



SIGNIFICANT IMPORTANCE OF Y-STR IN SEXUAL OFFENCES; A NARRATIVE REVIEW

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ABSTRACT

Background/Objective: The male identification chromosomes are X, Y but in sexual assailants' cases, Y-STR chromosome is specified for investigation. The main advantage of Y-STR approach is the ability to detect the male component in mixed DNA profiles (two or more than two profiles) samples are collected from different sexual assailants' crime scenes like, e.g., sodomy, gang-rape cases, male and female mixed sample's like, Blood stains, Semen stains, Saliva stains and other fragile samples. In DNA analysis of such samples are gated mixed STR profiles, although differential extraction is one of the methods for differentiating the male and female DNA parts. **Methods;** A comprehensive literature narrative review is conducted to interpreted the knowledge on the application of Y-STR intron (non-coding) part of male chromosome in sexual assault cases, Child abuse crime cases. Y chromosome is a crucial in forensically and Genetically by its unique characteristics (inherited from paternal side) includes, exclusively presence small size and limited numbers of genes, SNP, STR, sex determination via the (SRY) gene, Haploid ("male-specific Y-STR haplotype paternal lineage) inheritance it's passed down directly from Father to its offspring's. However, this linearly transmitted genetic material is not mixed during reproduction and also used to study male lineage and ancestry. **Results;** The moral of this study evaluates the applicability of Y-STR in Forensic criminal investigation such as, Y-STR Haplotype reference Database and Results (YHRD), male DNA Y-STR in Genetics and criminal case study in sexual offenses. **Conclusion;** Y-STR plays a crucial role in Forensics science criminal investigation like, Gang-rape sexual assault, Child abuse & sexual assault cases, Murder cases, Genetics lineage, rubbery cases and also Kinship cases study.

Keywords: Y Chromosome, Y-STR, Sexual offences, YHRD, Forensic Genetic, Kinship, Case Study.

INTRODUCTION

The analysis of Y-chromosome STRs (Y-STR) is widely an essential tool in forensic genetics, particularly in sexual crime cases. Genetically; males hold one pair of sex chromosome-X and Y-located within the nucleus of each cell, those chromosomes are directly transferred from parents ("X from Mother & Y from Father") respectively. During fertility when an X and Y chromosome normally are phenotypically male and having two x chromosomes are phenotypically female, either having

same number of autosomal 22 pair of chromosomes. A haplotype is the set of short-tandem repeat (STR) alleles typed on single Y chromosome (1). Y-chromosome is widely used in forensic DNA analysis particularly in cases where standard autosomal DNA profiling is not informative. Gen fragment of Y-chromosome is applied for identification as a trace contributor between mixed DNA (male and female) collected from crime scene; Such as, sexual assault cases, identify the paternal lineage of male perpetrator, highlight involvement of multiple contributor, unknown, kinship, missing person, mass disaster victim identification as well as historical cases involving men (2).

Pakistan South Asia Country were selected as a focus of this study due to their complementary characteristics in terms of population genetics and forensic infrastructure. Pakistan has relatively well-established Y-STR population database; however, despite several completed population studies, the available data remain limited in both geographic coverage and the number of loci analyzed, preventing, a comprehensive understanding of the country's genetic diversity (3). The markers are valuable in situations where male DNA is present in minimal quantities or overwhelmed by a large proportion of female DNA. The male DNA Y-STRs enable the precise identification of male genetic profiles, even a complex sample, offering a robust approach for forensic investigation.

This article examines the forensic application and importance of Y-STRs in account of sexual offences, like sodomy, gang-rape cases, mass disaster, kinship, Y-STR Haplotype Reference Database (YHRD) also discussing their advantages, limitations, and provide recommendations to maximize the importance and impact on the criminal justice system.

METHODS

In June 2025, a comprehensive search was conducted in Google Scholar, PubMed (U.S National Library of Medicine) without restrictions on publication date or language to identify the relevant literature of the application on Importance of Y-STR in sexual offences. The key words used included, Y chromosome, Y-STR Forensic Genetics, DNA mixture, sexual crime, Kinship, Autosomal markers, and YHRD (Y-STR Haplotype Reference Database). The review focused on recent advances in Y-STRs application, their limitations and their Forensic impact in solving complex cases, particularly those involving sexual crimes.

The inclusion criteria comprised peer-reviewed scientific articles that addressed the application of Y-STRs in Forensic Genetic, particularly sexual crimes with emphasis on DNA mixture analysis, YHRD Database, RM-STR marker articles discussing relevant ethical, legal, and methodological aspect were also included.

Exclusion criteria involved duplicate records articles not directly related to the forensic application of Y-STRs (e.g., studies with a purely evolutionary or clinical Database), papers without full-text access, and systemic reviews lacking original data or discussion. The literature synthesis was performed through a narrative approach, organized around the following thematic areas; (1) genetic and biological characteristics of Y-STR markers; (2) application of Y-STR analysis in mixed and degraded DNA; (3) forensic utility of Y-STRs in sexual and violent crime investigations; (4) interpretation frameworks and population databases including YHRD and, (5) Limitations ethical Considerations, and emerging advancements in Y-STR typing.

Analytical approaches of Y-STR in sexual assault evidence

Mainly in sexual offence cases investigator collect five types of samples like, peripheral & menstrual blood, semen, saliva and vaginal & rectal secretion from assault victim by the help of Sexual Assault & Evidence Collection Kit (SAECK). However, many forensic laboratories face new challenges to separate mixed sample collected as evidence like, epithelial cells, non-epithelial cells (gland secretion). Most of the Forensic laboratories are engaging different types of extraction methods are used to different chemical lysis in DNA protocol for processing. Often sexual assault samples are separate by the help of Predominant Differential Extraction (DE) organic method, DE is a modified extraction method that separate female epithelial cells and sperm cells. It breaks open the female epithelial cells with an incubation in an SDS/Proteinase K, dithiothreitol DTT mixture (4). Interpreting mixture often one of the most complex aspect of forensic DNA analysis. Unlike

autosomal DNA short tandem repeat markers, Y-STR haplotype mixtures are more difficult to analyze because the Y chromosome does not undergo recombination. Consequently, a Y-STR profile is assessed as a single haplotype rather than an individual locus. The discriminatory capacity of Y-STR is notably high, it approximately 99.9% when using 10 forensically selected Y-STR markers, and increasing to about 99.99% when markers are analyzed (5).

The Y-Chromosome and its Application in Forensic Genetics

Normal males possess one Y-chromosome (XY), while females lack it (XX). The Y-chromosome with a relatively small size of about ~50-60 Mb including ~24 Mb of euchromatin and 30 Mb of heterochromatin regions, both arms consist pseudo-autosomal region (PARs) and also contain non-recombining region (NRY), it transferred from father to its offspring without significant changes, unless a mutation occur at a given locus. The Y-chromosome exists to determine maleness by specifying the development of the testis early in embryogenesis, majority of the Y-chromosome does not undergo recombination and is always in a haploid state. These characteristics makes Y-STRs short tandem repeats valuable tools in identification of male/female DNA mixtures, ancestral origin analysis, family genealogical study, as well as historical study in the field of forensic investigation (6)-(7)-(8).

Genetic Structure and Inheritance Pattern of Y-STR Markers

The Y chromosome is uniquely inherited along the paternal lineage without undergoing meiotic recombination across most of its length, except for the pseudo autosomal regions (PARs). The non-recombining region of the Y chromosome (NRY) contains the majority of forensic Y-STRs loci and is transmitted as a single haplotype from father to son. This haploid inheritance pattern makes Y-STRs fundamentally different from autosomal STRs, where alleles segregate independently and recombine between generations(9). Y-STR markers are composed of short tandem repeats located primarily within non-coding intronic regions of the Y chromosome. Due to the absence of recombination, all Y-STR loci are inherited together, forming a stable paternal lineage marker. This characteristic is particularly useful in forensic investigations involving male-specific identification, lineage tracing, and kinship analysis (1).

However, the lack of recombination also reduces the individual discriminatory power of Y-STRs compared to autosomal STRs. As a result, Y-STR profiles cannot uniquely identify an individual but rather indicate a shared paternal lineage. Despite this limitation, Y-STRs remains indispensable in cases where autosomal STRs fail to provide informative results, particularly in sexual assault investigation involving male-female DNA mixtures.

Real Case Studies of Y-STRs in Forensic Investigation

The DNA & Serology Department, at Punjab Forensic Science Agency (PFSA) Lahore, Pakistan, is a state of art Forensic Facility allowing law investigation and courts to deliver justice to the people. The DNA Database at PFSA is an exclusive facility in Pakistan, which submitted items at including clothing, vaginal & Anal swabs, from victim and suspects. During examination of evidences a preliminary and confirmatory test were performed to solve high profiled crimes. Forensic DNA Y-STRs Database have proven to be essential in various forensic investigations, particularly in cases involving sexual crimes where male DNA is a crucial part of the evidence. Below are two Significant cases in which Y-STRs Played a central role in solving crimes and identifying suspects.

Foreign DNA Rapid Case: Punjab Forensic Science Agency

In March, 2017, a nine-year-old minor girl was raped in district khusab (Province Punjab), indicate brutal sexual violented victim was examined by medico-legal section. Police apprehended a suspect who was excluded as the source of foreign male DNA from tested evidence items, an unknown male DNA profile obtained from evidence items was uploaded to PFSA DNA Database to maintain record. After that, a suspect was arrested by Karachi (Province Sindh), police in another rape case, DNA profile of suspected was searched in the PFSA Database. This generated DNA profile matched with the foreign DNA profile Obtained from evidence items victim(10).

Mortality of Zanib Ansari; High Profile Sexual Assault case

One of the land mark case Zanib's solved with the help of DNA Y-STR Database of Punjab Forensic Science Agency (PFSA). Zanib Ansari, a Seven year (07) old from Kasur, Panjab was missing on January 4, 2018, while enroot to her Quran class. Serial killer suspect had committed 12 case of Sexual Assault and murders within a radius of 2.5 kilometer. A total of 1187 sample were collected for DNA analysis, where as the 814th sample to be of the criminal's data. However, the use of Y-STRs allowed investigators to trace the paternal lineage of the perpetrator, ultimately leading to the identification of the culprit. This case demonstrated the value of Y-STRs not only in identifying suspects directly, but also in locating relatives, which enabled the resolution of a long-unsolved case. These examples highlight how Y-STRs are essential in investigating cold or archived cases, particularly when DNA mixtures are involved or when the offender's profile is difficult to access due to predominant presence of female DNA. The application of Y-STRs not only in identifying perpetrators but also helps exclude suspects and strengthen links to the crime scene, offering a powerful tool for investigators. The integration of Y-STRs with other forensic technologies, such as investigative genealogy, has become increasingly useful in resolving old and complex cases(11)-(12).

Y-STRs in Mixed and Degraded DNA Sample

One of the most significant forensic advantages of Y-STR analysis lies in its ability to detect male DNA in samples overwhelmed by female DNA. In sexual assault cases, vaginal, anal, or oral swabs often contain a high proportion of female epithelial cells, which can mask the male contributor when using autosomal STR profiling. Y-STR selectively amplify male DNA, thereby enabling the identification of male genetic material even when present in trace amount(13). Y-STR analysis is also effective in degraded samples commonly encountered in forensic casework, such as aged stains, environmentally exposed evidence, or samples subjected to chemical or biological degradation. The relatively small amplicon sizes of modern Y-STR kits increase the likelihood of successful amplification from compromised DNA(14). In cases involving multiple male contributors, such as gang-rape or group assaults, Y-STR profiles may reveal complex haplotype mixtures. Although interpretation can be challenging, the detection of multiple male haplotypes provides critical investigative leads that may otherwise be missed using autosomal markers alone.

Rapidly Mutating Y-STRs (RM Y-STR) and Their Forensic Significance

Rapidly mutating Y-STRs (RM Y-STRs) represent a major advancement in forensic Y-chromosome analysis. These loci exhibit mutation rates significantly higher than conventional Y-STR markers, often exceeding 1×10^{-2} mutation per generation. This elevated mutation rate enhances discriminatory power related males, such as brothers, cousins, or paternal relatives(3). RM Y-STR are particularly valuable in sexual assault cases where the suspect belongs to the same paternal lineage as other potential contributors. By incorporating RM Y-STRs into standard Y-STR panels, forensic analysts can differentiate between male relatives who would otherwise share identical Y-STR haplotypes(15). The integration of RM Y-STR into forensic practice has significantly improved the resolution of Y-STR analysis, bridging the gap between lineage markers and individual identification while retaining male specificity.

Statistical Interpretation and Evidential value of Y-STR Profiles

The interpretation of Y-STR results relies on haplotype frequency estimation rather than locus-by-locus allele frequencies. Since Y-STR loci are inherited together, the evidential value is expressed as the frequency of the observed haplotype within a relevant population database(16). Likelihood ratios (LRs) are commonly used to evaluate the strength of Y-STR evidence by comparing the probability of observing the haplotype under competing hypotheses. However, due to shared paternal inheritance, Y-STR evidence must be interpreted cautiously and in conjunction with other forensic findings(17). The statistical weight of Y-STR evidence increases when rare haplotypes are observed or when results are combined with autosomal STRs, mitochondrial DNA, or case-specific contextual information.

Y-STR Haplotype Reference Database (YHRD) and Population Genetics

The Y-STR Haplotype Reference Database (YHRD) is the largest publicly accessible repository of Y-chromosomal haplotypes, containing data from diverse global populations. YHRD plays a crucial role in estimating haplotype frequencies and supporting statistical interpretation in forensic casework (<https://yhrd.org/>). Population-specific variation in Y-STR haplotype highlights the importance of regional databases, particularly for countries such as Pakistan, where population substructure, endogamy, and consanguinity may influence haplotype distributions. Expanding Y-STR datasets from South Asian Populations would significantly enhance the reliability of forensic interpretation in this region(9).

Limitations and Challenges of Y-STR Analysis

Despite its advantages, Y-STR analysis has inherent limitations. The inability to distinguish between paternally related males remains a key challenge, particularly in communities with strong paternal lineage structures. Additionally, the interpretation of mixed Y-STR profiles can be complex when multiple male contributors are present(14).

Mutation events, although relatively rare in conventional Y-STRs, may lead to exclusions or mismatches in kinship analysis. Furthermore, Y-STR evidence alone cannot provide individualization and should not be considered definitive proof of identity(18).

Ethical, Legal, and Courtroom Considerations

The use of Y-STRs raises important ethical and legal considerations, particularly regarding privacy and familial searching. Because Y-STRs reveal paternal lineage information, their use may inadvertently implicate male relatives of a suspect(16). In courtrooms, forensic experts must clearly communicate the limitations of Y-STR evidence to avoid misinterpretation by judges and juries. Transparent reporting, appropriate statistical support, and contextual explanation are essential to ensure fair judicial outcomes.

Emerging Technologies and Future Perspectives

Next-generation sequencing (NGS) and massively parallel sequencing (MPS) technologies are transforming forensic Y-chromosome analysis. These approaches enable simultaneous analysis of Y-STRs, Y-SNPs, and sequence-level variation, offering enhanced resolution and interpretative power (9).

Future Perspectives

The progress in the identification of RM Y-STRs represents a Significant advancement in the field of forensic genetics, enabling a more precise discrimination between individuals belonging to the same paternal lineage and substantially expanding the potential for solving complex cases. However, the successful implementation of these advances requires not only modernized laboratory infrastructure and validated methodologies but also the continuous training and qualification of forensic experts to ensure the accurate interpretation and application of genetic data.

Simultaneously, the expansion of genetic databases and the use of mass screening strategies, such as those employed in large-scale investigations (e.g., DNA dragnets), must be approached with extreme caution, particularly regarding informed consent, the risk of unauthorized secondary use of genetic data, and the potential stigmatization of minority groups.

The increasing adoption of Y-STR analysis and the enhancement of genetic databases should not be regarded as a standalone solution, but rather as a complementary tool, achieving its full potential only when integrated into a judicial system that is ethical, technically proficient, and socially responsible. In this context, international cooperation plays a vital role, facilitating not only the exchange of best practices in both laboratory and judicial domains but also the harmonization of data protection standards and ethical procedures, with special consideration given to the national contexts of Pakistan. The Future of forensic genetic and of Y-STR analysis will inevitably depend on maintaining a careful balance between technological innovation, scientific rigor, and protection of fundamental rights. This

balance is essential not only to ensure technical reliability but also to strengthen public trust and uphold the legitimacy of using genetic data in judicial processes.

DISCUSSION

This study presented highlights the importance of the Y chromosome and Y-STRs in forensic genetics, especially in complex cases of sexual crimes. The ability to identify male DNA in mixtures containing large quantities of female genetic material represents a significant scientific advance. However, despite the well-established applications of this method, it is essential to critically assess the technical and contextual limitations that may affect the reliability of the result.

Among the technical challenges, allelic dropout is a prominent issue, especially in degraded or low-template sample. This phenomenon compromises the completeness of genetic profiles, directly impacting the capacity for comparison and individual identification. Furthermore, DNA degradation (frequently observed in samples exposed to adverse environment conditions), may impair the uniform amplification of all loci, resulting in partial profiles and limiting the scope of forensic interpretation. The high sensitivity of amplification protocols employed in Y-STR typing, while enabling the analysis of extremely small quantities of genetic material, also heightens the risk of contamination. This necessitates rigorous laboratory protocols and stringent quality control measures during both the collection and analysis phases.

Another critical consideration involves the representativeness of population databases, such as the Y chromosome Haplotype Reference Database (YHRD), which are fundamental for estimating the probative value of a genetic profile. Underrepresented populations (such as PFSA) may introduce sampling bias and compromise the statistical robustness of the analyses, ultimately limiting the evidential value in forensic casework and increasing the risk of misinterpretation, particularly when inferring geographic origin. Beyond these technical limitations, it is also essential to consider the ethical and legal implications associated with the use and expansion of genetic database. In Pakistan, Law Qanoon-e-Shahadat Order (Articles 59 and 164), establishes clear guidelines for the collection, storage, use, and deletion of DNA profiles, aiming to balance the need of criminal investigations with the protection of fundamental rights, particularly regarding privacy and informed consent.

In Pakistan, personal data protection is regulated according to the Bill, 2023 (PDPB), which classifies genetic data as “sensitive data” and requires explicit consent for its processing. However, to date, no specific regulation has been enacted to address the unique challenges inherent to the forensic use of genetic profiles, leaving significant gaps in the protection of citizens, rights and in the establishment of best practices for forensic applications in judicial processes.

CONCLUSIONS

The expansion and diversification of Y-STR databases, combined with investments in technology and professional training, are essential to maximize the potential of these tools. At the same time, implementing clear and transparent ethical regulations will be indispensable to ensure public trust in the use of these technologies. By combining these efforts, it will be possible to strengthen forensic investigations and offer more robust and fair judicial responses of Pakistan.

The Y chromosome and Y-STRs emerge as indispensable tool in forensic genetics, offering precision in identifying male individuals and inferring paternal lineages. Their application is especially relevant in sexual crime cases, where the unique characteristics of Y-STRs enable effective analysis even in situations involving complex DNA mixtures. Additionally, recent advancements, such as RM Y-STR, have enhanced their discriminatory capacity, enabling differentiation between individuals with genetic connections and significantly increasing the scope of forensic investigations.

The statistics from Pakistan clearly illustrate the urgency of effective strategies to address sexual crimes. In Pakistan, the steady increase in reported cases, especially child sexual abuse, reflects a severe social issue, with most victims being young women and the perpetrators adult men. In Pakistan, the prevalence of sexual crimes such as child abuse and rape, with nearly all suspects being male, reinforces the need for robust genetic identification methods. These data, along with South Asia trends

showing a rise in sexual violence, underline the importance of tools such as Y-STRs in supporting criminal justice.

Therefore, the use of Y-STRs, combined with extensive and representative population databases, emerges as a viable and effective solution to assist forensic investigations, especially in contexts where conventional methods face limitations. By integrating advanced technologies with a detail understanding of regional realities, as demonstrated by the cases in Pakistan, it is possible to significantly improve the response to sexual crimes and strengthen the justice system.

While this article has focused on the specific realities of Pakistan, its findings can be extended to other countries that share similar forensic and infrastructural challenges. These insights may contribute to the development of forensic strategies tailored to local contexts while ensuring the protection of individual rights and ethical standards in the handling of genetic data.

Furthermore, investigations should be accompanied by clear regulations on the ethical use of genetic information, ensuring privacy and non-discrimination. The future of forensic investigations will depend on the combination of these advanced technologies with the public policies that ensure efficiency and equity across legal systems.

Data Availability Statement: No new data were created or analyzed in this study.

Conflicts of Interest: The author declares no conflicts of interest.

Abbreviations:

DE	Differential Extraction method
DTT	Dithiothreitol
SAECK	Sexual Assault Evidence Collection Kit
SDS	Sodium Dodecyl Sulphate
SNP	Single Nucleotide Polymorphism
SRY	Sex-Determining Region Y Chromosome
YHRD	Y chromosome Haplotype Reference Database
Y-STR	Y Chromosome Short Tandem Repeat

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