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" EVALUATING DNA EXTRACTION METHODS IN FORENSIC INVESTIGATIONS: BEST PRACTICES, CHALLENGES AND LEGAL IMPLICATIONS "

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ABSTRACT:

DNA extraction forms the cornerstone of forensic science, serving as the initial and most critical step in analyzing genetic evidence. This study examines various DNA extraction methods, ranging from conventional techniques such as phenol-chloroform and salting out to modern approaches like silica column-based and magnetic bead-based extractions. The choice of method depends on factors including the biological source, DNA quality, and intended downstream applications. Different types of DNA—genomic, mitochondrial, nuclear, cell-free, and plasmid—are explored for their distinct forensic relevance and specific isolation protocols. The forensic significance of DNA lies in its ability to accurately identify individuals, exonerate the innocent, and assist in mass disaster victim identification. Yet, the expanding use of genetic data introduces complex legal and ethical challenges, particularly concerning privacy, consent, and protection against self-incrimination, as recognized under Indian and international legal frameworks. This study further reviews landmark case laws and statutory provisions to evaluate the admissibility, reliability, and evidentiary value of DNA in judicial proceedings. It also assesses the transformative impact of emerging technologies—including automation, artificial intelligence, microfluidics, and next-generation sequencing—on the precision and efficiency of forensic DNA analysis. The research concludes that while technological progress has significantly enhanced the speed and accuracy of DNA profiling, it must be balanced with robust legal safeguards and ethical principles. Harmonizing scientific innovation with judicial

integrity remains essential to uphold justice and ensure that DNA evidence continues to serve as a trustworthy pillar of forensic investigation.

KEYWORDS:

DNA Extraction, Forensic DNA Analysis, Genetic evidence, DNA Profiling, Criminal Justice System

1.1 INTRODUCTION:

Deoxyribonucleic Acid (DNA) is the fundamental genetic material that governs biological function and heredity in all living organisms. DNA extraction is a fundamental process in molecular biology, serving as the initial step in countless scientific investigations. Friedrich Miescher[1] was the first scientist to isolate DNA while studying the chemical composition of cells. In 1869, he isolated leukocytes from material obtained on fresh surgical dressings and began examining the proteins within these cells. While carrying out this work, he discovered a previously unknown substance in the cell nucleus, which he named "nuclein." This substance, later identified as DNA, displayed unique properties distinct from proteins. his scientific finding, along with the isolation protocols used, was published in 1871 in collaboration with his mentor, Felix Hoppe-Seyler. However, it was only in 1958 that Meselson and Stahl developed a routine laboratory procedure for DNA extraction. They performed DNA extraction from bacterial samples of *Escherichia coli*[2] using a salt density gradient centrifugation protocol. Since then, the extraction and study of DNA have become crucial in various scientific fields[3]. The efficiency, purity, and yield of extracted DNA significantly influence the reliability of downstream applications such as polymerase chain reaction (PCR), DNA sequencing, genotyping, and forensic profiling for DNA fingerprinting, enabling the identification of suspects and establishing paternity.

DNA extraction involves the disruption of cellular structures to release nucleic acids, followed by the removal of proteins, lipids, and other contaminants. Numerous techniques have been

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[1] R. Dahm, 'Friedrich Miescher and the Discovery of DNA' (2005) 278(2) *Developmental Biology* 274
[2] K. Elkins, *Forensic DNA Biology* (Oxford Academic Press, Kidlington, England 2013) 43–45.
[3] M. Dairawan and P.J. Shetty, 'The Evolution of DNA Extraction Methods' (2020) 8(1) *American Journal of Biomedical Science & Research* 39.

developed to optimize this process, ranging from conventional organic solvent-based methods (e.g., phenol-chloroform extraction) to advanced commercial kits using silica columns or magnetic beads. The selection of a suitable DNA extraction method is often guided by the type of biological sample, the required DNA quality, the presence of inhibitors, and the intended analytical application.

In forensic science, DNA extraction holds paramount importance, as it facilitates the identification of individuals, establishment of biological relationships, and resolution of criminal cases[4]. Newer techniques that are more reliable and efficient have facilitated the advancement in knowledge about the human genome and played a major role in the advent of various fields in science such as gene-editing and personalized medicine. However, forensic samples often present challenges such as low quantity, degradation, and contamination, necessitating the use of highly sensitive and reliable extraction methods. Furthermore, best practices in sample handling, contamination prevention, and quality control are essential to ensure the integrity of the extracted DNA, especially in legal and investigative contexts.[5]

This study aims to provide a comprehensive overview of various DNA extraction techniques, evaluate their suitability for different biological samples, and outline best practices for high-quality DNA recovery. It further seeks to critically examine the challenges, limitations, and forensic significance of these methods, along with their legal implications in forensic investigations. By understanding and optimizing DNA extraction protocols, researchers and forensic practitioners can enhance the accuracy, efficiency, and credibility of genetic analysis.

1.2 STATEMENT OF PROBLEM:

DNA extraction is a crucial step in forensic investigations, yet the process is often hindered by degraded, contaminated, or low-quality samples. The existence of multiple extraction methods creates uncertainty regarding their efficiency and suitability for different forensic scenarios. Additionally, improper handling may affect the reliability and admissibility of DNA evidence in courts. The increasing use of DNA evidence also raises important legal and ethical concerns, particularly relating to privacy, consent, and potential misuse of genetic information. Hence,

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[4] W. Goodwin, A. Linacre and S. Hadi, *An Introduction to Forensic Genetics* (John Wiley & Sons, Chichester 2011).
[5] W.R. Kuperus, K.H. Hummel and J.M. Roney, 'Crime Scene Links through DNA Evidence: The Practical Experience from Saskatchewan Casework' (2003) 36(1) *Canadian Society of Forensic Science Journal* 19.

there is a need to evaluate DNA extraction methods, identify best practices, and examine their forensic and legal implications.

1.3 RESEARCH OBJECTIVES:

1. To analyze various DNA extraction methods used in forensic investigations, including conventional and modern techniques.
2. To evaluate the efficiency and reliability of DNA extraction methods, particularly in handling degraded and low-quality forensic samples.
3. To identify best practices for obtaining high-quality DNA while minimizing contamination and ensuring accuracy in forensic analysis.
4. To examine the challenges and limitations associated with DNA extraction in forensic contexts.
5. To assess the legal and ethical implications of DNA extraction and its admissibility as evidence in judicial proceedings.

1.4 RESEARCH METHODOLOGY:

The present study adopts a **doctrinal and analytical research methodology**, primarily qualitative in nature, focusing on the evaluation of DNA extraction methods in forensic investigations along with their legal and ethical implications. A descriptive approach is used to explain various conventional and modern DNA extraction techniques, while an analytical approach is employed to assess their efficiency, reliability, and associated challenges, particularly in handling degraded forensic samples. The research is based entirely on **secondary sources of data**, including standard textbooks on forensic science and molecular biology, peer-reviewed journal articles, case laws, statutory provisions, and reports from governmental and international organizations. Data has been collected through library research and online academic databases.

The scope of the study is limited to the scientific evaluation of DNA extraction techniques and their relevance in forensic investigations, with a specific focus on legal admissibility and ethical concerns within the Indian context, supplemented by relevant international perspectives. However, the study is subject to certain limitations, including reliance on secondary data without experimental validation, and the possibility that rapidly evolving technological advancements in DNA analysis may not be fully captured within the scope of this research.

CHAPTER II

BASIC PRINCIPLES OF DNA EXTRACTION

DNA extraction is the process of isolating deoxyribonucleic acid (DNA) from cells or tissues using physical, chemical, or enzymatic methods. It is a fundamental step in molecular biology and genetic analysis, as it enables the study and use of an organism's genetic material[6].. The quality and quantity of extracted DNA play a crucial role in ensuring the accuracy of downstream applications such as polymerase chain reaction (PCR), sequencing, genotyping, and forensic profiling. DNA can be obtained from a wide range of biological samples depending on the purpose of analysis[7]. Common sources include:

1. Blood – Whole blood or dried bloodstains
2. Saliva and Buccal Swabs – Non-invasive sources for human DNA
3. Semen and Vaginal Swabs – Frequently used in sexual assault investigations
4. Tissues – Derived from biopsies or forensic remains
5. Hair, Bone, and Teeth – Useful in cases involving degraded or skeletal remains
6. Urine, Feces, and Nails – Less common but valuable in specific situations
7. Touch DNA – Skin cells transferred onto objects through contact

The human body consists of trillions of cells, most of which contain a nucleus, except red blood cells. Each nucleated cell carries two copies of an individual's genome, which can be used to generate a DNA profile. However, different sample types present unique challenges in terms of DNA quantity, quality, and the presence of inhibitors, all of which must be considered during the extraction process.

DNA used for forensic genetic analysis should possess the following ideal characteristics[8]:

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[6] J.M. Butler, 'Fundamentals of Forensic DNA Typing' (2010) 55(4) *Journal of Forensic Sciences* 1019.
[7] A. Dhaliwal, 'DNA Extraction and Purification' (2013) *Materials and Methods* 3
[8] S. Tan and B. Yiap, 'DNA, RNA, and Protein Extraction: The Past and The Present' (2009) *Journal of Biomedicine and Biotechnology* 1

1. High level of polymorphism
2. Easy and cost-effective to analyze
3. Simple to interpret and compare across laboratories
4. Low mutation rate

2.1 BASIC PRINCIPLES OF DNA EXTRACTION:

1. **Cell Lysis**- This step involves breaking open the cell membrane and nuclear envelope to release DNA. It typically uses detergents or enzymes (like Proteinase K) in a buffer solution to disrupt lipid layers.
2. **Removal of Proteins and Inhibitors**- Once the DNA is released, contaminants such as proteins, lipids, and polysaccharides are removed. This can be achieved using organic solvents (e.g., phenol-chloroform), proteases, or silica/magnetic bead-based purification systems.
3. **DNA Precipitation and Purification**- DNA is precipitated out of the solution using alcohol (usually ethanol or isopropanol) and salt. The precipitated DNA is then washed to remove residual impurities and rehydrated in a suitable buffer for storage or analysis.

Together, these principles ensure the extraction of high-quality DNA, which is essential for the success of subsequent analytical procedures. DNA extraction serves as the cornerstone of numerous scientific and forensic procedures[9]. The quality and integrity of the extracted DNA directly affect the accuracy and reliability of downstream applications such as: Polymerase Chain Reaction (PCR), DNA sequencing and genotyping, Cloning and genetic engineering,

2.2 TYPES OF DNA FOR EXTRACTION PURPOSES:

The type of DNA targeted for extraction depends on the nature of the sample, the intended application, and the cellular compartment from which the DNA originates. Each type of DNA has unique structural and functional characteristics that influence the choice of extraction method and downstream analytical approaches[10].

[9] Stuart H. James, Jon J. Nordby and Suzanne Bell, *Forensic Science: An Introduction to Scientific and Investigative Techniques* (3rd edn., CRC Press 2009).
[10] Geoffrey M. Cooper and Robert E. Hausman, *The Cell: A Molecular Approach* (7th edn., Sinauer Associates 2019).

1. Genomic DNA (gDNA):

Genomic DNA refers to the complete set of DNA found within the nucleus of a cell, encompassing both coding (exons) and non-coding regions (introns and regulatory sequences). It contains the full genetic blueprint of an organism.

- **Applications**: Whole-genome sequencing, gene expression analysis, genetic testing, and forensic DNA profiling.
- **Extraction Considerations**: Requires methods that preserve long DNA fragments and minimize shearing; commonly extracted from blood, saliva, tissue, and cultured cells.

2. Mitochondrial DNA (mtDNA):

Mitochondrial DNA is a small circular molecule found in the mitochondria, separate from nuclear DNA. It is maternally inherited and exists in multiple copies per cell.

- **Applications**: Forensic identification (especially in degraded samples), evolutionary studies, and maternal lineage analysis.
- **Extraction Considerations**: mtDNA is more abundant in degraded or old samples (e.g., bones, hair shafts), making it useful in forensic anthropology and disaster victim identification.

3. Nuclear DNA:

Nuclear DNA refers specifically to DNA housed within the cell nucleus. It includes the chromosomal DNA that encodes the majority of an organism's genetic information.

- **Applications**: STR profiling in forensics, genetic disease testing, and human identification.
- **Extraction Considerations**: High-molecular-weight nuclear DNA is essential for detailed genetic analyses and must be extracted with care to prevent degradation.

4. Plasmid DNA (in microbes):

Plasmid DNA is a small, circular form of DNA that exists outside the main chromosomal DNA, primarily in bacteria and certain eukaryotic microorganisms. It commonly contains genes that provide advantages such as antibiotic resistance or enhanced survival capabilities.

- **Applications**: Genetic engineering, cloning, recombinant protein production, and vaccine development.
- **Extraction Considerations**: Plasmid DNA must be separated from chromosomal DNA using alkaline lysis and specific purification techniques (e.g., miniprep, midiprep, or maxiprep kits).

Understanding the types of DNA is essential for selecting appropriate extraction methods and designing reliable molecular experiments. While genomic and nuclear DNA dominate most applications, mitochondrial and plasmid DNA play crucial roles in forensic analysis, medical diagnostics, and biotechnology[11]. The nature, size, and abundance of each DNA type influence the complexity and sensitivity required in the extraction protocol.

CHAPTER III

DNA EXTRACTION TECHNIQUES

DNA extraction techniques are designed to isolate deoxyribonucleic acid (DNA) from biological samples for analysis. The method used depends on factors such as sample type, required DNA quality, cost, and downstream applications. These techniques are broadly classified into **conventional (manual)** and **modern (commercial or automated)** methods[12].

Steps in DNA Extraction[13]:

- **Cell Lysis:** Disruption of cells to release DNA.
- **Separation of DNA from Debris:** Removing of membrane lipids, cell debris and other unwanted material.

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[11] Robin Williams and Paul Johnson, *Genetic Policing: The Use of DNA in Criminal Investigations* (Willan Publishing 2008).
[12] N. Gupta, 'DNA Extraction and Polymerase Chain Reaction' (2019) 36(2) *Journal of Cytology* 116.
[13] David E. Krane and Mei-Huey J. Chu, *DNA Techniques in Forensic Science* (CRC Press 2006)

- **Binding and Purification:** Using specific methods to bind and purify DNA.
- **DNA Precipitation:** Releasing the purified DNA from the binding matrix.

3.1 CONVENTIONAL DNA EXTRACTION METHODS:

These methods involve the manual use of chemical reagents and centrifugation steps. Though labor-intensive, they are widely used for their effectiveness and cost-efficiency.

3.1.1 Phenol-Chloroform Extraction:

- **Principle**: Uses phenol and chloroform to denature proteins and separate them from DNA.
- **Steps**: Cell lysis → Protein denaturation → Centrifugation → Aqueous phase collection → DNA precipitation with alcohol.

This method yields **high molecular weight DNA** that is suitable for a variety of downstream applications, including PCR, cloning, and sequencing. However, phenol-chloroform extraction requires caution due to the use of **toxic and volatile organic solvents**, and it is not ideal for automation or high-throughput processing. Despite these limitations, it remains a **gold standard for DNA purification** when purity and integrity are critical, especially in molecular biology and forensic research.

3.1.2 Salting-Out Method:

The **salting-out method** is a widely used, non-toxic, and cost-effective technique for extracting DNA from biological samples such as blood, tissues, or cultured cells[14].

- **Principle**: Uses high concentrations of salt (e.g., sodium acetate) to precipitate proteins, leaving DNA in solution.
- **Process**: Cells are lysed; salt (e.g., sodium acetate) is added; proteins precipitate and are removed by centrifugation.

The salting-out method is favored in many laboratories because it avoids the use of toxic organic solvents like phenol or chloroform, making it safer and more environmentally friendly.

[14] M.R. Green and J. Sambrook, 'Isolation of High-Molecular-Weight DNA Using Organic Solvents' (2017) *Cold Spring Harbor Protocols* prot093450.

It also produces reasonably pure and high-molecular-weight DNA that is suitable for common molecular biology applications, including PCR, restriction digestion, and genotyping. Still, its simplicity, low cost, and efficiency make salting-out a popular method in teaching, diagnostic, and research labs[15].

3.2 MODERN AND COMMERCIAL DNA EXTRACTION METHODS:

These techniques use ready-to-use kits and are optimized for consistency, speed, and safety. They are widely adopted in clinical and forensic laboratories.

3.2.1 Silica Column-Based Extraction:

The **silica column-based DNA extraction method** is a modern and widely adopted technique that utilizes the chemical affinity of DNA to silica surfaces under high-salt conditions. It is the foundation of many commercial DNA extraction kits due to its **efficiency, speed, and high purity** of extracted DNA.

- **Principle**: DNA binds to silica under chaotropic salt conditions while contaminants are washed away.
- **Process**: Lysis → Binding to silica column → Washing → Elution.

Silica column-based extraction offers several advantages: it is **fast, easy to perform, requires minimal manual handling,** and is scalable for different sample sizes. Nevertheless, this method has become the **gold standard in clinical, forensic, and molecular biology laboratories,** where reproducibility and DNA quality are paramount.

3.2.2 Magnetic Bead-Based Extraction:

Magnetic bead-based DNA extraction is an advanced and highly efficient method that uses **magnetically responsive particles** to isolate DNA from biological samples[16]. This technique is particularly favoured in **automated and high-throughput laboratory settings** because of

S. Miller, D. Dykes and H. Polesky, 'A Simple Salting Out Procedure for Extracting DNA from Human Nucleated Cells' (1988) 16(3) *Nucleic Acids Research* 1215.

[16] Hawkins T (1998) DNA purification and isolation using magnetic particles. United States.

its scalability, precision, and ability to deliver high-quality DNA suitable for sensitive downstream applications.

- **Principle**: DNA binds to silica-coated magnetic beads; magnets are used to separate DNA from impurities.
- This method is suitable for automation and high-throughput applications, allowing for quick and efficient DNA isolation.

3.2.3 Boiling Method:

- **Principle**: Boiling ruptures cells to release DNA.
- **Advantages**: Rapid and requires no reagents.
- **Disadvantages**: Yields low-quality, degraded DNA; not suitable for sensitive assays.

3.2.4 Paper-Based DNA Extraction (FTA Cards):

In this method, DNA is bound to a paper matrix for transport and storage, later eluted for further processing. This approach is used for fieldwork and sample collection in remote areas.

- **Principle**: Cells are lysed on filter paper; DNA is absorbed and can later be eluted.
- **Applications**: Field sample collection, long-term storage.
- **Advantages**: Easy transport and storage; non-toxic.
- **Disadvantages**: Lower yield; limited to small-scale applications.

Choosing the appropriate DNA extraction technique is crucial for ensuring high-quality DNA for accurate downstream analysis. Traditional methods are suitable for cost-effective lab work, whereas modern kits and automated systems offer enhanced speed and reproducibility. Emerging techniques provide innovative solutions for field and clinical diagnostics. Each method presents trade-offs between cost, complexity, and output, making method selection a critical aspect of molecular biology and forensic science.

3.3 DNA EXTRACTION TECHNIQUE FROM CHALLENGING SAMPLES:

Extracting DNA from challenging samples such as degraded tissues, forensic traces (like bones, teeth, and touch DNA), formalin-fixed paraffin-embedded (FFPE) tissues, ancient remains, and low-cellularity fluids requires specialized techniques to overcome issues like low yield, fragmentation, and the presence of inhibitors[17]. Specialized extraction methods and modifications are required to maximize yield, integrity, and purity from such problematic sources.

1. In forensic science, methods like the Chelex 100 resin technique are employed for trace samples, as it binds metal ions and prevents DNA degradation, though it yields single-stranded DNA. For low-concentration or degraded samples, silica column or magnetic bead-based kits with carrier RNA enhance recovery and purity[18].
2. Hard tissues such as bones and teeth undergo decalcification with EDTA followed by proteinase K digestion and either organic extraction or kit-based purification. FFPE tissues, which suffer from formalin-induced crosslinking, require deparaffinization with xylene, heat-induced reversal of crosslinks, and enzymatic digestion before extraction using specialized kits[19].
3. Ancient DNA (aDNA) samples demand strict contamination control and the use of silica or magnetic bead-based methods, often with extended incubation times and damage-repair enzymes. Similarly, cell-free DNA (cfDNA) from plasma or serum, which is highly fragmented and present in minute quantities, requires high-sensitivity extraction kits with prior centrifugation and the addition of carrier RNA[20]. These adapted and often sample-specific protocols are essential to obtaining high-quality DNA for use in forensic investigations, clinical diagnostics, archaeological research, and molecular biology.

3.4 BEST PRACTICES FOR HIGH QUALITY DNA EXTRACTION:

- Ensuring high-quality DNA extraction is critical for the accuracy and reliability of downstream applications such as PCR, sequencing, and forensic analysis. The first step toward achieving

[17] Walker F M, Hsieh K. Advances in directly amplifying nucleic acids from complex samples. Biosensors (Basel) 2019;9(04):117. doi: 10.3390/bios9040117

[18] P.S. Walsh, D.A. Metzger and R. Higuchi, 'Chelex 100 as a Medium for Simple Extraction of DNA for PCR-Based Typing' (1991) 10(4) *BioTechniques* 506.

[19] K. Inoue and M. Sasaki, 'DNA Extraction from Challenging Forensic Samples' (2015) 60(2) *Journal of Forensic Sciences* 290

[20] B. Budowle, A.J. Eisenberg and A. van Daal, 'Validating Forensic DNA Testing Methods' (2009) 6(9) *Electrophoresis* 1627

optimal results is the **use of fresh or properly preserved biological samples**, as degradation significantly affects DNA yield and integrity.

- It is essential to **select an appropriate extraction method** based on the sample type—whether it's soft tissue, blood, bones, or forensic trace evidence. Employing **clean, nuclease-free reagents and equipment** prevents contamination and degradation of nucleic acids.
- During cell lysis, proper use of detergents, enzymes (like proteinase K), and optimized incubation times enhances DNA release and prevents shearing. **Avoiding vortexing or excessive pipetting** of lysates helps preserve high molecular weight DNA.
- In methods involving binding matrices (like silica columns or magnetic beads), **strict adherence to protocol timings, washing steps, and elution conditions** ensures purity and concentration[21]. Additionally, using carrier RNA can improve DNA recovery, especially from low-yield samples.
- Post-extraction, DNA should be quantified (e.g., using spectrophotometry or fluorometry) and assessed for purity, and **stored properly at -20°C or -80°C** to prevent degradation. Maintaining a **contamination-free environment**, especially when working with forensic or ancient DNA, is also crucial.
- By integrating these best practices, researchers and forensic analysts can consistently obtain high-quality DNA suitable for sensitive and precise genetic analysis.

CHAPTER IV

FORENSIC AND LEGAL SIGNIFICANCE OF DNA EXTRACTION

DNA evidence has emerged as one of the most powerful tools in modern forensic science, significantly transforming the investigation and adjudication of criminal cases. Its ability to uniquely identify individuals with a high degree of accuracy has made it particularly valuable in serious offences such as sexual assault, homicide, and mass disaster identification. By analyzing biological materials like blood, semen, saliva, or hair, forensic experts can establish a direct link between the accused, the victim, and the crime scene[22].

[21] K. Doyle, *The Source of Discovery: Protocols and Applications Guide* (Promega, Madison 1996).
[22] R. Dabney, M. Meyer and S. Pääbo, 'Ancient DNA Damage' (2013) 14(3) *Cold Spring Harbor Perspectives in Biology* a012567.

At the same time, the growing reliance on DNA profiling raises important legal and ethical considerations. Courts must ensure that such evidence is collected, analyzed, and presented in accordance with established procedures to maintain its reliability and admissibility. Additionally, concerns relating to privacy, consent, and protection against self-incrimination require careful judicial scrutiny[23]. Thus, while DNA evidence strengthens the pursuit of justice through scientific precision, its use must be balanced with constitutional safeguards and legal principles.

4.1 FORENSIC SIGNIFICANCE:

DNA Extraction can helps in forensic investigations by,

1. **Individual Identification-** DNA is unique to every individual (except identical twins), making it the most reliable biological marker for identifying suspects, victims, or unknown remains.
2. **Linking Suspects to Crime Scenes-** DNA extracted from bloodstains, hair, saliva, semen, skin cells (touch DNA), or other biological traces can connect a suspect directly to the scene of a crime[24].
3. **Exoneration of the Innocent-** DNA evidence has been used to overturn wrongful convictions, ensuring justice for innocent individuals through post-conviction testing.
4. **Analysis of Minute or Degraded Samples-** Advanced DNA extraction techniques allow forensic experts to work with degraded, old, or trace-level samples, such as from skeletal remains or decades-old case files.
5. **Kinship and Paternity Testing-** DNA is used to establish biological relationships, which is critical in missing persons cases, disaster victim identification, or inheritance disputes.
6. **Cold Case Resolution-** Improved DNA extraction methods enable investigators to re-analyze evidence from unsolved cases, sometimes decades later, using better technology.
7. **Mass Disaster Identification-** DNA extraction plays a crucial role in identifying victims in mass casualty incidents (natural disasters, terrorist attacks) where bodies may be fragmented or decomposed.

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[23] S. Tan and B.C. Yiap, 'DNA, RNA, and Protein Extraction: The Past and The Present' (2009) *Journal of Biomedicine and Biotechnology* 1.
[24] National Human Rights Commission (India), *Advisory on Use of DNA Technology in Forensic Investigation* (2021).

8. **Sexual Assault and Rape Cases**- DNA extracted from semen, vaginal swabs, or clothing is essential for identifying perpetrators and corroborating victim testimony.

These are the ways in which DNA Extraction will play a crucial role in crime investigation with personal identification by extracting and analyzing the biological evidence left behind the crime scene.

4.2 LEGAL SIGNIFICANCE:

4.2.1 Indian Laws and Provisions:

1. The Code of Criminal Procedure (CrPC), 1973:

- **Section 53 & 53A**: Allows **medical examination** of accused (including collection of DNA) in certain cases like rape and bodily offenses.
- **Section 164A**: Pertains to **medical examination of rape victims**, including collection of forensic samples.

2. Indian Evidence Act, 1872:

- **Section 45**: Recognizes opinions of forensic and scientific experts, including DNA analysts, as relevant evidence.
- **Section 65B**: Governs electronic records; chain of custody and proper authentication are crucial for admissibility of DNA reports.

3. DNA Technology (Use and Application) Regulation Bill, 2019[25]**:**

- **This Act** Regulates DNA data usage for forensic and civil purposes.
- **Proposes**:
    - National and regional DNA Data Banks.
    - DNA Regulatory Board for oversight.
    - Safeguards on privacy, consent, and removal of DNA profiles.

[25] *DNA Technology (Use and Application) Regulation Bill*, 2019 (India)

- **But it c**oncerns over misuse, data breach, and violation of privacy due to weak safeguards.

4. Bharatiya Sakshya Adhiniyam, 2023: **Section 39** of the Act gives admissibility in opinions of experts, including fingerprint experts, can be used in courts.
5. **Identification of Prisoners Framework:** The Criminal Procedure (Identification) Act, 2022 authorizes the collection of **biometric and biological samples**, including DNA, from convicts and certain categories of accused persons expands the scope of identification beyond traditional fingerprints to include **modern forensic techniques**.

4.2.2 Case laws and Judgments:

1. **Selvi v. State of Karnataka (2010):**[26]

Issue: Whether compulsory scientific techniques like nacro analysis, polygraph, and brain mapping violate Article 20(3) of the Constitution.

Judgment:

- The Court held that **testimonial evidence** (like polygraph tests) cannot be compelled.
- However, **DNA, blood samples, and fingerprints are considered physical evidence,** and **compulsory collection does not violate Article 20(3).**
- **This case** established the constitutionality of **compelled DNA collection** for forensic purposes.

2. **Justice K.S. Puttaswamy v. Union of India (2017) :**[27]

Issue: Recognition of the **right to privacy** as a fundamental right under the Constitution.

Judgment:

- Any **DNA extraction and storage** must respect the individual's **bodily integrity and privacy** under Article 21.
- **This case also established that** DNA profiling must be governed by **procedural safeguards** and proportionality principles. This case was a landmark judgment gave

[26] *Selvi v. State of Karnataka, (2010) 7 SCC 263.*
[27] **Justice K.S. Puttaswamy v. Union of India (2017) 10 SCC1**

recognition to forensic investigation for the collection of samples and equal protection to right to privacy of individual rights.[28]

3. Krishan Kumar Malik v. State of Haryana (2011):[29]

Issue: Whether DNA profiling can be relied upon in a rape case to establish the involvement of the accused.

Judgment:

- The Supreme Court upheld the use of DNA evidence as **scientifically reliable** and useful in proving guilt.
- **This case** reinforced the admissibility and importance of DNA extraction in sexual assault cases.

4. Bhabani Prasad Jena v. Orissa State Commission for Women (2010):[30]

Issue: Whether courts can compel a person to undergo a DNA test to determine **paternity.**

Judgment:

- DNA testing cannot be ordered **routinely**.
- The court must balance **individual privacy** and **the need for justice**.
- **Significance**: Highlighted ethical limitations and privacy concerns in non-forensic DNA extraction.

4.2.3 International Instruments and Guidelines:

[28] M. Khosla, "DNA Profiling and the Right to Privacy," *Indian Journal of Constitutional Law*, Vol. 7 (2013), pp. 125–138.
[29] **Justice K.S. Puttaswamy v. Union of India (2017) 10 SCC1**
[30] **Bhabani Prasad Jena v. Orissa State Commission for Women (2010) 8 SCC633**

1. Universal Declaration on the Human Genome and Human Rights, 1997 (UNESCO)

- Emphasizes protection of human dignity in genetic testing and bans discrimination based on genetic characteristics[31].

2. European Court of Human Rights – S. and Marper v. United Kingdom (2008)

- Holding: Retention of DNA profiles of innocent individuals violates **Article 8 (Right to Privacy)** of the European Convention on Human Rights.[32]
- Relevance: Set a global precedent for **limiting DNA database misuse** and respecting individual rights.
- **Maryland v. King (2013, US)**: Upheld the routine collection of DNA at arrest — used as valid forensic evidence.
- **R. v. Adams (1996, UK)**: DNA evidence admissible but required strong statistical support due to mixed samples.

Courts are increasingly accepting DNA extraction as **conclusive or corroborative evidence**, especially in rape, murder, and paternity disputes — provided it is backed by **scientific integrity and legal compliance**.

CHAPTER V

LEGAL AND ETHICAL ISSUES

DNA extraction, especially in the forensic and biomedical context, raises several important legal and ethical issues. These concerns revolve around privacy, consent, misuse of genetic information, and proper handling of evidence. Below are the key considerations:

5.1 LEGAL ISSUES:

1. **Lack of Comprehensive Legislation:**

[31] United Nations Educational, Scientific and Cultural Organization (UNESCO), *Universal Declaration on the Human Genome and Human Rights*, 1997, Article 5.
[32] S and Marper v. United Kingdom, (2008) ECHR 1581, (2008) ECHR 1581 — held that indefinite retention of DNA samples violates Article 8 of the European Convention on Human Rights (right to privacy).

- India currently lacks a standalone, fully enacted law governing DNA use.
- The **DNA Technology (Use and Application) Regulation Bill, 2019,** is pending, though it aims to regulate DNA data banks and safeguard individuals' rights.

2. **Compelled DNA Collection:**
   - Under **Section 53 and 53A of the CrPC**, law enforcement can collect biological samples from accused persons without consent in certain criminal cases (e.g., rape).
   - Raises questions of bodily autonomy and the **right against self-incrimination** under **Article 20(3) of the Constitution.**
3. **Chain of Custody & Evidentiary Integrity:**
   - Legally admissible DNA evidence requires an unbroken chain of custody.
   - Poor documentation, mishandling, or contamination may render the evidence inadmissible under the **Indian Evidence Act**.
4. **Inclusion in DNA Databases:**
   - Without judicial oversight, indefinite retention of DNA profiles (including of innocents or acquitted persons) can violate the **right to privacy** (recognized in *Puttaswamy v. Union of India*, 2017).
5. **International Data Sharing:**
   - Cross-border exchange of forensic DNA data (e.g., via INTERPOL) must comply with data protection laws and international treaties to prevent misuse or unauthorized access.

5.2ETHICAL ISSUES:

1. **Consent and Autonomy:**
   - In forensic contexts, **non-consensual collection** (e.g., from suspects) is ethically debated, especially when dealing with vulnerable populations[33].

2. **Right to Privacy:**

[33] **International Society for Forensic Genetics**, "Recommendations on Ethical Issues in Forensic Genetics" (2010) 4(4) *Forensic Science International: Genetics* 273–280.

- DNA carries sensitive information, including genetic predispositions and ancestry.
- Unauthorized extraction or analysis may amount to an invasion of personal and family privacy.

3. **Risk of Genetic Discrimination:**
   - Misuse of extracted DNA by employers, insurers, or state agencies could lead to discrimination based on health or hereditary factors.
   - Highlights the need for ethical safeguards and regulatory oversight.[34]

4. **Data Storage and Retention:**
   - Ethical concerns arise over how long DNA samples and profiles should be stored.
   - Long-term retention of innocent individuals' DNA raises issues of surveillance and profiling.

CHAPTER VI
CHALLENGES AND ADVANCES IN DNA EXTRACTION

6.1 CHALLENGES IN DNA EXTRACTION FROM BIOLOGICAL SAMPLES:

DNA extraction from biological samples poses numerous challenges, particularly in forensic contexts where the integrity and reliability of evidence are paramount. However, the process begins with DNA extraction—a stage that is critical and often vulnerable to multiple challenges. Given the wide range of sample types encountered in forensic scenarios (e.g., blood, saliva, hair, bones), each with varying DNA content and quality, the extraction process must overcome significant biological and environmental obstacles.

- One of the most significant issues is **DNA degradation**, which occurs due to environmental exposure such as heat, moisture, UV light, or microbial activity, especially when samples are recovered from crime scenes after prolonged periods. *Selvi v. State of Karnataka*,

[34] **Graeme Laurie**, *Genetic Privacy: A Challenge to Medico-Legal Norms* (Cambridge University Press, 2002) 197.

(2010) 7 SCC 263 — emphasizes constitutional protection of bodily integrity, indirectly raising concerns about handling degraded and invasive samples.

- **Low quantity of DNA** is another major hurdle; forensic samples often contain minimal biological material—like touch DNA or shed skin cells—making extraction and amplification difficult. The challenge here is to extract a sufficient quantity of usable DNA without contamination or loss. Highly sensitive amplification techniques like PCR can be used, but these often amplify noise along with signal.
- Additionally, the presence of **PCR inhibitors** such as heme (in blood), melanin (in hair), or humic substances (in soil) can interfere with enzymatic reactions, resulting in poor-quality DNA profiles.
- **Mixed DNA samples** from multiple individuals, frequently encountered in sexual assault or violent crimes, complicate the extraction process further and require advanced statistical interpretation. Separating individual DNA profiles in such mixtures is highly complex and prone to errors. Statistical deconvolution methods are required, and even then, conclusive results are not always possible.
- The risk of **contamination**—either at the crime scene, during transport, or in the laboratory—also threatens the credibility of DNA evidence. The presence of extraneous DNA can compromise the entire forensic analysis. Best practices such as the use of gloves, sterile equipment, and clean rooms are crucial to mitigate this risk.
- The **type of biological material** significantly influences extraction success; while blood and saliva are rich in DNA, bone, hair shafts, and decomposed tissues require specialized protocols.
- Furthermore, the **time elapsed since deposition** of the sample affects DNA quality, with older samples more likely to be degraded. The longer a biological sample is left in uncontrolled environmental conditions, the higher the likelihood of DNA degradation. This is a particular issue in cold cases and disaster victim identification where evidence may be recovered years after the incident.
- **Infrastructural limitations** in forensic labs—such as lack of advanced equipment, trained personnel, and high-end technologies like Next-Generation Sequencing (NGS)—restrict the effective extraction and analysis of DNA, especially in resource-constrained settings.

Together, these challenges highlight the complexity of extracting DNA in a forensic framework and the need for continuous technological and procedural improvements[35].

6.2 TECHNOLOGICAL ADVANCES AND FUTURE DEVELOPMENTS:

Advances in DNA extraction techniques have opened the door to a wide range of applications across various fields, offering more efficient, reliable, and versatile methods for obtaining high-quality DNA[36]. In forensic investigations, DNA extraction from evidence such as hair, blood, or tissue samples is critical for identifying suspects and solving crimes. Advances in extraction techniques enable successful DNA retrieval from degraded or limited samples, providing reliable results that can stand up in court[37]. This has revolutionized the field of forensic science, leading to more accurate convictions and exonerations.

1. Automation and robotics have been integrated into modern forensic laboratories, enabling machines like Qiagen QIAcube and Thermo Fisher KingFisher to perform DNA extraction with minimal human intervention, reducing errors and ensuring consistency.
2. Microfluidic or "lab-on-a-chip" technologies now allow DNA extraction and analysis to be performed on small, portable platforms using minute sample volumes. These are increasingly used in fieldwork and disaster zones for rapid forensic analysis[38].
3. Magnetic nanoparticle-based techniques are being developed for selective and efficient binding of DNA, improving yields especially from degraded or trace biological materials.
4. Portable DNA extraction and sequencing tools, like the Oxford Nanopore MinION, are enabling real-time, on-site genetic analysis, which is transformative for rapid suspect identification and disaster victim identification in remote or time-sensitive situations.

CHAPTER VII

CONCLUSION

DNA extraction is a fundamental process in forensic science and biological research, enabling the isolation of genetic material from diverse biological sources. The choice of extraction method—

[35] S. Krimsky and T. Simoncelli, *Genetic Justice: DNA Data Banks, Criminal Investigations, and Civil Liberties* (Columbia University Press 2011) 112–134

[36] S.J. Kinaston *et al.*, 'Emerging DNA Technologies in Forensic Science' (2021) 50 *Forensic Science International: Genetics* 102383.

[37] H.C. Lee and R.E. Gaensslen, *Advances in Forensic Science* (CRC Press 2001).

[38] J.M. Butler, *Advanced Topics in Forensic DNA Typing: Methodology* (Elsevier Academic Press 2011) 525–540.

ranging from traditional techniques like phenol-chloroform and salting out to modern approaches such as silica column and magnetic bead-based methods—significantly affects the quality and reliability of DNA obtained, particularly in degraded or complex samples.

In forensic investigations, DNA evidence plays a vital role in identifying suspects, exonerating the innocent, and resolving criminal cases. Its admissibility depends not only on scientific accuracy but also on proper procedures, chain of custody, and compliance with legal and constitutional safeguards, including privacy and protection against self-incrimination. While technological advancements like automation and AI have enhanced efficiency and precision, they also raise ethical concerns regarding data protection and consent.

In conclusion, DNA extraction is more than just a laboratory process—it is a crucial intersection of science, law, and ethics. Its continued evolution must be guided not only by innovation but also by a strong commitment to legal standards, human rights, and scientific integrity.

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