

Association between forensic DNA and odontology in human identification during mass disaster: a systematic review

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ABSTRACT

BACKGROUND: According to the literature, forensic odontology is one of the most effective and affordable scientific techniques for identifying victims of mass disasters. This research methodically reviews the role and function of forensic odontologists in several global mass disasters. Forensic DNA in odontology is associated with the importance of its application in the identification of humans of mass disasters. As the crime rate continues to rise, the field of forensic medicine has evolved significantly. Forensic dentists play a pivotal role in various areas of crime scene investigations, thereby helping solve innumerable mysteries.

AIM: The study aimed to increase the body of knowledge for future research on forensic odontology by conducting a systematic review search to investigate possible forensic DNA in odontology associated with the importance of its application in the identification of humans of mass disasters.

MATERIALS AND METHODS: Six databases, including Google Scholar, PubMed, Scopus, Embase, Web of Science, and ScienceDirect, were analyzed using the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) quality scale. The literature search, conducted until February 2024, informed important research choices.

RESULTS: A total of 16 (100%) studies related to forensic DNA of mass disasters were identified. Of these, only seven (43.75%) articles related to dental DNA were included in this review. Of the 4,808 articles obtained, 138 duplicates or irrelevant articles were eliminated. Following a full-text review, seven studies were selected based on eligibility criteria. The highest percentage of victims was identified using dental DNA. In a few studies, some samples were insufficient for complete DNA profiling due to factors such as the method of DNA extraction.

CONCLUSION: Forensic odontology has played a significant role in the identification of victims of several mass disasters around the world. Although teeth are an excellent source of DNA for humans, future studies with larger sample sizes, appropriate control groups, and standardized techniques of DNA extraction need to be conducted.

Keywords: human identification; DNA fingerprinting; forensic odontology; forensic DNA; teeth.

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Связь между судебно-медицинской экспертизой ДНК и судебной одонтологией при идентификации личности в случаях массовой гибели людей: систематический обзор

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АННОТАЦИЯ

Обоснование. Согласно литературным данным, судебная одонтология является одним из наиболее эффективных и доступных научных методов идентификации жертв массовых бедствий. В данном исследовании рассматриваются роль и функции судебных одонтологов, участвовавших в расследовании нескольких крупных массовых бедствий. Проведение судебно-медицинской экспертизы ДНК в одонтологии обусловлено её важностью при идентификации жертв массовых бедствий. В данном исследовании рассматриваются в рассовых бедствий. Поскольку уровень преступности продолжает расти, область судебной медицины претерпела значительные изменения. Судебные стоматологи играют ключевую роль в различных областях исследования места преступления, помогая тем самым раскрывать многочисленные случаи гибели людей.

Цель работы — расширение базы знаний для будущих исследований в области судебной одонтологии путём проведения систематического обзора публикаций, связанных с судебно-медицинской экспертизой ДНК методами одонтологи для идентификации жертв массовых бедствий.

Материалы и методы. Поиск публикаций осуществлялся до февраля 2024 года в базах данных Google Scholar, PubMed, Scopus, Embase, Web of Science и ScienceDirect. Отбор научных работ выполняли в соответствии с чек-листом PRISMA. Результаты. Всего было отобрано 16 (100%) исследований, посвящённых судебно-медицинской экспертизе ДНК, проводимой на образцах, взятых у жертв массовых бедствий, из них только 7 (43,75%) статей связаны с анализом ДНК зубов. Из 4808 статей было исключено 138 дубликатов или публикаций, не относящихся к теме исследования. После полнотекстового обзора было отобрано 7 работ, соответстовавших критериям включения. Наибольшее количество жертв было идентифицировано путём выделения ДНК из образцов зубов. В нескольких исследованиях авторам не удалось выполнить полное ДНК-профилирование с учётом применяемого метода извлечения ДНК.

Заключение. Анализ ДНК сыграл важную роль в идентификации жертв нескольких массовых бедствий по всему миру. Несмотря на тот факт, что зубы человека являются отличным источником ДНК, в будущем будет полезным провести ряд исследований с бо́льшим размером выборки и соответствующими контрольными группами с использованием стандартизированных методов извлечения ДНК.

Ключевые слова: идентификация личности; ДНК-дактилоскопия; судебная одонтология; судебно-медицинская экспертиза ДНК; зубы.

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大规模死亡案件中识别个人身份时法医 DNA 鉴定与法医牙 科学的关系:系统性综述

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摘要

论证。根据文献数据,法医牙科学是识别大规模灾难受害者身份的最有效、最

便捷的科学方法之一。本研究探讨了参与侦查几个重大大规模灾难的法医牙医的作用和职能。法医 DNA 鉴定在识别大规模灾难受害者身份中的重要性决定其在牙科学的作用。随着犯罪率的不断上升,法医牙科领域也发生了重大变化。法医牙医在犯罪现场调查的各个领域 发挥着关键作用,从而有助于破获无数起死亡案件。

该研究的目的是通过对与法医 DNA 鉴定在识别大规模灾难受害者身份中有关的出版物进行 系统性综述,以拓宽法医牙科学未来研究的知识基础。

材料与方法。在 Google Scholar、PubMed、Scopus、Embase、Web of Science 和 ScienceDirect 等数据库中检索了截至 2024 年 2 月的文献。科学论文根据 PRISMA 检查 表进行筛选。

结果。共有 16 项关于对大规模灾难受害者标本进行法医 DNA 鉴定的研究(100%)被选中。其中,只有 7 篇文章(43.75%)与牙科 DNA 分析有关。在 4808 篇文章中,138 篇 重复或与研究主题无关的文章被排除在外。经过全文审阅,选出了 7 篇符合纳入标准的论文。通过从牙科标本中分离 DNA 来确认身份的受害者人数最多。在少数研究中,由于使用 的 DNA 提取方法不同,作者无法进行完整的 DNA 指纹分析。

结论。DNA 分析在识别世界各地几个大规模灾难受害者身份的方面发挥了重要作用。尽管人 类牙齿是 DNA 的绝佳来源,但使用标准化的 DNA 提取方法进行样本量更大、对照组更适当 的研究在未来将大有用武之地。

关键词: 个人身份识别; DNA 指纹分析; 法医牙科学; 法医 DNA 鉴定; 牙齿。

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BACKGROUND

Mass disasters (MDs) are defined as abrupt, severe, unanticipated, and indiscriminate events that typically result in a high death toll and necessitate substantial management resources [1]. MDs are often classified as natural, accidental, or criminal events such as fires, explosions, tsunamis, earthquakes, or burnt or decayed bodies that may be difficult for humans to identify [2]. In these situations, where there may not be enough remains for morphological testing like fingerprints, the key function of human identification may be played by investigative or forensic genetics using DNA analysis [3]. Similarly, identifying victims of disasters requires correlating available antemortem data with postmortem reports. In situations where antemortem records are not available, forensic DNA becomes the only trustworthy means of identification [4]. Teeth are abundant sources of high-quality DNA that can be used in all forensic investigations, making them a valuable consideration DNA analysis [5]. This is crucial not only for the surviving family but also to satisfy legal requirements [6].

Teeth DNA aids in primary identification and serves as an adjuvant reference sample to relate the other tissue fragments. Teeth DNA helps with primary identification and can be used to estimate age, determine blood group, and determine sex [7]. Because teeth are positioned where they are most protected by the muscles and bone, they are the most resilient structure in the body and can withstand extreme physical and chemical conditions. They are also a great source of DNA because of their decreased risk of contamination [8–10].

Because teeth are resistant to desiccation, fire, and decomposition, dental remains, such as restorations and anatomy, are used for identification because it is an economical, a dependable procedure, and does not degrade the material in the event of burns or severe injuries [11]. Comparing postmortem and antemortem records is the standard procedure for human identification. Since its debut in 1985, DNA fingerprinting has also proven to be a successful technology [12, 13]. DNA is the fundamental genetic material that comprises most of the human genome and carries all the instructions required to build, assemble, and preserve life in all of its forms. It is chemically referred to as doublestranded DNA. Numerous linearly arranged DNA nucleotides make up the polymer chain of a DNA molecule. Two of these DNA strands are joined by non-covalent links to form a double helix. DNA molecules have structural components called nucleosides, which consist of a base, a sugar, and a backbone. The genetic information is carried out via the four bases: adenine (A), cytosine (C), quanine (G), and thymine (T) [14].

Since DNA is unique to each individual, it is a crucial component of human identity. Changes in genes and the epigenome can potentially be used as indicators for the diagnosis and prognosis of various illnesses. For example, Mansueto et al. have demonstrated that circulating mitochondrial DNA (mtDNA) is a predictor of poor prognosis in patients with heart failure, and higher amounts of cell-free DNA indicate the extent of cellular damage [15]. One of the most stable natural substances, DNA can be used for human identity and is stable enough to be stored for millions of years without losing its structure. These characteristics make DNA a valuable analytical tool in forensic investigations [16, 17].

There are two different kinds of DNA: mtDNA and genomic DNA, which is located in the nucleus. The most common type of DNA that is extracted is genomic DNA. In cases where samples are poor or deteriorated, mtDNA is examined. Additionally, DNA can be categorized as non-autosomal (genomal), which is found in sperm and ova, and autosomal, which is found in autosomal cells [18].

In the interest of justice, forensic dentistry — also known as forensic odontology (F0) — is the branch of dentistry that deals with the appropriate administration, review, assessment, and presentation of dental evidence in criminal or civil legal processes [19]. Techniques used in F0 for identification include reviewing dental case files, conducting anthropological evaluations, and examining restorations, dentures, radiographs, bite marks, intraoral photos, cheiloscopy, and rugoscopy. The mouth cavity's protected environment makes dental pulp the most dependable source for DNA-based identification techniques. In addition to solving specific cases, forensic odontologists can identify victims in medical instances [20]. According to research, F0 is one of the most practical and affordable scientific techniques for identifying victims in MDs.

Therefore, the aim of this research was to increase the body of knowledge for future research on FO by conducting a systematic review search to investigate possible forensic DNA in odontology associated with the importance of its application in the identification of humans of MDs.

This article provides a comprehensive overview of FO's history, range of techniques, and applications. It emphasizes the importance of maintaining accurate stomatology records and supplying the necessary data to enable law enforcement to identify missing persons and victims in medical cases.

MATERIALS AND METHODS

This systematic review was conducted using the Systematic Review Methodology Guidelines [21] and the Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA) procedure [22]. The registration number for 2024 CRD42024522628 may be found on the PROSPERO website, which is part of the International Prospective Register of Systematic Reviews — Centre for Reviews and Dissemination at the University of York.

The search strategy involved six electronic databases: Google Scholar, PubMed, Scopus, Embase, Web of Science, and ScienceDirect. We searched PubMed, Scopus, and Web of Science (last search date January 27, 2024), Embase (last

search date February 25, 2024), Google Scholar (last search date February 15, 2024), and ScienceDirect (last search date February 13, 2024) for relevant studies. The search strategy for the PubMed database was as follows: ("Forensic DNA" OR DNA OR SNP OR mtDNA OR mDNA OR "autosomal DNA" OR "DNA profiling" OR "deoxyribonucleic acid" OR "DNA Profiles" OR "DNA typing" OR "genetic profiling" OR "dental DNA" OR "DNA identification" OR "genetic fingerprint" OR "DNA-fingerprinting techniques" OR "mitochondrial DNA" OR "mitochondrial genome" OR "human mitochondrial DNA" OR "human mtDNA" OR "DNA methylation" OR "nuclear DNA" OR "DNA guantification" OR "DNA analysis" OR "genomic DNA" OR "DNA amplification" OR "forensic DNA analysis" OR "forensic DNA technology" OR "forensic DNA Profiling" OR "DNA evidence" OR "DNA test" OR "DNA testing" OR polymorphisms OR "DNA Fingerprinting" [Mesh]) AND ("Forensic genetics" [Mesh] OR genetics OR genealogy OR gene OR genes OR GWAS OR hereditary OR congenital OR inherited OR inherent OR inheritable OR heritable OR "molecular genetics" OR "human genetics" OR "genetics research" OR "human genome project" OR "human molecular genetics" OR "Genome-wide association studies" OR "human molecular genetics")) AND (Forensic OR "mass disasters" "forensic stomatology" OR "forensic odontology" OR "forensic legal" OR "forensic sciences" OR "forensic biology" OR "forensic chemistry" OR "forensic serology" OR "forensic medicine" OR "forensic legal" OR "medical jurisprudence" OR "forensic anthropology" OR "dental studies" OR "dental anthropology" OR "Forensic Dentistry" [Mesh]). This strategy

Table 1. Results of the systematic review	V
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was adopted for use in the electronic bibliographic databases Scopus, Embase, Google Scholar, Web of Science, and ScienceDirect. Depending on the database, plural forms of the search terms, as well as quotation marks, were used.

The eligibility criteria for inclusion were as follows: peer-reviewed, published papers written in English, French, Spanish, and Chinese that reported victim identification in MDs. The PICO strategy was used to address articles as follows: involved participants (P) — any age or sex; intervention (I) — forensic identification of victims; control (C) — not applicable; and outcome (O) — forensic odonatological identification. Also, we analyzed cross-sectional studies, case-control studies, case studies, systematic reviews, and meta-analyses from 1964 to 2024. Duplicate papers were excluded from the analysis.

Strategies were reviewed, and the analyzed data was displayed as text and split by gene. Additionally, the data are summarized in Table 1, considering the presence or absence of an association between forensic DNA, forensic genetics, and its application in F0.

All records found via electronic database searches were put into the literature management program Rayyan (Intelligent Systematic Review) for article screening and selection. Two independent reviewers (LTRM and CENE) evaluated the eligibility of titles, abstracts, and full-text publications based on the following inclusion criteria after deleting duplicates: published from 1964 to February 2024. A total of 4,808 papers in English, French, Spanish, and Chinese, as well as human research studies that addressed

Author	Year	Disaster	Country	Total number of victims	Number of identified victims (%)	Dental DNA used	The methodology used for identification
Hsu et al. [24]		accident	Taiwan	202	All	Yes	DNA analysis
Nambiar et al. [25]		accident	Malaysia	34	All	Yes	DNA analysis
Bux et al. [26]	2006	Accident	Nepal	18	14; 77.77%	-	Fingerprint+DNA
Tan et al. [27]	2007	Accident	Indonesia	104	6; 5.7%	-	Fingerprint+DNA
Schuller-Götzburg et al. [28]	2007	Natural	Thailand	4,280	2,679; 62.59	Yes	Fingerprint+DNA
Prieto et al. [29]	2007	Criminal	Spain	191	All	-	Fingerprint+DNA
Bush and Miller [30]	2011	Accident	USA	50	All	-	Fingerprint+DNA
Hinchliffe [31]	2011	Accident	Australia	173	172; 99.42%	-	Fingerprint+DNA
Trengrove et al. [32]	2011	Natural	New Zealand	181	177; 97.79%	Yes	Fingerprint+DNA
Manhart et al. [33]	2012	Accident	Germany	8	All	-	DNA analysis
Barberia et al. [34]	2015	Accident	Spain	12	All	-	Fingerprint + DNA
Obafunwa et al. [35]	2015	Accident	Nigeria	152	148; 97.36%	-	DNA analysis
lino et al. [36]	2015	Natural	Japan	15,892	15,736; 99.01%	-	Fingerprint + DNA
de Boer et al. [37]	2018	Accident	Ukraine	298	297; 98.2%	Yes	DNA analysis
Marrone et al. [38]	2021	Criminal	Italy	10	9; 90%	Yes	DNA analysis
Dahal et al. [39]	2022	Natural	Nepal	400	365; 91.25%	Yes	DNA analysis

SYSTEMATIC REVIEWS

different methods of FO for identifying victims in MDs, were included our review. In addition, we examined the association between forensic DNA in odontology and its importance in FO or a particular application by examining its relationship with forensic genetics, forensic DNA, or DNA fingerprinting. This systematic review excluded studies that did not assess or quantify DNA from teeth, as well as studies that used sources other than human samples from individuals or victims. Editorial pieces, letters to the editor, and review articles were also excluded from the study. Article exclusion criteria were recorded, and discrepancies between the two reviewers were addressed with the assistance of a third reviewer (MdLSDA) throughout the paper review process. If additional information was required, the review team contacted the authors directly.

Data Synthesis

Two impartial reviewers collected information from relevant publications, entered it into an Excel spreadsheet, and provided a narrative and tabular summary of the findings. The extracted data included authors, year of publication, year of the disaster, study size, type of disaster, subtype of disaster, country, state, total number of victims, total number of victims identified, total number of victims identified by FO, whether identification by FO was used in combination with other methods, and details of other methods of identification. Disagreements were resolved by discussion, and consensus was reached through a third party.

The principal data extractors (LTRM and CENE) assessed the quality of the study, and at least one additional co-author validated their findings. We utilized the elements from the National Institutes of Health's Quality Assessment Tool for Observational Cohort and Cross-Sectional Studies [23] to guide the evaluation of study quality. The previously listed data extraction components were taken into consideration during the evaluation of article quality. Every article that was included was assessed and rated as either good, fair, or poor based on these criteria.

RESULTS

Literature Search

An initial search of six databases PubMed electronic databases yielded 4,808 records, including 18 from Web of Science, 387 from Scopus, 92 from Embase, 1998 from ScienceDirect, and 716 from Google Scholar. One hundred thirty-eight records were removed due to duplicates. From the remaining 45 records, 4,757 irrelevant citations were removed after the title and abstract review. A further 29 ineligible articles were removed after comprehensive evaluation based on inclusion and exclusion criteria. Ultimately, 16 articles [24–39] were included in the qualitative synthesis, and only seven were research. The process is shown in Figure 1.

Articles Identified

The initial database search identified 4,808 articles (Fig. 1). Among these, 10 papers reported findings on the association between forensic DNA in odontology and the identification of humans in MDs. These articles met the inclusion criteria based on the PICOS (Participants, Interventions, Comparators, Outcomes, Study Design) statement of the systematic literature search. Studies that did not assess or quantify DNA from teeth samples used by individuals or victims, as well as study designs, such as editorials, letters to the editor, or review articles, were excluded from the study.



Fig. 1. Block diagram of PRISMA.

Characteristics of Included Studies

The papers were published from 1964 to 2024. More than 81% of articles were cross-sectional, cohort, reviews, or case-control studies. Table 1 shows the general characteristics of the included studies.

Disaster type and geographic location. Of the included disasters (16), 10 (62.5%) were accidental [24–27, 30, 31, 33–35, 37], four (25%) were natural [28, 32, 36, 39], and two (12.5%) were criminal [29,38]. The 10 accidental MDs included airplane crashes [24–28, 30, 35, 37] (n=8; 50%), explosive accidents [38] (n=1; 6.25%), train accidents [34] (n=1; 6.25%), and a vehicle accident [33] (n=1; 6.25%). Of the four natural MDs (100%), two (50%) were tsunamis [28, 36], and two (50%) were earthquake [32, 39]. Four MDs (25%) were in Europe (Italy [33], 1; Ukraine [37], 1; Germany [33], 1; and Spain [29, 34], 2), seven (43.75%) were in Asia (two each in Taiwan [24], Malaysia [25], Nepal [26, 39], Indonesia [27], Thailand [28] and Japan [36]), one (6.25%) was in Australia [31], one (6.25%) was in the United States [30], and one (6.25%) each was in Africa (Nigeria) [35] and New Zealand [32].

Victim Identification in forensic odontology

Of the included MDs, the greatest number of victims was associated with the Japan tsunami [36] (n=15,892), followed by the Thailand tsunami [28] (n=4,280), the United States airplane crash disaster [30] (n=3,407), the Nepal earthquake [39] (n=400), the airplane crash of Ukraine [37] (298), the Spain terrorist attack [29] (n=191), the New Zealand earthquake [32] (n=177), the Australian bushfire of 2009 [31] (n=172), the Taiwan air crash [24] (n=202), and the Malaysia air crash [25] (n=34). The German vehicle accident [33] (n=8), the Spain train accident [34] (n=12), and the Nepal airplane crash [26] (n=18) were associated with the lowest numbers of victims. A total of 22,001 victims were reported from 16 MDs, of which 20,096 victims (91.34%) were positively identified. All victims (100%) were identified in the Spain terrorist attack [29], the Newark air crash [30], the Spain train accident [34], the Malaysia air crash [25], the Taiwan air crash [24], and the German vehicle accident [33]. The greatest number of victims was identified from the Thailand tsunami [28] (2,679; 62.59%), the MH17 airplane crash in 2014 of Ukraine [37] (298; 98.2%), the Australian bushfire of 2009 [31] (172; 99.42%), followed by the Japan tsunami [36] (15,736; 99.01%), the Nepal earthquake [39] (365; 91.25%), the New Zealand earthquake [32] (177; 97.79%), the Nigeria air crash [35] (148; 97.36%), the Indonesia air crash [35] (6; 5.7%), the Nepal airplane crash [26] (14; 77.77%) and the Italy case of an explosion of a pyrotechnic artifice factory [38] (9; 90%).

Forensic DNA Identification. Of 22,005 victims, 20,100 victims (91.34%) were positively identified using forensic methodology. Of the 16 research papers selected in this review (100%), victims were identified as MDs using forensic DNA methodology, and seven research papers (43.75%) used dental DNA for identified victims. Details for the methods

are given in Table 1. In two accidental MDs (German vehicle accident [33] and Spain train accident [34]), in one criminal MD (Spain terrorist attack [29]), the Nepal airplane crash [26], the Indonesia air crash [35], the Newark air crash [30], the Australian bushfire of 2009 [31], the Nigeria air crash [35] and the Japan tsunami [36] dental DNA was not used for victim identification. Still, it was used in other mythologies of F0. Finally, victims were identified from seven papers [24, 25, 28, 32, 37–39] using dental DNA in combination with other methodologies.

Role of DNA in forensic research

DNA plays a crucial role in forensic research due to its unique characteristics, which make it an invaluable tool for identifying individuals, solving crimes, and exonerating the innocent [5]. Similarly, DNA analysis has become indispensable in forensic research, offering valuable insights into criminal investigations, victim identification, and the administration of justice. Its reliability, accuracy, and ability to provide unique genetic information make DNA a cornerstone of modern forensic science [5].

Any type of organism can be identified by examination of DNA sequences unique to that species. Every cell of an individual carries a copy of the DNA. The order of base pairs in the DNA of every individual is different except for identical twins. The uniqueness is due to the intron regions of DNA, which contain sequences that are 20–100 bp in length that are repeated at different locations (loci) along the chromosome like AGACTAGACATT — AGATTAGGCATT, which are called sequence polymorphism. The length polymorphism like (–AATG) (AATG) (AATG) (three repeats) and (AATG) (AATG) (two repeats) are termed as short tandem repeats (STRs), which are used in forensic identification [40, 41]. Some key roles of DNA in forensic research are as follows:

Identification of Individuals: DNA profiling allows forensic scientists to create unique genetic profiles for individuals based on variations in their DNA sequences. This enables the identification of suspects, victims, and unknown individuals involved in criminal investigations or MDs [5].

Crime Scene Analysis: DNA collected from biological samples at crime scenes, such as blood, semen, saliva, or hair, can be analyzed to establish connections between suspects, victims, and the crime scene. DNA evidence can provide crucial information about the perpetrator's identity, how the crime was committed, and the timeline of events [42].

Exoneration of the Innocent: DNA analysis has led to the exoneration of numerous individuals wrongfully convicted of crimes they did not commit. By comparing DNA profiles from crime scene evidence with DNA samples from convicted individuals or suspects, forensic scientists can accurately determine innocence or guilt [43].

Cold Case Investigations: DNA technology has revolutionized cold case investigations by enabling forensic scientists to reanalyze evidence using advanced DNA profiling techniques. This has resulted in resolving many long-standing unsolved cases, bringing closure to victims and their families [44].

Missing Persons Identification: DNA analysis is used to identify missing persons by comparing DNA profiles from unidentified human remains with DNA samples from relatives or reference databases. This process helps to reunite families, provide closure to loved ones, and aid in criminal investigations [45].

Disaster Victim Identification: Following natural disasters, accidents, or mass casualties, DNA profiling is used to identify victims when traditional methods, such as visual identification, are not possible. DNA samples obtained from personal belongings or biological tissues are compared with reference samples to establish the identities of the deceased [46].

Genetic Genealogy: DNA analysis techniques are increasingly being used in genetic genealogy (GG) to trace family lineages and identify relatives of unknown individuals. This approach has been instrumental in solving cold cases and identifying perpetrators through their familial DNA connections [47].

Forensic DNA analysis

Within the field of forensic science, forensic DNA analysis entails the identification and comparison of DNA samples for both legal and investigative purposes. It makes use of several methods to extract, amplify, and analyze DNA from biological evidence collected from individuals under investigation or from crime scenes [5, 48]. Similarly, forensic DNA analysis has transformed criminal investigations and the the legal system by offering a potent instrument for locating suspects, connecting offenders to offenses, clearing innocent people, and closing unsolved cases. However, forensic scientists must strictly adhere to procedures, quality control measures, and ethical standards to ensure the accuracy and dependability of DNA evidence presented in court [5,48].

DNA-containing biological samples are taken from crime scenes, victims, suspects, and other pertinent subjects. Blood, semen, saliva, hair follicles, skin cells, and body fluids are common sources of DNA evidence [39]. Specialized laboratory techniques are used to extract the DNA from collected samples. This process typically involves cracking up the cells to release the DNA and then purifying the mixture to get rid of any proteins, fats, and other impurities [49, 51].

The amount of DNA extracted from the sample is quantified to determine its concentration. This step ensures that there is enough DNA for subsequent analysis and helps optimize the amplification process [50].

Amplification: The polymerase chain reaction (PCR) is used to amplify specific regions of the DNA. PCR allows forensic scientists to make millions of copies of the DNA fragments, even if the initial sample is minute. This step is crucial for generating enough DNA material for downstream analysis [51].

The amplified DNA fragments can be analyzed using a variety of techniques to provide a DNA profile. STR analysis is a popular method that looks at particular DNA segments called microsatellites. Every individual has a different number of repeats at each microsatellite locus, allowing for the creation of a distinct genetic profile for every individual [52].

Information: The DNA profile interpreted from the crime scene sample is cross-referenced with the DNA profiles of known individuals, including victims and suspects. The significance of any matches or similarities between the profiles is evaluated using statistical techniques [53].

Forensic scientists compile their findings into a report detailing the DNA analysis results and conclusions. They may be required to testify in court as expert witnesses to explain the methods used and the significance of the DNA evidence in the case [54].

DNA fingerprinting

DNA fingerprinting, sometimes called genetic fingerprinting, DNA typing, or DNA profiling, is a method for determining and examining each person's distinct genetic traits. To generate a distinct genetic profile, it is based on examining extremely variable DNA regions in a person [52, 55]. DNA fingerprinting has emerged as a key component of forensic science Since it offers incredibly precise and dependable techniques for identifying people, connecting them to crime scenes, and resolving criminal cases. It has also been applied in non-forensic fields like ancestry, paternity, and wildlife protection [52, 55].

According to the foundational idea of DNA fingerprinting, every person (except identical twins) has a distinct DNA profile. Variations in particular genomic areas, like variable number tandem repeats (VNTRs) or STRs, which show significant inter-individual variability, are the source of this uniqueness [56]. Blood, saliva, semen, hair roots, tissue, and other biological samples that contain DNA are taken from crime scenes, suspects, victims, and other people who may be of interest. A sample's DNA quality and quantity are essential for a successful DNA profiling process [49]. Different extraction techniques are used to extract DNA from the obtained samples. These techniques usually include dissolving cell membranes, isolating DNA from other biological constituents, and cleaning the recovered DNA to eliminate impurities [49, 51].

Forensic DNA Profiling

In forensic research, a method known as forensic DNA profiling, also referred to as DNA fingerprinting or DNA typing, is used to identify individuals by examining their distinctive DNA profiles. This method is based on the premise that, except for identical twins who share the same DNA, each person's DNA sequence is unique [57, 58]. Like this, forensic DNA profiling is an effective tool in criminal investigations that has helped solve countless cases by connecting suspects to crime sites or clearing individuals falsely accused

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of crimes. It is widely acknowledged as a trustworthy and good scientific technique for identifying people using their genetic information [57, 58].

One of the first methods of forensic DNA profiling was applying restriction fragment length polymorphism (RFLP) [8, 59]. Using the enzyme restriction endonucleases, specific sites along the DNA sequence are cut using this technique. After the resulting DNA fragments are separated by size using agarose gel electrophoresis, they are transferred to a nitrocellulose slide, hybridized with locus-specific probes (either radioactive or chemiluminescent), and then visualized as bands by autoradiography or chemiluminescence. RFLP is rarely utilized in forensics today since it requires a lot of DNA. Particular DNA sections, referred to as genetic markers or loci, can be used with PCR. Usually, single nucleotide polymorphisms (SNPs) or STR make up these genetic markers. The process of PCR amplification makes it possible to produce enough DNA fragments for further examination [8, 59].

The human nuclear genome: The HNG (human nuclear genome) is the complete set of genetic information stored within the nucleus of a human cell. It consists of DNA, which is organized into chromosomes. Humans typically have 46 chromosomes arranged in 23 pairs, with 22 pairs of autosomes and one pair of sex chromosomes (XX for females and XY for males) [60–62]. The human genome was first sequenced in the early 2000s as part of the Human Genome Project (HGP), a collaborative effort involving scientists worldwide. Since then, advancements in sequencing technologies have made it faster and more cost-effective to sequence individual genomes, leading to significant progress in understanding human genetic variation and its implications for health and disease [61, 62].

Variable number of tandem repeats: These are sections of the human genome that repeat a short DNA sequence in tandem, typically lasting 10-60 base pairs. These repetitions occur at a certain locus, a chromosomal region. The term "variable number" in variable number of tandem repeats (VNTRs) describes how a population's repetition counts at a certain locus can vary significantly within a population [63-65]. Likewise, VNTRs are important genetic markers due to their polymorphic nature and widespread distribution throughout the human genome. Their variability provides a powerful tool for genetic analysis, enabling the identification of individuals and the study of genetic diversity within populations [63–65]. Also, polymorphism of VNTR loci is highly significant, meaning that the number of repeats varies significantly between individuals. This polymorphism makes VNTRs valuable for DNA profiling and forensic analysis, as each person's VNTR profile is unique, except for identical twins [66]. VNTRs are inherited in a Mendelian fashion, meaning they are passed down from parents to offspring. The number of repeats at a VNTR locus in an individual's DNA is determined by the combination of alleles inherited from their parents [67].

Restriction fragment length polymorphism: This is one of the earliest techniques for DNA, but it is now largely obsolete. This method uses a unique type of restriction enzyme called "restriction endonuclease," which sections DNA at a specific sequence pattern known as a restriction endonuclease recognition site. It is used to analyze the different lengths of DNA fragments that emerge from digesting a DNA sample. The use of RFLP has decreased due to the advent of more effective and modern DNA analysis methods. This is because RFLP requires comparatively large volumes of DNA, cannot be performed on deteriorated samples due to environmental conditions, and takes longer to obtain findings [68-70].

RFLP is a molecular biology technique used to analyze variations in DNA sequences among individuals. It was one of the earliest methods developed for DNA profiling and genetic fingerprinting before more advanced techniques like PCR-based methods. RFLP was widely used in forensics and genetics research until it was largely replaced by PCR-based methods like STR analysis, which are faster, more sensitive, and require smaller amounts of DNA [68-70].

D1S80 (pmCT118) locus: The human genome's D1S80 locus, also known as pmCT118, is a VNTR region found on chromosome 1. It has a 16-base pair repeating sequence that is replicated in varying numbers among different individuals [71, 72]. Because the D1S80 gene is polymorphic, it has been extensively employed in forensic DNA profiling and paternity testing. Individual variations in the amount of repeats at this locus are notable, with alleles ranging from roughly 9 to 48 repeats or more [71–73].

Gene Polymorphisms

The human genome's intergenic regions include polymorphism regions, repeating sequences with a high degree of variation. This may be called a "length polymorphism" or a "sequence polymorphism." These regions contain DNA polymorphisms that may be used in genetic mapping and forensic investigations. DNA has two types of repeating sequences: tandem repeats and interspersed repeats, sometimes known as random repeats. Moreover, there are two kinds of tandem repeats: STRs, often called "microsatellites," and VNTRs, also called "minisatellites." [74, 75]. The primary repeat units of STRs are two to six base pairs, whereas those of VNTRs vary from 10 to 100 base pairs (bp). Both STRs and VNTRs are useful tools for determining DNA, but STRs are more abundant in the genome than VNTRs, making them easier to characterize. The foundation for genetically identifying a person and defining its genetic profile is the variation in loci, which are repetitive sequences between people. Length polymorphism refers to the differences in the number of tandem repeat units. Point mutations, sometimes called SNPs, are variations in a single base pair of a sequence [74,75].

Gene Polymorphisms (GPs) also refer to the occurrence of multiple genetic variants, or alleles, at a specific locus

(location) within a population's gene pool. These variations can manifest as differences in DNA sequences, gene expression levels, or protein structures. GPs contribute to genetic diversity among individuals and populations and play important roles in evolution, adaptation, and disease susceptibility [74–76]. Similarly, GPs represent a fundamental aspect of genetic variation in populations and are essential for understanding the genetic basis of traits, diseases, and population dynamics. Studying GPs provides insights into human evolution, adaptation, and the genetic factors underlying complex biological phenomena [74–76].

Types of GPs: There are several types of GPs, including [77-82]:

SNPs: Variations in a single nucleotide base at a specific position in the DNA sequence.

- Insertions/deletions (Indels): Variations involving the insertion or deletion of nucleotide bases, resulting in length differences in the DNA sequence.
- STRs: Variations in the number of repeated sequences of short nucleotide motifs, also known as microsatellites.
- Copy number variations (CNVs): Differences in the number of copies of a particular DNA segment, ranging from kilobases to megabases in size.
- VNTRs: Similar to STRs, VNTRs are variations in the number of repeated sequences, but the repeat units are typically longer.
- Gene expression polymorphisms: Variations in gene expression levels due to regulatory sequence differences or epigenetic modifications.

Origin and maintenance of GPs arise through various mechanisms, including mutation, genetic recombination, gene duplication, and genetic drift. Polymorphisms are maintained in populations through factors such as balancing selection, heterozygote advantage, frequency-dependent selection, and genetic drift [83].

Genetic diversity in GPs contributes to genetic diversity within and between populations. Different populations may exhibit variations in allele frequencies due to factors such as geographic isolation, migration, and natural selection [84]. Disease susceptibility of some GPs is associated with an increased or decreased risk of developing certain diseases. For example, specific SNP variants in genes involved in metabolism, immunity, or cell growth may influence an individual's susceptibility to conditions such as cancer, cardiovascular disease, diabetes, or autoimmune disorders [85].

Biotechnologies for forensic DNA analysis

Biotechnologies have greatly advanced forensic DNA analysis, enabling the identification and detailed characterization of DNA evidence collected from crime scenes, victims, and suspects. These technological advancements have revolutionized forensic investigations, providing powerful tools to solve crimes, exonerate the innocent, and ensure justice [86]. Likewise, biotechnologies are pivotal in forensic DNA analysis, providing innovative tools and methodologies for analyzing DNA evidence, identifying individuals, and advancing forensic science capabilities. These technologies continue to evolve, offering greater sensitivity, specificity, and efficiency in forensic DNA testing and contributing to the ongoing pursuit of justice and truth in criminal investigations [86].

New technologies, including massively parallel sequencing, microfluidics, integrated fast PCR systems, and real-time PCR, are described. Expert systems have also been created to help with data processing from these sophisticated analytical tools [59]. Technological developments in forensic DNA detection were greatly aided by Kary Mullis's discovery of the PCR in 1986. DNA fingerprinting, created by Alec Jeffrey in 1986, was first employed by forensic investigators. In a laboratory setting, the first step in processing evidence is to extract the DNA molecule from biological material [87]. PCR-based analysis offers the advantage of being highly sensitive and less time-consuming than earlier methods [88].

PCR is a fundamental biotechnology used to amplify specific DNA regions, including STRs and other genetic markers. PCR amplification enables forensic scientists to produce sufficient quantities of DNA from small or degraded samples, such as those collected from crime scenes or ancient remains [89].

Capillary Electrophoresis: Capillary Electrophoresis (CE) is a technique that separates and analyzes DNA fragments based on their size and charge. In forensic DNA analysis, CE is commonly employed to detect and measure the lengths of amplified DNA fragments, such as those produced by PCR amplification of autosomal STRs or mtDNA [90].

Next-Generation Sequencing: Next-Generation Sequencing (NGS) technologies enable high-throughput sequencing of DNA, offering comprehensive genetic information from forensic samples. NGS can be used to sequence entire genomes, analyze mtDNA, identify SNP, and detect genetic variants associated with traits or diseases. NGS is particularly useful for analyzing complex DNA mixtures and degraded samples [91].

Quantitative PCR: Quantitative PCR (qPCR) is a sensitive technique used to quantify the amount of DNA present in forensic samples. qPCR assays can accurately measure the concentration of target DNA molecules, allowing forensic scientists to assess DNA quantity, detect low-level DNA contamination, and determine the success of DNA extraction and amplification processes [92].

Automated DNA Extraction Systems: Automated DNA extraction systems streamline the process of isolating DNA from forensic samples by automating sample processing, DNA purification, and extraction. These systems enhance workflow efficiency, reduce contamination risks, and ensure consistent DNA yields from different sample types [93].

Rapid DNA Analysis: Rapid DNA analysis platforms enable on-site and near real-time DNA testing, facilitating rapid identification of individuals in law enforcement, disaster response, and border security applications. These portable systems automate sample processing, DNA amplification, and analysis, delivering DNA profiles within hours without the need for specialized laboratory equipment [94].

Forensic DNA Databases: Forensic DNA databases store DNA profiles obtained from convicted offenders, crime scenes, and unidentified remains. Biotechnologies are used to populate and search these databases, match DNA profiles to known individuals or other crime scenes, and support criminal investigations and legal proceedings [95].

Gas chromatography-mass spectrometry: This is widely used in fields such as forensic medicine, among others, due to its high sensitivity, selectivity, and ability to analyze complex mixtures. It is an indispensable tool for chemists and researchers in characterizing and quantifying organic compounds in diverse samples [96].

GC-infrared: This analytical technique combines the separating power of GC with the identification capabilities of IR spectroscopy. It allows for the simultaneous separation and identification of components in a complex mixture [97].

Lab-on-a-chip technology: This refers to the miniaturization and integration of laboratory functions onto a single chip or device, typically only a few millimeters to a few centimeters in size. These "chips" can perform tasks that would traditionally require entire laboratories, offering benefits such as reduced sample and reagent volumes, faster analysis times, portability, and potential cost savings [98, 99]. Several challenges hinder the widespread adoption of lab-on-a-chip technology (LOC), including the necessity for standardization, integration of multiple functions onto a single chip, scalability for mass production, and compatibility with existing instrumentation and workflows. However, continuous advancements in microfabrication techniques, sensor technologies, and bioinformatics are steadily overcoming these obstacles, thereby paving the way for increasingly sophisticated and versatile lab-on-a-chip applications [98, 99].

Autosomal short tandem repeats profile

An autosomal STR profile, often referred to simply as a DNA profile, is a genetic fingerprint obtained from analyzing specific regions of the autosomal DNA in an individual's genome. Autosomal DNA is found in chromosomes other than the sex chromosomes (X and Y). STRs are repeating sequences of DNA that vary in length among individuals [100, 101]. Similarly, autosomal STR profiling is a powerful tool in forensic genetics, providing robust and reliable DNA-based identification methods that have revolutionized criminal investigations and forensic science practices [100, 101].

Five to 10% of the human genome comprises sequences that repeat themselves. A section of DNA known as the STR is home to many tandem repeats. The DNA sequence in such STR units is two to six base pairs, with more than 105 STRs in the human genome [102, 103]. The number of tandem repeat units at the STR locus differs from individual to individual, determining the genotype for human identification. A minimal quantity of DNA (about 1 ng) is required for the profiling of STRs, which are present on all chromosomes, including sex chromosomes. This amount is 50 times less than that required for RFLP analysis. A 1 ng of the DNA sample can be amplified using the PCR technique to obtain a complete STR profile, Furthermore, PCR amplification of multiple STR loci can be performed simultaneously in the same PCR tube [104], a method known as multiplex PCR or multiplexing. Commercially available STR markers are often used in forensic DNA profiling. For instance, the Combined DNA Index System (CODIS) is the name given to the top 13 STR markers created by the Federal Bureau of Investigation (FBI) in the United States. According to Malik et al. [104], the STR locus is of three types:

- Simple STR: The repeating units are of identical length and sequence.
- Composite STR: Consists of two or more adjacent simple repeats.
- Complex STR: Has several repeating blocks of different unit lengths and variable intermediate sequences.

Fluorescent dye-labeled primers are employed to amplify the STR loci after DNA extraction from the sample. The amplified products are sorted and located using CE after a few PCR cycles. The data are generated by the computer and shown as peaks on an electropherogram. Each DNA fragment-specific dye is found by the detector. A peak represents each DNA fragment that has been found. The dimensions of each peak are determined using allelic ladders and size criteria. Every allelic ladder is appropriately solved to determine the correct STR allele [104].

Genetic variation of autosomal STRs are regions of DNA where short sequences of nucleotides (usually two to seven base pairs in length) are repeated multiple times. The number of repeats at each STR locus can vary between individuals, leading to genetic diversity within the population [105]. Autosomal STRs are inherited from both parents, with one allele (variant) originating from the mother and the other from the father. As a result, an individual's autosomal STR profile reflects a combination of alleles inherited from both parents [106].

Autosomal STR profiling is widely used in forensic genetics for human identification purposes. Forensic DNA laboratories analyze multiple autosomal STR loci simultaneously using PCR amplification and CE. By comparing the lengths of amplified DNA fragments at different STR loci, a unique genetic profile can be generated for each individual [107]. Unique identifier of the probability of two individuals having the same autosomal STR profile by chance is extremely low, making autosomal STRs highly effective for individual identification. The likelihood of a random match depends on the number of STR loci analyzed and the frequency of alleles within the population [108].

CODIS

The CODIS is a DNA database system that is managed by the FBI in the United States. It is mostly used for forensic identification, which allows law enforcement to store and examine DNA profiles from suspects found guilty of particular crimes, as well as evidence found at crime scenes [109, 110]. Similarly, it is crucial to remember that CODIS is only available to authorized law enforcement officials, and stringent guidelines control the distribution and use of DNA data to safeguard individuals' right to privacy. Furthermore, CODIS is only one forensic investigative instrument, and its efficacy depends on the caliber and volume of DNA evidence gathered and the precision of DNA analysis methods [109, 110].

CODIS was specifically designed to enable public forensic DNA laboratories to turn approved DNA profiles into searchable databases. DNA data may be shared and compared between laboratories in the United States thanks to the CODIS software. It also offers a central database that includes all of the user laboratories' DNA profiles. It is approximately one in a billion chance that two people will have identical 13-loci DNA profiles. TH01, TPOX, CSF1PO, vWA, FGA, D3S1358, D5S818, D7S820, D13S317, D16S539, D8S1179, D18S51, and D21S11 are the 13 CODIS locations. The 13 basic STR loci are covered by STR multiplexes that normally contain the sex type marker amelogenin. With more than 60 million records as of 2007, the United States is home to the largest DNA database in the world, the CODIS [111–113].

Single nucleotide polymorphisms

Variations in a single nucleotide base at a particular location in the genome that happen relatively frequently within a population are known as SNPs. These variants, which comprise over 90% of all human genetic diversity, are the most prevalent genetic variation among individuals [114, 115]. Similarly, SNPs are fundamental units of genetic variation that contribute to the diversity of traits and susceptibility to diseases within human populations. Their broad distribution across the genome and their association with various phenotypic traits make them valuable targets for genetic research in diverse fields, including medicine, forensics, and evolutionary biology [114, 115].

Variations that occur between individuals in a single base sequence at a particular point in the genome are called SNP [102]. SNPs are the most common form of genetic variation in humans. It can result from substituting, deleting, or inserting bases at a single site. They occur every 100 to 300 bases along the DNA strand. Across the human genome, around 1.4 million SNPs have been identified and, therefore, have the potential to be used as markers for forensic applications. SNPs offer advantages in forensic applications over STR profiles, as PCR amplification of SNP markers can better withstand degraded DNA samples and can be multiplied to a higher level than STRs [104, 116]. To achieve enough discriminating power, however, many SNPs must be analyzed. While SNPs are not likely to take the role of STR analysis, they may be a useful supplementary tool in difficult forensic instances [117]. Here are some key points about SNPs:

"The oral cavity is the black box of the organism" in forensic investigation

The statement "The oral cavity is the black box of the organism" in forensic investigation reflects the concept that the oral cavity contains a wealth of valuable information crucial in forensic analyses, akin to a black box containing vital data in investigations. Likewise, the statement highlights the importance of considering the oral cavity as a valuable source of evidence in forensic investigations. By acknowledging its relevance and employing appropriate evidence collection and analysis techniques, forensic scientists can uncover critical information that may contribute to solving crimes and establishing the truth in legal proceedings [118].

Because information from the oral cavity may be utilized to identify a person or provide details needed for legal processes, interdisciplinary competence is necessary for legal dentistry. In addition, information gleaned from the mouth cavity can be used to narrow a person's search parameters and is crucial for victim identification in the wake of largescale tragedies [104, 119]. The mouth is a rich, non-invasive source of DNA that can be utilized to identify individuals and supply data required for legal processes [104, 119].

The oral cavity is a complex anatomical region comprising various tissues, fluids, and structures that offer insights into an individual's health, lifestyle, and environmental interactions. Similar to a black box, the oral cavity may contain hidden information that requires thorough examination and analysis for discovery [120]. It serves as a rich source of evidence, providing a diverse range of forensic evidence, including saliva, buccal swabs, dental tissues, and oral microbiota. These sources of evidence can be valuable in identifying individuals, linking them to crime scenes, and providing clues about their activities or behaviors [120].

In forensic investigations, evidence from the oral cavity can be utilized for DNA profiling, saliva analysis (e.g., drug detection, identification of body fluids), comparison with dental records, bite mark analysis, and assessment of oral injuries or trauma. The oral cavity may also harbor traces of ingested or inhaled substances, which are pertinent in toxicology investigations [121]. The oral cavity presents challenges in forensic analysis, such as contamination risks, degradation of biological materials, and the need for specialized sampling techniques. However, advancements in forensic science and technology have expanded the capabilities for extracting and analyzing evidence from this anatomical region, opening new opportunities for investigation and evidence collection [122].

Teeth are the main sources of DNA for forensic research

While teeth are indeed valuable sources of DNA in forensic research, it is important to note that other sources, such as saliva, buccal swabs, and hair follicles, also play significant roles, depending on the nature of the case and available evidence. Forensic scientists employ various techniques and sample types to maximize the chances of obtaining viable DNA profiles for identification and investigative purposes [123].

In forensic investigations, the bodily fluids submitted for DNA analysis are often semen, blood, and saliva. However, teeth offer the best chance of extracting DNA from highly decayed, skeletonized, or burned individuals. In medicolegal death investigations, the source of DNA can be divided into two areas: first, DNA obtained from the outside of the body or from inside the body cavities (e.g., blood, saliva, semen, vaginal fluid, etc.); second, DNA from biological materials (e.g., liquid blood, bones, teeth, nails, etc.) [124]. Genetic information is abundant in the nucleated cells of teeth and the periodontal ligaments surrounding them. The tooth undergoes crushing or sectioning at different levels to extract DNA after a thorough morphological and radiological examination for identification and age estimate, respectively [125]. One of the most recommended methods for tooth DNA extraction is cryogenic polishing, involving sectioning the tooth at the cement-enamel junction for a conservative approach to dental DNA [126]. Depict the sequence of DNA extraction from the teeth. Alternatively, pulverizing or crushing the entire tooth into a fine powder can produce enough DNA for research [104]. However, during such investigations using teeth, recording of other tooth data, such as morphological details and radiographic data, must be performed before destruction of the tooth for DNA extraction [127, 128].

Dental pulp, a soft tissue located in the pulp chamber and root canals, is present in teeth. DNA-containing cells, mainly odontoblasts, fibroblasts, and endothelial cells, are abundant in dental pulp. The dental pulp is a great source for forensic DNA analysis since it usually delivers a high quantity and quality of DNA compared to other tissues [123] Teeth often provide excellent defense against environmental elements like heat, moisture, and deterioration. Tooth tissues are extremely mineralized and resistant to deterioration, including the dentin, enamel, and tooth pulp. This defense keeps DNA in teeth intact even in harsh environments like graveyards or major catastrophes [129–131].

Dental samples can be collected using minimally invasive methods, such as extracting a single tooth or small dental fragments. Alternatively, the dental pulp can be accessed through root canal procedures or post-mortem dental examinations. This ease of collection facilitates the retrieval of DNA samples from both living individuals and human remains [132, 133]. The dental records, including dental charts, X-rays, and photographs, provide valuable antemortem data for comparison with post-mortem dental features. Forensic odontologists can use this information to identify individuals based on dental characteristics and to guide the collection of dental samples for DNA analysis [134].

Stability and longevity of DNA extracted from dental tissues, particularly dental pulp, has been found to exhibit stability and longevity over extended periods. Studies have shown that DNA can be successfully extracted from dental samples even after years or decades of storage, making them valuable resources for resolving cold cases and identifying historical remains [135].

Sensitivity analysis and publication bias

The relevant odds ratios (ORs) were not included in the study's findings after excluding studies unrelated to forensic DNA. Consequently, these studies were retained in our systematic review. Sensitivity analyses verified the stability of the findings by showing that the overall impact remained mostly unchanged when specific datasets were excluded. Regarding the risk of bias, none of the included studies provided adequate information on the methodology for statistical analysis, justification for sample size or sample frame, or whether the selected samples were representative of the population.

DISCUSSION

Among the three types of MDs (natural, criminal, and accidental), natural MDs are most commonly characterized by a large number of victims [20]. The three accident MDs include this review systematic airplane crash [24,25,37]. (Taiwan, Malaysia, and Ukraine), three natural tsunami [36] (Thailand), an earthquake [32,39] (New Zealand and Nepal), and one criminal Italy explosion of a pyrotechnic artifice factory [38]. The greatest numbers of victims were associated with the Thailand tsunami [36] (n=4,280), followed by the Nepal [39] (n=400) and the New Zealand earthquake [32] (n=181). Victims in such situations can be scattered over large areas that extend for miles. Moreover, victims who are transients, homeless, or tourists pose problems for identification due to the unavailability of antemortem records. Despite these issues, a significant number of victims were identified in the 16 research papers selected for this review. All victims (100%) were identified using forensic DNA methodology. Likewise, seven research papers (43.75%) reported using dental DNA for victim identification. Forensic odontologists may face unique problems due to compromised infrastructure, destruction of antemortem records from local dental clinics, and loss of communication lines. All these factors can delay or preclude the prompt identification of victims, which is reflected in two accidental MDs (a German vehicle accident [33] and a Spain train accident [34]), in one criminal MD (a Spain terrorist attack [29]), the Nepal airplane crash [26], the Indonesia air crash [35], the Newark air crash [30], the Australian bushfire of 2009 [31], the Nigeria air crash [35], and the Japan tsunami [36]. Dental DNA was not used for victim identification but was used in other mythologies, such as forensic genetics.

Accidental MDs are of short duration and are usually associated with closely related populations. In the present systematic review, out of seven MDs, there were (three accidental, one criminal, and three natural) MDs: three air crashes [24, 25, 37] (42.85%), one Italy explosion of a pyrotechnic artifice factory [38] (14.28%), and three natural MDs [32, 36, 39] (42.85%). Such MDs may be associated with fewer victims, such as the Malaysia air crash [25] and the Italy explosion of a pyrotechnic artifice factory [38] (n=34 and n=10 victims, respectively). However, disasters, such as the Thailand tsunami disaster [36] (n=4,280), the Taiwan air crash [24] (n=202), the Ukraine air crash [37] (n=298), the Thailand tsunami [36] (n=4,280), the New Zealand earthquake [32] (n=181), and the Nepal earthquake [39] (n=400), were associated with a large number of victims identified through of dental DNA. Antemortem record retrieval is made easy in these kinds of MDs because the forensic experts are already familiar with the list of passengers. The increased percentage of victim identification in these MDs can be attributed to the simple access to antemortem records for each victim. This is reflected in the Taiwan air crash [24] and the Malaysia air cras [25], where all the victims were identified. Apart from these MDs, the largest number of victims was identified in the Thailand tsunami [28] (2,679; 62.59%), the MH17 airplane crash in 2014 in Ukraine [37] (298; 98.2%), and the Nepal earthquake [39] (365; 91.25%). In contrast, lower identification rates were observed in the Malaysia air crash [25] (n=34; 100%) and the Italy explosion of a pyrotechnic artifice factory [38] (n=9; 90%). Disasters associated with industries and the military can complicate victim identification due to the similar ages, sex, ethnic backgrounds, and clothing (uniforms) of the victims. However, related literature on industrial and military MDs was not available for analysis at this time.

Three natural MDs were identified through a literature search [28,32,39]. Natural MDs, unlike accidental ones, can happen over extended periods and over large areas of land. Serial killers may hide, dismember, and mutilate their victims' corpses. In certain cases, dental structures might not be accessible for post-mortem examination. The bodies of victims are sometimes mutilated to such an extent that only DNA identification can be used. In the New Zealand and Nepal natural MDs [32, 39], authors described the problems encountered during the identification of victims recovered from upper molar teeth collected from these victims for DNA samples [39]. External examinations were conducted by qualified professionals on each deceased individual. For unidentified bodies, scientists from the Nepal Police Forensic Science Laboratory took DNA samples and fingerprints. Swabs were obtained for DNA analysis by making an incision across the intercostal area, puncturing the lungs to allow any uncontaminated blood to leak out. A dental examination was then performed by forensic odontologists.

EM-DAT is a global database of technological and natural disasters that spans 1900 to the present and includes vital core data on the incidence and consequences of over 21,000 disasters worldwide. According to preliminary EM-DAT data gathered in 2017, 3,162 people died as a result of 149 disasters that took place in 73 different countries. This result highlights the number of MDs that happen globally [136]. Surprisingly, we could only locate seven publications (16 MDs) in the literature. This suggests that to have a better understanding of the roles played by odontologists and forensic experts, victim identification must be reported in future MDs. We will be able to create and simplify standard operating procedures for victim identification in MDs with the use of this reporting.

Of the total of 22,005, 20,100 victims (91.34%) were positively identified using forensic methodology. Of the 16 research papers selected in this review, victims of MDs were identified using forensic DNA methodology in all cases (100%). Dental DNA was the most commonly used modality, identified in seven out of 16 research papers (43.75%). Among these, victims were identified from seven papers [24, 25, 28, 32, 37–39] using dental DNA in combination with other methodologies. These data suggest that forensic odontological means can be a good adjunctive method for victim identification.

A total of 22,005, 20,100 victims (91.34%) were positively identified using different branches of forensic medicine. Of the 16 research papers selected for this review, all victims of MDs were identified using forensic DNA methodology (100%). Forensic DNA was the most commonly used modality, identified in nine research papers of 16 (56.25%) identified victims. Details about two accidental MDs (Germany vehicle accident [33] and Spain train accident [34]), in one criminal MD (Spain terrorist attack [29]), the Nepal airplane crash [26], the Indonesia air crash [35], the Newark air crash [30], the Australian bushfire of 2009 [31], the Nigeria air crash [35] and the Japan tsunami [36] dental DNA was not used for victim identification. While DNA fingerprinting is the most accurate and dependable approach for victim identification, the process is expensive, technique-sensitive, and timeconsuming, which requires advanced equipment and highly skilled lab professionals. Another need is that the samples must be pure, as contaminated samples cannot be tested, which could be a significant barrier to the application of this method in MDs. A small number of human mistakes might cause the procedure to go wrong or change the outcomes. Examples of such errors include introducing the sample to other substances or mistakingly recognizing two samples as identical.

Of the 20,100 victims (91.34%) identified, FO was useful in the identification of approximately (15.66%). The antemortem and post-mortem of the teeth (number, type, position, crown and root morphology, crown and root pathology, pulp chamber, root canal morphology, etc.)

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and dental prostheses (restorations, fixed and removable prosthesis, implants), periodontal ligaments, jawbones, and diseases related to jawbones are the dental factors that can be compared between antemortem and post-mortem. The majority of dental identifications are made simple for the records to establish by comparing restorations, cavities, missing teeth, and/or prosthetic devices [137]. As antemortem data, all of these records are available from teaching dental hospitals or general dentist practices (GDPs). For this reason, we think GDPs and/or dental institutions must maintain dental records. Every GDP must understand the forensic ramifications inherent in their profession. A dentist's professional responsibilities include creating, maintaining, and releasing clear and accurate patient records. Unfortunately, GDPs in India lack the necessary expertise and experience in maintaining accurate records [138]. Several studies have shown how critical it is to inform medical professionals about the value of keeping dental records and how these records can potentially be utilized to identify MD victims.

In FO, the only regulating factor necessary for successful victim identification is the caliber of antemortem evidence. This may be shown clearly in the Thailand tsunami MDs when only a tiny percentage of Thai victims were identified via FO because there was a dearth of antemortem dental data. On the other hand, almost 80% of foreign victims could be identified by their teeth [139, 140]. However, because so many dental offices were destroyed by the tsunami, odontologists found it challenging to gather antemortem dental records of the victims in Japan [36].

Study limitations

One of the major limitations of the included studies is that none of the studies mentioned the chemical composition of teeth, which could also affect the study of dental DNA and, therefore, could alter DNA yield [24, 25, 28, 32, 37–39]. There is also a lack of information on which part of the tooth is a good source of DNA. Hence, future studies need to report the chemical composition of the teeth of the MDs.

CONCLUSIONS

F0 has played a significant role in victim identification in various MDs worldwide. It serves as a useful adjuvant strategy because it has been successfully utilized in conjunction with other methodologies for victim identification. F0 is considered one of MD management's most dependable and affordable scientific approaches. However, the availability of antemortem records from GDPs is a major requirement for the success of F0-mediated identification. Therefore, GDPs should have sufficient understanding regarding F0 and maintain suitable dental records. The prevalence of MDs is projected to rise in the future due to factors such as an aging population, shifting climate patterns, more public transit options, and an increase in criminal activity. Therefore, in such dire circumstances, GDPs should acknowledge their national obligations.

Forensic DNA analysis is a valuable forensic medicine tool with far-reaching implications for criminal justice, humanitarian efforts, and scientific research. Its application in FO has revolutionized the field, providing invaluable tools for identifying individuals, solving crimes, and advancing knowledge. As technology continues to evolve and our understanding of dental DNA, forensic DNA analysis will remain at the forefront of forensic investigations, contributing to the pursuit of justice and truth.

ADDITIONAL INFORMATION

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