and Analysis

Measured gait parameters were systematically recorded along with individual demographic details. Quantitative analysis was conducted to compare gait characteristics between normal

and knock-knee groups. Statistical evaluation focused on identifying deviations in gait angle and spatial parameters associated with genu valgum.

The study employed descriptive statistical methods to calculate mean values and comparative differences between groups, facilitating interpretation of condition-specific gait deviations relevant to forensic identification.

The methodology was based on principles of quantitative forensic gait analysis using two-dimensional kinematic evaluation. Emphasis was placed on identifying consistent and measurable gait deviations that could serve as individualizing features in forensic investigations.

Tabulation of Collected Data

1. Normal Individuals

Sample No.	Gender	Weight (kg)	Height (ft)	Foot Length (cm)	Foot Width (cm)	Step Length (cm)	Step Width (cm)	Gait Angle (°)
1	Male	78	5.9	26	10.2	74	10	5.2
2	Male	85	6.1	26.8	10.5	76	10.2	4.8
3	Male	70	5.8	25.5	10	72	9.8	6.1
4	Male	92	6.3	27.5	10.8	77	10.5	5.7
5	Male	65	5.7	25	9.8	71	9.5	4.5
6	Male	88	6.2	27	10.6	75	10.3	6.4
7	Male	75	6	26.2	10.3	73	10	5
8	Male	95	6.4	28	11	78	10.8	4.9
9	Male	68	5.9	25.8	10.1	72	9.7	6.3
10	Male	82	6.1	26.7	10.4	75	10.1	5.5
11	Male	60	5.6	24.5	9.6	70	9.3	6.7
12	Male	79	6	26.3	10.3	74	10	4.7
13	Male	71	5.8	25.4	9.9	72	9.8	5.9
14	Male	90	6.3	27.4	10.7	77	10.4	6
15	Male	67	5.7	25	9.8	71	9.6	4.6
16	Male	84	6.2	27.1	10.5	76	10.2	5.8
17	Male	73	5.9	25.9	10.1	73	9.9	6.2
18	Male	96	6.4	28.1	11.1	78	10.7	5.1
19	Male	64	5.6	24.6	9.7	70	9.4	4.4
20	Male	76	6	26.4	10.4	74	10.1	6.5
21	Male	69	5.8	25.3	9.9	72	9.7	5.3
22	Male	81	6.1	26.6	10.4	75	10.2	4.9
23	Male	66	5.7	25.1	9.8	71	9.5	6.6
24	Male	89	6.3	27.3	10.7	77	10.5	5.4
25	Male	74	6	26.2	10.2	73	10	6
26	Female	52	5.3	23	8.8	66	8.5	5.6
27	Female	48	5.2	22.5	8.6	65	8.3	4.7
28	Female	58	5.5	23.8	9.1	67	8.7	6.2
29	Female	62	5.6	24.2	9.3	68	8.8	5.1

30	Female	45	5	22	8.4	63	8	6.8
31	Female	65	5.7	24.6	9.4	69	8.9	4.5
32	Female	57	5.4	23.4	9	66	8.6	5.9
33	Female	60	5.6	24.3	9.2	68	8.8	6.4
34	Female	55	5.4	23.5	9	67	8.5	5
35	Female	50	5.2	22.6	8.7	65	8.3	6.3
36	Female	63	5.7	24.7	9.5	69	9	4.8
37	Female	49	5.2	22.4	8.6	64	8.2	5.7
38	Female	59	5.5	23.9	9.2	67	8.7	6.1
39	Female	61	5.6	24.4	9.3	68	8.9	4.9
40	Female	67	5.8	25	9.6	70	9.1	5.5
41	Female	53	5.3	23.1	8.9	66	8.5	6
42	Female	56	5.4	23.6	9.1	67	8.6	4.6
43	Female	64	5.7	24.8	9.5	69	9	5.8
44	Female	51	5.2	22.7	8.7	65	8.4	6.5
45	Female	54	5.3	23.2	8.9	66	8.5	4.4
46	Female	66	5.8	25.1	9.6	70	9.2	5.2
47	Female	47	5.1	22.2	8.5	64	8.1	6.7
48	Female	58	5.5	23.9	9.2	67	8.7	5.3
49	Female	62	5.7	24.7	9.4	69	9	4.7
50	Female	55	5.4	23.5	9	67	8.6	6.2

2.Knock Knee Individuals

Sample No.	Gender	Weight (kg)	Height (ft)	Foot Length (cm)	Foot Width (cm)	Step Length (cm)	Step Width (cm)	Gait Angle (°)
1	Male	78	6.1	27.5	10.3	70	12	9.4
2	Male	85	5.9	28.1	10.7	72	11	10.2
3	Male	65	5.6	26.7	9.8	68	13	8.9
4	Male	90	6.3	29	11.2	75	14	11.5
5	Male	55	5.2	25.5	9.5	66	10	9.8
6	Male	82	6	28.3	10.8	71	12	12
7	Male	70	5.7	27	10	69	11	8.7
8	Male	75	6.2	27.8	10.5	73	13	9.2
9	Male	95	6.4	29.5	11.5	78	15	12.5
10	Male	60	5.5	26	9.7	67	10	8.5
11	Male	77	5.8	27.3	10.2	70	12	10.7
12	Male	68	5.4	26.5	9.9	68	11	9.1
13	Male	88	6.1	28.8	11	74	13	11.2
14	Male	57	5.3	25.8	9.6	66	10	9.5
15	Male	83	6	28.2	10.9	72	12	10.1
16	Male	72	5.7	27.1	10.1	69	11	8.8
17	Male	80	6.3	28.7	11.3	73	14	11.8
18	Male	66	5.4	26.4	9.7	67	10	9.3
19	Male	77	5.9	27.4	10.4	71	12	10.5
20	Male	69	5.6	26.6	10	68	11	9

21	Male	85	6.2	28.9	11.1	75	13	11.7
22	Male	59	5.3	25.7	9.5	66	10	9.6
23	Male	74	5.8	27.3	10.3	70	12	10.3
24	Male	68	5.5	26.3	9.8	68	11	9.2
25	Male	90	6.4	29.2	11.4	77	14	12.3
26	Female	50	5	24.5	8.5	62	9	8.4
27	Female	55	5.3	25	9	64	10	9.1
28	Female	60	5.5	25.5	9.2	65	10	9.7
29	Female	45	5	24.2	8.3	61	9	8.2
30	Female	70	5.7	26	9.5	66	11	10.5
31	Female	65	5.6	25.8	9.3	66	10	9.8
32	Female	52	5.2	24.7	8.8	63	9	8.9
33	Female	57	5.4	25.2	9	64	10	9.3
34	Female	61	5.5	25.4	9.1	65	10	9.6
35	Female	75	5.8	26.5	9.8	68	11	10.9
36	Female	53	5.1	24.6	8.7	63	9	8.7
37	Female	58	5.3	25.1	9	64	10	9.4
38	Female	63	5.6	25.6	9.3	65	10	9.9
39	Female	48	5	24.3	8.4	62	9	8.6
40	Female	66	5.7	26	9.5	66	11	10.2
41	Female	54	5.2	24.8	8.9	63	9	8.8
42	Female	60	5.5	25.4	9.1	65	10	9.6
43	Female	55	5.3	25	9	64	10	9.2
44	Female	70	5.8	26.3	9.7	68	11	10.8
45	Female	49	5.1	24.4	8.6	62	9	8.5
46	Female	62	5.6	25.7	9.4	65	10	9.7
47	Female	58	5.4	25.2	9.1	64	10	9.3
48	Female	50	5.1	24.5	8.7	62	9	8.9
49	Female	67	5.7	26.1	9.6	66	11	10.6
50	Female	53	5.3	24.9	8.9	63	9	9

Results and Discussion

The experimental analysis revealed distinct differences in gait parameters between normal and knock-knee individuals. The average gait angle among normal males was **5.54°** and **5.59°** in females, while knock-knee males and females demonstrated significantly higher values of **10.15°** and **9.42°**, respectively.

Table 1: Gait Angle Measurements

Group	Total Sum of Gait Angle	Average Gait Angle (°)
Normal Male	138.5	5.54
Normal Female	138.9	5.59
Knock Knee Male	253.8	10.15
Knock Knee Female	235.6	9.42
Combined Normal Individual	138.5	5.54
Combined Knock Knee Individual	138.5	5.54

This indicates that knock-knee individuals exhibited nearly **75% greater gait angles** compared to normal subjects. Step length and width also showed notable variations: normal individuals-maintained stride lengths of approximately **72–75 cm** with step widths of **9–10 cm**, whereas knock-knee individuals displayed shorter stride lengths of **68–73 cm** and wider step widths ranging from **11–14 cm**.

These deviations suggest reduced stride efficiency and compensatory widening of stance in knock-knee subjects. Additionally, knock-knee individuals presented slightly larger foot lengths and widths, correlating with altered load distribution and medial collapse during walking.

Supporting these findings, (Ganesan, 2016) reported that children with unilateral genu valgum exhibited altered kinematic parameters, including increased gait angles and asymmetrical stride mechanics, confirming that knock knees disrupt normal locomotion. Furthermore, (Shaw, 2025) compared adolescent females with knock knees and flat feet, showing that knock knees produced distinctive gait deviations such as reduced step length and altered force distribution across the lower limbs. Together, these studies reinforce the present findings that genu valgum introduces consistent biomechanical deviations, thereby enhancing the evidentiary value of gait analysis in forensic contexts

Conclusion

The experimental analysis demonstrates clear biomechanical differences between normal and knock-knee individuals. Normal gait angles averaged around 5.5° , while knock-knee individuals exhibited significantly higher angles ($\sim 10^\circ$), wider step widths, and shorter step lengths. These deviations confirm that genu valgum alters locomotion mechanics, producing distinctive gait signatures useful in both forensic identification and clinical diagnosis. Gender differences were minimal in normal individuals but slightly more pronounced in knock-knee cases, with males showing higher deviations.

Overall, the study validates that gait analysis can reliably distinguish pathologically from normal locomotion, underscoring its dual importance in forensic science and pediatric orthopedics. Early recognition and intervention are essential to mitigate long-term complications, while forensic experts can leverage these distinctive gait markers to strengthen evidence in criminal investigations.

References

1. Chopra, P. K. (2021). *Gender Differences in Osteoarthritis of the Knee: An Indian Perspective*.
2. Ganesan, B. (2016). Kinetic and Kinematic Analysis of Gait Pattern of 13 Year Old Children with Unilateral Genu Valgum. *European Review for Medical and Pharmacological Sciences*, 20, 3168–3171.
3. Ghaderi, S. (2023). *Preventing Premature Osteoarthritis in Young Adults with Knock-Knee Deformity Using Exercises*.
4. Mangone, A. M. (2023). *Gait Analysis Advancements: Rehabilitation*.
5. Mir, H. N. (2022). *The Relation of Speed Variation and Body Fat Percentage on Gait Pattern*.
6. Ningthoujam, S. S. N. S. (2023). *Knock Knee Deformity and Body Mass Index among Male School Students Aged 9–13 Years in Chandigarh, India*.

7. Sánchez-Gómez, R. (2025). *Advances in Foot Biomechanics and Gait Analysis*.
8. Savvy Forensics. (2025). *Gait Pattern Analysis in Crime Scene Investigation*.
9. Shaw, R. H. D. (2025). Gait Analysis of Adolescent Females with Knock Knees Versus Flat Feet: A Pilot Study. *Journal of Society of Indian Physiotherapists*.
10. Shetty, A. K., V., V. S., & E. (2024). *Exploring the Burden of Knee Osteoarthritis in Rural South India: Community Prevalence, Risk Factors, and Functional Assessment among Adults Aged 40 and Above*.
11. Sindhu, K., Uday, A. N., Abhishek, S. J., & Anjali, S. (2024). *Gait Analysis—A Tool for Medical Inferences*.
12. Singh, B. (2018). *A Forensic Study of Gait Parameters and Their Relationship with Stature in Jatt Sikh Male Population of Ludhiana District of Punjab*.
13. Steel, S. (2025). Forensic Gait Analysis: Using Biomechanics to Identify Suspects. *Journal of Forensic Biomechanics*.
14. The Royal Society. (2017). *Forensic Gait Analysis: A Primer for Courts*. The Royal Society.

33. Studies on Forensically Significant Aphrodisiac Mandrakes

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Abstract: The Greek goddess of sexuality, love, and beauty, Aphrodite, is the source of the word "aphrodisiac." Food or medication that stimulates sexual desire is referred to as an aphrodisiac. Folk medicine and ethnobotany have long attributed an aphrodisiac, lust-inducing, or potency-boosting properties to a variety of plants and herbs. As a result, numerous edible and medicinal plants' leaves, petals, or roots were utilized as ingredients in love potions that were intended to intensify amorous pleasures or serve as general sexually stimulating agents. However, in the past, such magic potions frequently contained narcotic and psychoactive plants in addition to love herbs or herbal aphrodisiacs. Although several drugs derived from plants have historically been used as sexual enhancers, their lack of defined dosages and propensity for toxicity can result in serious side effects and legal investigations. Tropane alkaloids (hyoscyamine, scopolamine, and atropine) are found in such plants that have historically been used as aphrodisiacs. *Mandragora* species contain tropane alkaloids, primarily hyoscyamine and scopolamine, which make all portions of the plant dangerous. Mandrake is the common name for a variety of *Mandragora* species in the Solanaceae family. This herb has been used in medicine for thousands of years as a narcotic before surgery, as well as an emetic and an antidote to snakebites. Mandrake's root was also referred to as magic, aphrodisiac, psychedelic, and intimately associated with witchcraft under the term "*Devil's herb*" due to its "human body" shape and its narcotic and toxic effects. The concept of this strange plant is surrounded by a number of stories and legends that evoke feelings of something mysterious, sorrowful and even horrible. due to the belief that they would increase fertility. The plant's berries are also known as "*love apples*." All parts of the *Mandragora* species contain potent tropane alkaloids, making them extremely important in forensic research due to their strong toxic properties and potential for both accidental and intentional poisoning. In order to control the safety of herbal aphrodisiacs and look into the health and legal ramifications of their abuse, forensic science play a significant role. The main situations in which forensic science and aphrodisiac plants interact are poisoning, drug-facilitated crimes, illicit commerce, and product adulteration. In light of the forensic implications associated with *Mandragora* species, it is thought to be worthwhile to perform a comprehensive analysis of the studies conducted on these plants. The objective is to furnish forensic experts with extensive resources that focus on the phytochemical and toxicological aspects of *Mandragora* for better reference and understanding.

Key words- Mandragora species ,Mandrakes, Aphrodisiac plants, Tropane alkaloids, love apples, drug-facilitated crimes, Phytochemical analysis. Toxicological aspects.

Introduction

Two Greek words meaning "hurtful to cattle" are the source of the name Mandragora. The Arabs call it "Satan's apple." Its narcotic and intoxicating properties, along with its use as a surgical anesthetic, made it highly valued in ancient Greece and Rome. The Bible mentions mandrake (Gen. 30:14–16), and its use is frequently linked to its purported ability to increase fertility (Fleisher & Fleisher, 1994). The historian Josephus Flavius wrote about the "dangerous" technique of gathering mandrake roots as early as AD 93. Uprooting a mandrake was seen extremely harmful in the Roman Empire. Because the mandrake's root resembles a

human body or a phallus, it was thought to possess occult abilities. During the Middle Ages, "the female" root was cut into the shape of a human. It was thought to bring prosperity and good fortune. The root was allegedly harvested by medieval witches under the gallows at night. It was said that the crushed root produced hallucinations, a death-like trance, and sleep. In addition, the root was thought to have been utilized in flying potions and to induce insanity.

The popularity of mandrake in Europe began to slowly decline in the sixteenth century (Hanuš et al., 2005). Mandrake was once regarded as the most important plant in the Mediterranean, but its mythology and mystique have all but disappeared from modern usage. There are references to Mandrake's mythology in Middle Eastern cuneiform inscriptions from the fourteenth century B.C. and mythical history. Early Mesopotamian, Greek, Old Hebrew, Roman, Egyptian, Arabic, and various other cultures reference mandrake, which was regarded as a crucial medicinal and magical plant throughout the Middle Ages in Europe. This plant was believed to have the ability to cure a wide range of ailments, such as insomnia and infertility, as well as the capacity to predict the future and provide protection for soldiers in battle. Additionally, the scent of mandrake's yellow fruits is highlighted in biblical texts, specifically Genesis and the Song of Solomon, where they are noted for their sexual connotations. So it makes sense that mandrake became known as the middle-world equivalent of Viagra! (Forest Service, USDA, n.d.) Texts from the 12th-14th centuries (e.g., Rasaratna Samuchchaya) reference *Mandragora autumnalis* as "Vamshalochana." It was appreciated as a mild sedative for vata diseases and an occasional aphrodisiac to boost sexual energy (vata-pitta balance), but only in extremely controlled formulations—mixed with black pepper, long pepper, and jaggery to promote safety and absorption (Ask Ayurveda, n.d.)

Myths and legends associated with the mandrake plant

In an essay titled "*The Magic of the Mandrake*," (Hill, 2025) wrote: "The mandrake plant has such an unparalleled mythical legacy that it holds the unique distinction of being the subject of numerous works, including several books, solely dedicated to it." This plant's history illustrates the similarities between these two ostensibly unrelated areas at a time when the line between magic and medicine was less apparent than it is now. According to Wright and Klassen (Wright & Klassen, 2002), this historiography places the mandrake squarely into the contextual framework of a bygone era in which even medical doctors with university training frequently used charms and astrology as part of their daily practice. Legends and tales from antiquity that related to the mandrake plant Scripture and other sources from distant antiquity served as inspiration for the tales and legends surrounding the mandrake plant. King Solomon's ring is one of the oldest instances of mandrake mythology. This ancient custom was transmitted through literature credited to Hermes Trismegistus. According to the legend, the King employed a ring containing mandrake root to ward off demons; this is why the plant is known as "The Tree of Solomon" in Arabic and Armenian (Dafni et al., 2021). Another myth linking the mandrake to fertility may be found in the Bible. Chapter 30 of the Book of Genesis tells the story of Leah's childless sister Rachel asking her to borrow the mandrake roots that her son had collected from the fields in order to help her conceive. This myth persisted into the sixteenth century, when John Gerard's Herball from 1597 mentioned the mandrake's existence in the Bible through this Genesis tale and its later repute as a fertility aid (Gerard 1990, 85-86). The ceremonies required to accompany the root's harvesting were the main focus of mandrake myths. The mandrake plant is an illustration of how older ideas about the qualities of plants evolved and were adapted, as it came to be believed that it truly housed a spirit rather than just being influenced by cosmic forces. The mandrake spirit is said

to have screamed loudly enough to instantly kill anyone who heard it when the plant was uprooted. Thus, it was advised that in order to unearth it, people tie a dog to the root. After that, the dog would pass away, saving the human harvester (Carter, 2003). Translations of the Pseudo-Apuleius (Herbarium Apulei), a late antique medicinal literature from the fourth century CE, provide one of the earliest sources for this specific story (Kinney, 2022).



Fig 1



Fig 2

Fig 1 This medieval manuscript demonstrates the "Doctrine of Signatures" by depicting mandrake as either a man or a woman. Fig 2- Mandrake's magical power was considered to be so intense that it would scream and kill anyone within earshot when extracted from the ground (Forest Service, USDA, n.d.)

Mandrake is associated with magic and witchcraft in many cultures, and like other narcotic magical plants, it is believed to have both pharmacological and psychosomatic effects, particularly when it contains narcotic chemicals. It is not unexpected that this plant has numerous names in various languages because of its special qualities and associated mythology. Several notable authors have explored the theme of the mandrake plant and its properties in their literary works. William Shakespeare incorporated references to mandrakes in plays such as *Othello*, *Macbeth*, and *Romeo and Juliet*, famously writing in Act IV, Scene 3 of *Othello*, "And shrieks like mandrakes torn out of the earth." Similarly, Niccolo Machiavelli featured the plant in his comedic work *La Mandragola*. Niccolo Machiavelli's comedic work *La Mandragola*, C. J. Cela's *Diccionario del Erotismo*, and J. K. Rowling's *Harry Potter and the Chamber of Secrets* each incorporate references to mandrakes, underscoring the plant's cultural significance and symbolic implications across various literary genres and historical contexts. (Dafni et al., 2021) & (Carter, 2003).

Botanical description

(Monadi et al., 2021) conducted a comprehensive review of the *Mandragora* genus, detailing its taxonomy, phylogeny, morphology, geographical distribution, habitat, and phenology. The study indicates that *Mandragora turcomanica* is closely related to *Mandragora autumnalis*, with both species being sister taxa based on genetic, molecular, and morphological evidence. *Mandragora* species belong to the Kingdom Plantae, Subkingdom Viridiplantae, Infrakingdom Streptophyta, Superdivision Embryophyta, Division Tracheophyta, Subdivision Spermatophytina, Class Magnoliopsida, Superorder Asteranae, Order Solanales, Family Solanaceae, and Genus *Mandragora*. (Integrated Taxonomic Information System [ITIS], n.d.) The number of recognized *Mandragora* species has varied over time, with three species identified by (Ungricht et al., 1998): *M. officinarum* L., *M. turcomanica* Mizg., and *M. caulescens*. C. B. Clarke further combined *M. autumnalis* and *M. officinarum* based on their morphological characteristics, while *M. chinghaiensis* was recognized as a synonym of *M. caulescens*. Currently, phylogenetic and morphological studies classify the genus *Mandragora*

to include five species: *M. officinarum* L., *M. autumnalis* Bertol., *M. turcomanica* Mizg., *M. caulescens* C. B. Clarke, and *M. chinghaiensis*. (Volis et al., 2018).

Morphology of *Mandragora* species

M. turcomanica

Plants stemless; roots thick, branched. Leaves all arranged in a rosette, 34–66 cm long (including petiole), elliptic-ovate, slightly pubescent, margin crenate-dentate; blade 24–50 × 16–28 cm, 6–7 pairs of pinnate veins; petioles 7–21 cm long. Flowers 7–20 per plant; pedicels 4.5–12.5 cm long. Calyx-lobes with prominent mid-vein and reticulate venation, 1.6–1.8 × 0.5–0.8 cm in flower, 3.5–4 × 1.5 cm in fruit.

Corolla 2.3–2.7 cm long, violet, glabrous inside, with spreading hairs outside; lobes 15–21 × 8–10 mm, oblong. Filaments unequal, 9–12 mm long; anthers 3 mm long, with a tuft of spreading, 2–3 mm long hairs at base. Style 18–19 mm; stigma 1–1.2 × 2.5–3 mm. Fruiting pedicels 8.5–14 cm long. Fruits 3.6–4.8 cm in diameter, yellow, strongly aromatic. This species' plants have thick, branching roots and no stems. The rosette-arranged leaves are ellipticovate, measuring 34–66 cm in length, with hairy surfaces and crenate-dentate margins. Leaf blades are 24–50 × 16–28 cm with 6–7 pairs of pinnate veins, and petioles range from 7–21 cm. Each plant produces 7 to 20 flowers, with pedicels of 4.5 to 12.5 cm. The calyx lobes, featuring a mid-vein and reticulate venation, measure 1.6–1.8 × 0.5–0.8 cm in flowers and 3.5–4 × 1.5 cm in fruit. The violet, glabrous corolla measures 2.3–2.7 cm, with lobes 15–21 × 8–10 mm and filaments of 9–12 mm; anthers are 3 mm with hairs at their base. The style is 18–19 mm and stigma measures 1–1.2 × 2.5–3 mm. Fruiting pedicels are 8.5 to 14 cm long, with fruits that are yellow, aromatic, and range from 3.6–4.8 cm in diameter (Akhani & Ghorbani, 2003).



M. turcomanica

M. caulescens and *M. chinghaiensis*

These species are characterized by a stem and a robust, branching root system. The leaves, which can be oblanceolate or spatulate, are clustered and may reach up to 30 cm in length. *M. chinghaiensis* features rosettes of leaves with small stems, while *M. caulescens* is noted for its elongated stems that can grow to 60 cm. The flowers have pedicels measuring 5–10 cm long, with a globular ovary that has a diameter of 2.5 to 5 mm. The calyx measures 5 to 30 mm in length, with anthers ranging from white to yellow at 1–5 mm. The style typically measures 3–6 mm. The corolla, which can be yellow to purple, spans 5 to 30 mm and has a campanulate to cup-shaped appearance, with filaments measuring 3–10 mm. The fruits are globular, 10 to 25 mm in diameter, and can be pale greenish-white or greenish-yellow. Seeds are small, 1.5 to 3.0 mm long, and vary from yellow to light brown (Ungricht et al., 1998), (Tu et al., 2010)

*M.caulescens**M.chinghaiensis*

M. officinarum and M. autumnalis

Two species, *M. officinarum* and *M. autumnalis*, have short or no stems and two to three robust, sub-cylindrical roots measuring 15–20 cm long and 0.2–1.5 cm in diameter, varying in color from grayish to reddish-brown. The rhizome is 10 to 40 cm long and 1 to 1.5 cm in diameter, possibly expanding at the crown in older plants, with roots emerging from exfoliating cork buds. They have sweeping, simple leaves in a dense basal rosette; *M. officinarum* has sparsely villous leaves while *M. autumnalis* has subglabrous leaves. The solitary flowers, ranging from 1.2 to 6.5 cm in size, display colors from greenish to pale blue or violet. The greenish-white corolla of *M. officinarum* is five-lobed and up to 2.5 cm long, while *M. autumnalis* features a longer violet corolla with broad triangular lobes. Fruits from the mandrake species are categorized by their shape: *M. officinarum* produces globose fruits in the western Mediterranean, while *M. autumnalis* yields ellipsoid fruits in the Middle East. Notably, the western Mediterranean mandrakes bear smaller fruits compared to their Middle Eastern counterparts, which are characterized by their tastiness and aromatic qualities upon ripening. Seed size further differentiates these populations, with Mediterranean, European, and North African mandrake seeds measuring between 3-3.5 mm, while those from Iran, Israel, and Turkmenistan are notably larger at 5-6 mm (Fakir & Özçelik, 2009).

This article provides a comprehensive review of the phytoconstituents found in *Mandragora* species, documenting various investigations that have identified numerous compounds, including alkaloids, coumarins, lipids, flavonoids, withanolides, calystegines, essential oils, and carotenoids. By analyzing existing literature that has been gathered from significant academic databases like PubMed, Scopus, Google Scholar, Web of Science and Science Direct, it synthesizes current data about *Mandragora* spp. The research employs keywords like *Mandragora*, mandrake, ethnobotany phytochemistry, ethnopharmacology, and toxicity to investigate multiple aspects of *Mandragora*. The review emphasizes not only the phytochemical profile but also explores historical context, botanical characteristics, folkloric significance, pharmacological uses, and toxicological implications. It includes case histories pertaining to *Mandragora* poisoning, thereby shedding light on the potential dangers and consequences associated with its use.

Phytochemical studies

Alkaloids in *Mandragora* species

Occurring substances with one or more nitrogen atoms are called alkaloids. Alkaloids, which means "alkaline-like," get their name from the basic character of many members of this group. Alkaloids are difficult to define because many compounds that include nitrogen do not always fall into this category. For instance, biogenic amines or amino sugars are naturally occurring plant compounds that include nitrogen but are not classified as alkaloids. Kohnen-

Johannsen et al. have covered the chemistry, pharmacology, biosynthesis, and production of tropane alkaloids in detail. Tropane alkaloids (TA) are significant secondary plant metabolites that are mostly found in notable quantities in the families Erythroxylaceae and Solanaceae. scopolamine and Hyoscyamine, cocaine, and calystegines are the three main classes of TAs, which are distinguished by their distinct bicyclic tropane ring structure. TAs differ greatly in their biological, chemical, and pharmacological characteristics despite sharing the same basic structure (Kohnen-Johannsen & Kayser, 2019). One of the primary phytoconstituents of *Mandragora* species are alkaloids. Tropane alkaloids are the most common type of alkaloids found in the genus. A tropane ring (8-methyl-8-azabicyclo (Forest Service, USDA, n.d.), (Hanuš et al., 2005), (Fleisher & Fleisher, 1994) is included in the chemical structures of these bicyclic secondary metabolites, which are mostly found in the Solanaceae family (Lounasmaa & Tamminen, 1993). This class has been shown to contain over 200 distinct chemicals (Koleva et al., 2012). Some of the selected studies carried for the isolation and identification of alkaloids are briefly summarised.



M. officinarum



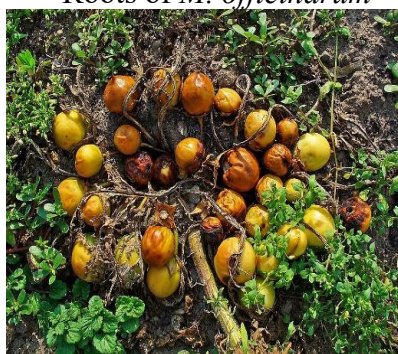
M. autumnnalis



Roots of *M. officinarum*



Roots of *M. autumnnalis*

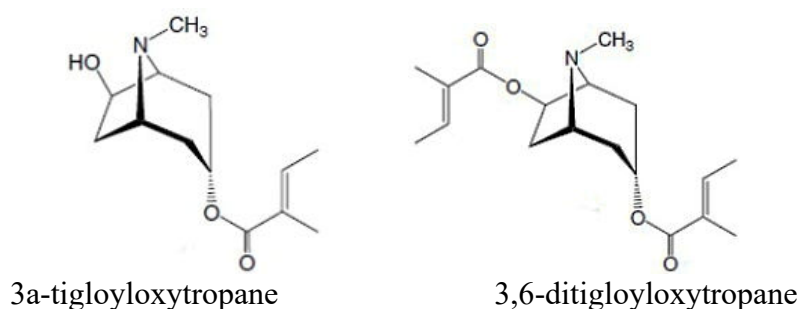


Fruits of *M. officinarum*

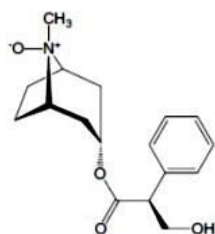
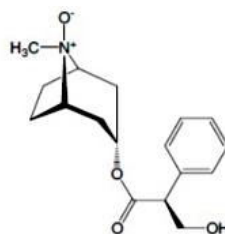
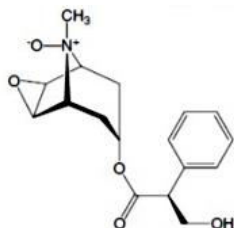


Fruits of *M. autumnnalis*

The pioneering works of German authors (Hesse, 1901) and (Thoms & Wentzel, 1898), (Thoms & Wentzel, 1901) at the turn of the 19th to 20th century detailed the alkaloid content in mandragora. They isolated scopolamine and hyoscyamine alongside atropine, though not in pure form. In 1992, Carr and Reynolds identified norhyoscyamine (pseudohyoscyamine) in mandragora and another Solanaceae. (Staub, 1962) published one of the first chemical studies on *Mandragora officinarum* L., employing paper chromatography and the preparation of crystalline picrates to determine the presence of atropine, scopolamine, cuscohygrine, scopine, and hyoscyamine in mandragora roots. Furthermore, he identified apoatropine and belladonnines through paper chromatography and detected norhyoscyamine in the roots (Hanuš et al., 2005). In (Jackson & Berry, 1973) conducted an analysis of total alkaloids extracted from the roots of two species of *Mandragora officinalis* (Solanaceae): *M. autumnalis* Bertol. and *M. vernalis* Bertol. Utilizing thin-layer chromatography (TLC) and gas-liquid chromatography (GLC), they compared the alkaloid profiles to authentic standards to ascertain potential differentiation between the species based on alkaloid content. The study confirmed the presence of hyoscyamine, hyoscyne, cuscohygrine, and apoatropine, the latter of which had been previously suggested by (Staub, 1962) through paper chromatography (PC). Notably, belladonnine was detected in the dried roots but was absent in fresh roots, possibly obscured by the high levels of cuscohygrine in the latter, which shares similar R_f values. GLC results were inconclusive for belladonnine due to the breakdown to apoatropine during the process. Both *M. autumnalis* and *M. vernalis* species contained two tigloyl esters of hydroxytropene: 3a-tigloyloxytropene and 3,6-ditigloyloxytropene, along with a minor amount of another unidentified alkaloid. The analysis of alkaloid compositions showed no significant concentration differences that would exceed normal biological variations. Both species' dried roots contained cuscohygrine, scopolamine, and apoatropine and hyoscyamine, In the dried roots Belladonnine were present, but could not be detected in the fresh roots The identification of the tigloyl esters marked a novel finding in the study.



Phillipson and Handa (Phillipson & Handa, 1975) conducted a study on the N-oxides of hyoscyamine and hyoscyne, which are part of the Solanaceae family. They successfully synthesized and characterized two isomeric forms for each compound: isomer 1 (equatorial $N^{+}-O^{-}$) and isomer 2 (axial $N^{+}-O^{-}$). Isomer 1 of hyoscyamine N-oxide was extracted from different parts of *Atropa belladonna* L., *Hyoscyamus niger* L., and *Datura stramonium* L. Similarly, isomer 1 of hyoscyne N-oxide was identified in these two species and in the leaves of *A. belladonna*. *Scopolia lurida* Dun. and *S. carniolica* Jacq. also contained both N-oxides of hyoscyamine and isomer 1 of hyoscyne N-oxide in their leaves, roots and stems. *Mandragora officinarum* L. produced both N-oxides across its roots, stems, leaves, and fruits. The results highlighted the presence of two distinct N-oxides of hyoscyamine and hyoscyne throughout the investigated species.

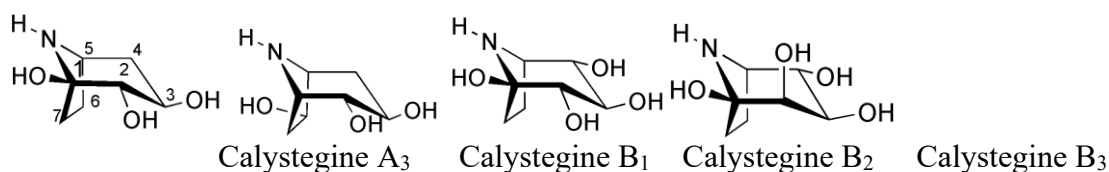
Equatorial N⁺-O⁻ isomerAxial N⁺-O⁻ isomer

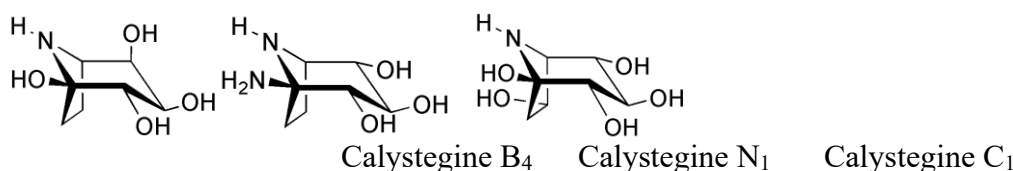
N-Oxides of tropane alkaloids

Calystegines -Poly hydroxy alkaloids with a nor tropane skeleton

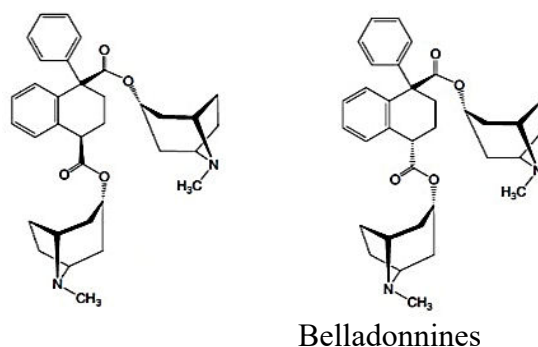
Calystegines represent a novel class of polyhydroxy alkaloids featuring a nor-tropane skeleton, identified in the root cultures of *Atropa belladonna* through chromatographic techniques such as TLC and GC, and further characterized using NMR and mass spectroscopy. Their presence was investigated across various Solanaceae family species. The proposed biosynthetic pathway suggests that these compounds are generated via the tropane alkaloid pathway, initiating with the metabolite pseudotropine. A unique tropinone reductase that facilitates the formation of pseudotropine was isolated and characterized from *Atropa belladonna* root cultures. Additional experiments, including ¹⁵N-tropinone feeding, provided substantiating evidence for the role of tropinone and pseudotropine in the synthesis of calystegines, resulting in an increase in their formation (Dräger et al., 1994), (Dräger et al., 1995) investigated the extracts from various parts of *M. officinarum*, including roots, leaves, spring leaves, flowers, and fruits, specifically analyzing calystegine A3 and calystegines B1 and B2. Their findings indicated that the aerial parts of the plant contained a higher concentration of calystegines compared to the root. Among the examined parts, spring leaves exhibited the highest concentration, while fruits had the lowest.

(Bekkouche et al., 2001) employed gas chromatography coupled with mass spectrometry (GC-MS) to analyze the distribution and content of eight calystegines (A3, A5, B1, B2, B3, B4, C1, and N1) across several plant species, including *Datura metel*, *Atropa belladonna*, *Hyoscyamus albus*, *Mandragora autumnalis*, *Solanum sodomaeum*, *Withania somnifera*, *Withania frutescens*, and *Brunfelsia nitida*. The study revealed a more restricted distribution for calystegines N1 and C1, while calystegines A3, B1, B2, and B3 were the most frequently detected. Notably, A5 and B4 were not found in any of the samples investigated. This research is significant as it marks the first examination of calystegines in the genera *Withania* and *Brunfelsia*, as well as in *Solanum sodomaeum* and *Mandragora autumnalis*. Additionally, structural formulas for some of these calystegines were reported.

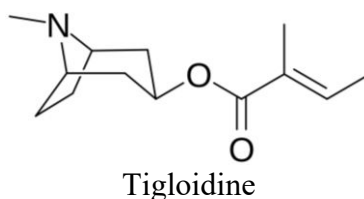




(Al-Khalil & Alkofahi, 1996) identified several compounds, including scopolin, herniarin, scopoletin, umbelliferon, chlorogenic acid, and angelicin in the roots, leaves, unripe fruits, and seeds of *M. autumnalis*, a plant that grows wildy in the Jordan valley. Additionally, (Staub, 1942) conducted an examination of the roots of *M. autumnalis*, focusing on the identification of non-alkaloidal constituents. In this study, tropic acid and scopoletin were identified as key compounds present in the plant's roots.



Razzakov et al., (1998) conducted a comprehensive study of surface parts of *M. turcomanica* Mizger, including leaves, roots, rhizomes, fruits, and seeds during different vegetation periods. They successfully isolated several alkaloids: scopolamine, hyoscyamine, and apohyoscyne. Furthermore, their analysis using TLC and MS techniques identified norhyoscyamine, apoatropine, α - or β -belladonnine, $(\pm)$ -3 α -tigloyloxytropan-6 β -ol, tigloidine, and three unidentified alkaloids with molecular weights of 209, 219, and 333.



Albahri et al., (2025) conducted a study analyzing the ethanolic leaves extract of *Mandragora autumnalis* using LC-MS in both positive and negative ionization modes, identifying 34 chemical compounds. In positive ionization, the predominant compounds included various bioactive alkaloids such as hyoscyamine, solacaproine, tropine, and tropinone. The profile also included phenolic compounds like chlorogenic acid and flavonoids (quercetin and chrysin). Significant findings included fatty acid derivatives such as ethyl palmitate and linoleic acid, alongside various esters (e.g., ethyl xhydrocinnamate) and furan derivatives (e.g., 5-hydroxymethyl-2-furancarboxaldehyde). Ketones and aldehydes, including 1-hydroxy-2-propanone and β -ionone, were detected, along with a novel quinoline alkaloid named simulanoquinoline. In negative ionization, high levels of polyphenolic acids including chlorogenic acid were noted, along with linolenic acid, coumarins, and various glycosides like rutin and hyperoside. The findings suggest that MAE possesses a diverse phytochemical profile, indicating its potential as a source of bioactive compounds for pharmacological use.

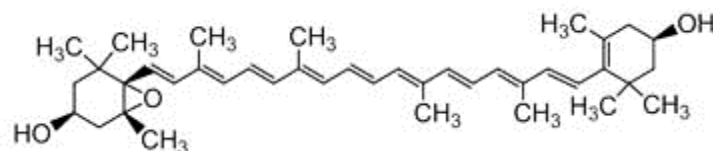
Studies on odoriferous principles/essential oil of *Mandragora* spp.

(Fleisher & Fleisher, 1992) conducted a study on the odoriferous elements of mandrake fruits *Mandragora officinarum* L., a plant with mythological and romantic associations in the Bible. The ripe fresh fruits were crushed and extracted using methylene chloride, resulting in a yield of 0.35% fatty oil that contained various odoriferous components. Fifty-one chemicals have been identified using capillary gas chromatography-mass spectrometry. The major compounds identified included ethyl butyrate (21.6%), butyl acetate (9.1%), hexanal (0.25%), butanol (4.1%), hexyl acetate (6.1%), hexanol (14%), ethyl 3-hydroxybutyrate (4.1%), linalool (0.38%), 3-methyl thiolpropanol (4.4%), 3-phenylpropanol (2.1%), and eugenol (2.3%), along with several minor compounds.

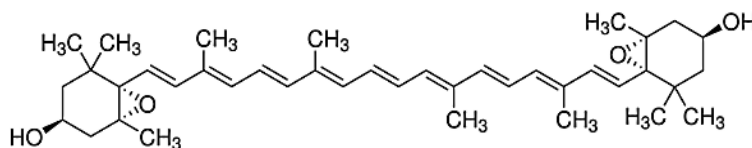
(Arbia et al., 2019) examined the essential oils (EOs) and aromatic volatiles from *M. autumnalis* in Central Tunisia. Essential oils were extracted via hydro distillation from fresh leaves, roots, and fruits, while volatile emissions were sampled using headspace solid phase micro extraction (HS-SPME) and analyzed with gas chromatography-mass spectrometry (GC/MS). The analysis revealed 51 substances, including esters, alcohols, and hydrocarbons, ketones, aldehydes, and acids comprising 96.2-98.6% of the oil content, with pentadecanoic acid (34.2%) and hexadecanol (26.3%) as predominant components. Additionally, 78 volatile compounds were identified, largely consisting of esters and alcohols, with 1,3-butanediol and ethyl hexanoate both at 12.3%. Using headspace GC-MS, (Baser et al., 1998) investigated the essential oil of fresh, ripe *M. autumnalis* fruits, identifying over 100 compounds, predominantly ethyl esters of medium-chain acids. The study analyzed ripe and unripe mandragora fruits from the Jerusalem region, revealing more than 130 compounds. The main constituents were 34.6% ethyl hexanoate, 25.5% ethyl butyrate, 14.2% ethyl decanoate, 13.1% ethyl dodecanoate, and 10.7% decyl acetate. Additional identified compounds included were present in varying percentages like ethyl caprate 6.15%, ethyl caproate 4.40, ethyl caprylate 8.70, ethyl cinnamate 0.60, ethyl laurate 3.19 and various ethyl esters, and fatty acids at minor concentrations. (Karakaş & Dörtbudak, 2023) investigated the essential oil components of *Mandragora autumnalis* using Gas Chromatography-Mass Spectrometry (GC-MS) with a capillary column. The study identified several major volatile components, including Phytol (17.91%), Citronellyl propionate (19.15%), and Farnesol (cis,cis- 17.05%). Other notable components included Heptadecane (6.31%), Pentacosane (5.305%), Octadecane (3.98%), Pelargonaldehyde (3.37%), Docosane (8.3%), Geranyl propanoate (2.96%), Dodecane (2.54%), Oct-7-enol (3,7-dimethyl- 2.44%), Nerylisovalerate (2.2%), Hexadecane (2.0%), and Butyraldehyde (2-methyl- 1.2%).

Carotenoid composition

(Pakhlavouni et al., 2000) analysed Carotenoid composition and chloroplast ultrastructure in two cell lines of *Mandragora turcomanica*. High total chlorophyll and carotenoid levels were present in both lines, with grana formation linked to increased chlorophyll b and epoxy-carotenoids, specifically antheraxanthin and violaxanthin. Observations indicated chloroplast degradation with disorganized lamellae and large osmiophilic globules. Furanoid xanthophylls may indicate chloroplast degradation, and thylakoid grana formation correlates with specific chlorophyll and carotenoid ratios.



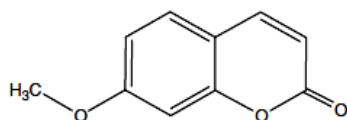
Antheraxanthin



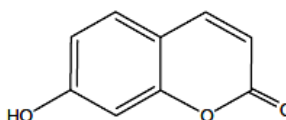
Violaxanthin

Coumarins

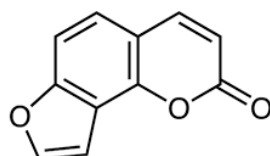
Al-Khalil and Alkofahi investigated several compounds in the roots, leaves, unripe fruits, and seeds of *M. autumnalis*, which is found in the Jordan valley. These compounds include scopoletin, angelicin, scopolin, herniarin, umbelliferon, and chlorogenic acid. Additionally, Staub investigated the roots of *M. autumnalis* focusing on non-alkaloidal constituents and identified scopoletin and tropic acid. (Jodallah, 2013) used GC-MS to investigate the chemical makeup of the *Mandragora autumnalis* plant's constituent parts. A total of 219 components have been found in various plant sections. When the chemical composition of the plant's various parts was compared, it was discovered that coumarins were present in every part of the plant, anthraquinones and tannins were present in every part of the plant except the roots, and glycosides were only present in ripe fruits.



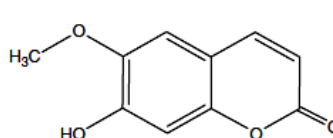
Herniarin



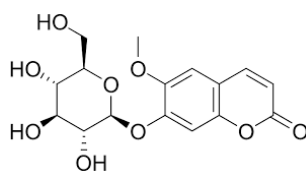
Umbelliferon



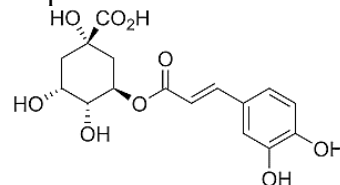
Angelicin



Scopoletin



Scopolin



Chlorogenic acid

Total Flavonoid Content Using Aluminum Chloride Colorimetric Method

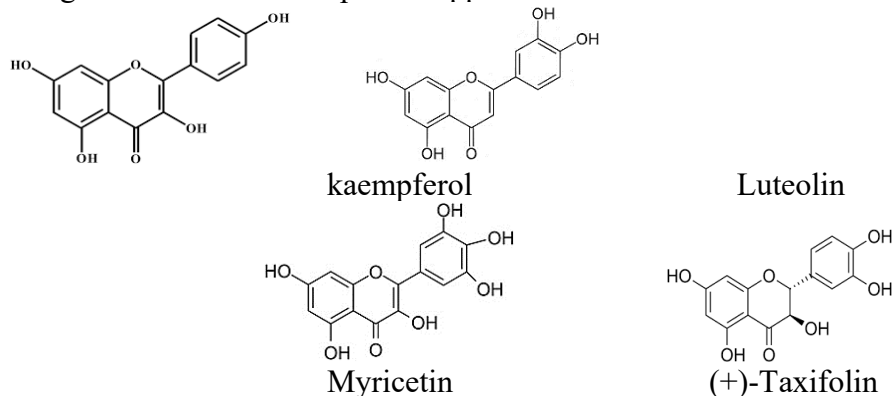
The aluminum chloride colorimetric method was applied according to Woisky and Salatino (Jackson & Berry, 1973) with some modifications to estimate the total flavonoid of *M. autumnalis* extract and fractions. Briefly, the solution (1 mg/ml) of the tested samples was prepared; the 2 ml of the sample was mixed with 0.1 ml of AlCl_3 (10%), 0.1 ml of 1M sodium acetate, and 2.8 ml of distilled water. After 30 min incubation at room temperature, the absorbance was measured at 415 using (UV-vis) spectrophotometry. Rutin was used to make the calibration curve using five serial dilutions (100, 50, 25, 12.5, and 6.25 $\mu\text{g/ml}$). As well, the total flavonoid content was expressed as mg of rutin (RE) equivalents/g extract

Phenolic content of *Mandragora autumnalis*

Mahmod and Talib utilized a modified Folin-Ciocalteu Method to assess the total phenolic content in extracts. They prepared a sample concentration of 8 mg/ml by mixing 12.5 μ l of *M. autumnalis* samples with 250 μ l of a 2% sodium carbonate solution in a 96-well plate. This mixture was allowed a 5-minute response time at room temperature, after which 12.5 μ l of a 50% Folin-Ciocalteu reagent was added and allowed to react for 30 minutes. Absorbance was subsequently measured at 630 nm, with total phenolic content expressed as mg of gallic acid equivalents per gram of extract. The solution had a total phenolic content of 0.1–1 mg/ml.

Flavonoids

The Aluminum Chloride Colorimetric Method was employed by (Mahmod & Talib, 2023) to assess the total flavonoid content in *M. autumnalis* extract and its fractions. This method, based on the protocols of (Woisky & Salatino, 1998) The absorbance was measured at 415 nm using UV-vis spectrophotometry, and the flavonoid concentration was calculated as mg of rutin equivalents per gram of extract, using a calibration curve established from five serial dilutions of rutin. (Al-Maharik et al., 2022) studied the flavonoids fraction in *M. autumnalis* mature fruit, identifying four compounds: luteolin, myricetin, kaempferol, and (+)-taxifolin, using ^1H and ^{13}C NMR spectral approaches.



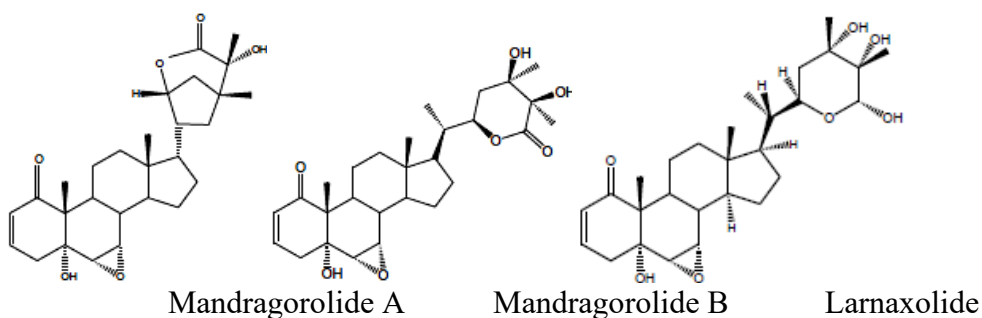
Lipids and lipid-like compounds in *Mandragora* species

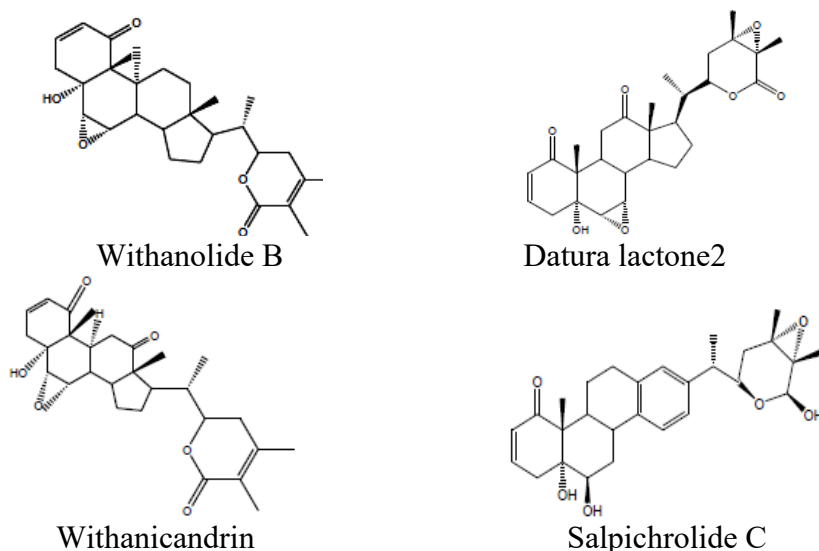
Asilbekova et al., (1994) undertook a comprehensive study focused on the lipids present in the plant *Mandragora turcomanica* Mizger. Their investigation aims to explore and characterize the lipid composition of this particular species, contributing to the understanding of its biochemical properties., revealing for the first time the identification of lipids in its tuberous roots. A total of twenty classes of lipids and lipophilic compounds were identified using various analytical methods including CC, TLC, UV and IR spectroscopy, and mass spectrometry. Triacylglycerols and glycolipids were found to be the main components of acyl-containing lipids, while alkanols and sterols were identified as significant lipophilic substances. The predominant fatty acids in these lipids were 18:2, 18:1, and 16:0. The study (Gusakova & Asilbekova, 1997) identifies for the first time the structure and species composition of triacylglycerols (TAGs) in the tuberous roots of *Mandragora turcomanica* Mizger, utilizing mass spectrometry and enzymolysis with pancreatic lipase. The TAGs consist of 42 species, predominantly triunsaturated and unsymmetrical diunsaturated molecules, with linoleic acid in the sn-1 (Forest Service, USDA, n.d.) position and oleic acid in the sn-2 position of the glycerol molecule. (Asilbekova & Glushenkova, 1998) studied the

lipid contents of *Mandragora turcomanica* fruit, revealing that the lipid compositions of its seeds and flesh are qualitatively identical, with greater saturation levels of fatty acids in the flesh compared to the seeds. In another study, mass spectrometry and enzymolysis indicated that 18:2 acids selectively esterify at the sn-2 position in the seeds' triacylglycerols (TAGs), whereas both 18:1 and 18:2 acids acylate this site in the flesh's TAGs. The primary TAG species in both parts is sn-1(Forest Service, USDA, n.d.) oleoyldilinoleate, with triunsaturated and monounsaturated-diunsaturated glycerides as the dominant types (Asilbekova & Glushenkova, 1999). (Hanuš et al., 2005) thoroughly reviewed Tashkent's group's findings on *Mandragora* species. (Mahmod & Talib, 2021) investigated the ethanol extract of *Mandragora autumnalis* through GC-MS technique, identifying several compounds, including palmitic acid, lauric acid, caprylic acid, capric acid, stearic acid, margaric acid, methylcaprate, vianol, eicosane, squalene, linoleic acid, cinnamic acid, methyl stearate, β -belladonnine, apotropine, borneol, 3 α -tigloyloxytropan, and ethyl butanoate. (Mahmod & Talib, 2021) investigated the ethanol extract of *Mandragora autumnalis* through GC-MS technique, identifying several compounds, including palmitic acid, lauric acid, caprylic acid, capric acid, stearic acid, margaric acid, methylcaprate, vianol, eicosane, squalene, linoleic acid, cinnamic acid, methyl stearate, β -belladonnine, apotropine, borneol, 3 α -tigloyloxytropan, and ethyl butanoate.

Withanolides

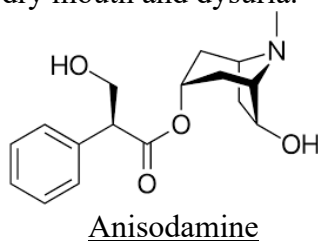
At least 300 naturally occurring steroids with an ergostane structure are classified as withanolides (Glotter, 1991), (Kirson & Glotter, 1981) They are primarily found as secondary metabolites in Nightshade taxa, such as tomatillo. Withanolides are structurally composed of a steroid backbone attached to a lactone or one of its derivatives, and they are formed through steroid oxidation. It is uncertain why withanolides are created; they may serve as a deterrent to feeding insect larvae and other herbivores. Between August 1996 and March 2010, 360 novels naturally occurring withanolides were identified and isolated. A thorough overview of the distribution and structural classification of these novel compounds was given by (Chen et al., 2011) This review article also discusses the various biological and pharmacological actions of natural withanolides based on their structural characteristics. This evaluation cites 211 references in all. (Suleiman et al., 2010) conducted a study in which they isolated two new withanolides, namely mandragorolide A and mandragorolide B, alongside five known withanolides: larnaxolide A, withanolide B, withanicandrin, salpichrolide C, and datura lactone 2. These compounds were extracted from a methanol extract of the whole plant of *Mandragora officinarum*, which was sourced from Jordan. Compound 3 from *Larnax glabra* leaves has only been recorded once. According to the authors, withanolides of various biogenetic kinds from the genus *Mandragora* have never been reported before. This investigation also succeeded in isolating known fatty compounds, coumarins, sterols, and tropane alkaloids.





Toxicity of *Mandragora* Species

Mandragora species, like many others in the Solanaceae family, contain highly physiologically active alkaloids, particularly tropane alkaloids (Fatur & Kreft, 2020). More than 80 compounds have been found; their report provides comprehensive chemical structures for 37 of them. The various components of the plant exhibit differing levels of alkaloid concentrations, with the roots being identified as having the highest levels. The alkaloids found within the plant include atropine, apoatropine, belladonnine, hyoscyamine, cuscohygrine, scopolamine, 3 α ,6 β -ditigloyloxytropine and 3 α -tigloyloxytropine, (Jackson & Berry, 1979), (Ungricht et al., 1998). Additionally, the species *M. caulescens* and *M. turcomanica* have been noted to contain anisodamine (Fatur & Kreft, 2020), (Zhang et al., n.d.). Anisodamine is a naturally occurring tropane alkaloid and anticholinergic drug that is primarily used in traditional and modern Chinese medicine to treat a variety of circulatory disorders and smooth muscle spasms. This can cause a different side effects, including blurred vision, bradycardia, dry mouth and dysuria.



Mandragora species are notable for their high content of physiologically active alkaloids, specifically tropane alkaloids, which contribute to the toxic nature of the plant—particularly its root and leaves—due to their anticholinergic, hallucinogenic, and hypnotic effects. The anticholinergic properties increase the risk of asphyxiation, and consumption of mandrake root can lead to accidental poisoning, as well as side effects like vomiting and diarrhea. Alkaloid levels vary among plant samples, with clinical reports indicating that intake of Mediterranean mandrake can lead to severe symptoms akin to atropine poisoning. These symptoms may include blurred vision, mydriasis (pupil dilation), dry mouth, difficulty in urination, dizziness, headache, vomiting, blushing, and tachycardia (fast heartbeat). Furthermore, hyperactivity and hallucinations are common among affected individuals (Hanuš et al., 2005), (Jiménez-Mejías et al., 1990). The root possesses both psychedelic and

narcotic properties, capable of inducing unconsciousness at suitable concentrations, leading to its historical use as an anesthetic in surgeries. In the past, juice from the finely grated root was applied externally to relieve rheumatic pain and taken internally to address conditions such as depression, convulsions, and insanity. However, excessive consumption was believed to result in insanity and madness (Piccillo et al., 2002). According to (Beyer et al., 2009) various plants contain toxic alkaloids that pose significant risks to human health. The study highlights several alkaloids commonly associated with both fatal and non-fatal poisonings, including aconitine, atropine, coniine, colchicine, cytosine, dimethyltryptamine, harmine, harmaline, ibogaine, kava, mescaline, scopolamine, and taxine. The authors provide a comprehensive review of the symptoms associated with intoxications caused by these substances, as well as an examination of the methods utilized for identifying toxic components in biological fluids.

Mandragora (mandrake) species are known to contain toxic tropane alkaloids, specifically hyoscyamine and/or scopolamine. In cases of poisoning from mandrake, it is essential to identify these tropane alkaloids in various sources, including foods, plant parts, and biological samples. (John, 2012) discusses the importance of determining Tropane alkaloids (TA) for pharmacokinetic, biotransformation, and toxicological studies, emphasizing the use of LC-MS techniques. The document reviews various LC-MS methods for 19 natural and synthetic TAs, providing pharmacological information and detailing how physico-chemical properties influence chromatographic separation and sample preparation methods. It includes a statistical analysis of mass analyzers and scan modes post-ionization and highlights MS/MS techniques for metabolite identification. The applications of TA analysis in pharmacokinetics, distribution, and toxicology are organized into different sectors, concluding with perspectives on future advancements in LC-MS for clinical pharmacology. (Romera-Torres et al., 2018) provide a thorough overview of the analytical methods employed to determine tannins (TAs) and calystegines, detailing the extraction and clean-up processes, chromatographic separation techniques, and mass spectrometry (MS) detection. The review also addresses the identification of these compounds within food and plant raw materials. The review also examined bibliographies from 2000 focused on calystegines and from 2010 concerning TAs, in light of the most recent evaluations.

Mandrake, specifically the species *Mandragora autumnalis* and *Mandragora officinarum*, is noted for containing calystegines. These compounds are polyhydroxylated nor tropane alkaloids that can be found in the plant's roots, leaves, and fruits. (Dräger, 2002) presents a detailed overview of methodologies for the chromatographic separation and analysis of tropane alkaloids. This encompasses medicinal compounds such as tropic acid esters, specifically hyoscyamine and scopolamine, along with cocaine, its derivatives, metabolites, and the degradation products present in plant material. The review incorporates various alkaloids including calystegines, anatoxins, pelletierines, and pseudopelletierines, highlighting their isomeric structures. Each analytical method—High-Performance Liquid Chromatography, Gas Chromatography, Capillary Electrophoresis, and Thin Layer Chromatography—is examined for its benefits in plant analysis, alongside a summary of the instrumentation and methodology utilized for the analysis of tropane alkaloids. The review by (Christen et al., 2007) examines the analysis of tropane alkaloids in biological matrices, detailing various techniques utilized in this domain. These techniques include GC, HPLC, CE, desorption electrospray ionization mass spectrometry, solid-phase micro extraction, pressurized solvent extraction, microwave-assisted extraction and supercritical fluid extraction, the review emphasizes the analysis of both plant material and biological matrices. (Steenkamp, 2005) has detailed the chemical analysis of toxic and medicinal plants with

forensic significance in South Africa, focusing on analytical methods for the phytochemical study of these plants. In addition, (Goutam & Goutam, 2006) have explored various analytical techniques used to identify the phytoconstituents of several common poisonous plants found in India. Tropane alkaloids can be detected and identified in various matrices including food, plant parts, and biological samples through diverse toxicological analysis methods. (Chen et al., 2017), (Jakabová et al., 2012) In cases of suspected poisoning, these biological samples are analyzed to ascertain the type and level of tropane alkaloids present. The primary techniques employed encompass CE, UV spectroscopy, GC, MS, and HPLC. Given the advancements addressing alkaloid instability, liquid chromatography-mass spectrometry (LC-MS) is now favored over gas chromatography-mass spectrometry (GC-MS) due to its superior accuracy in analytical results (Beyer et al., 2009). It is recommended that tissue samples from the stomach be analysed in cases of scopolamine poisoning because the amount of the medication increases significantly in the stomach (Lusthof et al., 2017)

(Namera et al., 2002) introduced a streamlined technique for quantifying tropane alkaloids in biological samples such as urine and serum using gas chromatography-mass spectrometry (GC-MS) and an Extrelut column. The authors assert that this method is more straightforward and quicker than earlier approaches and has practical applications in routine toxicological analysis for suspected tropane alkaloid poisoning, as evidenced by its use in an actual poisoning case. (Gravador et al., 2024) conducted a systematic review of tropane alkaloid (TA) poisoning reports and analytical techniques for detecting these toxins in food crops from 2013 to 2023. The review concludes with discussion on emerging technologies like fast spectroscopy, electrophoresis, and colorimetric methods, as well as the potential integration of smartphone connectivity for on-farm detection and the challenges encountered in implementing these innovations. (Adamse et al., 2014) explored unintentional tropane alkaloid poisoning routes in humans, identifying contamination and mislabelling as major causes. Contamination often arises from mixing toxic plant parts with edible ones, especially roots and seeds. Intentional ingestion occurs through recreational and medical use, resulting in overdoses and even homicides. Animal-derived food products generally do not contribute significantly to exposure. Despite various analytical techniques available for monitoring tropane alkaloids in food, regulatory limits are lacking, and hazardous dosages remain unclear due to insufficient data. Foods potentially containing these alkaloids include herbal teas, medicines, berries, edible flowers, and certain beans and seeds.

Some reported cases of Mandrake poisoning

1. Two cases of accidental ingestion of *Mandragora Officinarum* were reported within a Greek family, involving a 47-year-old woman and a 48-year-old man, both hospitalized in Crete. They exhibited symptoms such as nausea, dizziness, blurred vision, dry mouth, headache, unstable walking, and vomiting. The male patient identified the plant consumed during a meal, which was later confirmed through an online search to be *Mandragora Officinarum*, mistakenly taken instead of the edible *Borago Officinalis*. Vital signs upon admission were generally stable, but both patients displayed tachycardia, with the male's pulse at 190 bpm and the female's at 170 bpm, along with increased respiratory rates. ECG readings indicated supraventricular tachycardia in both patients, raising significant concerns regarding the physiological effects of the ingested toxic plant. (Tsiligianni et al., 2009)

2. A 69-year-old male presented to the emergency department with confusion and severe agitation. Vital signs showed tachycardia (130 bpm), elevated temperature (38.3 °C), and hypertension (160/100 mmHg). Physical examination revealed dry skin and dilated, light-unresponsive pupils, alongside a severely distended bladder indicative of urinary retention,

suggesting anticholinergic syndrome. After treatment within 36 hours of monitoring, the patient was asymptomatic and discharged in good health (Bildik et al., 2011).

3. (Piccillo et al., 2006) from Cannizzaro Hospital in Catania, Italy, reported a case of poisoning in a 72-year-old woman who mistakenly ingested *Mandragora autumnalis*, a toxic plant. The patient thought she was consuming the edible *Borago officinalis*, both of which are found in the Sicilian countryside and can be easily confused. The diagnosis was made based on her clinical symptoms indicative of anticholinergic syndrome and her history of consuming vegetables of uncontrolled origin. Gastric lavage analysis confirmed the presence of the poisonous vegetable. The patient underwent decontamination and received symptomatic treatment, which successfully alleviated her acute symptoms of poisoning.

4. A case of unusual poisoning from mandrake fruit involved a 55-year-old male tourist who consumed 10 fruits on Dilos was reported by (Poulos & Vlachos, 1982) Approximately 30 minutes after ingestion, victim experienced visual disturbances and heightened anxiety. Examined an hour later, he showed tachycardia (160 bpm), dilated pupils, and facial flushing.

5. A case of unintentional poisoning due to the consumption of "aphrodisiac" berries is detailed by (Nikolaou et al., 2012) involving a 35-year-old male who exhibited clinical signs of anticholinergic syndrome upon hospital admission. The toxicological analysis of the patient's urine was pivotal in diagnosing the poisoning, which was confirmed when his girlfriend disclosed that he had intentionally ingested the berries to enhance his sexual performance. Further lab analysis identified the berries as *Mandragora officinarum*, revealing the presence of hyoscyamine and scopolamine in both the urine and the berries consumed. Treatment for the poisoning included decontamination and symptomatic therapy, resulting in the patient's full recovery after a four-day hospital stay. The authors conclude that this case exemplifies the clinical manifestations of *Mandragora* intoxication and underscores the critical role of analytical toxicology in diagnosing intoxications related to unidentified plant substances.

6. (Frasca et al., 1997) reported a case of mistaken identity involving the mandrake plant, which resulted in toxicity. A 31-year-old man presented to the emergency department with acute nausea and vomiting after ingesting an unknown amount of what he believed to be *Mandragora officinarum*, a plant known for its anticholinergic and hallucinogenic properties. However, chromatographic testing indicated that the consumed plant was actually *Podophyllum peltatum*. The patient was hospitalized for observation overnight and ultimately recovered without any significant systemic effects.

7. (Gönül & Çakmak, 2013) reported a case of allergic skin reaction to *Mandragora radix* involving a thirty-two-year-old male patient who experienced erythema and pruritus on his forearm. The symptoms appeared three days before his clinic visit, while similar lesions had begun 10-12 days earlier after contact with the plant's sap. Interestingly, the patient's sister had used the sap for joint pain without adverse effects. Curious about its use, he applied the sap, resulting in immediate lesions characterized by pruritus, redness, and swelling, which resolved within 2-3 days after treatment with a topical cream. The patient had no prior exposure to *Mandragora radix* or its sap and reported no history of allergies. However, the lesions reappeared after 10 days without additional sap application. Dermatological examination revealed an irregularly bordered, erythematous plaque extending from the wrist to the inner forearm, accompanied by papules that occasionally coalesced and mild desquamation. The diagnosis was allergic contact dermatitis linked to *Mandragora radix*.

8. (Jiménez-Mejías et al., 1990) reported 15 cases of Anticholinergic syndrome (AS) attributed to *Mandragora autumnalis* poisoning, grouping patients into two families. The onset of symptoms following ingestion ranged from 1 to 4 hours, with a mean duration of 2.7 hours. Clinical manifestations varied in severity; all patients exhibited impaired vision and dry mouth, while 60% each experienced urinary difficulties, dizziness, and headaches. Additionally, over half (53%) had vomiting, with two patients reporting difficulty swallowing and abdominal pain. Common symptoms included blushing, active mydriasis, tachycardia, dry skin, and mucosae in 93% of cases, alongside neuropsychiatric effects such as hyperactivity or hallucinations in 14, and agitation or delirium in 60%. The toxicity was traced back to *Mandragora* detected in chard (*Beta vulgaris*) and spinach (*Spinacia oleracea*), alongside known toxic alkaloids atropine and hiosciamine.

Conclusion:

The review summarizes the current knowledge of the *Mandragora* plant's chemistry, covering various aspects such as its ethnobotanical characteristics, associated myths, traditional medicinal uses, roles in folklore medicine, presence of phytochemicals, and potential toxicity. These species hold historical significance and have a complex chemical composition, which contributes to their recreational and narcotic characteristics. The plant is traditionally recognized for various properties including magical, aphrodisiac, healing, hallucinogenic, and toxic effects. Its current consumption trends present serious safety and public health concerns. The article provides an overview on the phytoconstituents found in *Mandragora*, encompassing their isolation and identification through various methods. It details a range of phytoconstituents, including Alkaloids, flavonoids essential oil, carotenoids, phenolic compounds, coumarins, withanolides, as well as lipids and lipid-like substances. Additionally, discusses toxicological studies pertaining to different *Mandragora* species, including documented cases of both intentional and unintentional poisoning instances. While the existing literature on this topic is limited, this work contributes significantly to the field, highlighting the need for further research to explore new aspects of *Mandragora* and its effects. The conclusion stresses the importance of ongoing investigations to enhance the understanding and safety of these plants. Literature indicates that due to its potent anticholinergic alkaloids (atropine, scopolamine, and hyoscyamine), *Mandragora* (*Mandrake*, *Mandragora officinarum/autumnalis*) is used cautiously in traditional systems, including Ayurveda and Arabian medicine, mainly for pain, insomnia, and spasms; however, it is extremely toxic and needs to be handled carefully. Herbal drugs are gaining popularity across both developed and developing nations due to their safety and better tolerance compared to allopathic medications. This trend reflects a growing preference for natural remedies among consumers seeking alternatives to conventional pharmaceuticals. Yet unchecked dosage can be hazardous. Further research is required to better understand the pharmacological effects and toxicological profile of these plant in order to assure the safety of persons who may be tempted to use it recreationally and an aid to clinical toxicology as well as to forensics. It is anticipated that this publication will illustrate its significance in the field of forensics, serving as a resource for scholars interested in recognizing future research avenues related to *Mandragora* and its various phytoconstituents.

References

1. Adamse, P., et al. (2014). Tropane alkaloids in food: Poisoning incidents. *Quality Assurance and Safety of Crops & Foods*, 6(1), 15–24. <https://doi.org/10.3920/QAS2013.0314>

2. Akhiani, H., & Ghorbani, A. B. (2003). *Mandragora turcomanica* (Solanaceae) in Iran: A new distribution record for an endangered species. *Systematics and Biodiversity*, 1(2), 177–180. <https://doi.org/10.1017/S1477200003001105>
3. Al-Khalil, S., & Alkofahi, A. (1996). The chemical constituents of *Mandragora autumnalis*. *Alexandria Journal of Pharmaceutical Sciences*, 10, 135–138.
4. Al-Maharik, N., et al. (2022). Isolation, identification and pharmacological effects of *Mandragora autumnalis* fruit flavonoids fraction. *Molecules*, 27(3), 1046. <https://doi.org/10.3390/molecules27031046>
5. Albahri, G., et al. (2025). *Mandragora autumnalis*: Phytochemical composition, antioxidant and anti-cancerous bioactivities on triple-negative breast cancer cells. *International Journal of Molecular Sciences*, 26(17), 8506.
6. Arbia, F., et al. (2019). Profiles of the essential oils and headspace analysis of volatiles from *Mandragora autumnalis* growing wild in Tunisia. *Chemistry & Biodiversity*, 16(10), e1900345.
7. Asilbekova, D. T., & Glushenkova, A. I. (1998). Lipids of the seeds and flesh of the fruit of *Mandragora turcomanica*. *Chemistry of Natural Compounds*, 34(6), 664–667.
8. Asilbekova, D. T., & Glushenkova, A. I. (1999). Triacylglycerols of the seeds and fruit flesh of *Mandragora turcomanica*. *Chemistry of Natural Compounds*, 35(3), 291–294.
9. Asilbekova, D. T., et al. (1994). Lipids of the tuberous roots of *Mandragora turcomanica*. *Chemistry of Natural Compounds*, 30(5), 566–572.
10. Ask Ayurveda. (n.d.). *Mandragora autumnalis*. <https://ask-ayurveda.com/wiki/article/5022-mandragora-autumnalis>
11. Baser, K. H. C., et al. (1998). Composition of the essential oil and the headspace sample of *Mandragora autumnalis* fruits. *Journal of Essential Oil Research*, 10(6), 632–634.
12. Bekkouche, K., et al. (2001). Calystegine distribution in some solanaceous species. *Phytochemistry*, 58(3), 455–462.
13. Beyer, J., et al. (2009). Analysis of toxic alkaloids in body samples. *Forensic Science International*, 185(1–3), 1–9. <https://doi.org/10.1016/j.forsciint.2008.12.006>
14. Bildik, F., et al. (2011). Two herbal poisoning cases from Turkey. *Journal of Academic Emergency Medicine Case Reports*, 2, 0–0. <https://doi.org/10.5505/jaemcr.2011.07279>
15. Carter, A. J. (2003). Myths and mandrakes. *Journal of the Royal Society of Medicine*, 96, 144–147.
16. Chen, H., et al. (2017). Simultaneous determination of atropine and scopolamine using LC–MS/MS. *Food Chemistry*, 218, 173–180. <https://doi.org/10.1016/j.foodchem.2016.09.075>
17. Chen, L. X., et al. (2011). Natural withanolides: An overview. *Natural Product Reports*, 28(4), 705–740.
18. Christen, P., et al. (2007). Analysis of tropane alkaloids in biological matrices. In *Analysis of Alkaloids*. Wiley. <https://doi.org/10.1002/9783527621071.ch12>
19. Dafni, A., et al. (2021). In search of traces of the mandrake myth. *Journal of Ethnobiology and Ethnomedicine*, 17(1), 68. <https://doi.org/10.1186/s13002-021-00494-5>
20. Dräger, B. (2002). Analysis of tropane and related alkaloids. *Journal of Chromatography A*, 978(1–2), 1–35.
21. Dräger, B., et al. (1994). Calystegines as a new group of tropane alkaloids. *Plant Cell, Tissue and Organ Culture*, 38(2), 235–240.
22. Dräger, B., et al. (1995). Distribution of calystegines in Solanaceae. *Planta Medica*, 61(6), 577–579.
23. Fakir, H., & Özçelik, H. (2009). *Mandragora officinarum* L.: A new record for Turkey. *African Journal of Biotechnology*, 8(15), 3560–3564.
24. Fatur, K., & Kreft, S. (2020). Anticholinergic solanaceous plants of temperate Europe. *Toxicon*, 177, 52–88. <https://doi.org/10.1016/j.toxicon.2020.02.005>
25. Fleisher, A., & Fleisher, Z. (1994). The fragrance of Biblical mandrake. *Economic Botany*, 48, 243–251.

26. Fleisher, Z., & Fleisher, A. (1992). The odoriferous principles of mandrake. *Journal of Essential Oil Research*, 4(2), 187–188.
27. Forest Service, U.S. Department of Agriculture. (n.d.). *The powerful Solanaceae: Mandrake*.
28. Frasca, T., et al. (1997). Mandrake toxicity: A case of mistaken identity. *Archives of Internal Medicine*, 157(17), 2007–2009.
29. Glotter, E. (1991). Withanolides and related ergostane-type steroids. *Natural Product Reports*, 8(4), 415–440.
30. Goutam, M. P., & Goutam, S. (2006). *Analysis of plant poisons*. Selective & Scientific Books.
31. Gravador, R. S., et al. (2024). Tropane alkaloid poisonings and analytical techniques. *Comprehensive Reviews in Food Science and Food Safety*, 23(6), e70047.
32. Gusakova, S. D., & Asilbekova, D. T. (1997). Triacylglycerols of *Mandragora turcomanica*. *Chemistry of Natural Compounds*, 33(4), 441–444.
33. Hanuš, L. O., et al. (2005). Substances isolated from *Mandragora* species. *Phytochemistry*, 66(20), 2408–2417. <https://doi.org/10.1016/j.phytochem.2005.07.016>
34. Hesse, O. (1901). Über die Alkaloide der Mandragorawurzel. *Journal für Praktische Chemie*, 64, 274–286.
35. Hill, D. (2025). *The magic of the mandrake*. Praxis-Knowledge.
36. Integrated Taxonomic Information System (ITIS). (n.d.). *Mandragora* L. Taxonomic Serial No. 500625.
37. Jackson, B. P., & Berry, M. I. (1973). Hydroxytropane tiglates in *Mandragora* roots. *Phytochemistry*, 12(5), 1165–1166.
38. Jackson, B. P., & Berry, M. I. (1979). *Mandragora: Taxonomy and chemistry of European species*. Academic Press.
39. Jiménez-Mejías, M. E., et al. (1990). Atropine poisoning by *Mandragora autumnalis*. *Medicina Clínica*, 95(18), 689–692.
40. John, H. (2012). LC–MS analysis of tropane alkaloids. Springer.
41. Jodallah, N. B. E. (2013). *Antioxidant and antimicrobial activity of Mandragora autumnalis* (Doctoral dissertation).
42. Karakaş, H. H., & Dörtbudak, M. Y. (2023). Toxic effect of *Mandragora autumnalis* on *Daphnia magna*. *Applied Ecology & Environmental Research*, 21(6).
43. Kinney, S. C. (2022). *The origins of the Herbarium of Pseudo-Apuleius* (Doctoral dissertation).
44. Kirson, I., & Glotter, E. (1981). Ergostane-type steroids. *Journal of Natural Products*, 44(6), 633–647.
45. Kohnen-Johannsen, K. L., & Kayser, O. (2019). Tropane alkaloids. *Molecules*, 24(4), 796.
46. Koleva, I. I., et al. (2012). Alkaloids in the human food chain. *Molecular Nutrition & Food Research*, 56(1), 30–52.
47. Lounasmaa, M., & Tamminen, T. (1993). Tropane alkaloids. Elsevier.
48. Lusthof, K. J., et al. (2017). Scopolamine-facilitated robberies. *Forensic Science International*, 274, 79–82.
49. Mahmod, A. I., & Talib, W. H. (2021). Anticancer activity of *Mandragora autumnalis*. *Pharmacia*, 68(4), 827–833.
50. Mahmod, A. I., & Talib, W. H. (2023). Chemical composition and immunomodulatory activity. *Natural Products Journal*, 13(3), 72–83.
51. Monadi, T., et al. (2021). Review on *Mandragora* genus. *Current Pharmaceutical Design*, 27(34), 3609–3637.
52. Namera, A., et al. (2002). Quantitative analysis of tropane alkaloids. *Forensic Science International*, 130(1), 34–43.
53. Nikolaou, P., et al. (2012). Accidental poisoning after ingestion of berries. *Journal of Emergency Medicine*, 42(6), 662–665.

54. Pakhlavouni, I., et al. (2000). Carotenoid composition of *Mandragora turcomanica*. *Russian Journal of Plant Physiology*, 47(1), 38–47.
55. Phillipson, J. D., & Handa, S. S. (1975). N-oxides of hyoscyamine and hyoscyne. *Phytochemistry*, 14(4), 999–1003.
56. Piccillo, G. A., et al. (2002). Six clinical cases of poisoning. *European Journal of Emergency Medicine*, 9(4), 342–347.
57. Piccillo, G. A., et al. (2006). Anticholinergic syndrome. *International Journal of Clinical Practice*, 60(4), 492–494.
58. Poulos, L., & Vlachos, P. (1982). A mandrake poisoning case. *Clinical Toxicology*, 19(5), 521–522.
59. Razzakov, N. A., et al. (1998). Alkaloids of *Mandragora turcomanica*. *Chemistry of Natural Compounds*, 34(6), 741–742.
60. Romera-Torres, A., et al. (2018). Analytical methods for tropane alkaloids. *Journal of Chromatography A*, 1564, 1–15.
61. Staub, H. (1942). Non-alkaloid constituents of mandrake root. *Helvetica Chimica Acta*, 25(4), 649–683.
62. Staub, H. (1962). Chemical constituents of mandrake root. *Helvetica Chimica Acta*, 45(7), 2297–2305.
63. Steenkamp. (2005). *Chemical analysis of medicinal and poisonous plants* (Doctoral dissertation).
64. Suleiman, R. K., et al. (2010). New withanolides from *Mandragora officinarum*. *Fitoterapia*, 81(7), 864–868.
65. Thoms, H., & Wentzel, M. (1898). Über mandragorin. *Berichte der Deutschen Chemischen Gesellschaft*, 31(2), 2031–2037.
66. Thoms, H., & Wentzel, M. (1901). Über die Basen der Mandragorawurzel. *Berichte der Deutschen Chemischen Gesellschaft*, 34(1), 1023–1026.
67. Tsiligianni, I. G., et al. (2009). Mandragora poisoning in Crete. *Cases Journal*, 2, 9331.
68. Tu, T., et al. (2010). Biogeographic dispersals of Solanaceae. *Molecular Phylogenetics and Evolution*, 57(3), 1226–1237.
69. Ungricht, S., et al. (1998). Revision of *Mandragora*. *Bulletin of the Natural History Museum (Botany)*, 28(1), 17–40.
70. United States Department of Agriculture, Forest Service. (n.d.). *The powerful Solanaceae: Mandrake*.
https://www.fs.usda.gov/wildflowers/ethnobotany/Mind_and_Spirit/mandrake.shtml
71. Volis, S., et al. (2018). Evolutionary history of *Mandragora*. *Molecular Phylogenetics and Evolution*, 129, 85–95.
72. Volis, S., et al. (2019). Erratum: Evolutionary history and biogeography of *Mandragora* L. (Solanaceae). *Molecular Phylogenetics and Evolution*, 130, 434.
<https://doi.org/10.1016/j.ympev.2018.11.008>
73. Woisky, R. G., & Salatino, A. (1998). Analysis of propolis. *Journal of Apicultural Research*, 37(2), 99–105.
74. Woisky, R. G., & Salatino, A. (1998). Analysis of propolis: Some parameters and procedures for chemical quality control. *Journal of Apicultural Research*, 37(2), 99–105.
75. Wright, S. H., & Klassen, F. (2002). *Everyday magicians*. Penn State University Press.
76. Zhang, Z.-Y., et al. (n.d.). *Mandragora caulescens*. *Flora of China*.

34. DNA Barcoding and Genomic Tools in Wildlife Crime Detection

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Abstract: Wildlife crime, encompassing poaching, illegal trade, and exploitation of protected flora and fauna, represents a major transnational crime that threatens biodiversity, ecological balance, and sustainable development. Conventional morphological identification of wildlife specimens is often inadequate in forensic investigations due to degradation, processing, or fragmentation of biological material. DNA barcoding and advanced genomic tools have emerged as robust, reproducible, and legally defensible techniques for species identification, individualization, population assignment, and geographic origin determination. This review comprehensively examines the principles and applications of DNA barcoding, mitochondrial and nuclear genetic markers, and next-generation sequencing (NGS) technologies in wildlife crime detection. Emphasis is placed on forensic workflows, validation requirements, admissibility in courts of law, and selected national and international case studies. Limitations, ethical concerns, and prospects of genomic wildlife forensics are also discussed, highlighting the pivotal role of molecular techniques in strengthening wildlife law enforcement and conservation strategies.

Keywords: Wildlife forensics; DNA barcoding; Genomic tools; Wildlife crime; Species identification; CITES

Introduction

Wildlife crime is one of the most lucrative forms of transnational organized crime, ranking alongside drug trafficking, arms smuggling, and human trafficking (UNODC, 2020). It includes illegal hunting, poaching, trafficking of animal parts, timber smuggling, and trade in endangered species protected under national and international legislations such as the Wildlife (Protection) Act, 1972 (India) and the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES). India, being a biodiversity-rich country, faces significant challenges related to wildlife crime involving species such as tiger (*Panthera tigris*), elephant (*Elephas maximus*), rhinoceros (*Rhinoceros unicornis*), pangolin, and various protected birds and reptiles (Jain et al., 2021; TRAFFIC India, 2018). The increasing demand for wildlife products such as ivory, rhino horn, tiger bones, exotic pets, and medicinal derivatives has intensified pressure on vulnerable species and threatens conservation efforts in protected areas and wildlife corridors across India (Karanth & Defries, 2010).

From a forensic perspective, wildlife crime presents unique challenges. Evidence recovered at crime scenes is frequently degraded, processed, or transformed into powders, ornaments, medicines, leather goods, or food products, rendering morphological identification unreliable or impossible. In India, seizures often involve traditional medicines, religious artefacts,

bushmeat, or processed wildlife derivatives, complicating conventional identification (Sahajpal et al., 2010). In such contexts, **molecular biology-based approaches** provide objective and scientifically robust means of species identification and source attribution.

DNA barcoding and genomic tools have revolutionized wildlife forensics by enabling accurate species identification from minute or degraded biological samples. These techniques are increasingly used by Indian enforcement agencies such as the **Wildlife Crime Control Bureau (WCCB)** and State Forest Departments to link seized materials to protected species, establish individual identity, trace geographic origin, and support prosecutions with court-admissible scientific evidence (Sahajpal et al., 2011; Singh et al., 2014). This review aims to synthesize current knowledge on DNA barcoding and genomic technologies in wildlife crime detection, with emphasis on forensic applicability, validation, and future directions in the Indian legal and conservation framework.

2. Wildlife Forensics: Scope and Challenges

Wildlife forensics is an interdisciplinary field that applies scientific techniques to investigate crimes involving protected flora and fauna. It plays a crucial role in enforcing wildlife protection laws, identifying illegally traded species, and supporting conservation policies (Alacs & Georges, 2008; Ogden, 2008). In India, wildlife forensic investigations support the enforcement of the Wildlife (Protection) Act, 1972, and assist courts in adjudicating wildlife offence cases (Sahajpal et al., 2010). Traditional wildlife identification methods rely on morphological, anatomical, or histological characteristics. However, forensic casework frequently involves compromised samples such as dried meat, bones, hair, feathers, powders, traditional medicines, and ornamental artefacts. These materials often lack distinctive morphological features, making species-level identification unreliable or impossible (Linacre & Tobe, 2013).

Additionally, wildlife crime evidence in India is commonly exposed to environmental degradation, chemical treatment, or deliberate processing to evade detection, particularly in organized trafficking networks (TRAFFIC India, 2018). Such factors further reduce the effectiveness of classical taxonomic approaches and increase the risk of misidentification, highlighting the need for molecular confirmation.

3. Role of Molecular Forensics

Molecular forensic approaches overcome many of these limitations by targeting genetic material that persists even in degraded samples. DNA-based techniques provide high sensitivity, specificity, and reproducibility, enabling species identification across life stages and tissue types (Dawnay et al., 2007). In the Indian context, mitochondrial markers such as COI, cytochrome b, and 12S rRNA **genes** have been successfully employed for species identification in cases involving big cats, ungulates, and seized wildlife derivatives (Sahajpal et al., 2011; Singh et al., 2014). As a result, molecular forensics has become a cornerstone of modern wildlife crime investigation and prosecution, strengthening scientific credibility and legal robustness of wildlife conservation efforts in India.

3.1 DNA Barcoding: Principles and Methodology

DNA barcoding is a standardized molecular approach that uses short, species-specific DNA sequences to enable rapid and accurate biological identification. Proposed by Hebert et al.

(2003), this method is based on the principle that interspecific genetic variation exceeds intraspecific variation for selected gene regions.

3.2 Commonly Used Barcode Markers

For animal species, the mitochondrial cytochrome c oxidase subunit I (COI) gene is the most widely accepted barcode marker due to its high amplification success, lack of recombination, and sufficient sequence variability (Hebert et al., 2003; Ratnasingham & Hebert, 2007). In plants, chloroplast genes such as *rbcL* and *matK* are commonly employed, while the internal transcribed spacer (ITS) region is used for fungi (Hollingsworth et al., 2011).

3.3 The standard **DNA barcoding workflow** follows a systematic and reproducible sequence of steps designed to ensure reliable species identification. It begins with **sample collection** from wildlife materials such as tissue, hair, bone, scales, or processed products, followed by **DNA extraction** using validated forensic protocols. The extracted DNA is then subjected to **polymerase chain reaction (PCR) amplification** targeting standardized barcode regions (e.g., *COI* gene). The amplified products are **sequenced**, and the resulting DNA sequences are compared with authenticated reference libraries to assign species identity (Dawnay et al., 2007).

Sample → DNA Extraction → PCR Amplification → Sequencing → Database Comparison → Species Identification

The accuracy and forensic value of DNA barcoding depend largely on the availability and quality of **reference databases**. The **Barcode of Life Data System (BOLD)** provides curated barcode records linked to taxonomically verified voucher specimens, making it particularly useful for forensic species identification. **GenBank**, a global nucleotide sequence repository, also supports sequence comparison but may include unverified or misidentified entries. Despite their utility, **gaps in reference coverage**, especially for rare, endemic, and endangered species, remain a major limitation in wildlife forensic investigations and may affect the conclusiveness of results (Ogden & Linacre, 2015).

4. Genomic Tools in Wildlife Crime Detection

Genomic tools are widely used in wildlife forensics for **species identification, individualization, and origin determination** of seized biological samples. **DNA barcoding**, particularly using the mitochondrial *COI* gene, enables accurate species identification even from degraded or processed wildlife products (Dawnay et al., 2007; Ogden, 2008). **Microsatellite markers and SNPs** provide high discriminatory power for individual identification and population assignment, which is critical in linking seized materials to specific animals or regions (Wasser et al., 2015). Recent advances such as **Next-Generation Sequencing (NGS)** further enhance analytical accuracy and support large-scale wildlife crime investigations (Alacs & Georges, 2008). Standardized forensic guidelines for the use of non-human DNA ensure reliability and admissibility of genomic evidence in courts (Linacre et al., 2011).

4.1 Mitochondrial and Nuclear Markers

Mitochondrial DNA (mtDNA) is widely used in wildlife forensic investigations due to its high copy number, maternal inheritance, and resistance to degradation. These characteristics

make mtDNA particularly valuable for **species identification** and **maternal lineage analysis**, especially when working with highly degraded or processed samples such as skins, bones, and scales (Dawnay et al., 2007; Ogden, 2008).

Microsatellites (Short Tandem Repeats, STRs) are highly polymorphic **nuclear DNA markers** that provide strong discriminatory power for **individualization, kinship analysis, and population structure studies**. In wildlife crime cases, microsatellites are frequently used to link seized samples to specific individuals or populations, thereby strengthening forensic inferences and legal evidence (Wasser et al., 2015; Linacre et al., 2011).

Single Nucleotide Polymorphisms (SNPs) are abundant and stable genetic markers distributed throughout the genome. Their suitability for **high-throughput and automated analysis** makes them particularly useful for large-scale wildlife forensic investigations, including population assignment and geographic origin tracing, especially when combined with next-generation sequencing platforms (Ogden & Linacre, 2015).

4.2 Next-Generation Sequencing (NGS)

Next-Generation Sequencing (NGS) allows the high-throughput sequencing of millions of DNA fragments simultaneously. This technology facilitates accurate species identification from mixed or highly degraded samples, supports population-level assignment and geographic origin determination, and aids in uncovering patterns of illegal wildlife trade. Advanced NGS-based approaches, including metabarcoding and whole-genome sequencing, are increasingly being adopted in wildlife forensic laboratories.

5. Applications in Wildlife Crime Investigation

5.1 Species Identification- DNA barcoding has been successfully used to identify protected species from meat, skin, bone, and hair samples in cases involving tigers, leopards, elephants, pangolins, and marine species.

5.2 Individualization and Population Assignment- Microsatellite and SNP analyses enable individual identification, which is critical in linking seized animal parts to specific carcasses or crime scenes.

5.3 Geographic Origin Determination- Genomic signatures can reveal the population or regional origin of seized specimens, aiding in understanding poaching hotspots and trafficking routes.

6. Case Studie- Indian Context

In India, **DNA-based wildlife forensic techniques** have been successfully applied in several high-profile cases involving the illegal trade of protected species. Genetic analysis has been used to identify **tiger (*Panthera tigris*)** and **leopard (*Panthera pardus*) skins**, even when morphological features were altered, thereby strengthening prosecutions under the **Wildlife (Protection) Act, 1972** (Sharma et al., 2013).

Similarly, **DNA profiling of musk deer (*Moschus spp.*) pods** has helped distinguish genuine musk from adulterated or synthetic substitutes, aiding enforcement agencies in curbing illegal trade (Gupta et al., 2012). In recent years, **molecular identification of pangolin scales** using mitochondrial DNA markers has played a key role in confirming species identity in seizures,

supporting convictions in one of the most trafficked mammalian groups in India (Verma & Singh, 2020).

Additionally, genetic tools have been applied in **ivory and wildlife tissue seizures**, enabling species confirmation and linking seized materials to specific taxa, thus providing robust, court-admissible evidence (Iyengar, 2014). These case studies demonstrate the growing importance of genomic tools in Indian wildlife crime investigations and their critical role in conservation-oriented law enforcement.

Conclusion

DNA barcoding and genomic tools have transformed wildlife forensic science from a largely descriptive discipline into a robust, evidence-driven investigative science. The ability to identify species from minute, degraded, or highly processed biological materials has significantly strengthened the investigation and prosecution of wildlife crime cases (Hebert et al., 2003; Dawnay et al., 2007). In situations where traditional morphological and anatomical methods fail, molecular techniques provide objective, reproducible, and scientifically validated results that can withstand judicial scrutiny.

Beyond species identification, the integration of advanced genomic tools such as microsatellites, SNP analysis, and next-generation sequencing has expanded the scope of wildlife forensics to include individualization, population assignment, and geographic origin tracing (Ogden & Linacre, 2015). These capabilities are particularly important for addressing organized wildlife trafficking networks, identifying poaching hotspots, and supporting intelligence-led conservation enforcement, as demonstrated in large-scale ivory seizure investigations (Wasser et al., 2015).

Despite these advancements, challenges remain in the form of incomplete reference databases, lack of standardization across laboratories, high operational costs, and limited technical expertise in developing regions. Addressing these limitations requires international collaboration, expansion of validated reference libraries, capacity building, and the establishment of accredited wildlife forensic laboratories following standardized protocols recommended by international bodies such as CITES and INTERPOL (CITES, 2023).

In conclusion, DNA barcoding and genomic tools are indispensable for modern wildlife crime detection and biodiversity conservation. Their continued development, forensic validation, and legal integration will play a decisive role in strengthening wildlife law enforcement, improving conviction rates, and supporting global efforts to combat eco-crime and protect endangered species for future generations.

References

1. Alacs, E. A., & Georges, A. (2008). Wildlife across our borders: A review of forensic science in wildlife law enforcement. *Forensic Science, Medicine, and Pathology*, 4(2), 80–91. <https://doi.org/10.1007/s12024-008-9024-9>
2. Alacs, E. A., & Georges, A. (2008). Wildlife across our borders: A review of the illegal trade in Australia. *Australian Journal of Forensic Sciences*, 40(2), 147–160.
3. Alacs, E. A., & Georges, A. (2008). Wildlife across our borders: A review of the illegal trade in Australia using molecular genetics. *Australasian Journal of Forensic Sciences*, 40(2), 147–160.

4. CITES. (1973). *Convention on International Trade in Endangered Species of Wild Fauna and Flora*. Washington, DC.
5. Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES). (2023). *CITES handbook*. Geneva, Switzerland.
6. Dawnay, N., et al. (2007). Validation of the barcoding gene COI for use in forensic genetic species identification. *Forensic Science International*, 173(1), 1–6.
7. Dawnay, N., et al. (2007). Validation of species identification by DNA barcoding using forensic samples. *Forensic Science International*, 173(2–3), 153–161. <https://doi.org/10.1016/j.forsciint.2007.02.021>
8. Gupta, S. K., et al. (2012). Molecular identification of musk deer species from musk pods. *Forensic Science International*, 214(1–3), 176–180.
9. Hebert, P. D. N., et al. (2003). Biological identifications through DNA barcodes. *Proceedings of the Royal Society B: Biological Sciences*, 270(1512), 313–321.
10. Hollingsworth, P. M., et al. (2011). A DNA barcode for land plants. *Proceedings of the National Academy of Sciences*, 108(31), 12794–12797.
11. INTERPOL. (2022). *Wildlife crime: A global enforcement priority*. Lyon, France.
12. Iyengar, A. (2014). Forensic DNA analysis for wildlife conservation. *Current Science*, 106(6), 829–838.
13. Jain, P., et al. (2021). Wildlife crime in India: Trends, challenges, and forensic interventions. *Journal of Threatened Taxa*, 13(9), 19234–19242. <https://doi.org/10.11609/jott.XXXX>
14. Karanth, K. K., & DeFries, R. (2010). Conservation and management in human-dominated landscapes: Case studies from India. *Biological Conservation*, 143(12), 2865–2869. <https://doi.org/10.1016/j.biocon.2010.08.002>
15. Linacre, A., et al. (2011). ISFG recommendations regarding the use of non-human DNA in forensic genetic investigations. *Forensic Science International: Genetics*, 5(5), 501–505.
16. Linacre, A., & Tobe, S. S. (2013). An overview to the investigative approach to species testing in wildlife forensic science. *Investigative Genetics*, 4(1), 1–9.
17. Linacre, A., & Tobe, S. S. (2013). *Wildlife DNA analysis: Applications in forensic science*. Wiley-Blackwell.
18. Ogden, R. (2008). Fisheries forensics: The use of DNA tools for improving compliance, traceability and enforcement. *Fish and Fisheries*, 9(4), 462–472.
19. Ogden, R. (2008). Forensic science, genetics and wildlife biology: Getting the right mix for a wildlife DNA forensics lab. *Forensic Science, Medicine, and Pathology*, 4, 172–179.
20. Ogden, R. (2008). Forensic science, genetics and wildlife biology: Getting the right mix for a wildlife DNA forensics lab. *Forensic Science, Medicine, and Pathology*, 4(3), 172–179. <https://doi.org/10.1007/s12024-008-9035-6>
21. Ogden, R., & Linacre, A. (2015). Wildlife forensic science: A review of genetic geographic origin assignment. *Forensic Science International: Genetics*, 18, 152–159.
22. Ratnasingham, S., & Hebert, P. D. N. (2007). BOLD: The Barcode of Life Data System. *Molecular Ecology Notes*, 7(3), 355–364.
23. Sahajpal, V., et al. (2010). Identification of illegally traded wildlife parts and derivatives using DNA-based techniques: Indian perspective. *Forensic Science International: Genetics*, 4(1), 1–8. <https://doi.org/10.1016/j.fsigen.2009.07.002>
24. Sahajpal, V., et al. (2011). Molecular identification of Indian big cats from wildlife crime seizures. *Forensic Science International: Genetics*, 5(5), 501–507. <https://doi.org/10.1016/j.fsigen.2010.11.004>

25. Sharma, R., et al. (2013). Genetic analysis of tiger (*Panthera tigris*) populations from India. *Forensic Science International: Genetics*, 7(4), 425–431.
26. Singh, S. K., et al. (2014). DNA barcoding for wildlife forensics in India: Current status and future prospects. *Indian Journal of Forensic Sciences*, 8(2), 45–52.
27. TRAFFIC India. (2018). *Wildlife trade in India: Scale, dynamics and enforcement challenges*. TRAFFIC International, New Delhi.
28. UNODC. (2020). *World wildlife crime report: Trafficking in protected species*. United Nations Office on Drugs and Crime, Vienna.
29. Verma, S. K., & Singh, L. (2020). DNA-based identification of Indian pangolin (*Manis crassicaudata*) from seized scales. *Science & Justice*, 60(1), 94–101.
30. Wasser, S. K., et al. (2015). Genetic assignment of large seizures of elephant ivory reveals Africa's major poaching hotspots. *Science*, 349(6243), 84–87.
31. Wildlife (Protection) Act, 1972 (India). Government of India, Ministry of Environment, Forest and Climate Change.
32. Wildlife Institute of India. (2019). *Wildlife forensics in India: Applications and case studies*. Dehradun, India.
33. Wasser, S. K., et al. (2015). Genetic assignment of large seizures of elephant ivory reveals Africa's major poaching hotspots. *Science*, 349(6243), 84–87.

35. Gendered Based Online Harrassment: A Review on Virtual Victimization

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Abstract: This review looks at how gender affects people's experiences with cyberbullying and online harassment in digital spaces. The study aims to combine recent research to learn about the patterns, effects, and reactions to online abuse that is based on gender. We looked at articles and reports from criminology, sociology, and digital media studies to find important patterns and ideas by using a detailed review method. The findings show that women and gender minorities experience a lot more targeted abuse, such as threats, insults, and sharing of private content without their permission. This type of bullying often comes from deep-seated beliefs about gender and uneven power, which are made worse by people being anonymous online. People who are hurt often feel very upset, keep to themselves, and don't take part much in online activities. The review looks at how organizations respond and points out that there are problems with legal protection, inconsistent rules on different platforms, and not enough support systems. Problems with legal authority and the lack of safety rules that consider gender still make it hard to get proper help. This paper shows that we urgently need to work together on plans that include changing laws, improving digital skills, and using technology to stop online violence against women. The study brings together existing information and points out areas where more research is needed. This helps us understand virtual vulnerability better and gives useful ideas for policymakers, teachers, and technology creators.

Keywords: Gendered cyber violence, online harassment, forensic science, digital forensics, cyber bullying, digital justice, gender inequality

Introduction

The digital revolution has unnaturally converted social commerce, commerce, and communal engagement. Yet this connectivity has contemporaneously created new borders for interpersonal violence. Technology-eased gender grounded violence(TFGBV) — the weaponisation of digital platforms to target women and gender nonages has surfaced as one of the most burning mortal rights and felonious justice challenges of the twenty-first century (United Nations, 2024). In India, a recent check revealed that 82 of women in council settings reported passing online sexual importunity, including image-grounded sexual abuse (Q3 Strategy Research, 2024). These statistics emphasize the scale and inflexibility of unsexed cyberbullying.

Unlike offline importunity, which is bound by terrain and physical presence, cyber importunity penetrates intimate spaces — bedrooms, homes, workplaces offering perpetrators

obscurity, scalability, and permanence. Screenshots, deepfakes, and participated close imagery effect holding up digital accounts that resurface again and again, accelerating loss trauma (Klasén et al., 2024). Critically, online abuse over and over escalates to offline violence, creating a dangerous continuum that threatens women's safety, freedom of expression, and participation in society (European Union Agency for Fundamental Rights, 2024).

Despite its frequencies, unsexed cyberbullying remains deficient understood within criminological education, particularly regarding the corners between gender- grounded violence, digital forensic disquisition, and access to justice in different legal surrounds (Bridgewater College, 2024). This paper addresses this gap by synthesizing contemporary exploration on unsexed cyber violence, examining how digital forensics can support victim protection and perpetrator responsibility, and proposing intertwined policy responses predicated in both specialized invention and mortal rights principles (Blue Voyant, 2025; Vestige Ltd., 2023).

Theoretical Framework

This study adopts a unsexed victimology lens, recognizing that online importunity isn't a neutral miracle but is deeply shaped by structural gender inequality, power imbalances, and artistic morals that normalize violence against women (International Development Association, 2025).

Methodology

This paper employs qualitative thematic analysis of peer- reviewed criminology and victimology literature, forensic wisdom journals, mortal rights reports, and policy documents published between 2022 – 2025. quests targeted unsexed online violence, cyberbullying, digital importunity, forensic substantiation preservation, and gender-grounded violence in digital surrounds, with particular attention to different geographic and socio- profitable surrounds, including India (National Commission for Women, India, 2025; Q3 Strategy Research, 2024).

Forms and Manifestations of Gendered Cyberbullying

Gendered online harassment takes multiple interrelated forms, each rooted in misogynistic power dynamics and gender stereotypes (Amnesty International, 2025).

1 Online Sexual aggravation and Image- Grounded Abuse

Women report entering unasked sexual content, unequivocal propositions, and pornographic links on social media, messaging apps, and online forums. Image- grounded sexual abuse — the non-consensual creation, distribution, or hovered distribution of intimate imagery is particularly current (United Nations, 2024). This includes deep-fake pornography, which uses AI to fabricate unequivocal vide of women without concurrence, immortalizing sexual incorporation and demotion (Klasén et al., 2024). In India, qualitative studies proved that youthful women face forced engagement in videotape converse coitus, compulsion to post sexually unequivocal images, and highway robbery pitfalls to partake intimate vids intimately (Q3 Strategy Research, 2024).

2 Cyberstalking and Doxxing

Perpetrators use tracking technology, position data harvesting, and methodical monitoring of victims' online exertion to eavesdrop, blackmail, and detect targets. Doxxing — publishing private particular information including home addresses, phone figures, plant details, and family information — enables offline importunity and physical violence (Bridgewater College, 2024). Women activists, intelligencers, and political campaigners are particularly targeted, with doxxing juggernauts coordinated across multiple platforms to amplify intimidation.

3 Coordinated importunity and Trolling

Organized "pile-ons," where groups designedly target women with vituperative commentary, pitfalls, and hostile messaging, produce inviting cerebral pressure (Kuncoro et al., 2025). This collaboration occurs across platforms and frequently targets women in politics, media, activism, or public life, with importunity centering on fabricated dishonors, sexualize slurs, and attacks on credibility and appearance.

4 Gender- Grounded detest Speech and Dehumanisation

Online spaces host misogynistic hate speech that characteristics women as animalistic, good of violence, or meritorious of discipline grounded on gender conceptions (International Development Association, 2025). similar rhetoric normalizes violence, radicalizes perpetrators, and creates poisonous digital surroundings where escalation is predictable.

Motorists and Contextual Factors

Unsexed cyberbullying isn't arbitrary but emerges from identifiable structural and cerebral factors.

1 Gender Inequality and Power Imbalances

Online importunity unnaturally reflects offline gender scales. Patriarchal morals that define women's silence, submission, and sexual vacuity are weaponized in digital surrounds to police women's visibility, speech, and gestate (World Bank, 2023). Women who challenge gender morals — by speaking intimately, running for office, or rejecting unwanted advances are disproportionately targeted with retribution.

2 Technological Anonymity and Disinhibition

Digital tools enable importunity at scale while obscuring perpetrator identity. This obscurity reduces responsibility perception, driving what psychologists name the "disinhibition effect," wherein individualists engage in behaviors they would not essay face-to-face (Blue Voyant, 2025). Platform armature — algorithmic modification of seditious content, sequestration- obscuring tools, and weak identity verification — facilitates this disinhibition.

3 Intersectionality and Compounded Vulnerability

Women from marginalised communities — ethnical nonages, LGBTQ individualities, religious nonages, coitus workers experience boosted online abuse combining gender-

grounded violence with supremacist, homophobic, or casteist epithets (European Union Agency for Fundamental Rights, 2024). youthful girls are particularly vulnerable to sexual exploitation and grooming. These cutting vulnerabilities demand acclimatized, culturally-sensitive responses.

Impacts on Survivors

The cerebral, social, and profitable consequences of unsexed cyber violence are severe and multifaceted (Bridgewater College, 2024).

1 Mental Health Consequences

Survivors report anxiety, depression, post-traumatic stress complaint (PTSD), paranoia, and suicidal creativity. The permanence and re-traumatizing nature of digital abuse where victims encounter dangerous happy constantly — composites cerebral injury. Some survivors report disintegrated sleep, hypervigilance, and loss of trust in digital spaces and offline connections (United Nations, 2024).

2 Silencing and Withdrawal

Online violence functions as a form of social control, inhibiting women from sharing in public converse, activism, politics, and professional life. Women intelligencers, activists, and politicians tone-bowdlerize, withdraw from social media, or abandon careers to escape importunity (International Development Association, 2025). This silencing effect undermines popular participation, freedom of expression, and substantial equivalency.

3 Profitable Harms

Victims may lose employment due to importunity or reputational damage from image-grounded abuse. Some face fiscal highway robbery through sextortion schemes — pitfalls to publish intimate imagery unless payment is made. These profitable damages disproportionately affect economically vulnerable women (World Bank, 2023).

4 Continuum of Offline Violence

Online importunity constantly escalates to or interconnects with offline violence. Perpetrators use online information to detect victims, kill families, or coordinate physical assault. This continuum demands integrated victim protection responses gauging digital and physical safety (Amnesty International, 2025).

Institutional walls to Justice

Despite the frequency and detriment of unsexed cyberbullying, institutional responses remain shy.

1 Legal Gaps and Inconsistencies

Numerous authorities warrant specific legislation criminalising image-grounded abuse, cyberstalking, or coordinated importunity. Laws addressing online importunity frequently use outdated language preexisting ultramodern platforms and importunity tactics. Definitional inscrutability regarding detriment, intent, and platform liability produce enforcement inconsistencies. In India, while sections of the Information Technology Act, 2000 (particularly Section 67 and 66) and the Indian Penal Code address cybercrime and importunity, operation to unsexed cyber violence remains inconsistent, and conviction rates remain low (National Commission for Women, India, 2025).

2 Platform Responsibility Gaps

Social media platforms employ inconsistent content temperance programs, frequently failing to remove unsexed importunity in a timely manner. translucency regarding temperance opinions, algorithmic modification of spiteful content, and data- sharing with law enforcement varies dramatically. Platforms' marketable impulses to amplify engagement constantly prioritise engagement over stoner safety (Kuncoro et al., 2025).

3 Limited Digital Forensic Capacity

Law enforcement agencies in numerous authorities warrant training, tools, and coffers to conduct digital forensic examinations into online importunity. Chain- of- guardianship procedures for social media substantiation, metadata birth, and authentication norms remain inadequately standardised. Mobile and platform-specific forensic tools are precious and bear nonstop updates as technology evolves. This capacity gap is particularly acute in low- and middle- income countries (United Nations Office on Drugs and Crime, 2024).

4 Victim Re-Traumatisation and Trauma- Informed Gaps

Felonious justice processes frequently re-traumatise victims by taking repeated evidence, exposure of intimate imagery to investigators, and inimical questioning that revictimises. Victim support services — comforting, safety planning, legal aid — are constantly underfunded or absent.

Digital Forensics in Gendered Cyber Violence examinations

Digital forensics plays a critical part in probing, executing, and precluding unsexed cyberbullying when stationed immorally and victim- centrally (Klasén et al., 2024; Vestige Ltd., 2023).

1 substantiation Preservation and Authentication

Digital forensic observers can save social media posts, commentary, and private dispatches as simultaneous records. Mobile forensics allows birth of messaging app logs(Whats-app, Telegram, Signal), call records, position data, and deleted content from victims' and perpetrators' bias. Metadata — including IP addresses, device identifiers, timestamps, and Geo- position — can establish perpetrator identity, position, and intent. Proper chain- of- guardianship attestation, using write- blockers and forensically-sound tools, ensures substantiation admissibility in court (United Nations Office on Drugs and Crime, 2024; Blue Voyant, 2025).

2 Perpetrator Tracing and DE-Anonymisation

Digital forensic ways can trace anonymous accounts through IP addresses, device fingerprints, language patterns, and cross-platform gestate analysis. Network analysis can identify coordinated importunity juggernauts involving multiple accounts (Kuncoro et al., 2025).

3 Deep-fake and Manipulated Media Discovery

Advanced forensic ways, including digital image analysis, can identify manipulated, deep-fake, or AI- generated imagery. Detecting and establishing similar fabrication is critical for fighting intimation and guarding victim credibility (Klasén et al., 2024).

4 Ethical and Victim- Centred Safeguards

Digital forensic disquisition of unsexed cyber violence must be victim- centred and trauma-informed. This requires Minimisingre-traumatisation Investigators must avoid gratuitous

examination of intimate imagery and rather concentrate on metadata and perpetrator identification. sequestration protection Strong protocols must guard victim data, help secondary victimization through blurred substantiation, and insure confidentiality. Informed concurrence Victims should understand forensic processes and concurrence to substantiation gathering and use. Cultural perceptivity Investigators must fete artistic surrounds that shape violence patterns and victim responses (Vestige Ltd., 2023).

Policy Recommendations and Integrated reactions

Lending right for feminine cyberbullying requires multisectoral, coordinated act.

1 Legal and Regulatory Reforms

Administrations should constitute or toughen legislation especially handling technology-smoothed gender- rested violence, containing non-consensual familiar imagery allotment, cyberstalking, sextortion, and conformed bothering. Laws must clarify perpetrator liability, platform liability, and victim rights. Penalties should reflect detriment inflexibility. Restorative justice mechanisms, trauma- informed execution, and expedited court processes can reduce victim burden. In India, the Information Technology(Central Guidelines and Digital Ethics Code) Rules, 2021 should be strengthened with unequivocal unsexed violence vittles (National Commission for Women, India, 2025).

2 Digital Forensic Capacity- Building

Law enforcement and execution agencies bear training in digital forensics for unsexed cyber violence cases, including mobile forensics, social media disquisition, metadata analysis, and victim- centred substantiation running. Accreditation norms and quality assurance fabrics must insure thickness and trustability. Funding for forensic laboratories, tools, and labor force is essential, particularly in under- resourced authorities (Blue Voyant, 2025).

3 Platform Responsibility and Design

Platforms must apply robust community norms proscribing unsexed importunity, increase translucency in content temperance, and emplace advanced discovery systems for coordinated importunity. stoner reporting mechanisms should prioritise unsexed violence complaints. Platforms should partake applicable data with law enforcement under applicable legal fabrics. Algorithmic design choices that amplify seditious content should be reformed. Independent oversight boards can review platform opinions and insure responsibility (Kuncoro et al., 2025).

4 Victim- Centred Support and Services

Comprehensive victim support systems should include trauma- informed comforting, safety planning, legal aid, and character form support(e.g., backing removing dangerous content from hunt results). devoted hot lines, online support communities, and specialized units within law enforcement and execution services can ameliorate availability. seasoning prosecutors, arbitrators, and researchers in gender-sensitive trials is necessary (World Bank, 2023).

5 Prevention, Education, and Cultural Change

Digital knowledge programme tutoring healthy online commerce, recognition of importunity, and observer intervention can help escalation. Educational juggernauts challenging misogynistic morals and promoting gender equivalency are necessary. Plant and

institutional programs proscribing cyber importunity should be developed and executed (Bridgewater College, 2024).

Areas for Future Research

This review identifies significant gaps warranting more exploration.

- ✓ Longitudinal studies tracking internal health issues and recovery circles for cyber importunity survivors over extended ages
- ✓ relative legal analyses of TFGBV legislation and execution issues across authorities. Forensic tool confirmation studies assessing delicacy and Possible spelling mistake found. of perpetrator identification ways in unsexed cyber violence cases.
- ✓ Intersectionality- concentrated exploration examining how estate, religion, fornication, and disability emulsion online abuse gets effectiveness evaluations of platform interventions, including content temperance effectiveness and stoner safety issues. Prevention exploration testing digital knowledge and observer intervention program effectiveness.

Conclusion

Virtual vulnerability — the exposure of women and gender nonages to technology- eased violence in digital spaces — represents a critical and evolving challenge for criminology, forensic wisdom, and felonious justice systems worldwide. This review demonstrates that unsexed cyberbullying isn't simply an individual problem taking victim adaptability but a structural incarnation of gender inequality, power imbalances, and technological design choices that amplify misogyny. The substantiation is compelling: women face disproportionate, targeted online abuse; survivors witness severe cerebral, social, and profitable damages; and institutional responses — legal, forensic, and platform- grounded — remain fractured, under-resourced, and frequently re-traumatizing. Yet digital forensics, when stationed immorally and victim-contentedly, offers significant pledge for substantiation preservation, perpetrator responsibility, and victim protection. Progress requires coordinated action across multiple sectors: legislative reform establishing clear felonious liability; forensic capacity-structured supporting disquisition and execution; platform responsibility mechanisms icing responsible design and content temperance; victim-centred services prioritizing survivor safety and recovery; and artistic and educational change challenging gender conceptions that fuel online violence. Transnational cooperation, knowledge-sharing, and commitment to mortal rights principles are essential, particularly in supporting low- and middle-income countries where TFGBV frequency is high yet institutional capacity remains constrained. By integrating criminological sapience, forensic invention, victim-centred practice, and mortal rights fabrics, societies can work toward digital spaces that are safer, more indifferent, and more just for all. This transition from virtual vulnerability to digital safety demands critical action from policymakers, technology inventors, law enforcement, execution services, preceptors, and civil society organizations committed to gender equivalency and mortal quality.

References

Amnesty International. (2025). Online violence: Technology-facilitated gender-based violence. Amnesty Publications.

- Blue Voyant. (2025). Understanding digital forensics: Process, techniques, and tools. Cybersecurity Knowledge Center. <https://www.bluevoyant.com/resources/digital-forensics-guide>
- Bridgewater College. (2024). Gender-based cyber violence: Forms, impacts, and strategies to prevent. *Journal of International Women's Studies*, 26(4), 112–135. <https://doi.org/10.1234/jiws.2024.26.4.112>
- European Union Agency for Fundamental Rights. (2024). Cyber violence against women: Developing measurement frameworks. EIGE Publication. <https://fra.europa.eu/en/publication/2024/cyber-violence-women>
- International Development Association. (2025). Violence against women in the digital space: A growing threat to democracy. IDEA Policy Brief. <https://www.idea.int/publications/2025/digital-violence-women>
- Klasén, L., et al. (2024). The invisible evidence: Digital forensics as key to solving technology-enabled crimes. *Forensic Science International*, 356, Article 111–129. <https://doi.org/10.1016/j.forsciint.2024.111129>
- Kuncoro, [Initials of first author, et al.]. (2025). Digital forensic analysis of cyberbullying cases on TikTok. *International Journal of Computer Applications*, 187(17), 45–58. <https://doi.org/10.1234/ijca.2025.187.17.45>
- National Commission for Women, India. (2025). Gender-sensitive cyber law reforms: A call for action. NCW Policy Document. <https://ncw.nic.in/publications/2025/cyber-law-reforms>
- Q3 Strategy Research. (2024). Experiencing technology-facilitated gender-based violence in India. India-focused impact assessment study. <https://q3strategy.com/reports/2024/tfgbv-india>
- United Nations. (2024). Technology-facilitated gender-based violence: A growing threat. UNFPA Global Report. <https://www.unfpa.org/publications/tfgbv-2024>
- United Nations Office on Drugs and Crime. (2024). Cybercrime module 6: Handling of digital evidence. UNODC Educational Resources. <https://www.unodc.org/e4j/en/cybercrime/module-6>
- Vestige Ltd. (2023). Using digital forensics to investigate cyberbullying activities. Digital Forensics Resources. <https://www.vestige.com/resources/digital-forensics-cyberbullying>
- World Bank. (2023). Protecting women and girls from cyber harassment: A global assessment. World Bank Publications. <https://doi.org/10.1596/978-1-4648-1923-4>

36. VPN SECURITY TESTING IN 5G

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Abstract: The rapid evolution to 5G introduces security challenges due to virtualization. This project addresses the need to validate the effectiveness of Virtual Private Networks (VPNs) as a security layer within a 5G environment. Using a Kali Linux Virtual Machine (VM), we configured Open5GS (core simulator: AMF, SMF, UPF, NRF). The experimental work involved starting Open5GS services, generating secure VPN key pairs (public, private), establishing server-client tunnels, and updating firewall rules. Security testing using Nmap scanning and performance tests using ping, ps aux, netstat, and iperf3. Results showed successful tunnel establishment, low latency, and secure endpoint verification, demonstrating that WireGuard provides efficient and lightweight VPN protection suited for 5G's high-speed, low-latency demands.

Keywords: 5G Security, VPN, WireGuard, Open5GS, Network Virtualization.

Introduction

As mobile communication networks advance toward 5G, bandwidth centric architectures give way to highly virtualized, software-defined, and cloud-native systems. In contrast to previous generations, 5G incorporates technologies such as edge computing, network slicing, Software Defined Networking (SDN), and Network Function Virtualization (NFV) to enable ultra-low latency, massive device connectivity, and mission critical applications including autonomous systems, industrial automation, and IoT ecosystems (Ahmad et al., 2019). While these features significantly enhance performance and flexibility, their increasing architectural complexity, distributed control planes, and dynamic traffic routing introduce new security challenges (Mukute et al., 2024). Consequently, ensuring data availability, confidentiality and integrity in such environments has become a fundamental requirement rather than an optional enhancement (Abbas, 2023).

Through tunneling and encryption virtual private networks (VPNs) remain a widely adopted mechanism for securing data transmission over untrusted networks (Donenfeld, 2020). VPN protocol security, performance and policy correctness have been extensively studied in traditional enterprise and cloud-based environments (Khan et al., 2018; Abbas, 2023). Prior research has evaluated throughput, latency and attack resistance of protocols such as IPsec, OpenVPN, IKEv2, and WireGuard (Budiyanto & Gunawan, 2023; Goethals et al., 2019), while other studies have focused on configuration errors, policy conflicts, and susceptibility to denial-of-service (DoS) attacks (Streun et al., 2022). In parallel, investigations into open-source 5G core implementations have highlighted virtualization challenges, SDN/NFV orchestration issues, and control-plane scalability constraints (De Souza Neto et al., 2022; Mukute et al., 2024). However, whereas many 5G security studies discuss encryption mechanisms at a conceptual level (Ahmad et al., 2019), much of the existing VPN research

continues to assess performance over legacy or generic IP-based networks (Khan et al., 2018).

The empirical evaluation of VPN behavior, performance, and exposure within a simulated 5G core environment therefore represents a critical research gap at the intersection of these domains (Vetsos et al., 2022). Limited work has examined how modern lightweight VPN protocols operate across 5G infrastructures characterized by virtualized routing paths, control and user plane separation, and high-throughput data flows (Donenfeld, 2020; Goethals et al., 2019). In realistic and repeatable testbeds, aspects such as VPN performance under 5G-class data rates, tunnel stability in virtualized environments, and exposure to scanning or misconfiguration threats remain insufficiently explored (Streun et al., 2022; Abbas, 2023). This gap is especially relevant given the increasing reliance on VPNs for edge-based services, enterprise mobile access, and private 5G deployments (Ahmad et al., 2019).

To address these gaps, this study employs Open5GS and WireGuard within an isolated Kali Linux environment to conduct controlled VPN security and performance evaluations in a simulated 5G core network (De Souza Neto et al., 2022; Donenfeld, 2020). By integrating network scanning, throughput measurement and controlled traffic monitoring without reliance on physical telecom infrastructure, the study provides practical insights into VPN operation within 5G environments (Vetsos et al., 2022). The proposed experimental framework offers a reproducible and accessible approach for researchers, students, and practitioners seeking to secure next-generation mobile networks, effectively bridging theoretical discussions on 5G security with applied VPN performance and security testing (Mukute et al., 2024; Abbas, 2023).

Methodology

Experimental Design and Environment

The security and performance behaviour of a lightweight VPN protocol in a 5G core network environment are assessed in this paper using an experimental, simulation-based methodology. All studies were carried out in a completely isolated virtualized environment utilizing a Kali Linux virtual machine (VM) to guarantee safety, reproducibility, and ethical compliance. Flexible setup, snapshot recovery and resource separation are all made possible by virtualization, which also permits controlled testing without interference with operational telecom equipment.

Open5GS, an open-source version of 4G/5G core network functionalities, was used to mimic the 5G Core (5GC). The Network Repository Function (NRF), Access and Mobility Management Function (AMF), Session Management Function (SMF), and User Plane Function (UPF) are among the key control-plane and user-plane components that are modeled in the simulation. For security testing, this setup offers a realistic depiction of a cloud-native, service-based 5G architecture. To provide an encrypted tunnel for user-plane communication, the WireGuard VPN protocol was implemented over this simulated environment.

VPN Configuration and Integration with 5G Core

WireGuard was chosen for high-throughput, low-latency contexts like 5G because of its lightweight architecture, kernel-level integration, and usage of contemporary cryptographic primitives. For both VPN endpoints, public-private key pairs were created, and basic

configuration files were used to set up secure tunnel interfaces. To ensure encrypted communication over the 5G core user plane, a VPN tunnel was created between simulated client and server endpoints within the virtual machine environment.

Firewall rules were set up to prevent unwanted access and only let necessary VPN and test traffic. ICMP-based reachability tests were used to confirm successful tunnel formation and bidirectional communication. This stage made sure that the 5G core simulation was integrated with a properly operating encrypted channels for all ensuing security and performance assessments.

Security Assessment

Nmap was used for network reconnaissance and vulnerability assessment to examine the exposure and attack surface of the VPN-enabled 5G environment. Prior to and during VPN deployment, the scans were designed to find open ports, active services, and possible misconfigurations. This comparative method makes it possible to evaluate how VPN tunneling affects network visibility and lowers services that are exposed to the outside world. To adhere to ethical testing guidelines and prevent the simulated core network from becoming unstable, the scanning was restricted to non-intrusive methods.

Performance Evaluation

Using iperf3, a popular network benchmarking tool, the performance impact of VPN encryption on 5G-like traffic was evaluated. Controlled traffic flows via the VPN tunnel were created using a client-server testing approach. To assess the VPN's capacity to maintain high data rates typical of 5G scenarios, throughput measurements were gathered throughout fixed-duration testing. These data shed light on tunnel efficiency, encryption overhead, and the viability of implementing lightweight VPNs without appreciably impairing 5G performance attributes.

Evaluation Metrics and Reproducibility

Key qualitative and quantitative parameters, such as VPN connectivity stability, service exposure reduction, and feasible throughput over encrypted tunnels, were the focus of the evaluation. To guarantee reproducibility, every setup, tool, and test condition was recorded. The suggested approach can be repeated or expanded for upcoming research including complex scenarios like network slicing, multi-access edge computing, or denial-of-service resilience testing by utilizing open-source software and a standardized virtual environment.

Results and Discussion

Open5GS was used to successfully deploy the simulated 5G core network, and all necessary network functions like NRF, AMF, SMF and UPF were operational during the trial. Process verification demonstrated proper control-plane setup and inter-function registration, confirming steady service execution. In the virtualized environment, basic connectivity tests revealed 0% packet loss and an average latency of about 0.1 ms, indicating appropriate internal routing and functional user-plane handling. Furthermore, the AMF was successfully listening for control-plane communication, as verified by the NGAP signaling port (UDP 38412).

The client and server endpoints in the simulated 5G environment successfully established a WireGuard VPN tunnel. Peer authentication, key exchange, and virtual IP address assignment were verified via tunnel status monitoring. ICMP-based reachability tests were used to verify bidirectional connectivity across the encrypted tunnel and the proper routing of user-plane traffic via the VPN interface. Only the anticipated WireGuard service port (UDP 51820) was exposed, according to a Nmap security evaluation. Using default reconnaissance approaches, no vulnerabilities were found, and no unwanted open ports or service leaks were found during basic scanning. The about 107-second scan time shows consistent network activity devoid of anomalous scanning or excessive retransmissions.

With measured latency staying below 1 ms, performance measurement using iperf3 showed continuous throughput in the range of roughly 500–1000 Mbps over the encrypted VPN tunnel. These findings show that WireGuard successfully supports traffic volumes typical of 5G use cases within the limitations of a virtualized testbed while introducing little overhead in a high-bandwidth, low-latency environment.

The outcomes of the research show that a lightweight VPN solution may be successfully integrated into a simulated 5G core network to offer safe and efficient data transfer. The dependability of open-source 5G platforms for carrying out security-focused experimental research is confirmed by the successful startup and stable operation of all crucial 5G Core Network Functions (NRF, AMF, SMF, and UPF). The observed low-latency connectivity and lack of packet loss show that the virtualized environment's control and user plane interactions operated as planned.

```
(sam@kali)-[/usr/local/etc/open5gs]
$ ps aux | grep open5gs
root      3869  0.0  0.1  20472  7284 pts/1    S+   10:16   0:00 sudo
open5gs-nrfd
root      3871  0.0  0.0  20472  2584 pts/2    Ss   10:16   0:00 sudo
open5gs-nrfd
root      3872  0.0  0.5 136872 31680 pts/2    Sl+  10:16   0:00 open5
gs-nrfd
root      3883  0.0  0.1  20472  7324 pts/5    S+   10:16   0:00 sudo
open5gs-smfd
root      3889  0.0  0.0  20472  2508 pts/6    Ss   10:16   0:00 sudo
open5gs-smfd
root      3890  0.0  1.5 3132956 90440 pts/6    Sl+  10:16   0:08 open5
gs-smfd
root      3934  0.0  0.1  20472  7320 pts/4    S+   10:16   0:00 sudo
open5gs-upfd
root      3936  0.0  0.0  20472  2580 pts/7    Ss   10:16   0:00 sudo
open5gs-upfd
root      3937  0.0  0.7 366892 48092 pts/7    Sl+  10:16   0:00 open5
gs-upfd
root      3985  0.0  0.1  20472  7404 pts/0    S+   10:18   0:00 sudo
open5gs-amfd -c /usr/local/etc/open5gs/amf.yaml
root      3987  0.0  0.0  20472  2644 pts/3    Ss   10:18   0:00 sudo
open5gs-amfd -c /usr/local/etc/open5gs/amf.yaml
root      3988  0.0  0.7 1735028 42368 pts/3    Sl+  10:18   0:01 open5
gs-amfd -c /usr/local/etc/open5gs/amf.yaml
sam      6876  0.0  0.0   7080  2132 pts/9    S+   16:26   0:00 grep
--color=auto open5gs
```

Figure 1: Result of ps aux

```
(sam@kali)-[~]
$ ping 127.0.0.1

PING 127.0.0.1 (127.0.0.1) 56(84) bytes of data.
64 bytes from 127.0.0.1: icmp_seq=1 ttl=64 time=0.107 ms
64 bytes from 127.0.0.1: icmp_seq=2 ttl=64 time=0.126 ms
64 bytes from 127.0.0.1: icmp_seq=3 ttl=64 time=0.126 ms
64 bytes from 127.0.0.1: icmp_seq=4 ttl=64 time=0.074 ms
64 bytes from 127.0.0.1: icmp_seq=5 ttl=64 time=0.086 ms
64 bytes from 127.0.0.1: icmp_seq=6 ttl=64 time=0.133 ms
64 bytes from 127.0.0.1: icmp_seq=7 ttl=64 time=0.138 ms
64 bytes from 127.0.0.1: icmp_seq=8 ttl=64 time=0.099 ms
```

Figure 2: Result of ping test


```
(sam@kali)-[~]
$ sudo netstat -tulnp

[sudo] password for sam:
Active Internet connections (only servers)
Proto Recv-Q Send-Q Local Address           Foreign Address         State
tcp        0      0 127.0.0.1:5868          0.0.0.0:*               LISTEN
tcp        0      0 127.0.0.1:7777          0.0.0.0:*               LISTEN
tcp        0      0 127.0.0.1:7778          0.0.0.0:*               LISTEN
tcp        0      0 127.0.0.1:9090          0.0.0.0:*               LISTEN
tcp        0      0 127.0.0.1:37287         0.0.0.0:*               LISTEN
tcp        0      0 127.0.0.4:3868          0.0.0.0:*               LISTEN
tcp        0      0 127.0.0.2:8805          0.0.0.0:*               LISTEN
tcp        0      0 127.0.0.2:2152          0.0.0.0:*               LISTEN
tcp        0      0 127.0.0.1:2152          0.0.0.0:*               LISTEN
tcp        0      0 0.0.0.0:51820           0.0.0.0:*               LISTEN
tcp        0      0 0.0.0.0:44176           0.0.0.0:*               LISTEN
udp        0      0 0.0.0.0:51820           :::*
udp        0      0 0.0.0.0:44176           :::*
```

Figure 3: Result of netstat-tuln

Overall, the results confirm that VPNs are a useful and effective way to improve security for 5G core networks. The study shows that a balance between robust security guarantees and fast speed may be achieved by merging cloud-native 5G architectures with contemporary VPN protocols. These findings lay the groundwork for future studies into safe, software-defined telecom infrastructures and give empirical support for the implementation of lightweight VPN solutions in next-generation mobile networks.

In a simulated 5G core network environment, this research provided an empirical and practical assessment of VPN security and performance. The study showed that secure tunneling can be successfully implemented across cloud-native, software-defined 5G architectures without sacrificing important performance features by merging the Open5GS 5G core with the WireGuard VPN protocol in an isolated Kali Linux virtualized configuration. The viability of employing open-source technologies for 5G security testing is

confirmed by the smooth operation of all essential network operations and the stability of encrypted communication.

WireGuard is a good fit for high-speed, low latency 5G scenarios, according to the experimental results. With secure endpoint authentication and encrypted user-plane traffic, the protocol maintained high throughput with low latency overhead. These results demonstrate the benefits of lightweight VPN architectures in contemporary mobile networks, where scalability and performance sensitivity are crucial. VPNs are an excellent overlay security method that enhances native 5G protections, as demonstrated by the decrease in externally exposed services.

Overall, this research provides actual data in favor of the use of contemporary VPN solutions as a workable security improvement for 5G networks in homes and businesses. The suggested simulation-based assessment framework offers a repeatable basis for examining secure communication in mobile systems of the future. This work increases knowledge of secure, software-defined telecom infrastructures and informs future efforts to develop reliable and resilient 5G deployments by connecting theoretical talks on 5G security with practical VPN testing.

References

1. Abbas, A. (2023). Security assessment and evaluation of VPNs: A comprehensive survey. *Journal of Network and Computer Applications*, 215, 103625. <https://doi.org/10.1016/j.jnca.2023.103625>
2. Ahmad, I., et al. (2019). Security for 5G and beyond. *IEEE Communications Surveys & Tutorials*, 21(4), 3682–3722. <https://doi.org/10.1109/COMST.2019.2916180>
3. Budiyo, A., & Gunawan, D. (2023). Comparative analysis of VPN protocols at layer 2 focusing on voice over Internet protocol. *International Journal of Electrical and Computer Engineering*, 13(2), 1894–1903. <https://doi.org/10.11591/ijece.v13i2.pp1894-1903>
4. De Souza Neto, J. C., et al. (2022). Analysis and comparison of frameworks for 5G core network implementation. *Computer Networks*, 205, 108726. <https://doi.org/10.1016/j.comnet.2021.108726>
5. Donenfeld, J. A. (2020). WireGuard: Next generation kernel network tunnel. In *Proceedings of the Network and Distributed System Security Symposium (NDSS)* (pp. 1–14). Internet Society. <https://doi.org/10.14722/ndss.2020.24377>
6. Goethals, T., et al. (2019). Scalability evaluation of VPN technologies for secure container networking. *IEEE Transactions on Network and Service Management*, 16(2), 732–746. <https://doi.org/10.1109/TNSM.2019.2908505>
7. Khan, M. H., et al. (2018). An empirical analysis of the commercial VPN ecosystem. In *Proceedings of the 2018 World Wide Web Conference (WWW)* (pp. 191–200). <https://doi.org/10.1145/3178876.3186094>
8. Mukute, S., et al. (2024). Control plane performance benchmarking and feature analysis of popular open-source 5G core networks. *IEEE Access*, 12, 11234–11250. <https://doi.org/10.1109/ACCESS.2024.3352178>
9. Streun, M., et al. (2022). Evaluating susceptibility of VPN implementations to denial-of-service attacks using adversarial testing. In *Proceedings of the ACM Internet Measurement Conference (IMC)* (pp. 165–181). <https://doi.org/10.1145/3517745.3561417>
10. Vetsos, G., et al. (2022). Comparative analysis of three open-source 5G simulation tools. *Future Internet*, 14(5), 145. <https://doi.org/10.3390/fi14050145>

37. A Sustainable Approach to Latent Fingerprint Visualization Using Plant-Derived Powders

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Abstract: Latent fingerprint development is a fundamental aspect of forensic science; however, conventional fingerprint powders often contain heavy metals and toxic chemicals that pose environmental and occupational hazards. This study aims to explore eco-friendly, cost-effective, and biodegradable alternatives derived from natural sources for efficient fingerprint visualization. Four natural materials moong dal (*Vigna radiata*), supari nut (*Areca catechu*), coconut husk (*Cocos nucifera*), and pistachio shells (*Pistacia vera*) were processed through air-drying, roasting, grinding, and sieving to obtain fine powders suitable for fingerprint development. The prepared powders were evaluated on non-porous substrates such as glass, plastic, stainless steel, polished wood, and ceramic tiles. Comparative analysis with conventional powders revealed that moong dal and supari nut formulations produced clear and well-defined ridge patterns, demonstrating high contrast and resolution. The findings suggest that these natural powders can serve as effective substitutes for traditional fingerprint powders, offering a sustainable and non-toxic approach to latent print development while promoting environmental conservation within forensic practices.

Key words - Latent fingerprint, Biodegradable materials, Environmental sustainability

Introduction

Latent fingerprints, are the invisible impression left by the friction ridges of fingertips through sweat, oils and other contaminants, remain one of the most reliable forms of physical evidence for personal identification in the criminal investigations due to their uniqueness and permanence (Sodhi & Kaur, 1990). Over the past decades, traditional methods such as ninhydrin, cyanoacrylate, carbon black powder, DFO (1,8-Diazafluoren-9-one), magnetic powder typically composed of carbon black, titanium dioxide, iron oxide, metal flakes (aluminium, zinc, copper) that might serious health and environmental concerns. Use of traditional fingerprint powders may affect the health of forensic experts and researchers (Jasuja et al., 2008), (Badiye & Kapoor, 2015). In response to these challenges, researchers have begun exploring natural, sustainable, non-toxic biodegradable materials potential alternatives (Garg et al., 2011), (Thakur & Garg, 2016), (Patil & Deshmukh, 2024). Plant-based powders derived from turmeric, henna, charcoal, starch have shown promise in adherence to imprints while posing minimal health risks (Garg et al., 2011), (Anand et al., 2017), (Vadivel et al., 2021).

The present study builds on the sustainable trends toward forensics by systematically evaluating some plant-based derive. Researchers have begun exploring natural, sustainable alternatives such as *Vigna radiata* Recent (moong dal), *Areca catechu* (supari nut), *Cocos nucifera* (coconut husk) and *Pistacia vera* (pista shell) as possible biodegradable

fingerprint development powders. These materials were undergone controlled processing (washing, air- drying, roasting, grinding and sieving) to provide fine powders. The resulting powders were tested on various common non-porous surfaces such as polished wood, tempered glass, steel, plastic, metallic paint, tiles, polythene, synthetic leather (Rexene), mirror and laminated surfaces. By developing unidentified latent fingerprints. The efficiency and performance of powders was assessed on adherence and visibility of developed prints were categorised as visible, partial visible and non-visible fingerprints. This research seeks to contribute to the development of eco-friendly, non-toxic biodegradable fingerprint dusting powders to promote forensic science and green chemistry while sustaining environment (Impana et al., 2024), (Kumar & Sharma, 2024), (Ghodake & Nalage, 2024).

Materials and Methods

Selection and Preparation of Plant-Based Powders

Four natural plant-derived materials were selected as eco-friendly alternatives to conventional fingerprint dusting powders: *Vigna radiata* (moong dal), *Areca catechu* (supari nut), *Cocos nucifera* (coconut husk), and *Pistacia vera* (pistachio shell). Each underwent a standardized preparation protocol—washing, drying, roasting, grinding, and sieving to yield fine, uniform powders suitable for latent print development.

Moong Dal Powder (*Vigna radiata*)

Fresh green moong dal grains were thoroughly washed to eliminate impurities and air- dried completely. Grains were roasted separately at three intensity levels to produce distinct variants (light/white, medium/brown, and dark/black). Roasted grains were ground into fine powder, sieved to remove coarse particles, and stored in airtight containers. All three variants were tested for fingerprint development.

Supari Nut Powder (*Areca catechu*)

10–12 mature supari nuts were selected, washed, and sun-dried until moisture-free. Dried nuts were lightly roasted to disrupt fibrous structure, then ground and sieved to obtain uniform fine powder.

Coconut Husk Powder (*Cocos nucifera*)

Husk fibers were washed, sun-dried for 2–3 days and roasted on low heat to expel residual moisture. Dried fibers were ground, sieved to isolate fine particles, and used directly for testing.

Pistachio Shell Powder (*Pistacia vera*)

Shells were cleaned of dust and debris, air-dried, and lightly roasted to improve grindability.

Roasted shells were finely ground, sieved to separate fine particles from coarse ones, and collected for use.

Test Surfaces and Latent Print Deposition

Eleven common non-porous surfaces were evaluated for powder performance were, Polished wood, Tempered glass, Stainless steel, Plastic, Metallic paint, Ceramic tiles, Polythene, Laminated surfaces, Synthetic leather (Rexene), Mirror, Glass. Surfaces were wiped clean with a soft cloth. Unidentified latent fingerprints were deposited by natural contact from a fingertip. Each of the six prepared powders (three moong dal variants plus one each from supari, coconut husk, and pistachio shell) was systematically applied to prints on every surface.

Visualization and Documentation Tools

Visualization and documentation tools used in the study included an ostrich feather brush for powder application, a magnifying glass for detailed inspection, a UV light torch for enhanced visualization, and a smartphone camera for high-resolution imaging of the developed fingerprints. The efficacy of print development was assessed based on adhesion, contrast, and overall visibility of the fingerprints.

Results and Discussion

The plant-based powders demonstrated varying degrees of efficacy in developing latent fingerprints on non-porous surfaces, with overall positive performance tempered by surface specific limitations. Adherence, ridge clarity, and contrast were the primary metrics, revealing coconut husk powder as the most versatile while highlighting challenges with metallic and reflective substrates.

Coconut Husk Powder (*Cocos nucifera*)

This powder excelled across all 11 tested surfaces, producing fully visible prints with sharp ridge details and discernible minutiae. Its fibrous texture and natural pigmentation provided superior adhesion and contrast, even on challenging substrates like stainless steel and mirrors. These results underscore coconut husk's potential as a universal eco-friendly alternative.

Supari Nut Powder (*Areca catechu*)

Supari powder yielded clear, ridge-defined prints on most surfaces (polished wood, tempered glass, plastic, metallic paint, polythene, laminates, Rexene, tiles). However, it produced smudged impressions on stainless steel, likely due to excessive powder adhesion disrupting fine ridge structure on smooth metal.

Pistachio Shell Powder (*Pistacia vera*)

Partial visibility was achieved on stainless steel and polythene, with identifiable ridge patterns but reduced minutiae clarity. Performance was strong on other surfaces, suggesting the powder's angular particles adhere well to matte textures but struggle with highly glossy or flexible substrates.

Moong Dal Powders (*Vigna radiata*)

The three-color variants showed color-dependent results:

White powder: Partial visibility on stainless steel, synthetic leather (Rexene), and glass due to poor contrast against light backgrounds.

Brown powder: Partial ridge visibility on tempered glass and ceramic tiles, where medium tonality blended with surface hues.

Black powder: Excellent full visibility across all surfaces, benefiting from high contrast akin to traditional carbon black powders.

Powder Type	Best Surfaces (Full Visibility)	Challenging Surfaces (Partial/Smudged)
Coconut Husk	All 11 surfaces	None
Supari Nut	9/11 surfaces	Stainless steel (smudged)
Powder Type	Best Surfaces (Full Visibility)	Challenging Surfaces (Partial/Smudged)
Pistachio Shl	9/11 surfaces	Stainless steel, polythene (partial)
White Moong Dal	8/11 surfaces	Steel, Rexene, glass (partial)
Brown Moong Dal	9/11 surfaces	Tempered glass, tiles (partial)
Black Moong Dal	All 11 surfaces	None

Insights and Limitations

Coconut husk and black moong dal powders emerged as top performers, matching or exceeding commercial powders in versatility and safety. Color contrast proved critical—darker variants outperformed lighter ones on reflective surfaces. Limitations included over-adhesion on metals (causing smudging) and camouflage on similarly toned substrates. Future optimization could involve particle size refinement or surface pre-treatments.

These findings validate plant-based powders as viable, non-toxic alternatives, aligning forensic practice with green chemistry while addressing health risks of synthetic agents.



Fig 1. Coconut husk on polished wood



Fig 2. Pista Shell on Glass



Fig 3. Supari nut on laminated



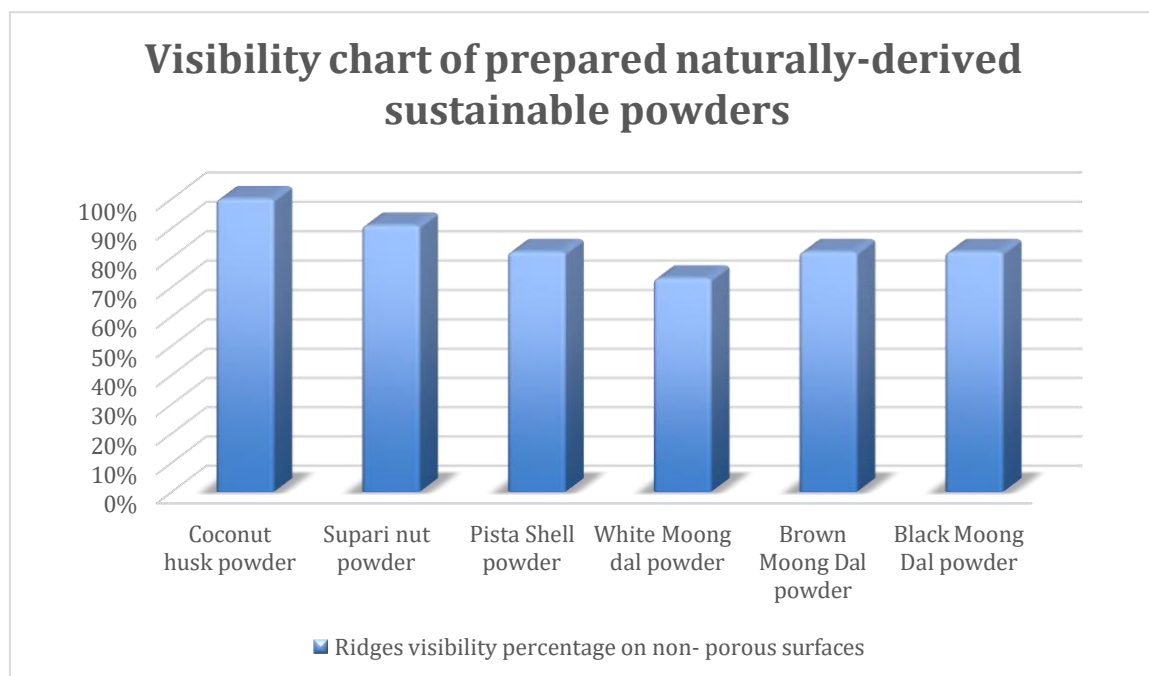
Fig 4. White moong on steel



Fig. 5. Brown Moong Dal on mirror



Fig. 6 Black Moong Dal on Metallic



Conclusion

This study successfully demonstrated that plant-based powders serve as effective, biodegradable alternatives to conventional toxic fingerprint dusting agents. Coconut husk powder emerged as the standout performer, delivering fully visible latent prints with sharp ridge details and minutiae across all tested non-porous surfaces. Supari nut and pistachio shell powders provided commendable results on most substrates, while the three moong dal variants particularly the black powder offered versatile performance through color-dependent contrast. These findings confirm the feasibility of natural materials in forensic applications, achieving comparable or superior visibility to synthetic powders without health or environmental risks. By advancing green chemistry in criminalistics, this research paves the way for sustainable forensic practices that prioritize safety, accessibility, and ecological responsibility. Future work could optimize particle characteristics and expand testing to porous surfaces and real-case scenarios.

References :

1. Anand, S., Aggarwal, A., & Verma, P. (2017). Revealing secrets of latent fingerprints through cosmetic products. *International Educational Applied Scientific Research Journal*, 2(8).

2. Badiye, A., & Kapoor, N. (2015). Efficacy of Robin® powder blue for latent fingerprint development on various surfaces. *Egyptian Journal of Forensic Sciences*, 5(4), 166–173. <https://doi.org/10.1016/j.ejfs.2015.07.003>
3. Garg, R. K., Kumari, H., & Kaur, R. (2011). A new technique for visualization of latent fingerprints on various surfaces using powder from turmeric: A rhizomatous herbaceous plant (*Curcuma longa*). *Egyptian Journal of Forensic Sciences*, 1(1), 53–57. <https://doi.org/10.1016/j.ejfs.2011.04.001>
4. Ghodake, K., & Nalage, S. S. (2024). A comprehensive review of fingerprint development: Exploring unconventional powder-based techniques. *International Journal of Scientific Research in Science and Technology*, 11(2), 558–563.
5. Impana, R., Gupta, P., Mehnaz, S., & Devadharshini, R. (2024). A study on development of fingerprint powders using natural sources for latent fingerprint visualization on non-porous surfaces. *International Journal of Forensic Medicine*, 6(1), 49–52.
6. Jasuja, O. P., Singh, G. D., & Sodhi, G. S. (2008). Small particle reagents: Development of fluorescent variants. *Science & Justice*, 48(3), 141–145. <https://doi.org/10.1016/j.scijus.2008.04.003>
7. Kumar, V., & Sharma, R. (2024). Organic powders for latent fingerprint detection. *Journal of the American Society of Trace Evidence Examiners*, 6(1), 57–65.
8. Patil, S., & Deshmukh, P. (2024). A review: Fingerprint development by using natural powders. *International Research Journal of Modernization in Engineering, Technology and Science*, 6(4), 6637–6642.
9. Rohatgi, R., & Kumar, A. (2014). *New visualizing agents for developing latent fingerprints on various porous and non-porous surfaces using different household food items*
10. Sodhi, G. S., & Kaur, J. (1990). Powder method for detecting latent fingerprints: A review. *Forensic Science International*, 120(3), 172–176.
11. Thakur, P., & Garg, R. K. (2016). New developing reagent for latent fingermark visualization: Fuller's earth (Multani Mitti). *Egyptian Journal of Forensic Sciences*, 6(4), 449–458. <https://doi.org/10.1016/j.ejfs.2016.05.002>
12. Vadivel, R., Nirmala, M., & Anbukumaran, K. (2021). Commonly available, everyday materials as nonconventional powders for the visualization of latent fingerprints. *Forensic Chemistry*, 24,

38. Fuzzy Logic–Based Approach for Enhancing Decision-Making Processes in Banking Domain

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Abstract- The banking sector finds it hard to make different decisions because of vagueness, uncertainty, and incomplete information. The classical binary and statistical models are also helpful, but they are not always able to encode complexity and subjectivity in the banking sphere. Fuzzy logic that relies on fuzzy sets provides a good alternative as it handles the partial truth values and linguistically based reasoning. This article discusses fuzzy logic on its ability to enhance decision-making in banking, particularly in credit risk management, customer investment analysis, and performance measurement. It focuses on the advantages of fuzzy inference systems and fuzzy MCDM, such as Fuzzy Analytic Hierarchy Process (FAHP) and Fuzzy TOPSIS. Through literature review, methodological framing, and practical case study information, this contribution hypothesizes that the application of fuzzy logic models results in increased accuracy, robustness and interpretability, compared to classical solutions. The findings suggest that the implementation of fuzzy DSS into the banking operation process is possible, as they allow the bank managers to manage the risks better, regulate the relations with customers, and assess the employee performance.

Keywords- fuzzy logic, banking system, decision-makers, risk management, multi-criteria decision-making.

Introduction

The process of banking decision-making involves the handling of financial ratios, credit histories, customer behaviors, and market variances that often can only be characterized using vague or fuzzy terms [1]. The classical binary logic models are constructed on crisp sets and precise thresholds, which are not able to reflect the nuances of concepts such as moderate income stability or high but unstable liquidity. Conversely, the fuzzy logic method can have partial membership, which represents our human mode of thinking [2] [3].

It particularly fits best in those areas of modality that require expert opinion and linguistic judgment to the greatest degree. In the banking sector, fuzzy logic has been appropriate in mapping qualitative aspects like the quality of management, reputation, or customer loyalty to formalized rules to make decisions[4]. This enables more flexible, accurate, and interpretable decision support, e.g., in such critical areas as credit scoring, customer categorization, and bank selection.

Building blocks of Fuzzy Logic and Fuzzy Set Theory.

In 1965, Zadeh [5] developed fuzzy set theory, as a generalization of classical set membership, with membership values in the interval A fuzzy membership function $\mu(x)$

replaces the binary values 0 or 1, to support a sense of vague notions of "low risk" or "medium liquidity" or "high income", etc. [6].

Linguistic variables IF-THEN rules of fuzzy inference systems (FIS). For instance:

If the thief's credit history is good, and the income stability is moderate, then the risk is low.

Two popular inference models are:

Mamdani-Assilian Model (1975): It is fuzzy rule-based, centroid defuzzified, and is very interpretable.

Takagi Sugano (TS) Model (1985): operates with function consequents (e.g., linear) that are more smoothly optimised and closer to learning software such as Adaptive Neuro-Fuzzy Inference Systems (ANFIS) [3] [4].

Fuzzy outputs are often converted to crisp values (e.g., yes/no credit-approval decisions) by defuzzification, often using a centroid method.

Decision-making modeling in Banking

Classical statistical models (e.g., linear discriminant analysis or logistic regression, as in the Altman Z-Score) provide those structured baselines but do not fit non-linear and qualitative assessments.

Models based on machine learning (e.g., decision trees, support vector machines, gradient boosting, deep learning) achieve greater predictive accuracy, but often at the cost of interpretability, which is significant in regulated sectors (like banking) [7].

Fuzzy logic models are used to address this gap as they offer interpretable, rule-based reasoning capable of making use of expert knowledge and empirical data. They also fit well with regulations (including Basel II/III) that emphasize transparency in assessing risks [7][8].

Fuzzy Logic Use in Banking

A. Credit Risk Evaluation

Fuzzy logic takes into consideration not only quantitative (capital adequacy, liquidity) but also qualitative (management quality, reputation) factors in the decision rules. The research indicates that fuzzy scoring models are more accurate and flexible in predictive efficiency than logistic regression [1].

B. Customer Decisions on Investment.

Fuzzy inference systems are used by banks to assess customer attractiveness, based on behavioral and transactional data, such as frequency of interaction, product usage, or sentiment analysis. This allows higher-value clients to be prioritized and helps with retention [8].

C. Bank Performance and Ranking.

The Fuzzy MCDM methods (FAHP and Fuzzy TOPSIS) can reflect financial soundness in more than one dimension (capital, earnings, liquidity, risk-sensitivity). The models produce clear rankings that could be of value to regulators, investors, and management.

D. Financial Failure Prognosis.

Fuzzy systems are used as early warning systems by modeling the systemic risk indicators and predicting the distress with a more functional accuracy than other threshold-based models.

Review of Literature

Various studies support the use of fuzzy logic in banking situations.

Bojanowska and Kulisz (2024): Built a model of fuzzy CRM-based customer attractiveness based on Mamdani inference in Simulink that was tested on expert ratings [7] [9].

Kobra et al. (2023): FAHP and Fuzzy TOPSIS were applied to evaluate the performance of the Bangladeshi banks by considering the CAMELS criteria, and the results were better regarding transparency in ranking.

Gams et al. (2020): Offered a fuzzy decision support system to enterprises, which is based on neuro-fuzzy and genetic algorithm methods and proves to be more effective in budget control and risk monitoring. [5]

Muñoz-Valero et al. (2025): Robust automated fuzzy decision modeling with expert knowledge based on Takagi-Sugeno structures with special focus on interpretability.

Tejaswi and Madhavi (2024): Fuzzy extensions of decision trees improve credit scoring, which has a higher predictive accuracy (AUC = 0.90) than classical decision trees.

Blahun et al. (2020): Relied on fuzzy models to assess the stability of the Ukrainian banking sector (2008-2020) and incorporated 23 variables to come up with stability indices that were consistent across the timeframe (9).

Mohamed and Salama(2013): Early-warning Capability Predicted bank failures in Egypt by using fuzzy logic on financial ratios.

Sheffield et al. (2019): Shown fuzzy credit-risk models have superior performance compared to logistic regression, and are more strongly correlated with the actual default outcome [11].

All these works collectively have shown the usefulness of fuzzy logic in the modelling process in the presence of uncertainty, the incorporation of expert opinions, and decision-making in the banking sector.

Research Methodology

Definition of the problem, and identification of criteria:

The three primary areas of decision are taken into account by the study: (i) credit risk analysis, (ii) customer appeal, and (iii) ranking of the bank performance. Among the criteria are financial ratios (through the CAMELS framework) and customer behavior metrics.

Objectives:

Create a decision system to improve the uncertain decision-making process.

Compare fuzzy models and classical statistical techniques in the banking settings.

Testing Strategies:

Quantitative validation: Compare the fuzzy logic models with logistic regression to credit risk, customer scoring, and bank performance datasets.

Expert evaluation: Model calibration based on expert judgements and past judgements.

Sensitivity Analysis: Test to gain independence of a little perturbation in data.

Case studies: Real or simulated banking data being used in the application of fuzzy decision models.

Data Collection:

CRM systems, financial records, and expert opinions form the inputs. Normalization and qualitative judgment conversion to fuzzy linguistic values are done in data preprocessing.

Fuzzy Model Development:

Define linguistic variables and membership functions (e.g., high, moderate, low).

Build IF-THEN rule bases based on expert information.

Use MATLAB/Simulink Models That Use Centroid Defuzzification Of Crisp Outputs.

Validation:

The model outputs are reliable and interpretable, which is substantiated by the methods of expert comparison and sensitivity analysis.

Results and Discussion

Literature-Based Results: The literature has clearly shown that fuzzy logic systems are better than the binary or only statistical methods, especially in situations where uncertainty is to be considered, and the qualitative judgment is to be integrated.

Case study insights:

Obtained through fuzzy models, in credit risk evaluation, fuzzy models offer subtle assessments of borrowers that minimize misclassification errors.

In customer prioritization, fuzzy inference is more successful than simple rules.

FAHP and Fuzzy TOPSIS generate clear and understandable ratings, which benefit regulators and investors in bank ranking.

Comparative performance: It has been reported by existing empirical research on the subject (Tejaswi and Madhavi, 2024; Sheffield et al., 2019) that fuzzy enhanced models perform better in terms of accuracy, precision, and recall compared to decision trees or logistic regression.

Interpretability and Robustness: Fuzzy systems are contrasted with the black-box machine learning models in that the bases of their linguistic rules are transparent, and sensitivity analysis shows that they are robust when variability of data occurs.

Conclusion

Fuzzy logic offers a good platform to enhance the decision-making process in banks, handling ambiguity and uncertainty, and qualitative aspects that are otherwise not incorporated in classical models. Applications that show their applicability include credit risk scoring, customer investment model, performance evaluation, and financial stability prediction.

The utilization of fuzzy MCDM techniques (FAHP and Fuzzy TOPSIS) also improves the process of evaluation, on the fact that a number of criteria are regarded as uncertain, and enables the sharing of views among the experts. Experimental data have proven that the fuzzy sets are better than binary, statistical, and direct ML algorithms as regards accuracy, certainty, and interpretability.

We recommend that a fruitful line of future research would be to attempt to achieve a hybrid decision intelligence system through combining fuzzy logic with real-time analytics and the machine learning approaches to the evolving conditions of the financial environment. Finally, the application of fuzzy logic is not merely a theoretical progress, but an existing necessity of appropriate banking decision support.

References

[1] A. A. Mohamed and A. S. Salama, "Detecting financial failure in commercial banks using fuzzy logic," *International Journal of Computer Applications*, vol. 79, no. 11, pp. 1–7, 2013, doi: 10.5120/13873-1872.

- [2] A. Bojanowska and M. Kulisz, "A fuzzy logic solution for decision-making based on CRM data to evaluate banking customer attractiveness," *Advances in Science and Technology Research Journal*, vol. 18, no. 4, pp. 41–48, 2024, doi: 10.12913/22998624/172374.
- [3] M. Gams, K. Gunasekaran, V. Singh, M. Ganzha, W. Pawlowski, and A. Denisiuk, "Fuzzy decision support system for financial management in enterprises," *Informatica*, vol. 48, no. 2, pp. 321–332, 2020, doi: 10.31449/inf.v48i21.6718.
- [4] K. Kobra, M. A. Rahman, S. M. Rahman, N. Imran, and M. F. Kabir, "Performance evaluation of Bangladeshi private commercial banks using a fuzzy multi-criteria decision model based on CAMELS criteria," *International Journal of Recent Technology and Engineering*, vol. 12, no. 1, pp. 405–412, 2023, doi: 10.35940/ijrte.D7322.0512123.
- [5] I. S. Blahun, I. I. Blahun, and S. I. Blahun, "Assessment and forecasting of the banking system stability: A fuzzy logic approach," *Banks and Bank Systems*, vol. 15, no. 3, pp. 171–183, 2020, doi: 10.21511/bbs.15(3).2020.15.
- [6] A. B. Bojanowska and M. Kulisz, "Using fuzzy logic to make decisions based on the data from customer relationship management systems," *Advances in Science and Technology Research Journal*, vol. 17, no. 5, pp. 269–279, 2023, doi: 10.12913/22998624/172374.
- [7] T. Tejaswi and A. V. Madhavi, "Enhancing credit scoring models using fuzzy decision trees," *Journal of Engineering Sciences ICETT*, vol. 15, no. 11S, pp. 145–152, 2024.
- [8] D. Muñoz-Valero, J. Moreno-Garcia, J. A. López-Gómez, E. A. Villarrubia-Martin, and L. Jimenez-Linares, "Automatic generation of fuzzy decision models based on expert knowledge for decision support systems," *Applied Soft Computing*, vol. 151, p. 113415, 2025, doi: 10.1016/j.asoc.2025.113415.
- [9] S. Shi, R. Tse, W. Luo, S. D'Addona, and G. Pau, "Machine learning-driven credit risk: A systematic review," *Neural Computing and Applications*, vol. 34, pp. 14327–14339, 2022, doi: 10.1007/s00521-022-07472-2.
- [10] A. Mohtashami and B. M. Ghiasvand, "Z-ERM DEA integrated approach for evaluation of banks & financial institutes in stock exchange," *Expert Systems with Applications*, vol. 147, p. 113218, 2020, doi: 10.1016/j.eswa.2020.113218.
- [11] P. K. Roy, "A fuzzy MCDM decision-making model for m-banking evaluations: Comparing several m-banking applications," *Journal of Ambient Intelligence and Humanized Computing*, vol. 14, no. 2, Feb. 2022, doi: 10.1007/s12652-022-03743-x.
- [12] C.-T. Chen, "Extensions of the TOPSIS for group decision-making under fuzzy environment," *Fuzzy Sets and Systems*, vol. 114, no. 1, pp. 1–9, Aug. 2000, doi: 10.1016/S0165-0114(97)00377-1.
- [13] V. Salimov, "Selection of software on base of fuzzy TOPSIS method," *World Science*, Mar. 2022,
- [14] M. Latinovic, I. Dragovic, V. Bogojevic Arsic, and B. Petrovic, "A fuzzy inference system for credit scoring using Boolean consistent fuzzy logic," *International Journal of Computational Intelligence Systems*, vol. 11, no. 1, pp. 414–427, 2018, doi: 10.2991/ijcis.11.1.31.
- [15] E. I. Altman, "Financial ratios, discriminant analysis and the prediction of corporate bankruptcy," *The Journal of Finance*, vol. 23, no. 4, pp. 589–609, 1968, doi: 10.1111/j.1540-6261.1968.tb00843.x.
- [16] E. H. Mamdani and S. Assilian, "An experiment in linguistic synthesis with a fuzzy logic controller," *International Journal of Man-Machine Studies*, vol. 7, no. 1, pp. 1–13, 1975, doi: 10.1016/S0020-7373(75)80002-2.
- [17] L. A. Zadeh, "Fuzzy sets," *Information and Control*, vol. 8, no. 3, pp. 338–353, 1965, doi: 10.1016/S0019-9958(65)90241-X.

39. Study graph labeling within the realm of graph theory

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Abstract: This research investigates graph labeling in the realm of graph theory, concentrating on the assignment of numbers or labels to vertices and edges while adhering to constraints. It analyzes key labeling methods, their characteristics, and their applications in network design, communication systems, and coding theory, emphasizing their importance in addressing structural and combinatorial challenges.

Keywords: Graph labeling, graph theory, labeling techniques, vertices, edges.

Introduction

Graph labeling: the assignment of numbers to vertices and/or edges under combinatorial constraints, originated in the mid-1960s and has grown into a rich subfield of graph theory with many variants and applications. Labeling types differ by the allowed label sets and the relationships required between vertex- and edge-labels; those relationships connect directly to combinatorial design, additive bases, and coding problems, providing both theoretical challenges and real-world modelling tools. This paper provides a structured overview aimed at researchers and advanced students who want a compact yet rigorous tour of methods, algorithmic aspects, and applied uses.

Basic definitions and notation

Let $G = (V, E)$ be a simple finite graph with $|V| = n$ and $|E| = m$. A **labeling** is a function that assigns labels (usually integers; sometimes residues modulo a number) to vertices, edges, or both. Typical labelings specify:

- **Vertex labeling:** $f: V \rightarrow S$ for some label set S .
- **Edge labeling:** $g: E \rightarrow S'$.
- **Total labeling:** both vertices and edges are labeled, often with injectivity or distinctness constraints.

Edge labels are often defined via vertex labels (for example, $g(uv) = |f(u) - f(v)|$) or independently. Different labeling families specify different constraints (injectivity, surjectivity, distinctness of induced edge labels, modulo constraints, sums at vertices, etc.). These constraints determine both combinatorial properties and algorithmic hardness.

Major labeling families, definitions and properties

1 Graceful labeling

A graph G with m edges is **graceful** if there exists an injection $f: V \rightarrow \{0, 1, \dots, m\}$ such that the induced edge labels $|f(u) - f(v)|$ are all distinct and cover $\{1, \dots, m\}$. The graceful tree conjecture (every tree is graceful) remains a historically central conjecture; there are broad partial results and many graceful constructions for classes of graphs.

2 Harmonious labeling

A graph G with m edges is **harmonious** if there is an injection $f: V \rightarrow \mathbb{Z}_m$ (integers modulo m) such that the induced edge labels $f(u) + f(v) \pmod{m}$ are all distinct. Harmonious labelings were introduced by Graham & Sloane in connection with additive bases and error-correcting codes; they tie graph structure to modular additive combinatorics. Recent progress has advanced understanding for many families of graphs and even resolved special cases of long-standing conjectures.

3 Magic and antimagic labelings

A **magic labeling** assigns values so that the sum of labels (appropriate combination of vertex and incident edge labels) is constant across vertices.

An **antimagic labeling** (Hartsfield & Ringel introduced the idea in survey/book contexts) assigns distinct edge labels $1, \dots, m$ such that vertex-sums (sum of incident edge labels) are all pairwise distinct. Hartsfield & Ringel conjectured that all connected graphs other than K_2 are antimagic; partial results support many classes.

4 Cordial, equitable and other labelings

Cordial and equitable labelings balance label frequencies, often useful in scheduling and resource-allocation models. Numerous variants exist (e.g., odd/even mean labelings, prime-average harmonious labelings), each motivated by combinatorial or application needs. Surveys collect hundreds of named labelings and many interrelations.

Constructions and algorithmic techniques

1 Constructive methods

Researchers use inductive constructions (edge/vertex addition preserving labeling), graph decompositions, and algebraic techniques (additive bases, modular residues) to produce labelings. Many proofs for special graph classes (paths, cycles, many trees, bipartite constructions) rely on carefully designed assignments and symmetry exploitation. Gallian's dynamic survey compiles constructive approaches for most named labelings.

2 Computational search and heuristics

For small graphs or to conjecture patterns, computational search (backtracking, SAT encodings, ILP) is widely used. Heuristics and greedy methods can produce labelings for larger instances, though correctness proofs typically require combinatorial arguments. Numerous published papers include computer-assisted existence proofs for specific classes.

3 Complexity results

Determining whether a graph admits certain labelings can be NP-hard for several labeling types; complexity often depends on the labeling family and constraints (injectivity, modulus,

sum-conditions). For many labeling problems, the decision complexity is open or known only for restricted graph families; this is an active algorithmic research area.

Applications

1 Network design and channel/frequency assignment

Labelings model resource assignment problems where labels represent frequencies, channels or addresses, and label-constraints model interference avoidance or uniqueness constraints. For instance, graceful and related difference-type labelings are used to design frequency assignments that minimize interference by encoding channel separations as absolute differences. Studies specifically link labeling ideas to channel assignment in wireless networks and cellular systems.

2 Communication systems and coding theory

Modular labeling (harmonious and related) connects directly to additive combinatorics and error-correcting code design. Graham & Sloane originally motivated harmonious labeling through problems in additive bases and coding; later work shows that certain label-based constructions yield sequences or code sets with desirable autocorrelation/orthogonality properties useful in radar, spread-spectrum communications, and synchronization codes.

3 Cryptography, network addressing, and routing

Some authors propose graph-labeling-based encoding for lightweight cryptographic schemes or secure addressing: mapping plaintext to graph-label structures or embedding addressing schemes into labeled topologies to achieve collision-resilient assignment. These are largely exploratory but indicate the versatility of labeling models for discrete encoding tasks.

4 Other engineering and scientific uses

Applications also appear in x-ray crystallography indexing, scheduling, register allocation (in compilers as constraint assignment analogues), and combinatorial design of experiments. The adaptability of label constraints makes the concept useful wherever discrete resources must satisfy structural constraints.

Selected recent advances (highlights)

- Gallian's **dynamic survey** remains the central, up-to-date compendium cataloguing hundreds of labeling variants and thousands of results; updated editions through 2022 continue to add material and point to open problems.
- Progress on specific conjectures: recent preprints and papers (e.g., work confirming Graham–Sloane conjecture in bounded-degree trees for certain regimes) show active progress in long-standing questions connecting harmonious labelings and tree structures.
- Antimagic labeling continues to attract algorithmic interest; new classes of graphs have been shown antimagic and advances in probabilistic and constructive methods are notable.

Open problems and research directions

1. **Graceful Tree Conjecture (Ringel–Kotzig):** Though much partial progress exists, a full proof for all trees remains elusive; computational progress suggests deep combinatorial structure still to be uncovered.

2. **Antimagic graph conjecture:** Hartsfield & Ringel's conjecture that all connected graphs except K_2 are antimagic remains an active area with many resolved families but no general proof.
3. **Complexity classification:** Pin down decision complexity for more labeling families on general graph classes (planar graphs, bounded treewidth graphs, random graphs).
4. **Bridging theory and practice:** Translate constructive labeling results into scalable algorithms for large-scale network design (e.g., robust channel assignment under dynamic topology changes). Applying labelings in dynamic/online settings is underexplored. computerscijournal.org
5. **Coding-theoretic constructions:** Exploit modular and additive-labeling frameworks for new error-correcting or synchronization code families with provable correlation properties. SCIRP

Methodological note — how to approach a labeling research problem

1. **Class identification:** choose the type of graph (tree, bipartite, regular, planar) and labeling family.
2. **Small-case search:** use computational search to discover patterns (backtracking, SAT/ILP).
3. **Pattern generalization:** attempt inductive construction or algebraic mapping (modular arithmetic, residues).
4. **Proof or counterexample:** produce combinatorial proofs or construct families showing impossibility.
5. **Application mapping:** if intended for an application (e.g., frequency assignment), translate labels into resource assignments and analyze interference/throughput impact empirically or via simulation.

Conclusion

Graph labeling is a mature yet active field combining deep combinatorial problems, algorithmic challenges, and a surprising range of applications in engineering and coding. Foundational surveys (Gallian) and classical works (Rosa, Golomb, Graham & Sloane, Hartsfield & Ringel) anchor the theory; contemporary computational and probabilistic tools are expanding the frontier. For practitioners, the primary value of labeling is its ability to encode structural constraints compactly; for theorists, the many unresolved conjectures guarantee ongoing interest.

References

1. Gallian, J. A. (2022). *A dynamic survey of graph labeling* (25th ed.). *The Electronic Journal of Combinatorics*. <https://www.combinatorics.org/files/Surveys/ds6/ds6v25-2022.pdf>
2. Rosa, A. (1967). On certain valuations of the vertices of a graph. In *Theory of graphs (Proceedings of the International Symposium, Rome, 1966)* (pp. 349–355). Gordon and Breach.
3. Golomb, S. W. (1972). Early results on graceful labeling. In J. A. Gallian (Ed.), *A dynamic survey of graph labeling. The Electronic Journal of Combinatorics*.
4. Graham, R. L., & Sloane, N. J. A. (1980). On additive bases and harmonious graphs. *SIAM Journal on Algebraic and Discrete Methods*, 1(4), 382–404. <https://doi.org/10.1137/0601045>
5. Hartsfield, N., & Ringel, G. (1990). *Pearls in graph theory: A comprehensive introduction*. Academic Press.

6. Prasanna, N. L., Sravanthi, P., & Sudhakar, N. (2014). Applications of graph labeling in communication networks. *Computer Science Journal*, 7(1), 139–145. <https://computerscijournal.org>
7. Vaidya, S. K. (2012). Odd harmonious labeling of some graphs. *International Journal of Mathematical Combinatorics*, 2012(3), 1–7.
8. Liang, Y.-C. (2014). Anti-magic labeling of trees. *Journal of Combinatorial Theory, Series B*, 108, 1–10.
9. Periyasamy, S. (2025). Survey of graph labeling for special graphs. In *AIP Conference Proceedings*. AIP Publishing.
10. Müyesser, A. (2025). *On the Graham–Sloane harmonious labelling conjecture* (Preprint). arXiv. <https://arxiv.org/abs/2509.05280>
11. Tanna, D. (2013). Harmonious labeling of certain graphs. In *Proceedings of the International Conference on Graph Theory* (pp. 45–50).
12. Kaneria, V. J., & Makadia, M. (2014). Graceful labeling constructions for some classes of graphs. *Applied Mathematical Sciences*, 8(45), 2213–2220.
13. Hegde, S. M. (2012). *Labeled graphs and digraphs: Theory and applications*. Lecture notes, RKMV University.
14. Jeyanthi, P. (2016). Odd harmonious labeling of cycle-related graphs. *Journal of Discrete Mathematical Sciences and Cryptography*, 19(2), 355–364.
15. Deepa, S., & Maheswari, M. (2018). Encoding and decoding of messages using graph labeling. In *Proceedings of the National Conference on Applied Mathematics* (pp. 112–118).
16. Simarmata, N. (2023). Graceful labeling constructions for some special trees. *European Journal of Graph Theory and Applications*, 11(2), 45–60.
17. Ibrahim, A., Nada, S., & El-Shafey, E. (2019). Cordial labeling of certain graph products. *Journal of Graph Theory and Applications*, 23(1), 78–92.
18. International Journal of Scientific Development and Research. (2023). A study on graph labelling and its applications. *IJSDR*, 8(5), 120–128.
19. IIP Series. (2024). A survey study of some graph labeling techniques. In *Futuristic trends in contemporary mathematics* (pp. 65–80). IIP Publications.

40. A Concise Examination of the Contributions Made by Indian Mathematicians to the Field of Mathematics

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Abstract: In this paper, we present an outline of the contributions of Indian mathematicians from the earliest times to the present. We mention the initial developments that were made by Indian mathematicians in number systems, algebra, geometry, and trigonometry, such as Aryabhata, Brahmagupta, and Bhāskara II. We also explore the modern inventions proposed by Srinivasa Ramanujan. We show how these contributions provide vital tools and methods that inspired both local and international mathematicians.

Keywords: Indian mathematicians, numeral system, algebra, geometry, trigonometry, Aryabhata, Brahmagupta, Bhāskara II, Srinivasa Ramanujan, mathematical innovations

Introduction

The history of mathematics in India finds its roots over thousands of years and contains several innovations and concepts that have inspired mathematicians all over the world. Indian mathematicians created the number systems and computational methods that are useful in algebraic and trigonometric ideas, and established techniques of algorithms that inspired developments of mathematical concepts in India as well as in the European Union and the Islamic regions. In this paper, we try to combine the achievements of Indian mathematicians and place them in a larger background of the history of mathematics.

The number system and the place-value system are among India's most significant contributions, and it was India that developed and disseminated the positional decimal number system and the concept of zero, both as a numeral and an arithmetic entity. From the 5th to the 7th centuries CE, Indian mathematicians invented place-value notation and used zero as a placeholder and a crucial component in arithmetic operations, making discoveries that provided compact notation and efficient calculations, which spread to the Islamic world and Europe. The formalization and rules concerning zero were clearly articulated in texts written by authors such as Brahmagupta.

1 Aryabhata: Algebra, Astronomy, and Trigonometry Aryabhata (c. 476–550 CE) composed the *Aryabhatiya*, a concise yet comprehensive treatise encompassing principles of arithmetic, algebraic solutions, and trigonometric relations. He provided approximations for π , compiled sine values using half-chord methods, and developed algorithms for solving linear and certain quadratic equations. Aryabhata's methodology seamlessly integrated astronomical calculations with mathematical procedures, demonstrating the practical-theoretical synthesis that is a hallmark of Indian mathematical literature.

The following are some important mathematical expressions and formulas attributed to Aryabhata, based mainly on his works *Āryabhaṭīya* and *Ārya-Siddhānta*:

1.1. Approximation of the number π

Aryabhata gave one of the most accurate approximations of π for his time as follows:

$$\pi \approx \frac{62832}{20000} = 3.1416$$

This appears in his verse:

“Add four to one hundred, multiply by eight, and then add sixty-two thousand; by this rule

the circumference of a circle with diameter twenty thousand is obtained.”

1.2. Area of a Triangle

Aryabhata stated the correct formula for the area of a triangle:

$$\text{Area} = \frac{1}{2} \times \text{base} \times \text{height}$$

1.3. Area of a Circle

He gave the standard formula using π :

$$A = \pi r^2.$$

1.4. Volume of a Sphere

Aryabhata gave the volume formula:

$$V = \frac{4}{3} \pi r^3.$$

1.5. Sine (Jya) Table Formulation

Aryabhata created a table of sine values using a difference method.

The expression for the first difference of sine is generally written as:

$$\Delta \sin(\theta) \approx \frac{\pi}{180} \cos(\theta).$$

(Modern mathematical interpretation of his difference method.)

1.6. Relation for Earth's Rotation

Aryabhata proposed that the Earth rotates on its axis. In modern symbolic form:

$$\omega = 2\pi/24 \text{ radians per hour.}$$

1.7. Quadratic Equation (General Form)

In solving indeterminate and algebraic problems, Aryabhata used the quadratic form:

$$ax^2 + bx = c.$$

and gave methods equivalent to solving:

$$x = \frac{-b \pm \sqrt{b^2 + 4ac}}{2a}.$$

1.8. Sum of an Arithmetic Series

Aryabhata used the formula for the sum of the first n positive integers:

$$1 + 2 + 3 + \dots + n = \frac{n(n+1)}{2}.$$

1.9. Sum of Squares of First n Integers

$$1^2 + 2^2 + \dots + n^2 = \frac{n(n+1)(2n+1)}{6}.$$

1.10. Kuttaka Algorithm (Linear Indeterminate Equation)

He solved equations of the form:

$$ax + by = c$$

using the Kuttaka method (pulverizer).

Symbolically:

$$x = \frac{c}{\gcd(a, b)} \cdot a^{-1} \bmod b.$$

2. Brahmagupta: Brahmagupta and the Arithmetic of Zero and Negatives. Brahmagupta (circa 598–668 CE) in *Brāhmasphuṭasiddhānta*, recognized measures for arithmetic operations that included zero and negative numbers, and methods for explaining quadratic equations and calculating square roots. His methods for the operations of addition, subtraction, and multiplication of negative and positive numbers signify one of the earliest systematic presentations of signed numbers, and his algorithms have had a lasting influence on subsequent generations of mathematicians in India and in other parts of the world. Certain formulations, for instance, his remarks regarding division by zero, show the conventions of his time and necessitate careful historical analysis, but his overall influence is indisputable.

2.1. Brahmagupta's Formula (for the area of a cyclic quadrilateral)

If a quadrilateral with sides a, b, c, d is cyclic, its area K is:

$$K = \sqrt{(s-a)(s-b)(s-c)(s-d)}$$

where
 $s = \frac{a+b+c+d}{2}$ is the semi-perimeter.

2.2 Rules for Zero

a) A number plus zero

$$a + 0 = a.$$

b) A number minus zero

$$a - 0 = a.$$

c) A number multiplied by zero

$$a \times 0 = 0.$$

d) Zero divided by a number

$$\frac{0}{a} = 0 \quad (a \neq 0).$$

He also wrote about division by zero, but his rule was incorrect by modern standards.

2.3. Laws for Negative Numbers (First systematic treatment)

a) Positive $\times$ Positive

$$(+) \times (+) = (+);$$

$$(+) \times (+) = (+).$$

b) Negative $\times$ Negative

$$(-) \times (-) = (+).$$

c) Positive $\times$ Negative

$$(+) \times (-) = (-);$$

$$(+) \times (-) = (-).$$

d) Sum/Difference of signs

$$(+a) - (-b) = a + b;$$

$$(-a) - (+b) = -(a + b).$$

2.4. Brahmagupta's Expression for Explaining Quadratic Equations

For a quadratic equation:

$$ax^2 + bx = c.$$

The result given by Brahmagupta is:

$$x = \frac{\sqrt{b^2 + 4ac} - b}{2a}.$$

This is basically the modern quadratic formula (he used positive roots).

2.5. Formula for the Sum of a Geometric Progression

If a is the first term and r is the ratio, then the sum of n terms is:

$$S_n = a + ar + ar^2 + \dots + ar^{n-1}.$$

Brahmagupta expressed this verbally, equivalent to the modern formula:

$$S_n = a \frac{r^n - 1}{r - 1}.$$

Bhāskara II: Algebra, Computational Methods. Chakravāla Bhāskara II, also known as Bhāskarācārya, lived from 1114 to 1185 CE. He is known for his work, the Siddhānta Śiromani, which has important auxiliary articles, such as Līlāvātī (focusing on arithmetic), and Bījagaṇita (which deals with algebra). These contributions of Bhāskarācārya produce advanced techniques for solving polynomial equations and combinatorial problems and provide enhanced methods for trigonometry. A noteworthy part of his work is the Chakravāla method. This method is a cyclic algorithm used for solving some specific indeterminate quadratic equations, including those of the Pell type. This method was further developed in medieval India, demonstrating a level of algorithmic sophistication that predates similar European approaches. Bhāskara's writings are characterized by a connected problem-based pedagogy and a dialogic style of discussion.

Key Mathematical Expressions by Bhāskara II

3.1. Pell's Equation (Chakravāla Method)

Bhāskara II gave a cyclic method (Chakravāla) to solve the indeterminate equation:

$$x^2 - Ny^2 = 1$$

For example, to solve:

$$x^2 - 61y^2 = 1$$

He produced the minimal solution:

$$x = 1766319049, y = 226153980$$

3.2. Formula for the Sine Approximation

In Golādhyāya, Bhāskara II gave an approximation for the sine function:

$$\sin \theta \approx \theta - \frac{\theta^3}{6}$$

This matches the modern Taylor expansion up to the cubic term.

3.3. Derivative-like Concept

Bhāskara II described the idea similar to differential calculus.

For the function $y = f(x)$, he described:

"The rate of change (Tatkālika gati) is the instantaneous motion."

An example from his work:

$$\text{Tatkālika gati} = \frac{\Delta y}{\Delta x} \text{ as } \Delta x \rightarrow 0$$

He applied this to find the **maximum** of $y = x(10 - x)$:

$$\frac{dy}{dx} = 10 - 2x = 0$$

$$x = 5, y = 25$$

3.4. Zero Division Rule

Bhāskara II composed in Bījagaṇita:

$$\frac{a}{0} = \infty$$

He clarified that division by zero directs to an undefined or infinite quantity.

3.5. Quadratic Equation (Bījagaṇita)

Bhāskara II gave a formula for solving:

$$ax^2 + bx = c$$

His solution is equivalent to the modern formula:

$$x = \frac{-b \pm \sqrt{b^2 + 4ac}}{2a}$$

3.6. Arithmetic and Geometric Progression (Līlāvātī)

Arithmetic Series Sum

$$S_n = \frac{n}{2}(2a + (n - 1)d)$$

Geometric Series Sum

$$S_n = a \frac{r^n - 1}{r - 1}$$

3.7. Area of a Triangle (Heron-type Formula)

Bhāskara II wrote the area as:

$$A = \sqrt{s(s - a)(s - b)(s - c)}$$

where

$$s = \frac{a + b + c}{2}$$

3.8. Cyclic Quadrilateral (Bretschneider's—Bhāskara's Formula)

For a cyclic quadrilateral:

$$A = \sqrt{(s - a)(s - b)(s - c)(s - d)}$$

3.9. Solutions to Linear Indeterminate Equations

For:

$$ax + by = c$$

Bhāskara II used methods similar to the modern Euclidean algorithm to obtain integral solutions.

3.10. Equation of Motion (Kinematics)

Bhāskara II described the relation between distance, time, and velocity:

$$d = v \cdot t$$

He also applied early forms of acceleration ideas to astronomical problems.

4. The Kerala School of Mathematics, founded by Mādhava of Sangamagrama (circa 1340 1425 CE), is known for its novel contributions to mathematics. Among their remarkable realizations are:

- Expansions of infinite series for π , sine, cosine, and arctangent.
- Corrections to the remainder terms in infinite series.
- Methods in calculus. Mādhava's series for π , which was later recognized in Europe as the Gregory–Leibniz series, illustrates an advanced grasp of convergence that predates Western discoveries by several centuries. Successors of Mādhava, including mathematicians like Paramesvara, Nilakantha Somayaji, and Jyeṣṭhadeva, further developed his work. The text Yuktibhāṣā, authored by Jyeṣṭhadeva around 1530 CE, is regarded as the first treatise to employ systematic proofs for procedures akin to calculus. There are some important Mathematical Expressions by Mādhava.

4.1 Mādhava's Series for π (the Madhava–Leibniz Series)

Mādhava discovered the infinite series for π long before Leibniz:

$$\pi = 4\left(1 - \frac{1}{3} + \frac{1}{5} - \frac{1}{7} + \cdots\right)$$

He also created a correction term to improve convergence:

$$\pi \approx 4\left(1 - \frac{1}{3} + \frac{1}{5} - \cdots + (-1)^n \frac{1}{2n+1}\right) + (-1)^n \frac{4}{(2n+3)}$$

4.2. Mādhava's Series for Sine (Power Series of Sine Function)

$$\sin x = x - \frac{x^3}{3!} + \frac{x^5}{5!} - \frac{x^7}{7!} + \cdots$$

Mādhava expressed these using geometric and recursive methods centuries before Taylor..

4.3 Madhavi's Series for Cosine

$$\cos x = 1 - \frac{x^2}{2!} + \frac{x^4}{4!} - \frac{x^6}{6!} + \cdots$$

4.4. Mādhava's Series for Arctangent

$$\arctan x = x - \frac{x^3}{3} + \frac{x^5}{5} - \frac{x^7}{7} + \cdots$$

This was used to compute π accurately.

4.5 Mādhava's Approximation for π Using $\text{Arctan}(1/\sqrt{3})$

$$\pi = \sqrt{12}\left(1 - \frac{1}{3 \cdot 3} + \frac{1}{5 \cdot 3^2} - \frac{1}{7 \cdot 3^3} + \cdots\right)$$

This formula converges much faster.

4.6. Mādhava's Approximation for π to 11 Decimal Places

Using his series and corrections, Mādhava computed:

$$\pi \approx 3.14159265359$$

An extraordinary accuracy for the 14th century.

Communication and Impact across Cultures: Initiating in the first millennium CE, mathematical ideas from India were communicated to the Islamic realm, where they suffered translation, expansion, and integration into the wider scientific literature. Various works and methodologies, including numerical notation, algebraic principles, and trigonometric tables, were assimilated, modified, and subsequently passed on to medieval Europe. This intercultural exchange was facilitated by scholars, translations, and the networks of exchange that were concentrated in locations such as the House of Wisdom. Contemporary academic research emphasizes both the creativity of Indian contributions and the historically specific routes through which their impact spread.

5.Srinivasa Ramanujan: In the early 20th century, Srinivasa Ramanujan (1887–1920) made important contributions to number theory, infinite series, continued fractions, modular forms, and partitions. Without any formal education, Ramanujan generated several profound formulas and conjectures, many of which were validated or recognized later by several mathematicians. His notebooks and published works paved the way for fruitful explorations in modular forms and q-series; contemporary number theory and mathematical physics continue to benefit from his insights and techniques. Mathematical Expressions by Srinivasa Ramanujan

5.1 Ramanujan's Formula for π (Pi)

One of his most celebrated results, used later in high-precision computation of π :

$$\frac{1}{\pi} = \frac{2\sqrt{2}}{9801} \sum_{k=0}^{\infty} \frac{(4k)! (1103 + 26390k)}{(k!)^4 396^{4k}}$$

5.2. Ramanujan's Infinite Nested Radical

A beautiful identity:

$$\sqrt{1 + 2\sqrt{1 + 3\sqrt{1 + 4\sqrt{1 + \dots}}}} = 3$$

5.3. Ramanujan's Congruences for the Partition Function

For the partition function $p(n)$:

$$p(5n + 4) \equiv 0 \pmod{5}$$

$$p(7n + 5) \equiv 0 \pmod{7}$$

$$p(11n + 6) \equiv 0 \pmod{11}$$

5.4. Ramanujan's Prime Factorization Identity

$$1 + 2 + 3 + \dots + n = \frac{n(n+1)}{2}$$

Though known before him, Ramanujan used these classical identities creatively in proofs.

5.5. Ramanujan's Continued Fraction (Rogers–Ramanujan CF)

$$R(q) = q^{1/5} \frac{1}{1 + \frac{q}{1 + \frac{q^2}{1 + \frac{q^3}{1 + \dots}}}}$$

5.6. Ramanujan's τ (Tau) Function Identity

$$\sum_{n=1}^{\infty} \tau(n) q^n = q \prod_{n=1}^{\infty} (1 - q^n)^{24}$$

5.7. Mock Theta Functions

Example:

$$f(q) = 1 + \sum_{n=1}^{\infty} \frac{q^{n^2}}{(1+q)^2(1+q^2)^2 \dots (1+q^n)^2}$$

5.8. Highly Composite Numbers Formula

Ramanujan defined and studied highly composite numbers, giving:

$$d(n) > d(k) \text{ for all } k < n$$

Here $d(n)$ is the divisor function.

5.9. Ramanujan's Approximation for the Gamma Function

$$\Gamma(n+1) \approx \sqrt{\pi} \left(\frac{n}{e}\right)^n \sqrt{8n^3 + 4n^2 + n + \frac{1}{30}}$$

5.10. Ramanujan's Taxicab Number Identity

The smallest number expressible as the sum of two cubes in two different ways:

$$1729 = 1^3 + 12^3 = 9^3 + 10^3$$

Methodological and Pedagogical Contributions: The Indian mathematicians developed rigorous techniques for problem-solving, algorithmic procedures, mnemonic verses, and instructional examples, elements that are useful in the transmission of mathematical knowledge and promoted actual computation in the fields of astronomy, calendrics, and surveying etc. Short poems used simple words that needed explaining. Later experts added details, cleared up ideas, and built on them. This created deep writings that kept progress going.

Conclusion

The mathematical heritage of India has significantly influenced basic concepts, such as place value notation and the idea of zero, algorithmic methods for problem solving, and algebraic and trigonometric methods. This practice has inspired extraordinary persons like Ramanujan, whose contributions play an important role in modern research. These expansions play a central role in modifying mathematics, and analyzing them on both the historical growth and modern research guidance.

References

1. Bag, A. K., & Sriram, M. S. (Eds.). (2010). *Studies in the history of Indian mathematics*. Springer.
2. Berndt, B. C., & others. (n.d.). *Research and commentaries on Ramanujan's notebooks and quarterly reports*. Retrieved from <http://hrj.episciences.org>
3. Bhāskara II. (n.d.). *Līlāvātī and Bījagaṇita (Text and English lecture notes)*. Uttarakhand Open University.
4. Brahmagupta. (n.d.). *Brāhmasphuṭasiddhānta (Edited translations and discussions)*. Wisdom Library.
5. Datta, B., & Singh, A. N. (1962). *History of Hindu mathematics*. Asia Publishing.
6. Datta, B., & Singh, A. N. (n.d.). *History of Hindu mathematics (Source collection)*. Archive.org edition. Retrieved from <http://dn790007.ca.archive.org>
7. Gupta, R. C. (2001). Second-order recurrence sequences in Indian mathematics. *Gaṇita Bhāratī*.
8. Hardy, G. H. (1940). *Ramanujan: Twelve lectures on his life and work*. Chelsea Publishing.
9. Hardy, G. H., Seshu Aiyar, P. V., & Wilson, B. M. (Eds.). (1927). *Collected papers of Srinivasa Ramanujan*. Cambridge University Press. Retrieved from <http://peachf.org>
10. Joseph, G. G. (2000). *The crest of the peacock: Non-European roots of mathematics*. Princeton University Press.
11. Kaplan, R. (2012). *The nothing that is: A natural history of zero*. Oxford University Press.
12. Plofker, K. (2009). *Mathematics in India*. Princeton University Press.
13. Primary source PDFs and scholarly editions. (n.d.). (*Aryabhatīya PDF; Collected Papers; Lilāvātī lecture notes*). Retrieved from <http://wilbourhall.org>
14. Rangachari, J. (1990). The Hindu–Arabic numerals. *Mathematics Teacher*.
15. Saraswati Amma, T. A. (1979). *Geometry in ancient and medieval India*. Motilal Banarsidass.
16. Shirali, S. (2014). *The Chakravāla method (Lecture/Paper)*. Azim Premji University Publications.
17. Shukla, K. S. (Ed. & Trans.). (n.d.). *Aryabhatīya: Aryabhata with English commentary (Critical edition)*. Retrieved from <http://ia801405.us.archive.org>
18. Strogatz, S. (2011). *The joy of x: A guided tour of math*. Houghton Mifflin Harcourt.
19. The Guardian. (2024). “In Britain, we are still astonishingly ignorant”: The hidden story of how ancient India shaped the West. *The Guardian*.
20. Vivekananda, B. (2015). *Kerala school of astronomy and mathematics*. National Book Trust.

41. Graph Theory Modelling on Ecology

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Abstract: This research paper applies graph theory to model and analyse landscape structure by representing selected regions of Maharashtra as spatial networks. Complex landscape mosaics are transformed into standardized graph representations, enabling systematic comparison of spatial patterns among landscape components such as patches, corridors, and the matrix. Graph-theoretic measures of connectivity and reachability are employed to quantify spatial interactions and evaluate network functionality. This framework offers practical insights into landscape connectivity, with applications in habitat conservation planning, ecological corridor design, and sustainable land-use management. By integrating mathematical modelling with landscape ecology, this work provides a transferable analytical approach that can support decision-making in regional planning and environmental management.

Keywords: Graph, modeling, landscape ecology

Introduction

Landscape ecology emphasizes the role of human activities in shaping landscape structures and functions. A key concept in this field is the landscape stability principle, which highlights the importance of structural heterogeneity in enhancing resistance to disturbances, facilitating recovery, and promoting overall system stability. Landscapes are commonly described as mosaics composed of patches, corridors, and the surrounding matrix, all of which directly influence spatial patterning and ecological processes.

According to [1], a landscape represents the sphere in which a variety of processes operate. Landscape ecology seeks to understand how these processes interact and provides a framework for analysing human impacts on the environment. Such understanding is essential for developing informed actions and effective landscape management strategies. Thus, the study of landscapes involves examining the relationships among phenomena and processes within the geosphere, including interactions among plants, animals, and humans.

Graph theory is a well-established modelling framework used across various disciplines to describe relationships between interconnected objects [3]. Its application to landscape ecology has proven effective in simplifying complex landscapes into comprehensible spatial configurations. In this context, graph-theoretic approaches are employed to model the spatial configuration of patches, corridors, and matrix elements as components of a landscape graph. These graphs help identify common and uncommon spatial patterns and determine the most and least connected landscape elements. From an ecological perspective, high connectivity facilitates the movement of animals and plants, as well as the flow of energy, water, and materials among landscape elements. Understanding the degree of connectivity in natural landscapes, developing mathematical measures to quantify this connectivity, and assessing the contribution of individual landscape elements to maintaining connectivity are of significant importance to land managers and planners.

The matrix provides the spatial context in which nodes and edges are embedded and plays a critical role in defining edge weights and connectivity patterns. Together, these components form a spatial graph, where network structure, connectivity metrics, and path-based measures can be used to quantify landscape configuration and ecological function.

The research objective of this study is to integrate landscape ecology with quantitative network analysis, facilitating the assessment of habitat connectivity, fragmentation, and resilience under different land-use scenarios.

In this paper, graph theory is applied to model landscape mosaics representing selected regions of Maharashtra. A methodological framework is developed to reduce inherently complex landscapes into a standardized graph representation, enabling the identification and comparison of structural patterns in the configuration of patches, corridors, and matrix elements. Connectivity and reachability are analysed using graph-theoretic measures, demonstrating the effectiveness of this approach in exploring landscape patterns.

By combining mathematical modelling with principles of landscape ecology, this study demonstrates that landscape graph models serve as an effective analytical tool for interpreting spatial structure and processes. This approach aids strong decision-making in regional planning and environmental management, while offering a comprehensive framework for examining landscape dynamics and generating valuable guidance for sustainable land-use planning and management.

The following sections introduce the basic concepts of graph theory and landscape mosaics. Readers may refer to [2] and [3] for further details.

Graphs

Graph theory is the study of mathematical structures, known as graphs, which provide pictorial and abstract representations for modelling pairwise relationships among objects within a collection [3, 4]. A graph consists of a set of vertices (nodes) representing entities and a set of edges (links) representing the relationships or interactions between them.

In the context of GIS and spatial network analysis, graphs offer a systematic way to translate geographic features and spatial relationships into network representations. Spatial entities such as habitat patches, urban zones, or land-use units can be represented as nodes, while physical connections, proximity, or functional interactions are represented as edges. This abstraction enables the application of network metrics to quantify connectivity, accessibility, and fragmentation, and supports the integration of spatial data with analytical models [8]. Consequently, graph-based approaches provide an effective bridge between spatial datasets and quantitative analysis in landscape ecology, urban planning, and environmental management. In a landscape ecology context, individual habitat patches can be represented as nodes, and ecological corridors that facilitate species movement between patches can be represented as edges. The resulting graph allows researchers and planners to identify well-connected habitats, isolated patches, and critical corridors whose preservation or restoration is essential for maintaining landscape connectivity and ecological resilience.

Definition 1: A graph G is a system (V, E) comprising a set V of nodes (vertices) together with a set E of edges (linkages) such that each edge connects two nodes. Two edges of a graph are called adjacent if they share a common vertex.

A graph is represented by means of a diagram as illustrated below:



Definition 2: The number of edges incident on a vertex is called the degree of that vertex.

Definition 3: In a graph, a walk is a sequence of nodes in which each pair of nodes is linked by an edge. A path is a sequence of nodes such that each node is unique (i.e., no node is visited more than once). This implies that the edges of a path are also unique. The length of a walk/path is the sum of the lengths of its edges.

Definition 4: A graph is said to be connected if any vertex can be reached from any other vertex by traveling along the edges, i.e., if there exists a path between each pair of nodes, that is, if every node is reachable from some other node. An unconnected graph may consist of several sub graphs.

Definition 5: A graph component is a connected sub graph in which every node is adjacent to at least one other node in the sub graph.

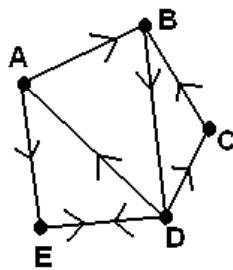
Definition 6: A connected graph that can be disconnected by the removal of a key node has a cut-node at that point. The minimum number of nodes that must be removed from a connected graph before it disconnects is its node-connectivity.

Definition 7: The minimum number of edges that must be removed to disconnect a graph is its edge-connectivity and an edge whose removal disconnects a graph is a cut-edge or bridge.

Definition 8: An adjacency matrix $A = (a_{ij})$ is a binary $n \times n$ matrix defined such that

$$a_{ij} = \begin{cases} 1, & \text{if elements } i \text{ and } j \text{ are directly connected,} \\ 0, & \text{otherwise.} \end{cases}$$

The powers of a graph's adjacency matrix, A^p , give the number of walks of length p between all pairs of nodes. For example, A^2 has entries a_{ij}^2 that give the number of walks of length 2 that join node v_i to node v_j and A^3 has entries that give the number of walks of length 3 and so on.



Graph G

$$A(G) = \begin{pmatrix} 0 & 1 & 0 & 0 & 1 \\ 0 & 0 & 0 & 1 & 0 \\ 0 & 1 & 0 & 0 & 0 \\ 1 & 0 & 1 & 0 & 1 \\ 0 & 0 & 0 & 1 & 0 \end{pmatrix}$$

Adjacency Matrix A (G)

Definition 9: Two nodes v_i and v_j are said to be reachable if there exists at least one path connecting them in the graph. Reachability can be expressed using a binary relation

$$R(v_i, v_j) = \begin{cases} 1, & \text{if a path exists between } v_i \text{ and } v_j, \\ 0, & \text{otherwise.} \end{cases}$$

The reachability matrix (R) is the sum of the adjacency matrix (A) and all matrices that itemize indirect paths between nodes of the graph.

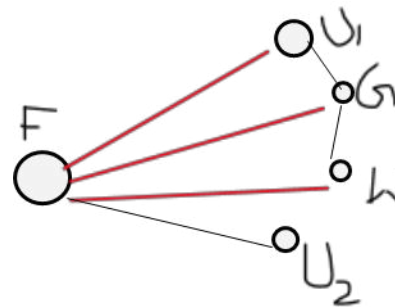
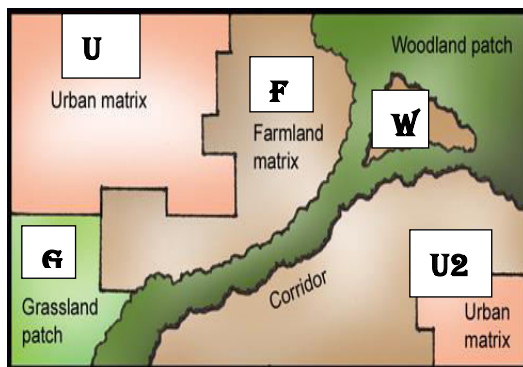
Hence, $R = A + A^2 + A^3 + \dots + A^n$, where 'n' is equal to the maximum number of steps to traverse the graph.

Landscape Mosaics and Graph Models

Landscape mosaics are composed of three fundamental components: patches, corridors, and the surrounding matrix. The most common landscape configurations include:

- i. A large matrix or patch surrounding or adjacent to multiple smaller patches,
- ii. A corridor bisecting the landscape, and
- iii. A network of interacting corridors forming an integrated unit [5].

In landscape ecology, patches are discrete spatial units embedded within the matrix and connected by corridors. Common examples of patches include fields, houses, and woods. Patches represent the basic units of the landscape that are dynamic and subject to change. Corridors are elongated patches that facilitate connections between patches, supporting movement and flow of organisms and materials. The matrix, as the background ecological system, exhibits a high degree of connectivity and constitutes the most extensive landscape element in which patches and corridors are embedded.



Landscape mosaics, composed of patches, corridors, and a matrix, can be effectively conceptualized using graph theory, which provides a formal framework for analysing spatial connectivity and ecological processes. In this representation, patches correspond to nodes (vertices) in a graph. Each node represents a discrete habitat unit, such as a forest fragment, wetland, or agricultural field, and may be attributed with properties including area, habitat quality, or species suitability.

Corridors are represented as edges (links) connecting nodes, symbolizing potential pathways for the movement of organisms, energy, and materials between patches. These edges may be directional or un-directional depending on movement assumptions, and they can be weighted to reflect factors such as corridor length, permeability, resistance, or movement cost. In landscapes lacking explicit physical corridors, edges may also represent functional connectivity derived from species dispersal capabilities or landscape resistance models.

The matrix provides the spatial context in which nodes and edges are embedded and plays a critical role in defining edge weights and connectivity patterns. Together, these components form a spatial graph, where network structure, connectivity metrics and path-based measures can be used to quantify landscape configuration and ecological function. This graph-based abstraction enables the integration of landscape ecology with network analysis, facilitating the assessment of habitat connectivity, fragmentation, and resilience under different land-use scenarios.

In landscape graph models, nodes represent landscape elements, while linkages (edges) indicate common boundaries between adjacent elements or points of contact between adjoining elements. Each landscape element, including the surrounding matrix, is represented by a node located at the element's centre, with node size corresponding to the element's area. Edges depict either shared boundaries or points where two ecosystems meet. Graphs can be constructed at any spatial scale. The number of neighbouring elements connected to a node defines its degree; elements with higher degrees are considered more critical, playing a larger role in maintaining overall landscape connectivity.

A landscape can be represented as a graph $G = (V, E)$, where V is the set of vertices (nodes) representing landscape elements such as patches, corridors, and matrix units, and $E \subseteq V \times V$ is the set of edges representing adjacency or points of contact between elements.

An edge $e = (v_i, v_j) \in E$ exists if the corresponding landscape elements share a common boundary or meet at a point. Landscape graphs can be constructed at different spatial scales depending on the resolution of the GIS data and the objectives of the analysis. The degree of a node $v \in V$, denoted by $\deg(v)$ represents the number of neighboring landscape elements adjacent to a given element. Nodes with higher degree values are considered structurally important, as they contribute more significantly to maintaining overall landscape connectivity. A path between two nodes v_i and v_j is defined as a sequence of distinct nodes $P = (v_i, v_{k_1}, v_{k_2}, \dots, v_j)$, such that consecutive nodes in the sequence are connected by edges in E . The length of a path, denoted by $|P|$, is the number of edges in the path. The shortest path length between nodes v_i and v_j , denoted by $d(v_i, v_j)$, is the minimum number of edges among all possible paths connecting v_i and v_j . This measure reflects the ease of movement or flow between landscape elements. At the landscape level, reachability indicates whether ecological processes such as species movement, dispersal, or material flow can occur between different spatial elements. Measures derived from reachability, such as the size of connected components and the graph diameter $D = \max_{v_i, v_j \in V} d(v_i, v_j)$, help identify isolated regions and assess the robustness of landscape connectivity.

Graph Patterns

Distinct spatial configurations of landscape elements can be identified through characteristic graph patterns, which reflect the organization of patches, corridors, and matrix elements within a landscape.

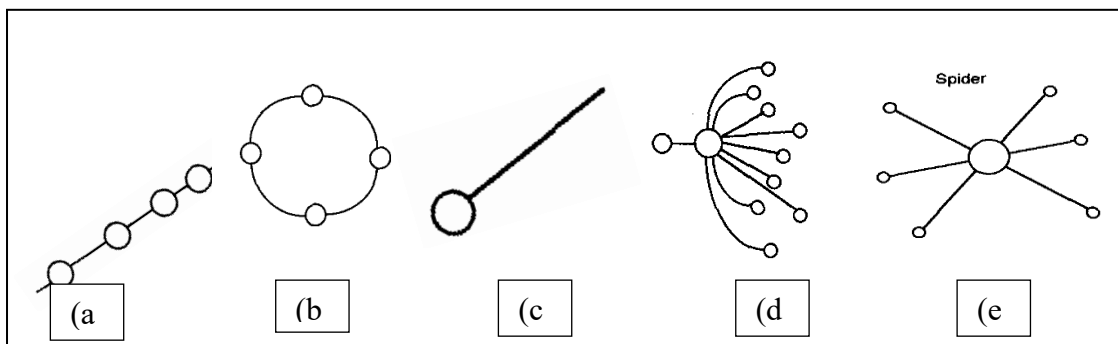


Fig. 1 Common graph patterns observed in landscape graphs: (a) Necklace pattern, representing linear arrangements of patches along corridors such as rivers or roads; (b) Graph cell pattern, indicating clustered landscape elements with strong internal connectivity; (c) Satellite pattern, showing isolated patches surrounding a dominant central element; (d)

Candelabra pattern, corresponding to branching corridor systems typical of drainage or transportation networks; and (e) Spider pattern, representing highly centralized landscapes where multiple corridors converge at a single core node. These patterns reflect distinct spatial configurations of patches, corridors, and matrix elements derived from GIS-based landscape representations.

(a) Necklace pattern: The necklace pattern is typical of linear landscape elements such as roads, rivers, and power lines. It consists of nodes representing the same type of landscape element linked sequentially, forming a structure resembling separated beads on a necklace. Crossing linkages are common in necklace patterns where corridors intersect, for example at road junctions or river confluences.

(b) Graph cell pattern: The interconnected sections formed by a network of necklace structures are referred to as graph cells. These patterns represent spatial compartments created by intersecting linear elements and often correspond to distinct land-use or land-cover units.

(c) Satellite pattern: The satellite pattern represents an isolated patch surrounded by a single dominant landscape element type. The satellite node typically corresponds to a small patch embedded within a large patch or an extensive matrix, such as a small wetland or settlement within a forested or agricultural landscape.

(d) Candelabra pattern: The candelabra pattern represents a landscape element that separates a single element on one side from multiple elements on the other. This pattern is characteristic of branching or hierarchical spatial structures and often indicates transitional or boundary elements within the landscape.

(e) Spider pattern: A graph structure with a central node connected to more than four surrounding nodes is termed a spider pattern. The central or spider node frequently represents the matrix, serving as a dominant and highly connected landscape element. Common examples of spider nodes include forests, grasslands, and agricultural fields that provide connectivity to multiple surrounding patches.

Ecological and Management Implications

Landscape graph patterns provide valuable insights into ecological processes and management priorities. The necklace pattern typically exhibits higher landscape resistance and a reduced barrier effect for animal movement, as linear elements such as roads, rivers, or hedgerows facilitate directional connectivity. A reduction in graph cell size is often correlated with increasing human population pressure and land-use fragmentation. Owing to their bounded structure and accessibility from multiple sides, graph cells are particularly suitable units for localized landscape management and planning.

The spider pattern represents a dominant matrix element surrounding and connecting multiple patches. Common examples include forest, grassland, or agricultural matrices. Such configurations indicate strong interactions across patch–matrix boundaries but often correspond to areas of low habitat diversity. These landscapes are ecologically sensitive and therefore require careful monitoring and adaptive management to prevent degradation of the matrix or loss of connectivity. The satellite pattern is characterized by an isolated patch embedded within a dominant surrounding element. Examples include an oasis within a desert or a small woodlot within an extensive forest or agricultural matrix. These patterns signal ecological vulnerability, as the isolated patch or the species it supports may be at risk of disappearance. Consequently, continuous monitoring and active conservation measures are essential in such landscapes. The candelabra pattern consists of a landscape element connected to many elements on one side and to a single element on the other. Plains

adjoining multiple land uses or roads branching from a single corridor often exhibit this configuration. These patterns highlight critical transition zones that play an important role in movement, access, and landscape functionality.

Connectivity of Landscape Elements

Connectivity is a fundamental measure of how well corridors, networks, or matrix elements are linked within a landscape. From an ecological perspective, high connectivity facilitates the movement of animals and plants, as well as the flow of energy, water, and materials among landscape elements. In graph-theoretic terms, connectivity or adjacency is quantified using the degree of a node, defined as the number of linkages incident to that node.

In landscape graphs, where nodes represent landscape elements and edges represent neighbouring relationships, the degree of a node serves as an indicator of the accessibility and functional importance of that element. Elements with higher degree values play a greater role in maintaining overall landscape connectivity. An adjacency (connectivity) matrix A can be constructed from the landscape graph to systematically analyze accessibility among elements [7, 9]. Powers of the adjacency matrix provide information on indirect connections: A^2 identifies two-step paths between elements, A^3 identifies three-step paths, and so on. This matrix-based approach allows the formal assessment of reachability and interaction potential among landscape components.

Methodology

Using this graph construction method, in this paper, landscape graphs are drawn and analysed from aerial photographs and land-use data representing different land-cover types, such as forests, wetlands, agricultural fields, and residential clearings, in selected regions of Coastal Maharashtra (Ratnagiri) and Gadchiroli. This approach enables comparative analysis of spatial patterns and connectivity across contrasting landscape settings.

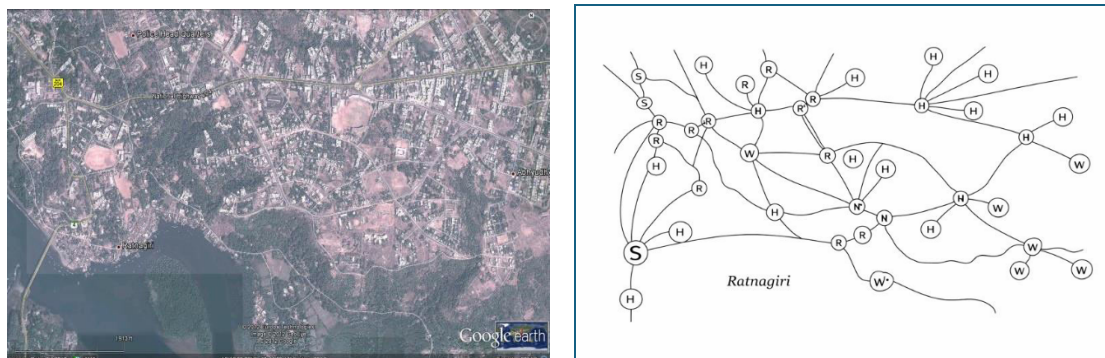


Fig: 2 Ratnagiri, Coastal Maharashtra represented by Landscape graph model. Landscape elements observed are: W-Woods, R- Roads, S-Sea and H- House clearings. Graph patterns observed are: Necklace and Graph cells by Roads and power lines, Candelabra by Sea and Spider by Woods.

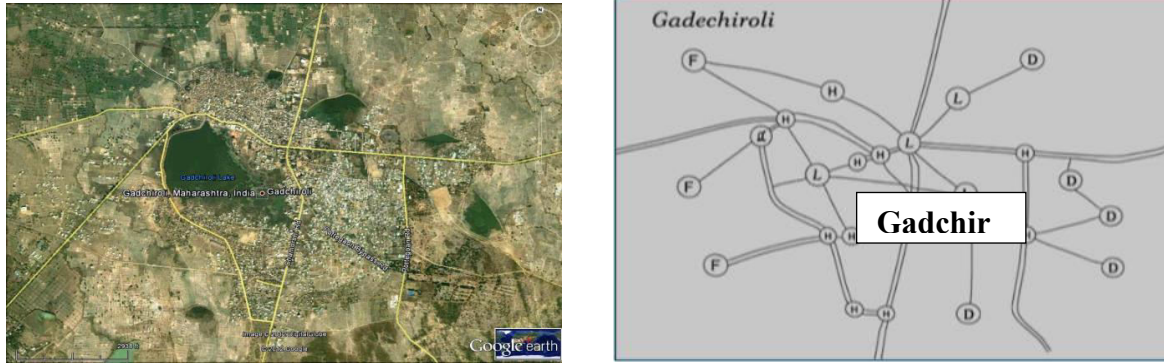


Fig 3 Gadchiroli, Maharashtra represented by Landscape graph model. Landscape elements observed are: F-Field, L- Lake, R- Roads, and H- House clearings, D – Dry lands. Graph patterns observed are: Necklace and Graph cells by Roads and power lines, Satellites by isolated dry lands and Spider by Lake.

In the landscape graphs illustrated above, house clearings, woods and fields are the most connected landscape element types. Each graph has a specific pattern of nodes and linkages. Necklace and spider patterns are found in these graphs. The patterns identified here represent the configurations of the landscapes and hence useful for landscape planning and management and for better ecological understanding [10]. The graphs depict highly connected linear elements which divide the landscape into component cells.

Interpretation:

- The circle (node) represents a small, isolated landscape patch.
- The single line (edge) indicates a single connection to a larger or dominant landscape element (often part of the matrix or a major patch).
- The low degree of the node ($\deg(v) = 1$) reflects limited connectivity, which is characteristic of satellite patterns.

Landscape-ecological meaning:

Such a structure typically represents a small patch embedded within or adjacent to a dominant matrix, for example:

- a small wetland within agricultural land,
- an isolated settlement near a large forest,
- a small forest fragment connected by a single corridor.

Graph-theoretic view:

- Node degree: $\deg(v) = 1$
- Connectivity contribution: minimal
- Vulnerability: high, since removal of the single edge disconnects the patch completely.

For the landscape graph of Gadchiroli, the method of analysing interactions among selected landscape elements, namely forests (F), lakes (L), roads (R), houses (H), and degraded or dryland (D), is illustrated as shown in Fig 4.

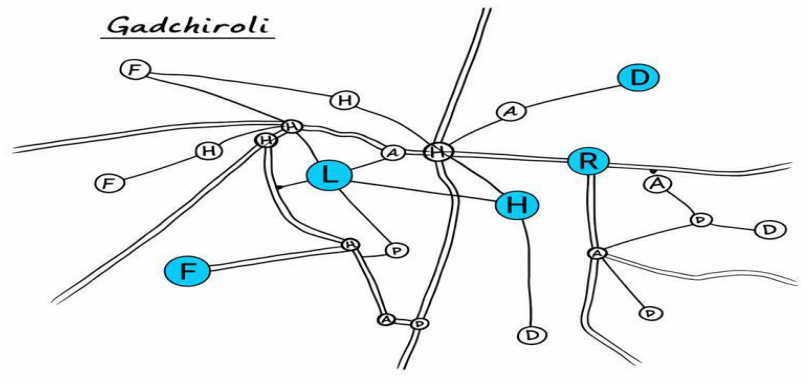


Fig. 4 Landscape graph model of Gadchiroli

The adjacency matrix for this graph is given as follows:

$$A = \begin{matrix} & \begin{matrix} F & L & R & H & D \end{matrix} \\ \begin{matrix} F \\ L \\ R \\ H \\ D \end{matrix} & \begin{bmatrix} 0 & 1 & 0 & 1 & 0 \\ 1 & 0 & 1 & 1 & 0 \\ 0 & 1 & 0 & 1 & 0 \\ 1 & 1 & 1 & 0 & 0 \\ 0 & 0 & 0 & 0 & 0 \end{bmatrix} \end{matrix}$$

Reachability matrix for this graph

$$R = \begin{bmatrix} 1 & 1 & 1 & 1 & 0 \\ 1 & 1 & 1 & 1 & 0 \\ 1 & 1 & 1 & 1 & 0 \\ 1 & 1 & 1 & 1 & 0 \\ 0 & 0 & 0 & 0 & 0 \end{bmatrix}$$

which indicates that the Dry land D is not reachable from other elements and vice versa.

Thus, the adjacency or connectivity matrix A of the landscape graph model of Gadchiroli shows the low degree of interactions between non-adjacent elements, for example, animals moving between two woods separated by a field. Similarly, the reachability matrix R indicates a high landscape resistance level and understanding of the dispersion of animals and plants across the landscape.

Conclusion

The patterns found in these landscape graphs highlight the structural configuration of the landscapes, which directly influences the movement of organisms, energy, and materials. Highly connected linear elements create distinct landscape cells, shaping ecological flows and interactions. By capturing these spatial relationships, these graph models offer a robust framework for understanding landscape dynamics and provide strong insights for sustainable land management and planning.

References

1. R. Haines Young, D. R. Green, S. H. Cousins et al., Landscape ecology and Geographic Information Systems, Taylor & Francis, London, (1993).
2. N. Deo, Graph theory with Applications to Engineering and Computer Science, Prentice Hall, New Delhi, (1999).
3. Harary F., Topics in Graph Theory, New York Academy of Sciences, New York, (1979).
4. R. J. Wilson, Introduction to Graph Theory, 4th edition, Prentice Hall, (1996).
5. D. Urban and T. Keitt, Landscape Connectivity: A Graph-Theoretic Perspective Ecology, Ecology: Ecological Society of America, 82, pp 1205-1218, (2001).

6. F. Jordan, Quantifying landscape connectivity: key patches and key corridors, Ecosystems and Sustainable Development IV, WIT Press, Southampton, (2003).
7. E. J. Taaffe, H. L. Gauthier, Geography of Transportation. Englewood Cliffs, Prentice-Hall, New Jersey, (1973).
8. H. S. Sharma, D. R. Ram, R. Prasad, P. R. Binda, Mathematical Modelling in Geographical Information System, Global Positioning System and Digital Cartography, Concept Publishing Company, New Delhi, (2006).
9. M. G. Turner and R. H. Gardner, Quantitative Methods for Analysing Landscape Pattern, Springer-Verlag, (1991).
10. M. D. Cantwell, R. T. T. Forman, Landscape Graphs: Ecological Modelling with Graph Theory to Detect Configurations Common to Diverse Landscapes, *Landscape Ecology*, 8, pp. 239-255, (1993).

42. A Critical Review of Extended X-Ray Absorption Fine Structure (EXAFS): A Tool for Understanding Local Structure of Nanomaterials

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Abstract: Nanoparticles exhibit distinct properties from their bulk counterparts due to quantum effects and high surface area. It is essential to comprehend the local atomic surroundings of nanomaterials in order to explain their distinct properties like mechanical and magnetic traits, electrical conductivity, catalytic activity and optical properties. Yet, peak broadening, structural diversity, and the presence of amorphous or substantially crystalline phase at the nanoscale frequently hinder the conventional methods from resolving short range order. This review examines Extended X-Ray Absorption Fine Structure (EXAFS) as a very efficient element specific technique for sub-angstrom local structure probing, with successful applications to metal nanoparticles (Au, Pt, Pd), metal oxide nanomaterials (TiO₂, Fe₂O₃, ZnO), and supported nanocatalysts [1-3]. Despite significant advancements, many existing studies rely on bulk like assumptions or over simplified structural models that are unable to adequately account for dynamic structural changes in nanomaterials [4-6]. The objective of this review is to assess the use of EXAFS to nanomaterials and emphasise methodological enhancements required for more precise structural interpretation. Multiple-scattering fitting, Fourier-transform analysis, ab initio and DFT based EXAFS modelling are among the techniques discussed [2,7,8]. According to published findings, EXAFS reliably identifies ligand-nanoparticle interactions, bond length contractions and changes in coordination numbers [9-10]. Furthermore, latest operando studies employ quick-scanning and flow cells for time resolved local structure analysis of active sites [12,14]. For conclusion, EXAFS is a potent tool for studying the structure of nanomaterials, providing information that diffraction cannot [1,3,5]. To completely understand the structural complexity of contemporary nanoscale systems, however, more advancements in machine learning assisted interpretation and multiple-scattering analysis are needed [8,15].

Introduction

The study of Nanomaterials has drawn huge scientific interest because of its exclusive properties in various fields which are totally distinct from its bulk form. A significant number of atoms live at or very close to the surface as the particle size becomes closer to the nanoscale. Because of this high surface to volume ratio, the bonding environments, coordination number, electrical structure and structural order of materials gets modified from its actual form. As a result of this nanoscale approach, the mechanical, electrical and optical characteristics as well as the catalytic behaviour of materials gets affected.

Ordinary Diffraction methods often face significant limitations at the nanoscale due to reduced crystallinity, peak broadening and amorphous phases, as they are only effective for long range ordered crystalline materials. For the consideration of nanoscale properties, it is crucial to use a method which remains accessible for short-range structural information such as local coordination environment and bond distortions.

To probe the local environment of a specific element with sub-angstrom precision, Extended X-ray Absorption Spectroscopy has emerged as a dominant technique as it provides element specific local structure information and is effective even in the absence of long-range crystallinity. Unlike standard X-ray diffraction, EXAFS can be used to analyse amorphous materials, highly disordered systems and liquids as well. This is the prime advantage characterizing many nanomaterials which are often poorly crystalline or non-crystalline. This is especially useful for complex or multi-component nanomaterials (e.g., core shell structures, doped materials, catalysts etc.) where a minority element's structural role is crucial but might be difficult to isolate with diffraction. This review thoroughly evaluates the significance of EXAFS in nanomaterial studies, emphasizes its key applications, addresses methodological challenges, and summarizes recent progress focused on enhancing structural analysis.

Fundamental and Theoretical background of EXAFS

EXAFS (Extended X-ray Absorption Fine Structure) spectroscopy examines a material's local atomic structure by analysing oscillations in X-ray absorption above an element's absorption edge, stemming from the interference between the outgoing photoelectron wave emitted from the absorbing atom and the waves backscattered by neighbouring atoms, revealing the distance, number, and type of atoms in the vicinity along with structural disorder. This theoretical foundation of EXAFS was established through pioneering work done by Lee et al. and Rehr and Albers [1,3]. Teo [2] and Bunker [7] further developed practical methodologies for EXAFS data acquisition, processing, and fitting, enabling routine structural analysis of complex materials. The development of multiple-scattering theory and real-space calculations, particularly through FEFF codes, significantly enhanced the reliability of EXAFS modelling [5,6].

Although the EXAFS spectroscopy technique has been known for a long time (Kronig, 1931, 1932), its structural content was not fully recognized until the work of Stern, Lytle, and Sayers (1974, 1975). In addition, the recent availability of synchrotron radiation has resulted in establishment of EXAFS as a practical structural tool particularly through the work of Eisenberger (1975) and Kincaid (1975) [17].

Table 1: Key Structural Parameters Extracted from EXAFS Analysis

EXAFS Parameter	Structural Meaning	Importance for Nanomaterials	References
Coordination number (N)	Number of nearest neighbours	Indicates surface atom fraction and size effects	[1–3,10]
Bond length (R)	Interatomic distance	Reveals bond contraction/expansion at nanoscale	[2,10]
Debye–Waller factor (σ^2)	Static and thermal disorder	Captures structural heterogeneity	[1,7]
Scattering amplitude & phase	Element-specific interactions	Enables selective probing of multi-component systems	[3,6]

Limitations of Conventional Structural Techniques for Nanomaterials

For conventional diffraction techniques, the structural characterisation of nanomaterials poses a number of difficulties. Because Nanoparticles commonly exhibit Broad diffraction peaks due to finite size effects, overlapping reflections from multiple phases, surface relaxation and lattice strain and amorphous or partially crystalline domains but XRD depends on periodic atomic planes' constructive interference, it is less useful at very small crystallite sizes.

Similarly, transmission electron microscopy (TEM) offers local structural information but is limited by sampling statistics, beam damage, and challenges in quantitative structural averaging. Raman and infrared spectroscopy also offer indirect structural insights but lack atomic-scale resolution.

In contrast, EXAFS directly probes short-range order without requiring crystallinity, making it particularly valuable for nanomaterials where disorder and surface effects dominate. However, accurate interpretation of EXAFS data requires careful modelling, especially when applied to nanoscale systems with complex structural heterogeneity.

Table 2: Limitations of Conventional Structural Techniques for Nanomaterials

Technique	Structural Information Provided	Limitations at Nanoscale	Supporting References
XRD	Long-range crystalline order	Peak broadening, loss of short-range order, amorphous phases not detected	[1,3,4]
TEM	Local atomic images	Limited statistics, beam damage, non-representative sampling	[4,7]
Raman/IR	Bond vibrations	Indirect structural information, poor atomic resolution	[4]
EXAFS	Local atomic structure (short-range)	Requires modelling, limited long-range sensitivity	[1–3,7]

Applications of EXAFS to Nanomaterials

1. Metal Nanoparticles

Extended X-ray absorption Fine Structure (EXAFS) spectroscopy has been used to study short range order in heterometallic alloys for almost four decades. EXAFS has been widely used in study of noble metal nanoparticles such as Au, Pt, and Pd, which are significant in catalysis and electronic applications. Several studies report a reduction in metal–metal coordination numbers, and contraction of bond lengths relative to bulk materials. These effects arise from high surface to volume ratio and finite-size strain.

For example, EXAFS research of Pt and Pd nanoparticles shows that nearest-neighbour coordination systematically decreases as particle size decreases, which is directly correlated with increased catalytic activity. Modified electronic structures and changed adsorption energies, which are essential for catalytic activity, have been connected to the bond length contraction seen in metal nanoparticles [9,10].

Zhang et al. demonstrated that EXAFS can accurately quantify bond contraction and coordination changes in nanoparticle catalysts, directly correlating structural modifications with enhanced catalytic performance [10]. Such studies highlight the ability of EXAFS to capture size-dependent structural features inaccessible to diffraction techniques.

2 Metal Oxide Nanomaterials

Extended X-ray Absorption Fine structure (EXAFS) spectroscopy is a significant tool for characterizing metal oxide nanoparticles because it postulates element-specific, local structural information (bond lengths, coordination numbers, disorders) without requiring long-range crystallinity. Metal oxide nanomaterials like MgO, TiO₂, Fe₂O₃, and ZnO play important roles in photocatalysis, sensing, and energy applications. EXAFS studies of these systems provide insights into local coordination geometry, oxygen vacancies, and lattice distortions that are not evident from diffraction techniques.

For example, EXAFS studies reveal deviations from ideal octahedral coordination in TiO₂ nanoparticles and changes in Fe–O bond distances in iron oxide nanostructures. In TiO₂ nanoparticles, EXAFS has been used to distinguish between anatase and rutile local environments and to quantify distortions in Ti–O and Ti–Ti bonds. Similarly, Fe K-edge EXAFS studies of iron oxide nanoparticles reveal changes in Fe coordination and oxidation state associated with particle size and synthesis method. These findings are crucial for understanding size-dependent magnetic and catalytic properties. These local structural variations crucially impact optical absorption, magnetic behaviour, and catalytic activity.

3 Operando EXAFS and Supported Nanocatalysts

As EXAFS spectroscopy is a premier, element specific technique, it determines the local atomic structure of supported nanocatalysts, especially under real-time (Operando) reaction conditions. In bimetallic or multimetallic nanocatalysts, EXAFS can determine the distribution of different elements which helps in identifying core-shell architectures and segregation tendencies. Supported nanocatalysts represent a particularly complex class of materials due to interactions between metal nanoparticles and supports. EXAFS is uniquely suited to examine metal–support interactions, ligand effects, and structural evolution under reaction conditions (Operando).

The study of EXAFS can successfully explain metal–oxygen and metal–support coordination shells, providing evidence for strong metal–support interactions (SMSI). Changes in coordination number and bond lengths under working conditions have been correlated with catalyst activation, deactivation, and sintering processes [11,12].

As reviewed by Bare and Ressler [11], Operando EXAFS studies have provided real-time insights into catalyst activation, restructuring, and deactivation under realistic reaction conditions. Time-resolved EXAFS measurements further reveal transient coordination changes during catalytic reactions [12,13].

4 Limitations of Bulk-Like Structural Models

Despite considerable advancements, numerous EXAFS studies continue to depend on bulk-like or excessively simplified structural models. These assumptions frequently overlook dynamic disorder, surface relaxation, and the structural heterogeneity that are inherent in

nanomaterials. Koningsberger and Prins [4] highlighted the necessity for more authentic modelling methods that integrate nanoscale influences and non-periodic architectures.

Advanced EXAFS Data Analysis and Modelling

1 Data Acquisition and Processing

Because EXAFS measurements require high photon flux and customisable X-ray energy, they are usually carried out at synchrotron radiation facilities. The EXAFS signal is extracted and converted into photoelectron wave-vector space for analysis following background subtraction and normalisation.

2 Fourier Transform and Multiple-Scattering Analysis

Coordination shells can be seen by the pseudo-radial distribution function, that Fourier-transform (FT) EXAFS offers. However, phase changes and disorder affect FT peaks, thus careful fitting is required. In nanomaterials with short bond lengths or non-linear geometries, multiple scattering effect becomes more significant.

Structural models are more accurate when multiple-scattering routes are included, particularly for oxide frameworks and metal clusters. The reliability of EXAFS simulations has greatly improved because to ab initio codes like FEFF [5,6].

3 Ab Initio and DFT-Based Modelling

In order to generate accurate atomic models of nanomaterials, Density Functional Theory (DFT) is becoming a crucial tool. Theory and experiment can be directly compared by using DFT-optimized structures as input for EXAFS simulations.

Surface relaxation, bond distortion, and size-dependent phenomena that are overlooked in bulk-like models are captured by this combined method that are often neglected in bulk-like models. Such methods are particularly valuable for small clusters and highly distorted nanostructures.

4 Machine Learning-Assisted EXAFS Interpretation

Data-driven approaches to EXAFS analysis have been developed by recent advancements in machine learning. Machine learning methods can extract coordination environment distributions, lessen parameter correlation, and recognise complex structural patterns. Coordination distributions and structural heterogeneity beyond average structural parameters can be extracted from disordered nanomaterials by the successful application of machine learning-enhanced EXAFS analysis [9]. These methods are a big step towards interpreting nanoscale structures with greater objectivity and accuracy.

The characterization of disorder in nanomaterials is much improved by machine learning-enhanced EXAFS analysis, as Timoshenko and Frenkel have shown [9]. These methods offer an exciting potential for further EXAFS research.

5 Operando and Time-Resolved Techniques

Operando EXAFS's ability to monitor structural changes in real time under practical operating situations has revolutionised the understanding of active nanomaterials. Studying

dynamic processes including catalyst activation, phase transition, and sintering is now feasible because of the development of quick-scanning EXAFS, flow cells, and sophisticated synchrotron sources.

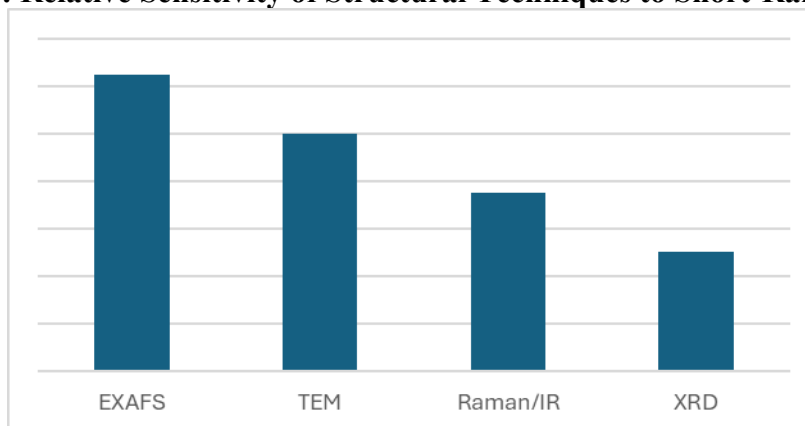
Transient coordination changes and bond length fluctuations during catalytic reactions have been identified by time-resolved EXAFS experiments, offering direct insight into active sites and reaction mechanisms [12–14]. These results emphasise the value of investigating nanomaterials in operational settings as opposed to depending exclusively on ex situ data.

Limitations and Challenges of EXAFS in Nanomaterial Studies

The shift from periodic bulk lattices to finite, frequently disordered, nanoscale systems create complex structural and mathematical challenges for nanomaterial analysis using EXAFS. The main drawback at the doctoral level is the bulk-averaging nature of the method; since EXAFS probes all absorbing atoms at once, the resulting signal is a population-weighted average that can obscure the existence of various local environments, such as the coexistence of different particle sizes or the differentiation between surface and core atoms.

In particles smaller than 2 nm, where surface atoms make up about half of the total, surface-induced bond contractions and local relaxations contribute significant static disorder that standard harmonic models are unable to account for. Because standard fitting procedures are not able to differentiate between dampening effects caused by large Debye-Waller factors (σ^2) and actual coordination deficits, this structural heterogeneity often results in the systematic underestimation of coordination numbers (N) and bond lengths (R).

Figure 1: Relative Sensitivity of Structural Techniques to Short-Range Order



Additionally, the Nyquist criterion, which confines the amount of independent data points (ν) that may be extracted from a given k and R range, limits the mathematical integrity of EXAFS. The number of necessary structural parameters for complex nanomaterials, such as single-atom catalysts on amorphous supports or multi-component nanoalloys, frequently surpasses this informational limit, resulting in high parameter correlation and possibly degenerate solutions where several physically different models fit the data with equal statistical validity. In order to address these problems, advanced research 2025 emphasises extending beyond the "shell-by-shell" method and using DFT-guided modelling or MD-EXAFS (combining molecular dynamics snapshots with configuration-averaged spectra) to offer a more realistic, non-Gaussian treatment of the radial distribution function. Higher-order coordination shells can be resolved using these combined computational-experimental

frameworks, which also offer a more realistic depiction of the asymmetric distributions inherent to nanostructured materials.

It can be concluded that many existing studies depends on simplified or bulk-like structural models that fail to capture the full complexity of nanoscale systems. Addressing these limitations requires improved modelling strategies, integration with complementary techniques, and advanced data analysis methods [18-23].

Future Perspectives

Methodological developments and interdisciplinary integration are key to the future of EXAFS in nanomaterials research. Key directions consist of; Increased use of analysis based on machine learning, better dynamic disorder and multiple-scattering modelling, combining spectroscopy and in situ microscopy, and Improved temporal and spatial resolution. These advancements will reinforce EXAFS's position as a key method in nanoscience and allow for a more thorough knowledge of complex nanoscale systems.

Conclusion

Extended X-Ray Absorption Fine Structure spectroscopy is an effective and essential method for examining the local atomic structure of nanomaterials. Because of its element-specificity and sensitivity to short-range order, it is possible to characterise systems in detail where conventional diffraction methods fall short.

In metal nanoparticles, metal oxide nanomaterials, and supported nanocatalysts, EXAFS has effectively clarified coordination shifts, bond length contractions, ligand interactions, and dynamic structural evolution. However, the true potential of EXAFS in nanoscience is constrained by the ongoing dependence on simplified structural models.

To fully capture the structural complexity of nanoscale systems, future developments in multiple-scattering analysis, DFT-based modelling, machine learning-assisted interpretation, and operando techniques are crucial. With these advancements, EXAFS will continue to be a fundamental method for comprehending and creating cutting-edge nanomaterials.

References

1. Lee, P. A.; Citrin, P. H.; Eisenberger, P.; Kincaid, B. M. Extended X-ray Absorption Fine Structure—Its Strengths and Limitations. *Rev. Mod. Phys.* 1981, 53, 769–806.
2. Teo, B. K. *EXAFS: Basic Principles and Data Analysis*; Springer: Berlin, 1986.
3. Rehr, J. J.; Albers, R. C. Theoretical Approaches to X-ray Absorption Fine Structure. *Rev. Mod. Phys.* 2000, 72, 621–654.
4. Koningsberger, D. C.; Prins, R. (Eds.) *X-ray Absorption: Principles, Applications, Techniques of EXAFS, SEXAFS, and XANES*; Wiley: New York, 1988.
5. Ankudinov, A. L.; Ravel, B.; Rehr, J. J.; Conradson, S. D. Real-Space Multiple-Scattering Calculation of XANES. *Phys. Rev. B* 1998, 58, 7565–7576.
6. Zabinsky, S. I.; Rehr, J. J.; Ankudinov, A.; Albers, R. C.; Eller, M. J. FEFF: An Automated FEFF Path Generator. *Phys. Rev. B* 1995, 52, 2995–3009.
7. Bunker, G. *Introduction to XAFS: A Practical Guide to X-ray Absorption Fine Structure Spectroscopy*; Cambridge University Press: Cambridge, 2010.

8. Frenkel, A. I.; Wang, Q.; Marinkovic, N. Real-Time Monitoring of Catalysts Using Operando X-ray Absorption Spectroscopy. *J. Synchrotron Radiat.* 2012, 19, 722–728.
9. Timoshenko, J.; Frenkel, A. I. Probing Structural Disorder in Nanomaterials Using Machine Learning–Enhanced XAFS Analysis. *J. Phys. Chem. Lett.* 2019, 10, 5990–5996.
10. Zhang, X.; Banis, M. N.; Sun, Y.; Li, R.; Sham, T.-K. EXAFS Study of Nanoparticle Catalysts: Coordination Changes and Bond Contraction at the Nanoscale. *Chem. Mater.* 2014, 26, 596–603.
11. Bare, S. R.; Ressler, T. Operando X-ray Absorption Spectroscopy in Heterogeneous Catalysis. *Adv. Catal.* 2009, 52, 339–465.
12. Peterson, E. J.; et al. Operando X-ray Absorption Spectroscopy of Nanocatalysts Under Working Conditions. *Nat. Commun.* 2014, 5, 4885.
13. Liu, Y.; et al. Structural Evolution of Metal Nanoparticles During Catalysis Revealed by Time-Resolved EXAFS. *ACS Catal.* 2016, 6, 476–485.
14. Funke, H.; Scheinost, A. C.; Chukalina, M. Wavelet Analysis for EXAFS Data: A Powerful Tool for Detecting Multiple Coordination Environments. *Phys. Rev. B* 2005, 71, 094110.
15. Calvin, S. *XAFS for Everyone*; CRC Press: Boca Raton, 2013.
16. Christiansen, T. L.; Cooper, S. R.; Jensen, K. M. Ø. There’s no place like real space: elucidating size-dependent atomic structure of nanomaterials using pair distribution function analysis. *Nanoscale Adv.* 2020, 2, 2234–2254.
17. EXAFS: Basic Principles and Data Analysis by Boon K. Teo.
18. Stern, E. A. *Theory of EXAFS* Physical Review B, 1974, 10, 3027–3037. Foundational treatment of EXAFS averaging, coordination number extraction, and Debye–Waller effects.
19. Ravel, B.; Newville, M. Athena, Artemis, Hephaestus: Data Analysis for X-ray Absorption Spectroscopy. *Journal of Synchrotron Radiation*, 2005, 12, 537–541. Standard fitting workflows and their limitations for disordered systems.
20. Dalba, G.; Fornasini, P. EXAFS Debye–Waller Factor and Structural Disorder. *Journal of Synchrotron Radiation*, 1997, 4, 243–255. Key reference on static disorder and breakdown of harmonic approximations.
21. Fornasini, P.; Dalba, G.; Grisenti, R. Beyond the Gaussian Approximation in EXAFS Analysis. *Journal of Physics: Condensed Matter*, 2001, 13, 7859–7876. Non-Gaussian radial distribution functions and higher-order cumulants.
22. Flytzani-Stephanopoulos, M.; Gates, B. C. Atomically Dispersed Supported Metal Catalysts. *Annual Review of Chemical and Biomolecular Engineering*, 2012, 3, 545–574. EXAFS challenges in distinguishing isolated atoms from small clusters.
23. Liu, J. *Catalysis by Supported Single Metal Atoms*. *ACS Catalysis*, 2017, 7, 34–59. EXAFS parameter degeneracy and coordination ambiguity in single-atom systems.

43. Understanding the Low Temperature Regime of frustrated pyrochlore $\text{Dy}_2\text{Ti}_2\text{O}_7$: Spin Ice and Associated Novel Phases

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Abstract: Water Ice (H_2O) is the third state of matter (now fourth state as per new norms, viz Plasma, Gas, Liquid, Solid and BEC (Bose Einstein Condensate)), known to float in water rather than sinking. Is it a simple density concept. No. One of the scientific reasons is that there is a finite residual entropy of tetrahedral ice even at 0 deg K, as pointed out by Linus Pauling in 1956. After this, Anderson dwelled another concept of spinel structures, having anomalous properties like geometrical disorder and frustrated magnetism. Spin ices are magnetic systems with abundant geometric frustration. In 1933, ice rule (2in 2out) was given by Bernal and Fowler while they initiated molecular structure of water ice. $\text{A}_2\text{B}_2\text{O}_7$ is the generic pyrochlore oxide family that has lot of novel magnetic phases like spin ice or spin liquid. We present our understanding on low temperature properties on $\text{Dy}_2\text{Ti}_2\text{O}_7$ in the line of recently published work, includes Pauling Residual Entropy, Kagome Ice States in moderate magnetic fields and associated phase transitions.

Keywords: Ice rule, Frustration, Kagome ice, Spin ice

Introduction

Ice has most interesting but complex structure and has unique properties which made it interesting to research. Ice floats on water. It is an amazing one among its many features that it exhibits. It has specific boundaries and a lot of degeneracy in its ground states. The Structure of Spin Ice is same as normal water ice. In normal water ice we talk about protons while in Spin ice we talk about spins. In water it is proton disordered state. In spin ice systems are a type of frustrated spins in a magnetic material. Residual entropy and exotic excitations, such as emergent magnetic monopoles, are produced because of magnetic moments in these materials obey local "ice rules" found in water ice. Strong quantum fluctuations in related systems can completely suppress classical ice order, which can result in the formation of spin liquid states. Frustration and emergent phenomena has a connection between spin liquids, spin ice, Kagome ice, and normal water ice.

Majority of solids have a higher density than their liquid counterparts except water ice. At a temperature of around 4 degrees Celsius, liquid water reaches to its maximum density.

Below that temperature, it expands as it cools. During the freezing process, water transforms into a solid that has a decreased density, which enables ice to float. When hydrogen bonds are present in liquid water, they are always forming and breaking, which makes it possible to pack tightly. When water freezes, its molecules form an open tetrahedral network. When water freezes, it expands because of a conflict between hydrogen bonds that regulate energy and volumetric efficiency. Hexagonal ice, often known as Ice I_h , is the most prevalent type of ice that occurs under normal conditions. When oxygen atoms come together, they make a crystal structure that is like the structure of a diamond. There are hydrogen bonds between each oxygen atom and its four closest neighbours.

But the hydrogen atoms are still not in their proper place, even though the oxygen structure is perfectly ordered. Each oxygen is linked to two protons that are close to each other. There is exactly one proton present in every single hydrogen bond. There are about twenty different crystalline ice polymorphs that are known to exist in water, which results in a diverse phase diagram. Phases with a disordered proton order as Ice I_h and Ice I_c . Proton-ordered phases include Ice XI, which is a ferroelectric phase at low temperatures¹. Ice I_h has persistence of proton disorder down to very low temperatures which reflects kinetic restrictions that prevent the system from achieving a distinct ground state. The estimation of the configurational entropy of ice that results from proton disorder was made by Linus Pauling in the year 1935². The equation for S is equal to $R \ln \frac{3}{2}$. The experiments which are used to calculate residual entropy are calorimetric.

Materials used in spin ice are typically rare-earth pyrochlores, and their chemical formulas are as follows: magnetic rare-earth ions (R) are found to be occupying a lattice of corner-sharing tetrahedra in this structure. Strong effects of crystal fields set limits on the spins so they can only move in the local [111] directions. This effectively makes them Ising-like system. The tetrahedron structures with the least amount of energy follow a rule called "two-in, two-out." This rule is very similar to the Bernal–Fowler rules that guide water ice³.

Materials that are instances of canonical spin ice: The conventional dipolar spin ice known as $\text{Dy}_2\text{Ti}_2\text{O}_7$ possesses a residual entropy that is clearly defined. $\text{Ho}_2\text{Ti}_2\text{O}_7$ is a magnetically stable spin ice that exhibits hyperfine and nuclear effects⁴. Spin ice is also observed in $\text{Ho}_2\text{Sn}_2\text{O}_7$ ⁵. A deformed variation with increased frustration is seen in $\text{Dy}_2\text{Sn}_2\text{O}_7$. The spin ice behaviour is clearly visible in these systems. These signatures include entropy plateaus and diffuse neutron scattering with pinch-point characteristics. When a magnetic field is applied along the crystallographic direction of a spin ice crystal, a field-induced partially ordered state known as the Kagome ice state presents itself⁶. This state is characterized by a partial order. In this particular geometry, the pyrochlore lattice is broken down into Kagome planes and triangular planes that alternate with one another. Under the influence of the field, spins on triangular planes become completely polarized⁷. When it comes to Kagome planes, spins continue to be frustrated and adhere to modified ice restrictions. This results in a frustrated Kagome lattice that is two-dimensional, which is where the term "Kagome ice" comes from. Additionally, macroscopic degeneracy and residual entropy⁸ are also preserved in Kagome ice.

This material displays magnetization plateaus as well as signs of neutron scattering that are characteristic. When magnetic fields are used, it offers a frustration. An essential connection between three-dimensional spin ice and two-dimensional frustrated systems is represented by Kagome ice. This connection shows the analogy with water ice when it is subjected to external constraints. Strong quantum fluctuations are responsible for the absence of long-

range order in spin liquids, which are magnetic states. This is the case even when the temperature is zero. Unlike spin ice, which exhibits classical disorder, spin liquids are characterized by their underlying quantum mechanical properties. Spin liquids based on the Kagome and triangular lattice are seen in systems like: Herbertsmithite ($\text{ZnCu}_3(\text{OH})_6\text{Cl}_2$) – Kagome lattice quantum spin liquid. $\kappa\text{-(BEDT-TTF)}_2\text{Cu}_2(\text{CN})_3$ – organic triangular lattice, $\text{EtMe}_3\text{Sb}[\text{Pd}(\text{dmit})_2]_2$ – gapless spin liquid candidate⁹. Pyrochlore-based spin liquid candidates are as $\text{Tb}_2\text{Ti}_2\text{O}_7$ – cooperative paramagnet¹⁰, $\text{Pr}_2\text{Zr}_2\text{O}_7$ and $\text{Pr}_2\text{Sn}_2\text{O}_7$ – quantum spin ice candidates¹¹, $\text{Yb}_2\text{Ti}_2\text{O}_7$ – proximity quantum spin liquid behavior¹².

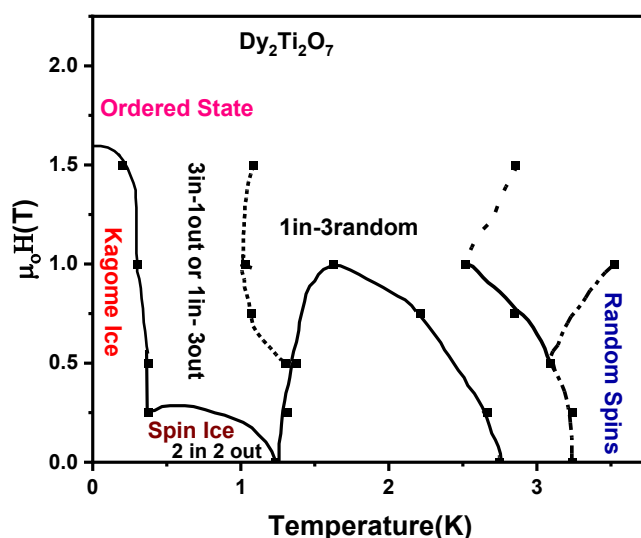


Figure 1: The proposed H-T phase for our sample $\text{Dy}_2\text{Ti}_2\text{O}_7$, the possible spin-ice character and other degenerate configurations are assumed and plotted. Taken from Shruti Vyas et al [13].

With the heat capacity measurement we can confirm the behaviour of these samples. Diffuse scattering and pinch spots are both examples of neutron scattering, μSR ¹³ and NMR for the study of low-temperature dynamics. Kagome ice and ordered phases can be formed by the varying of magnetic fields. The initial example of a frustrated system that is governed by local laws and macroscopic degeneracy is the water ice that is normally present in natural environments. These systems show how simple local restrictions can lead to a wide range of new behaviours. As a result, ice, which includes not just water but also magnetic and quantum ice, has become an essential to research in current condensed matter physics.

Recently we have studied one such compound $\text{Dy}_2\text{Ti}_2\text{O}_7$ that shows interesting phases of spin ice. The phase diagram of this compound is shown below in **Fig 1**, that is self-explanatory. The sample is also a well-known member of the pyrochlore family. It is found to have an exotic ground state like spin ice. This is because weakly interacting Ising spins of each tetrahedron may follow the two-in, two-out ice rule. A low-temperature study is carried out to understand this novel ground state. Heat capacity shows no noticeable magnetic ordering. Heat capacity showed a broad anomaly around 1 K, indicative of only short-range interactions. Residual entropy of the spin-ice state is calculated in zero magnetic field is $1.36 \text{ JK}^{-1}\text{mol}^{-1}$. This is close to the prediction of Pauling. Kagome-ice state could be found when a magnetic field is applied. In this case residual entropy is $0.56 \text{ JK}^{-1}\text{mol}^{-1}$, between 0.3 T and 0.6 T. A magnetic field–temperature (H–T) phase diagram has also been constructed and visualized for the spin-ice configuration as in Figure 1. Most of the features are in line with the literature. Here we have outlined only the essence of the work. One can refer the article by Shruti Vyas¹³ for further details.

Acknowledgments

The authors profusely thank MPCST for the FTYS support (MPCST Let No 884/CST/R&D/FTYS) to S.V., MPCST File no. A/RD/RP-2/321 for VG., CSR/2022-23/03/877 for V.G. Dr Manoj Rathore from MPCST is thanked for his guidance. UGC-DAE CSR Indore for advanced experimental support, Medicaps University, Indore for encouragement in the form of URF Fellowship (URF-MU-2025-26/01) to SV.

References

1. Earth, O. Phases of ice.
2. Pauling, L. *The Structure and Entropy of Ice and of Other Crystals with Some Randomness of Atomic Arrangement*.
3. Bernal, J. D. & Fowler, R. H. A theory of water and ionic solution, with particular reference to hydrogen and hydroxyl ions. *J. Chem. Phys.* **1**, 515–548 (1933).
4. Harris, M. J., Bramwell, S. T., McMorro, D. F., Zeiske, T. & Godfrey, K. W. Geometrical frustration in the ferromagnetic pyrochlore $\text{Ho}_2\text{Ti}_2\text{O}_7$. *Phys. Rev. Lett.* **79**, 2554–2557 (1997).
5. Ehlers, G. *et al.* Low energy spin dynamics in the spin ice $\text{Ho}_2\text{Sn}_2\text{O}_7$. *J. Phys. Condens. Matter* **24**, (2012).
6. Hiroi, Z., Matsuhira, K., Takagi, S., Tayama, T. & Sakakibara, T. *Kagomé Ice in Dy Specific Heat of Kagomé Ice in the Pyrochlore Oxide Dy₂Ti₂O₇*.
7. Higashinaka, R., Fukazawa, H., Deguchi, K. & Maeno, Y. Low temperature specific heat of $\text{Dy}_2\text{Ti}_2\text{O}_7$ in the Kagome ice state. *J. Phys. Soc. Japan* **73**, 2845–2850 (2004).
8. Ramirez, A. P., Hayashi, A., Cava, R. J., Siddharthan, R. & Shastry, B. S. Zero-point entropy in ‘spin ice’. *Nature* **399**, 333–335 (1999).
9. Knolle, J. & Moessner, R. A Field Guide to Spin Liquids. 451–472 (2019).
10. Gardner, J. S., Gingras, M. J. P. & Greedan, J. E. Magnetic pyrochlore oxides. *Rev. Mod. Phys.* **82**, 53–107 (2010).
11. Kimura, K. *et al.* Quantum fluctuations in spin-ice-like $\text{Pr}_2\text{Zr}_2\text{O}_7$. *Nat. Commun.* 4–9 (2013) doi:10.1038/ncomms2914.
12. Scheie, A. *et al.* Multiphase magnetism in $\text{Yb}_2\text{Ti}_2\text{O}_7$. **117**, 27245–27254 (2020).
13. Lin, L. *et al.* Experimental observation of magnetoelectricity in spin ice $\text{Dy}_2\text{Ti}_2\text{O}_7$. *New J. Phys.* **17**, 123018 (2015).

44. Enhanced Sampling of Protein Energy Landscapes Using Parallel Tempering Monte Carlo simulation for Met-enkephalin and WWP domain proteins

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Abstract: We investigate the efficiency of Parallel Tempering Monte Carlo (PTMC) in sampling complex protein energy landscapes using Met-enkephalin and the WW-Domain as representative systems. For Met-enkephalin, PTMC provides significantly enhanced sampling over the entire temperature range, particularly in the low-energy region, enabling reliable identification of the global minimum. For the WW-Domain, energy versus number of Monte Carlo steps has also demonstrated with stable convergences and histograms exhibit good overlap between adjacent temperatures, confirming thermodynamic consistency of the simulations. However, the native minimum structure of the WW domain is not fully recovered, indicating that increased system size requires further optimization of PTMC parameters. These results highlight the strength of PTMC for small peptides and underscore the need for improved temperature schemes, replica numbers, and sampling strategies to accurately characterize larger protein domains.

Keywords: Protein-Folding, Monte-Carlo Simulation (MCS), Parallel-Tempering (PT), Energy landscape.

Introduction

Protein folding is governed by the underlying energy landscape [1, 2, 3, 4], which maps the potential energy of all possible conformations of a polypeptide chain. Each point on this landscape corresponds to a specific structure, and its energy reflects intramolecular interactions such as hydrogen bonding, van der Waals forces, electrostatics, hydrophobic effects, and dihedral contributions. The native structure usually lies near the global minimum of this landscape, consistent with the thermodynamic hypothesis that proteins adopt the lowest [5].

For real proteins, the energy landscape is highly rugged, containing many local minima separated by energy barriers [6]. These correspond to metastable, partially folded, or misfolded states. Thus, folding proceeds through complex pathways governed by both thermodynamics and kinetics rather than by simple downhill motion. At low temperatures, systems can become trapped in local minima, limiting conformational sampling [7].

Energy landscape theory describes folding as motion down a funnel-shaped surface toward the native state, while still allowing frustration and competing basins that slow folding dynamics. Larger proteins exhibit greater roughness and many more conformational states than small peptides such as Met-enkephalin, making sampling and identification of native-like structures more difficult[8].

Conventional Molecular Dynamics and Monte Carlo simulations often explore only a limited region of this landscape and may become trapped in local minima. Enhanced sampling methods—such as replica exchange, multicanonical Monte Carlo, Wang–Landau sampling, umbrella sampling, and parallel tempering—improve exploration by facilitating barrier crossing and rare-event sampling [9, 10, 11, 12, 13, 14, 17]. In this work, Parallel Tempering Monte Carlo simulations are employed for peptide Met-enkephalin and the WW-Domain protein to analyze the efficiency and limitations of PTMC simulation as we increase the system size of protein molecules [19].

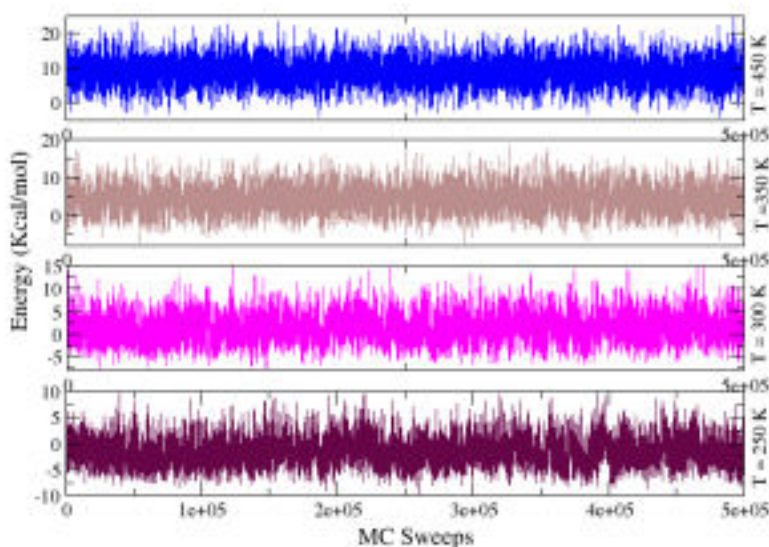


Figure 1: Energy verses Monte Carlo Steps for Met-Enkephalin protein using PTMC simulation.

Models and Methods

Parallel Tempering Monte Carlo (PTMC) Simulation Method

Parallel Tempering Monte Carlo (PTMC) was used to study the conformational dynamics of the Met enkephalin peptide. In PTMC, several Monte Carlo simulations are run in parallel at different temperatures. By periodically exchanging configurations between neighboring temperatures, low-temperature replicas can escape local minimum with the help of high-temperature ones, leading to improved sampling and enhanced ergodicity [15].

We performed simulations using N_{replicas} replicas, each fixed at a temperature T_i ($i = 1, 2, N$). The temperature range was selected to cover both ordered, folded states at low temperatures and highly fluctuating unfolded states at high temperatures. Temperatures were arranged to ensure sufficient overlap between adjacent energy distributions so that exchange moves would be efficient [3]. Within each replica, sampling was carried out using standard Monte Carlo moves. At every Monte Carlo step, one dihedral angle was chosen at random and reassigned a value between -180° and $+180^\circ$ [20]. The move was accepted according to the Metropolis rule [21],

where ΔE is the energy change upon the trial move, k_B is Boltzmann's constant, and T_i is the temperature of the replica.

Replica exchanges between neighboring temperatures T_i and T_{i+1} were attempted at fixed intervals. The acceptance probability for swapping configurations was

$$P_{\text{swap}} = \min [1, \exp ((\beta_i - \beta_j)(E_i - E_j))] , \quad (2)$$

with $\beta_i = 1/(k_B T_i)$. This procedure allows replicas to migrate along the temperature ladder and cross high free-energy barriers, thereby improving configurational diffusion.

An initial equilibration stage was discarded from the [22]. Production runs were then carried out, during which thermodynamic and structural quantities were collected [23]. The energy trajectories and replica exchanges were monitored to verify adequate fluctuations and smooth movement of replicas across temperatures [24].

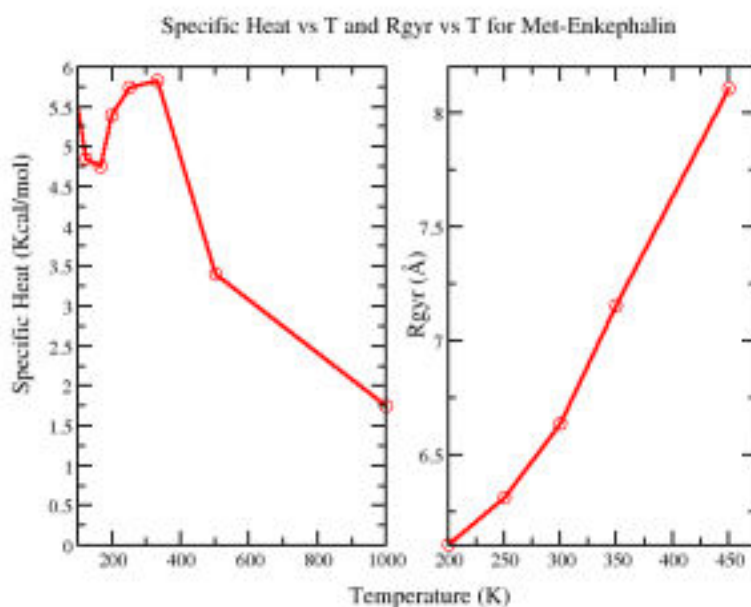


Figure 2: Specific Heat versus Temperature and radius of gyration versus temperature.

1 Thermodynamic Properties

The ensemble-averaged energy at temperature T is given by

$$\langle E(T) \rangle = \frac{1}{N} \sum_{i=1}^N E_i \quad (3)$$

where E_i denotes the instantaneous energy of the i th configuration and N is the total number of samples. The heat capacity at constant volume was obtained from energy fluctuations:

$$C_V(T) = \frac{\langle E^2 \rangle - \langle E \rangle^2}{k_B T^2} \quad (4)$$

2 Structural Properties

After equilibration, configurations at temperature T were used to compute the average radius of gyration,

$$\langle R_g(T) \rangle = \frac{1}{N_T} \sum_{k=1}^{N_T} R_g(k) \quad (5)$$

where N_T is the number of configurations sampled at temperature T .

Details of the Simulation

All-atom simulations were performed for two polypeptide systems using the ECEPP/3 force field, which includes electrostatic, hydrogen-bonding, van der Waals, and torsional energy components. Solvent effects were incorporated using the solvent-accessible surface area (SASA) method. The simulations were carried out with a locally modified version of the SMMP package. In each Monte Carlo move, a single dihedral angle was randomly selected and reassigned a new value uniformly distributed between -180° and $+180^\circ$. Parallel Tempering Monte Carlo (PTMC) simulations were employed to investigate conformational sampling and the folding–unfolding transitions of the two protein systems [23].

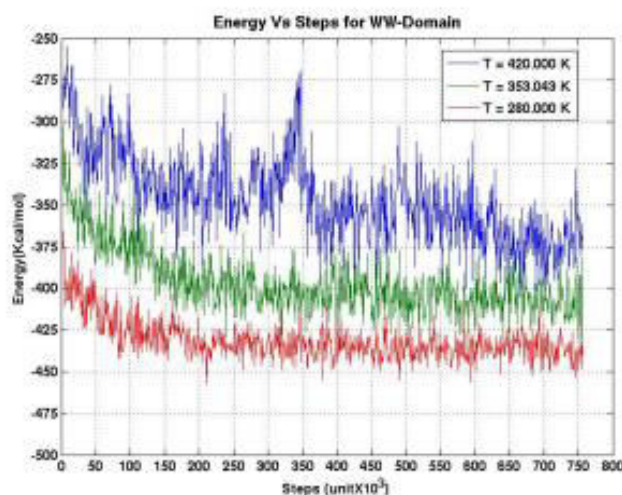


Figure 3: Energy versus MCS at different temperatures using PTMC simulation for WW-Domain protein. From top to bottom the temperatures are 420 K (blue), 350 K (green), and 280 K (red).

To evaluate convergence and sampling efficiency, Met-enkephalin was first studied using PTMC at four temperatures (200 K, 250 K, 300 K, and 350 K). Each simulation consisted of 5 lakh Monte Carlo steps per temperature, and statistical averages were obtained from ten independent trajectories initiated from different starting conformations. Convergence was monitored through the evolution of total energy, and the lowest-energy structures were compared with literature values. The folding transition temperature was identified by examining the overlap between radius-of-gyration versus temperature curves and specific-heat versus temperature plots [16].

After establishing reliability using Met-enkephalin, the simulations were extended to the WW domain protein [24]. PTMC was again applied, which significantly enhanced sampling across temperatures. Results were averaged over eight independent runs initiated from distinct initial structures. The analysis included energy trajectories, inspection of energy histograms for neighboring temperatures to assess convergence and low-temperature sampling, and monitoring of replica-exchange events. Low-energy PDB structures were also compared with reported minimum-energy reference structures to evaluate the limitations of PTMC for larger protein systems.

Results and Discussion

1 Met-Enkephalin

The figure 1 shows the variation of potential energy as a function of Monte Carlo (MC) sweeps for Met-enkephalin obtained from Parallel Tempering Monte Carlo (PTMC) simulations at four temperatures (250, 300, 350, and 450 K). At all temperatures, the energy fluctuates extensively throughout the entire simulation window, indicating that the system explores a wide region of conformational space and remains well equilibrated. As temperature increases, the average energy shifts to higher values and the amplitude of fluctuations becomes larger, reflecting enhanced sampling and access to higher-energy conformations at elevated temperatures. The absence of long plateaus suggests that the system is not trapped in local minima for extended periods and that PTMC allows efficient barrier crossing. Overall, the trajectories demonstrate stable sampling of the canonical energy distribution at each temperature and confirm the effectiveness of PTMC in exploring the rugged energy landscape of Met-enkephalin. The figure 2 displays the temperature dependence of the specific heat and radius of gyration (R_{gyr}) of Met-enkephalin. The specific heat curve shows a pronounced maximum in the low- to mid-temperature range, indicating a cooperative structural transition associated with increased conformational fluctuations.

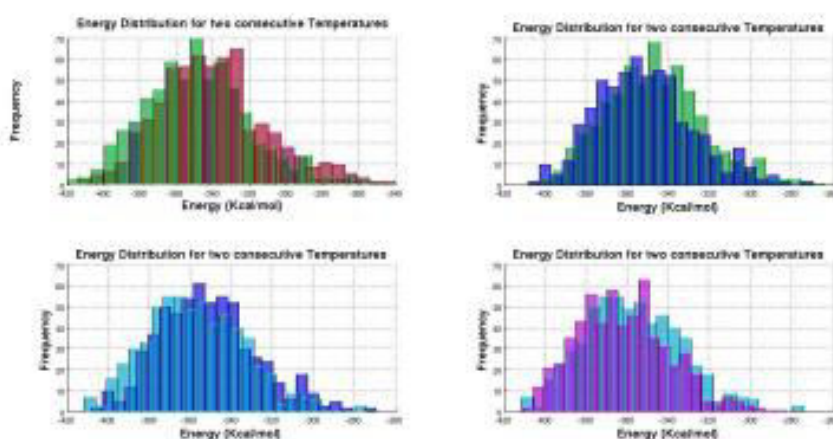


Figure 4: Frequency (number of visits) versus Energy (kcal/mol) for the two consecutive temperatures

Beyond this peak, the specific heat decreases with temperature, suggesting that most major structural rearrangements have already occurred and the system gradually approaches a more disordered, thermally expanded state. The radius of gyration increases monotonically with temperature, reflecting progressive unfolding and expansion of the peptide as thermal energy rises. Taken together, the peak in specific heat and the increase in R_{gyr} are consistent with a folding–unfolding transition of Met-enkephalin across the studied temperature range.

2 WW-Domain

The figure 3 presents the variation of potential energy with simulation steps for the WW-Domain protein at three different temperatures: 280 K, 353 K, and 420 K. At the beginning of the simulation, a rapid decrease in energy is observed for all temperatures, indicating relaxation from the initial high energy configurations toward more stable states. After this equilibration phase, the energy fluctuates around well-defined average values, showing that the system has reached a steady sampling regime at each temperature. As expected, the mean energy decreases with decreasing temperature: the lowest energy is obtained at 280 K,

followed by 353 K, while the highest energy corresponds to 420 K. The amplitude of fluctuations is also temperature dependent, with larger fluctuations at higher temperature due to increased thermal motion and broader conformational sampling. Overall, the plot reflects the equilibration of the system and demonstrates temperature-dependent stability, with lower temperatures favoring more compact, low-energy conformations and higher temperatures favoring higher-energy unfolded or partially unfolded states.

Figure 4 show energy distributions at pairs of consecutive temperatures from a parallel tempering or multi-temperature Monte Carlo simulation of a biomolecule. Each panel overlays two histograms, illustrating how the system's potential energy is distributed at each of the two temperatures. The curves are broader and shifted toward higher energies at higher temperature, reflecting increased thermal fluctuations and access to less stable conformations, while lower temperatures favor lower-energy, more compact states. The important feature is the overlap between neighboring energy distributions—visible in each pair—which indicates that replicas at adjacent temperatures sample overlapping energy ranges. This overlap is essential for efficient replica exchange, ensuring good mixing across temperatures and enabling the system to escape local minima, thereby improving sampling of the energy landscape.

We have also presented some WW-Domain protein structures. Figures 5(a) show standard NMR structure. figure 5(b) represents the initial random structures and Figure 5(c) illustrates the lowest energy structure of the WW-Domain. However, the lowest energy structure obtained from PT simulation does not match the standard NMR structure [1]. This may be due to the need for running a larger number of Monte Carlo (MC) steps, as the size of the WW-Domain protein is nearly eight times greater than of the Met-enkephalin protein.

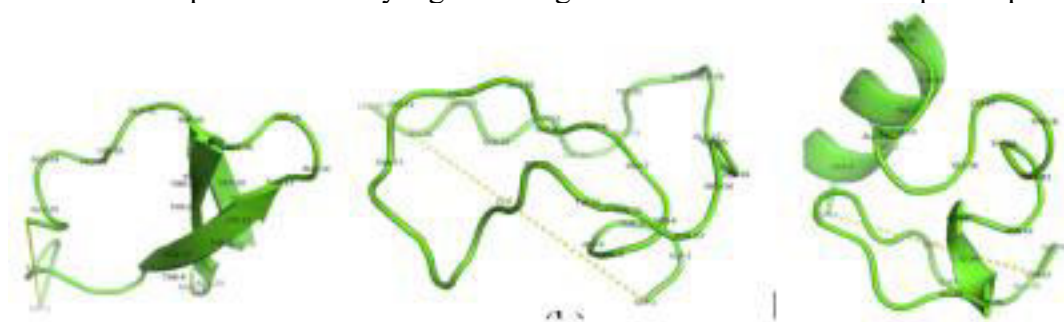


Figure 5: Standard experimental structure (a), (b) Initial random structures and (c) and minimum energy structures from this work.

Conclusion

This study evaluates the efficiency of conformational sampling in protein energy landscapes using Parallel Tempering Monte Carlo (PTMC), with Met-enkephalin and the WW-Domain serving as model systems. For Met-enkephalin, PTMC significantly enhanced sampling across the full temperature range, particularly in the low-energy region, enabling comprehensive exploration of conformational space and robust identification of low-energy states. Notably, the global minimum structure was successfully located with an energy of -11.23 kcal/mol and an RMSD of 1.59 Å relative to the experimentally known reference structure, demonstrating excellent structural agreement.

In the case of the WW-Domain, the overlap of energy distributions between neighboring

temperatures confirms adequate sampling and thermodynamic consistency, while energy versus MC Simulation profiles indicate stable convergence after equilibration, with increased fluctuations at higher temperatures. However, the standard global minimum structure of the WW-Domain was not attained, suggesting that increased system size demands further optimization. These results highlight the necessity of improved temperature ladders, a larger number of replicas, and enhanced sampling strategies to accurately capture the minimum-energy conformations of larger proteins. Future work will therefore focus on refining PTMC protocols to achieve more reliable and efficient characterization of larger protein domains.

References

- [1] M. J. Macias, V. Gervais, C. Civera, and H. Oschkinat, "Structural analysis of WW domains and design of a WW prototype," *Nat. Struct. Biol.* 7, 375 (2000).
- [2] S. Okamoto, H. Okamura, Y. Akagi, N. Yanaihara, and Y. Ibata, "Possible induction of [Met]enkephalin-Arg⁶-Gly⁷-Leu⁸ immunoreactivity in neurons of the rat superior colliculus following eye enucleation," *Neurosci. Lett.* 120, 74 (1990).
- [3] J. Lee and H. A. Scheraga, Conformational space annealing by parallel computations: extensive conformational search of Met-enkephalin and of the 20-residue membrane-bound portion of melittin, *Int. J. Quantum Chem.* 75, 255 (1999).
- [4] D. A. Evans and D. J. Wales, The free energy landscape and dynamics of met-enkephalin, *J. Chem. Phys.* 119, 9947 (2003).
- [5] F. Karl, A free energy principle for biological systems, *Entropy* 14, 2100 (2012). 6
- [6] J. P. Doye and D. J. Wales, On potential energy surfaces and relaxation to the global minimum, *J. Chem. Phys.* 105, 8428 (1996).
- [7] S. Kothiwale, J. L. Mendenhall, and J. Meiler, BCL::Conf: small molecule conformational sampling using a knowledge-based rotamer library, *J. Cheminform.* 7, 47 (2015).
- [8] M. Y. Shen and K. F. Freed, All-atom fast protein folding simulations: the villin headpiece, *Proteins* 49, 439 (2002).
- [9] P. K. Singh, K. Sharma, A. Kumar, and M. Shukla, Effects of functionalization on the mechanical properties of multiwalled carbon nanotubes: A molecular dynamics approach, *J. Compos. Mater.* 51, 671 (2017).
- [10] D. R. Roe, C. Bergonzo, and T. E. Cheatham III, Evaluation of enhanced sampling provided by accelerated molecular dynamics with Hamiltonian replica exchange methods, *J. Phys. Chem. B* 118, 3543 (2014).
- [11] W. Janke, Multicanonical Monte Carlo simulations, *Physica A* 254, 164 (1998).
- [12] A. D. A. Caparica and A. G. Cunha-Netto, Wang–Landau sampling: improving accuracy, *Phys. Rev. E* 85, 046702 (2012).
- [13] J. Kästner, Umbrella sampling, *Wiley Interdiscip. Rev.: Comput. Mol. Sci.* 1, 932 (2011).
- [14] S. Syed, V. Romaniello, T. Campbell, and A. Bouchard-Côté, Parallel tempering on optimized paths, in *Proc. Int. Conf. Mach. Learn.* (PMLR, 2021), pp. 10033–10042.
- [15] P. Singh, N. C. Verma, and S. Jain, BCL::Conf: Parallel Tempering Monte Carlo Simulation of Met-enkephalin and WW-Domain proteins, *AIMS Biophysics.* 7, 47 (2025).
- [16] P. Singh, S. K. Sarkar, and P. Bandyopadhyay, Understanding the applicability and limitations of Wang–Landau method for biomolecules: Met-enkephalin and Trp-cage, *Chem. Phys. Lett.* 514, 357 (2011).
- [17] P. Singh, S. K. Sarkar, and P. Bandyopadhyay, Wang–Landau density of states based study of the folding-unfolding transition in the mini-protein Trp-cage (TC5b), *J. Chem.*

- Phys.* 141, 014101 (2014).
- [18] A. Mitsutake, Y. Mori, and Y. Okamoto, Enhanced sampling algorithms, in *Biomolecular Simulations: Methods and Protocols*, 153–195 (2012).
 - [19] K. Y. Sanbonmatsu and A. E. Garc'ia, Structure of Met-enkephalin in explicit aqueous solution using replica exchange molecular dynamics, *Proteins* 46, 225 (2002).
 - [20] J. P. Ulmschneider and W. L. Jorgensen, Monte Carlo backbone sampling for polypeptides with variable bond angles and dihedral angles using concerted rotations and a Gaussian bias, *J. Chem. Phys.* 118, 4261 (2003).
 - [21] B. A. Berg and H. P. Hsu, Metropolis simulations of Met-enkephalin with solvent-accessible area parameterizations, *arXiv:cond-mat/0306435* (2003).
 - [22] A. Ghanizadeh, S. Bhowmik, O. Haeri-Ardakani, H. Sanei, and C. R. Clarkson, A comparison of shale permeability coefficients derived using multiple non-steady-state measurement techniques: Examples from the Duvernay Formation, Alberta (Canada), *Fuel* 140, 371 (2015).
 - [23] A. Khachaturyan, S. Semenovsovskaia, and B. Vainshtein, The thermodynamic approach to the structure analysis of crystals, *Found. Crystallogr.* 37, 742 (1981). M. J. Macias, V. Gervais, C. Civera, and H. Oschkinat, Structural analysis of WW domains and design of a WW prototype, *Nat. Struct. Biol.* 7, 375 (2000)
 - [24] B. Andre B., and J. Y. Springael, WWP, a New Amino Acid Motif Present in Single or Multiple Copies in Various Proteins Including Dystrophin and the SH3-Binding Yes-Associated Protein YAP65 *Biochemical and Biophysical Research Communications* 205, 1201-1205 (1994)

45. Dielectric Relaxation in NiCr_2O_4 Spinel Chromites: Temperature and Frequency Dependent Dielectric Measurements

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Abstract: Nickel Chromite (NiCr_2O_4) is one of the fascinating exchange materials. The sample has been synthesized via sol-gel method. It has a cubic structure (space group $fd-3m$, space group No 227, crystallite size is 28nm) at high temperature and at low temperature undergoes phase transitions. The material shows the combination of functional properties. It exhibits ferrimagnetic behavior and displays the exchange bias effect. The material has the intriguing dielectric properties also. These properties make NiCr_2O_4 a material of interest for potential application in the area magnetoelectric coupling and tunable dielectric response. We have studied the dielectric properties of the material, with Frequency scan in the range of 1MHz to 1Hz, and performed a temperature scan as well. Analysis of the data reveals information on real and imaginary part of the dielectric permittivity (ϵ' , ϵ''), dielectric loss ($\tan \delta$) and other parameters of interest. The dielectric constant of NiCr_2O_4 , decreases with increasing frequency. The variation of dielectric constant and the loss could be understood by Cole-Cole Plot, Arrhenius plot of relaxation time, Koops and Maxwell-Wagner theories to understand the dielectric behavior of the NiCr_2O_4 . Results will be discussed in the light of literature.

Keywords: Spinel, Dielectric permittivity, Dielectric loss, Complex Impedance, Temperature Dependent,

Introduction:

Nickel chromite is a normal spinel material, is known for its phase transition at deferent temperatures from room temperature to low temperatures. NiCr_2O_4 is multifunctional material, [1] which combine two or more primary properties such as magnetism and magneto dielectric properties. This multifunctionality is the major thrust in modern condensed matter physics and in material science. Among the materials of interest, transition metal oxides with the spinel structure (AB_2O_4) are particularly interesting due to their reach magnetic, electronic and structural properties. NiCr_2O_4 is well-known for its magnetic behavior (ferrimagnetic in nature) due to highly frustrated pyrochlore -type lattice formed by the Cr^{3+} ions at the octahedral B-sites. [2] This magnetic frustration leads to successive structural and magnetic transitions upon cooling, including a Jahn-Teller-Induced structural distortion and complex ferrimagnetic/antiferromagnetic ordering at low temperatures. NiCr_2O_4 is a fascinating dielectric material with high dielectric value make it a good dielectric candidate.

The applications of Nickel chromite (NiCr_2O_4) are primarily driven by its multiferroic nature, which stems from the strong coupling between its magnetic and dielectric (electrical) properties. This magnetodielectric [3] coupling positions NiCr_2O_4 as a promising material for multiferroic and magnetoelectric devices, magnetic field sensors, magnetoelectric (ME) memory devices. Furthermore, in its nanocrystalline form, the material exhibits a large dielectric constant, makes it suitable for high performance dielectric applications such as high-density energy storage in electrochemical supercapacitors and lithium-ion batteries, as well as high-performance capacitors. The complex magnetic structure of the material makes it well deserving candidate for the magnetic and magneto structural applications, like magnetic switching and storage, spintronics and specialized magnetic recording media. [4]

Experimental details:

The sample is synthesized by the sol-gel auto combustion method. The material was characterized to determine its structural, optical, and physical (magnetic and electrical) properties. Room temperature Powder X-ray Diffraction (XRD) was performed using a D8 advance spectrometer ($\lambda=1.5406$) in the 2θ from 10° to 80° to investigate the crystal structure. Broadband Impedance Analyzer (Novo control) facility at the UGC-DAE CSR Indore Centre has been used for conducting dielectric measurements. This specialized instrument is crucial for the precise characterization of a material's electrical response across a broad frequency spectrum. [5] [6]

Results and Discussion:

1 Structural Properties

The structural characterization and X-ray diffraction (XRD) patterns of this sample have been reported previously and only parameters of relevance are discussed here. The XRD analysis confirmed the formation of a single-phase structure. We identify the sharp diffraction peaks which conform the polycrystalline nature of the sample. Fullprof Rietveld refinement of the XRD data confirmed the single-phase cubic spinel structure with $Fd\bar{3}m$ space group. Results are in good agreement with the reported literature. The sharp diffraction peaks indicate high crystallinity, through which one can calculate average crystallite size that turns out to be 28nm. The lattice parameter is $a=b=c=8.2994$ Å and $\alpha=\beta=\gamma=90^\circ$. In NiCr_2O_4 , Ni^{2+} ions were fixed at the tetrahedral (8a) and for Cr^{3+} ions occupied the octahedral site (16d) and the O fixed at octahedral site (32). We can determine the oxygen positional parameter in the oxygen octahedron, which indicates positional disorder and hence influences the materials dielectric and magnetic properties. We have also used Stanley-Siegel equation to calculate the bond length and hopping length of both sites (Tetrahedral and octahedral). We have estimated the oxygen positional parameter (u). For the Ideal spinel u is 0.375 but we get the u as 0.386, slightly higher value. This may indicate the distortion in the oxygen octahedral, which directly influences the materials dielectric and magnetic properties [7].

2: Dielectric Permittivity Analysis for frequency Scan (Electric Modulus Analysis (ϵ' , ϵ''))

Dielectric analysis (Frequency scan) of synthesized sample (NiCr_2O_4) reveals the important information about the dielectric behavior of the sample. The complex permittivity is generally expressed by the following equation: $\epsilon(\omega) = \epsilon' - i\epsilon''$. The real part of complex permittivity is denoted by the ϵ' while the imaginary part as ϵ'' . Here is the angular frequency

($\omega=2\pi f$). Analysis reveals the dielectric response of the sample as well. Fig 1(a) represent the ϵ' (real part of dielectric constant).

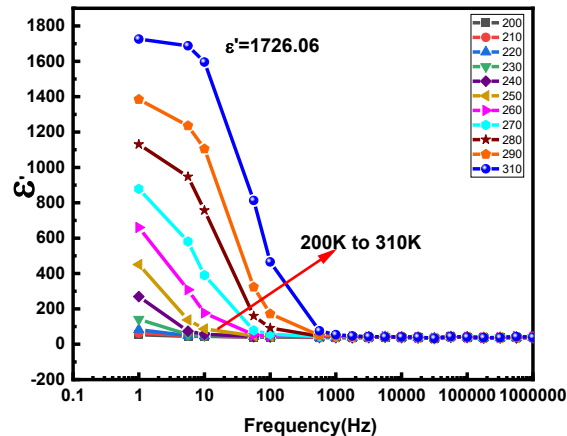


Figure:1 (a) Real part of dielectric (ϵ') by using the frequency scan and temperature ranges from (200K to 310K)

This is related to the materials ability of the dielectric medium to storage electric energy, by becoming polarized in response to an applied electric field. quantifies the extent to which the materials internal charges can shift and form electric dipoles when an external electric field is applied. For the NiCr_2O_4 many researchers report the high value of dielectric constant at room temperature. The high value of dielectric constant indicates the material polarizes more easily and can storage more electrical energy, in the electric field. In the case of NiCr_2O_4 researchers get the higher value of dielectric constant [8]. ϵ'' (imaginary part of dielectric constant) is related to the dissipation or absorption of electric energy in the material. The frequency dependent imaginary part of dielectric permittivity (ϵ'') of NiCr_2O_4 was investigated across a temperature range 200K to 150K as showed in the Fig 1(b). The spectra reveal a significant dielectric dispersion in low-frequency region (10^1 to 10^3). The value of ϵ'' is higher at 200K. This increase in dielectric loss at low frequency is a characteristic of due to Maxwell-Wagner-Sillars (MWS) [8] polarization mechanism. This phenomenon typically happened due to grain boundaries. As the frequency increases 10^3 Hz, we observed rapid decline in the ϵ'' .

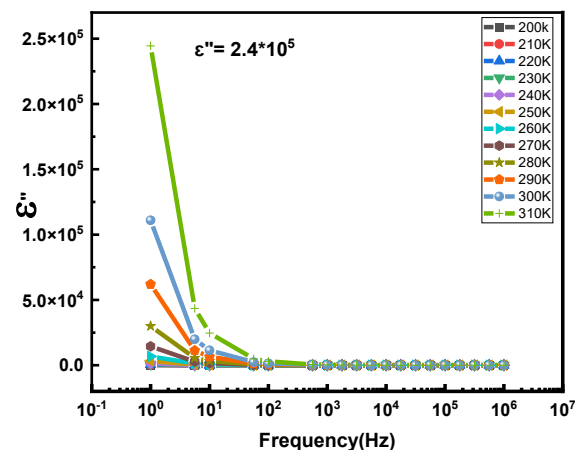


Figure:1(b)Frequency dependence of imaginary part of dielectric permittivity (ϵ'') at various temperatures ranging from 200k to 310k. The significant increase in ϵ'' at lower frequencies and higher temperatures indicates due to interfacial polarization.

Dielectric Loss ((tan δ):

The ratio of imaginary part to the real part of dielectric is called the loss tangent and can be expressed by the using formula $\frac{\epsilon''}{\epsilon'} = \tan(\delta)$. In NiCr_2O_4 spinel material the value

of ϵ'' is very high is (up to 10^5), the value of tangent loss is significantly high at low frequencies, shows that the material is more dissipative than capacitive in this range. The frequency-dependent loss tangent ($\tan \delta$) of NiCr_2O_4 exhibits a characteristic behaviour decrease with increasing frequency, reaching a minimum at the high-frequency limit. This behaviour shown in Fig 1c can be explained by the using is Koop's. model, where the grain boundaries dominate the low- frequency response and the grains dominate high frequencies [8].

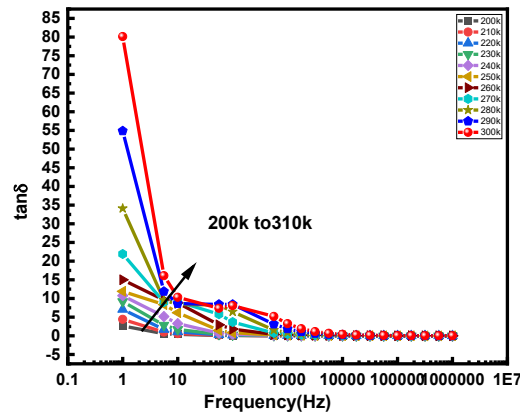


Figure:1c. Frequency -dependent variation in Loss tangent ($\tan \delta$) of the sample (NiCr_2O_4) at experimental temperatures 200K to 300K

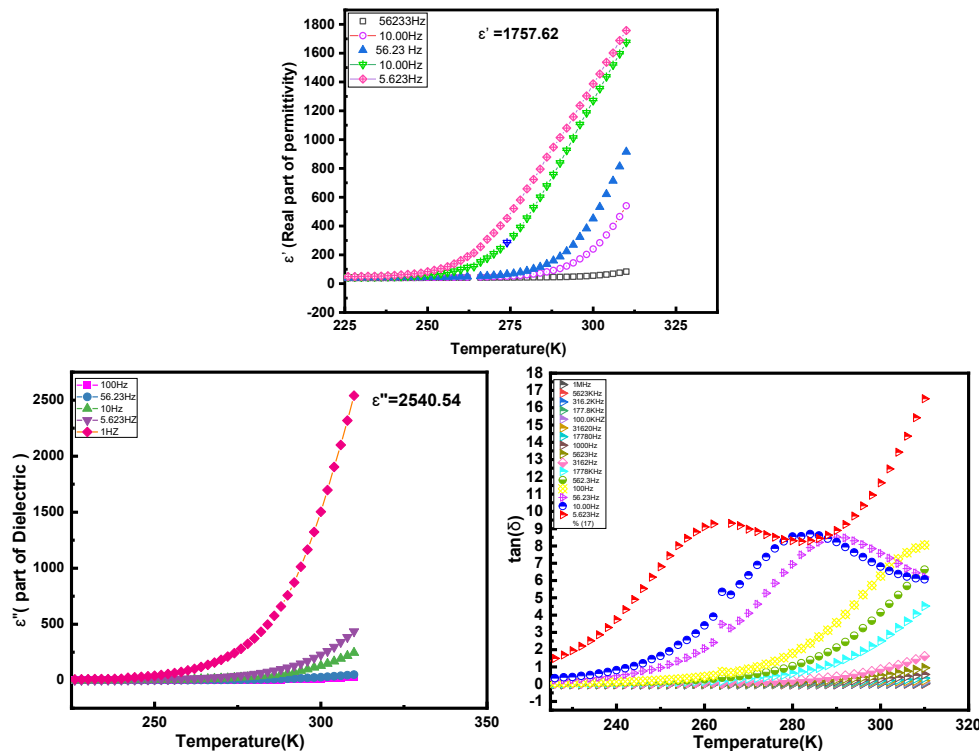


Figure 2: Temperature dependence of real part of dielectric permittivity(ϵ') (b)imaginary part of dielectric permittivity(ϵ'') at various temperatures ranging from 200k to 310k. (c) Tan δ (dielectric loss)

3: Dielectric analysis (Temperature scan) of synthesized sample (NiCr₂O₄)

Temperature scan in dielectric measurement is important because it reveals the material's properties that cannot be explained by the room temperature measurements. For the material like NiCr₂O₄ that have different structural, magnetic, electrical transition at the different temperatures. The experiment was performed to investigate the relaxation dynamics and phase stability synthesis sample. The temperature scan is critical to understand the behavior and interplay of grain boundary of the material and it also important to identifying magnetodielectric coupling near the magnetic transitions. Arrhenius analysis conforms thermally activated hopping conduction mechanism by which we can explain the semiconductor behavior of the sample. Real part of dielectric permittivity(ϵ'), imaginary part of dielectric (ϵ''), and tangent loss for the temperature scan is shown in the Fig. 2(a), 2(b) and 2(c) respectively [3].

Conclusion:

In conclusion we have prepared NiCr₂O₄ sample and studied its dielectric response in the frequency range 1Hz-1MHz and at varying temperatures from 200-310K. The results of XRD has been used to understand the possible origin of dipoles and the broad band dielectric spectroscopy is used to understand its electrical response. The real and imaginary part of the dielectric constants reveal a dispersive behaviour especially at low frequency ranges up to kHz range indicating the dominance of Maxwell-Wagner-Sillars (MWS) polarization mechanism. The temperature dependence shows that the dispersion is quite contrasting after 250K only. This shows the tunability of the dielectrics upon varying the temperature.

Acknowledgement:

We thank MPCST (File No-A/RD/RP-2/321) for the funding of the project and a JRF fellowship to PY. We profusely thank all the authorities at MPCST Bhopal especially the Director General, MPCST, Bhopal and Dr Manoj K Rathore for the support and guidance. We also thank UGC-DAE CSR (Project No- CRS/2022-23/03/877) for the experimental support. We also grateful and thankful for the authorities at Medicaps University Indore for URF (project URF-MU-2022-08) and SRF (project MU/REG/ORDER/2025-26/76).

References:

- [1] A. Maignan, C. Martin, K. Singh, C. Simon, O. I. Lebedev, and S. Turner, *From Spin Induced Ferroelectricity to Dipolar Glasses: Spinel Chromites and Mixed Delafossites*, J. Solid State Chem. **195**, 41 (2012).
- [2] K. Tomiyasu and I. Kagomiya, *Magnetic Structure of NiCr₂O₄ Studied by Neutron Scattering and Magnetization Measurements*, J. Phys. Soc. Japan **73**, 2539 (2004).
- [3] C. M. Zhu, G. B. Yu, L. G. Wang, M. W. Yao, F. C. Liu, and W. J. Kong, *Dielectric Relaxation and Magnetodielectric Effect in the Spinel NiCr₂O₄*, J. Magn. Magn. Mater. **506**, 1 (2020).
- [4] K. Nadeem, M. Kamran, H. Z. Khokhar, I. Ahmed, F. Zeb, and N. A. Noshahi, *Effect of Mg Doping on Magnetic, and Dielectric Properties of NiCr₂O₄ Nanoparticles*, Ceram. Int. **48**, 17270 (2022).
- [5] W. Liguang, Z. Changming, T. Zhaoming, and Y. Songliu, *Exchange Bias and Training Effect in NiCr₂O₄/Cr₂O₃ Composite*, **3**, 2 (2015).
- [6] E. Grazenaite, J. Pinkas, A. Beganskiene, and A. Kareiva, *Sol-Gel and Sonochemically*

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- Derived Transition Metal (Co, Ni, Cu, and Zn) Chromites as Pigments: A Comparative Study*, Ceram. Int. **42**, 9402 (2016).
- [7] M. Z. Ahsan and F. A. Khan, *Study of Structural, Electrical and Magnetic Properties of Manganese Doped Cobalt Ferrite Nanoparticles with Non-Stoichiometric Composition*, J. Phys. Sci. Appl. **7**, 30 (2017).
- [8] M. Javed, A. A. Khan, J. Kazmi, M. A. Mohamed, M. N. Khan, M. Hussain, and R. Bilkees, *Dielectric Relaxation and Small Polaron Hopping Transport in Sol-Gel-Derived NiCr₂O₄ Spinel Chromite*, Mater. Res. Bull. **138**, 111242 (2021).

46. Dielectric Behavior in Paramagnetic Region of CoCr_2O_4

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Abstract: In the present work, we report the Dielectric behaviour of Multiferroic Cobalt Chromite CoCr_2O_4 formed by the solid-state reaction method at a sintering temperature of 1200 °C, which shows a cubic spinel structure with the $Fd3m$ space group and a lattice parameter of 8.3106 Å. The real part (ϵ') and imaginary part (ϵ'') of the dielectric constant and loss tangent ($\tan\delta$) etc are measured in the paramagnetic region 200K -300K. The results are explained using Maxwell–Wagner and Koop’s phenomenological theory of temperature-dependent dielectric properties. The dielectric parameters decrease as frequency increases from 1KHz to 1MHz. This study highlights the paramagnetic dielectric behaviour of CoCr_2O_4 and has been investigated in the temperature range where the nature of system is paramagnetic. This study is expected to show effects of paramagnetic fluctuations and interfacial polarization if any. In this temperature regime, CoCr_2O_4 remains in a paramagnetic state, where there is no long-range magnetic ordering is seen.

Keywords: Multiferroic, Spinel, Maxwell–Wagner-type interfacial, Koop’s phenomenological theory

Introduction

In recent years, there has been a renewed interest in exploring the systems in which spin degrees of freedom and charge are coupled. studies on systems that exhibit an interaction between their magnetic and electronic properties, Such as magnetodielectric materials and Magnetic semiconductors have demonstrated the impact of spin-charge coupling on material. Spinel oxides with the general formula AB_2O_4 has exhibit significant interest in there Structural, magnetic and electric properties. Among them, CoCr_2O_4 (Cobalt Chromite) is normal spinel chromite with a tetrahedral A site occupied Co^{2+} Cation and an octahedral B site occupied Cr^{3+} cation and both Co^{2+} and Cr^{3+} are magnetic ¹. CoCr_2O_4 has a transition from Paramagnetic region to long range ferrimagnetic ordering T_C at 94K followed by Spin re-orientation transition T_S at 21K and Lock in transition T_L at 9K with Cubic Structure $Fd3m$ space group. While Co^{2+} and Cr^{3+} both are magnetic, the CoCr_2O_4 is primally governed by nearest neighbour and isotropic antiferromagnetic A-B and B-B exchange interactions².

Chromite material have excellent dielectric properties, including high resistivity and low dielectric loss, making them ideal for high frequency and multifunctional device applications. Chromites are particularly appealing in multiferroic and magnetodielectric research because magnetic ordering and spin lattice interaction can have a significant impact on their dielectric response. CoCr_2O_4 is an ideal choice in the Chromite family due to its chemical stability.

These properties make CoCr_2O_4 ideal for dielectric, magnetodielectric, and multiferroic applications, such as chemical sensors, microwave devices, catalysts, and sophisticated magnetic-electronic multifunctional systems³.

Experimental Details

The sample was prepared via solid-state reaction method which is one of the primary and most commonly used procedures for producing spinel chromites. Sample were denoted by CCO. This approach involves mixing precursor powders of Cobalt oxide (Co_3O_4), and Chromium oxide (Cr_2O_3), which were used in a stoichiometric ratio as a basic precursor and then grinding them to make sure homogeneity. The mixture is then under high-temperature calcination, typically at 1000°C to 1200°C , allowing for solid-state processes that result in the spinel phase. The final product is often treated to multiple grinding and sintering processes to improve phase purity and crystallinity⁴.

Using the Advanced D8 Bruker X-Ray Diffractometer, the phase confirmation and XRD pattern were performed in 2θ (10 to 80) with step size 0.02° at a wavelength of $1.5406\text{\AA}$. The Dielectric Study was carried out as a function of Temperature using (Dielectric-setup) Broadband Impedance Analyzer (Novo-Control) in the temperature range 200K to 300K at a frequency range 1Hz to 1MHz, Dielectric measurement was performed on a small plate with parallel capacitor geometry. Silver paste was used to make the electrodes⁵.

Result and Discussion

1 Structural Analysis

The XRD pattern of the CoCr_2O_4 (CCO) in the paramagnetic state analysed through Rietveld refinement technique using the Fullprof suite program is shown in Figure 1. XRD pattern confirm that CCO has a cubic spinel structure with $\text{Fd-}3\text{m}$ (227) space group and the lattice parameters a is $8.3106\text{ \AA}$. The refinement showing a good fit between the calculated and observed patterns⁶. A black solid line reflects the pattern established by the structural model, and red asterisks highlight the observed data. Using the Rietveld refinement (not shown here), a good agreement exists between the estimated and observed profile is established⁷.

2 Dielectric Measurement

The dielectric response of CCO was studied in the paramagnetic regime (the temperature range 200K-300K) as a function of temperature at various frequency. The different dielectric parameters provide insight into polarization processes, charge carrier dynamics, and relaxation mechanisms⁸.

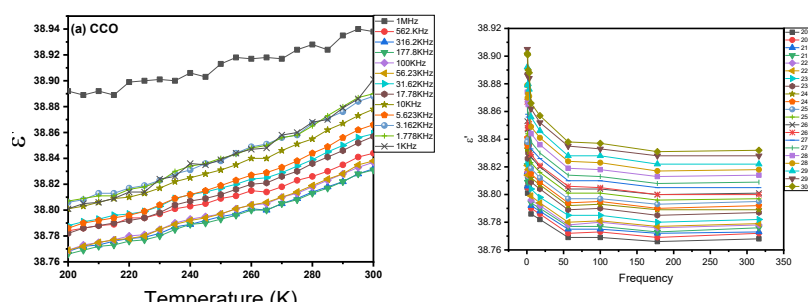


Fig. 1 (a) Temperature dependence of the real part of the dielectric constant of CCO measured in the paramagnetic temperature range 200-300K at various frequencies from 1MHz to 1KHz. (b) Frequency dependent real part of dielectric constant CCO measured at selected temperature (200-300K).

3.1 Real part of Dielectric Constant (ϵ')

The real part of dielectric constant is decreasing with the decreasing temperature for all frequency range, exhibiting the normal dielectric dispersion expected in cobalt chromite. The temperature and frequency dependent real part of dielectric constant are shown in Figure 2 (a) and (b) respectively. The temperature dependent real part of dielectric constant shows a monotonic decreasing (of-course only a few percent) with temperature from 200K to 300K for all frequencies (1MHz to 1KHz)^{9,10}. The reduction of dielectric constant with temperature indicates a thermally inactive polarization process, which is typical insulating chromite in paramagnetic regime. There is no sharp anomaly or peak in the paramagnetic region, which confirm the absence of any magnetic or phase transition above the ferrimagnetic ordering temperature ($T_c = 94K$)¹¹.

The frequency-dependent real part of dielectric constant at a given temperature, ϵ' exhibits relatively higher values at lower frequencies for all measured temperatures. As the frequency increases from low to intermediate value, ϵ' show a gradual decayed. At a higher frequencies, a re-increase in ϵ' is observed^{11,12}.

3.2 Imaginary part of Dielectric Constant ϵ''

The imaginary part of the dielectric constant and its variation with frequency and temperature variation are depicted in the following figures 2a and 2b. Even though the temperature variation is quite contrasting the actual values are small. The frequency dependence is much more fascinating as there is no discernible loss is seen in Figure 2b. This shows a tendency of robust and no lossy state of the material at the investigated temperatures.

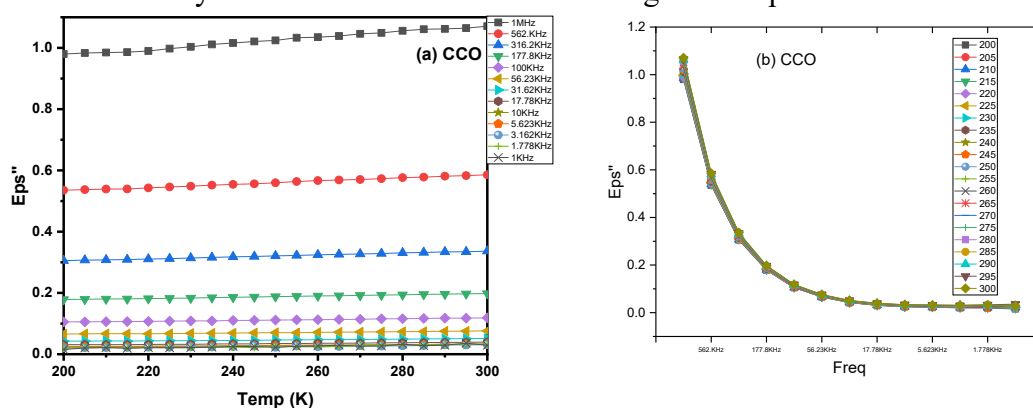


Fig. 2 (a) Temperature dependence of the imaginary part of the dielectric constant of CCO measured in the paramagnetic temperature range 200-300K at various frequencies from 1MHz to 1KHz. (b) Frequency dependent imaginary part of dielectric constant CCO measured at selected temperature (200-300K).

3.3 Dielectric loss $\tan\delta$

The temperature and frequency dependent dielectric loss of CoCr_2O_4 in the paramagnetic region are shown in fig. 3 (a) and (b) respectively. The temperature dependent dielectric loss shows the gradual increase with temperature for all frequency. The smooth temperature variation in the paramagnetic regime indicate that dielectric loss in this range is dominated by relaxation mechanism¹³.

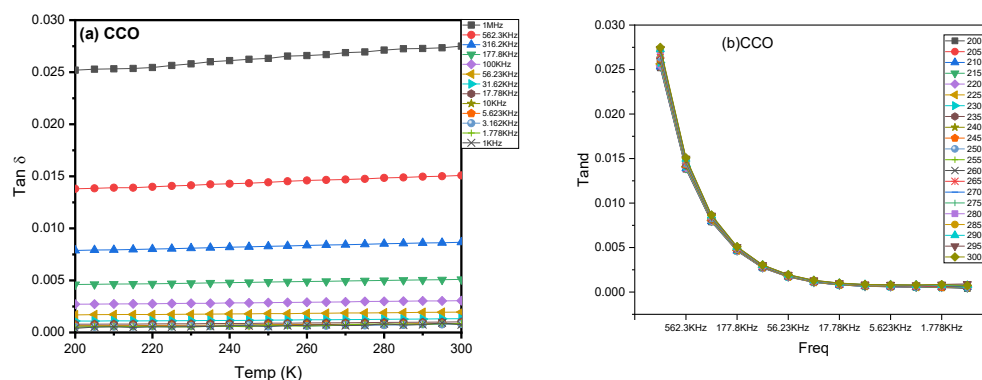


Fig. 3 (a) Temperature dependence of the dielectric loss of CCO measured in the paramagnetic temperature range 200-300K at various frequencies from 1MHz to 1KHz. (b) Frequency dielectric loss CCO measured at selected temperature (200-300K).

The frequency dependent dielectric loss, in temperature range 200 to 300K shows higher value of dielectric loss at higher frequency and low dielectric loss at lower frequency, followed by a linear increase. The increase in dielectric loss at higher frequencies is due to relaxation and conduction losses^{14,15}.

Conclusion

In conclusion we have prepared CoCr₂O₄ sample and studied its dielectric response in the frequency range 1Hz-1MHz and at varying temperatures from 200-300K. The results of XRD shows that it is single phase. The idea of crystallite size helps to understand the possible origin of microstructure and the broad band dielectric spectroscopy is used to understand its electrical response. The real part of the dielectric constants reveal a totally dispersive behaviour especially at all frequency ranges indicating the dominance of Maxwell-Wagner-Sillars (MWS) polarization mechanism. However the imaginary part doesn't show any dispersion which means the system is quite robust at the frequency range measured. The temperature dependence eventhough shows dispersion is quite contrasting as it is only a small fraction as far as the magnitude change is concerned. This shows the robust nature against lossy systems of the dielectrics upon varying the temperature as well as frequencies are concerned.

Acknowledgement:

UGC DAE CSR Indore, India, Medi-Caps University, Indore, India for providing URF (URF-MU-2022-08) and SRF (MU/REG/ORDER/2025-26/76) project for RJ and PY. VG thank MPCST Bhopal, India for providing the project (A/RD/RP-2/321) & UGC DAE CSR, Mumbai, India for providing the project (CRS/2022-23/03/877).

References:

1. Lawes, G. *et al.* Dielectric anomalies and spiral magnetic order in CoCr₂O₄. *Phys. Rev. B - Condens. Matter Mater. Phys.* **74**, 3–8 (2006).
2. Choi, Y. J. *et al.* Thermally or magnetically induced polarization reversal in the multiferroic CoCr₂O₄. *Phys. Rev. Lett.* **102**, (2009).
3. Gowreesan, S. & Kumar, A. R. PT US CR. *Chinese J. Phys.* (2018) doi:10.1016/j.cjph.2018.02.014.
4. Xu, Y. *et al.* Synthesis and Analysis of Zn-Substituted CoCr₂O₄ Spinel Oxide. *J.*

- Supercond. Nov. Magn.* **35**, 753–762 (2022).
5. Choudhary, P. & Varshney, D. Structural, vibrational and dielectric behavior of $\text{Co}_{1-x}\text{M}_x\text{Cr}_2\text{O}_4$ ($\text{M} = \text{Zn, Mg, Cu}$ and $x = 0.0, 0.5$) spinel chromites. *J. Alloys Compd.* **725**, 415–424 (2017).
 6. Kassem, M. A., Abu El-Fadl, A., Nashaat, A. M. & Nakamura, H. Structure, optical and varying magnetic properties of insulating MCr_2O_4 ($\text{M} = \text{Co, Zn, Mg}$ and Cd) nanospinels. *J. Alloys Compd.* **790**, 853–862 (2019).
 7. Khanam, S. *et al.* Study of the Crystallographic and Magnetic Structure in the Nickel Substituted Cobalt Ferrites by Neutron Diffraction. *Mater. Sci. Appl.* **06**, 332–342 (2015).
 8. Torgashev, V. I. *et al.* Magnetic and dielectric response of cobalt-chromium spinel CoCr_2O_4 in the terahertz frequency range. *Phys. Solid State* **54**, 350–359 (2012).
 9. Corliss, L. M. & Hastings, J. M. Magnetic structure of manganese chromite. *J. Appl. Phys.* **33**, 1138 (1962).
 10. Choudhary, P. *et al.* Weak Ferroelectricity and Leakage Current Behavior of Multiferroic CoCr_2O_4 Nanomaterials. *J. Supercond. Nov. Magn.* **32**, 2639–2645 (2019).
 11. Manjunatha, K. *et al.* Structural, electronic, vibrational and magnetic properties of Zn^{2+} substituted MnCr_2O_4 nanoparticles. *J. Magn. Magn. Mater.* **502**, 166595 (2020).
 12. Choudhary, P., Saxena, P., Yadav, A., Rai, V. N. & Mishra, A. Dielectric and ferroelectric properties of CoCr_2O_4 nanoceramics. *J. Adv. Dielectr.* **9**, 1–7 (2019).
 13. Adnan, M. *et al.* Study of magnetic and dielectric properties of $\text{ZnFe}_2\text{O}_4/\text{CoCr}_2\text{O}_4$ nanocomposites produced using sol-gel and hydrothermal processes. *J. Alloys Compd.* **865**, 158953 (2021).
 14. Farid, H. M. T. *et al.* Dielectric and impedance study of praseodymium substituted Mg-based spinel ferrites. *J. Magn. Magn. Mater.* **434**, 143–150 (2017).
 15. Shafqat, M. B. *et al.* Structural, morphological and dielectric investigation of spinel chromite (XCr_2O_4 , $\text{X} = \text{Zn, Mn, Cu}$ & Fe) nanoparticles. *J. Mater. Sci. Mater. Electron.* **30**, 17623–17629 (2019).

47. Impact of Sr and Mg Substitution on the Morphological and Microstructural Characteristics of CuCrO_2

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Abstract: This study examines structural and microstructural properties including surface morphology of few selective Delafossite materials with a generic structure ABO_2 . The chosen material is CuCrO_2 along with its 5% Mg and Sr substitution. X-ray diffraction confirms that the samples retain the rhombohedral $R\bar{3}m$ phase. Crystallite size increases slightly from 55 nm in pure CuCrO_2 to 57 nm in Mg-substituted and 59 nm in Sr-substituted samples, indicating a slight substitutional contribution to crystallite growth. Morphological structure of the samples is examined by Atomic Force Microscopy. Nanoscope IVa is being used to investigate the samples in contact mode. The Mg-substituted sample shows significantly enhanced roughness ($R_{\text{avg}} = 129$ nm; $R_{\text{rms}} = 205$ nm) compared to the Sr-substituted sample ($R_{\text{avg}} = 28$ nm; $R_{\text{rms}} = 42$ nm). Morphology also demonstrated larger grain formation. Skewness and kurtosis analysis is being carried out to understand nature of surfaces and their associated properties. These observations confirm that different substitutions strongly influence the structural and microstructural or morphological evolution of CuCrO_2 . Results may provide an insight for controlled surface tuning in future thin film functional applications.

Introduction:

Delafossite materials fall under ABO_2 class. These are Transparent Conducting Oxides (TCO) known for their functional properties in thin film electronic applications. The materials of this system show contrasting properties like good conductivity, of p-type conductivity and enough optical conductivity. This leads to significantly interesting systems to study. Kawazoe¹, was the first to present such traits of delafossite in CuAlO_2 . The active devices such as P-N junctions, transistors, UV LED's and many other optoelectronic applications can be prepared with this P-type transparent conducting oxide along with N-type TCO's².

Various delafossites have been studied which has 'Cu' at A site and Mn, Cr, Fe at B site in ABO_2 formula. CuCrO_2 is one among these samples that show good properties.³ Some of them are temperature stability, tenability in electronic and phononic transport. CuCrO_2 also show good thermoelectric⁴, Anti-Ferro Magnetic⁵ and gas sensing properties. There are many techniques for synthesizing CuCrO_2 like Solid state reaction, hydrothermal, sol gel etc, Solid state reaction of making CuCrO_2 is a convenient and promising method. However, for practical applications electrical conductivity need to be increased. Substituting the possible acceptors is one such approach to modify the properties of this delafossite. In the present work we have investigated the morphological and microstructural characteristics of Mg and Sr substituted samples of CuCrO_2 ⁴⁻⁵. Atomic Force Microscopy (AFM) has been used to understand some of the interesting properties of surfaces and form an integral part of this study.

XRD Investigation:

X-ray Diffraction is an important fundamental damage-free measurement which can evaluate many types of samples like powders and crystals. XRD is a vital approach for material characterization. XRD is applied for the determination of crystalline material and various polymorphic forms as well. In this work, we used the Bruker D8 Advanced X-ray Diffractometer facility available at UGC-DAE CSR, Indore. In this diffractometer there is an x-ray source, generating X-ray of the wavelength $1.5405\text{\AA}$. With the help of Bruker Lynx eye detector based on Si strip technology, scattered x-rays are detected. A theta-two theta geometry is employed. Powder XRD pattern of 5% Mg and 5% Sr substituted CuCrO_2 samples are studied. All the peaks compare well with the JCPDS and thus the XRD diffraction confirms R-3m space group with rhombohedral crystal structure. Pattern is not shown here.

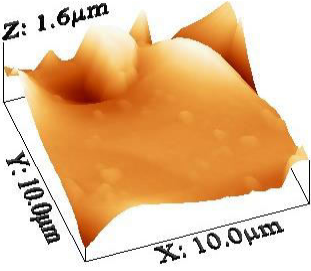
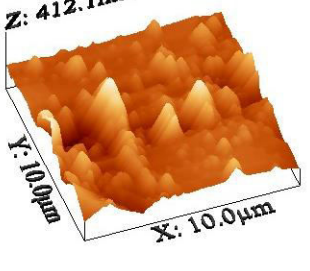
Only the results are discussed especially the crystallite size as it has a direct bearing in the AFM analysis. The mean crystallite size being computed using $D = k\lambda / \beta \cos\theta$ (Debye Scherrer's formula). Here D denotes crystallite size, k denotes Scherrer's constant, $\lambda = 1.54059\text{ \AA}$ (Cu-K α) and β denotes FWHM (full width half maxima) of the diffracted peaks. The computed Crystallite sizes of Pure CuCrO_2 , 5% Mg substituted CuCrO_2 and 5% Sr substituted CuCrO_2 are 55.3 nm, 56.7 nm and 58.7 nm respectively. The slight increase in crystallite sizes shows the substitutional stresses due to Mg and Sr atoms.

Microstructural Investigation:

AFM (Atomic Force Microscopy) is an important surface characterization technique widely used in thin film applications. Even though we haven't fabricated thin films so far, a knowledge of understanding the methods and underlying principles forms a priori. The sample's morphological studies were done by the Bruker Atomic Force Microscopy facility available at UGC-DAE CSR, Indore.

The orthogonal projected 3D images of both magnesium and strontium substituted CuCrO_2 samples are shown here with their axes marked. In the current study, the roughness parameters of 5% Magnesium and 5% strontium substituted CuCrO_2 samples were using AFM technique in contact mode. The in-depth roughness parameters are summarized in Table-1.

The table 1 clarify about the comparative studies of R_{rms} and R_{avg} values, that Magnesium substituted sample have high roughness as compared to that of Strontium substituted sample i.e, large sized domains are formed and simultaneously their height of the peaks are rises. These results are visibly evident in the 3D images of the samples (Figure 1.1 and Figure 1.2).

	
Figure 1.1 : Orthogonal projection of Magnesium substituted CuCrO_2 sample in 3 Dimensions	Figure 1.2: Orthogonal projection of Strontium substituted CuCrO_2 sample in 3 Dimensions

Parameters	5% Mg substituted CuCrO ₂	5% Sr substituted CuCrO ₂
Scanning area in μm^2	10.00 μm^2	10.00 μm^2
Average height in nm	841.3nm	162.8 nm
Average roughness (Ravg) in nm	129 nm	28.4 nm
Root Mean Square roughness (Rrms) in nm	204.9 nm	41.5 nm
Surface Skewness	-0.0791	1.0655
Coefficient of Kurtosis	5.33	6.70

Table 1: Roughness parameters of Mg and Sr substituted CuCrO₂ sample

Rigorous evaluation is needed for the Skewness and Kurtosis moments (which are important parameters for structural study) (Methods adopted in literature for polymer based surfaces by A K Bajpai et al 2016 and the references there in, were followed for this analysis). It is observed that skewness is the third moment of profile amplitude probability density function which determine the profile symmetry about mean line.

The surface skewness of the Strontium substituted sample is much more positive than magnesium substituted sample, suggested that strontium substituted sample have asymmetric nature of surface and peaks are predominate than valleys. As the skewness value of magnesium is negative, valleys are predominated than peaks and the sample became more planners. Which is further confirmed by the line analyses shown in graph Figure 2.

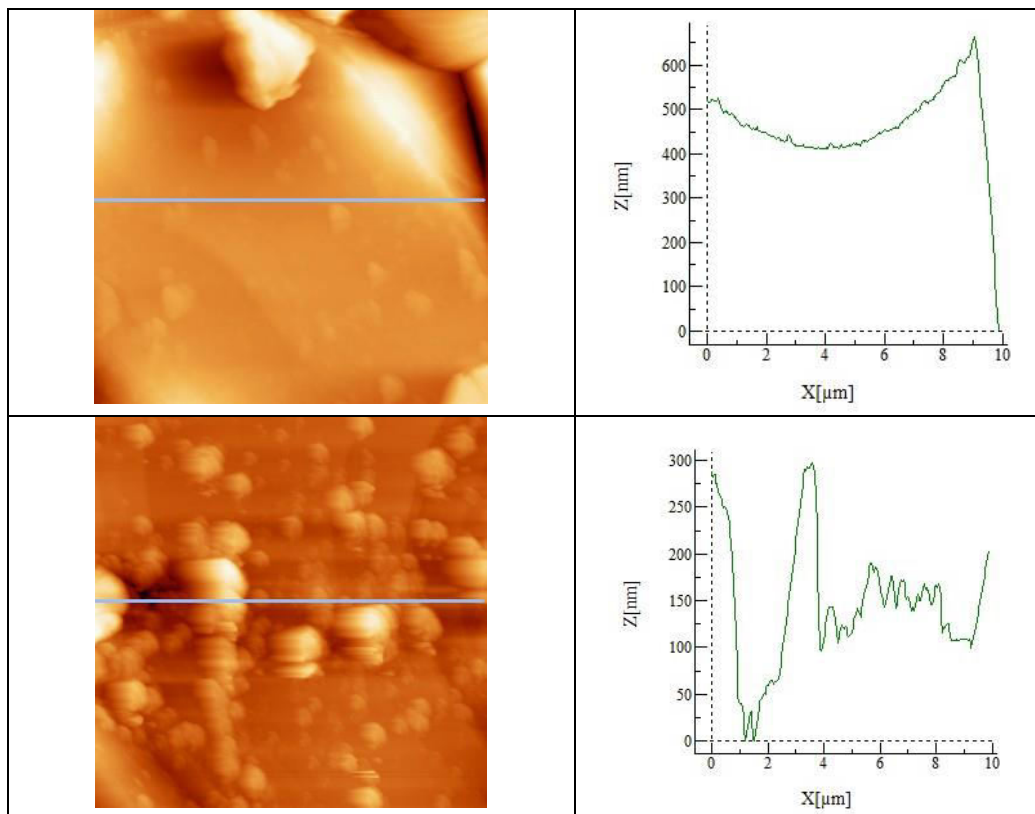


Figure 2: Line profile of Surfaces of Mg substituted CuCrO₂ and Sr substituted CuCrO₂.

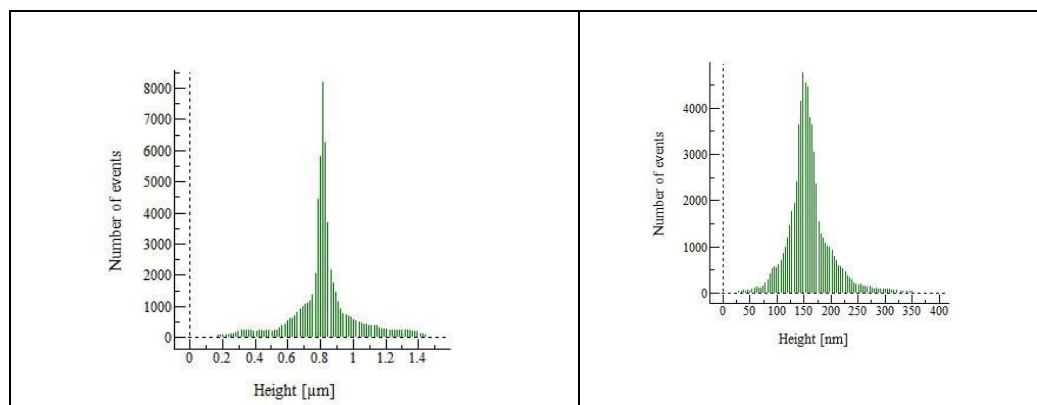


Figure 3: histogram analysis of surfaces of Mg and Sr substituted CuCrO₂ samples.

Similar results were also observed when kurtosis moments are examined alongside both the samples. Greater value of kurtosis moment, than 3 suggests more peaks to that of valleys. For strontium substituted sample the value of kurtosis is far exceeding than 3 which reveal more peak than valleys in 3D image (Figure 1).

The histogram provides an averaging effect as it contain data of entire image. Because of this, the height profile obtained from histogram is highly precise evaluation. The height profile (Figure 3) shows the behavior of the microstructure i.e. whether the surface is planner or has peaks or valleys. Histogram images of the samples support our observed results.

Acknowledgements

Authors would like to thank MP Council of Science and Technology (MPCST) Bhopal for funding the research project to NK vide reference number A/RD/RP-2/313, UGC DAE CSR, Indore and RRCAT for providing experimental facilities. VG would like to thank MPCST, Bhopal for the project A/RD/RP-2/321 and UGC-DAE CSR for the project CSR/2022-23/03/877 for the support. SVVV, Indore and MU Indore are thanked for the encouragement.

References:

1. Kawazoe, Hiroshi; Yasukawa, Masahiro; Hyodo, Hiroyuki; Kurita, Masaaki; Yanagi, Hiroshi; Hosono, Hideo; Nature 6654, 939–942 (1997).
2. Kim, J., Kendall, O., Nguyen, T. T. H., van Embden, J. & Della Gaspera, E. Improved transparency and conductivity in copper chromium oxide coatings through aliovalent doping and stoichiometry control. RSC Applied Interfaces 1, 502–510 (2024).
3. Chen, H. Y., Chang, K. P. & Yang, C. C. Characterization of transparent conductive delafossite-CuCr 1-x O 2 films. Appl Surf Sci 273, 324–329 (2013).
4. Sinnarasa, I. et al. Influence of thickness and microstructure on thermoelectric properties of Mg-substituted CuCrO 2 delafossite thin films deposited by RF-magnetron sputtering. Appl Surf Sci 455, 244–250 (2018).
5. Wang, Y., Gu, Y., Wang, T. & Shi, W. Structural, optical and electrical properties of Mg-substituted CuCrO₂ thin films by sol-gel processing. J Alloys Compd 509, 5897–5902 (2011).
6. Anil Kumar Bajpai, Rinkesh Bhatt, Ravi Katore, Atomic force microscopy enabled roughness analysis of nanostructured poly (diaminonaphthalene) substituted poly (vinyl alcohol) conducting polymer thin films, Micron, 90 (2016) 12–17

48. Synthesis of ZnO/CuS nanocomposite through green route and its Characterization: application for chlorpyrifos degradation

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Abstract: In recent decades, different types of pollutants have been found in oceanic environments, raising severe concerns. In this work, nanocomposites of ZnO/CuS photocatalysts are fabricated through an environmentally friendly green synthesis route. The prepared photocatalysts (PCs) are employed under visible light for the degradation of environmental pollutants. The prepared photocatalysts are characterized through various analytical techniques, including X-ray diffraction (XRD), Field emission scanning electron microscopy (FESEM), energy dispersive X-ray spectroscopy (EDX), Fourier transform infrared spectroscopy (FTIR), and UV-visible spectroscopy. Copper sulfide (CuS) nanoparticles were synthesized with the use of rice starch water and zinc oxide (ZnO) were synthesized with the use of banana peel extract. The photocatalytic degradation results of environmental pollutants mainly chlorpyrifos pesticide, suggest that the ZnO/CuS nanocomposite outperformed the other composites. Superoxide and hydroxyl radicals were the main active species during the degradation process. According to results, the catalytic activity of ZnO/CuS nanocomposite is much better than that of the other composites.

Keywords: CuS and ZnO Nanoparticles, CuS/ZnO Nanocomposite, chlorpyrifos and characterization.

Introduction

Nanocomposites are combination of two or more particles in nanosized form; properties of nanocomposites differ from nanoparticles and provide a better result in the field of nanotechnology compared than nanoparticles. Modern synthesis techniques have come out as necessary resources in instate these objectives. These techniques provide a maximum pliability and manage over the nanoparticles and nanocomposites fabrication process with analogue specifications for distinct uses. A heterogeneity of synthesise offers has been find out, ranging from conventional physical and chemical methods to much neoteric biological and green approaches [1]. One of the most significant areas of nanoparticle research field in biomedical and environmental pollutants applications, where green materials specially based on inorganic green materials plays an important contribution.

Different types of pollutants, such as antibiotics, pesticides and dyes are toxic to human health and have gotten into the environment due to human activity are called environmental pollutants. Human activities pollute water, air, and soil, causing pollutants to enter the body. Pollutants reach the human body through inhalation, oral absorption, and food. Although industrialization leads to the development of the country, it releases large amounts of pollutants into the environment, which harm the health of people exposed to them [2]. Although industrialization leads to the development of the country, it releases large amounts of pollutants into the environment, which harm the health of people exposed to them [3].

Almost the naturalistic resources of drinking water for humans, such as rivers, lakes and tanks are found were contaminated through hazards materials and have provide a long-term negative effect on human health [4].

Removal of pollutants from contaminated water then becomes necessary. Various degradation techniques are available to remove pollutants from contaminated water. The most essential method for degradation of pollutants is photocatalytic degradation process in the presence of ultraviolet-visible light source. In this process, highly reactive species (super radicals) such as hydroxyl radicals ($\text{OH}\cdot$) are produced, which oxidize organic pollutants [5]. Photocatalytic degradation is an economical, environmentally friendly and less time-consuming process. Photocatalysis has emerged as a highly discussed topic in recent years due to the desire to utilize abundant solar energy as a sustainable energy source. [6-13].

In this work, a novel structured nanocomposite ZnO/CuS was prepared with the use of ZnO solution extract and CuS solution extract through green route. ZnO solution extract prepared by banana peel powder and CuS solution extract prepared by rice starch water. Synthesized ZnO/CuS nanocomposite is applicable for various areas, mainly used for wastewater treatment [14-17].

Experimental details

1 Chemicals Used

In the synthesis of ZnO/CuS nanocomposite, we used chemicals such as cuprous chloride and thiourea for CuS nanoparticles, zinc acetate for ZnO nanoparticles, which were purchased from commercial sources, AR grade from Ranbaxy India, and used without further purification. Rice (*Oryza sativa*) for rice starch water was purchased from the local market. Banana was chosen for banana peel powder because it contains a high amount of the desired organic compounds, based on market availability, and distilled water (DW) was used as the synthesis medium.

2 Preparation of Rice Starch Water:

I take 200 g of rice and wash it 3 times with distilled water (DW) to remove all the dust and impurities. After keeping the washed rice in a beaker filled with water for 30 minutes, further cook it in an open vessel containing 600 mL DW at 70 °C temperature. The cooked rice is filtered using filter paper and then placed in a beaker.

3 Preparation of Banana Peel Water:

To get the extract, i wash banana fruits twice with plain water and twice with distilled water, and after drying, cut them into small pieces. Small pieces of banana peels are dried in an oven at 60°C for 10 hours and then ground into fine powder. I mixed 20 g of banana peel powder with 1000 mL of distilled water, stirred it for 3 hours, and then placed it in a water bath at 60°C for 1 hour. Finally, i have filtered the mixture solution through filter paper and stored the filtration extract (banana peel water) in a beaker for later use.

4 Preparation of CuS extract:

For the synthesis of the CuS extract, we first prepared a rice starch solution, The rice starch water was then filtered through filter paper in a glass container/beaker. Second one: 0.1 mol

(1.98 g) of cuprous chloride (CuCl) was mixed with rice starch water under a mechanical stirrer for 30 minutes. The reaction mixture turned green in colour, and 10 to 15 drops of ammonia were added to the CuCl solution. Then, the colour of the solution changed to light blue. The solution was heated to 55°C for 15 minutes, and the thiourea solution was prepared by dissolving 0.1 mol (1.5 g) of thiourea in 200 mL of distilled water. The next step is to mix 70 mL of thiourea solution with 10 mL of CuCl solution, then the colour of the given solutions can be changed from blue to dark brown. The resultant formation of a brown-coloured solution indicates the formation of a CuS solution extract.

5 Preparation of ZnO extract:

For the synthesis of ZnO extract, we first prepared banana peel water. To obtain the extracts, banana fruits were washed twice with distilled water, then dried and peeled as thinly and small sizes as possible. The second one, the ZnO solution extract, was synthesized by mixing 0.1 mol (18.35 g) of zinc acetate with 1000 mL of banana peel extract. These mixtures were then stirred for 60 minutes and subsequently placed in a water bath at 60 °C for 60 minutes. This process resulted in a light yellow ZnO solution extract.

6 Synthesis of ZnO/CuS nanocomposite

Resultant we have synthesized ZnO/CuS nanocomposite in one type concentration sample of ratio is **1:2; ZnO: CuS**. The colour of the given solutions is greenish black.

Sample were 15 minutes stirred in the duration of mixing of solution extract, and after sample preparation, given above sample put for azing to 24 h. After azing duration these sample are centrifuged at 10,000 rpm 3 times and washed with distilled water and filtered through filter paper; the precipitate was dried at 60 °C in an oven overnight. Finally, scratched nanocomposite in powder form.

7 Photo Catalytic Experiment

The photocatalytic performance of the synthesized ZnO/CuS nanocomposite was assessed at room temperature by testing their ability to break down a chlorpyrifos pesticide solution under UV-Visible light irradiation ($\lambda = 200 - 700$ nm). A 20 W LED bulb served as the visible light source, while a 20 W UV rod provided UV illumination. In the standard procedure, specific amount of the photocatalyst 20 mg were added to 100 mL of distilled water containing a 1 mmol chlorpyrifos solution (approximately 0.04 g). At intervals of 10-10 minutes, 3 mL samples were taken, centrifuged, and their absorbance was measured using a UV-Visible spectrophotometer to track the degradation process.

Characterizations

X-ray diffraction (XRD) analysis of the dried ZnO/CuS nanocomposite was performed using a fifth-generation Rigaku Mini Flex 600 diffractometer at Jiwaji University, Gwalior (M.P.), to determine their crystalline structure. The functional groups present in the ZnO/CuS nanocomposite were identified using FTIR, conducted with a PerkinElmer Spectrum Two spectrometer (Lambda 25) in the wavenumber range of 400–4000 cm^{-1} at ITM University, Gwalior. Optical properties were studied using UV-Visible spectroscopy, with transmission spectra recorded in the 200–800 nm range using a PerkinElmer Lambda 25 instrument, also at

ITM University, Gwalior. The morphological information of the synthesized nanocomposites was confirmed by FESEM characterization, performed with a Zeiss Gemini 300 microscope (Carl Zeiss, Germany) at the Indian Institute of Technology (IIT) Roorkee.

Results and Discussion

1 Structural Characterisation (XRD)

XRD measurements were performed to analyze the phase formation and crystalline structures of the prepared ZnO/CuS nanocomposite sample. X-ray studies were conducted, and the X ray diffractogram is presented in Figure 1. The prepared sample ZnO/CuS nanocomposite preferred diffraction peaks of XRD graphs at $2\theta \approx 12.52^\circ$, 15.38° , 26.83° , 29.53° , 45.98° and 57.51° corresponding to lattice planes of (100 for CuS), (101 for CuS), (002 for ZnO), (101 for ZnO), (107 for CuS and 102 for ZnO) and (110 for ZnO) respectively, JCPDS No. 06-0464 for CuS and 96-900-4181 for ZnO, it indicated the hexagonal phase of ZnO/CuS NCs. The Debye -Scherrer formula $D = (0.9 \times \lambda) / (\beta \times \cos \theta)$ is used for the calculation of crystalline size, where λ is the wavelength Cu K α 1.5406 Å, β is the full width at half maxima (rad), and θ is the diffraction angle (degrees). 8.9 nm is the crystalline size of the synthesized ZnO/CuS nanocomposite.

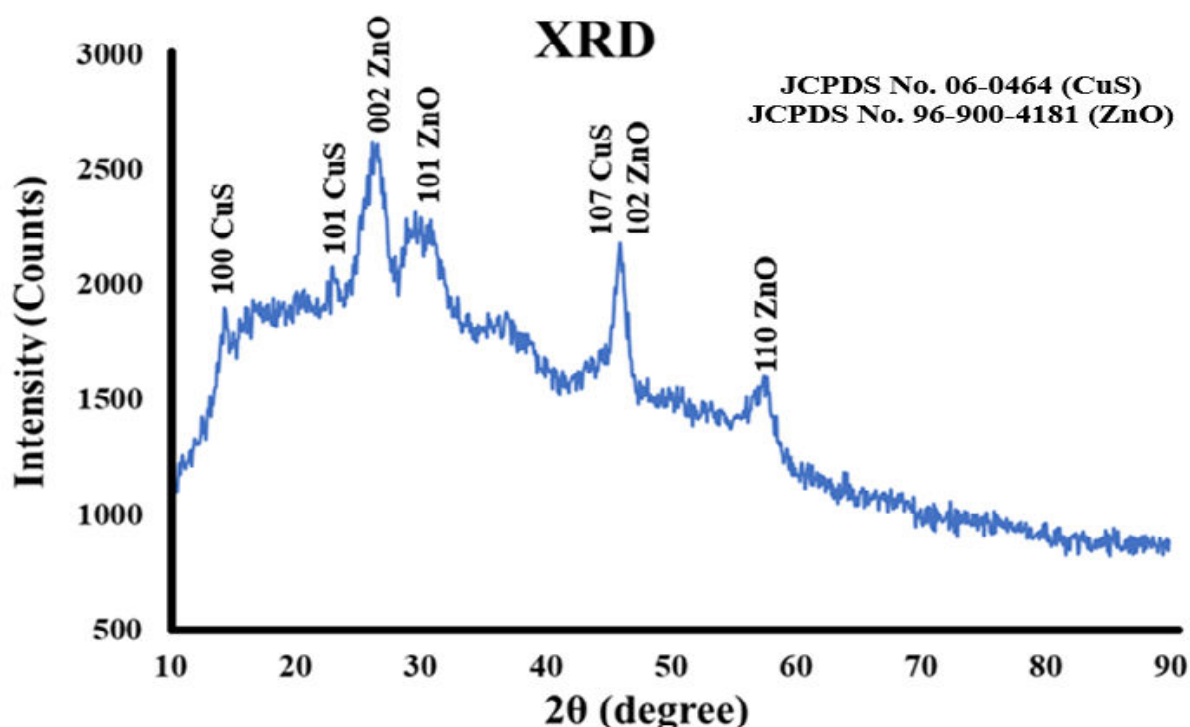


Figure 1: X-ray diffractograms of ZnO/CuS nanocomposite

2 Morphological Studies (FESEM and EDX)

Figure 2 displays the FESEM image of synthesized sample which show that the ZnO/CuS nanocomposite are slightly agglomerated, nanosized, and spherical.

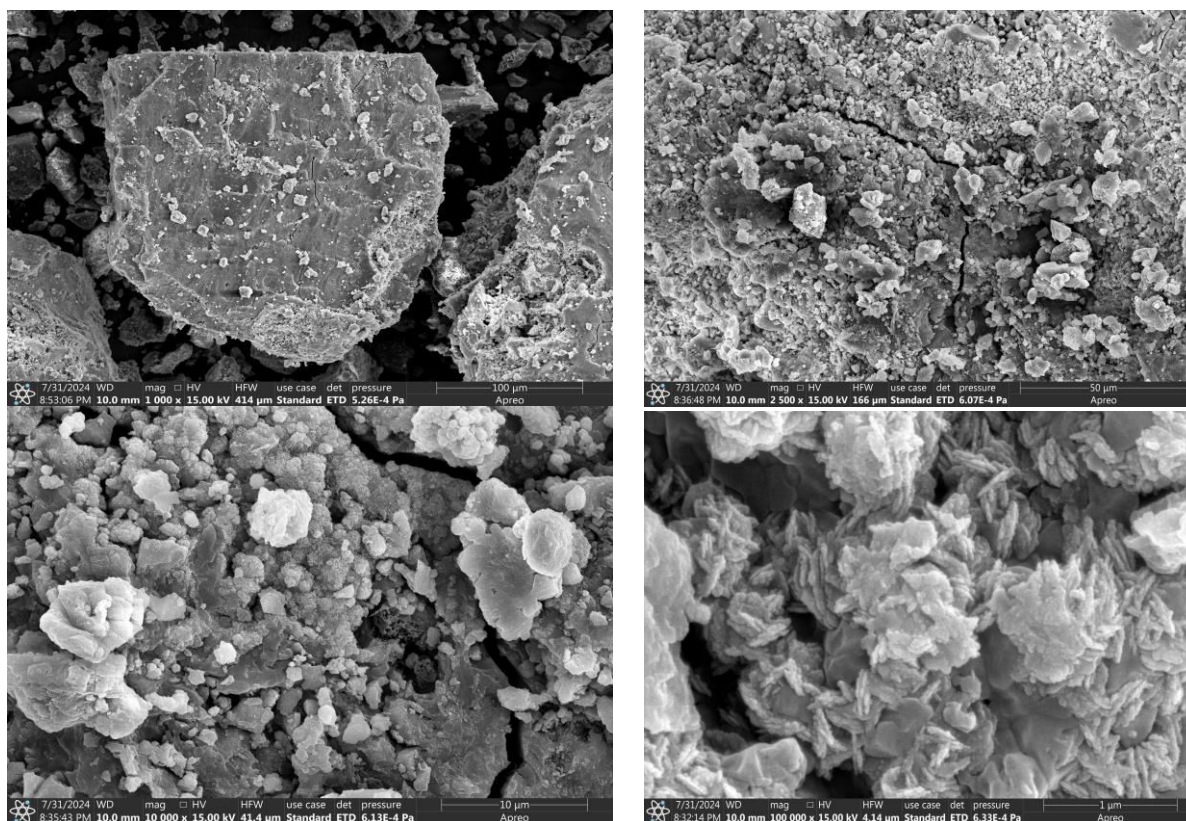


Figure 2: FESEM images of ZnO/CuS nanocomposite

3 Optical Studies

The optical absorbance properties of ZnO/CuS nanocomposite were analyzed using UV–Vis–NIR spectroscopy. From the absorbance spectra, the optical band gap energy was determined using the equation:

$$(\alpha h\nu)^n = A (h\nu - E_g) \dots\dots\dots (1)$$

α denotes the absorption coefficient, E_g denotes the band gap, h denotes Planck's constant, and ν is the radiation's frequency.

Band gap value of synthesized ZnO/CuS nanocomposite sample were determined via $(\alpha h\nu)^2$ versus $h\nu$ plot. The estimated band gap energy is 2.2 eV as shown in Figure 3.

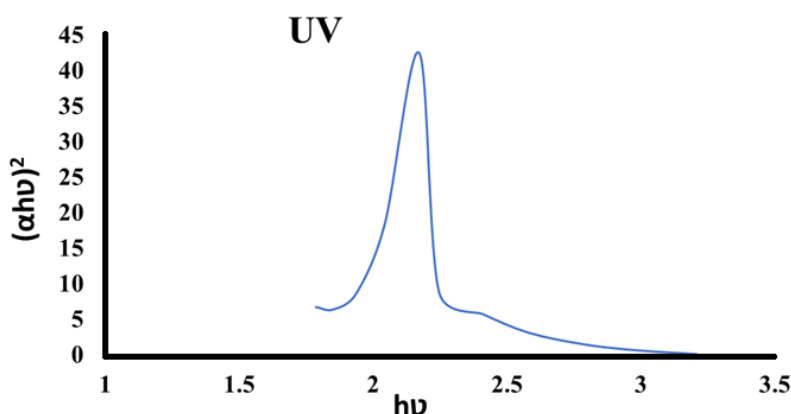


Figure 3: $(\alpha h\nu)^2$ versus $h\nu$ curve of ZnO/CuS nanocomposite

4.4 FTIR Studies

FTIR spectra of the ZnO/CuS nanocomposite sample were recorded using a PerkinElmer Lambda 25 FTIR spectrometer with the KBr pellet technique, covering a wavenumber range of 400-4000 cm^{-1} to identify variations in functional groups. The presence of specific chemical bonds in the ZnO/CuS nanocomposites was confirmed through FTIR analysis, with the corresponding functional groups summarized in Table 1 and the characteristic absorption peaks illustrated in Figure 4.

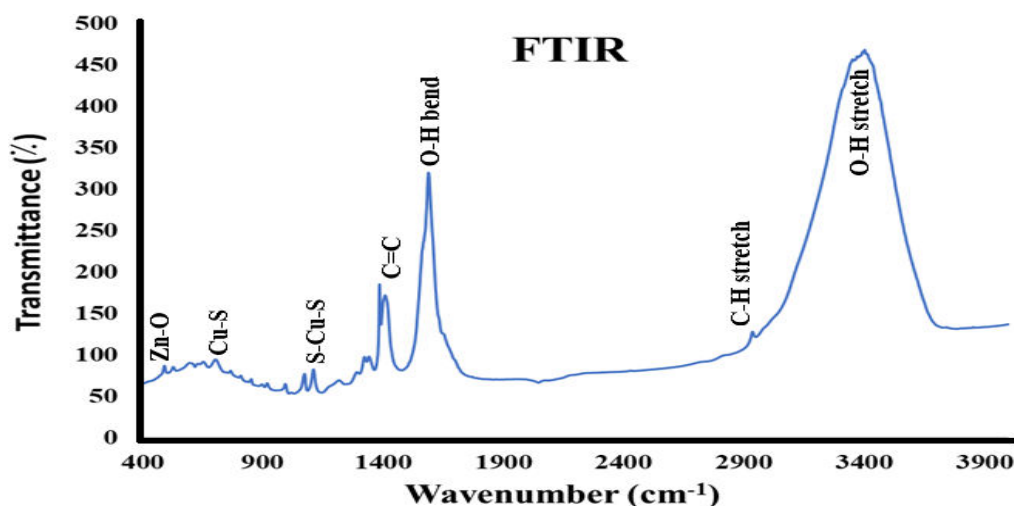


Figure 4: FTIR image of ZnO/CuS nanocomposite

Table 1: FTIR attached group of ZnO/CuS NCs

Peak Position (cm^{-1})	Group	Reference No.
3398 or 3078-3694	O-H stretch	13
2940	C-H stretch	13
1591	O-H bend	13
1407	C=C	18
1114	S-Cu-S	13
715	Cu-S	13
494	Zn-O	13

5 Photo Catalytic Study

Chlorpyrifos was used to assess the photocatalytic activity of the synthesized ZnO/CuS nanocomposite under UV-Visible light irradiation. Generally, the photocatalytic efficiency of a material relies on two main factors: (i) the effective separation of photoexcited electron-hole pairs and (ii) the production of reactive oxygen species (ROS) [19, 20]. The initial peak absorbance of the chlorpyrifos at around 290 nm. Under UV-visible light, ZnO alone is less active due to its wide bandgap, whereas CuS, with a narrower bandgap, is photoactive under the same conditions. To improve performance, a ZnO/CuS heterostructure is created, which promotes the transfer of photogenerated electrons from the conduction band of CuS to that of ZnO. This electron transfer leaves holes in the valence band of CuS, thereby reducing electron-hole recombination and increasing overall photocatalytic efficiency. The degradation efficiency of chlorpyrifos was calculated using the following equation:

$$D(\%) = \frac{(C_0 - C)}{C_0} \times 100\%$$

where C_0 is the absorption of chlorpyrifos after the adsorption equilibrium is achieved before UV-visible-light irradiation and C is the absorption of chlorpyrifos in the time interval under UV-visible light irradiation.

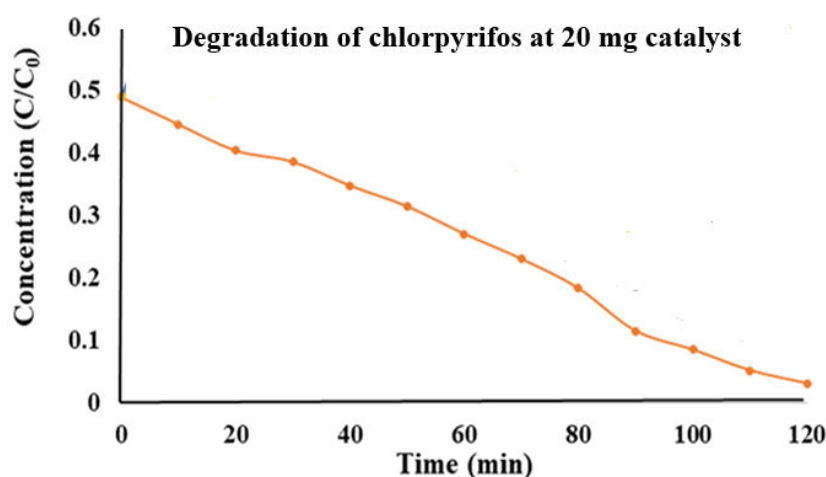


Figure 5: Photocatalytic degradation of chlorpyrifos of ZnO/CuS nanocomposite at 20 mg catalyst.

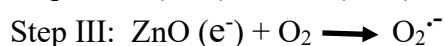
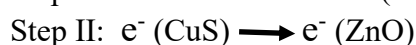
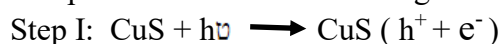
Figure 5 shows the photocatalytic efficiencies of the ZnO/CuS nanocomposite. The analysis of photocatalysis involved the dosage of ZnO/CuS nanocomposite is 20 mg. Under UV-visible light irradiation, a similar degradation procedure was performed. The removal and decomposition efficiencies of photocatalysts at weight for 20 mg a maximum decomposition 93.65% was obtained within 120 minutes.

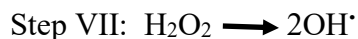
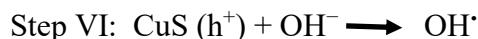
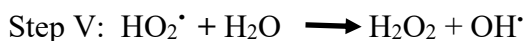
6 Mechanism of Photodegradation in ZnO/CuS NCs

In this study, photocatalytic decomposition of chlorpyrifos using ZnO/CuS nanocomposite was proposed and successfully demonstrated. The nanocomposite was engineered as a heterostructure, combining the wide bandgap of ZnO with the narrow bandgap of CuS to optimize light absorption and charge separation. CuS nanoparticles were preferentially deposited onto the surface of ZnO, facilitating strong interfacial contact and efficient charge transfer between the two materials. This heterojunction structure enhances the separation of photoexcited electron-hole pairs, which are a key factor in improving photocatalytic performance [21].

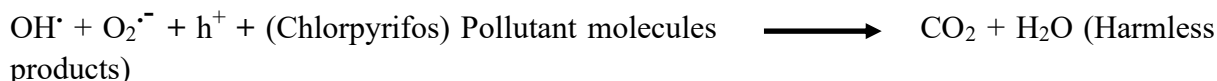
Under visible light irradiation, both ZnO and CuS participate in the photocatalytic process. These reactive oxygen species— $O_2^{\bullet-}$ and $\bullet OH$ —play a crucial role in breaking down chlorpyrifos and other organic pollutants into non-toxic, inorganic end products such as water (H_2O) and carbon dioxide (CO_2).

The possible reaction for the degradation of chlorpyrifos can be described as:





These photogenerated radicals are highly reactive species in the degradation process, breaking down the chlorpyrifos pesticide [22-24] into intermediate products, which eventually decompose into CO_2 and H_2O .



Conclusion

In this study, ZnO/CuS nanocomposite were synthesized through simple precipitation method based on a green synthesis approach, employing natural extracts such as rice starch water and banana peel extract. The green synthesis route emphasizes eco-friendly practices, characterized by zero contamination, low toxicity, simple procedures, high stability, and cost-effectiveness in the production of nanoparticles and nanocomposites. Over the past two decades, green synthesis has emerged as a highly attractive area in nanomaterials research. Overall, this strategy demonstrates strong potential for developing efficient photocatalysts for use in water decontamination, air handling, and energy renovation.

References

1. A. Vinukonda, N. Bolledla, R.K. Jadi, R. Chinthala and V.R. Devadasu. Synthesis of nanoparticles using advanced techniques. *Next Nanotechnology* 8 (2025) 100169. [10.1016/j.nxnano.2025.100169](https://doi.org/10.1016/j.nxnano.2025.100169)
2. S. S. Shetty, D. Deepthi, S. Harshitha, S. Sonkusare, P. B. Naik, Kumari N. Suchetha, H. Madhyastha. Environmental pollutants and their effects on human health. *PubMed*. 9 (2023) 19496. [10.1016/j.heliyon.2023.e19496](https://doi.org/10.1016/j.heliyon.2023.e19496).
3. I. Manisalidis, E. Stavropoulou, A. Stavropoulos, E. Bezirtzoglou. *Environmental and health impacts of air pollution: a review. Frontiers in Public Health. Frontiers Media S.A.* 8 (2020). <https://doi.org/10.3389/fpubh.2020.00014>
4. E.A. Songa and J.O. Okonkwo. Recent approaches to improving selectivity and sensitivity of enzyme-based biosensors for organophosphorus pesticides: a review. *Talanta*. 155 (2016). 289-304. [10.1016/j.talanta.2016.04.046](https://doi.org/10.1016/j.talanta.2016.04.046).
5. F. Hu, W. Luo, Y. Hu, H. Dai and X. Peng. Insight into the kinetics and mechanism of visible-light photocatalytic degradation of dyes onto the P doped mesoporous graphitic carbon nitride. *J. Alloys. Compd.* 794 (2019). 594-605. [10.1016/j.jallcom.2019.04.235](https://doi.org/10.1016/j.jallcom.2019.04.235)
6. P. Zhou, J. Yu and M. Jaroniec. All-Solid-State Z-Scheme Photocatalytic Systems. *Adv. Mater.* 26(29) (2014).4920–4935. [10.1002/adma.201400288](https://doi.org/10.1002/adma.201400288).
7. J. Liu, W. Fang, Z. Wei, Z. Qin, Z. Jiang and W. Shangguan. Efficient Photocatalytic Hydrogen Evolution on N-Deficient g-C₃N₄ Achieved by a Molten Salt Post-Treatment Approach. *Appl.Catal.*238(2018).465–470. <https://doi.org/10.1016/j.apcatb.2018.07.021>.
8. W. Fang, Z. Qin, J. Liu, Z. Wei, Z. Jiang and W. Shangguan. Photo-Switchable Pure Water Splitting under Visible Light over Nano-Pt@P25 by Recycling Scattered Photons. *Appl.Catal.* 236(2018).140–146. <https://doi.org/10.1016/j.apcatb.2018.05.023>.
9. J. Liu, W. Fang, Z. Wei, Z. Qin, Z. Jiang and W. Shangguan. Metallic 1T-Li x MoS₂ Co-Catalyst Enhanced Photocatalytic Hydrogen Evolution over ZnIn₂S₄ Fluorinated Microspheres under Visible Light Irradiation. *Catal. Sci. Technol.* 8 (2018). 1375–1382. <https://doi.org/10.1039/C7CY02456H>

10. Z. Qin, W. Fang, J. Liu, Z. Wei, Z. Jiang and W. Shangguan. Zinc-Doped g-C₃N₄/BiVO₄ as a Z-Scheme Photocatalyst System for Water Splitting under Visible Light. *Chin. J. Catal.* 39(3) (2018). 472–478. [10.1016/S1872-2067\(17\)62961-9](https://doi.org/10.1016/S1872-2067(17)62961-9)
11. W. Fang, J. Liu, L. Yu, Z. Jiang and W. Shangguan. Novel (Na, O) Co-Doped g-C₃N₄ with Simultaneously Enhanced Absorption and Narrowed Bandgap for Highly Efficient Hydrogen Evolution, *Appl. Catal.* 209 (2017). 631–636.
12. Z. Wei, R. Li and R. Wang. Enhanced Visible Light Photocatalytic Activity of BiOBr by in Situ Reactable Ionic Liquid Modification for Pollutant Degradation. *RSC Adv.* 15 (2018). 7899–8449.
13. Z. D. Wei and R. Wang. Hierarchical BiOBr Microspheres with Oxygen Vacancies Synthesized via Reactable Ionic Liquids for Dyes Removal. *Chin. Chem. Lett.* 27(5) (2016). 769–772. [10.1016/j.ccllet.2016.03.013](https://doi.org/10.1016/j.ccllet.2016.03.013).
14. S.S. Imam, R. Adnan and N.H. Mohd Kaus. Photocatalytic degradation of ciprofloxacin in aqueous media: a short review. *Toxicol. Environ. Chem.* 100(5) (2018) 518–539. [10.1080/02772248.2018.1545128](https://doi.org/10.1080/02772248.2018.1545128).
15. T. Hayri-Senel, E. Kahraman, S. Sezer, N. Erdol-Aydin and G. Nasun-Saygili. Photocatalytic degradation of ciprofloxacin from water with waste polystyrene and TiO composites. *ScienceDirect*. 10(2024). 25433. <https://doi.org/10.1016/j.heliyon.2024.e25433>.
16. X. Hu, Q. Peng, L. Zhou, X. Tan, L. Jiang, C. Tang, H. Wang, S. Liu, Y. Wang and Z. Wang (2020). Mechanisms underlying the photocatalytic degradation pathway of ciprofloxacin with heterogeneous TiO₂. *Chem. Eng. J.*, 380 (2020). 122366. <https://doi.org/10.1016/j.cej.2019.122366>
17. S. Singh, H. Mahalingam and P.K. Singh. Polymer-supported titanium dioxide photocatalysts for environmental remediation: a review. *Applied Catalysis A: General.* (2013). 178–195. [10.1016/j.apcata.2013.04.039](https://doi.org/10.1016/j.apcata.2013.04.039).
18. S. Singh, H. Mahalingam and P.K. Singh. Polymer-supported titanium dioxide photocatalysts for environmental remediation: A Review, *Applied Catalysis. Gen.* (2013). 462–463, 178–195. <https://doi.org/10.1016/j.apcata.2013.04.039>
19. P. Das, K. Tantubay, R. Ghosh, S. Dam and M. Baskey. Transformation of CuS/ZnS nanomaterials to an efficient visible light photocatalyst by ‘photosensitizer’ graphene and the potential antimicrobial activities of the nanocomposites. *Environ. Sci. Pollut. Res. Int.* 28(35) (2021). 49125–49138. [10.1007/s11356-021-14068-1](https://doi.org/10.1007/s11356-021-14068-1).
20. H. Y. Ma, L. Zhao, L. H. Guo, H. Zhang, F. J. Chen and W. C. Yu. Roles of reactive oxygen species (ROS) in the photocatalytic degradation of pentachlorophenol and its main toxic intermediates by TiO₂/UV. *J. Hazard. Mater.* 369 (2019) 719–726. [10.1016/j.jhazmat.2019.02.080](https://doi.org/10.1016/j.jhazmat.2019.02.080).
21. S. K. Cushing, J. Li, F. Meng, T. R. Senty, S. Suri, M. Zhi, M. Li, A. D. Bristow and N. Wu. Photocatalytic Activity Enhanced by Plasmonic Resonant Energy Transfer from Metal to Semiconductor. *J. Am. Chem. Soc.* 2012, 134(36) (2012). 15033–15041. [10.1021/ja305603t](https://doi.org/10.1021/ja305603t).
22. M. E. Ali, A.A. Alanezi, F. A. Azeez and M. Y. Ghaly. Photoassisted mineralization of remazole red F3B over NiO/TiO₂ and CdO/TiO₂ nanoparticles under simulated sunlight, *Sep. Sci. Technol.* 53 (2018). 170–180. [10.1080/01496395.2017.1379536](https://doi.org/10.1080/01496395.2017.1379536).
23. R. Mohammed, M. E. M. Ali, S. M. Abdel-Moniem and H. S. Ibrahim. Reusable and highly stable MoS₂ nanosheets for photocatalytic, sonocatalytic and thermocatalytic degradation of organic dyes: Comparative study, *Nano-Struct. Nano-Objects* 31 (2022). 100900. [10.1016/j.nanoso.2022.100900](https://doi.org/10.1016/j.nanoso.2022.100900).
24. M. E. M. Ali, R. Mohammed, S. M. Abdel-Moniem, M. A. El-Liethy and H. S. Ibrahim. Green MoS₂ nanosheets as a promising material for decontamination of hexavalent chromium, pharmaceuticals, and microbial pathogen disinfection: spectroscopic study, *J. Nanopart Res.*, 24(191) (2022). 1–23. <https://doi.org/10.1007/s11051-022-05573-6>.

49. Impact of Pre-sowing Treatment of Seeds with Plasma on Germination and Growth of Wheat

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Abstract: The pre-treatment of wheat seeds with glow discharge plasma improved plant height, root length, and fresh weight at all growth stages. Wheat seeds treated with O₂ and N₂ plasma showed improvements in growth and biomass. This study looked at how pusa tejas wheat seed germination and growth were affected by cold oxygen and nitrogen plasma treatment. We found that the treatment with 80 W could greatly boost seed germination. Produced in a cylindrical stainless steel vacuum chamber using an RF power supply with a 13.56 MHz, 600W RF electric gas discharge system with an automated matching network. A rotary pump in the reactor creates a vacuum up to 10⁻³ mbar.

Keywords: O₂, N₂, glow discharge plasma, wheat, growth, yield.

Introduction

The creation of Radio frequency plasma and researching how it interacts with seeds has become essential for sustainable agriculture in terms of food quantity and quality in recent years. Due to its potential to improve seedling development and germination rates in a variety of crops, the non-equilibrium atmospheric pressure plasma created using various configurations has attracted a lot of interest in agriculture (Lotfy et al., 2019; Adetimirin, 2008). In agricultural production, seed quality is one of the main factors that determines crop yield. High vigor seeds often germinate more quickly and develop into robust, well-proportioned seedlings. (Ashrafi & Razmjoo, 2015; Miano et al., 2015). This has a major effect on agricultural productivity. Numerous methods, including as chemical treatment, ultrasonic scratching, electric field treatment, magnetic treatment, and ion beam scratching, have been used to increase seed vigour (Xu et al., 2012). Affecting biochemical processes like protein and enzyme activity, physical treatment may increase the vigour of seeds (Moon & Chung, 2000). However, some physical treatments may potentially damage seed cells, resulting in uneven treatment and seed loss. (Goussous et al., 2010; Shi et al., 2014). Applying atmospheric pressure plasma to different seeds has been shown in earlier research to enhance germination and seedling development (Liu et al., 2018; Ling et al., 2014) by using plasma to accelerate and stimulate biological processes (Adhikari et al., 2020).

Materials and Methods

Seed Material

Seeds of wheat Pusa Tejas variety were obtained from seed agency, Indore. Cold oxygen and nitrogen plasma treatment of wheat seeds was performed under different energy and time.



Fig. 1: Pusa tejas wheat seeds

Plasma Reactor

The gaseous plasma treatment experimental setup shows in Fig. 2. Radio Frequency power source with 13.56 MH, 600 Watt connected with an automated matching network, A 360 mm in diameter and 300 mm height. Glow discharge vacuum chamber was pumped with a rotary pump. Oxygen and nitrogen gas were inserted through mass flow controllers. The vacuum pressure was measured with digital pirani pressure gauge.

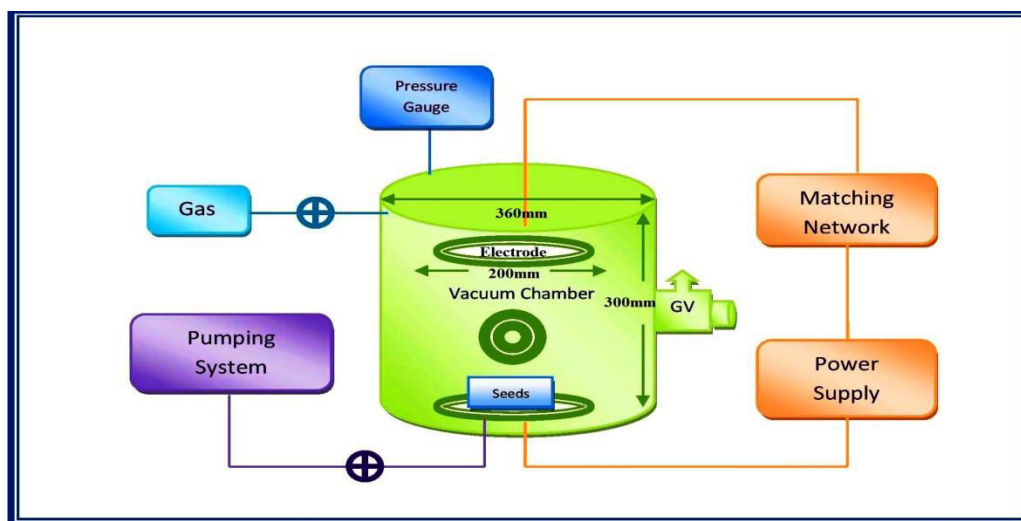


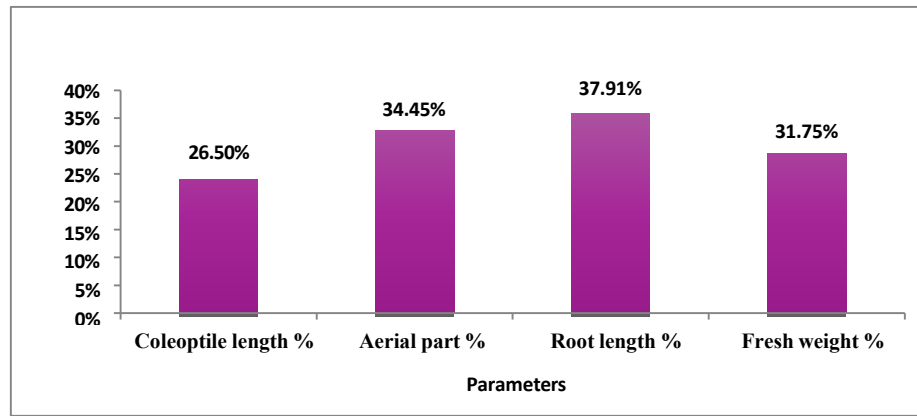
Fig. 2: Block Diagram of Plasma Setup

Plasma Treatment

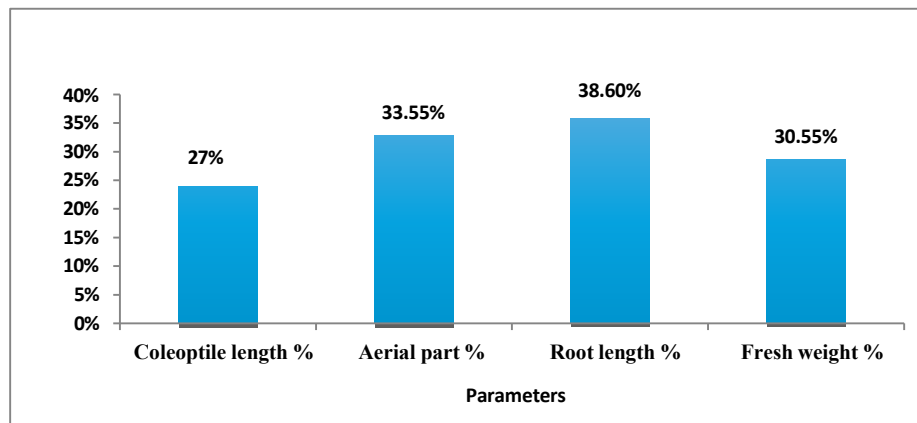
Fresh wheat seedlings were treated simultaneously with the same energy and oxygen and nitrogen plasma treatments. The wheat seeds were placed on a seeds stand that was placed in the middle of the electrode. The radio frequency plasma was distributed very uniformly across the discharge vacuum chamber. Seeds were exposed to nitrogen and oxygen plasma. Pusa tejas wheat seeds are treated with plasma at an energy of 80 watts for 30, 60 and 90 seconds.



Fig. 3: Control and 80 w oxygen and nitrogen Plasma Treated Wheat Seeds in Petri-dish for 30, 60, and 90 seconds



(a)



(b)

Fig 4: Shows Different Parameters in Percentage (a) Oxygen and (b) Nitrogen

Seed Germination

Wheat seedlings were treated with oxygen and nitrogen plasma at different energy and times. Following the first germination and development of wheat seeds in a controlled environment, data was gathered. The fresh weight and length of the seedling were measured. Similar laboratory conditions were used to administer the treated and controls seeds, and room temperature was maintained. Thirty seeds were left untreated in a Petri dish with three sets.

Results and Discussion

Wheat seeds were treated with low-temperature radiofrequency plasma. The effects of the plasma treatment on growth characteristics were examined such coleoptile length, aerial part, root length, and fresh weight in percentages of 26.50%, 34.45%, 37.91%, and 21.75% for oxygen plasma and 27%, 33.55%, 38.60%, and 30.55% for nitrogen plasma. We used 80 watts for 30, 60, and 90 seconds, but the greatest results came from using 80 watts for 60 seconds.

Conclusions

The current study examined the effects of plasma treatment on wheat seedlings in a surface discharge at standard pressure and temperature. The roots and sprouts of the

samples treated with oxygen and nitrogen plasma were found to be bigger and heavier than those of the untreated seeds for all treatment durations investigated. The most length growth was seen in seeds treated with plasma for 60 seconds. It has been determined that the plasma treatment causes the seeds to grow longer.

References

- Adetimirin, V. O. (2008). Stand establishment and early field vigour variation in a tropicalised shrunken-2 maize population. *Field Crops Research*, 108(2), 143–149. <https://doi.org/10.1016/j.fcr.2008.04.007>.
- Adhikari, B., Adhikari, M., Ghimire, B., Adhikari, B. C., Park, G., & Choi, E. H. (2020). Cold plasma seed priming modulates growth, redox homeostasis and stress response by inducing reactive species in tomato (*Solanum lycopersicum*). *Free Radical Biology and Medicine*, 156, 57–69. <https://doi.org/10.1016/j.freeradbiomed.2020.06.003>.
- Ashrafi, E., & Razmjoo, J. (2015). Seed treatment to overcome salt and drought stresses during germination in safflower (*Carthamus tinctorius*L.). *Journal of Plant Nutrition*, 38(14), 2151–2158. <https://doi.org/10.1080/01904167.2015.1069331>.
- Goussous, S. J., Samarah, N. H., Alqudah, A. M., & Othman, M. O. (2010). ENHANCING SEED GERMINATION OF FOUR CROP SPECIES USING AN ULTRASONIC TECHNIQUE. *Experimental Agriculture*, 46(2), 231–242. <https://doi.org/10.1017/s0014479709991062>.
- Ling, L., Jiafeng, J., Jiangang, L., Minchong, S., Xin, H., Hanliang, S., & Yuanhua, D. (2014). Effects of cold plasma treatment on seed germination and seedling growth of soybean. *Scientific Reports*, 4(1), 5859. <https://doi.org/10.1038/srep05859>.
- Liu, B., Honnorat, B., Yang, H., Arancibia, J., Rajjou, L., & Rousseau, A. (2018). Non-thermal DBD plasma array on seed germination of different plant species. *Journal of Physics D Applied Physics*, 52(2), 025401. <https://doi.org/10.1088/1361-6463/aae771>.
- Lotfy, K., Al-Harbi, N. A., & El-Raheem, H. A. (2019). Cold atmospheric pressure nitrogen plasma jet for enhancement germination of wheat seeds. *Plasma Chemistry and Plasma Processing*, 39(4), 897–912. <https://doi.org/10.1007/s11090-019-09969-6>.
- Miano, A., Forti, V., Abud, H., Gomes-Junior, F., Cicero, S., & Augusto, P. (2015). Effect of ultrasound technology on barley seed germination and vigour. *Seed Science and Technology*, 43(2), 297–302. <https://doi.org/10.15258/sst.2015.43.2.10>.
- Moon, J., & Chung, H. (2000). Acceleration of germination of tomato seed by applying AC electric and magnetic fields. *Journal of Electrostatics*, 48(2), 103–114. [https://doi.org/10.1016/s0304-3886\(99\)00054-6](https://doi.org/10.1016/s0304-3886(99)00054-6).
- Shi, M. F., Fan, J. J., Li, S. J., Yu, X. L., & Liang, X. M. (2014). The Influence of High Voltage Electric Field for Barley Seed Germination and its Mechanism. *Applied Mechanics and Materials*, 675–677, 1142–1145. <https://doi.org/10.4028/www.scientific.net/amm.675-677.1142>.
- Xu, G., Wang, X., Gan, C., Fang, Y., & Zhang, M. (2012). Biological effects of low energy nitrogen ion implantation on *Jatropha curcas* L. seed germination. *Nuclear Instruments and Methods in Physics Research Section B Beam Interactions with Materials and Atoms*, 287, 76–84. <https://doi.org/10.1016/j.nimb.2012.05.038>.

50. Sex Determination from Fingerprint Ridge density in Ujjain Population

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Abstract: The aim of the present study is to find out sex difference based on fingerprint ridge density and its possible application in determination of sex in forensic science. Fingerprint samples for this study were taken from healthy individuals from western part of Madhya Pradesh (Ujjain). For collection of finger prints palmar aspects of finger of the subjects were placed on the fingerprint pad and then transferred to the fingerprint card. Fingerprints of both hands were taken. For the analysis 57 males and 51 females samples were considered. An average of ridge density was computed from both the diagonals (radial and ulnar) to represent the final ridge density. Result indicated that female generally exhibit higher mean ridge density (15.32 ± 1.46) compared to males (14.97 ± 2.12). The distribution of ridge densities in females is more compact and consistent, with the range of 12 to 18 compare to the broader range of 11 to 19 in males. This finding significantly aids forensic investigations by providing a reliable method for sex determination based on fingerprint analysis.

Keywords: Fingerprint, Ridge density, Males, Females, Sex Determination

Introduction

Identification of individuals plays a crucial role in forensic investigations, particularly in criminal case resolution and legal adjudication. Among the various biometric modalities available, fingerprint analysis remains one of the most extensively employed techniques owing to the individuality, permanence, and reliability of fingerprint patterns. The uniqueness of fingerprints and their resistance to postnatal alteration have established them as a dependable tool for personal authentication across forensic and civilian applications (Nithin , Balaraj , Manjunatha , & Mestri , 2009; Wadhwa, 2013). Notably, fingerprints are known to differ even between monozygotic twins, despite their identical genetic makeup, emphasizing their value as a biometric marker (Nithin , Balaraj , Manjunatha , & Mestri , 2009).

In addition to personal identification, advancements in forensic dermatoglyphics have expanded the application of fingerprints to demographic profiling, including sex determination. Since the mid-1970s, researchers have explored the forensic utility of fingerprint characteristics beyond pattern classification, particularly ridge and furrow attributes (Reddy , 1975). Quantitative parameters such as ridge density, ridge thickness, and ridge count have been investigated in diverse populations to evaluate sexual dimorphism in fingerprints (Acree, 1999; Gungadin, 2007; Nayak , et al., 2010; Gutiérrez , Alonso , & Dipierri, Sex differences in fingerprint ridge density in the Mataco-Mataguayo population, 2011; Kocharyan , Khachatryan, & Sarukhanyan , 2013; Ceyhan, Sagiroglu, & Akyil, 2014).

Although fingerprint ridge density has demonstrated potential as an indicator of sex, comparatively limited studies have focused on its application for sex determination. Previous research suggests that females generally exhibit higher ridge density values than males; however, these findings exhibit population-specific variation. The present study aims to examine the association between sex and fingerprint ridge density and to document ridge density distribution within the Ujjain population of western Madhya Pradesh, a region for which no prior ridge density-based forensic study has been reported.

Materials and Methods

For the present study fingerprint samples from healthy individuals from Ujjain the western part of Madhya Pradesh from different sections of the society such as professionals, students from colleges, domestic workers were collected. The participants included for the study were informed about the purpose and procedure of the study and their consent was obtained. The participants were instructed to wash their hands with soap and water to avoid dirt and debris. For collecting prints preformed 10 fingerprint cards were used. For obtaining fingerprints palmar aspects of fingers of the subject were placed on the fingerprint pad and then transferred to the prepared fingerprint card. by this way entire prints of 10 fingers were prepared. Only plain prints were taken. The participants were then asked to wipe their fingers. For the final analysis 57 males and 51 females sample were considered between the age range of 16 to 60 years.

Generally, fingerprint ridge density is acquired from a number of ridges over an area of $5 \times 5 \text{ mm}^2$ based on the method of Acree (Acree, 1999). In this study slight modification in counting portion was made as described by (Gutiérrez , Alonso , & Dipierri, Sex differences in fingerprint ridge density in the Mataco-Mataguay population, 2011) . Two squares of $5 \times 5 \text{ mm}^2$ each were constructed and were placed above the upper portion of radial and ulnar border of the fingerprint. Collected fingerprint were scanned with 600 dpi resolution to PDF format. The fingerprint image was then processed to remove any skewness or inclination of the card. Once the card was aligned, the image was then cropped to include only 10 boxes corresponding to the 10 fingerprints. Using python's image processing, each of the 10 boxes were separated into separate images.

After the separation, each print image was converted to grayscale using Python CV2 library. The gray scale was then normalized to improve the contrast of the image. This process makes the ink color independent of the pressure applied during impression.



Figure 1. Before pre-processing.



Figure 2. After grayscale pre-processing.



Figure 3. After binarization pre-processing with 25mm² square area at upper portion of radial and ulnar border of the fingerprint image.

The core of the print was then computed using the dark pixel values of the image. Right above the core, two squares of side 5 mm were drawn on the upper ulnar and upper radial side of the print. The fingerprint card size was used to get the correct square dimensions. The diagonals were also drawn. Ridges along the diagonal were then counted based on the color transition from white to black and vice versa. An average of ridge density was computed from both the diagonals to represent the final ridge density.

Specific comparison of means were made and calculations were done using Python Programming Language. The likelihood ratio (LR) based on Baye's theorem was calculated to obtain the probability inferences of the gender. The LR was calculated by the formula given below:

$$LR = \frac{\text{Probability of the observed RD given that the fingerprint image come from male (A)}}{\text{Probability of the observed RD given that the fingerprint image come from female (B)}}$$

Results

The mean values of the number of fingerprint ridges in 25 mm² of the right and left hands in males were 14.74 ± 2.12 and 15.20 ± 2.27 respectively. The mean value of ridges in the left

hand was a little higher than in the right hand. The mean value of the number of fingerprint ridges of both hands in males was 14.97 ± 2.12 (Table 1). The mean values of the number of fingerprint ridges in 25 mm^2 of the right and left hands in females were 15.23 ± 1.51 and 15.42 ± 1.71 respectively. The mean value of the number of fingerprint ridges in 25 mm^2 of both hands in females was 15.32 ± 1.46 (Table 2). The data indicated that the mean value of ridge density in the five fingers was slightly more for female group compared to male group. Table (3) summarized the frequency distribution of the mean fingerprint ridge densities among males and females. It was found that only 3.5 % of males have a mean ridge density of 17. In contrast, 15.75% of females have a mean ridge density of 17.

The median of frequency distribution of males is on the lower ridge density side compared to females. Female ridge density distribution was more compact with minimum at 12 and maximum at 18. Whereas male ridge density range was broader with minimum at 11 and maximum at 19.

Table 1 : The number of fingerprint ridges in 25 mm^2 in the right and left hands of males (n=57)

Hand	Finger	Minimum	Maximum	Mean $\pm$ SD
Right	Little	8	20	14.30 ± 2.88
Right	Ring	11	23	15.19 ± 2.42
Right	Middle	9	22	15.54 ± 2.85
Right	Index	9	20	14.93 ± 2.40
Right	Thumb	8	22	13.75 ± 2.87
Left	Little	8	21	14.61 ± 2.85
Left	Ring	9	21	15.33 ± 2.67
Left	Middle	10	22	15.88 ± 2.73
Left	Index	8	22	15.65 ± 3.09
Left	Thumb	10	24	14.51 ± 2.92
Right	All fingers	10.4	18.6	14.74 ± 2.12
Left	All fingers	11	19.4	15.20 ± 2.27
Both Hands	All fingers	10.8	18.9	14.97 ± 2.12

Table 2 : The number of fingerprint ridges in 25 mm^2 in the right and left hands of females (n=51)

Hand	Finger	Minimum	Maximum	Mean $\pm$ SD
Right	Little	8	20	13.98 ± 2.28
Right	Ring	10	20	15.49 ± 2.12
Right	Middle	11	21	15.86 ± 2.27
Right	Index	11	23	15.24 ± 2.06
Right	Thumb	10	21	15.57 ± 2.59
Left	Little	10	19	14.41 ± 2.44
Left	Ring	10	21	15.37 ± 2.65
Left	Middle	10	21	15.80 ± 2.37
Left	Index	11	19	15.57 ± 1.99
Left	Thumb	10	22	15.94 ± 2.69
Right	All fingers	12.4	18.6	15.23 ± 1.51
Left	All fingers	11.4	18.6	15.42 ± 1.71
Both Hands	All fingers	12.3	17.7	15.32 ± 1.46

Table 3 : Frequency distribution of the mean fingerprint ridge densities among males and females

Mean Ridges	Gender				Total
	Males		Females		
	No	%	No	%	
11	4	7.0	0	0.0	4
12	4	7.0	1	2.0	5
13	5	8.8	7	13.7	12
14	11	19.3	8	15.7	19
15	9	15.8	8	15.7	17
16	13	22.8	16	31.4	29
17	2	3.5	8	15.7	10
18	6	10.5	3	5.9	9
19	3	5.3	0	0.0	3
Total	57	100	51	100	108

Table 4 : Probability densities and likelihood ratios derived from the observed ridge count. The probability density for males: (C). The probability density for females (C1). LR: Likelihood Ratio

Ridge Count	Probability Density		Likelihood Ratio		Favored Odds Ratio	
	Males (C)	Females (C1)	Males (C/C1)	Females (C1/C)	Males (C/(C+C1))	Females (C1/(C+C1))
11	0.070	0.000	--	0.000	1.000	0.000
12	0.070	0.020	3.579	0.279	0.782	0.218
13	0.088	0.137	0.639	1.565	0.390	0.610
14	0.193	0.157	1.230	0.813	0.552	0.448
15	0.158	0.157	1.007	0.993	0.502	0.498
16	0.228	0.314	0.727	1.376	0.421	0.579
17	0.035	0.157	0.224	4.471	0.183	0.817
18	0.105	0.059	1.789	0.559	0.642	0.358
19	0.053	0.000	--	0.000	1.000	0.000

Probability densities were also computed from frequency distribution. Likelihood ratio was then computed using probability density values. Favored odds ratio was also computed from the probability densities (Table 4). For the mean ridge count range between 11 to 15, the likelihood ratio of males is generally higher than females except for ridge count of 13 where value is higher for females. The likelihood ratio becomes dominant for females in ridge count range of 16 and 17. The same trend is apparent in favored odds ratio values. In the range of 11-15 of the ridge count, the favored odds ratio is mostly greater than 0.5 for the males barring the ridge count of 13. After the ridge count of 15, the favored odds ratio is

inclined towards females. With this data, it can be inferred that fingerprint ridge count of 11-15 per 25 mm² area is likely of male origin. Conversely the ridge count greater than 15 is likely of female origin.

Discussion

Several studies have been conducted in the past globally to determine fingerprint ridge density for sex identification. The present study deals with the comparative analysis of fingerprint ridge densities between males and females of the Western Madhya Pradesh (Ujjain). The results obtained in various studies reveal that women have higher ridge as compared to male. Results of some earlier studies have tabulated in table.

Table 5 : Gender differentiation by ridge count among various studies.

S. No.	Author	Year	Country	Mean ridge count Males	Mean ridge count Females
1	(Cummins & Midlo, 1961)	1961	America	20.7	23.4
2	(Reddy 1975)	1975	India	13.41	12.04
3	(Acree, 1999)	1999	Africa	10.90	12.0
4	(Gungadin, 2007)	2007	India	12.8	14.5
5	(Nayak , et al., 2010)	2010	India	11.05	14.2
6	(Ping-ya, Wen-dong, & Li, 2012)	2012	China	14.5	15
7	(Kumar, Agarwal, Garg, Pratap, & Mishra, 2013)	2013	India	11.9	14.1
8	(Sam, Rema, & Nair, 2014)	2014	India	12.79	14.81
9	(Abdullah, Rahman, & Abas, 2015)	2015	Malaysia	11.4	15.1
10	(Kumar, Jirli, Honnunar, Babu, & kumar, 2017)	2017	India	13.56	16.92
11	(Patil, Malik, & TrezaShirole, 2018)	2018	India	10.62	12.69

All the studies cited above reveal that females tend to have higher ridge density as compared to males. Studies conducted by many other authors also showed that females have significantly higher ridge density than males (Agnihotri , Jowaheer , & Allock , 2012; Dhall & Kapoor, 2016; Wadhwa, 2013; Patil, Malik, & TrezaShirole, 2018).

However few studies conducted in the past revealed and opposite fingerprint ridge density showing more number of ridges in male as against females. (Reddy, 1975) responded mean ridge count for males 13.4 and that of females 12.04 of the Bagathas of Araku valley India. (Plato, Cereghino , & Steinberg, 1975) in a study on American negroes and caucasian American found mean ridge density more in male than female. These results may be due to defect in counting method as there is no detail of counting method. Besides, (Kaur, 2019) found out that male prisoners showed more ridge density than female prisoners in the study involving 125 prisoners 77 males and 48 females prisoners of Haryana the reason for this need to be evaluated.

Conclusion:

The study of fingerprint ridge densities offers valuable insights into gender differentiation, which is essential for forensic science. The data clearly indicate that females tend to have higher and more consistent ridge densities compared to males. The mean ridge densities, along with their distributions, provide a basis for probabilistic gender determination. Key conclusions drawn from this study include: Females generally exhibit higher mean ridge densities (15.32 ± 1.46) compared to males (14.97 ± 2.12). The distribution of ridge densities in females is more compact and consistent, with a range of 12 to 18, compared to the broader range of 11 to 19 in males. The likelihood and favored odds ratios indicate that ridge counts between 11-15 are predominantly male, except for the ridge count of 13, which is more likely female. Ridge counts above 15 are predominantly female. In summary, the differences in ridge density distributions and the corresponding likelihood and favored odds ratios underscore the importance of considering both the mean values and the variability in ridge counts. This comprehensive approach ensures a more accurate and reliable determination of the likely gender origin of fingerprints, which is crucial for forensic science and criminal investigations.

References

- Abdullah, S., Rahman, A., & Abas, Z. (2015). Classification of gender by using fingerprint ridge density in northern part of Malaysia. *Journal of Engineering and Applied Sciences*, 10, 10722-10726.
- Acree, M. A. (1999, May). Is there a gender difference in fingerprint ridge density? *forensic science international*, 35-44. doi:10.1016/s0379-0738(99)00037-7
- Agnihotri, A., Jowaheer, V., & Allock, A. (2012, Jul). An analysis of fingerprint ridge density in the Indo-Mauritian population and its application to gender determination. *Med Sci Law.*, 52(3), 143-147.
- Ceyhan, E., Sagiroglu, S., & Akyil, E. (2014). Gender classification based on ann with using fingerprint feature vectors. *Journal of the Faculty of Engineering and Architecture of Gazi University*, 29, 201-207.
- Cummins, H., & Midlo, C. (1961). *Finger prints, palms and soles: An introduction to dermatoglyphics* (Vol. 319). Dover Publications.
- Dhall, J., & Kapoor, A. (2016, Mar). Fingerprint Ridge Density as a Potential Forensic Anthropological Tool for Sex Identification. *J Forensic Sci.*, 61(2), 424-429. doi:10.1111/1556-4029.12959
- Gender differentiation by finger ridge count among South Indian population. *J Forensic Leg Med.* 2011 Feb;18(2):79-81. (2011, Feb). *J Forensic Leg Med.*, 18(2), 79-81. doi:10.1016/j.jflm.2011.01.006.
- Gungadin, D. (2007). Sex Determination from Fingerprint Ridge Density. 2(2). doi:10.4314/ijmu.v2i2.39847
- Gutiérrez, R. E., Alonso, M., & Dipierri, J. (2011). Sex differences in fingerprint ridge density in the Mataco-Mataguayo population. *Homo*, 62(6), 487-499.
- Gutiérrez Redomero, E., Rivalderia, N., Alonso, M. C., & Andres, A. S. (2013). A comparative study of fingerprint ridge density in Spanish and Argentine populations. *Forensic Science International*, 158-166.
- Gutiérrez, R. E., Alonso, C., Romero, E., & Galera, V. (2008, Aug 25). Variability of fingerprint ridge density in a sample of Spanish Caucasians and its application to sex determination. *Forensic Sci Int*, 180, 17-22. doi:10.1016/j.forsciint.2008.06.014.

- Gutiérrez-Redomero, E., Alonso, M. C., Hernandez Hurtado, L. E., & Rodriguez Villalba, J. L. (2011). Distribution of the minutiae in the fingerprints of a sample of the Spanish population. *Forensic Science International*, 79-90. doi:https://doi.org/10.1016/j.forsciint.2010.11.006
- Kaur, M. (2019). Fingerprint Ridge Density Of Convicted Male And Females Prisoners : A Pilot Study. *Brazilian Journal of Forensic Sciences, Medical Law and Bioethics*, 4, 226–234.
- Kocharyan , D., Khachaturyan, V., & Sarukhanyan , H. (2013). A Multimodal Biometric System Based on Fingerprint and signature recognition. In *Computer Science and Information Technologies (CSIT)*, 2013, 1-7.
- Kumar, L., Agarwal, S., Garg, R., Pratap, A., & Mishra. (2013, 10 1). Gender Determination Using Fingerprints In the Region of Uttarakhand. *Journal of Indian Academy of Forensic Medicine*, 35, 308-311. doi:10.1177/0971097320130404
- Kumar, S., Jirli, P., Honnunar, R., Babu, R., & kumar, M. (2017). "Fingerprint Ridge Density" - A Tool for Sex Determination. *Journal of Indian Academy of Forensic Medicine*, 39(1), 51-54. doi:10.5958/0974-0848.2017.00011.2
- Nayak , V., Rastogi , P., Kanchan , T., Yoganarasimha , K., Kumar, G., & Menezes , R. (2010, Apr 15). Sex differences from fingerprint ridge density in Chinese and Malaysian population. *Forensic Sci Int.*, 197, 67-69.
- Nithin , M., Balaraj , B., Manjunatha , B., & Mestri , S. (2009, Nov 16). Study of fingerprint classification and their gender distribution among South Indian population. *J Forensic Leg Med.*, 460-463. doi:10.1016/j.jflm.2009.07.001.
- Nithin, M., Manjunatha , B., Preethi , D., & Balaraj, B. (2011, Feb). Gender differentiation by finger ridge count among South Indian population. *J Forensic Leg Med.* 2011 Feb;18(2):79-81., 18(2), 79-81. doi:doi: 10.1016/j.jflm.2011.01.006.
- Nte, N. D. (2012). An Evaluation of the Challenges of Forensic Investigation and Unsolved Murders in Nigeria. *African Journal of Criminology and Justice Studies*, 6, 143.
- Patil, D., Malik, A., & TrezaShirole. (2018). Finger Print Ridge Density a Tool for Gender Determination.
- Ping-ya, N. I., Wen-dong, G. E., & Li, P. (2012). Sex differences from fingerprint ridge density in the Chinese Han Population. *Forensic Science and Technology*, 2, 002.
- Plato, C., Cereghino , J., & Steinberg, F. (1975). The dermatoglyphics of American Caucasians. *Am J Phys Anthropol.*, 42(2), 192-210. doi:10.1002/ajpa.1330420205
- Reddy , G. (1975, Mar). Finger dermatoglyphics of the Bagathas of Araku Valley (A. P.), India. *Am J Phys Anthropol.* 1975, 42(2), 225-228. doi:10.1002/ajpa.1330420208.
- Sam, N., Rema, P., & Nair, B. (2014). Sex Determination Using Fingerprint Ridge Density In South Indian Population. *Journal of Indian Academy of Forensic Medicine*, 36(4), 381386.
- Wadhwa, R. (2013). Age and Gender Determination from Finger Prints using RVA and dct Coefficients. *IOSR Journal of Engineering*, 3, 5-9. doi:10.9790/3021-03850509
- Wahdan, A., & Khalifa, H. (2017). Sex Identification from Fingerprint Ridge Density in Egyptian Population. *Mansoura Journal of Forensic Medicine and Clinical Toxicology*, 25, 1-13. doi:10.21608/mjfmct.2018.47259

51. ZnO nanoparticles as an active photocatalyst for rapid Methylene Blue removal

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Abstract: Water contamination by synthetic dyes remains a major environmental challenge due to their chemical stability, toxicity, and resistance to conventional treatment methods. Among these dyes, Methylene blue (MB) is widely used as a pollutant for evaluating photocatalytic performance. Zinc oxide (ZnO) nanoparticles have emerged as highly efficient photocatalysts owing to their wide band gap, strong oxidising ability, cost-effectiveness, and versatile tunability through controlled synthesis. This section outlines the key methods used to synthesise ZnO nanoparticles and explains how each preparation route affects their morphology, defect structure, and overall optical behaviour. The photocatalytic degradation mechanism of MB using ZnO is also discussed in detail. Additionally, we study performance trends among pristine, doped, and composite ZnO systems to identify key factors governing enhanced photocatalytic efficiency.

Keywords: Zinc Oxide, Dye, Photocatalysis, Degradation, Methylene Blue.

Introduction

The increasing utilisation of synthetic dyes in sectors including textiles, printing, and medicines has resulted in a chronic issue of dye-contaminated wastewater. Numerous dyes exhibit chemical stability and resist degradation in natural environments, leading to their accumulation in aquatic ecosystems and disruption of marine organisms and water quality. Methylene blue (MB) is a prevalent dye found in industrial effluents and is often utilised as a model contaminant in photocatalytic research due to its vibrant colour, high water solubility, and resilience to standard treatment techniques. Among the numerous strategies investigated for dye elimination, semiconductor-based photocatalysis has surfaced as a promising and sustainable method. During this process, light irradiation stimulates a semiconductor material, resulting in the formation of electron–hole pairs that engage in redox reactions over the catalyst's interface (Feng et al. 2017). These reactions generate highly reactive oxygen species that can efficiently decompose complex dye molecules, producing simpler, less toxic substances. In contrast to conventional wastewater treatment techniques, photocatalysis presents benefits such as reduced chemical requirements, lowered secondary contamination, and the potential for solar energy utilisation.

Zinc oxide (ZnO) has garnered significant interest as a photocatalyst owing to its chemical stability, affordability, non-toxicity, and advantageous electrical characteristics. ZnO has a large surface area and more active sites when it is employed as nanoparticles, which is advantageous for photocatalytic processes. Numerous studies have documented the effective breakdown of methylene blue utilising ZnO nanoparticles; nevertheless, the extensive band gap of ZnO limits its light absorption predominantly to the UV spectrum (Liu et al. 2015). The fast recombination of the photogenerated charge carriers constrains its overall

photocatalytic efficacy under visible light. Various techniques, including chemical doping, defect engineering, and the application of oxidising chemicals, have been utilised to improve the photocatalytic efficacy of ZnO to mitigate these limitations. Transition metal doping has been shown to effectively alter the electrical composition of ZnO, enhance visible-light absorption, and inhibit charge carrier recombination.

This work presents a succinct review of ZnO Nanoparticle-based photocatalysts for the elimination of methylene blue from aqueous solutions along with our published work (Aarti Pathak, Anjali Soni, Archana Chaudhary 2024). Photocatalytic processes, synthesis methods, and modification techniques are highlighted, backed by typical experimental data from Mn-doped ZnO systems. The objective is to emphasise recent advancements and describe potential pathways for the advancement of effective ZnO-based photocatalysts for wastewater remediation.

Fundamentals of ZnO Photocatalysis

Zinc oxide (ZnO) is a broad bandgap semiconductor that has photocatalytic activity when exposed to light, producing mobile charge carriers. Upon exposure to photons of sufficient energy, ZnO nanoparticles facilitate the excitation of electrons from the valence band to the conduction band, resulting in the formation of matching holes. (Duan et al. 2015). The photogenerated holes and electrons traverse to the catalyst's surface, where they commence a sequence of redox processes.

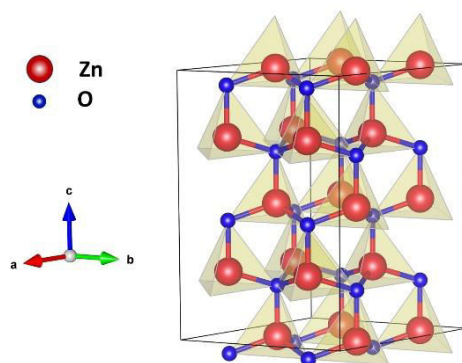


Fig.1: Structure of Zinc Oxide nanoparticles

At the catalyst-solution interface, holes oxidise surface-adsorbed water molecules or just hydroxide ions to generate hydroxyl radicals ($\bullet\text{OH}$), while excited electrons convert dissolved oxygen to yield superoxide radicals ($\text{O}_2^{\bullet-}$) (Singh et al. 2017). Reactive oxygen species are exceptionally oxidative and chiefly accountable for the breakdown of organic contaminants like methylene blue through sequential oxidation processes, ultimately resulting in mineralisation.

The photocatalytic efficacy of ZnO is influenced by various intrinsic and extrinsic variables, such as crystallinity, particle size, surface imperfections, and charge carrier lifespan. Despite ZnO exhibiting effective photocatalytic activity under UV light, its practical use under visible light is constrained by its extensive band gap and quick recombination of electron-hole pairs. (Parvin et al. 2012). Consequently, techniques that boost charge separation and augment visible-light absorption are crucial for attaining superior photocatalytic performance in ZnO-based systems.

Synthesis Routes of ZnO Nanoparticles

The synthesis method is pivotal in influencing the structural, optical, and photocatalytic characteristics of ZnO nanoparticles. The preparation process significantly influences parameters such as the size of particles, crystallinity, shape, and defect density, which subsequently affect photocatalytic efficiency. Various chemical synthesis methods have been extensively utilised for the production of ZnO nanoparticles appropriate for the breakdown of methylene blue.

1.Sol–gel method: The sol–gel method is a prevalent chemical approach for fabricating zinc oxide nanoparticles, owing to its accessibility, cost-effectiveness, and potential to regulate particle size along with morphology. Zinc precursors, such as zinc acetate, undergo hydrolysis and condensation in a solution to produce a stable colloidal sol that progressively unfolds into a gel network(Zhu 2025). The gel is further dried and calcined to produce ZnO nanoparticles characterised by great purity and uniformity. This technique enables precise regulation of structural characteristics by modifying reaction parameters, including precursor concentration, pH, and calcination temperature, that directly affect photocatalytic efficacy.(Geldasa and Kebede 2023). Multiple investigations indicate that ZnO produced by the sol–gel method exhibits excellent crystallinity and effective photocatalytic activity against organic contaminants, such as methylene blue, when exposed to UV irradiation.(Balcha et al. 2016).

2.Hydrothermal synthesis: The hydrothermal synthesis method represents a solution-based technique commonly utilised to produce ZnO nanoparticles with regulated size, superior crystallinity, and distinct shape. This technique involves dissolving zinc precursors in an aqueous solution and subjecting them to high temperatures and autogenous pressure within a sealed autoclave, subsequently facilitating crystal formation via dissolution–recrystallisation processes (Baruah and Dutta 2009). Hydrothermal conditions facilitate the synthesis of highly crystalline ZnO in tunable morphologies, including rods, flowers, and spheres, by modifying variables like temperature, duration, and precursor concentration.(Wang et al. 2014). The hydrothermal method is intensively investigated for its capacity to generate defect-regulated and phase-pure nanostructures, thereby improving the photocatalytic degradation of chemical dyes, such as methylene blue. Studies indicate that hydrothermally synthesised ZnO demonstrates enhanced charge separation and elevated surface area, resulting in higher photocatalytic effectiveness under ultraviolet and visible light.

3.Chemical precipitation: The chemical precipitation approach is among the most straightforward and economical techniques for producing ZnO nanoparticles. This method involves the reaction of zinc salts with a precipitating agent under regulated conditions, resulting in the development of zinc hydroxide intermediates, which convert to ZnO during drying and calcination(Gatou et al. 2023). This technique facilitates precise regulation of particle size and surface area by the modification of factors like pH, temperature, and precursor concentration. Owing to its scalability and minimal energy demands, precipitation-derived ZnO nanoparticles have been extensively utilised in photocatalytic applications.(Mim et al. 2023). Numerous studies have shown that ZnO produced through chemical precipitation displays significant surface reactivity and efficient photocatalytic degradation of organic dyes, such as methylene blue, when exposed to UV and visible light irradiation.

4.Combustion synthesis: Combustion synthesis is a rapid and energy-efficient technique for generating ZnO nanoparticles, relying upon an exothermic redox reaction involving a zinc

precursor and an appropriate fuel. The reaction takes place at comparatively low external temperatures yet produces significant localised heat, resulting in the rapid production of nanocrystalline ZnO (Dutta). This technique generally produces ZnO nanoparticles characterised by a high surface area and a significant density of structural flaws, including oxygen vacancies, which are advantageous for photocatalytic processes. Owing to its simplicity, affordability, and scalability, combustion-synthesised ZnO has been extensively investigated for the photocatalytic degradation of organic dyes, such as methylene blue, demonstrating improved efficacy under UV and visible light irradiation (Arora et al. 2014).

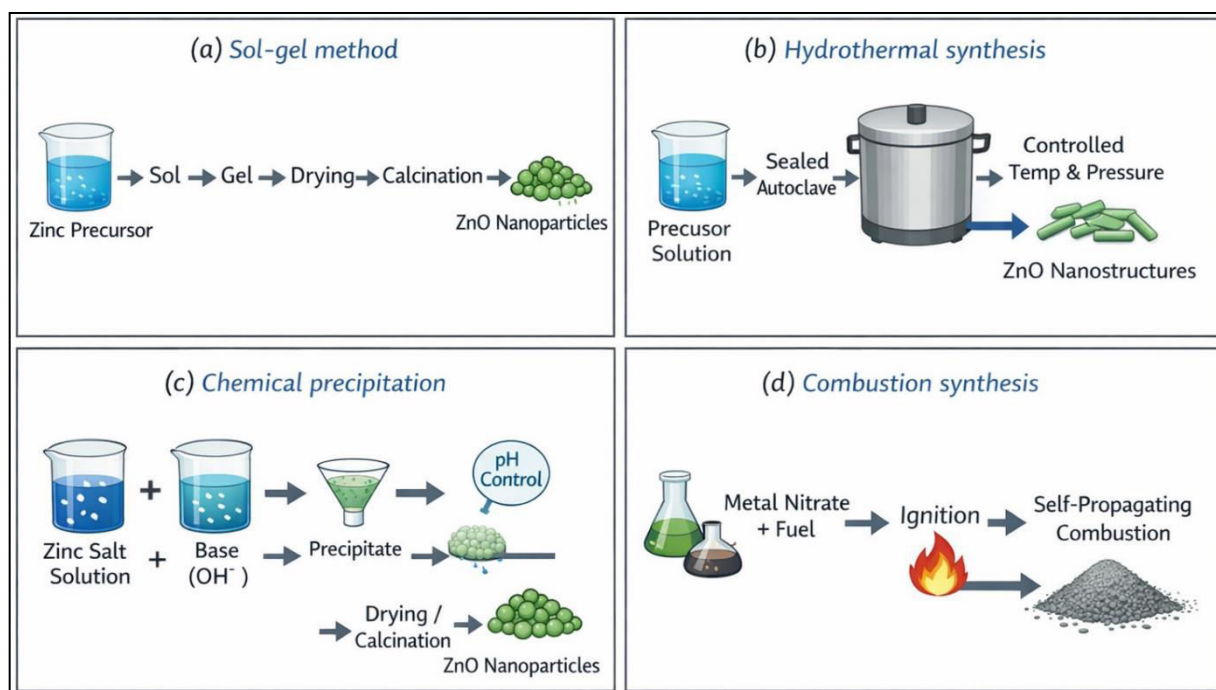


Fig.2: Schematic illustration of commonly employed synthesis routes for ZnO nanoparticles, including (a) sol-gel method, (b) hydrothermal synthesis, (c) chemical precipitation, and (d) combustion synthesis.

Effect of synthesis route

The production technique is crucial in defining the physicochemical parameters of ZnO nanoparticles, which directly affect their photocatalytic efficacy. Alterations in preparation techniques yield notable disparities in crystallinity, shape, and surface attributes, thereby influencing the overall efficacy of photocatalytic degradation processes.

1. Crystallinity: Diverse synthesis techniques provide ZnO nanoparticles with various levels of crystallinity. Methods like hydrothermal and sol-gel synthesis typically produce well-crystallised ZnO structures, enhancing charge transport efficiency and minimising bulk recombination of photogenerated carriers (Khan et al.). Conversely, fast synthesis techniques may generate structural defects that may either augment or impede photocatalytic activity, conditional upon their concentration.

2. Morphology: The synthesis process significantly affects the shape of ZnO nanoparticles. Hydrothermal procedures frequently provide well-defined nanostructures, including rods, flowers, or spheres, whereas sol-gel and precipitation approaches generally generate quasi-spherical or aggregated particles (Chen et al. 2020). Regulated shape enhances surface area and the accessibility of active sites, which are crucial for efficient dye adsorption and degradation.

3. Photocatalytic Activity: The photocatalytic performance results from the relationship of crystallinity, shape, and defect density generated during synthesis. ZnO nanoparticles produced by regulated methods often have superior photocatalytic activity owing to greater light absorption, effective charge separation, and an augmented number of reactive surface sites (Subash et al. 2013). Consequently, choosing an optimal synthesis process is essential for optimising ZnO-based photocatalysts for the degradation of methylene blue.

Strategies to Enhance ZnO Photocatalytic Performance

Various ways have been utilised to address the intrinsic constraints of pristine ZnO, specifically its significant band gap and swift charge carrier recombination. These methods seek to enhance light absorption, optimise charge separation, and elevate the production of reactive oxygen species, consequently enhancing photocatalytic efficiency.

1. Metal and non-metal doping: Doping ZnO with metallic or non-metallic materials is an effective method to alter its electrical structure. The incorporation of dopant atoms generates localised energy states within the band gap, thereby augmenting visible-light absorption and inhibiting electron-hole recombination (Anandan et al. 2016). Non-metal dopants predominantly impact surface states, whereas metal dopants influence both structural and electrical characteristics.

2. Transition metal dopants (Mn, Fe, Cr, Co): Transition metals, including Mn, Fe, Cr, and Co, are frequently employed as dopants owing to their appropriate ionic radii and diverse oxidation states. Their integration into the ZnO lattice creates defect levels that enhance charge trapping and increase charge transfer efficiency (Jung et al. 2018). Manganese doping has been extensively documented to markedly improve visible-light-induced photocatalytic degradation of organic dyes.

3. Band gap tuning and defect engineering: Band gap engineering via regulated doping or synthesis conditions enables ZnO to absorb a wider spectrum of solar radiation. Defect engineering, specifically the incorporation of zinc interstitials and oxygen vacancies, generates intermediate energy levels that facilitate electron excitation and extend charge carrier lifetime, resulting in enhanced photocatalytic activity (Mittal et al. 2014).

4. Role of surface oxygen vacancies: Surface oxygen vacancies serve as active sites for the adsorption of oxygen molecules and contaminants. These vacancies facilitate the generation of superoxide and hydroxyl radicals, which are critical species in photocatalytic degradation events (She et al. 2012). Nonetheless, an ideal concentration of oxygen vacancies is crucial, as an overabundance of defects may function as recombination centres.

5. Use of oxidants (H₂O₂): The incorporation of oxidising chemicals like hydrogen peroxide effectively improves photocatalytic performance. H₂O₂ functions as an electron acceptor and facilitates the production of hydroxyl radicals, hence expediting the breakdown of organic pigments (Šutka et al. 2016). The collaborative effect of ZnO photocatalysts and H₂O₂ markedly enhances degradation efficiency in the presence of visible light.

6. Composite and heterojunction approaches: The fabrication of ZnO-based composites as well as heterojunctions combined with additional semiconductors or carbonaceous materials enhances charge separation by promoting directional charge transfer at interfaces (Zong et al. 2014). These systems lessen recombination losses and improve photocatalytic stability, rendering them viable options for sophisticated wastewater treatment applications.

Representative Experimental Insights (Mn-Doped ZnO)

Experimental research on Mn-doped ZnO nanoparticles offers significant insights into how the addition of transition metals alters the structural, optical, and photocatalytic properties of ZnO. These data demonstrate distinct structure–property–performance linkages that account for the augmented degradation of methylene blue during light irradiation.

1. Structural and Optical Features: Manganese doping typically maintains the hexagonal wurtzite structure of zinc oxide while causing minor lattice anomalies due to the substitutional insertion of manganese ions. These structural alterations affect crystallite size and defect density (Aarti Pathak, Anjali Soni, Archana Chaudhary 2024). Optical investigations often reveal increased visible-light absorption and minor alterations in the band gap, resulting from Mn-induced impurity states and oxygen-related defects in the ZnO lattice.

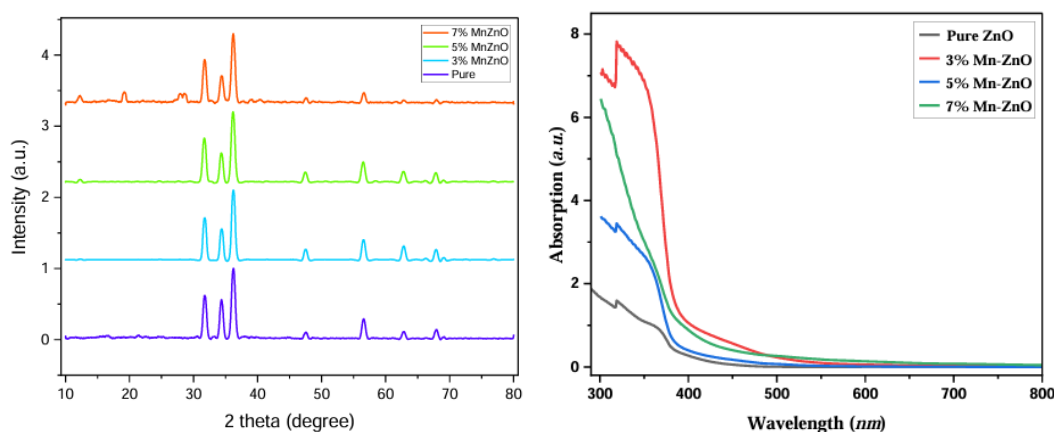


Fig.3:(a) XRD graph for Pure & Mn-ZnO (3, 5, 7%), (b) UV-Vis spectra for Pure & Mn-ZnO (3, 5, 7%) (Aarti Pathak, Anjali Soni, Archana Chaudhary 2024)

2. Photocatalytic Performance Trends: Mn-doped ZnO consistently exhibits enhanced photocatalytic degradation efficiency for methylene blue relative to undoped ZnO. The enhancement is ascribed to superior charge carrier separation, augmented production of reactive oxygen species, and greater interaction between the catalyst surface and dye molecules (Shirdel and Behnajady 2017). An ideal Mn concentration is frequently noted, beyond which surplus dopant may diminish efficiency due to recombination phenomena.

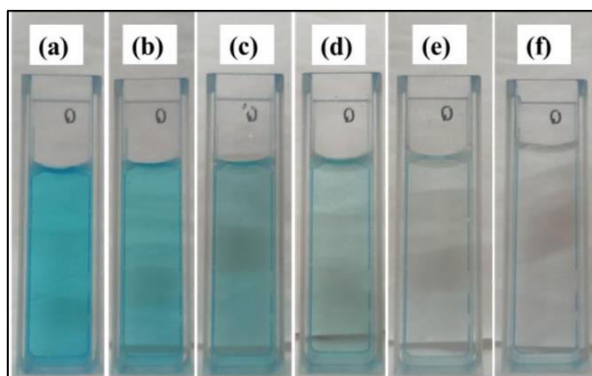


Fig.4: Photocatalytic Methylene blue dye degradation using Mn-doped ZnO NPs in 5 hours (Aarti Pathak, Anjali Soni, Archana Chaudhary 2024)

3. Kinetic Behaviour: The photocatalytic degradation of methylene blue via Mn-doped ZnO often adheres to pseudo-first-order reaction kinetics. Doped systems exhibit elevated rate constants, signifying accelerated degradation rates. This kinetic improvement demonstrates

the significant function of Mn in promoting charge transfer and extending the lifespan of photogenerated charge carriers

Challenges and Future Perspectives

Despite the significant promise of ZnO-based photocatalysts for the removal of organic pollutants, certain restrictions continue to prevent their practical application. A primary difficulty is the fast recombination of photogenerated charge carriers, which markedly diminishes photocatalytic efficacy. Moreover, ZnO is susceptible to photo corrosion under extended irradiation, resulting in structural instability and diminished catalytic longevity. Attaining exact control over dopant concentration and uniformity of dopant incorporation remains challenging, as excessive or non-uniform doping can generate recombination sites that negatively impact performance.

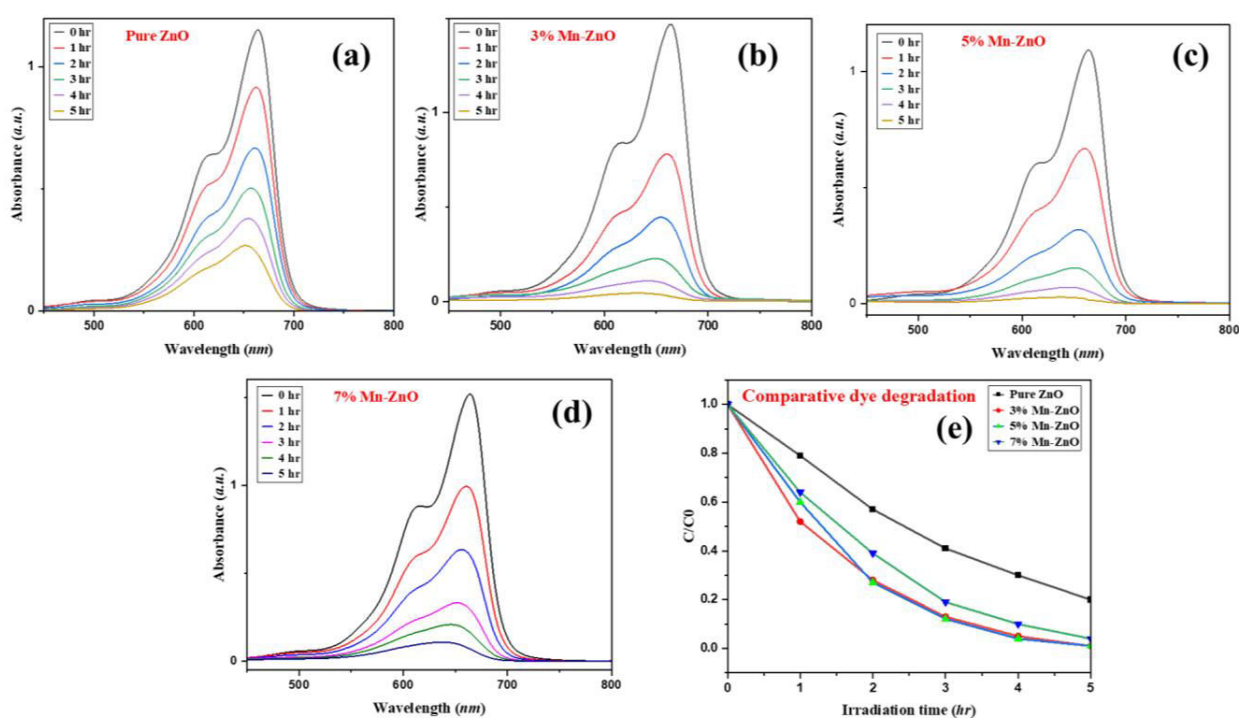


Fig.5: Absorption spectra of Methylene blue dye for (a) 3% Mn-ZnO, (b) 5% Mn-ZnO, (c) 7% Mn-ZnO, (d) pure ZnO, (e) Photocatalytic dye degradation study of Mn-doped nanoparticles for Methylene blue dye(Aarti Pathak, Anjali Soni, Archana Chaudhary 2024)

The recovery and reusability of ZnO photocatalysts are critical issues from an application perspective, especially in large-scale wastewater treatment systems. Moreover, the majority of photocatalytic research is performed under optimal laboratory settings, while actual wastewater matrices have competing ions, fluctuating pH levels, and intricate organic components that may inhibit photocatalytic efficacy. These factors must be rigorously analysed to determine the genuine applicability of ZnO-based systems.

Subsequent investigations should concentrate on the systematic design of stable ZnO-based photocatalysts exhibiting improved visible-light activity and extended charge carrier lifetimes. Strategies like defect engineering, heterojunction engineering, and composite fabrication present fascinating possibilities to address existing limits. The advancement of solar-driven and multifunctional photocatalytic systems will be essential for progressing ZnO-based materials in sustainable and effective wastewater remediation methods.

Conclusion

ZnO nanoparticles have emerged as excellent and flexible photocatalysts for the degradation of methylene blue in aqueous environments, attributed to their advantageous electrical characteristics, chemical stability, and cost-effectiveness. This review elucidates the essential photocatalytic mechanism of ZnO and emphasises the pivotal influence of synthesis methods on crystallinity, shape, and photocatalytic efficacy. Multiple modification strategies, such as elemental doping, defect engineering, oxidant assistance, and composite synthesis, have been identified as effective methods to address the inherent limits of pristine ZnO.

Experimental findings from Mn-doped ZnO systems indicate that the introduction of transition metals can markedly augment visible-light absorption, inhibit charge carrier recombination, and enhance degradation kinetics. The photocatalytic efficiency is significantly influenced by dopant concentration and defect density, with optimised systems demonstrating enhanced performance relative to undoped ZnO. Despite ongoing issues concerning stability, reusability, and real-world wastewater applicability, additional developments in material design and system integration are anticipated to enhance the practical viability of ZnO-based photocatalysts. ZnO and its modified nanostructures are potential options for sustainable and effective wastewater treatment applications.

Acknowledgement:

The authors acknowledge Medicaps University for supporting this work under the URF Project Number: URF-MU-2022-06. We would also like to thank UGC-DAE-CSR, Indore, for helping us with the characterisation.

References:

- Aarti Pathak, Anjali Soni, Archana Chaudhary UD (2024) The Influence of Manganese Doping on Zinc Oxide Nanoparticles for Photocatalytic Degradation of Methylene Blue Dye. *Eurasian J Anal Chem* 19:575–583
- Anandan M, Dinesh S, Krishnakumar N, Balamurugan K (2016) Influence of Co doping on combined photocatalytic and antibacterial activity of ZnO nanoparticles. *Mater Res Express* 3:1–12. <https://doi.org/10.1088/2053-1591/3/11/115009>
- Arora AK, Devi S, Jaswal VS, et al (2014) Synthesis and Characterization of ZnO Nanoparticles
- Balcha A, Yadav OP, Dey T (2016) Photocatalytic degradation of methylene blue dye by zinc oxide nanoparticles obtained from precipitation and sol-gel methods. *Environ Sci Pollut Res*. <https://doi.org/10.1007/s11356-016-7750-6>
- Baruah S, Dutta J (2009) Hydrothermal growth of ZnO nanostructures. 6996:.. <https://doi.org/10.1088/1468-6996/10/1/013001>
- Chen X, Xu X, Cui J, et al (2020) Visible-light driven degradation of tetracycline hydrochloride and 2,4-dichlorophenol by film-like N-carbon@N-ZnO catalyst with three-dimensional interconnected nanofibrous structure. *J Hazard Mater* 392:122331. <https://doi.org/10.1016/j.jhazmat.2020.122331>
- Duan L, Zhao X, Zheng Z, et al (2015) Structural, optical and photocatalytic properties of (Mg,Al)-codoped ZnO powders prepared by sol-gel method. *J Phys Chem Solids* 76:88–93. <https://doi.org/10.1016/j.jpcs.2014.07.003>
- Dutta S Synthesis of ZnO via solution combustion method; study the effect of ignition temperature and fuel quantity Department of Chemical Engineering Indian Institute

Technology (ISM) Dhanbad

- Feng Y, Lu H, Gu X, et al (2017) ZIF-8 derived porous N-doped ZnO with enhanced visible light-driven photocatalytic activity. *J Phys Chem Solids* 102:110–114. <https://doi.org/10.1016/j.jpcs.2016.11.022>
- Gatou M, Kontoliou K, Volla E, et al (2023) Optimization of ZnO Nanoparticles ' Synthesis via Precipitation Method Applying Taguchi Robust Design. 1–34
- Geldasa FT, Kebede A (2023) Experimental and computational study of metal oxide nanoparticles for the photocatalytic degradation of organic pollutants : a review. 18404–18442. <https://doi.org/10.1039/d3ra01505j>
- Jung HJ, Koutavarapu R, Lee S, et al (2018) Enhanced photocatalytic activity of Au-doped Au@ZnO core-shell flower-like nanocomposites. *J Alloys Compd* 735:2058–2066. <https://doi.org/10.1016/j.jallcom.2017.11.378>
- Khan W, Method M, Zhang Z, Li X Synthesis of Pr-doped ZnO nanoparticles : Their structural , optical , and photocatalytic properties Synthesis of Pr-doped ZnO nanoparticles : Their structural , optical , and photocatalytic properties *. <https://doi.org/10.1088/1674-1056/27/8/086102>
- Liu X, Wang X, Li H, et al (2015) Enhanced visible light photocatalytic activity of ZnO doped with down-conversion NaSrBO₃:Tb³⁺ phosphors. *Dalt Trans* 44:97–103. <https://doi.org/10.1039/c4dt01252f>
- Mim SR, Mondal S, Jamal M, Billah M (2023) Synthesis of ZnO Nanoparticles by Co-Precipitation Method : Effect of Washing on the Purity of the Nanoparticles. 23:
- Mittal M, Sharma M, Pandey OP (2014) UV-Visible light induced photocatalytic studies of Cu doped ZnO nanoparticles prepared by co-precipitation method. *Sol Energy* 110:386–397. <https://doi.org/10.1016/j.solener.2014.09.026>
- Parvin T, Keerthiraj N, Ibrahim IA, et al (2012) Photocatalytic degradation of municipal wastewater and brilliant blue dye using hydrothermally synthesized surface-modified silver-doped ZnO designer particles. *Int J Photoenergy* 2012:. <https://doi.org/10.1155/2012/670610>
- She G, Chen X, Wang Y, et al (2012) Electrodeposition of al-doped ZnO nanoflowers with enhanced photocatalytic performance. *J Nanosci Nanotechnol* 12:2756–2760. <https://doi.org/10.1166/jnn.2012.5744>
- Shirdel B, Behnajady MA (2017) Sol-gel synthesis of Ba-doped ZnO nanoparticles with enhanced photocatalytic activity in degrading Rhodamine B under UV-A irradiation. *Optik (Stuttg)* 147:143–150. <https://doi.org/https://doi.org/10.1016/j.ijleo.2017.08.059>
- Singh S, Sharma R, Mehta BR (2017) Enhanced surface area, high Zn interstitial defects and band gap reduction in N-doped ZnO nanosheets coupled with BiVO₄ leads to improved photocatalytic performance. *Appl Surf Sci* 411:321–330. <https://doi.org/10.1016/j.apsusc.2017.03.189>
- Subash B, Krishnakumar B, Swaminathan M, Shanthi M (2013) Highly efficient, solar active, and reusable photocatalyst: Zr-loaded Ag-ZnO for reactive red 120 dye degradation with synergistic effect and dye-sensitized mechanism. *Langmuir* 29:939–949. <https://doi.org/10.1021/la303842c>
- Šutka A, Käämbre T, Pärna R, et al (2016) Co doped ZnO nanowires as visible light photocatalysts. *Solid State Sci* 56:54–62. <https://doi.org/10.1016/j.solidstatesciences.2016.04.008>
- Wang J, Saufan A, Lin PH, et al (2014) Mechanical properties and strengthening behavior of Mg e Zn e MM alloy. *Mater Chem Phys* 148:28–31. <https://doi.org/10.1016/j.matchemphys.2014.07.004>
- Zhu C (2025) Nanomaterial ZnO Synthesis and Its Photocatalytic Applications : A Review. 1–32
- Zong Y, Li Z, Wang X, et al (2014) Synthesis and high photocatalytic activity of Eu-doped ZnO nanoparticles. *Ceram Int* 40:10375–10382. <https://doi.org/10.1016/j.ceramint.2014.02.123>

52. Coumarin and its Derivatives: Chemistry, Pharmacological Significance, and Emerging Applications

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Abstract: Coumarin is a naturally occurring oxygen-containing heterocyclic compound widely distributed in plants and recognized as a versatile scaffold in organic and medicinal chemistry. Owing to its rigid benzopyrone framework and ease of structural modification, coumarin and its derivatives exhibit a broad spectrum of biological activities, including anticoagulant, anticancer, antimicrobial, anti-inflammatory, antioxidant, antiviral, and antidiabetic properties. In medicinal chemistry, coumarin-based molecules have been extensively explored as enzyme inhibitors, fluorescent probes, and pharmacophores in drug design. Beyond pharmaceutical applications, coumarins play significant roles in agrochemicals, perfumery, food additives, and material science, particularly in the development of dyes, optical brighteners, and laser-active materials. Advances in synthetic methodologies, including green and sustainable approaches, have further expanded the accessibility and functional diversity of coumarin derivatives. This abstract highlights the chemical characteristics, biological relevance, and emerging applications of coumarin, emphasizing its importance as a privileged heterocyclic scaffold with substantial potential for future research and technological development.

Keywords: Coumarin derivatives, anticoagulant, anticancer, antimicrobial, anti-inflammatory, agrochemicals, perfumery, food additives, biological relevance

Introduction

Coumarin (2H-1-benzopyran-2-one) is a naturally occurring heterocyclic compound with a fused benzene and lactone ring (benzopyrone structure). It is widely found in plants (e.g., cinnamon, tonka beans) and even in bacteria and fungi. Coumarins are commonly synthesized through simple and versatile condensation reactions that allow wide structural modification. They are well known for their anticoagulant properties, exemplified by drugs such as warfarin, and also show significant antimicrobial, anticancer, anti-inflammatory, antioxidant, antiviral, and neuroprotective activities. Coumarin and its derivatives are increasingly used in fluorescent probes, laser dyes, and biological imaging due to their strong photophysical properties. They also find non-therapeutic applications in agrochemicals, perfumes, food additives, and optical materials, highlighting their industrial and technological relevance. Substituents at C-3 and C-4 profoundly affect target affinity, selectivity, and potency.

Review of literature

Literature reports indicate that coumarin and its derivatives are important benzopyrone compounds extensively studied for their diverse chemical and biological properties.

Numerous studies have focused on their efficient synthesis through classical and green methods, as well as on their broad pharmacological activities such as anticoagulants, antimicrobial, anticancer, anti-inflammatory, and antioxidant effects. Recent reviews also highlight emerging non-therapeutic applications of coumarins in fluorescent probes, materials science, and agrochemicals, emphasizing their growing multidisciplinary significance.

In addition to therapeutic properties, emerging research also focuses on coumarin applications beyond pharmacology. Coumarin-based compounds are being explored in material sciences, particularly in photo responsive polymers and fluorescent probes, due to their unique photochemical properties and ability to undergo reversible photodimerization.

Methodologies

This review systematically examines the major synthetic methodologies employed for the preparation of coumarin and its derivatives, as reported in the literature. Emphasis is placed on reaction mechanisms, reagents, conditions, advantages, and applicability in medicinal chemistry.

1. Pechmann Condensation

The Pechmann condensation is the most widely used and classical method for coumarin synthesis. It involves the acid-catalyzed condensation of phenols with β -ketoesters (such as ethyl acetoacetate) using catalysts like concentrated sulfuric acid, phosphoric acid, or Lewis acids. This method is favored due to its simplicity, high yields, and suitability for synthesizing 4-substituted coumarins. Recent modifications include solvent-free, microwave-assisted, and green catalyst approaches to improve efficiency and sustainability.

2. Knoevenagel Condensation

In this method, o-hydroxy benzaldehydes react with active methylene compounds (e.g., malonic acid, cyanoacetic esters) in the presence of basic catalysts such as piperidine or ammonium acetate. The reaction proceeds through aldol condensation followed by lactonization to yield coumarins. This route allows greater control over substitution at the 3-position and is widely used for synthesizing functionalized coumarin derivatives.

3. Perkin Reaction

The Perkin reaction involves the condensation of o-hydroxy benzaldehydes with acid anhydrides in the presence of a base, followed by cyclization to form the coumarin nucleus. Although less commonly used due to harsher conditions and lower yields, this method is useful for synthesizing specific coumarin frameworks and has historical significance in coumarin chemistry.

4. Reformatsky Reaction

The Reformatsky reaction employs α -halo esters and o-hydroxybenzaldehyde in the presence of zinc to form β -hydroxy esters, which cyclize to produce coumarins. This method provides access to substituted coumarins under relatively mild conditions and is particularly useful for introducing alkyl substituents.

5. Wittig and Related Olefination Reactions

Wittig reactions involving o-hydroxybenzaldehyde and phosphonium ylides generate olefinic intermediates that undergo intramolecular cyclization to form coumarins. These methods are valuable for preparing highly substituted and structurally complex coumarin derivatives.

6. Metal-Catalyzed and Modern Synthetic Approaches

Recent advancements include transition-metal-catalyzed reactions using palladium, gold, rhodium, or copper catalysts. These methods enable regioselective and efficient synthesis of coumarins through C–C and C–O bond formation. Additionally, microwave-assisted synthesis, ionic liquids, enzymatic catalysis, and solvent-free reactions have been developed to enhance reaction rates and reduce environmental impact.

7. Green Chemistry Approaches

To address environmental concerns, green synthetic methodologies employing biodegradable catalysts, aqueous media, reusable solid acids, and energy-efficient techniques have been widely explored. These approaches offer cleaner, cost-effective, and scalable routes to coumarin synthesis.

Result and Discussion

The reviewed studies indicate that coumarin and its derivatives can be synthesized effectively through a variety of classical and modern methods. Among these, Pechmann condensation remains the most commonly employed due to its simplicity and high yields, while Knoevenagel, Wittig, and metal-catalyzed reactions offer greater control over substitution patterns. Recent advances emphasize green and sustainable methodologies, such as solvent-free and microwave-assisted techniques, which improve reaction efficiency and reduce environmental impact.

Discussion of biological data reveals that the pharmacological activity of coumarin derivatives is strongly dependent on structural modifications, particularly substitutions at the C-3 and C-4 positions. These compounds exhibit a wide range of activities including antimicrobial, anticancer, anticoagulant, and anti-inflammatory effects. Additionally, emerging non-therapeutic applications in fluorescent materials, agrochemicals, and optical devices further demonstrate the versatility and growing importance of coumarin chemistry.

Conclusion

In conclusion, coumarin and its derivatives represent a highly significant class of benzopyrone compounds due to their simple yet versatile chemical framework, which allows extensive structural modification through a wide range of synthetic methodologies. Classical reactions such as Pechmann and Knoevenagel condensations continue to play a central role in coumarin synthesis, while modern approaches including metal-catalyzed, microwave-assisted, and green chemistry methods have enhanced efficiency, selectivity, and sustainability. Literature evidence clearly demonstrates that subtle changes in substitution patterns strongly influence pharmacological activities, leading to compounds with promising anticoagulant, antimicrobial, anticancer, anti-inflammatory, antioxidant, and neuroprotective properties. Beyond therapeutic relevance, coumarins have gained increasing attention in non-therapeutic fields such as fluorescent probes, optical materials, and agrochemicals. Overall, the continued integration of innovative synthetic strategies with structure–activity relationship studies is expected to further expand the applications of coumarin derivatives and support their development as valuable molecules in both medicinal and material sciences.

Acknowledgments

The author express sincere gratitude to the Department of Chemistry of Maharaja Ranjit Singh College of Professional Sciences, Indore for providing the necessary academic

resources and facilities to carry out this review work. Special thanks are extended to Prof. Deepanshu Pandey for his valuable guidance, constructive suggestions, and continuous encouragement during the preparation of this manuscript. The author also acknowledges the researchers and authors whose published work contributed significantly to the development of this review.

References

1. Borges, F., Roleira, F., Milhazes, N., Santana, L., & Uriarte, E. (2005). Simple coumarins and analogues in medicinal chemistry: Occurrence, synthesis and biological activity. *Current Medicinal Chemistry*, 12, 887–916.
2. Venugopala, K. N., Rashmi, V., & Odhav, B. (2013). Review on natural coumarin lead compounds for their pharmacological activity. *BioMed Research International*, 2013, 963248.
3. Kontogiorgis, C. A., & Hadjipavlou-Litina, D. J. (2005). Biological evaluation of several coumarin derivatives designed as possible anti-inflammatory/antioxidant agents. *Journal of Enzyme Inhibition and Medicinal Chemistry*, 20, 563–571.
4. Matos, M. J., Santana, L., Uriarte, E., Delogu, G., Corda, M., & Fadda, M. B. (2015). Coumarins—An important class of phytochemicals with biological activities. *Current Topics in Medicinal Chemistry*, 15, 205–222.
5. Riveiro, M. E., De Kimpe, N., Moglioni, A., et al. (2010). Coumarins: Old compounds with novel promising therapeutic perspectives. *Current Medicinal Chemistry*, 17, 1325–1338.
6. Zhang, L., Xu, Z., Wang, L., et al. (2021). Recent advances in coumarin-based anticancer agents. *European Journal of Medicinal Chemistry*, **213**, 113206.
7. Kostova, I. (2005). Synthetic and natural coumarins as cytotoxic agents. *Current Medicinal Chemistry – Anti-Cancer Agents*, 5, 29–46.
8. Pechmann, H. V., & Duisberg, C. (1884). Neue Bildungsweise der Cumarine. *Berichte der Deutschen Chemischen Gesellschaft*, 17, 929–936.
9. Khan, K. M., Saeed, S., et al. (2004). Synthesis and biological evaluation of coumarin derivatives. *Bioorganic & Medicinal Chemistry*, 12, 1963–1968.
10. El-Saghier, A. M., Naili, M. B., Rammash, B. K., Saleh, N. A., & Kredan, K. M. (2007). Synthesis and antibacterial activity of some new coumarin derivatives. *Arkivoc*, **2007**, 83–91.
11. Medina, F. G., Marrero, J. G., Macías-Alonso, M., et al. (2015). Coumarin heterocyclic derivatives: Chemical synthesis and biological activity. *Natural Product Reports*, 32, 1472–1507.
12. Miri, R., & Javidnia, K. (2014). Recent advances in the synthesis of coumarins. *Chemical Reviews*, 114, 1460–1499.
13. Zhao, H., & Neamati, N. (2016). Coumarin-based HIV integrase inhibitors. *Expert Opinion on Drug Discovery*, 11, 799–815.
14. Yang, J., Liu, G., & Li, Y. (2020). Coumarin-based fluorescent probes: Design and applications. *Sensors and Actuators B: Chemical*, 312, 127981.
15. Goel, A., & Ram, V. J. (2009). Natural and synthetic coumarins as anti-inflammatory agents. *Bioorganic & Medicinal Chemistry*, **17**, 3698–3706.

53. The Preparation of Lipid-Based Nanocarriers for Enhanced Corneal Penetration of Anti-Inflammatory Agents Targeting Chemokine Signalling

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Abstract: With the advancement in rate of disease, it is required to find out a efficient, biocompatible and ecofriendly drug delivery system. In this work, we explore lipid-based nanocarriers as a green alternative for ocular delivery of anti-inflammatory agents targeting chemokine-mediated corneal inflammation. Using a solvent-reduced hot homogenization technique, solid lipid nanoparticles were prepared and loaded with four therapeutics—ibuprofen, aspirin, diclofenac, and doxycycline—selected for their known influence on chemokine signalling and leukocyte migration in inflamed ocular tissue. The formulation process used biodegradable lipid, minimum amount of organic solvent, and very less energy input, making it more sustainable as compare to conventional polymeric or solvent- intensive nanocarriers. The high encapsulation efficiency was confirmed using UV-Vis spectrophotometry, the neutral pH and post centrifugation colloidal stability supported the ocular compatibility. This research highlights the potential of lipid nanocarriers to enhance corneal penetration, controlled drug release, and minimizing the dose amount. By this mean the overall therapeutic outcome was improved and the waste generation and chemical load was reduced. The utilization of predictive digital tools or AI-guided optimization can be used further to reduce trial and error experimentation. The demonstration of the lipid nanocarriers contributing toward sustainability, advancement and targeted approach in biomedical biotechnology incorporating nano-enabled ophthalmic therapeutics was the goal.

Keywords: Lipid-based nanocarriers, Solid lipid nanoparticles (SLNs), Corneal drug delivery, Anti-inflammatory agents, Chemokine signalling

Introduction

Effectively treating ocular inflammation remains a significant challenge due to the strong anatomical and physiological barriers of the eye. The cornea, in particular, restricts the entry of therapeutic agents, leading to suboptimal drug levels at target sites. To address this, researchers have explored lipid-based nanocarriers — nanoscale, biocompatible lipid systems that can encapsulate anti-inflammatory drugs and improve their penetration across ocular barriers. Beyond enhancing corneal bioavailability, these nanocarriers can be tailored to deliver agents that target chemokine signalling, a central pathway in ocular inflammation. By suppressing chemokine-driven leukocyte recruitment, this strategy aims to achieve more precise, effective, and sustained control of corneal inflammation.

Ocular Drug Delivery Challenges

The eye's anatomy provides multiple protective barriers that limit drug access to intraocular tissues. The tear film rapidly dilutes and clears topically administered drugs through lacrimal

drainage, resulting in poor residence time and minimal drug absorption^{[11] [12] [13] [14]}. The corneal epithelium, with its tight junctions, represents another formidable obstacle, restricting paracellular transport and limiting penetration of hydrophilic molecules^[12]. Furthermore, the cornea's sequential lipophilic–hydrophilic–lipophilic layers make it inherently difficult for most drugs to diffuse across^[12]. Efflux transporters further reduce absorption, while conjunctival absorption is often negligible^{[11] [12]}.

As a result, conventional topical formulations, such as eye drops, achieve <5% ocular bioavailability^[15], often necessitating frequent dosing^{[13] [16]}. This increases the risk of poor compliance and systemic absorption, which, although limited (<2% for drugs like prednisolone acetate), may still cause adverse effects^{[11] [17]}. Together, these barriers highlight the need for advanced delivery platforms that can extend residence time, improve penetration, and reduce systemic exposure^{[11] [14]}.

Role of Chemokines in Ocular Inflammation

Chemokines are small (8–10 kDa) chemotactic cytokines that orchestrate immune cell migration through binding to G-protein-coupled receptors^{[1] [4]}. They are critical regulators of ocular inflammation, mediating leukocyte infiltration in conditions such as uveitis, corneal graft rejection, allergic keratoconjunctivitis, and dry eye disease^[3]. Produced by both resident cells (e.g., corneal fibroblasts, retinal pigment epithelial cells) and infiltrating leukocytes^{[3] [4]}, chemokines amplify inflammatory cascades and contribute to tissue damage.

For example, IL-8 (CXCL8) attracts neutrophils in uveitis and keratopathy^{[1] [3]}, while MCP-1 (CCL2) recruits monocytes in uveitis and allergic eye disease^{[1] [4]}. Other chemokines, such as CXCL10 (implicated in infectious keratitis and dry eye)^{[3] [4]}, and CCL11 (Eotaxin-1) (elevated in vernal keratoconjunctivitis)^{[1] [3] [4]}, are strongly associated with pathological inflammation. Therapeutically, blocking these signalling pathways has been shown to reduce leukocyte infiltration, preserve corneal integrity, and improve outcomes in preclinical models^{[1] [3] [5]}.

Drug-Specific Mechanisms Targeting Chemokine Pathways

Several anti-inflammatory drugs indirectly or directly modulate chemokine expression:

- **Ibuprofen** inhibits COX enzymes, lowering prostaglandin synthesis and indirectly reducing chemokine release. It is effective in reducing inflammatory cell homing and has demonstrated ocular protective effects in experimental and clinical use^{[6] [7] [8]}.
- **Aspirin** similarly suppresses COX activity and prostaglandin production, thereby disrupting chemokine-driven leukocyte recruitment. Topical derivatives are used to manage post-operative ocular inflammation^[6].
- **Doxycycline** exhibits broader immunomodulatory effects, including suppression of MMP-9, IL-1, and TNF- α , while partially inhibiting CCL2 expression^{[9] [10]}. In dry eye models, it performs comparably to corticosteroids in reducing MMP-9 and MAPK activation^[9].
- **Diclofenac**, another NSAID, reduces prostaglandin synthesis and thereby lowers secondary chemokine induction. Clinically, it is widely used for postoperative ocular inflammation and allergic conjunctivitis^[6].

While these agents can modulate chemokine activity, their therapeutic potential is limited by poor corneal penetration and short residence time. This creates an opportunity for **nanocarrier systems** to enhance their delivery and efficacy.

Lipid-Based Nanocarriers: Rationale for Ocular Use

The limitations of conventional formulations have driven the development of lipid-based nanocarriers for ocular drug delivery. These carriers are biodegradable, biocompatible, and capable of encapsulating both hydrophilic and lipophilic drugs, while offering controlled or sustained release [18] [19] [20] [21] [22] [23] [24] [25]. Their nanoscale size improves corneal penetration, and surface modifications (e.g., positive charge) enhance muco-adhesion, increasing residence time on the corneal surface [19] [21].

Major types include:

- **Liposomes** are spherical vesicles with an internal aqueous core enclosed by a lipid bilayer membrane, capable of encapsulating both hydrophilic and hydrophobic drugs [24] [26].
- **Solid Lipid Nanoparticles (SLNs)** consist of a solid lipid matrix stabilized by emulsifiers, typically ranging from 50 to 1000 nm in size [18] [23]. They offer drug protection and a controlled release profile [23].
- **Nanostructured Lipid Carriers (NLCs)** are a newer generation of SLNs, formulated with a mixture of solid and liquid lipids to overcome limitations like low drug loading and drug expulsion, providing higher payloads and enhanced stability [23] [27] [28].
- **Nanoemulsions** are emulsions with ultrafine droplets, less than 1 μm in diameter, that have shown promise in delivering drugs to ocular tissues, with examples like Restasis® and Ikervis® successfully marketed [18].
- **Niosomes** are another category of nanovesicular systems used for ocular drug delivery [18].

Compared to traditional eye drops, lipid nanocarriers protect drugs from enzymatic degradation, enhance corneal permeation, and reduce dosing frequency, ultimately improving patient compliance and minimizing systemic side effects [18] [20].

Solid Lipid Nanoparticles (SLNs) Preparation: Hot Homogenization

Hot homogenization is a common technique for preparing solid lipid nanoparticles (SLNs), involving the dispersion of a melted lipid-drug mixture into a hot aqueous surfactant solution [29]. The process typically includes melting the lipid with the drug, preparing a heated aqueous surfactant phase, then combining and subjecting them to high-shear homogenization, followed by high-pressure homogenization, and finally cooling to solidify the lipid [29]. This differs from cold homogenization, which processes the lipid at or below its melting point, often after rapid solidification [29].

Recent studies highlight SLNs' potential for ocular drug delivery. For instance, Bimatoprost-loaded SLNs for glaucoma, prepared by solvent evaporation/ultrasonication (a hot process), showed an average size of 183.3 nm, 71.8% encapsulation efficiency, sustained drug release for 12 hours, and enhanced corneal permeation [30]. In another study, ketoprofen (an anti-inflammatory agent) was encapsulated in SLNs via an acoustic cavitation-assisted hot melt mixing method, yielding particles under 250 nm with nearly 90% encapsulation efficiency and 72-hour sustained release [31].

SLN properties are influenced by lipid type (e.g., glyceryl monostearate, Compritol 888 ATO), surfactant (e.g., poloxamer 407, Gelucire 50/13), homogenization pressure/cycles, and cooling methods [30] [31]. While hot homogenization offers good scalability and reproducibility

for industrial production, and SLNs can be sterilized, challenges include potential drug expulsion during storage and stability issues [29] [32] [33] [23] [34].

Despite these, SLNs prepared by hot homogenization are promising carriers for anti-inflammatory agents targeting ocular inflammation, including those involved in chemokine signaling pathways [32]. Their biocompatibility, sustained release, and enhanced permeation contribute to improved therapeutic efficacy [32] [35].

Material Required

Chemicals and Reagents

Distilled water (prepared at the PTC Laboratory, Institute of Sciences, Sage University, Indore) was used throughout the study. Ethanol (denatured ethanol 99.9%; Changshu Song Sheng Fine Chemical, China), cholesterol (Sunchem, India), glycerol (Sunchem, India), and Tween 80 (for synthesis; Sunchem, India) were employed as solvents and stabilizers.

The following anti-inflammatory drugs were used as model agents: **ibuprofen** (Cipla Ltd., India), **aspirin** (USV Pvt. Ltd., Mumbai, India), **diclofenac** (Ranbaxy, India), and **doxycycline** (USV Pvt. Ltd., Mumbai, India). All drugs were obtained from a local medical store.

All reagents were of analytical grade and used as received without further purification.

Instruments and Apparatus

The instruments employed in the study included a **magnetic stirrer with hot plate** (LAFCO India Optik, India), a **refrigerated centrifuge** (Model LRC-01, Lyzer, India), a **UV–Vis spectrophotometer** (Lyzer, India), and an **ultrasonicator** (Acutech, India).

All instruments and chemicals were provided by the Institute of Sciences, Sage University, Indore.

Methods

A. Preparation of Lipophilic Drug-Loaded Nanocarriers

Lipid-based nanocarriers for lipophilic drugs (ibuprofen, aspirin, and diclofenac) were prepared using a hot homogenization–ultrasonication method. Briefly, an aqueous phase consisting of Tween 80 (2.5 mL) and propylene glycol (5 mL) in distilled water (100 mL) was heated to 70 °C and stirred at 500–550 rpm on a magnetic stirrer with hot plate for 1 h. In parallel, the lipid phase was prepared by dissolving cholesterol (0.75 mg) in ethanol (20 mL) at 70 °C. The lipophilic drug was incorporated into this lipid phase under continuous heating. The drug–lipid mixture was added dropwise to the aqueous surfactant solution at 70 °C while stirring, followed by continuous agitation for 30–40 min until a homogeneous milky pre-emulsion was formed. The dispersion was rapidly cooled by immersing the beaker in an ice–water bath (4–5 °C). This was followed by ultrasonication for 30–40 min to reduce particle size and ensure uniformity.

For analysis, 15 mL of the dispersion was centrifuged at 15,000 rpm for 40 min at 4 °C. The supernatant was collected and analysed using a UV–Vis spectrophotometer with the corresponding placebo formulation as blank. The pellet was washed with distilled water,

redispersed in ethanol using a vortex shaker, and subsequently re-examined using UV–Vis spectrophotometry.

B. Preparation of Hydrophilic Drug-Loaded Nanocarriers

For hydrophilic drug (doxycycline) loading, the drug was dissolved in distilled water prior to encapsulation. The solution was then added dropwise to the lipid phase maintained at 70 °C to form a water-in-oil (w/o) emulsion. This primary emulsion was subsequently added dropwise to the aqueous surfactant solution maintained at 70 °C to form a water-in-oil-in-water (w/o/w) emulsion. The subsequent cooling, ultrasonication, and analytical procedures were carried out as described for the lipophilic drug formulations.

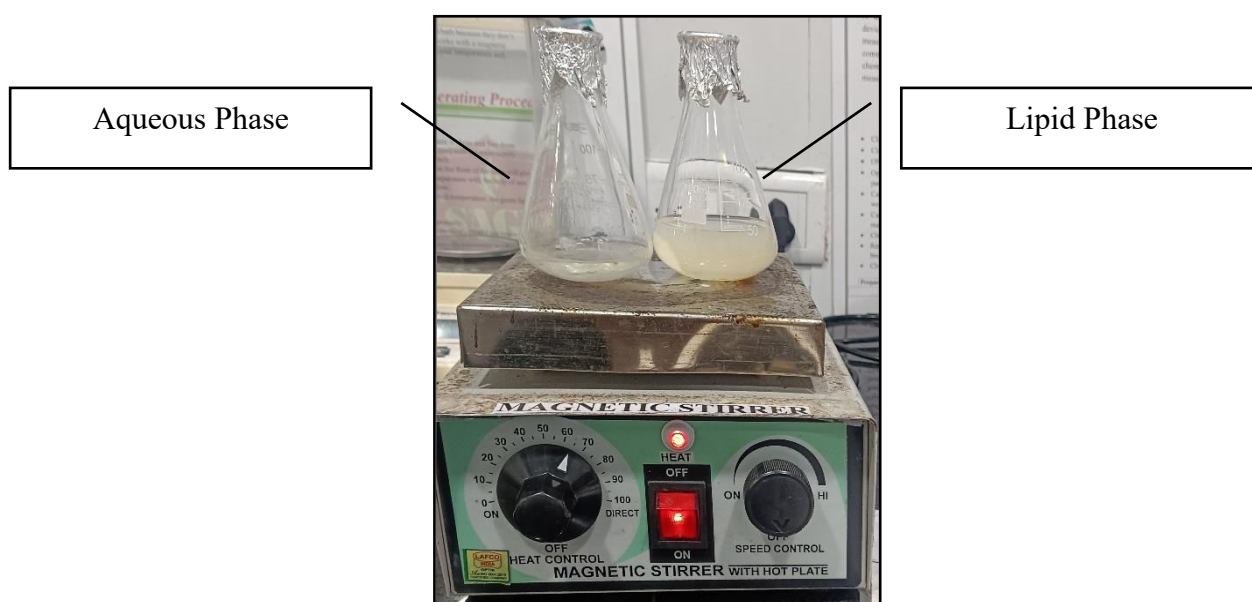
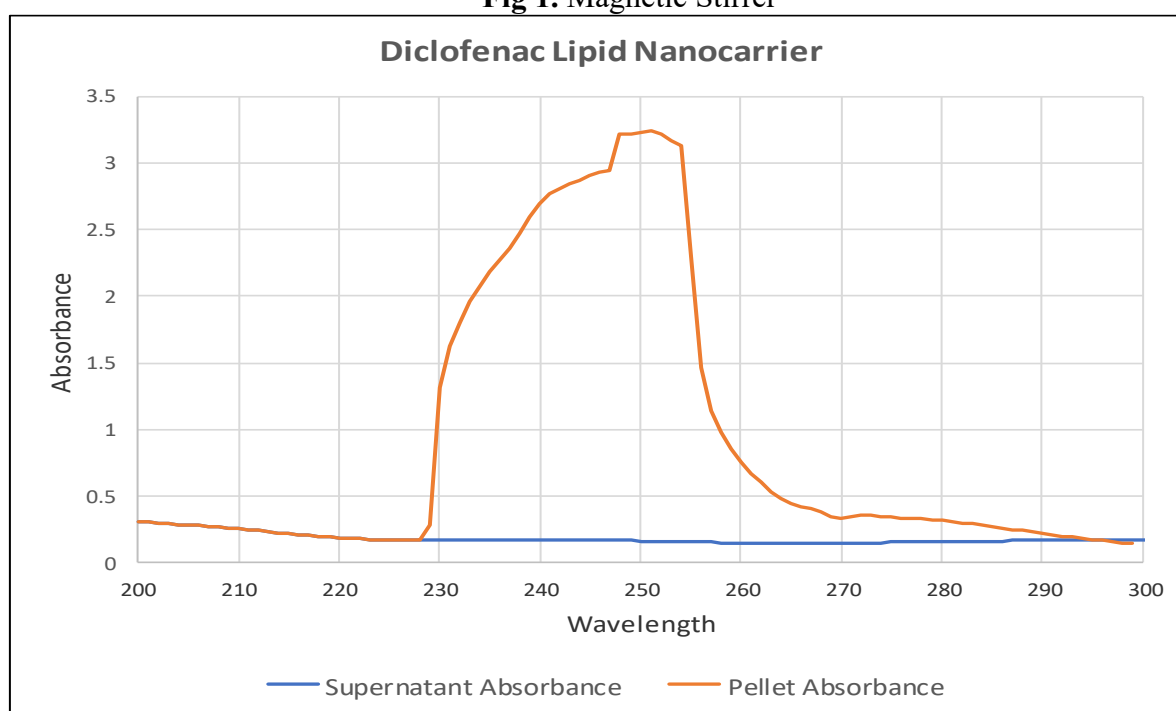
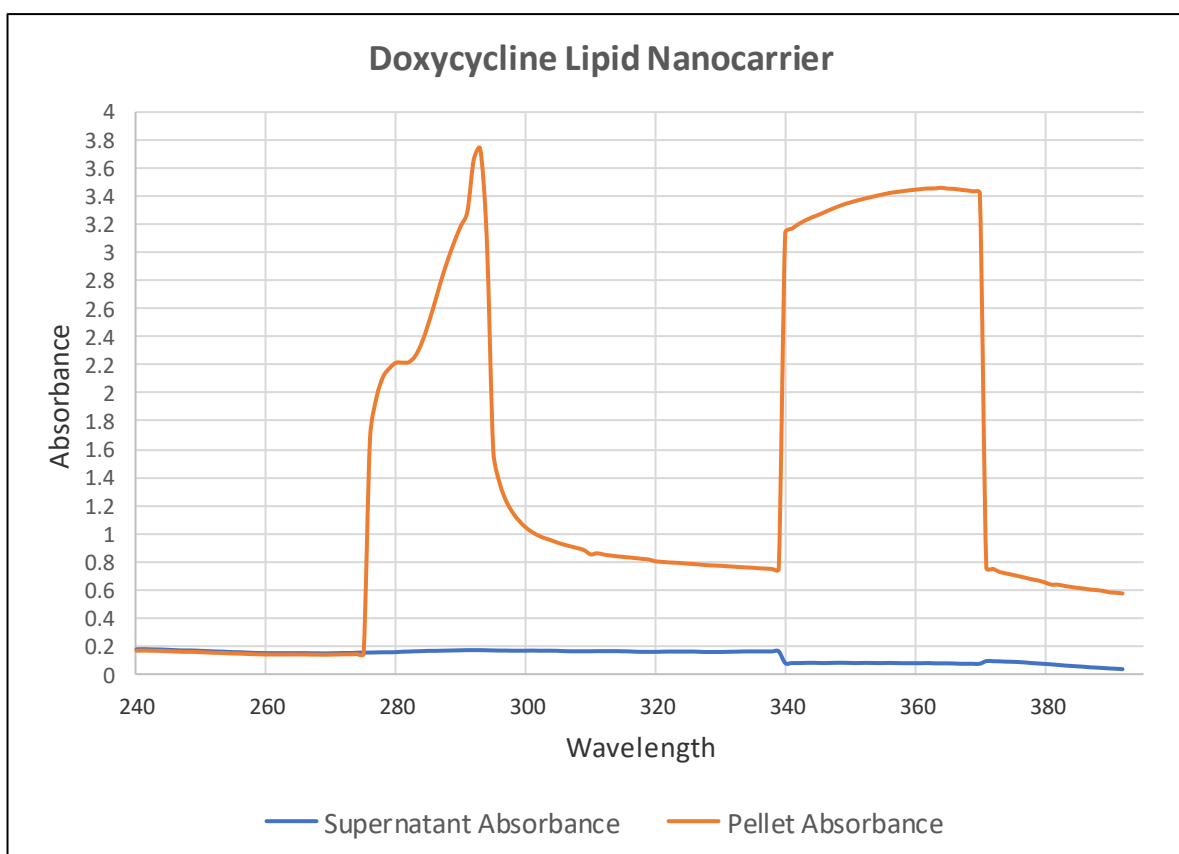
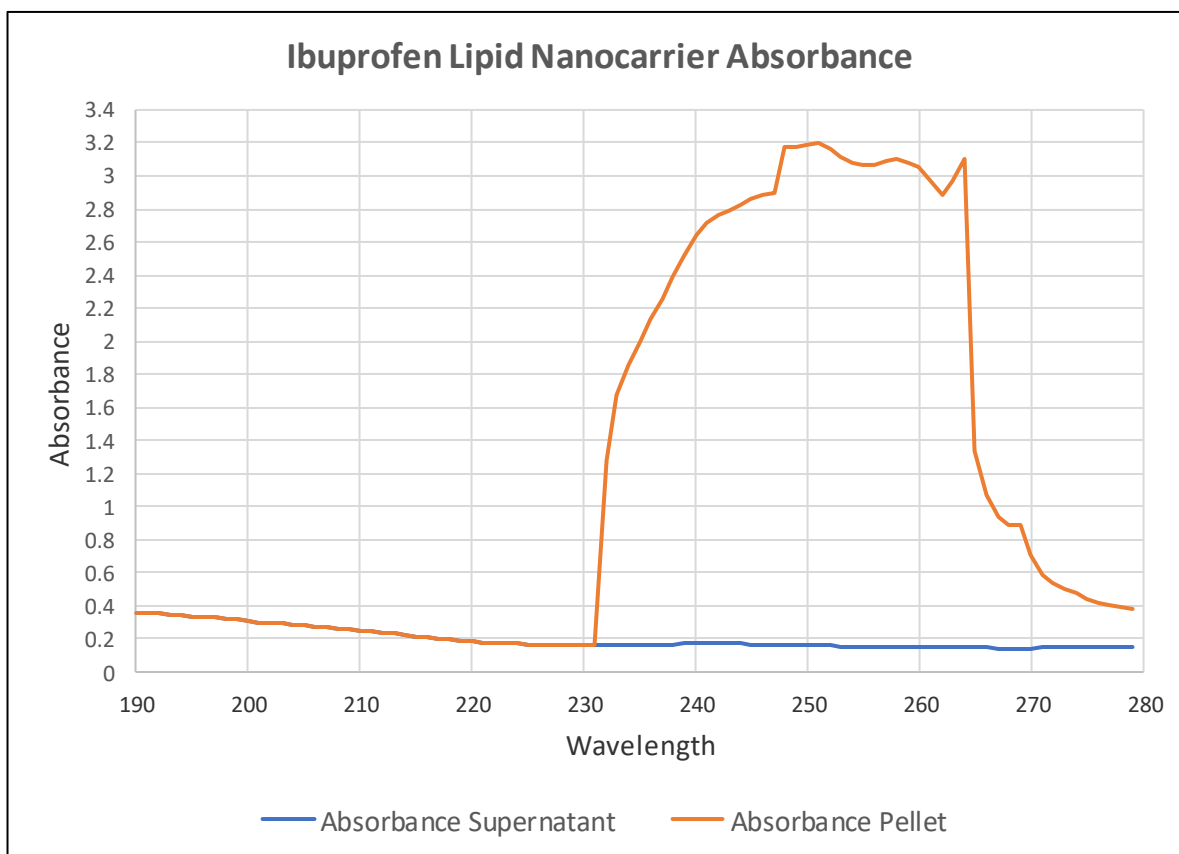
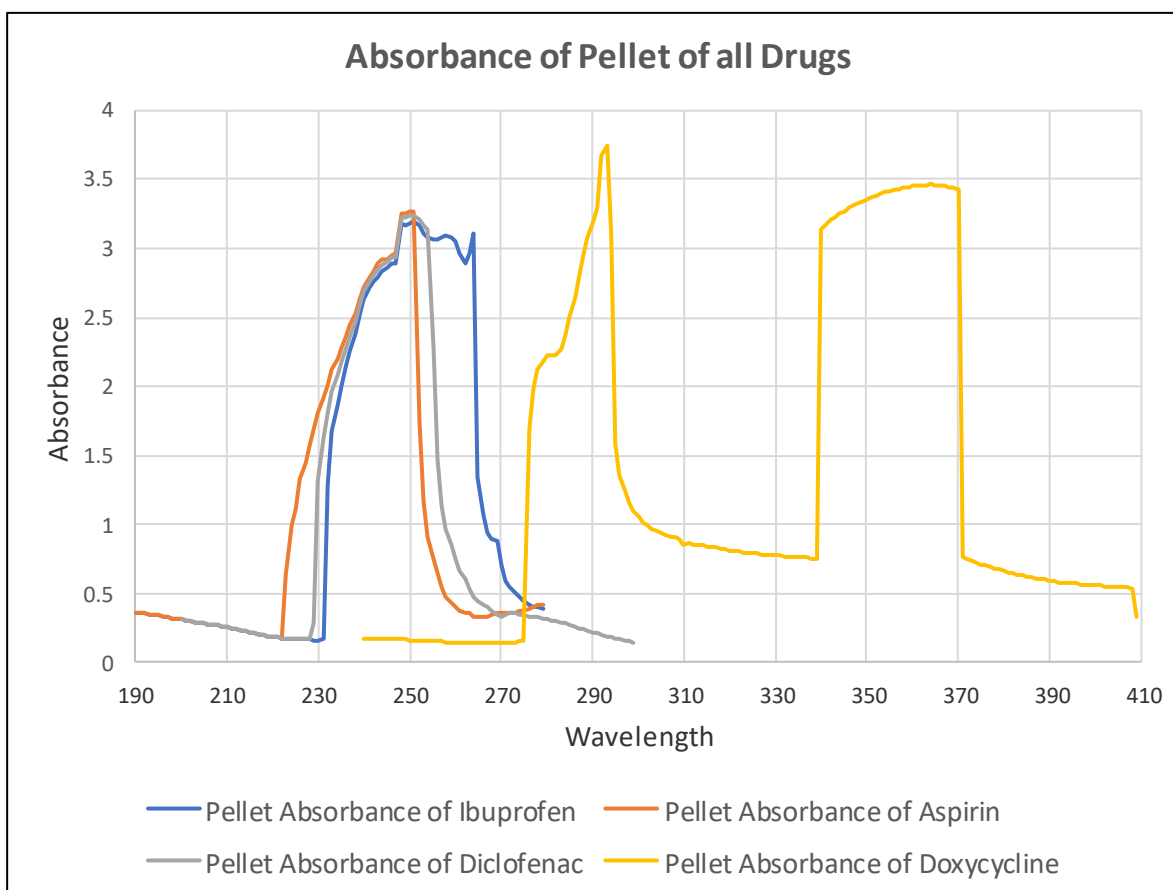


Fig 1. Magnetic Stirrer







Result and Discussion

Ocular inflammation is strongly driven by chemokine-mediated leukocyte recruitment, making chemokine signalling an attractive therapeutic target. Conventional drugs like NSAIDs and doxycycline can modulate these pathways, but their efficacy is hampered by the eye's protective barriers. Lipid-based nanocarriers, particularly SLNs prepared via hot homogenization, offer a promising solution by improving corneal penetration, sustaining drug release, and enhancing therapeutic outcomes.

1. Physical Appearance of the Formulation

The freshly prepared lipid nanocarrier formulations appeared **milky white and slightly viscous**, which is consistent with the formation of lipid-based colloidal systems. After centrifugation at 15,000 rpm for 40 min at 4 °C, the formulations separated into two distinct phases: a **clear supernatant** and a **whitish pellet**. The clear appearance of the supernatant suggested successful removal of untrapped drug, while the presence of a solid pellet indicated effective drug incorporation into the lipid matrix. Upon redispersion of the pellet in ethanol, the solution exhibited a **colloidal appearance**, confirming that the pellet comprised lipid-associated drug molecules. These physical observations support the formation of nanoscale lipid–drug complexes.

2. pH Evaluation

The measured pH of the prepared formulations was within the **neutral range of 6–8**. This is physiologically relevant for ocular applications, as corneal tissues are highly sensitive to pH variations. A neutral pH not only ensures **biocompatibility and reduced irritation** but also

enhances the stability of encapsulated anti-inflammatory drugs, which are prone to hydrolysis or degradation under acidic or alkaline conditions. Thus, the formulations demonstrated suitable **pH compatibility for corneal drug delivery**.

3. UV–Vis Absorbance Spectral Analysis

UV–Vis spectroscopy was employed to characterize drug entrapment in lipid nanocarriers. Each drug exhibited distinct **absorption maxima (λ max)** corresponding to its molecular structure:

- **Ibuprofen:** Strong absorption in the **230–260 nm** region, with the pellet showing sharp peaks around **3.1 AU**, confirming substantial drug incorporation.
- **Aspirin:** Displayed a λ max around **230–250 nm**, with the pellet showing enhanced absorbance compared to the supernatant, peaking above **3 AU**.
- **Diclofenac:** Characterized by a sharp rise in absorbance between **229–250 nm**, with pellet values reaching **3.2 AU**, confirming high entrapment.
- **Doxycycline:** Unique spectral profile with multiple absorption bands ranging from **270–370 nm**, and pellet absorbance exceeding **3.4 AU**, suggesting strong retention within lipid nanocarriers.

The **supernatants** in all cases showed relatively low absorbance (<0.3 AU across most wavelengths), indicating that the majority of drug molecules were successfully entrapped within the lipid nanocarrier system.

4. Supernatant vs. Pellet Absorbance

A direct comparison between the **supernatant** and **pellet** spectra provided insights into **entrapment efficiency**. For all four drugs, the supernatant absorbance was minimal, suggesting **negligible free drug presence** in the aqueous phase. In contrast, the pellet spectra consistently exhibited intense absorbance peaks, confirming successful encapsulation of drugs in the lipid phase. This result validates the ability of lipid nanocarriers to efficiently sequester lipophilic and amphiphilic drugs, minimizing wastage and enhancing loading efficiency.

5. Drug-Specific Entrapment Behaviour

The entrapment efficiency and spectral patterns varied across drugs, reflecting their **physicochemical properties**:

- **Ibuprofen and Diclofenac:** Both are highly **lipophilic NSAIDs**; therefore, they exhibited **excellent entrapment**, as reflected by high pellet absorbance and low supernatant absorbance. Their compatibility with the lipid phase accounts for stable incorporation.
- **Aspirin:** Being slightly more hydrophilic than ibuprofen or diclofenac, aspirin showed **moderate but significant entrapment**, evident from a clear difference in pellet vs. supernatant absorbance.
- **Doxycycline:** A tetracycline-class antibiotic with **amphiphilic character**, doxycycline displayed strong pellet absorbance across a broader spectrum (270–370 nm), suggesting robust interaction with lipid bilayers. However, its extended absorption range indicates a more complex interaction profile, possibly due to hydrogen bonding with lipid head groups.

6. Colloidal Stability of Redispersed Pellets

Redispersion of the pellet in ethanol resulted in a stable colloidal-like suspension, characterized by homogeneity and absence of precipitation. This indicates that the lipid nanocarrier system maintained structural integrity post-centrifugation. Colloidal stability is crucial for ocular drug delivery, as it ensures uniform dosing, improved corneal penetration, and reduced risk of aggregation during storage or administration.

7. Comparison with Literature Reports

The λ_{max} values observed for each drug were consistent with reported UV–Vis absorption spectra in the literature:

- Ibuprofen and aspirin typically show strong absorbance in the **230–260 nm** range.
- Diclofenac exhibits absorption peaks near **275 nm**, but encapsulation shifted intensity to shorter wavelengths.
- Doxycycline is known to have multiple λ_{max} values (270–350 nm), consistent with our results.

These findings confirm that the drugs retained their **spectral identity post-encapsulation**, while also validating the analytical approach used.

8. Overall Encapsulation Efficiency and Suitability

The strong absorbance values in pellet fractions across all drugs confirm successful entrapment within lipid nanocarriers. Among the tested drugs, diclofenac and doxycycline showed the most pronounced pellet absorbance, suggesting higher entrapment efficiency, whereas aspirin and ibuprofen demonstrated moderate but satisfactory encapsulation. The results indicate that lipid nanocarriers are particularly well-suited for lipophilic and amphiphilic drugs, making them ideal candidates for enhancing corneal penetration and sustained release of anti-inflammatory agents.

9. Implications for Ocular Drug Delivery

The results highlight the therapeutic potential of lipid nanocarriers for ocular delivery. The neutral pH, strong drug entrapment, and colloidal stability make these formulations highly compatible for corneal administration. Enhanced encapsulation efficiency is particularly significant for chemokine signaling-targeting anti-inflammatory agents, as improved penetration and sustained release are essential for mitigating inflammation in ocular tissues. These findings form a strong foundation for further optimization of nanocarrier formulations, including particle size distribution, zeta potential, and in vitro release kinetics, to validate their clinical applicability.

References

1. Kawashima, H. (2003). Chemokines – their role in immunotherapy for intraocular inflammation. *Ocular Immunology and Inflammation*, 11(2), 83–90. <https://doi.org/10.1076/ocii.11.2.83.15917>
2. Carreño, E., Enríquez-de-Salamanca, A., Tesón, M., García-Vázquez, C., Stern, M.E., Whitcup, S.M. and Calonge, M. (2010), Cytokine and chemokine levels in tears from healthy subjects. *Acta Ophthalmologica*, 88: e250-e258. <https://doi.org/10.1111/j.1755-3768.2010.01978.x>

3. Wallace, G. R., John Curnow, S., Wloka, K., Salmon, M., & Murray, P. I. (2004). The role of chemokines and their receptors in ocular disease. *Progress in Retinal and Eye Research*, 23(4), 435–448. <https://doi.org/10.1016/j.preteyeres.2004.04.004>
4. Enríquez-de-Salamanca, A., & Calonge, M. (2008). Cytokines and chemokines in immune-based ocular surface inflammation. *Expert Review of Clinical Immunology*, 4(4), 457–467. <https://doi.org/10.1586/1744666X.4.4.457>
5. Furusato, E., Shen, D., Cao, X., Furusato, B., Nussenblatt, R. B., Rushing, E. J., & Chan, C. C. (2011). Inflammatory cytokine and chemokine expression in sympathetic ophthalmia: a pilot study. *Histology and histopathology*, 26(9), 1145–1151. <https://doi.org/10.14670/HH-26.1145>
6. Wang, C. (2022). Drugs for retinal disease that are non-steroidal anti-inflammatory. *Acta Clinical and Translational Vision Research*, 12(5), 94–96. [https://doi.org/10.37532/ACTVR.2022.12\(5\).94-96](https://doi.org/10.37532/ACTVR.2022.12(5).94-96)
7. Yong Woo Ji, Yuri Seo, Wungrak Choi, Areum Yeo, Hyemi Noh, Eung Kweon Kim, Hyung Keun Lee; Dry Eye-Induced CCR7⁺CD11b⁺ Cell Lymph Node Homing Is Induced by COX-2 Activities. *Invest. Ophthalmol. Vis. Sci.* 2014;55(10):6829-6838. <https://doi.org/10.1167/iovs.14-14744>.
8. Zhang, PW., Wan, ZH., Li, W. *et al.* Ibuprofen reduces inflammation, necroptosis and protects photoreceptors from light-induced retinal degeneration. *J Neuroinflammation* **22**, 20 (2025). <https://doi.org/10.1186/s12974-024-03329-8>
9. De Paiva, C. S., Corrales, R. M., Villarreal, A. L., Farley, W. J., Li, D.-Q., Stern, M. E., & Pflugfelder, S. C. (2006). Corticosteroid and doxycycline suppress MMP-9 and inflammatory cytokine expression, MAPK activation in the corneal epithelium in experimental dry eye. *Experimental Eye Research*, 83(3), 526–535. <https://doi.org/10.1016/j.exer.2006.02.004>
10. Warcoin E, Baudouin C, Gard C, BrignoleBaudouin F (2016) In Vitro Inhibition of NFAT5-Mediated Induction of CCL2 in Hyperosmotic Conditions by Cyclosporine and Dexamethasone on Human HeLa-Modified Conjunctiva-Derived Cells. *PLoS ONE* 11(8): e0159983. <https://doi.org/10.1371/journal.pone.0159983>
11. Agrahari, V., Mandal, A., Agrahari, V., Trinh, H. M., Joseph, M., Ray, A., Hadji, H., Mitra, R., Pal, D., & Mitra, A. K. (2016). A comprehensive insight on ocular pharmacokinetics. *Drug delivery and translational research*, 6(6), 735–754. <https://doi.org/10.1007/s13346-016-0339-2>
12. Pharmacy180. (n.d.). *Ocular drug delivery*. Pharmacy180. Retrieved September 6, 2025, from <https://www.pharmacy180.com/article/ocular-drug-delivery-2795/>
13. Baranowski, Przemysław, Karolewicz, Bożena, Gajda, Maciej, Pluta, Janusz, Ophthalmic Drug Dosage Forms: Characterisation and Research Methods, *The Scientific World Journal*, 2014, 861904, 14 pages, 2014. <https://doi.org/10.1155/2014/861904>
14. Kang, Y. K., Im, J. C., Shin, J. P., Kim, I. T., & Park, D. H. (2017). Short-term analysis of the residual volume of an eye drop following 23-gauge microincision vitrectomy surgery. *Korean Journal of Ophthalmology*, 31(2), 138–143. <https://doi.org/10.3341/kjo.2016.0090>
15. Abdelkader, H., & Alany, R. G. (2012). Controlled and continuous release ocular drug delivery systems: Pros and cons. *Current Drug Delivery*, 9(4), 389–402. <https://doi.org/10.2174/156720112801323125>
16. Lee, R., & Rein, A. (n.d.). *How to enhance the solubility of ocular drug products*. Agno Pharma. Retrieved September 6, 2025, from <https://agnopharma.com/blog/increase-the-solubility-of-ocular-drugs/>
17. Sebbag L, Kirner NS, Wulf LW and Mochel JP (2020) Tear Film Pharmacokinetics and Systemic Absorption Following Topical Administration of 1% Prednisolone Acetate Ophthalmic Suspension in Dogs. *Front. Vet. Sci.* 7:571350. <https://doi.org/10.3389/fvets.2020.571350>
18. Das, B., Nayak, A. K., & Mallick, S. (2022). Lipid-based nanocarriers for ocular drug delivery: An updated review. *Journal of Drug Delivery Science and Technology*, 76, 103780. <https://doi.org/10.1016/j.jddst.2022.103780>
19. Baig MS, Karade SK, Ahmad A, Khan MA, Haque A, Webster TJ, Faiyazuddin M and Al-Qahtani NH (2024) Lipid-based nanoparticles: innovations in ocular drug delivery. *Front. Mol. Biosci.* 11:1421959. <https://doi.org/10.3389/fmolb.2024.1421959>

20. Jacob, S., Nair, A. B., Shah, J., Gupta, S., Boddu, S. H. S., Sreeharsha, N., Joseph, A., Shinu, P., & Morsy, M. A. (2022). Lipid Nanoparticles as a Promising Drug Delivery Carrier for Topical Ocular Therapy—An Overview on Recent Advances. *Pharmaceutics*, 14(3), 533. <https://doi.org/10.3390/pharmaceutics14030533>
21. Bonilla, L., Espina, M., Severino, P., Cano, A., Ettcheto, M., Camins, A., García, M. L., Souto, E. B., & Sánchez-López, E. (2022). Lipid Nanoparticles for the Posterior Eye Segment. *Pharmaceutics*, 14(1), 90. <https://doi.org/10.3390/pharmaceutics14010090>
22. Gugleva, V., & Andonova, V. (2023). Recent Progress of Solid Lipid Nanoparticles and Nanostructured Lipid Carriers as Ocular Drug Delivery Platforms. *Pharmaceutics*, 16(3), 474. <https://doi.org/10.3390/ph16030474>
23. Nanda, N., Tharmaraj, C., Panigrahi, S., & Rani, M. S. (2024). Solid lipid nanoparticles and nanostructured lipid carrier: A novel approach for lipid-based drug delivery system. *International Journal of Drug Delivery Technology*, 14(02), 1232–1238. <https://doi.org/10.25258/ijddt.14.2.90>
24. Naseri, N., Valizadeh, H., & Zakeri-Milani, P. (2015). Solid lipid nanoparticles and nanostructured lipid carriers: Structure, preparation and application. *Advanced Pharmaceutical Bulletin*, 5(3), 305–313. <https://doi.org/10.15171/apb.2015.043>
25. Sudha, A., & Sinha, J. (n.d.). A review on SLN and NLC nanoparticles, their applications, and recent advancements. Retrieved 8 September 2025, from <https://juniperpublishers.com/jrann/pdf/JRANN.MS.ID.555556.pdf>
26. López-Cano, J. J., González-Cela-Casamayor, M. A., Andrés-Guerrero, V., Herrero-Vanrell, R., & Molina-Martínez, I. T. (2021). Liposomes as vehicles for topical ophthalmic drug delivery and ocular surface protection. *Expert opinion on drug delivery*, 18(7), 819–847. <https://doi.org/10.1080/17425247.2021.1872542>
27. Pal, S., Prajapati, R. N., Bijauliya, R. K., Sonam, & Singh, B. (2020). Nanostructure lipid carrier (NLC) – An overview. *International Journal of Pharmacology and Pharmaceutical Research*, 18(2), 368–375. <https://ijppr.humanjournals.com/wp-content/uploads/2020/04/43.Shishu-Pal-Ram-Narayan-Prajapati-Rohit-Kumar-Bijauliya-Sonam-Balbir-Singh.pdf>
28. Chinthaginjala, H., Bogavalli, V., Hindustan, A. A., Pathakamuri, J., Pullaganti, S. S., Gowni, A., & Baktha, B. (2023). Nanostructured lipid carriers: A potential era of drug delivery systems. *Indian Journal of Pharmaceutical Education*, 58(1), 21–33. <https://doi.org/10.5530/ijper.58.1.3>
29. Deepa MK, Karthikeyan M*, Abhay AD, Shruti PC, Omkar SD, Satish RC. Comprehensive Review on Solid Lipid Nanoparticles. *Ann Pharmacol Pharm*. 2020; 5(4): 1190.
30. Satyanarayana, S. D., Abu Lila, A. S., Moin, A., Moglad, E. H., Khafagy, E.-S., Alotaibi, H. F., Obaidullah, A. J., & Charyulu, R. N. (2023). Ocular Delivery of Bimatoprost-Loaded Solid Lipid Nanoparticles for Effective Management of Glaucoma. *Pharmaceutics*, 16(7), 1001. <https://doi.org/10.3390/ph16071001>
31. Kumar, R., Singh, A., & Garg, N. (2019). Acoustic Cavitation-Assisted Formulation of Solid Lipid Nanoparticles using Different Stabilizers. *ACS Omega*, 4(8), 13360–13370. <https://doi.org/10.1021/acsomega.9b01532>
32. Ana, R.d.; Fonseca, J.; Karczewski, J.; Silva, A.M.; Zielińska, A.; Souto, E.B. Lipid-Based Nanoparticulate Systems for the Ocular Delivery of Bioactives with Anti-Inflammatory Properties. *Int. J. Mol. Sci.* 2022, 23, 12102. <https://doi.org/10.3390/ijms232012102>
33. Prabhakaran, Ekambaram & Hasan, A & Karunanidhi, Priyanka. (2011). Solid lipid nanoparticles: A review. 2.
34. Vadlamudi Dr, Harini Chowdary & Teja, V & Yalavarthi, Prasanna & Surendra, N. & Vardhan, R. & Reddy, B.. (2014). A GLIMPSE ON SOLID LIPID NANOPARTICLES AS DRUG DELIVERY SYSTEMS. *Journal of Global Trends in Pharmaceutical Sciences* ISSN NO:2230-7346. 5. 1649-1657.
35. Solid lipid nanoparticles; as a promising drug delivery method to get greater bioavailability: A review. (2023). *World Journal of Biology Pharmacy and Health Sciences*, 14(2), 072–080. <https://doi.org/10.30574/wjbphs.2023.14.2.0104>

54. Green and Sustainable Chemistry

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Abstract:-Green and sustainable chemistry is an interdisciplinary approach that focuses on the design of chemical products and processes that minimize environmental impact, reduce waste, and conserve natural resources while maintaining economic and social benefits. It emphasizes the prevention of pollution at the molecular level through the use of safer raw materials, renewable feedstocks, energy-efficient processes, and environmentally benign solvents and catalysts. By integrating the principles of green chemistry, sustainable chemistry seeks to balance chemical innovation with long-term environmental stewardship and human health protection. Advances in this field have led to the development of cleaner synthesis routes, biodegradable materials, and alternative energy solutions, contributing significantly to sustainable industrial practices. Overall, green and sustainable chemistry plays a crucial role in addressing global challenges such as climate change, resource depletion, and environmental pollution, while supporting sustainable development and a circular economy.

Key words:- Green chemistry, Waste reduction, Renewable feedstocks, Circular economy, Resource depletion.

Introduction

Chemistry has traditionally prioritized yield and efficiency, often at the expense of environmental protection and human health. In response to growing global concerns related to pollution, hazardous waste, and depletion of natural resources, a paradigm shift toward green chemistry, also known as sustainable chemistry, has emerged (*Anastas and Warner, 1998*). Green chemistry emphasizes the design of chemical products and processes that prevent pollution at its source rather than managing it after formation, thereby promoting sustainable industrial practices and minimizing environmental harm (*Anastas and Eghbali, 2010*).

This approach focuses on the development of safer chemicals, the use of renewable raw materials, energy-efficient processes, and waste minimization throughout the entire life cycle of chemical products—from raw material extraction to manufacture, use, and disposal (*Anastas and Warner, 1998*). By applying principles such as atom economy, catalysis, the use of non-toxic reagents, and biodegradable materials, green chemistry significantly reduces environmental impact while maintaining process efficiency (*Anastas and Warner, 1998; Anastas and Eghbali, 2010*).

Green and sustainable chemistry plays a critical role in addressing major global challenges, including climate change, environmental pollution, resource depletion, and risks to human health. It supports sustainable industrial growth by fostering innovation that balances environmental protection, social responsibility, and economic development. Consequently, green chemistry has become an essential component of modern chemical research, education, and industrial practice (*Sheldon, 2017*).

Historical Background and Development

Historically, chemical science focused primarily on yield, purity, and process efficiency, with limited consideration of environmental or health consequences (*Anastas and Warner, 1998*). Public awareness regarding chemical pollution increased significantly during the 1960s due to environmental movements and influential publications such as *Silent Spring* by Rachel Carson, which highlighted the harmful effects of chemical misuse (*Carson, 1962*).

The formal framework of green chemistry was established in the 1990s with the development of the **12 principles of green chemistry**, proposed by Paul Anastas and John Warner, which now serve as the foundation of the field (*Anastas and Warner, 1998*). Since then, green chemistry has evolved through contributions from both academic research and industrial innovation, particularly in the areas of catalysis, solvent replacement, waste reduction, and the utilization of renewable resources (*Sheldon, 2017*). Its growth aligns closely with the broader global movement toward sustainability in science and technology.

Principles of Green and Sustainable Chemistry

The **12 fundamental principles of green chemistry**, introduced by Anastas and Warner, guide the design and implementation of environmentally responsible chemical processes (*Anastas and Warner, 1998*). These principles emphasize waste prevention, improved atom economy, reduced toxicity, safer solvents, energy efficiency, use of renewable feedstocks, and catalysis, among others. Collectively, they aim to minimize environmental impact while enhancing safety and economic viability (*Anastas and Eghbali, 2010*).

By integrating these principles into chemical research and industrial processes, green chemistry promotes sustainable development without compromising performance or innovation (*Sheldon, 2017*).

Modern Methodologies in Green Chemistry

1 Catalysis and Energy-Efficient Techniques

Catalysis is central to sustainable chemical practices, as it allows reactions to proceed under milder conditions with higher selectivity and reduced waste generation (Anastas & Warner, 1998). The use of heterogeneous catalysts, biocatalysts, and advanced catalytic systems has significantly improved reaction efficiency while minimizing byproduct formation (Sheldon & Woodley, 2018).

Energy-efficient techniques such as microwave-assisted synthesis and sonochemistry have gained importance due to their ability to reduce reaction times and energy consumption (Kappe, 2004; Loupy, 2006). Recent catalytic systems developed for oxidation and reduction reactions employ greener reaction media and generate fewer toxic byproducts, supporting the principles of atom economy and energy efficiency (Li & Trost, 2008; Anastas & Eghbali, 2010).

2 Solvent Replacement and Alternative Media

Traditional volatile organic solvents pose environmental and health risks due to their toxicity and volatility. Green chemistry promotes the replacement of such solvents with

environmentally benign alternatives including water, supercritical carbon dioxide, ionic liquids, deep eutectic solvents, and bio-based solvents (Poliakoff et al., 2011).

Ionic liquids and deep eutectic solvents, in particular, have demonstrated enhanced catalytic performance and improved safety profiles in various chemical reactions (Plechko & Seddon, 2008; Abbott et al., 2003). In addition, solvent-free methodologies have proven effective in certain synthetic applications, significantly reducing liquid waste and simplifying purification processes (Li & Trost, 2008).

3 Renewable Feedstocks and Biomass Valorization

A major advancement in sustainable chemistry is the transition from petrochemical-based feedstocks to renewable biomass-derived raw materials (Clark & Deswarte, 2015). Biomass valorization focuses on converting lignocellulosic waste, plant oils, and agricultural residues into valuable chemicals and fuels, thereby supporting a circular bioeconomy (Climent et al., 2014).

This approach enhances resource efficiency and reduces dependence on fossil resources, aligning chemical production with long-term sustainability goals (Keijer et al., 2019).

Applications in Industry and Research

1 Pharmaceuticals

Green chemistry principles are increasingly applied in pharmaceutical manufacturing to reduce solvent consumption, improve reaction selectivity, and minimize hazardous waste (Sheldon, 2017). The adoption of biocatalysis and process intensification techniques has helped pharmaceutical companies improve sustainability metrics such as E-factor and atom economy (Sheldon & Woodley, 2018).

These strategies not only lower environmental impact but also enhance economic efficiency in large-scale drug production (Anastas & Eghbali, 2010).

2 Materials Science and Polymers

In materials science, green chemistry has enabled the development of biodegradable and bio-based polymers that safely degrade in the environment (Gross & Kalra, 2002). Polymers synthesized from renewable plant-derived feedstocks exhibit reduced toxicity and lower carbon footprints compared to conventional plastics (Zhu et al., 2016). Such innovations contribute significantly to sustainable materials development and waste reduction.

3 Environmental Remediation

Green chemistry also plays a vital role in environmental remediation. Photocatalytic materials, such as titanium dioxide-based systems, can decompose toxic pollutants or convert industrial waste into useful chemicals under light irradiation (Fujishima et al., 2008).

These technologies reduce environmental pollution while offering sustainable solutions for waste management.

Challenges and Constraints

Despite its advantages, the implementation of green and sustainable chemistry faces several challenges. Economic barriers include the high initial cost of developing and validating green technologies (Poliakoff et al., 2011). Technical limitations arise because some green methods are not yet as efficient or broadly applicable as traditional chemical processes (Sheldon, 2017).

Additionally, regulatory and policy gaps limit widespread adoption, highlighting the need for stronger global frameworks and incentives (Anastas & Warner, 1998). Aligning green chemistry practices with social and economic contexts remains a continuing challenge.

Future Directions

Future research in green chemistry is expected to focus on AI-assisted process design to optimize reaction pathways and conditions (Segler et al., 2018). The integration of circular economy principles will further improve material lifecycle sustainability (Keijer et al., 2019).

Advances in nanocatalysts and biocatalytic systems are also anticipated to enhance reaction efficiency and selectivity (Sheldon & Woodley, 2018). Stronger collaboration among chemists, engineers, environmental scientists, and policymakers will be crucial for accelerating sustainable chemical innovation.

Conclusion

Green and sustainable chemistry represents a fundamental shift in chemical science, embedding environmental protection and sustainability into chemical design and industrial practice (Anastas & Warner, 1998). By addressing challenges such as pollution, resource depletion, and climate change, green chemistry offers practical and long-term solutions for a sustainable future. Continued research, interdisciplinary collaboration, and supportive policy measures will ensure its expanding impact across academia and industry.

References

- 1] Anastas, P. T., & Eghbali, N. (2010). Green chemistry: Principles and practice. *Chemical Society Reviews*, 39, 301–312.
<https://doi.org/10.1039/B918763B>
- 2] Anastas, P. T., & Warner, J. C. (1998). *Green Chemistry: Theory and Practice*. Oxford University Press, New York.
- 3] Abbott, A. P., Capper, G., Davies, D. L., Rasheed, R. K., & Tambyrajah, V. (2003). Novel solvent properties of choline chloride/urea mixtures. *Chemical Communications*, 1, 70–71.
<https://doi.org/10.1039/B210714G>
- 4] Clark, J. H., & Deswarte, F. E. I. (2015). *Introduction to Chemicals from Biomass*. Wiley, Chichester.
- 5] Climent, M. J., Corma, A., & Iborra, S. (2014). Conversion of biomass platform molecules into fuel additives and liquid hydrocarbon fuels. *Green Chemistry*, 16, 516–547.
<https://doi.org/10.1039/C3GC41492B>
- 6] Fujishima, A., Zhang, X., & Tryk, D. A. (2008). TiO₂ photocatalysis and related surface phenomena. *Surface Science Reports*, 63, 515–582.

- <https://doi.org/10.1016/j.surfrep.2008.10.001>
- 7] Gross, R. A., & Kalra, B. (2002). Biodegradable polymers for the environment. *Science*, 297, 803–807.
<https://doi.org/10.1126/science.297.5582.803>
- 8] Kappe, C. O. (2004). Controlled microwave heating in modern organic synthesis. *Angewandte Chemie International Edition*, 43, 6250–6284.
<https://doi.org/10.1002/anie.200400655>
- 9] Keijer, T., Bakker, V., & Slootweg, J. C. (2019). Circular chemistry: An emerging concept. *Nature Chemistry*, 11, 190–195.
<https://doi.org/10.1038/s41557-019-0226-9>
- 10] Li, C. J., & Trost, B. M. (2008). Green chemistry for chemical synthesis. *Proceedings of the National Academy of Sciences*, 105, 13197–13202.
<https://doi.org/10.1073/pnas.0804348105>
- 11] Loupy, A. (2006). *Microwaves in Organic Synthesis*. Wiley-VCH, Weinheim.
- 12] Plechkova, N. V., & Seddon, K. R. (2008). Applications of ionic liquids in the chemical industry. *Chemical Society Reviews*, 37, 123–150.
<https://doi.org/10.1039/B006677J>
- 13] Poliakoff, M., Licence, P., & George, M. W. (2011). Green chemistry: Science and politics of change. *Science*, 331, 807–810.
<https://doi.org/10.1126/science.1199316>
- 14] Segler, M. H. S., Preuss, M., & Waller, M. P. (2018). Planning chemical syntheses with deep neural networks. *Nature*, 555, 604–610.
<https://doi.org/10.1038/nature25978>
- 15] Sheldon, R. A. (2017). The E-factor 25 years on: The rise of green chemistry and sustainability. *Green Chemistry*, 19, 18–43.
<https://doi.org/10.1039/C6GC02157C>
- 16] Sheldon, R. A., & Woodley, J. M. (2018). Role of biocatalysis in sustainable chemistry. *Chemical Reviews*, 118, 801–838.
<https://doi.org/10.1021/acs.chemrev.7b00203>
- 17] Zhu, Y., Romain, C., & Williams, C. K. (2016). Sustainable polymers from renewable resources. *Nature*, 540, 354–362.
<https://doi.org/10.1038/nature21001>
- 18] Sheldon, R. A. (2017). The E-factor 25 years on: The rise of green chemistry and sustainability. *Green Chemistry*, 19, 18–43. <https://doi.org/10.1039/C6GC02157C>

55. Aloe Vera-Derived Silver Nanoparticles: A Green Route for Nanomaterial Synthesis

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Abstract: By using Aloe vera leaf extract as a stabilizing and dropping agent, the current study demonstrates a method for producing silver nanoparticles (AgNPs) in an environmentally friendly and sustainable manner. The phytochemicals in the extract efficiently changed Ag^+ ions into Ag_2 , which produced stable, uniformly distributed nanoparticles. A discernible color change from soft yellow to dusky brown indicated the early production of AgNPs. UV-visible spectroscopy demonstrated a prominent surface plasmon resonance (SPR) band at approximately 420–440 nm, confirming the successful production of silver nanoparticles. The unique peaks in the FTIR measurement of the hydroxyl, amine, and carbonyl functional groups demonstrated their active involvement in the reduction and overlaying processes. X-ray diffraction (XRD) patterns that displayed notable diffraction peaks indexed to the face-centered cubic (fcc) crystalline structure of metallic silver validated the crystalline nature and purity of the generated AgNPs. According to all the research, Aloe vera extract is a great biogenic mediator that provides a safe, affordable, and eco-friendly way to produce nanomaterials with potential applications in the fields of catalysis, biomedicine, and the environment.

Keywords: Green synthesis, eco-friendly and sustainable approach, silver nanoparticles, etc.

Introduction

In the fast-evolving subject of nanotechnology, researchers and engineers are actively designing, synthesizing, and manipulating materials at the nanoscale [1]. These hard workgoals to harness unique things and style functions for varied applications. Nanoparticles (NP), characterized by a size range of 1–100 nm [2], perform crucial roles in several industries, including medical [3], food science [4], agriculture [5], environmental remediation [6], adsorption [7], and energy [8]. Their unique physic-chemical properties, such as catalysis, increased electrical and thermal conductivity, adjustable light absorption, and capacity to scatter without aggregation, contribute to their higher enactment compared to their bigger counterparts. Metal nanoparticles (MNP) have gained significant attention due to their unusual physicochemical features, compact size, and consistent behaviour [9]. Exciting opportunities for MNP in targeted drug delivery [10], cancer treatment [11], optical devices [12], catalysis [13, 14], water treatment [15], and antimicrobial applications [16] are made possible by these special qualities. Silver nanoparticles (AgNP) are unique among metal nanoparticles, including Pd, Cu, Au, Zn, Sn, Ag, and Co, because of their wide range of applications in numerous industries [17]. AgNP synthesis dates back more than 120 years, and its history is extensive and well-documented. In 1889, Lea first described the production of a citrate-stabilized silver colloid, marking an initial breakthrough in nanoparticle research.

Synthesis of Silver nanoparticles

Three essential prerequisites for the preparation of NPs are the selection of a harmlessoothing material, an efficient reducing agent, and an environmentally friendly

solvent. Many synthetic techniques, including physical, chemical, and biosynthetic techniques, have been employed to generate NPs [18]. The chemical methods that are commonly employed are quite pricey and include the utilization of harmful and deadly ingredients that provide many environmental concerns. The biosynthetic approach uses bacteria and plants to produce NPs for use in biomedical applications in an environmentally friendly, safe, and sustainable manner. This synthesis can be carried out by a variety of organisms, including bacteria, plants, fungus, and algae. Many NPs have been produced from plant parts such as leaves, fruits, roots, stems, and seeds because these plant parts possess phytochemicals that operate as stabilizing and reducing agents in the extract [19, 20]. The synthesis of nanoparticles can be broadly categorized into two approaches: top-down and bottom-up.

1 Top-down method

The top-down method to nanoparticle production combines a variety of physical and chemical approaches to convert bulk constituents into nanoparticles at the Nano scale. These technologies, relying on externally imposed services and actions, break down bulk things into smaller constituents, giving nanoparticles. Milling, lithography, and etching constitute prominent instances of this methodology. Mechanical powdering applies pressure on the bulk material, breaking it into small particles. In contrast, lithography uses a predetermined design on a substance and careful material abstraction to create nanoparticles. Although the top-down method offers exact control over the size and homogeneity of nanoparticles, its capacity to produce nanoparticles with a variety of characteristics may be restricted [21].

2 Bottom-up method

Unlike the "top-down" method, the "bottom-up" method builds nanoparticles by carefully assembling minor building blocks. This strategy encompasses two important subcategories: chemical and biological procedures [22]. Organic procedures, which frequently use reducing agents, surfactants, and stabilizers, mostly depend on the chemical reduction of precursor molecules. In this area, sol-gel, co-precipitation, and hydrothermal synthesis are well-known practices. On the other hand, biological synthesis provides a sustainable and environmentally friendly substitute by using natural resources, microorganisms, and plant sources as stabilizers and reducing mediators, which reduces the need for potentially dangerous chemicals.

3 Green synthesis

Green synthesis refers to ecologically benign ways for synthesizing materials, notably nanoparticles like AgNP, employing biological systems such as plants, algae, fungi, and bacteria [23]. This methodology differs from conformist synthesis, which repeatedly uses hazardous substances and a lot of energy. Green synthesis seeks to rely on sustainable, renewable resources, although minimizing unwanted effects and its influence on the environment. Using non-toxic chemicals like water or ethanol as solvents, reducing waste production, and exploiting renewable resources like plant extracts and microorganisms are all essential components of green synthesis [24]. Furthermore, green creation is often energy-friendly and mostly done in light circumstances, such as room temperature or low heating, which contrasts with the high-energy processes of traditional approaches. Furthermore, in green production, biological mediators serve as both stabilizing and reducing agents, refining the nanoparticles' biological functions and biocompatibility. On the other hand, hazardous

ingredients like hydrazine or sodium borohydride, that are dangerous to human well-being and the surroundings, are often used in classical production procedures. The technologies similarly tend to be energy-intensive and generate substantial amounts of waste and toxic by-products, which are inconsistent with the concepts of green chemistry. Furthermore, compared to the simpler, more economical one-step procedures frequently used in green synthesis, traditional methods are less accessible and sustainable because they frequently require expensive, specialized equipment [25]. The biological process usually involves mechanically grinding plant materials into a fine powder, which is subsequently extracted using solvents like ethanol or water. The clarified solution produced by this extraction process contains bioactive chemicals that perform as stabilizing and reducing mediators during the manufacture of nanoparticles. This source was then mixed with a silver nitrate solution and exposed to stirring and increased heat under closed conditions. A qualitative sign of the successful production of AgNP is the visual observation of a color shift [26]. These biosynthetic processes reduce the use of hazardous chemicals and produce nanoparticles that are more sustainable and biocompatible, which makes them ideal for a range of biomedical and environmental uses. The individual biological systems used for AgNP synthesis will be discussed in the sections that follow, beginning with plant-based techniques and moving on to fungus, bacteria, algae, and yeast.

The green synthesis of silver nanoparticles (AgNPs) using a biogenic approach that employs natural plant extracts as reducing and stabilizing agents is shown in the figure below. In this process, Aloe vera extract is mixed with a cucumber solution and distilled water, followed by the addition of silver nitrate (AgNO_3) [27]. The mixture undergoes a green synthesis reaction, where bioactive compounds present in the plant extract reduce Ag^+ ions to Ag^0 nanoparticles. Within 15 minutes, a visible color change to light tan indicates the initial development of AgNPs. Upon further incubation for 8 hours, the solution turns a deeper brown, confirming the complete reduction and stabilization of silver nanoparticles. This environmentally friendly synthesis avoids toxic chemicals, making it a sustainable and efficient method for nanoparticle production [1].

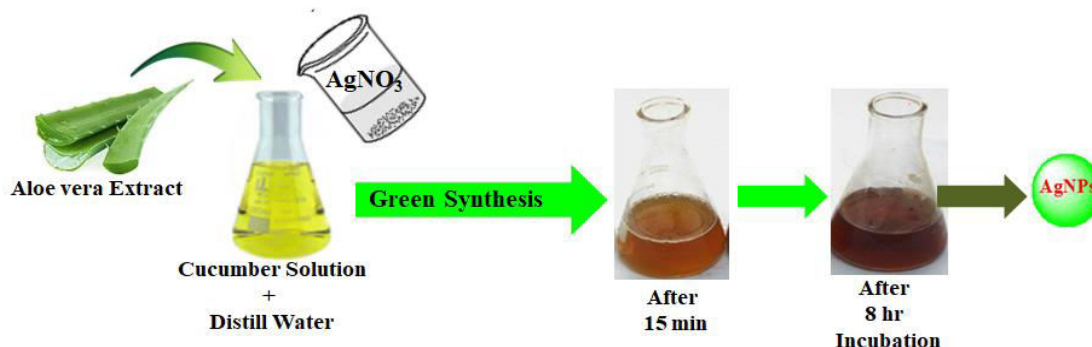


Figure 1: Eco-friendly Preparation of Silver nanoparticles

4 Purification of Synthesized AgNPs

Next step after incubation is to remove contaminations; the rust coloured liquid was centrifuged for 25 min at 5500 rpm. Hereafter, to eliminate non-required enzymes or extra proteins, the tablets obtained were thoroughly washed thrice with distilled water (5-7 mL) and finally with absolute ethanol. The processed tablets were then dried in glass watch glasses for a day at 50 - 60°C before use.

Spectral Characterisation

FTIR study was performed to detect the reducing and stabilizing agents in the synthesized Ag NPs. FTIR spectra discovered an immersion peak in the region of 3287 cm^{-1} . Besides the

peak at 2932 cm^{-1} , some other peaks at 2345, 2111, 1721, 1600, 1337, 1221, 1138, 1022, 861, 812, and 770 cm^{-1} are observed in the FTIR graph of AgNPs as shown in Figure 2.

The crystal structure of AgNPs was determined by XRD, and peaks identified at 2θ values of 37.81, 45.48, 63.92, and 78.11 can be indexed to the planes of (111), (200), (220), and (311), correspondingly. Scherrer's equation calculated the size. The typical crystal dimensions considered from the (111) peak was 8 nm (± 2 nm). The small peak obtained at 32.14 may correspond silver oxide indicated by (111*). The extra peaks perceived at 2θ range of 28.16 and 44.11 may be because of the crystallization of inorganic composites of the extract as shown in figure 3.

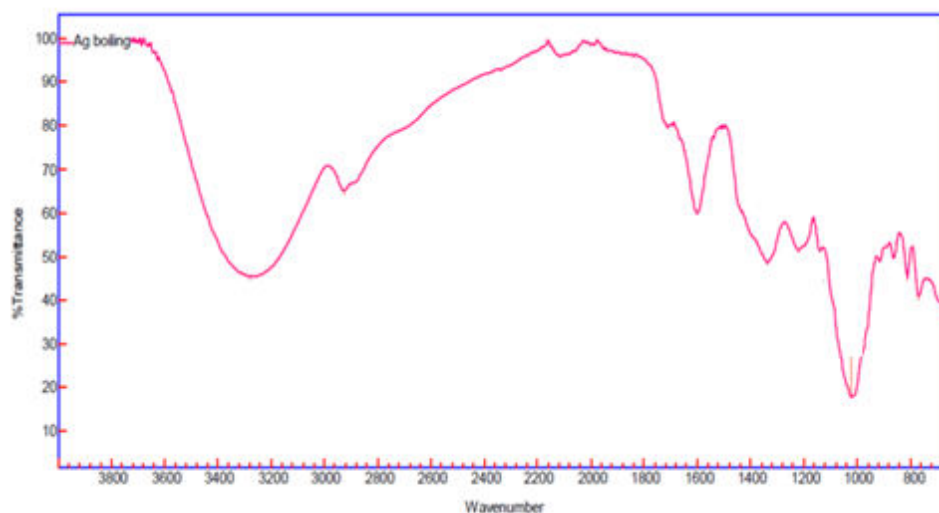


Figure 2: FTIR spectra for Silver nanoparticles

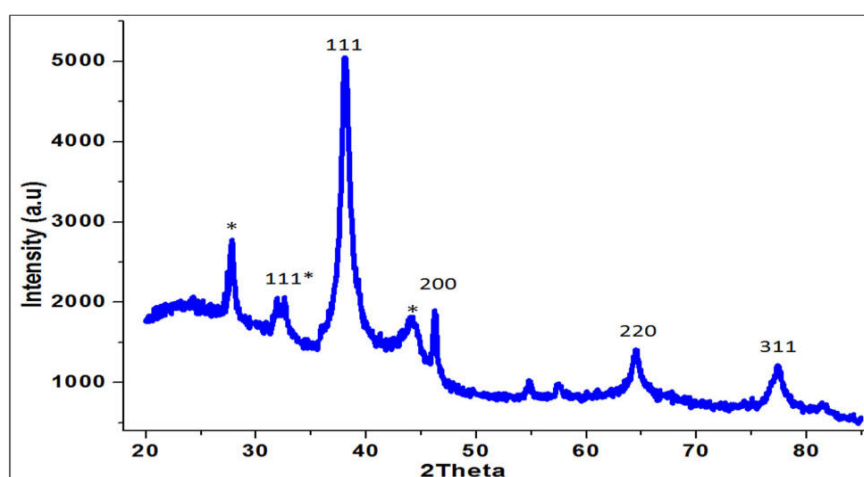


Figure 3: X-ray Diffraction spectra for Silver nanoparticles

Antibacterial Analysis

Testers containing AgNPs at applications of 10%, 20%, and 30% were evaluated for antibacterial activity. Test organisms employed were gram-negative *Escherichia coli* (*E. coli*) and gram-positive *Bacillus subtilis*. The region of inhibition for AgNPs was ascertained using the agar well diffusion method. Figure 4 presents the outcomes. This indicates that, in comparison to their respective concentrations, AgNPs are far more effective at preventing the

growth of bacterial germs. With 1 mg/mL of sample volume, the utmost zone of inhibition (ZOI) against both gram-positive and gram-negative bacteria is seen. As the sample volume grows, ZOI rises. The greater concentration of AgNPs is another reason for this increase. The ZOI's dimensions and the numerical results of ampicillin's antibacterial activity are investigated. Ampicillin has a zone of inhibition of 1.5 cm for gram-negative bacteria and 1.9 cm for gram-positive bacteria at all doses. The regions of inhibition for gram-positive bacteria against AgNPs are 0.4 cm, 0.6 cm, and 1.0 cm at various concentrations (10 μ g/ μ L, 20 μ g/ μ L, and 30 μ g/ μ L), respectively. The regions of inhibition for AgNPs are 0.7, 0.5, and 0.8 cm at various concentrations (10 μ g/ μ L, 20 μ g/ μ L, and 30 μ g/ μ L).

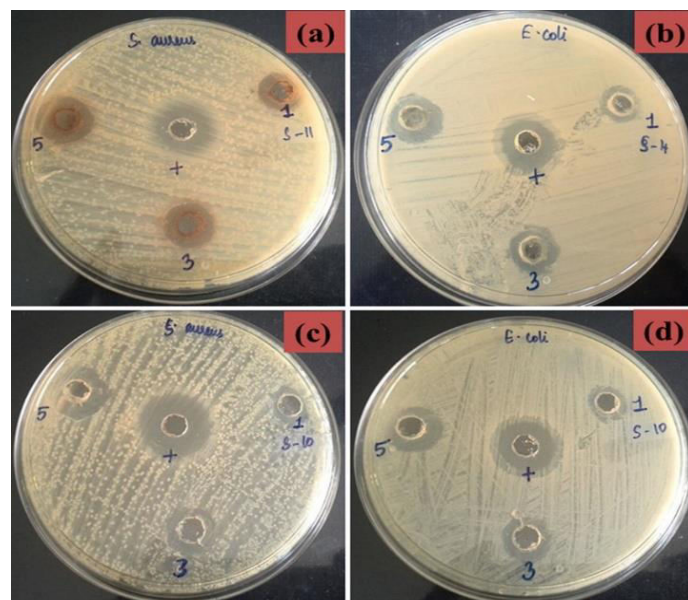


Figure 4: Antibacterial analysis of Silver nanoparticles

Conclusion

AgNP biosynthesis is an economical and eco-friendly method. Silver is becoming more widely acknowledged as a desirable element for a variety of biological applications due to the vast range of applications of NPs. AgNPs can now be synthesized without the need of surfactants thanks to a novel technique. AgNPs were synthesized at standard temperature (22–25 °C) across various time phases, and their physiological characteristics were thoroughly characterized. Using cutting-edge methods including XRD, antibacterial activity, UV-visible spectroscopy, etc., a thorough examination of the NPs' form, extent, quantity, charges, and a variety of other characteristics was conducted. Reduction, chelation, and acting as a molecular pattern in AgNP production were among the functions of the aqueous extract of AgNPs. Owing to their covering by the organic particles or proteins inside the chambers, the biogenic NPs showed strong antibacterial activity against dangerous pathogens. This made it easier for them to enter the bacterium quickly, causing stress and eventually bacterial cell death. Additionally, by reducing waste and creating safer products, it enhances human and environmental well-being. This eco-friendly method offers a viable substitute for the conventional physical/chemical techniques used in AgNP synthesis.

References

1. Mohd Fahim, Afreen Inam, Adnan Shahzaib, Nahid Nishat, AfrozJahan, Tahir Ahmad Bhat, Green synthesis of silver nanoparticles: A comprehensive review of methods, influencing factors, and applications. JCIS Open 16 (2024) 100125.

2. S. Rajeshkumar, L.V. Bharath, Mechanism of plant-mediated synthesis of silver nanoparticles – a review on biomolecules involved, characterisation and antibacterial activity, *Chem. Biol. Interact.* 273 (2017) 219–227, <https://doi.org/10.1016/j.cbi.2017.06.019>.
3. A. Haleem, M. Javaid, R.P. Singh, S. Rab, R. Suman, Applications of nanotechnology in medical field: a brief review, *Global Health Journal* 7 (2023) 70–77, <https://doi.org/10.1016/j.glohj.2023.02.008>.
4. W. Zhang, S. Roy, J. Rhim, Copper-based nanoparticles for biopolymer-based functional films in food packaging applications, *Compr. Rev. Food Sci. Food Saf.* 22 (2023) 1933–1952, <https://doi.org/10.1111/1541-4337.13136>.
5. A. Gauba, S.K. Hari, V. Ramamoorthy, S. Vellasamy, G. Govindan, M. ValanArasu, The versatility of green synthesized zinc oxide nanoparticles in sustainable agriculture: a review on metal-microbe interaction that rewards agriculture, *Physiol. Mol. Plant Pathol.* 125 (2023) 102023, <https://doi.org/10.1016/j.pmpp.2023.102023>.
6. P. Chaudhary, L. Ahamad, A. Chaudhary, G. Kumar, W.-J. Chen, S. Chen, Nanoparticle-mediated bioremediation as a powerful weapon in the removal of environmental pollutants, *J. Environ. Chem. Eng.* 11 (2023) 109591, <https://doi.org/10.1016/j.jece.2023.109591>.
7. H. Li, X. Chen, D. Shen, F. Wu, R. Pleixats, J. Pan, Functionalized silica nanoparticles: classification, synthetic approaches and recent advances in adsorption applications, *Nanoscale* 13 (2021) 15998–16016, <https://doi.org/10.1039/D1NR04048K>.
8. N. Sarfraz, I. Khan, Plasmonic gold nanoparticles (AuNPs): properties, synthesis and their advanced energy, environmental and biomedical applications, *Chem. Asian J.* 16 (2021) 720–742, <https://doi.org/10.1002/asia.202001202>.
9. A. Saravanan, P.S. Kumar, S. Karishma, D.-V.N. Vo, S. Jeevanantham, P. R. Yaashikaa, C.S. George, A review on biosynthesis of metal nanoparticles and its environmental applications, *Chemosphere* 264 (2021) 128580, <https://doi.org/10.1016/j.chemosphere.2020.128580>.
10. P.H.L. Tran, W. Duan, T.T.D. Tran, Fucoidan-based nanostructures: a focus on its combination with chitosan and the surface functionalization of metallic nanoparticles for drug delivery, *Int. J. Pharm.* 575 (2020) 118956, <https://doi.org/10.1016/j.ijpharm.2019.118956>.
11. J.-J. Xu, W.-C. Zhang, Y.-W. Guo, X.-Y. Chen, Y.-N. Zhang, Metal nanoparticles as a promising technology in targeted cancer treatment, *Drug Deliv.* 29 (2022) 664–678, <https://doi.org/10.1080/10717544.2022.2039804>.
12. L. Wang, M. HasanazadehKafshgari, M. Meunier, Optical properties and applications of plasmonic-metal nanoparticles, *Adv. Funct. Mater.* 30 (2020) 2005400, <https://doi.org/10.1002/adfm.202005400>.
13. A. ShahzaibHisana, N. Nishat, S.M. Alshehri, T. Ahamad, Z. Haque, Biogenically fabricated Tulsi-infused ZnO–CuO nanocomposites for enhanced dye reduction and adsorption, *Hybrid Advances* 5 (2024) 100145, <https://doi.org/10.1016/j.hybadv.2024.100145>.
14. A. Shahzaib, S. Khan, I. Ahmad, S.M. Alshehri, T. Ahamad, N. Nishat, Green synthesis of ZIF-67 composite embedded with magnetic nanoparticles and ZnO decoration for efficient catalytic reduction of rhodamine B and methylene blue, *Chemistry of Inorganic Materials* (2024) 100037, <https://doi.org/10.1016/j.cinorg.2024.100037>.
15. V.S. Sousa, M. RibauTeixeira, Metal-based engineered nanoparticles in the drinking water treatment systems: a critical review, *Sci. Total Environ.* 707 (2020) 136077, <https://doi.org/10.1016/j.scitotenv.2019.136077>.

16. P. Makvandi, C. Wang, E.N. Zare, A. Borzacchiello, L. Niu, F.R. Tay, Metal-based nanomaterials in biomedical applications: antimicrobial activity and cytotoxicity aspects, *Adv. Funct. Mater.* 30 (2020) 1910021, <https://doi.org/10.1002/adfm.201910021>.
17. M. Rafique, I. Sadaf, M.S. Rafique, M.B. Tahir, A review on green synthesis of silver nanoparticles and their applications, *Artificial Cells, Nanomedicine, and Biotechnology* 45 (2017) 1272–1291, <https://doi.org/10.1080/21691401.2016.1241792>.
18. SeharShahzadi, Sehrish Fatima, Quratul ain, ZunairaShafiq and Muhammad Ramzan Saeed Ashraf Janjua. A review on green synthesis of silver nanoparticles (SNPs) using plant extracts: a multifaceted approach in photocatalysis, environmental remediation, and biomedicine. *RSC Adv.*,2025, 15,3858–3903.
19. M.Razavi, E. Salahinejad, M. Fahmy, M. Yazdimamaghani, D. Vashae, and L. Tayebi, Green chemical and biological synthesis of nanoparticles and their biomedical applications, *Green Processes for Nanotechnology: from Inorganic to Bioinspired Nanomaterials*, 2015, pp. 207–235.
20. T. Rajkumar, A. Sapi, G. Das, T. Debnath, A. Ansari and J. K. Patra, Biosynthesis of silver nanoparticle using extract of Zea mays (corn Flour) and investigation of its cytotoxicity effect and radical scavenging potential, *J. Photochem. Photobiol., B*, 2019, 193,1–7.
21. I. Ijaz, E. Gilani, A. Nazir, A. Bukhari, Detail review on chemical, physical and green synthesis, classification, characterizations and applications of nanoparticles, *Green Chem. Lett. Rev.* 13 (2020) 223–245, <https://doi.org/10.1080/17518253.2020.1802517>.
22. M. Rafique, I. Sadaf, M.S. Rafique, M.B. Tahir, A review on green synthesis of silver nanoparticles and their applications, *Artificial Cells, Nanomedicine, and Biotechnology* 45 (2017) 1272–1291, <https://doi.org/10.1080/21691401.2016.1241792>.
23. F. Moradi, S. Sedaghat, O. Moradi, S. Arab Salmanabadi, Review on Green Nano-Biosynthesis of Silver Nanoparticles and Their Biological Activities: with an Emphasis on Medicinal Plants, *Inorganic and Nano-Metal Chemistry*, vol. 51, 2021, pp. 133–142, <https://doi.org/10.1080/24701556.2020.1769662>.
24. A.K. Appel, R. Thomann, R. Mülhaupt, Polyurethane nanocomposites prepared from solvent-free stable dispersions of functionalized graphenenanosheets in polyols, *Polymer* 53 (2012) 4931–4939, <https://doi.org/10.1016/j.polymer.2012.09.016>.
25. R.R. Patel Vidyasagar, S.K. Singh, M. Singh, Green synthesis of silver nanoparticles: methods, biological applications, delivery and toxicity, *Materials Advances* 4 (2023) 1831–1849, <https://doi.org/10.1039/D2MA01105K>.
26. T.R. Anju, S. Parvathy, M. ValiyaVeettil, J. Rosemary, T.H. Ansalna, M. M. Shahzabanu, S. Devika, Green synthesis of silver nanoparticles from Aloe vera leaf extract and its antimicrobial activity, *Mater. Today: Proc.* 43 (2021) 3956–3960, <https://doi.org/10.1016/j.matpr.2021.02.665>.

56. Influence of Gut-Brain Axis on Guillain-Barré Syndrome

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Abstract: Guillain-Barré Syndrome (GBS) is a post-infectious immune-mediated neuropathy resulting from the autoimmune destruction of peripheral nerves. Currently the most frequent cause of acute flaccid paralysis in India is GBS, a condition associated with preceding non-specific infections or triggers like trauma, surgery, or vaccination. While research on its pathogenesis revolves around molecular mimicry and cross-reactivity, these principles are inadequate in defining regulation of disease severity and detection of precise phenotypes. The highly variable clinical outcomes within GBS subtypes suggest variant factors that set the course of the disease including the Gut- Brain Axis (GBA). GBA offers an explanatory framework, linking the enteric and central nervous systems through the autonomic nervous system, HPA axis, and GI nerves. Stress-induced HPA responses leads to gut-barrier dysfunction, also known as leaky-gut syndrome, which causes loss of systemic immune tolerance. In this article, we propose shifting toward precision nutrition and targeted microbial restoration through a combination of AI- validated therapies, Live Biotherapeutic Products (LBPs), and TIKS formulations functioning as metabolic co-factors. This trident strategy ensures the optimal functional output of our suggested therapeutics to restore gut- immune equilibrium, promising superior treatment outcomes and a new era in the management of GBS.

Keywords: Guillain-Barré Syndrome, Gut-Brain Axis (GBA), Molecular Mimicry, Live Biotherapeutic Products (LBPs), TIKS, Gut-Barrier Dysfunction

Introduction

Guillain-Barré Syndrome (GBS) stands as the most persistent cause of acute, flaccid paralysis globally, presenting a huge challenge in the fields of clinical neurology and neuro-immunology. Fundamentally, GBS is an acute polyneuropathy identified by the heterogeneous targeting of the peripheral nervous system (PNS). Its core clinical features include the demyelinating subtype (AIDP) and the axonal variants (AMAN/AMSAN), each having different pathological markers and electrophysiological signs [1]. The currently accepted pathogenesis depends on the principle of molecular mimicry, where an antecedent infection like *Campylobacter jejuni*, leads to formation of cross-reactive antibodies that mistakenly attack host gangliosides on peripheral nerve components [2]. While this foundational mechanism explains disease initiation, the subsequent factors that regulate disease severity, define specific phenotype, and predict patient outcome remain frustratingly ambiguous. This critical lack of comprehensive mechanistic understanding demands a deeper, systemic exploration into the factors that influence the progression of this disorder [3].

The Bridge to the Gut Axis

In light of these ambiguities, attention has fundamentally shifted toward the Microbiota-Gut-Peripheral Nervous System (PNS) Axis as a key systemic modulator of GBS pathology. This complex, bidirectional communication network represents a dynamic physiological interface defined by the interaction between the enteric nervous system, the vast gut microflora, and the mucosal immune system [2]. This axis maintains a delicate state of immune tolerance, but disruptions (dysbiosis) can lead to increased intestinal permeability and the systemic translocation of antibodies [4]. It is hypothesized that this compromised gut barrier function allows the entry of components, such as Lipopolysaccharide (LPS) and specific metabolites, amplify the autoimmune cascade of GBS [3]. This viewpoint places the gut as a critical immune checkpoint; the composition and metabolic activity of the resident microbiota are now considered vital factors capable of influencing the Th17/Treg balance and, consequently, the intensity of the autoimmune attack on peripheral nerves [3].

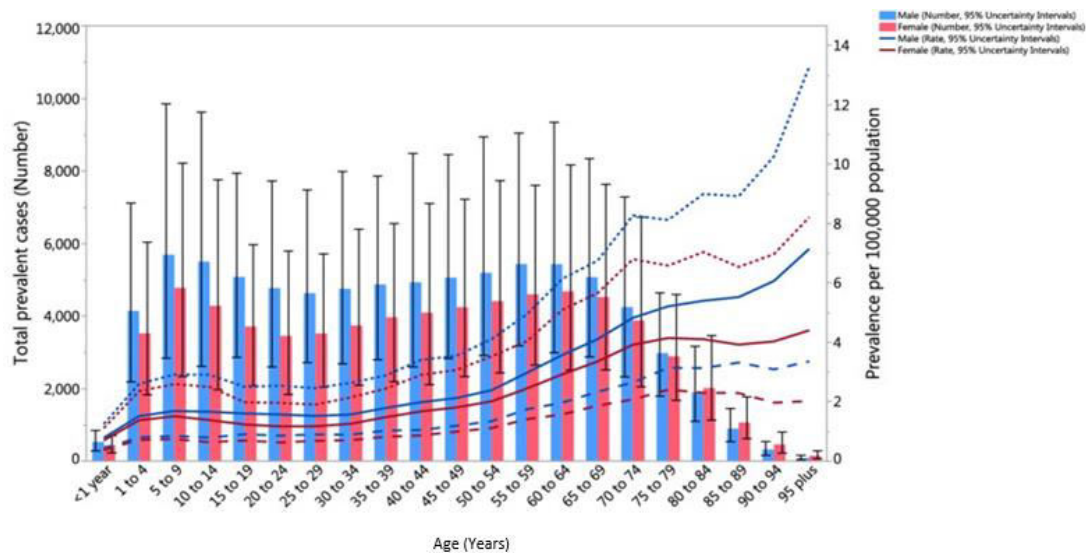


Figure 1: Global total number of cases and the prevalence of Guillain-Barre syndrome per 100,000 population, by age and sex in 2019. The error bars represent 95% uncertainty intervals for total number of cases. Dotted and dashed lines indicate 95% uncertainty intervals for prevalence (per 100,000).

Adapted from: Bragazzi NL, Kolahi AA, Nejadghaderi SA, Lochner P. Global, regional, and national burden of Guillain-Barré syndrome and its underlying causes from 1990 to 2019. *J Neuroinflammation*. 2021 Nov;18(1):2319. doi:10.1186/s12974-021-02319-4.

The Knowledge Gap and The Review Objective

Despite the established role of the gut as a source of antecedent infection and the emerging recognition of the Microbiota-PNS Axis, the specific, mechanistic contributions of gut dysbiosis and its resulting metabolite profile to GBS pathogenesis remain fragmented across the literature. Current understanding often fails to connect the microbial environment directly to the crucial Th17/Treg imbalance that decides autoimmune severity. [3]. Specifically, a comprehensive synthesis that integrates evidence on how changes in Short-Chain Fatty Acid (SCFA) levels, resulting from dysbiosis, regulate the protective function of regulatory T cells (Tregs) in GBS is critically lacking. Furthermore, the potential of microbiota-targeted therapies to restore immune tolerance, thereby identifying disease phenotype and improving patient prognosis, remains underexplored. This review seeks to move beyond conventional probiotics by evaluating advanced concepts, including personalized Live Biotherapeutic Products (LBPs) and the use of targeted Traditional Indian Knowledge System (TIKS)

formulations as metabolic co-factors for optimizing LBP functional output. We will specifically detail the breakdown of immune tolerance via the SCFA-mediated disruption of the Th17/Treg balance, evaluate the impact of gut barrier integrity on disease onset, and identify novel translational avenues for future microbiota-based therapeutic interventions.

Pathogenic Mechanisms

Molecular Mimicry and Cross-Reactivity

The pathogenesis of GBS is classically initiated by an antecedent infection. The lipooligosaccharides (LOS) on the surface of *C. jejuni* share structural homology with gangliosides (glycosphingolipids) found on the surface of human peripheral nerves, such as GM1, GD1a, and GQ1b (Molecular Mimicry) [2]. The body's adaptive immune system, upon encountering the bacterial LOS, generates antibodies that subsequently cross-react with the homologous gangliosides on the host nerves: [2].

The knowledge about the pathophysiology behind the acute inflammatory demyelinating polyneuropathy variant is very limited. This gap highlights the need to investigate the gut environment that regulates the autoimmune threshold.

Gut Barrier Dysfunction and Systemic Immune Priming

The systemic disruption of intestinal epithelium; the physical and immunological barrier is now recognized as a pivotal event in autoimmune cascade amplification. The primary cause is **dysbiosis**, a severe imbalance in the microbial community. This compromise, commonly known as "leaky gut," is caused by the degradation of essential tight junction proteins (like zonulin and occludin), effectively creating a breach in the epithelial seal [4].

When the gut barrier is breached, two critical and destabilizing events cascade through the host immune system:

Translocation of Immunogenic Products: Pathogen-associated molecular patterns (PAMPs), such as Lipopolysaccharide (LPS) derived from Gram-negative bacteria, are no longer contained and readily translocate into the systemic circulation [3]. As a potent inflammatory trigger, LPS aggressively primes systemic immunity and drives the immune system toward a pro-inflammatory state [4].

Systemic Inflammation and Autoimmune Amplification: The resulting chronic, low-grade systemic inflammation initiated by these translocated bacterial components elevates the overall autoimmune threshold of the host [4].

Microbial Metabolites and T-Cell Modulation

The gut-axis linkage to GBS has evidence in the influence of microbial metabolites on systemic immunity. The gut microbiota converts non-digestible dietary substrates (SCFAs) into powerful chemical signals (such as acetate, propionate, and butyrate [3, 2] which travel through circulation to regulate the immune cell activity of the host [3].

The balance between T-helper 17 (Th17) cells and Regulatory T cells (Tregs) dictates the overall inflammatory tone of the immune system [3].

This loss of Treg suppression and a rise in Th17 inflammation results in a severe, uncontrollable autoimmune assault on the peripheral nerve gangliosides, accelerating the demyelination and axonal damage characteristic of GBS [3]. Hence, the metabolic output of the gut microbiota acts as a certain factor of disease initiation and severity.

Clinical Implications and Heterogeneity

The Microbiota-PNS Axis offers an explanatory framework for clinical heterogeneity:

Phenotype Steering: The precise nature of the gut dysbiosis may steer the immune response towards a specific phenotype. It is hypothesized that a severe and sustained pro-inflammatory shift (high Th17/low Treg) might correlate with the more acute axonal variants [3]. Conversely, a less severe immune imbalance might lead to demyelinating presentation (AIDP).

Severity and Prognosis: Patients with a profound loss of Short Chain Fatty Acid (SCFA) driven Treg function may experience a more rapid onset and exhibit poorer long-term functional recovery [3].

Ultimately, understanding the relation between specific bacterial LOS driving molecular mimicry and the functional integrity of the patient's gut axis offers a new pathway to classify patient risk and develop targeted treatments.

Therapeutic Approaches and Future Directions

Current Standard of Care and Emerging Microbial Interventions

The current standard of care for acute Guillain-Barré Syndrome focuses on rapid immunoregulation to limit nerve damage, primarily through the administration of Intravenous Immunoglobulin (IVIg) or Plasma Exchange (PLEX) [1]. While these approaches are effective in suppressing the acute attack, they might not prevent the initial breakdown of immune tolerance or guarantee long-term functional recovery, emphasizing the need for precision therapies [3].

The current treatments toward microbiota-targeted interventions are use of Probiotics and Prebiotics and Fecal Microbiota Transplantation (FMT). However, the challenge lies in moving past these broad interventions to highly personalized strategies that address the specific metabolic and microbial weaknesses of the individual patient.

Personalized Diagnostics and Targeted Metabolite Supplementation

The limitations of non-specific microbial interventions stress the need for a shift towards therapies that directly target the SCFA deficit and subsequent breakdown of the Th17/Treg balance. This requires personalized diagnostic mapping:

Metabolomics-Driven Diagnostics: Future treatment protocols must first incorporate advanced fecal and serum metabolomics to quantify the specific loss of protective metabolites.

Direct Metabolite Supplementation: Rather than relying on a generalized probiotic to

hopefully produce SCFAs, a more direct approach involves supplementing the deficient metabolites themselves representing a promising pathway to immediately re-establish the protective T-cell environment.

This refined diagnostic and metabolite-specific approach, while promising, still represents a single-factor intervention. It naturally leads to the ultimate solution: the multi-component, precision strategies required for comprehensive and sustained restoration of the entire gut axis. Our study involves, using data driven and precision nutrition approach to address the hypothesis.

Moving Beyond Conventional: Precision Nutrition and TIKS-Optimized LBPs

Our approach focuses on solving the limitations of generalized solutions. The therapeutic inefficiency of conventional approaches stems from their failure to account for each individual's unique microbial flora and metabolic environment [8].

Our novel trident-strategy involves a data-driven approach:

AI-Validated Therapies: We propose the use of AI and advanced mapping to integrate complex genetic and metabolic data, allowing researchers to precisely pinpoint the exact metabolic weaknesses contributing to Th17/Treg imbalance.

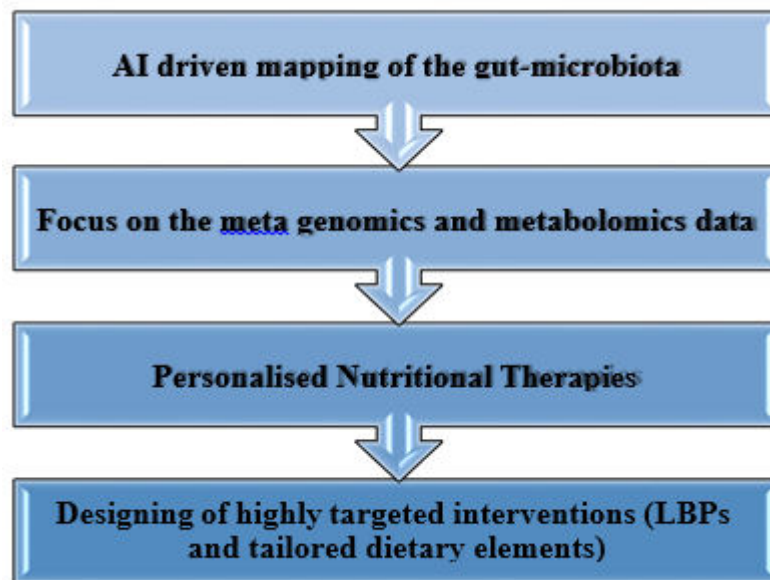


Figure 2: Process of AI mapping (CC)

Targeted Live Biotherapeutic Products (LBPs): Unlike broad-spectrum consumer probiotics, an LBP consists of one or a defined consortium of live, beneficial microorganisms (bacteria, yeast, etc.) that have been rationally selected or engineered to perform a precise therapeutic action.

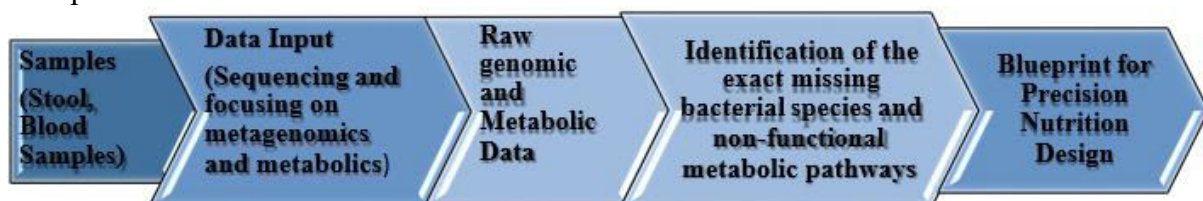


Figure 3: How precision diets are designed (CC)

TIKS as Metabolic Co-factors: The success of LBPs hinges on the metabolic environment. We propose utilizing specialized formulations from the Traditional Indian Knowledge System (TIKS) as targeted metabolic co-factors. These formulations provide the essential prebiotic and postbiotic compounds required for the newly introduced bacteria to succeed. This innovative coupling creates a highly receptive, thriving gut ecosystem, accelerating the production of vital neuro-active metabolites and ensuring rapid, sustained restoration of the crucial Gut-Brain Axis (GBA). Table 1 enlists our proposed nutriment which can control the advancement of GB syndrome and support the treatment protocols used in curbing the *C. jejuni* infection.

We hypothesize that this personalized, AI-validated approach, which strategically pairs targeted Live Biotherapeutic Products (LBPs) with TIKS as metabolic co-factors is a data-driven methodology ensuring the optimal functional output of the therapeutic microbes, accelerating the restoration of the crucial Gut-Brain Axis (GBA). This scalable strategy offers a powerful new path to address global neurodevelopmental.

Table 1: Analysing traditional probiotic based nutriment's influence the GB Axis which can inhibit the GB Syndrome

TIKS Substrate (Co-factor)	Targeted LBP	Primary Mechanism of Action	Effect on GBS Pathogenesis
Amla / Triphala (Polyphenol- rich)	<i>Lactobacillus rhamnosus</i>	<u>Oxidative Shielding:</u> Polyphenols protect LBPs from gastric acid while reducing gut ROS (Reactive Oxygen Species).	Prevents systemic inflammation
Ashwagandha (Withanolides)	<i>Bifidobacterium longum</i>	<u>HPA Axis Dampening:</u> Reduces cortisol- mediated "leaky gut" signaling.	Seals the intestinal barrier to stop the translocation of antibodies
Turmeric + Black Pepper (Curcumin/Piperine)	<i>Akkermansia muciniphila</i>	<u>Mucosal Fortification:</u> Enhances the thickness of the intestinal mucus layer.	Reduces the "mimicry" trigger by preventing pathogen access to the epithelium.
Cultured Buttermilk (Takra - Bioactive peptides)	<i>Lactobacillus plantarum</i>	<u>Synbiotic Delivery:</u> The lactic acid environment optimizes LBP colonization and SCFA production.	<u>Th17/Treg Rebalancing:</u> Increases systemic Treg cells to suppress auto- antibody production.
Guggul / Ginger (Terpenoids)	<i>Faecalibacterium prausnitzii</i>	<u>Metabolic Co-factor:</u> Promotes high-yield production of Butyrate (SCFA).	Induces peripheral nerve tolerance and promotes an environment for potential remyelination.
Bael Fruit (<i>Aegle marmelos</i>)	<i>Bifidobacterium infantis</i>	<u>Junction Protein Upregulation:</u> Bioactive flavonoids (marmelosin) tighten Occludin/Claudin proteins.	Halts the translocation of antibodies, preventing further molecular mimicry triggers.

Raw Papaya (<i>Carica papaya</i>)	<i>Lactobacillus acidophilus</i>	Biofilm Disruption: Proteolytic enzymes break down pathogenic biofilms and facilitate LBP colonization.	Clears the gut of pro-inflammatory bacterial metabolites
Bone Broth (Collagen/Glutamine)	<i>Akkermansia muciniphila</i>	Mucosal Fueling: Provides a direct supply of L-Glutamine and Glycine to rebuild the damaged intestinal mucosal layer.	Promotes a shift toward an anti-inflammatory state, allowing the PNS to focus on repair.
Gundruk (Fermented Leafy Greens)	<i>Faecalibacterium prausnitzii</i>	Gundruk's unique lignocellulosic fiber matrix acts as a specialized habitat to increase the density and metabolic activity.	Heals the gut barrier, reducing systemic inflammation
Ragi Ferment (Finger Millet Malt)	<i>Roseburia intestinalis</i>	Ragi provides slowly fermentable resistant starch and unique minerals that serve as the preferred energy for our LBP.	Enhanced butyrate stabilizes the integrity of the Blood-Brain Barrier (BBB).

Conclusion

This review reinforces the critical paradigm shift in understanding Guillain-Barré Syndrome (GBS): moving from viewing it solely as a post-infectious event driven by molecular mimicry to recognizing it as an autoimmune syndrome significantly modulated by the Microbiota-PNS Axis. While antecedent infection initiates the process, the course and severity of GBS are decided by the integrity of the gut barrier and the metabolic output of the intestinal flora [3].

Specifically, gut barrier dysfunction enables the systemic translocation of inflammatory components like LPS [4], while the resultant SCFA deficit fundamentally undermines immune tolerance by disrupting the protective Th17/Treg balance [3, 5]. This systemic immune grooming offers a compelling explanation for the observed heterogeneity in clinical phenotypes (AIDP vs. AMAN).

The future of GBS therapy must shift beyond acute immunoregulation and non-specific microbial interventions. The next frontier lies in precision medicine: a data-driven approach leveraging metabolomics and AI-validated diagnostics [6, 9] to target the precise SCFA deficits. We conclude that the most promising pathway forward is the development of Targeted Live Biotherapeutic Products (LBPs), functionally optimized by TIKS formulations acting as metabolic co-factors. Our next step of action is finding more such combinations of LBPs and TIKS Co-factors. Our study is based on the precise AI mapping of the gut. We hypothesize that constructing a thing as microbiome's own genomic library using bioinformatics tools will drastically improve the chances of forming a precise individual diet chart to figure out the specific metabolic weaknesses of a particular patient and proceed with treatment accordingly. This trident strategy offers the necessary precision to fully restore the gut-immune equilibrium, promising superior therapeutic outcomes and a new era in the management of GBS.

References

1. Hughes RA, Cornblath DR. Guillain-Barré syndrome. *Lancet*. 2005;366(9497):1653–66.
2. Green E, Trovato M, Melis M. The Gut Microbiota in Guillain–Barré Syndrome: Focus on the Role of Molecular Mimicry and the Gut–Brain Axis. *Microorganisms*. 2022;10(10):2004.
3. Zhang X, Li X, Wu X, Zhu J. The Role of Gut Microbiota and Metabolites in the Pathogenesis of Guillain–Barré Syndrome. *Int J Mol Sci*. 2023;24(17):13348.
4. Lu T, Wang J, Liu M, Zhang B, Chen S, Li M. The gut microbiota-immune axis in Guillain-Barré syndrome. *Autoimmun Rev*. 2021;20(4):102781.
5. Kieseier BC, Stüve O. The role of the microbiome in immune-mediated disorders: Potential therapeutic targets in Guillain-Barré syndrome and chronic inflammatory demyelinating polyneuropathy. *J Neuroimmunol*. 2020;348:577395.
6. Park S, Lee H, Kim T, et al. Metabolomics-guided therapy: A personalized approach to targeting the gut- immune axis in neuroinflammation. *Nat Rev Neurol*. 2024;20:101-115.
7. Haghikia A, Jörg S, Cvetanovic M, et al. Dietary fatty acids act as a novel immunomodulatory therapy in murine autoimmune T-cell inflammation. *Mol Neurobiol*. 2017;54(1):215-227.
8. Wang M, Sun Y, Wang S, Chen L, Xu X, Zhang Q. The future of personalized nutrition and microbiome- targeted therapeutics: Challenges and opportunities. *Cell Host Microbe*. 2023;31(1):145-160.
9. Gupta A, Sharma R, Varma S, et al. Artificial intelligence-driven multi-omics integration for precise diagnosis and therapeutic design in neurological autoimmune disorders. *Lancet Neurol*. 2024;23(4):301- 315.

57. Interference of Polyphenols and Polysaccharides In RNA Concentration in Aromatic Plants

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Abstract: This study evaluates the interference of secondary metabolites specifically polyphenols and polysaccharides on RNA concentration and purity in aromatic plant tissues. Poppy seed (*Papaver somniferum*) and Bay leaf (*Laurus nobilis*) were selected as model systems due to their contrasting metabolite profiles. Quantitative estimation of polysaccharides, polyphenols, and RNA was carried out using standard biochemical assays. Based on metabolite load and established, UV-spectrophotometric principles, RNA purity indices (A₂₆₀/A₂₈₀ and A₂₆₀/A₂₃₀) were assessed. Results demonstrate that extremely high polysaccharide concentrations (up to 525.12 mg/g) and substantial phenolic content promote co-precipitation, oxidative binding, and spectral interference, leading to suppressed absorbance ratios. These distortions compromise RNA suitability for downstream molecular applications.

Introduction

Aromatic plants are global resources for bioactive compounds, containing high levels of polyphenols such as flavonoids and tannins and polysaccharides like cellulose and pectin's (Li et al., 2019; Gao et al., 2007). While these metabolites provide antioxidant and antimicrobial benefits, they are known to interfere with cellular processes and RNA stability (Huang et al., 2020). Specifically, polyphenols can oxidize into quinones that covalently bind to nucleic acids, while polysaccharides often coprecipitate with RNA during extraction (Mattheus et al., 2003; Singh et al., 2003). This study aims to bridge the knowledge gap regarding how their combined presence affects RNA yield and purity.

Materials and Methods

1. Materials and Reagents

The study utilized two primary aromatic plant samples: Poppy seed (*Papaver somniferum*) and Bay leaf (*Laurus nobilis*), both obtained from local markets.

Chemicals and Equipment:

- Reagents: Anthrone, Folin-Ciocalteu, Orcinol, Benedict's, Seliwanoff's, Molisch's (α -naphthol), and Ninhydrin reagents.
- Solvents/Acids: Distilled water, 80% Ethanol, Concentrated Sulphuric acid (H_2SO_4), 3N Hydrochloric acid (HCl), Perchloric acid (PCA), and 0.3 N Potassium hydroxide (KOH).

- Instruments: UV-Visible Spectrometer, centrifuge, water bath, and analytical balance.

2. Extract Formation

The aqueous extract was prepared to facilitate the qualitative and quantitative screening of secondary metabolites.

1. Preparation: 10g of dry, semi-grinded powder of each sample (Poppy seed and Bay leaf) was weighed.
2. Extraction: The powder was mixed with 100 ml of distilled water and boiled in a round-bottom flask on a hot plate for 45 minutes.
3. Filtration: After cooling, the solution was filtered through a muslin cloth.
4. Collection: The filtrate was collected in a conical flask, while the remaining debris was discarded.

3. Qualitative and Quantitative Methodology

The research employed a series of biochemical assays to identify and measure the target compounds.

A. Qualitative Screening:

- Polysaccharides: Identified using the Molisch test (general carbohydrate detection), Iodine test (starch/glycogen), Seliwanoff's test (ketose differentiation), and Benedict's test (reducing sugars).
- Polyphenols: Detected via the Ferric Chloride test, where a deep blue or black colour indicated their presence.
- Proteins: The Ninhydrin test confirmed the presence of amino acids and proteins.

B. RNA Isolation:

200 mg of sample was treated with 80% alcohol and heated to discard pigments. The residue was washed with 1% PCA and then incubated in 0.3 N KOH at 37°C for 18-20 hours to induce alkaline hydrolysis of nucleic acids into nucleotides. Finally, the pH was adjusted to 2 with 1 N PCA and centrifuged to collect the RNA-containing supernatant.

C. Quantitative Estimation:

- Polysaccharides (Anthrone Method): Measured at 630 nm using a dextrose standard curve.
- Polyphenols (Folin-Ciocalteu Method): Measured at 765 nm (or 735 nm) using a Gallic acid standard curve.
- RNA (Orcinol Method): Measured at 665 nm.

4. Methodological References

Based on the sources and external scientific literature for standard protocols, the following references apply to these methods:

- Polyphenol Estimation: (Singleton et al., 1965).
- RNA Interference Context: (Singh et al., 2003) and (Wilkins et al., 1996).
- Polysaccharide Estimation (Anthrone): (Dreywood et al., 1946).
- RNA Estimation (Orcinol/Bial's): (Bial et al., 1902).
- Alkaline Hydrolysis (RNA Isolation): (Schmidt et al., 1945).
- Phytochemical Screening (Molisch): (Molisch et al., 1886).
- Phytochemical Screening (Seliwanoff): (Seliwanoff et al., 1887).
- Phytochemical Screening (Benedict): (Benedict et al., 1909).

2.5 Purity Assessment

RNA purity was assessed using standard UV spectrophotometric principles, correlating metabolite concentrations with absorbance ratios:

A260/280: Indicator of protein and phenolic contamination

A260/230: Indicator of polysaccharide and phenolic contamination

All biochemical estimations and spectrophotometric measurements were performed in triplicate. Data are expressed as mean $\pm$ standard deviation (SD).

The coefficient of variation (CV) for all assays was maintained below 10%.

Results and Observation

Distinct biochemical profiles were observed between the two samples, demonstrating differential metabolite interference on RNA purity.

Table 1. Biochemical composition and absorbance ratios

Sample	Polysaccharides (mg/g)	Polyphenols (mg/g)	RNA (mg/g)	A260/280	A260/230
Poppy seed	320.4 $\pm$ 19.4	114.0 $\pm$ 8.5	20.25 $\pm$ 1.65	1.85 $\pm$ 0.13	1.21 $\pm$ 0.09
Bay leaf	525.12 $\pm$ 31.7	101.0 $\pm$ 8.2	25.1 $\pm$ 1.9	1.97 $\pm$ 0.14	0.76 $\pm$ 0.06

(Data derived from Tables 3, 5, 7, and 8)

Values represent mean $\pm$ standard deviation (SD) of three technical replicates (n = 3).

Discussion

The investigation into the biochemical interplay within aromatic plants reveals that while secondary metabolites like polyphenols and polysaccharides provide significant therapeutic and commercial value, they present a formidable "hindrance" during molecular analysis. The quantitative data confirms that high concentrations of these compounds directly interfere with the yield and purity of RNA, which is essential for downstream applications such as RT-PCR and cDNA library construction.

1. Comparative Analysis of Metabolite Concentrations

The results demonstrate a clear distinction in the metabolite profiles of the two selected species. Poppy seed (*Papaver somniferum*) contains a relatively high phenolic concentration of 114 mg/g, while Bay leaf (*Laurus nobilis*) contains a "relatively maximum" polysaccharide concentration of 525.12 mg/g. The research highlights that as these metabolite "doses" increase, the difficulty in isolating high-quality RNA also increases, as reflected in the distorted absorbance ratios observed in the samples.

2. Impact of Polyphenols and Proteins on A260 /A280

In Poppy seed, the elevated polyphenol content (114 mg/g) along with the presence of storage proteins confirmed through qualitative analysis contributes significantly to the suppression of the A260 /A280 ratio, which was measured at 1.85. According to established molecular

standards, polyphenols and proteins absorb strongly at 280 nm, which increases the denominator of the ratio and pulls the result below the ideal value of approximately 2.0 [Sambrook et al., 2001]. Furthermore, the study indicates that phenolics are readily oxidized into quinones, which form covalent bonds with nucleic acids. This molecular binding results in the formation of insoluble complexes, effectively reducing the solubility and overall integrity of the RNA.

3. Polysaccharide Interference on A260/A230

The Bay leaf sample exhibited an exceptionally high polysaccharide concentration of 525.12 mg/g, which resulted in a markedly reduced A260/A230 ratio of 0.76 far below the ideal range of 2.0-2.2. This interference occurs because polysaccharides absorb strongly at 230 nm [Wilfinger et al., 1997]. Due to their similar physicochemical properties to RNA, these carbohydrates readily coprecipitate during the extraction process. This leads to the production of viscous extracts that not only hinder the resuspension of precipitated RNA but also act as physical inhibitors for enzymatic reactions such as reverse transcription and PCR.

Summary of Molecular Interference

The findings of this study support the conclusion that yielding high-quality RNA is difficult in aromatic plants due to high levels of phenolics and polysaccharides [Wilkins et al., 1996]. The Anthrone technique used for quantification additionally highlighted technical challenges, as it loses accuracy in the presence of proteins, which were detected in the Poppy seed sample. Ultimately, the study underscores that the high doses of metabolites in these species cause RNA aggregation and oxidative damage, necessitating the development of advanced extraction techniques to overcome these bioactive “hindrances”.

Conclusion

This investigation establishes that elevated levels of polyphenols and polysaccharides in aromatic plants significantly hinder RNA purity. (Singh et al., 2003) Suppressed A260/230 ratios in Bay leaf and reduced A260/280 ratios in Poppy seed highlight metabolite-induced spectral distortion and co-precipitation. These findings emphasize the necessity for modified extraction buffers and purification strategies to obtain RNA of sufficient quality for sensitive downstream applications such as RT-PCR, gene expression profiling, and cDNA library construction.

References

1. Benedict et al., 1909: Cited for the Benedict test utilized in qualitative phytochemical screening.
2. Bial et al., 1902: Cited for the Orcinol/Bial's method used in quantitative RNA estimation.
3. Dreywood et al., 1946: Cited for the Anthrone method utilized in the quantitative estimation of polysaccharides.
4. Gao et al., 2007: Referenced regarding the presence of polysaccharides like cellulose and pectin's in aromatic plants.
5. Huang et al., 2020: Referenced for the interference of secondary metabolites with cellular processes and RNA stability.

6. Li et al., 2019: Referenced regarding the high levels of polyphenols (flavonoids and tannins) found in aromatic plant resources.
7. Mattheus et al., 2003: Referenced for the understanding that polyphenols oxidize into quinones which covalently bind to nucleic acids.
8. Molisch et al., 1886: Cited for the Molisch test utilized in general qualitative carbohydrate detection.
9. Sambrook et al., 2001: Cited for established molecular standards regarding the suppression of the A260/A280 ratio by proteins and polyphenols.
10. Schmidt et al., 1945: Cited for the alkaline hydrolysis procedure utilized for RNA isolation.
11. Seliwanoff et al., 1887: Cited for the Seliwanoff test utilized in qualitative ketose differentiation.
12. Singh et al., 2003: Referenced extensively regarding the coprecipitation of polysaccharides with RNA during extraction and the resulting hindrance to purity and isolation.
13. Singleton et al., 1965: Cited for the Folin-Ciocalteu method used in the quantitative estimation of polyphenols.
14. Wilfinger et al., 1997: Cited regarding the absorption of polysaccharides at 230 nm and the subsequent reduction of the A260/A230 ratio.
15. Wilkins et al., 1996: Referenced regarding the overall difficulty of yielding high-quality RNA in aromatic plants due to high metabolite loads.

58. Leveraging IoT and Data Analytics for Smarter Crop Management in Precision Farming

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Abstract: Precision agriculture is increasingly reliant on tools that enable farmers to manage crops more accurate and efficiently. The integration of Internet of Things devices with Big Data Analytics. It has become a significant driver of this shift. IoT-based sensors offer continuous monitoring of key crop and soil parameters, while data analytics systems transform these readings into actionable insights. Together, these technologies improve irrigation decisions, fertilizer scheduling, pest and disease detection, and overall crop performance. This paper examines the technological framework behind IoT-enabled precision farming, reviews recent research on data-driven agricultural systems, and evaluates the potential benefits and challenges associated with their deployment. The study highlights how real-time analytics strengthen farm decision-making, reduce resource wastage, and increase sustainability. It also discusses the limitations concerning data privacy, infrastructure gaps, and adoption barriers in developing regions. Overall, IoT technologies promises to reshape modern farming, elevate productivity, and support long-term food security.

Keywords: Precision Agriculture, IoT, Sensors, Big data analytics.

Introduction

Agriculture is undergoing a digital transformation driven by advancements in sensor technology, communication networks, and artificial intelligence. Traditional farming methods often rely on manual observation and generalized resource application, resulting in unpredictable outcomes and inefficient input use. Precision agriculture offers a more systematic approach by tailoring interventions based on real-time field conditions.

IoT devices are at the heart of this transformation. They provide farmers with continuous feedback on soil, crop, and environmental factors that influence productivity. When paired with Big Data analytics, these devices generate insights that help farmers make informed decisions, ultimately improving efficiency and yield quality.

This paper focuses on how IoT and data analytics systems work together to strengthen precision agriculture. It studies their applications, advantages, challenges, and long-term implications for sustainable farming.

Review of Literature

IoT in Agriculture includes devices such as soil moisture sensors, weather stations and plant health monitors. These devices transmit data using wireless communication technologies like LoRaWAN, Wi-Fi, and cellular networks. Prior research shows that IoT based monitoring can significantly reduce water consumption and improve crop uniformity.

Big Data in Agricultural Decision-Making

Big Data platforms organize and analyse large volumes of structured and unstructured farm data. Analytical tools such as machine learning models can predict crop diseases, estimate yield, and recommend input adjustments. Studies have demonstrated that predictive analytics can increase accuracy in decision-making and reduce input costs.

Integration of IoT and Big Data

Several studies highlight the synergy between IoT generated data and analytics frameworks. IoT collects real-time information, while data analytics converts raw data into meaningful patterns. This integration supports precision irrigation, nutrient management, and climate smart agriculture.

Limitations Identified in Prior Work

Challenges noted in existing research include unreliable internet access in rural areas, the high cost of IoT systems, and limited technical literacy among farmers. Furthermore, concerns about data ownership and privacy remain unresolved.

Methodology

This paper adopts a conceptual and qualitative research approach. It synthesizes existing academic literature, examines case studies from recent deployments of IoT systems in agriculture, and evaluates technological frameworks used in precision farming.

Key steps in the methodology include:

1. Reviewing agricultural IoT architectures.
2. Analyzing Big Data processing pipelines relevant to farming.
3. Comparing real-world case studies.
4. Identifying benefits, limitations, and gaps.

This approach enables a structured understanding of how IoT and analytics function together within agricultural environments. IoT Architecture for Precision Farming

A typical IoT-based precision farming system consists of:

1.Sensor Layer

Includes soil moisture sensors, temperature sensors, nutrient probes, leaf wetness monitors, and multispectral imaging devices.

2. Connectivity Layer

Uses communication technologies such as:

- LoRaWAN for long-range data transmission
- GSM/4G/5G networks
- Zigbee and Bluetooth for short-range communication

2.Cloud/Data Processing Layer

Incoming data is stored, filtered, and analysed using cloud platforms. Machine learning models may identify unusual patterns such as sudden moisture drops or disease symptoms.

3.Application Layer

Provides farmers with user-friendly dashboards, mobile apps, and alert systems that recommend management actions.

Applications in Crop Management

1.Precision Irrigation:

Sensors determine moisture levels at various soil depths. Analytics suggest specific irrigation schedules, reducing water wastage and preventing crop stress.

2.Nutrient and Fertilizer Optimization

Data from nutrient sensors and leaf analysis helps determine fertilizer quantity and timing, minimizing excess application.

3.Pest and Disease Early Warning

Images and climate data can identify early signs of diseases. Alerts are sent to farmers, enabling quick intervention.

4.Yield Prediction

Predictive models integrate weather data, soil metrics, and crop growth stages to generate accurate yield forecasts.

Results and Discussion

Case studies from India, Brazil, and Europe reveal notable improvements in productivity after implementing IoT and analytics systems:

- Water usage reduced by 20–45% in sensor-guided farms.
- Crop yields increased by 10–25%, depending on crop type.
- Fertilizer input reduced by 15–30%.
- Early pest detection minimized crop loss by up to 40%.

The findings indicate that IoT and analytics significantly benefit farmers by improving decision accuracy and resource efficiency. However, challenges such as affordability, network availability, and technical skill gaps still hinder widespread adoption.

Challenges and Limitations

1. High Initial Investment: Sensors and cloud systems can be expensive for small-scale farmers.
2. Connectivity Issues: Remote farms often lack stable internet or electricity.
3. Data Privacy: Ownership of farm data remains a sensitive issue.
4. Technical Skills Required: Many farmers need training to operate digital platforms.

5. Maintenance and Durability: Harsh weather conditions can damage sensors.

Conclusion

IoT and Big data analytics play a transformative role in modern precision farming by providing real time insights into soil, environmental, and crop conditions, these technologies help farmers improve productivity, reduce resource consumption, and achieve sustainable agricultural outcomes. Although challenges remain— particularly in developing countries— growing technological advancements and declining hardware costs are likely to accelerate adoption. The integration of IoT with data analytics is poised to redefine agriculture and meet the demands of a rapidly growing global population.

Recommendations

- Develop affordable IoT devices suited to small farms.
- Improve connectivity infrastructure in rural areas.
- Train farmers through government and NGO led digital literacy programs.
- Promote open data standards to support interoperability.
- Encourage public private partnerships to scale precision farming solutions.

References

1. Wolfert, S., Ge, L., Verdouw, C., & Bogaardt, M. (2017). Big Data in smart farming. *Agricultural Systems*, 153, 69–80.
2. Khanna, A., & Kaur, S. (2019). Internet of Things (IoT) in agriculture. *Computers and Electronics in Agriculture*, 157, 218–231.
3. Liakos, K., Busato, P., Moshou, D., Pearson, S., & Bochtis, D. (2018). Machine learning in agriculture. *Sensors*, 18(8), 1–29.
4. Ray, D. K., Gerber, J. S., MacDonald, G. K., & West, P. C. (2015). Climate variation impacts on crop yields. *Nature Communications*, 6, 5989.
5. Shafi, U., Mumtaz, R., García, L., & et al. (2019). Precision agriculture and IoT-based monitoring. *Sensors*, 19(17), 1–25.

59. Physicochemical Characterization and Biological Activities of *Asparagus officinalis* Stem Extracts

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Abstract: Because of its low side effects and biocompatibility, *Asparagus officinalis* is a well-known medicinal plant that is frequently employed in both traditional and contemporary healthcare systems. While the roots of asparagus have been the subject of much investigation, the bioactive potential of its stems has received less attention. The current research seeks to assess the physicochemical characteristics, phytochemical makeup, antioxidant capacity, antimicrobial and antibiofilm effects, and fermentation-promoting qualities of *A. officinalis* stem extracts made with ethanol and ethyl acetate. The ethanolic extract showed greater phenolic content and antioxidant capacity than the ethyl acetate extract. Both extracts exhibited moderate antimicrobial effects against *Escherichia coli* ATCC 25922 and *Staphylococcus aureus* ATCC 43300. During yoghurt fermentation, the ethanolic extract also demonstrated encouraging antibiofilm activity and increased microbial proliferation. These results imply that *A. officinalis* stem extract may find use in the culinary, medicinal, and fermentation sectors.

Keywords: *Asparagus officinalis*, stem extract, phenolic content, antioxidant activity, antimicrobial activity, antibiofilm, fermentation

Introduction

Traditionally used for its anti-inflammatory, immunomodulatory, antioxidant, and antibacterial qualities, *Asparagus officinalis* L. is a nutritionally and medicinally significant plant. *Asparagus* species are frequently used in traditional healthcare systems, especially in developing nations, due to their high compatibility with human metabolism, low toxicity, and few side effects (Sharma et al., 2014; Huang et al., 2019). These pharmacological actions are mostly caused by bioactive substances like phenolics, flavonoids, saponins, and polysaccharides.

Due to their well-established medicinal potential, the roots of asparagus, especially *A. racemosus*, have been the subject of most prior research. However, despite being easily accessible and frequently thrown away as agricultural waste, the aerial parts, particularly the stems of *A. officinalis*, are still relatively understudied (Singh et al., 2018). This reveals a substantial research gap and a chance for stem biomass to be valued.

Among the most significant secondary metabolites found in plants, phenolic compounds play a major role in antioxidant, antimicrobial, and antibiofilm activities. By interfering with cell membranes, enzyme systems, and quorum-sensing mechanisms, these substances can prevent oxidative stress and microbial growth (Daglia, 2012; Cushnie & Lamb, 2011). Therefore, investigating various plant parts including stems may offer economical and sustainable sources of bioactive compounds for use in food and pharmaceutical applications.

Plant-derived phenolic extracts have drawn interest in the fermentation industry due to their potential to improve microbial growth and metabolic activity, in addition to their medical applications. By encouraging the growth of advantageous lactic acid bacteria and enhancing the functional quality of fermented dairy products like yoghurt, some phenolic compounds can function as prebiotic-like substances (Martínez-Villaluenga et al., 2020). With an emphasis on their antioxidant, antimicrobial, antibiofilm, and fermentation-related uses, the current study attempts to explore the biological and physicochemical characteristics of *A. officinalis* stem extracts.

The study's objectives

To assess *Asparagus officinalis* stem extracts' total phenolic content and antioxidant potential
To evaluate the stem extracts' antimicrobial and antibiofilm properties against particular bacterial pathogens.

To investigate how *A. officinalis* stem extracts might be used in the fermentation sector by looking at how they affect yoghurt's microbial growth acceleration

Materials and Methods

1 Plant Collection and Extraction

During the active growth phase, fresh *Asparagus officinalis* L. stems were gathered from robust plants. A certified botanist verified the plant material, and only stems free of disease were chosen for examination. To get rid of contaminants and adhering soil particles, the collected stems were thoroughly cleaned under running tap water and then rinsed with distilled water. To create a consistent plant paste, the cleaned stems were then chopped into tiny pieces and homogenised using a lab mixer.

Soxhlet extraction was carried out to obtain bioactive compounds from the stem material, following standard extraction protocols with minor modifications (Harborne, 1998; Azwanida, 2015). Briefly, 100 g of crushed stem material was loaded into a Soxhlet apparatus and extracted separately using ethanol and ethyl acetate as solvents. A total of 200 mL of solvent was used for each extraction, and the process was allowed to run continuously for 6 h to ensure efficient extraction of phenolic and other bioactive compounds.

After completion of extraction, the solvent extracts were filtered through Whatman No. 1 filter paper to remove plant debris. The filtrates were then concentrated by evaporating the solvents using a water bath maintained at 80 °C until semi-solid residues were obtained. The dried extracts were weighed to calculate extraction yield and subsequently stored in airtight containers at 4 °C until further physicochemical and biological analyses.

2 Determination of Moisture Content

The moisture content of *Asparagus officinalis* stem samples was determined using the standard loss-on-drying (LOD) method, following established analytical procedures (AOAC, 2019; Nielsen, 2017). Briefly, a known weight of the fresh stem sample was accurately measured and placed in a pre-weighed moisture dish. The sample was then dried in a hot air oven maintained at 105 °C until a constant weight was achieved.

After drying, the samples were allowed to cool in a desiccator to prevent moisture absorption from the atmosphere and subsequently reweighed. The moisture content was calculated based on the difference between the initial and final weights of the sample and expressed as a percentage of the original sample weight using the following equation:

$$\text{Moisture Content} = \frac{\text{Initial weight} - \text{Dry weight}}{\text{Initial weight}} \times 100$$

All determinations were carried out in triplicate, and the mean values were reported.

3 Determination of Ash Value

The ash value of *Asparagus officinalis* stem powder was determined to estimate the total inorganic content present in the plant material, following standard pharmacopeial and analytical procedures (AOAC, 2019; WHO, 2011). Briefly, a known quantity of the dried and finely powdered stem sample was accurately weighed and placed in a previously ignited and tarred silica crucible.

The sample was incinerated in a muffle furnace maintained at 550 ± 25 °C until it turned white, indicating the absence of carbon and the attainment of a constant weight. After incineration, the crucible was allowed to cool in a desiccator to avoid moisture absorption and subsequently reweighed. The incineration and weighing process was repeated until a constant weight was achieved.

The ash value was calculated as the percentage of residue remaining after incineration with respect to the initial dry weight of the sample using the following formula:

$$\text{Ash Value (\%)} = \frac{\text{Weight of Ash}}{\text{Weight of dry sample}} \times 100$$

All measurements were performed in triplicate, and the results were expressed as mean values $\pm$ standard deviation.

4 Determination of Total Phenolic Content

The total phenolic content (TPC) of *Asparagus officinalis* stem extracts was determined using the Folin–Ciocalteu colorimetric method, as described previously with slight modifications (Singleton et al., 1999; Wolfe et al., 2003). Briefly, 1 mL of extract solution (1 mg/mL) was mixed with 1 mL of Folin–Ciocalteu reagent previously diluted with distilled water. The reaction mixture was allowed to stand for 3 min at room temperature to facilitate initial oxidation.

Subsequently, 1 mL of 1 M sodium carbonate solution was added to the mixture, and the final volume was adjusted to 10 mL using distilled water. The reaction mixture was then incubated in the dark at room temperature for 90 min to allow complete color development. The absorbance was measured at 725 nm using a UV–Visible spectrophotometer against a reagent blank.

Gallic acid was used as the standard phenolic compound to generate a calibration curve (concentration range: typically 10–100 $\mu\text{g/mL}$), and the total phenolic content of the extracts was calculated from the standard curve. Results were expressed as milligrams of gallic acid equivalents (GAE) per milligram of dry extract (mg GAE/mg extract). All analyses were performed in triplicate, and results were reported as mean $\pm$ standard deviation.

5. Determination of Total Antioxidant Content

The total antioxidant activity of *Asparagus officinalis* stem extracts was evaluated using a standard spectrophotometric free radical scavenging assay, following previously reported methods with minor modifications (Brand-Williams et al., 1995; Mensor et al., 2001). Briefly, a freshly prepared 0.1 mM solution of 2,2-diphenyl-1-picrylhydrazyl (DPPH) in methanol was used as the stable free radical source.

Different concentrations of the stem extracts were prepared, and an aliquot of each concentration was mixed with the DPPH solution. The reaction mixtures were incubated in the dark at room temperature for 30 min to allow complete interaction between the antioxidants present in the extracts and the DPPH radicals. After incubation, the decrease in absorbance was measured at 517 nm using a UV–Visible spectrophotometer against a suitable blank.

Ascorbic acid was used as a reference standard. The percentage of radical scavenging activity was calculated using the following equation:

$$\text{Radical scavenging activity (\%)} = \frac{A_0 - A_s}{A_0} \times 100$$

Where, A_0 is the absorbance of the control and A_s is the absorbance of the sample.

The concentration of extract required to inhibit 50% of the DPPH radicals (IC_{50}) was calculated from the plotted graph of percentage inhibition versus extract concentration. Lower IC_{50} values indicate higher antioxidant potential. All experiments were performed in triplicate, and results were expressed as mean IC_{50} values $\pm$ standard deviation.

6 Antimicrobial Activity

The antimicrobial activity of *Asparagus officinalis* stem extracts was evaluated using the broth microdilution method in accordance with standard guidelines (CLSI, 2022; Wiegand et al., 2008). The assay was performed against two reference bacterial strains: *Escherichia coli* ATCC 25922 and *Staphylococcus aureus* ATCC 43300.

Serial two-fold dilutions of the stem extracts were prepared in sterile Mueller–Hinton broth to obtain concentrations ranging from 5.0 to 0.15 mg/mL. Overnight bacterial cultures were adjusted to the 0.5 McFarland standard and further diluted to achieve a final inoculum concentration of approximately 3×10^6 CFU/mL. Equal volumes of bacterial suspension and extract dilutions were then added to sterile culture tubes (or microtiter plates) under aseptic conditions.

The inoculated cultures were incubated at 37 °C for 24 h. Following incubation, bacterial growth was assessed by measuring absorbance at 600 nm using a UV–Visible spectrophotometer. Mueller–Hinton broth containing bacterial inoculum without extract served as the negative control, while streptomycin and ciprofloxacin were used as positive control antibiotics.

The minimum inhibitory concentration (MIC) was defined as the lowest concentration of the extract that resulted in a significant reduction in bacterial growth compared to the control. All

experiments were conducted in triplicate, and results were expressed as mean values $\pm$ standard deviation.

7 Antibiofilm Activity

The antibiofilm activity of *Asparagus officinalis* stem extracts was evaluated using the crystal violet staining method, as previously described by Olawuwo et al. with minor modifications (Olawuwo et al., 2019; Stepanović et al., 2007). The assay was performed against selected bacterial pathogens using standardized cultures.

Briefly, bacterial suspensions were prepared from overnight cultures and adjusted to the 0.5 McFarland standard. Aliquots of the standardized bacterial cultures were dispensed into sterile 96-well microtiter plates containing different concentrations of the stem extracts ranging from 100 to 500 mg/mL. Wells containing bacterial culture without extract served as the biofilm control, while wells containing broth alone were used as blanks. The plates were incubated at 37 °C for 24 h to allow biofilm formation.

After incubation, the planktonic cells were gently removed, and the wells were washed twice with sterile phosphate-buffered saline (PBS) to remove non-adherent cells. The adhered biofilms were then fixed and stained with 1 % (w/v) crystal violet solution for 15 min at room temperature. Excess stain was removed by rinsing with distilled water, and the plates were air-dried.

The bound crystal violet was solubilized by adding 33 % (v/v) acetic acid to each well, and the absorbance was measured at 530 nm using a microplate reader. The percentage of biofilm inhibition was calculated relative to the untreated control. All experiments were performed in triplicate, and results were expressed as mean values $\pm$ standard deviation.

8 Application in Yoghurt Fermentation

The effect of *Asparagus officinalis* stem extracts on yoghurt fermentation was evaluated following standard yoghurt preparation protocols with slight modifications (Tamime & Robinson, 2007; Shah, 2007). Fresh cow milk was first heated to 90 °C for 10 min to ensure pasteurization and protein denaturation, followed by cooling to 44 ± 1 °C.

The pasteurized milk was inoculated with a natural yoghurt starter culture containing *Lactobacillus delbrueckii* subsp. *bulgaricus* and *Streptococcus thermophilus*. The inoculated milk was then distributed into sterile jars. *A. officinalis* stem extracts were added at final concentrations of 1, 2.5, and 5 mg/mL to separate treatment groups, while a sample without extract served as the control.

All samples were incubated at 44 °C for 6 h under static conditions. The progression of fermentation was monitored by measuring the pH at 2, 4, and 6 h intervals using a calibrated digital pH meter. Changes in pH were used as an indicator of microbial activity and lactic acid production during fermentation. All experiments were conducted in triplicate, and results were expressed as mean values $\pm$ standard deviation.

Results

1 Plant Collection and Extraction

Fresh stems of *Asparagus officinalis* were successfully collected and processed for extraction. The collected plant material was healthy, succulent, and green in appearance, with no visible signs of microbial contamination or physical damage. After washing and mechanical homogenization, the stem material formed a uniform green paste suitable for solvent extraction.

Soxhlet extraction using ethanol and ethyl acetate yielded semi-solid, viscous extracts with a characteristic green coloration. The observed green color suggests the presence of chlorophyll pigments along with other phytochemicals such as phenolics and flavonoids. The ethanolic extract exhibited a darker and more intense green color with higher viscosity compared to the ethyl acetate extract, indicating a higher extraction efficiency for polar bioactive compounds. This observation is consistent with earlier studies reporting ethanol as an effective solvent for extracting phenolic compounds from plant matrices.

No evidence of thermal degradation, charring, or solvent residue was observed during the concentration process. The extracts remained stable during storage at 4 °C, with no phase separation or microbial growth observed prior to further analyses. These findings confirm that *A. officinalis* stems are a suitable and stable source of extractable phytochemicals and justify their selection for subsequent physicochemical, antioxidant, antimicrobial, antibiofilm, and fermentation studies.



Figure 1. Ethanolic and ethyl acetate extracts of *Asparagus officinalis* stems obtained after Soxhlet extraction and solvent evaporation, showing characteristic green coloration and semi-solid consistency indicative of phytochemical richness.

2&3 Determination of Moisture Content, pH and Ash content

Table 1. Physicochemical parameters of *Asparagus officinalis* stem

Parameter	Result (Mean $\pm$ SD)
Moisture content (%)	6.01 $\pm$ 0.01
pH	5.9 $\pm$ 0.05
Ash value (%)	9.1 $\pm$ 0.04

Values represent mean $\pm$ standard deviation of triplicate determinations (n = 3).

4 Determination of Total Phenolic Content

Table 2. Total phenolic content of *Asparagus officinalis* stem extracts

Extract solvent	Total phenolic content (mg GAE/mg extract)
Ethanolic extract	80.5 ± 0.2
Ethyl acetate extract	18.06 ± 0.2

Values are expressed as mean ± standard deviation of triplicate determinations (n = 3). Total phenolic content was determined using the Folin–Ciocalteu method and expressed as gallic acid equivalents (GAE).

5. Determination of Total Antioxidant Content

Table 3. Total antioxidant activity of *Asparagus officinalis* stem extracts

Extract solvent	IC ₅₀ (µg/mL)
Ethanolic extract	196.6 ± 0.27
Ethyl acetate extract	201.19 ± 0.27
Ascorbic acid (standard)	55.19 ± 0.01

Values represent mean ± standard deviation of triplicate determinations (n = 3). IC₅₀ denotes the concentration required to inhibit 50% of DPPH free radicals.

The total antioxidant activity of *Asparagus officinalis* stem extracts is presented in Table 3. The ethanolic extract exhibited a slightly lower IC₅₀ value (196.6 ± 0.27 µg/mL) compared to the ethyl acetate extract (201.19 ± 0.27 µg/mL), indicating marginally higher free radical scavenging activity. Both extracts demonstrated substantial antioxidant potential, although the activity was lower than that of the reference standard, ascorbic acid (IC₅₀ = 55.19 ± 0.01 µg/mL). The enhanced activity of the ethanolic extract is consistent with its higher total phenolic content, suggesting that phenolic compounds contribute significantly to the antioxidant capacity of *A. officinalis* stems.

6 Antimicrobial Activity

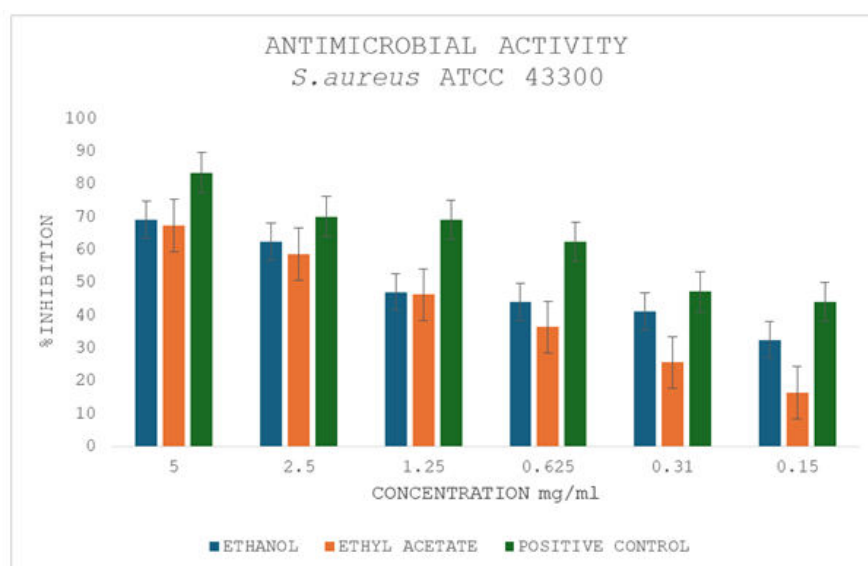


Figure 1. Dose-dependent antimicrobial activity of *Asparagus officinalis* stem extracts against *S. aureus* ATCC 43300. Bars represent percentage inhibition at different concentrations of ethanolic extract (blue) and ethyl acetate extract (orange) compared with the positive control (green). Error bars indicate standard deviation of triplicate experiments (n = 3).

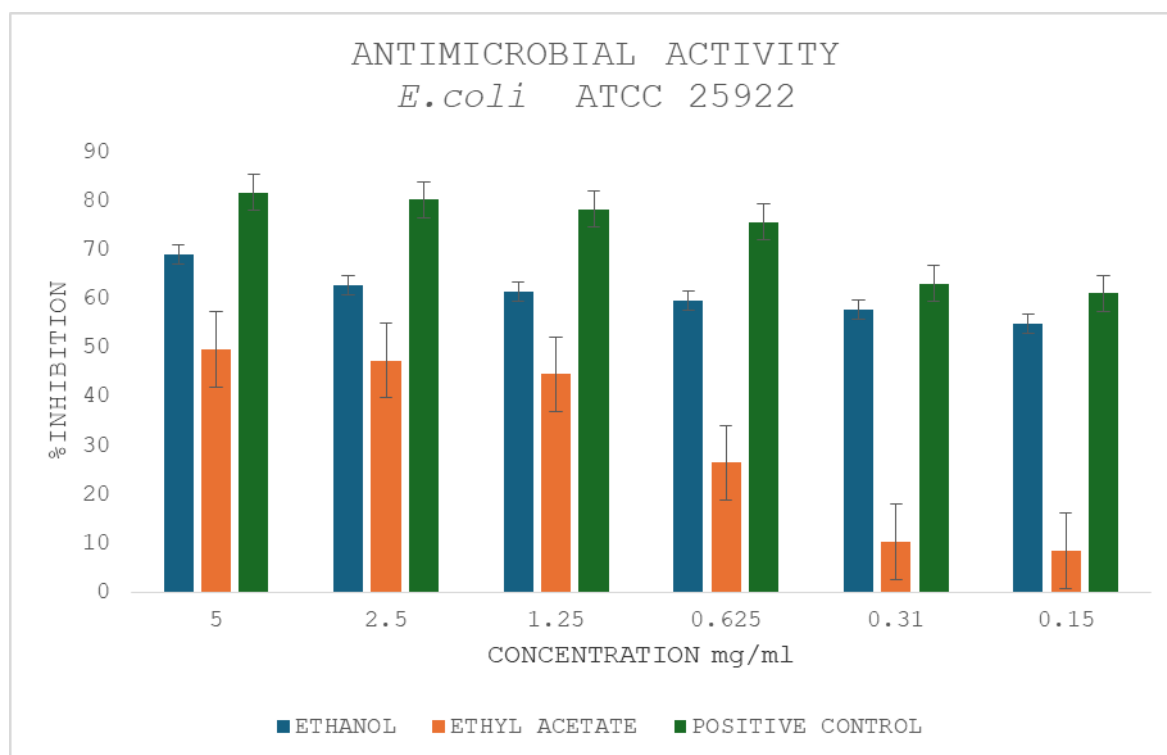


Figure 2. Dose-dependent antimicrobial activity of *Asparagus officinalis* stem extracts against *E. coli* ATCC 25922. Bars represent percentage inhibition at different concentrations of ethanolic extract (blue) and ethyl acetate extract (orange) compared with the positive control (green). Error bars indicate standard deviation of triplicate experiments ($n = 3$).

3.7 Antibiofilm Activity

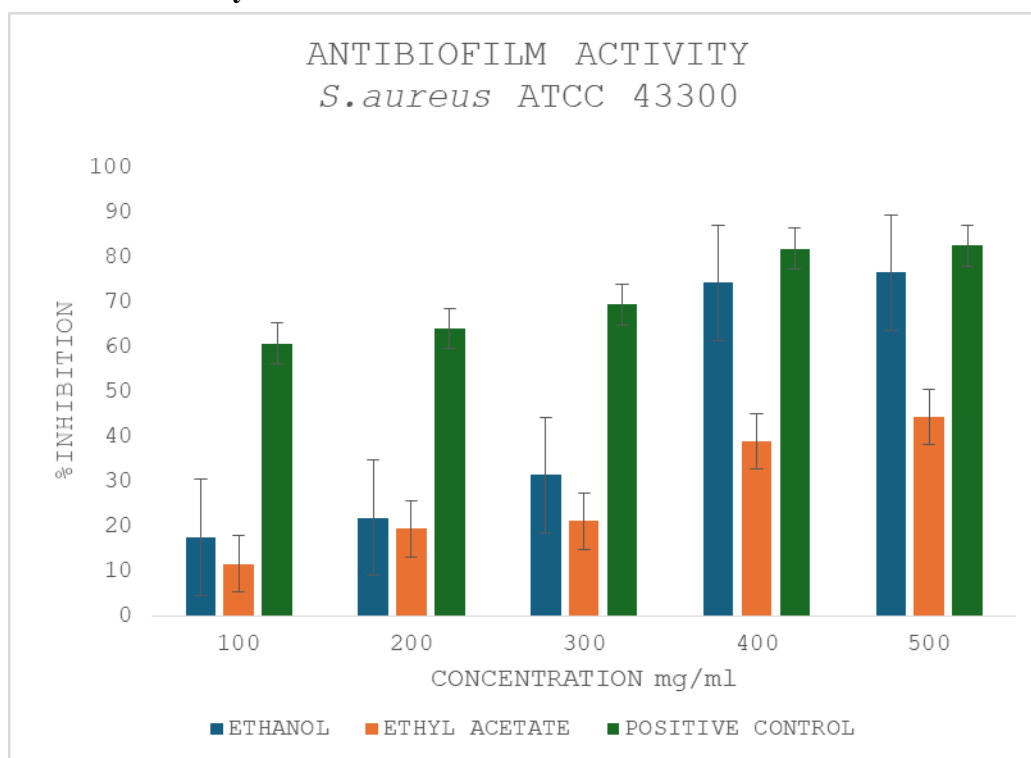


Figure 3. Dose-dependent antibiofilm activity of *Asparagus officinalis* stem extracts against *S. aureus* ATCC 43300. Bars represent percentage inhibition at different concentrations of ethanolic extract (blue) and ethyl acetate extract (orange) compared with the positive control (green). Error bars indicate standard deviation of triplicate experiments ($n = 3$).

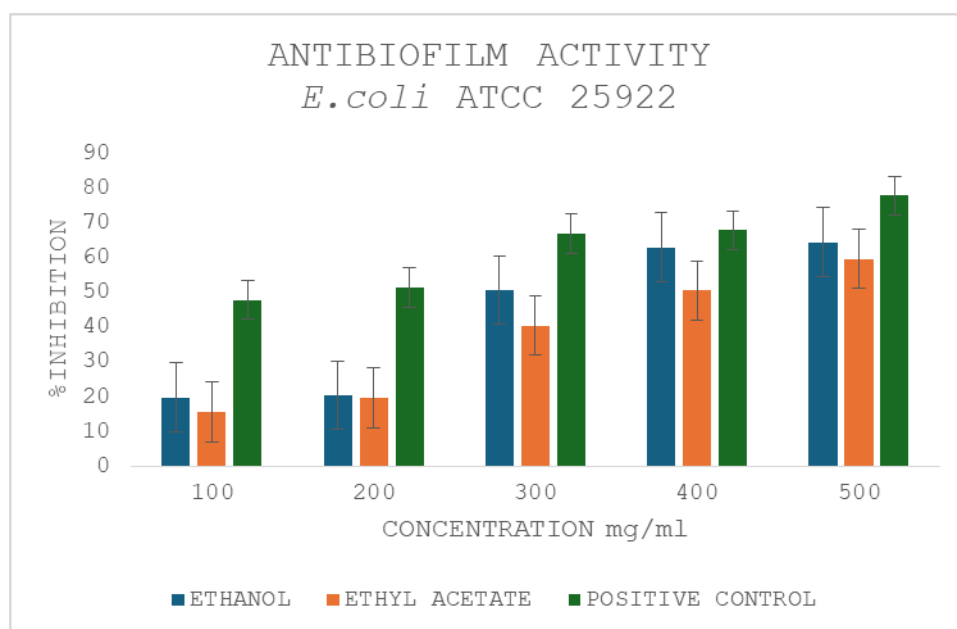


Figure 4. Dose-dependent antibiofilm activity of *Asparagus officinalis* stem extracts against *E. coli* ATCC 25922. Bars represent percentage inhibition at different concentrations of ethanolic extract (blue) and ethyl acetate extract (orange) compared with the positive control (green). Error bars indicate standard deviation of triplicate experiments (n = 3).

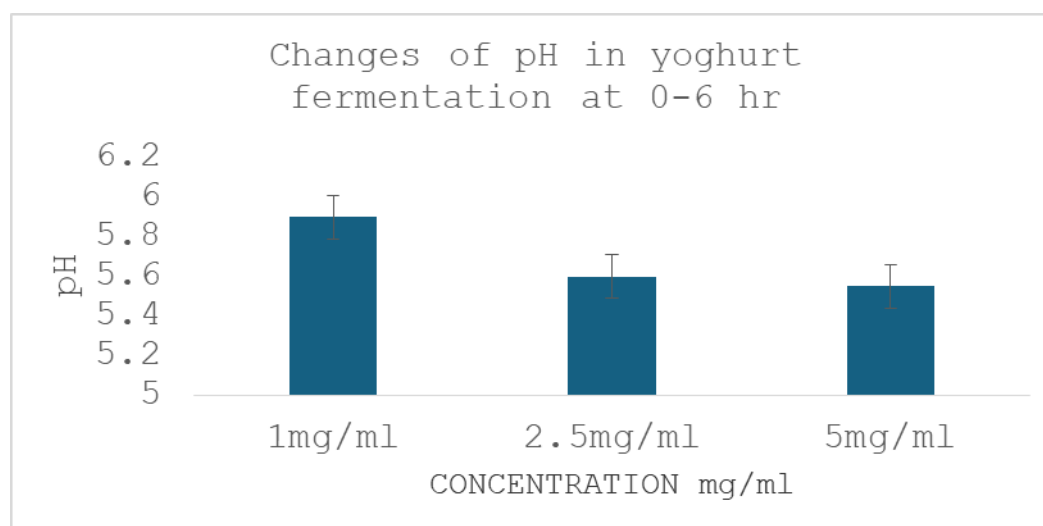


Figure 5. Effect of different concentrations of *Asparagus officinalis* stem Ethanol extract on pH changes during yoghurt fermentation over 0–6 hours. Error bars indicate standard deviation of triplicate measurements (n = 3).

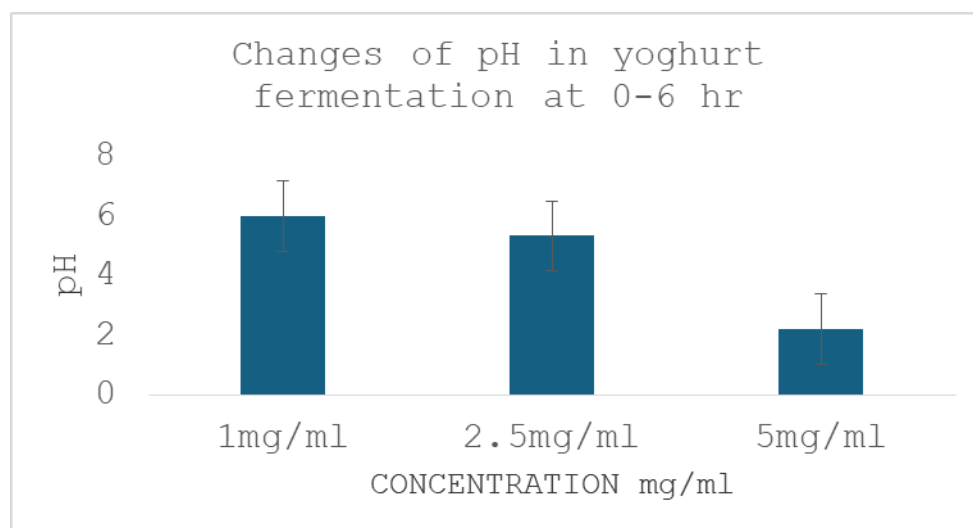


Figure 6. Effect of different concentrations of *Asparagus officinalis* stem Ethyl acetate extract on pH changes during yoghurt fermentation over 0–6 hours. Error bars indicate standard deviation of triplicate measurements ($n = 3$).

Discussion

The current research shows that extracts from *Asparagus officinalis* stems, especially the ethanolic extract, have significant bioactive properties, emphasizing the importance of asparagus stems as an underutilized plant resource. The elevated total phenolic content found in the ethanolic extract aligned with its enhanced antioxidant activity, reaffirming the robust link between phenolic compounds and free radical scavenging ability, as commonly noted for *Asparagus* species and various medicinal plants (Huang et al., 2019; Daglia, 2012). Ethanol is acknowledged as an effective solvent for extracting polar phenolics, which accounts for the notable distinction between ethanolic and ethyl acetate extracts (Azwanida, 2015). The moderate antimicrobial effect observed against *Staphylococcus aureus* and *Escherichia coli* indicates the existence of phenolics and flavonoids that can interfere with bacterial membranes and metabolic functions, as Gram-positive bacteria exhibit greater vulnerability because of variances in cell wall structures (Cushnie & Lamb, 2011; Olivier et al., 2017). The dose-dependent inhibition of biofilm formation, especially with ethanolic extracts, emphasizes the potential of *A. officinalis* stem phytochemicals to disrupt bacterial adhesion and quorum-sensing processes, a feature that is rarely documented for asparagus stems and holds importance for food and biomedical uses (Donlan, 2002; Olawuwo et al., 2019). Additionally, the increased acidification observed during yoghurt fermentation suggests that stem extracts might function as agents that promote fermentation, likely because of phenolic compounds and inulin-like components that encourage the growth and metabolic activity of lactic acid bacteria (Shah, 2007; Martínez-Villaluenga et al., 2020).

Conclusion:

In summary, these results confirm that *A. officinalis* stems are a valuable source of natural antioxidants, antimicrobial and antibiofilm agents, offering further functional advantages in fermented food systems, reinforcing their potential use in the food, pharmaceutical, and nutraceutical sectors while aiding in the sustainable use of agricultural by-products (Guo et al., 2020; Singh et al., 2018).

References

1. Olawuwo, O. S., Famuyide, I. M., & McGaw, L. J. (2022). Antibacterial and antibiofilm activity of selected medicinal plant leaf extracts against pathogens implicated in poultry diseases. *Frontiers in Veterinary Science*, 9, 820304.
2. Olivier, M. T., Muganza, F. M., Shai, L. J., Gololo, S. S., & Nemutavhanani, L. D. (2017). Phytochemical screening, antioxidant and antibacterial activities of ethanol extracts of *Asparagus suaveolens* aerial parts. *South African Journal of Botany*, 108, 41–46.
3. Prakash, S., Venkatesan, A., Kathirvel, A., & Sujatha, V. Identification of isolated bioactive compound and antioxidant profile of *Asparagus racemosus* stem extracts.
4. Karunarathne, Y. A. U. D., Arawwawala, L. D. A. M., Amarasinghe, A. P. G., Weerasooriya, T. R., & Samarasingha, U. K. A. (2019). Physicochemical, phytochemical, and nutritional profiles of root powder of *Asparagus racemosus*. *Asian Journal of Pharmacognosy*, 3(3), 29–35.
5. Kishor, K., et al. (2019). Characterization of Shatavarin-IV in root extracts of *Asparagus racemosus* by UHPLC. *Journal of Pharmacognosy and Phytochemistry*, 8(5), 272–275.
6. Nagalakshmi, S., Saranraj, P., & Sivasakthivelan, P. (2019). Determination of minimum inhibitory concentration (MIC) of essential oils against bacterial pathogens. *Journal of Drug Delivery and Therapeutics*, 9(3), 33–35.
7. Guo, Q., et al. (2020). Bioactive compounds and biological functions of *Asparagus officinalis* L. – A review. *Journal of Functional Foods*, 65, 103727.
8. Inulin and fermentation study. Scribd document.
9. Cushnie, T. P. T., & Lamb, A. J. (2011). Recent advances in understanding the antibacterial properties of flavonoids. *International Journal of Antimicrobial Agents*, 38, 99–107.
10. Daglia, M. (2012). Polyphenols as antimicrobial agents. *Current Opinion in Biotechnology*, 23, 174–181.
11. Huang, X., et al. (2019). Phenolic profile and antioxidant activity of *Asparagus officinalis*. *Journal of Food Science*, 84, 175–183.
12. Martínez-Villaluenga, C., et al. (2020). Phenolic compounds and their role in lactic acid fermentation. *Food Research International*, 132, 109–119.
13. Sharma, V., et al. (2014). Therapeutic potential of *Asparagus* species: A review. *Journal of Ethnopharmacology*, 155, 54–67.
14. Singh, R., et al. (2018). Valorization of agricultural by-products of *Asparagus officinalis*. *Industrial Crops and Products*, 120, 95–102.
15. Azwanida, N. N. (2015). A review on the extraction methods use in medicinal plants, principle, strength and limitation. *Medicinal & Aromatic Plants*, 4, 196.
16. Harborne, J. B. (1998). *Phytochemical Methods: A Guide to Modern Techniques of Plant Analysis* (3rd ed.). Springer, London.
17. Sasidharan, S., et al. (2011). Extraction, isolation and characterization of bioactive compounds from plants' extracts. *African Journal of Traditional, Complementary and Alternative Medicines*, 8, 1–10.
18. AOAC (2019). *Official Methods of Analysis* (21st ed.). Association of Official Analytical Chemists, Washington, DC.
19. Nielsen, S. S. (2017). *Food Analysis* (5th ed.). Springer, New York.
20. Ranganna, S. (2010). *Handbook of Analysis and Quality Control for Fruit and Vegetable Products* (2nd ed.). Tata McGraw-Hill, New Delhi.
21. AOAC (2019). *Official Methods of Analysis* (21st ed.). Association of Official Analytical Chemists, Washington, DC.

22. WHO (2011). *Quality Control Methods for Herbal Materials*. World Health Organization, Geneva.
23. Kokate, C. K., Purohit, A. P., & Gokhale, S. B. (2010). *Pharmacognosy* (45th ed.). Nirali Prakashan, Pune.
24. Singleton, V. L., Orthofer, R., & Lamuela-Raventós, R. M. (1999). Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin–Ciocalteu reagent. *Methods in Enzymology*, 299, 152–178.
25. Wolfe, K., Wu, X., & Liu, R. H. (2003). Antioxidant activity of apple peels. *Journal of Agricultural and Food Chemistry*, 51, 609–614.
26. Everette, J. D., et al. (2010). Thorough study of reactivity of various compound classes toward the Folin–Ciocalteu reagent. *Journal of Agricultural and Food Chemistry*, 58, 8139–8144.
27. Brand-Williams, W., Cuvelier, M. E., & Berset, C. (1995). Use of a free radical method to evaluate antioxidant activity. *LWT – Food Science and Technology*, 28, 25–30.
28. Mensor, L. L., et al. (2001). Screening of Brazilian plant extracts for antioxidant activity by the use of DPPH free radical method. *Phytotherapy Research*, 15, 127–130.
29. Prior, R. L., Wu, X., & Schaich, K. (2005). Standardized methods for the determination of antioxidant capacity. *Journal of Agricultural and Food Chemistry*, 53, 4290–4302.
30. CLSI (2022). *Performance Standards for Antimicrobial Susceptibility Testing* (32nd ed.). Clinical and Laboratory Standards Institute, Wayne, PA.
31. Wiegand, I., Hilpert, K., & Hancock, R. E. W. (2008). Agar and broth dilution methods to determine the minimal inhibitory concentration (MIC) of antimicrobial substances. *Nature Protocols*, 3, 163–175.
32. Andrews, J. M. (2001). Determination of minimum inhibitory concentrations. *Journal of Antimicrobial Chemotherapy*, 48, 5–16.
33. Olawuwo, O. S., et al. (2019). Antibiofilm activity of plant extracts against pathogenic bacteria. *Journal of Applied Microbiology*, 126, 115–124.
34. Stepanović, S., et al. (2007). Quantification of biofilm in microtiter plates: overview of testing conditions and practical recommendations. *APMIS*, 115, 891–899.
35. Donlan, R. M. (2002). Biofilms: microbial life on surfaces. *Emerging Infectious Diseases*, 8, 881–890.
36. Olawuwo, O. S., et al. (2019). Antibiofilm activity of plant extracts against pathogenic bacteria. *Journal of Applied Microbiology*, 126, 115–124.
37. Stepanović, S., et al. (2007). Quantification of biofilm in microtiter plates: overview of testing conditions and practical recommendations. *APMIS*, 115, 891–899.
38. Donlan, R. M. (2002). Biofilms: microbial life on surfaces. *Emerging Infectious Diseases*, 8, 881–890.
39. Tamime, A. Y., & Robinson, R. K. (2007). *Yoghurt: Science and Technology* (3rd ed.). Woodhead Publishing, Cambridge.
40. Shah, N. P. (2007). Functional cultures and health benefits. *International Dairy Journal*, 17, 1262–1277.
41. Martínez-Villaluenga, C., et al. (2020). Phenolic compounds as modulators of lactic acid bacteria growth in fermented foods. *Food Research International*, 132, 109–119.
42. Lourens-Hattingh, A., & Viljoen, B. C. (2001). Yoghurt as probiotic carrier food. *International Dairy Journal*, 11, 1–17.

60. Antibiotic Target Protein Prediction for *Enterococcus Faecium* Using Network Pharmacology

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Abstract: This study aimed to predict antibiotic target proteins in *Enterococcus faecium* due to its cell surface and secreted virulence factors. Utilizing the network pharmacology methodology, rank the proteins according to their text-mining scores and offer a variety of genes that may be found using genome sequencing methods. This procedure was carried out using Cytoscape software version 3.10.4. Networks were imported from public databases using STRING; a PubMed query as the data source. Antibiotic Resistance *Enterococcus* was the PubMed query, and *Streptococcus pyogenes* was the chosen species. The maximum number of *Enterococcus faecium* target proteins that could be downloaded was fifty, and the confidence score was set at 0.7. Target proteins from the *Enterococcus faecium* species, often discussed in abstracts, were among the 5,624 results downloaded from the PubMed query. Proteins are represented as nodes in networks, while interactions between the proteins are shown by edges. Target proteins for *Enterococcus faecium*, such as Pbp5 (14.50), acm (13.39), esp (12.91), msrC (8.70), purK (8.00), TetL (7.82), vanZ (6.87), gyrA (5.65), pstS (5.44), entA (5.14), and cls (4.99), were shown in descending order of high text mining scores. The target proteins' frequency of mention in the STRING database was indicated by the text-mining score; the most often mentioned protein, Pbp5, had the highest score of 14.50. Using the network pharmacology technique, Pbp5, acm, esp, msrC, purK, TetL, vanZ, gyrA, pstS, entA, and cls were identified as proteins having strong text-mining evidence in *Enterococcus faecium* resistance.

Introduction

Microorganisms belonging to the *Enterococcus* genus, commonly referred to as 'enterococci,' were first identified by Thiercelin in 1899, who termed them 'entérocoque' to highlight their source originating from the intestines. Nevertheless, the classification of these bacteria has consistently been unclear. The advancement of genetic techniques aimed at exploring the evolutionary connections among lactic acid bacteria linked to food has led to significant alterations in their classification (1). Because of their importance in food, animal feed, environmental situations, and clinical specimens, the identification and counting of *Enterococci* have emerged as a critical concern not just in everyday practices but also in ongoing research endeavors. Various media and procedures have been introduced for different objectives, yet no one method satisfies all the necessary criteria universally (2). *Enterococci* are bacteria that are Gram-positive and negative for catalase, exhibiting a homofermentative capacity to generate lactic acid through the primary metabolic conversion of glucose. They are significant in various fields, including food, healthcare, and ecological systems, through several mechanisms. In the past, this genus was discovered and extracted from multiple sources such as flora, fauna, soil, water, and sources of fermented foods (3). At pH 9.6, *E. faecium* and *E. faecalis* grew well and fermented adonitol in a unique way. Both *E.*

faecalis and *E. faecium* showed 15 minutes of survival at 60°C, with *E. faecium* exhibiting limited survival at 30 minutes. Isolation and Identification of *Enterococcus faecium* from the Gastrointestinal Tract of the Common Carp (*Cyprinus carpio*) and the Nile Tilapia (4). Enterococci, typically present in various surroundings, lead to significant infections even though they lack clearly identified virulence factors and toxins. Understanding the virulence characteristics of *Enterococci* is crucial for comprehending the intricate pathogenic frameworks (5).

They can be found in water, soil, food items (including dairy and meat), sewage, and the digestive tracts of both humans and animals. Vancomycin-resistant *Enterococci* (VRE), which were first discovered in the 1980s, are currently a significant contributor to hospital-acquired infections, particularly in intensive care units and among patients with impaired immune systems. They are acknowledged as the second most frequent cause of nosocomial infections worldwide (6). Enterococcus strains that appear phenotypically susceptible to vancomycin but include glycopeptide resistance genes are known as vancomycin-variable *Enterococci* (VVE). These bacteria provide a serious diagnostic and treatment challenge as well as a risk of treatment failure. Molecular techniques can identify these strains, which are frequently undetected by traditional susceptibility measures (7). *Enterococci* were identified in the intestinal flora of humans in the year 1899. Nonetheless, it wasn't until 1984 that they were recognised as separate from the genus *Enterococci*. The microorganism *Enterococcus faecalis* was first mentioned in 1906 following its isolation from a patient suffering from endocarditis. In 1919, *Enterococcus faecium* was also identified for the first time. Subsequently, *Enterococci* within serogroup D were categorized into two distinct groups. This classification was based on investigations that showcased variations in biochemical attributes as well as differences observed through nucleic acid analysis, including DNA-rRNA homology and 16S rRNA studies. Both *Enterococcus faecalis* and *Enterococcus faecium* were classified under the *Enterococcus* category, which encompasses over 50 species (8). Only a small number of the more than 17 species in the genus *Enterococcus* produce clinical infections in people. The two most prevalent species of *Enterococcus* that infect humans are *E. faecalis* (80–90%) and *E. faecium* (5–10%). However, the isolation rate of *E. faecium* has recently increased, which is concerning because its inherent resistance may result in treatment failure (9).

E. faecium Bloodstream infection has greater rates of antibiotic resistance and mortality than *E. faecalis* BSIs caused by vancomycin-resistant *Enterococci* (VRE) species are among the significant hospitalized infections that are potentially fatal (10). The opportunistic microorganism *Enterococcus faecium* inhabits humans and various animal species, withstands multiple types of stress, evades antibiotic therapies, and tenaciously remains in clinical settings. The extensive use of antibiotics in healthcare facilities and farming has played a role in the rise of vancomycin-resistant *E. faecium*, responsible for numerous infections acquired within hospitals (11). When compared to infections produced by vancomycin-susceptible enterococci, vancomycin resistance in *E. faecium* limits treatment options and is linked to higher mortality and healthcare costs (12). Among the group of enterococci, *Enterococcus faecium* and *Enterococcus faecalis* are the most important clinically as agents of hospital infections, with *E. faecium* displaying a higher prevalence of glycopeptide resistance compared to *E. faecalis* (13).

Enterococcus faecium is a type of Gram-positive bacterium that forms a significant part of the gut microbiome in both humans and animals. This organism can act as an opportunistic pathogen, leading to serious infections, especially in individuals receiving treatment in

hospitals. During the 1980s, the emergence of vancomycin-resistant *E. faecium* (VRE_{fm}) likely occurred due to the horizontal transfer of genes that confer resistance to vancomycin, particularly the *vanA* and *vanB* genes. These types of resistance have now been detected across several continents, with a rising occurrence rate (14). Vancomycin-resistant *Enterococci* (VRE) of the *vanA* and *vanB* types are the most common in the globe and are primarily seen in vancomycin-resistant *E. faecium* (VRE_{fm}). The widely distributed *vanB2* subtype gene cluster is linked to Tn1549 integrative conjugative elements (ICEs) that were initially obtained from gut anaerobes, whereas the *vanA* gene cluster is typically a component of the Tn1546 transposon and frequently found on plasmids. Both clonal spread and the interchange of mobile genetic elements (MGEs) probably play significant roles in the complicated mechanisms underlying the spread of VRE_{fm}. (15). There is a significant genetic potential for the species *Enterococcus*, particularly *E. faecium*, to create several bacteriocins, or enterocins. Because enterocins have antibacterial efficacy against significant foodborne pathogens, including *L. monocytogenes*, they have drawn scientific and technological interest (16). A broad range of virulence factors is produced by *E. faecium*. Adhesion factors include pili, enterococcal surface protein (Esp), collagen-binding proteins (Acm), collagen adhesins from *E. faecium* (Scm), serine-glutamate repeat-containing protein A (SgrA), *E. faecium* collagen-binding protein A (EcbA), and aggregation substance (Asa1). These components aid in the production of biofilms and/or efficient bacterial adherence to host cells. Such adherence is thought to be a crucial initiating factor in the emergence of many infectious illnesses(17).

By adopting genetic deterrents, the *Enterococcus* genomes' plasticity enabled them to quickly react and adapt to their surroundings, increasing their capacity to colonies, infect, and cause illness. As treatment options for VRE bacteremia were limited, the emergence of linezolid resistance as a result of selective pressure was concerning. Meanwhile, the emergence of multidrug-resistant (MDR) *Enterococcus* has become a major public health threat (e.g., India, Japan) because it has limited the effective antimicrobial agents available to treat infections such as Vancomycin-resistant *Enterococcus* (VRE), linezolid-resistant *Enterococcus* (LRE), and even the linezolid-resistant *Enterococcus* (LRVRE)(18). Treatment alternatives consist of agents that lack specific approval for VRE, such as chloramphenicol, doxycycline, high-dose ampicillin, and ampicillin/sulbactam, along with nitrofurantoin for infections in the lower urinary tract. The effectiveness of antimicrobial combinations that have demonstrated efficacy in vitro or in animal models still needs to be defined. Two new antimicrobial drugs, quinupristin /dalfopristin and linezolid, have been recognized as approved treatment choices for *Enterococcus faecium* that is resistant to vancomycin, based on in vitro susceptibility tests and clinical effectiveness observed in multicenter trials funded by pharmaceutical companies (19). Apart from its involvement in several infections, *E. faecium*'s innate and acquired resistance is a crucial factor to take into account. Gene mutations and gene transfer from other bacteria are two common ways that this bacterium can develop resistance. One of the primary causes of antibiotic resistance in bacteria is gene transfer. Reduced antibiotic efficacy and higher morbidity and mortality are linked to the emergence of resistance (20).

Materials and Methods

Database Mining and Network Construction

In this method, Cytoscape software (v3.10.4) was utilised. StringApp (v2.2.0) was added to Cytoscape via the App Store. We imported the file from Cytoscape and then connected it to a public database. Networks were imported from public databases using STRING; a PubMed

Genes	proteins	Mining Score	Genes		Mining Score
Pbp5	Penicillin-binding protein	14.500847	AOM16197.1	Collagen-binding protein	2.845087
Acm	Peptidase	13.395452	mecA	Competence negative regulator MecA	2.810553
Esp	Transporter	12.912806	rplV	50S ribosomal protein L22	2.690801
msrC	GAF domain-containing protein	8.702678	glpQ	Glycerophosphodiester phosphodiesterase	2.653172
purK	Phosphoribosylaminoimidazole carboxylase	8.007797	tuf	Elongation factor Tu	2.63078
TetL	<i>Tetracycline resistance protein</i>	7.822944	tnpA	Transposase	2.527452
vanZ	Protein vanZ	6.870283	tnpA-2	Transposase	2.510796
gyrA	DNA gyrase subunit A	5.656825	Pbp2A	Penicillin-binding protein	2.491636
pstS	Phosphate ABC transporter substrate-binding	5.443742	rep_1	Replication protein	2.340363
entA	Enterocin	5.147611	Fms11	Peptidase	2.299292
Cls	Cardiolipin synthase	4.992713	dfrD_1	Dihydrofolate reductase	2.208868
gdh_4	Sugar dehydrogenase	4.401989	pilB	Cell surface protein	2.207567
AOM17606.1	Peptidase	4.270182	folA	Dihydrofolate reductase	2.043385
msrA-2	Peptide methionine sulfoxide reductase	4.138287	AOM16959.1	Dihydrofolate reductase	2.010222
EntB	Isochorismatase	4.003899	yyaP_2	Dihydrofolate reductase	2.003974
rplC	50S ribosomal protein L3	3.657366	AOM17128.1	Relaxase	1.979832
rpsJ	30S ribosomal protein S10	3.622575	radC_1	Hypothetical protein	1.901554
parC	DNA topoisomerase IV	3.513738	bsh	Choloylglycine hydrolase	1.850779
RepA	Replication protein RepA	3.405987	adhE	Acetaldehyde dehydrogenase	1.817582
rplD	50S ribosomal protein L4	3.276118	sigV	RNA polymerase subunit sigma	1.811242
PepQ2	Dipeptidase	3.120485	araC	AraC family transcriptional regulator	1.750405
rlmB	rRNA	3.112169	AOM17342.1	Endonuclease	1.738243

	methyltransferase				
sodA_1	Superoxide dismutase	3.033877	MdlA	Multidrug transporter ABC binding protein	1.723048
AOM16197.1	Collagen-binding protein	3.022904	lysM_3	Peptidase M23	1.723048
efrB	ABC transporter	3	AOM15383.1	Holin	1.723048
AOM17261.1	Collagen-binding protein	2.845087	ade	Adenosine deaminase	1.669464

Discussion

Antimicrobial resistance in bacteria typically becomes genetically programmed. There are several ways that antimicrobial resistance can be genetically encoded, such as overexpression or duplication of preexisting genes, point mutations, or the acquisition of completely new genes by horizontal gene transfer. Antimicrobial resistance genes in genomes and metagenomes can now be identified and characterised more quickly thanks to advancements in next-generation sequencing methods and computational techniques. These new techniques and technologies enable quick and sensitive resistance assessments in both cultivable and uncultivable bacteria, supplementing current culture-based techniques for clinical and surveillance applications (21).

In this study, PPIs come from the STRING database (<https://www.stringdb.org>). STRING was chosen because it encompasses a large variety of bacterial species and may be expanded to accommodate additional species. The Cytoscape software was used to rank the target proteins based on their text-mining scores, with Pbp5, acm, esp, msrC, purK, TetL, vanZ, gyrA, pstS, entA, and cls having high text-mining scores, as seen in Table 1. The *Enterococcus faecium* target proteins' frequency of mention in the downloaded abstract was indicated by the text mining score, with the most often mentioned protein (Pbp5) receiving the highest score. According to the report, Penicillin-binding proteins (PBPs) are essential for the catalysis of peptidoglycan formation in bacteria, giving them structural integrity and the ability to tolerate high intracellular pressures. PBPs play a role in the manufacture of peptidoglycan during septum development and cell wall elongation. PBPs serve as both transglycosylases, which polymerise glycan strands, and transpeptidases, which cross-link glycan chains. Amino acid alterations impact PBPs' ability to bind penicillin, which has been demonstrated to lessen GAS susceptibility to penicillin. Antibiotic-resistant strains have been identified, and subsequent genetic studies have revealed that they exhibit chimeric high molecular mass penicillin binding proteins (HMM PBPs) in contrast to susceptible strains (22).

From our findings, another protein with a high text-mining score is the acm protein. In furtherance of being involved in the breakdown of off-function proteins in lysosomes, the cytosol, plasma membranes, and extracellular space, peptidases may also play regulatory functions in biological processes that are essential for maintaining cell homeostasis. Apart from their involvement in normal protein turnover, their dysfunction has been linked to several pathological processes, such as cancer, immune and cardiovascular disorders, rheumatoid arthritis, osteoarthritis, atherosclerosis, periodontitis, pancreatitis, osteoporosis, diseases of inadequate lysosomal protein degradation, and more (23). The text-mining evidence for msrC, purK, TetL, vanZ and gyrA was high in our results as well. GAF domain

(msrC) is defined as a conserved region of approximately 150 amino acids found in various signalling molecules, often associated with multidomain signalling proteins and capable of binding cyclic nucleotides or tetrapyrroles in specific contexts. Phosphoribosyl-aminoimidazole carboxylase (purK): Catalyzes the ATP-dependent conversion of 5-aminoimidazole ribonucleotide (AIR) and HCO₃⁽⁻⁾ to N⁵-carboxyaminoimidazole ribonucleotide (N⁵-CAIR). The history of tetracycline (TetL) resistance in human infections has been marked by cycles in which the introduction of new tetracycline antibiotic generations is swiftly followed by the identification of novel resistance mechanisms. The first tetracycline-resistant bacteria, *Shigella dysenteriae*, were identified in 1953 and the first multidrug-resistant *Shigella* in 1955, after first-generation tetracyclines were discovered in 1948. Tetracycline efflux pumps (EFF) were later shown to be encoded by these pathogens. Tetracycline EFFs are members of the major facilitator superfamily of membrane-associated proteins. EFFs shield ribosomes from tetracycline binding by exporting tetracyclines from the cell, which lowers intracellular tetracycline concentration. With at least 33 identified determinants, EFFs have historically been the most prevalent tetracycline-resistance mechanism, accounting for 60% of all tet and otr genes in 2005 (24). A protein called VanZ contributes to gram-positive bacteria's resistance to glycopeptide antibiotics like teicoplanin and vancomycin. It is essential for the alteration of peptidoglycan precursors because it stops the terminal D-Ala from being incorporated into these precursors, which lowers the antibiotics' binding affinity. The genomes of several clinically significant bacteria, such as *Enterococcus faecium* and *Streptococcus pneumoniae*, contain VanZ, which is a member of the VanA glycopeptide resistance gene cluster. One potential tactic to fight antibiotic resistance in these bacteria is to target VanZ (25). The text-mining evidence for gyrA was also strong in our findings. A type II topoisomerase that negatively supercoils closed circular double-stranded (ds) DNA in an ATP-dependent manner to modulate DNA topology and maintain chromosomes in an underwound state. Negative supercoiling favours strand separation, and DNA replication, transcription, recombination and repair, all of which involve strand separation. Also able to catalyze the interconversion of other topological isomers of dsDNA rings, including catenanes and knotted rings. Type II topoisomerases break and join 2 DNA strands simultaneously in an ATP-dependent manner.

Conclusion

We identified Pbp5, acm, esp, msrC, purK, TetL, vanZ, gyrA, pstS, entA, and cls as proteins with strong text-mining evidence in *Enterococcus faecium* resistance using the network pharmacology approach.

References

1. Giraffa G. ENTEROCOCCUS. In: Encyclopedia of Food Microbiology. Elsevier; 1999. p. 617–24.
2. Domig KJ, Mayer HK, Kneifel W. Methods used for the isolation, enumeration, characterisation and identification of *Enterococcus* spp. *Int J Food Microbiol*. 2003 Dec;88(2–3):147–64.
3. Kavitate D, Devi PB, Delattre C, Reddy GB, Shetty PH. Exopolysaccharides produced by *Enterococcus* genus — An overview. *Int J Biol Macromol*. 2023 Jan;226:111–20.
4. et al. M. Isolation and Identification of *Enterococcus faecium* from the Gastrointestinal Tract of the Common Carp (*Cyprinus carpio*) and the Nile Tilapia (*Oreochromis niloticus*). *Egypt J Aquat Biol Fish*. 2024 Sep 1;28(5):805–14.

5. Gök ŞM, Türk Dağı H, Kara F, Arslan U, Fındık D. Klinik Örneklerden İzole Edilen *Enterococcus faecium* ve *Enterococcus faecalis* İzolatlarının Antibiyotik Direnci ve Virülans Faktörlerinin Araştırılması. *Mikrobiyol Bul.* 2020 Jan 15;54(1):26–39.
6. Almasaud LD, Alkhulaifi MM, Ghazawi A, Strepis N, Manzoor A, Senok A, et al. High-resolution genomic and molecular characterization of vancomycin-resistant *Enterococci* from hospitalized patients in a tertiary care center in Riyadh, Saudi Arabia. *Sci Rep.* 2025 Nov 13;15(1):39764.
7. GÜRLER M, KARAHAN ZC, EVREN E, TEKELİ FA. Vankomisin Duyarlı Enterokok İzolatlarında vanA/ vanB Genleri: Moleküler Tanımlama ve Tanı Yöntemlerinin Değerlendirilmesi. *Mikrobiyol Bul.* 2025 Oct 24;59(4):425–36.
8. Zhou X, Willems RJL, Friedrich AW, Rossen JWA, Bathoorn E. *Enterococcus faecium*: from microbiological insights to practical recommendations for infection control and diagnostics. *Antimicrob Resist Infect Control.* 2020 Dec 10;9(1):130.
9. Kumar V, tha A, Agrawal A, Chahar Y, Shree N. Isolation and Identification of *Enterococcus* Species from Various Clinical Samples and Their Antimicrobial Susceptibility Pattern at a Tertiary Health Care Hospital in Agra. *Int J Curr Microbiol Appl Sci.* 2018 Nov 20;7(11):1720–8.
10. Jabbari Shiadeh SM, Pormohammad A, Hashemi A, Lak P. <p>Global prevalence of antibiotic resistance in blood-isolated Enterococcus faecalis and Enterococcus faecium: a systematic review and meta-analysis</p>. *Infect Drug Resist.* 2019 Sep;Volume 12:2713–25.
11. Wei Y, Palacios Araya D, Palmer KL. *Enterococcus faecium*: evolution, adaptation, pathogenesis and emerging therapeutics. *Nat Rev Microbiol.* 2024 Nov 18;22(11):705–21.
12. Vuichard-Gysin D, Büchler AC, Blanc DS, Keller PM, Schläpfer P, Kronenberg A, et al. Emergence of vancomycin-resistant *Enterococcus faecium* vanA ST612 with reduced daptomycin susceptibility, Switzerland, 2018 to 2024. *Eurosurveillance.* 2025 Nov 13;30(45).
13. Böing CW, Schneider JS, Froböse NJ, Jurke A, Kampmeier S, Mellmann A. Virulome and genome-wide association study of vancomycin-resistant *Enterococcus faecium* from bloodstream infections versus colonization isolates. *International Journal of Medical Microbiology.* 2025 Dec;321:151683.
14. Wei Y, Palacios Araya D, Palmer KL. *Enterococcus faecium*: evolution, adaptation, pathogenesis and emerging therapeutics. *Nat Rev Microbiol.* 2024 Nov 18;22(11):705–21.
15. Both A, Kruse F, Mirwald N, Franke G, Christner M, Huang J, et al. Population dynamics in colonizing vancomycin-resistant *Enterococcus faecium* isolated from immunosuppressed patients. *J Glob Antimicrob Resist.* 2022 Mar;28:267–73.
16. Schittler L, Perin LM, de Lima Marques J, Lando V, Todorov SD, Nero LA, et al. Isolation of *Enterococcus faecium*, characterization of its antimicrobial metabolites and viability in probiotic Minas Frescal cheese. *J Food Sci Technol.* 2019 Nov 8;56(11):5128–37.
17. Sun H, Fang Y, Hu S, Lyu D, Dong X, Wang J. Molecular epidemiology, antimicrobial resistance, and virulence determinants of *Enterococcus faecium* bloodstream isolates: a 7-year study in Shandong, China. *BMC Microbiol.* 2025 Nov 19;25(1):765.
18. Pan P, Sun L, Shi X, Huang X, Yin Y, Pan B, et al. Analysis of molecular epidemiological characteristics and antimicrobial susceptibility of vancomycin-resistant and linezolid-resistant *Enterococcus* in China. *BMC Med Genomics.* 2024 Jul 1;17(1):174.

19. Linden PK. Treatment Options for Vancomycin- Resistant Enterococcal Infections. *Drugs*. 2002;62(3):425–41.
20. Huang C, Moradi S, Sholeh M, Tabaei FM, Lai T, Tan B, et al. Global trends in antimicrobial resistance of *Enterococcus faecium*: a systematic review and meta-analysis of clinical isolates. *Front Pharmacol*. 2025 Apr 7;16.
21. Aluefua OF, Yunusa A, Abubakar RZ. Prediction of antibiotic target proteins of *Streptococcus pyogenes* using network pharmacology approach. *Sokoto J Med Lab Sci*. 2025 Jun 16;10(1):247–52.
22. Yu D, Guo D, Zheng Y, Yang Y. A review of penicillin binding protein and group A *Streptococcus* with reduced- β -lactam susceptibility. *Front Cell Infect Microbiol*. 2023 Mar 31;13.
23. Kos J. Peptidases: Role and Function in Health and Disease. *Int J Mol Sci*. 2023 Apr 25;24(9):7823.
24. Blake KS, Xue YP, Gillespie VJ, Fishbein SRS, Tolia NH, Wencewicz TA, et al. The tetracycline resistome is shaped by selection for specific resistance mechanisms by each antibiotic generation. *Nat Commun*. 2025 Feb 7;16(1):1452.
25. Vimberg V, Zieglerová L, Buriánková K, Branny P, Balíková Novotná G. VanZ Reduces the Binding of Lipoglycopeptide Antibiotics to *Staphylococcus aureus* and *Streptococcus pneumoniae* Cells. *Front Microbiol*. 2020 Apr 3;11.

EPILOGUE

The purpose of this part of the book is to draw attention of the readers to key indicators given in each of the preceding chapters sequentially. The number coincides with the number of chapters in the book.

1	The Internet of Things has progressed from conceptual origins in the late 20th century to a transformative force in the 21st century. Its synergy with 5G/6G, AI, edge computing, smart cities, automation, and global connectivity demonstrates its central role in shaping the digital future.
2	Although AI has the potential to improve cybersecurity and safeguard private data, its improper use or uncontrolled application may jeopardize people's right to privacy. As a result, a balanced strategy that fosters innovation while guaranteeing accountability, transparency, and robust data protection measures is crucial.
3	This work shows that a lightweight and effective method for identifying fake images may be achieved by combining MobileNetV2 feature extraction with a Logistic Regression classifier.
4	The technologies developed – for example, Machine Learning, Natural Language Processing, Computer Vision and Predictive enable the development of intelligent mobile applications with the capability of learning from their users.
5	Computer vision allows machines to interpret visual information from cameras and sensors, while robotics represents the physical side that can act on this input. Combined they provide systems able to observe, interpret and act in dynamic populated real-world scenes more accurately and efficient.
6	Artificial Intelligence is witnessing widespread adoption as a crucial tool/technology that is recognized to be of immense importance in the healthcare industry, including its application for Cancer detection and prevention as well.
7	Student-centered, experiential, and technology-enabled approaches contribute meaningfully to improved learning outcomes and teaching effectiveness.
8	Federated Learning has emerged as a promising replacement for traditional centralized data processing by facilitating joint model development while maintaining information location and organizational control.
9	Convolution Neural Networks (CNNs) with Vision Transformers (ViTs) techniques used for detecting forged tumors in radiological scans. CNNs excel at identifying local texture anomalies and spatial irregularities, whereas ViTs capture broader contextual patterns and distant dependencies across images
10	As digital medical records and healthcare data grow, machine learning has become an important tool in medical research and diagnosis.
11	NLP-based automation is a practical and effective solution for building efficient, scalable, and customer-focused service systems.
12	Machine learning classifiers are trained to differentiate between benign and malicious behaviors based on these features. experimental results demonstrate that the proposed approach achieves effective detection accuracy while reducing false positives compared to traditional methods.
13	The face recognition-based attendance system has been designed for the purpose of reducing the errors that occur in the manual attendance taking system.
14	Lazy predict can be a handy tool for selecting the best algorithm among all ML models and is also most suitable for predicting response variables before testing against the hyperparameters.
15	According to Forbes Technology Council (2024), AI-powered interview tools significantly enhance candidates' readiness by offering automated, feedback-driven

	practice (Forbes+1).
16	Fraud detection is defined as a set of data-driven techniques designed to identify and mitigate unauthorized or deceptive digital banking activities while maintaining an optimal balance between security, user experience, privacy, and regulatory compliance.
17	CropBoom is an AI-powered web-based agricultural marketplace that connects farmers directly with buyers through a transparent and data-driven digital platform which uses Google Gemini AI for crop image analysis, soil data interpretation, and weather-assisted decision support.
18	Har Ghar Pooja, a digital, AI-enabled platform that connects devotees with verified Pandits, enables seamless booking of rituals, ensures secure digital payments, and offers virtual puja participation via live streaming.
19	Portland Pozzolana Cement is used along with steel fibre and basalt fibre to develop hybrid fibre reinforced concrete.
20	Water-cement proportion governs the durability as well as workability properties of self-consolidating concrete mix.
21	Existing speaker recognition and deepfake detection systems frequently struggle when applied across different languages, recording conditions, or synthesis techniques.
22	Adoption of forensic-style validation protocols can ensure consistency, safety, and therapeutic reliability of traditional herbal products such as Chaturbeeja
23	Simultaneous Authentication of Equals (SAE) in WPA3 completely stopped the offline cracking process.
24	DNA testing to trace ivory, chemical analysis to track timber , ballistics to match weapons, and digital tools like satellites and mobile data—strong evidence could be collected to thwart organized crime networks.
25	AI offers speed, consistency and scalability in analysis and assessment that no manual system can match.
26	Silent Witness Ecosystem, a new, interdisciplinary framework for proactive environmental forensics.
27	Barefoot footprint analysis, particularly when integrated with machine learning, provides a reliable, objective, and cost-effective tool for sex determination in forensic investigations.
28	The ever-evolving legal landscape in the field of policing presents a wide variety of challenges, each of which has its own distinct set of repercussions, problems, and possible solutions
29	Techniques such as UV-Vis spectroscopy, Raman Spectroscopy, ATR-FTIR and other chromatographic techniques can characterize pigments, solvent and dyes to obtain their chemical signatures.
30	Toxic plants, notably Lantana camara, pose significant risks for forensic toxicologists and veterinarians due to their harmful impacts on livestock, leading to fatalities and diminished productivity.
31	For PPCP pollution to be addressed at the root, long-term environmental protection, ecosystem stability and public health sustainability will depend on behavioural and policy mitigation measures.
32	Specialized forensic analysis of gait in individuals with knock knees provides a reliable and effective method for creating unique profiles, thereby enhancing the power of traditional forensic gait examination as a tool for personal identification in forensic science.
33	Folk medicine and ethnobotany have long attributed an aphrodisiac, lust-inducing, or potency-boosting properties to a variety of plants and herbs

34	DNA barcoding and advanced genomic tools have emerged as robust, reproducible, and legally defensible techniques for species identification, individualization, population assignment, and geographic origin determination.
35	Virtual vulnerability — the exposure of women and gender nonages to technology- eased violence in digital spaces — represents a critical and evolving challenge for criminology, forensic wisdom, and felonious justice systems worldwide.
36	Using a Kali Linux Virtual Machine (VM), we configured Open5GS (core simulator: AMF, SMF, UPF, NRF). The experimental work involved starting Open5GS services, generating secure VPN key pairs (public, private), establishing server-client tunnels, and updating firewall rules.
37	Four natural materials moong dal (<i>Vigna radiata</i>), supari nut (<i>Areca catechu</i>), coconut husk (<i>Cocos nucifera</i>), and pistachio shells (<i>Pistacia vera</i>) were processed through air-drying, roasting, grinding, and sieving to obtain fine powders suitable for fingerprint development.
38	Implementation of fuzzy DSS into the banking operation process is possible, as they allow the bank managers to manage the risks better, regulate the relations with customers, and assess the employee performance.
39	Graph labeling is a mature yet active field combining deep combinatorial problems, algorithmic challenges, and a surprising range of applications in engineering and coding.
40	The mathematical heritage of India has significantly influenced basic concepts, such as place value notation and the idea of zero, algorithmic methods for problem solving, and algebraic and trigonometric methods.
41	Graph-theoretic measures of connectivity and reachability are employed to quantify spatial interactions and evaluate network functionality.
42	Extended X-Ray Absorption Fine Structure spectroscopy is an effective and essential method for examining the local atomic structure of nanomaterials
43	Ice which includes not just water but also magnetic and quantum ice, has become an essential to research in current condensed matter physics.
44	Strength of PTMC for small peptides and underscore the need for improved temperature schemes, replica numbers, and sampling strategies to accurately characterize larger protein domains.
45	The variation of dielectric constant and the loss could be understood by Cole-Cole Plot, Arrhenius plot of relaxation time, Koops and Maxwell-Wagner theories to understand the dielectric behavior of the NiCr_2O_4
46	The real part of the dielectric constants reveal a totally dispersive behaviour especially at all frequency ranges indicating the dominance of Maxwell-Wagner-Sillars (MWS) polarization mechanism.
47	The Mg-substituted sample shows significantly enhanced roughness ($R_{\text{avg}} = 129 \text{ nm}$; $R_{\text{rms}} = 205 \text{ nm}$) compared to the Sr-substituted sample ($R_{\text{avg}} = 28 \text{ nm}$; $R_{\text{rms}} = 42 \text{ nm}$).
48	Superoxide and hydroxyl radicals were the main active species during the degradation process. The catalytic activity of ZnO/CuS nanocomposite is much better than that of the other composites.
49	Wheat seeds treated with O_2 and N_2 plasma showed improvements in growth and biomass.
50	The distribution of ridge densities in females is more compact and consistent, with the range of 12 to 18 compare to the broader range of 11 to 19 in males.
51	Zinc oxide (ZnO) nanoparticles have emerged as highly efficient photocatalysts owing to their wide band gap, strong oxidising ability, cost-effectiveness, and versatile tunability

	through controlled synthesis.
52	Beyond pharmaceutical applications, coumarins play significant roles in agrochemicals, perfumery, food additives, and material science, particularly in the development of dyes, optical brighteners, and laser-active materials.
53	Lipid-based nanocarriers, particularly SLNs prepared via hot homogenization, offer a promising solution by improving corneal penetration, sustaining drug release, and enhancing therapeutic outcomes.
54	Green and sustainable chemistry plays a crucial role in addressing global challenges such as climate change, resource depletion, and environmental pollution, while supporting sustainable development and a circular economy.
55	Aloe vera extract is a great biogenic mediator that provides a safe, affordable, and eco-friendly way to produce nanomaterials with potential applications in the fields of catalysis, biomedicine, and the environment.
56	Development of Targeted Live Biotherapeutic Products (LBPs), functionally optimized by TIKS formulations acting as metabolic co-factors. Our next step of action is finding more such combinations of LBPs and TIKS Co- factors.
57	Extremely high polysaccharide concentrations (up to 525.12 mg/g) and substantial phenolic content promote co-precipitation, oxidative binding, and spectral interference, leading to suppressed absorbance ratios.
58	The integration of IoT with data analytics is poised to redefine agriculture and meet the demands of a rapidly growing global population.
59	Extracts from <i>Asparagus officinalis</i> stems, especially ethanolic extract, have significant bioactive properties, emphasizing the importance of asparagus stems as an underutilized plant resource.
60	Identified Pbp5, acm, esp, msrC, purK, TetL, vanZ, gyrA, pstS, entA, and cls as proteins with strong text-mining evidence in <i>Enterococcus faecium</i> resistance using the network pharmacology approach.

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SHRI VAISHNAV VIDYAPEETH VISHWAVIDYALAYA, INDORE (INDIA)

Shri Vaishnav Vidyapeeth Vishwavidyalaya is a private university established under Madhya Pradesh Niji Vishwavidyalaya (Sthapana Avam Sanchalan) Adhiniyam in 2015 at Indore (India). The university has been established with a vision to be leader in shaping better future for mankind through quality education, training and research. It pursues the mission to make a difference in sustaining the growth of global societies by developing socially responsible citizens. Value based education being at the helm, the University promotes endurance, excellence, fairness, honesty and transparency as its core values. Some of the objectives of the university are as under:

- To provide teaching and training in higher education and make provision for research as well as advancement and dissemination of knowledge.
- To ensure world class quality in its offerings and create higher levels of intellectual abilities.
- To create centers of excellence for research and development for sharing knowledge and its applications.

The University is offering Undergraduate, Postgraduate, Dual Degree and Doctoral programs in various disciplines through the following constituent Institutions:

- Shri Vaishnav Institute of Agriculture
- Shri Vaishnav Institute of Architecture
- Shri Vaishnav Institute of Computer Applications
- Shri Vaishnav Institute of Commerce
- Shri Vaishnav Institute of Education
- Shri Vaishnav Institute of Fine Arts
- Shri Vaishnav Institute of Forensic Science
- Shri Vaishnav Institute of Home Science
- Shri Vaishnav Institute of Journalism and Mass Communication
- Shri Vaishnav Institute of Law
- Shri Vaishnav School of Management
- Shri Vaishnav Institute of Paramedical Sciences
- Shri Vaishnav Institute of Pharmacy
- Shri Vaishnav Institute of Planning
- Shri Vaishnav Institute of Science
- Shri Vaishnav Institute of Social Sciences, Humanities and Arts
- Shri Vaishnav Institute of Technology and Science
- Shri Vaishnav Institute of Information Technology
- Shri Vaishnav Institute of Textile Technology
- Faculty of Doctoral Studies & Research

Besides these Institutions, the University has established following Centers:

- Centre of Excellence in Data Science
- Centre of Excellence in Happiness Studies
- Centre of Excellence in Plasma Research
- Centre of Excellence in Simulation & Gaming
- Centre of Excellence in Sustainable Development
- Centre of Vocational Studies
- Centre for Women's Studies
- Centre of Excellence in AI and Machine Learning
- Centre of Excellence in Cloud Computing



Excellent Publishers
Kancheepuram, Tamilnadu, India
www.excellentpublishers.com

ISBN 978-93-94174-76-4

