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Website: www.medicolegalupdate.org

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Published at

Institute of Medico-legal Publications

Logix Office Tower, Unit No. 1704, Logix City Centre Mall,
Sector- 32, Noida - 201 301 (Uttar Pradesh)

MLU Vol 25 No 2, April-June 2025

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Silent Scorcher: A Case of Heat Stroke Fatality-Case Report

Arjun Rajeev¹, Sujith Sreenivas C²

¹Junior Resident Medical Officer, ²Professor and Police Surgeon, Department of Forensic Medicine and Toxicology, Government Medical College, Kozhikode, Kerala, India

How to cite this article: Rajeev A, Sreenivas SC, Silent Scorcher: A Case of Heat Stroke Fatality - Case Report. 2025;25(2):1-5.

ABSTRACT

Background: Heat stroke is a life-threatening condition resulting from prolonged exposure to high temperatures, often exacerbated by physical exertion and inadequate hydration. Despite being preventable, it can lead to fatal outcomes if not promptly identified and treated. This case study highlights a fatal instance of heat stroke to emphasize its clinical manifestations, risk factors, and forensic importance.

Case Presentation: A 65-year-old male was found unresponsive after prolonged outdoor activity in extreme heat. Initial assessment revealed hyperthermia, altered mental status, and multi-organ dysfunction. Despite aggressive cooling measures and critical care interventions, the patient succumbed to complications arising from heatstroke, including rhabdomyolysis and acute kidney injury. The autopsy confirmed findings consistent with severe hyperthermia-induced organ damage.

Discussion: This case underscores the importance of early recognition and immediate management of heat stroke. Key contributing factors, such as environmental conditions, individual susceptibility, and delayed medical attention, are explored. The study also highlights the need for public awareness, preventive strategies, timely interventions to reduce the incidence of heat-related fatalities and diagnostic aspects in forensic.

Conclusion: Heat stroke remains a silent yet significant public health concern, particularly in regions experiencing rising temperatures. This case serves as a critical reminder of the deadly consequences of heat exposure and the urgent need for preventive action and how to diagnose such a case.

Keywords: Heat stroke, hyperthermia, rhabdomyolysis, acute kidney injury.

Introduction

Heat stroke is a life-threatening medical condition characterized by an elevated core body temperature typically over 40°C, accompanied by central nervous system dysfunction¹. It occurs in two forms: classic heatstroke, commonly seen in the elderly or very young, and exertional heat stroke, more prevalent in physically active individuals². Between 2000-2019 approximately 489,000 heat related deaths occur each year globally, with 45% of these in Asia and 36% in Europe.

According to India's crime record bureau, of 8060 accidental deaths attributable to natural causes in 2022, 9.1% were attributed to "Heat stroke". In 2021 India had reported 374 deaths due to heatstroke, which rose to 730 in 2022.

The case study presented here explores a fatal incident of heat stroke in a middle-aged individual, highlighting the critical role of forensic pathology in identifying the cause of death and the importance of this condition.

Corresponding author: Arjun Rajeev, Junior Resident Medical Officer, Department of Forensic Medicine and Toxicology, Government Medical College, Kozhikode, Kerala, India

E-mail: arjunmrajeev97@gmail.com

Submitted: Jan 25, 2025;

Accepted: Apr 2, 2025

Published: April-17-2025

Case Report

We report a 65-year-old male who had a sudden episode of collapse at a construction site where he was working presented to the emergency department. Upon arrival, his core body temperature was recorded at 42.1°C. Aggressive cooling measures were initiated, but the patient's condition rapidly deteriorated, leading to cardiovascular collapse and eventual death.

Autopsy Findings

Postmortem examination revealed no underlying medical conditions that could have predisposed the individual to heat stroke. Rectal temperature was measured using a thermometer measuring 40 deg C. The core body temperature within the pelvic region after opening the body seen to be elevated even after refrigeration. Other findings- The scalp showed petechial hemorrhages within the loose areolar tissues. Brain stem was softened with hemorrhages within the pons, medulla and caudate nucleus (Fig:1). The white matter of the cerebral hemispheres were markedly softened with multiple hemorrhages. There were no signs of raised intracranial pressure.



Fig. 1: Hemorrhages within the white matter

Petechial hemorrhages were seen on epicardial surface of heart. Subendocardial hemorrhages seen involving the papillary muscles and subaortic region (Fig:2)

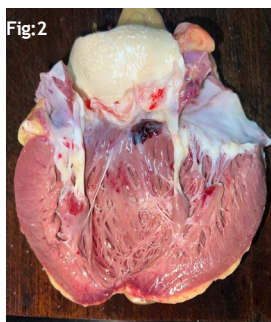


Fig. 2: Subendocardial hemorrhages

Myocardium of left ventricle appeared pale. Focal areas of pulmonary hemorrhages were seen (Fig:3). Both kidneys were pale, swollen and had loss of corticomedullary differentiation (Fig:4). Surface of liver showed slippage of capsule (Fig:5). Mucosa of intestines showed petechial hemorrhages. Urinary bladder contained 3- 5ml reddish brown colored urine. All internal organs appeared congested.



Fig. 3: Focal pulmonary hemorrhages



Fig. 4: Loss of CMD



Fig. 5: Slippage of capsule

Histopathological Examination

Dermis showing hemorrhages and congested vessels(fig:6). Skeletal muscle showed congestion with areas of muscle necrosis (fig:7) Heart showed multiple foci of cardiac myocyte necrosis (fig:8). Kidneys showed acute tubular necrosis(fig:9). Lungs showed pulmonary oedema and intraalveolar hemorrhage(fig:10).

Histopathological Findings

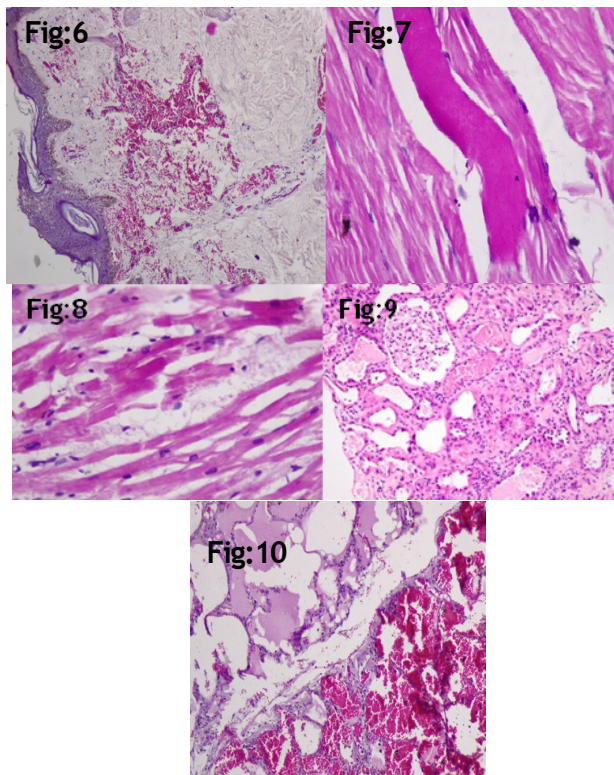


Fig. 6: Dermis showing hemorrhages and congested vessels; Fig. 7: Skeletal muscle showed congestion with areas of muscle necrosis; Fig. 8: Heart showed multiple foci of cardiac myocyte necrosis Fig. 9: Kidneys showed acute tubular necrosis; Fig. 10: Lungs showed pulmonary oedema and intraalveolar hemorrhage

Urinalysis report revealed RBCs were detected in large numbers; Myoglobin detected in urine (Myoglobinuria). Other culture and serology reports were unremarkable

The cause of death was determined to be exertional heat stroke, with the immediate trigger being the prolonged physical exertion and environmental heat exposure during possible construction work.

Discussion

Heat Stroke in Forensic Medicine Pathophysiology of Heat Stroke

The pathophysiology of heat stroke involves a complex cascade of events, including impaired thermoregulation, increased metabolic heat production and a breakdown of the body's homeostatic mechanism.³ During heatstroke, the body's core temperature rises rapidly, typically exceeding 40deg C, accompanied by central nervous dysfunction, organ failure and tissue damage. This physiological response is triggered by prolonged exposure to high ambient temperatures, often exacerbated by physical exertion and lack of adequate hydration.⁵

The pathophysiology of heat stroke is characterized by the inability of the body to dissipate heat effectively. As described in the literature, heat is gained from metabolic activities and the environment, and the body typically dissipates heat through sensible loss (conduction, convection and radiation) and insensible loss (evaporation of sweat).⁴ However, in heat stroke, one or more of the mechanisms may become impaired, leading to a rapid rise in body temperature.⁹

Importance of Comprehensive Forensic Analysis in Diagnosing Heat Stroke

A thorough and comprehensive forensic analysis is crucial in accurately diagnosing heat stroke. Due to the complex pathophysiology and the potential for heat stroke to mimic other causes of death, a multifaceted approach is necessary to differentiate heat stroke from other conditions. This comprehensive analysis involves a detailed examination of the circumstances surrounding the incident, a comprehensive autopsy, and comprehensive toxicological, histopathological and environmental assessments. By considering all available evidence and ruling out other potential causes of death, forensic experts can make a definitive diagnosis of heat stroke, which is essential for determining the cause and manner of death.

Differentiating Exertional Heat Stroke and Classic Heat Stroke

Heat stroke can be classified into two main types: exertional heat stroke and classic heat stroke. Understanding the key differences between these

two subtypes is essential for accurate diagnosis and appropriate management.

Exertional heat stroke typically occurs in individuals engaged in strenuous physical activity, such as athletes, laborers, or military personnel, who are exposed to high ambient temperatures and high humidity. In these cases, the increased metabolic heat production from exercise, combined with the environmental heat load, overwhelms the body's thermoregulatory mechanisms, leading to a rapid rise in core body temperature.

In contrast, classic heat stroke is more commonly observed in the elderly, young children, or individuals with underlying medical conditions, such as cardiovascular or neurological disorders. This type of heatstroke is often associated with prolonged exposure to high environmental temperatures, without the significant physical exertion seen in the exertional form.⁹

The clinical presentation and pathophysiological mechanisms underlying these two subtypes of heat stroke can differ, which may influence the diagnostic approach and the management strategies employed by forensic experts and healthcare professionals.

Acute Organ Damage in Heat Stroke Victims

Heat stroke can lead to rapid and severe damage to multiple organ systems.⁸ The elevated core body temperature and associated physiological disturbances can result in acute organ dysfunction, including:

- **Central nervous system:** Encephalopathy, seizures, coma
- **Cardiovascular system:** Myocardial injury, arrhythmias, hypotension
- **Respiratory system:** Acute respiratory distress syndrome, pulmonary edema
- **Renal system:** Acute kidney injury, rhabdomyolysis
- **Hepatic system:** Acute liver injury, coagulopathy
- **Gastrointestinal system:** Intestinal ischemia, gastrointestinal bleeding

Prompt recognition and aggressive management of these acute organ complications are crucial to preventing further deterioration and improving outcomes in heat stroke.

Histopathological Findings in Heat Stroke Cases

The neuropathological changes associated with heat stroke are complex and can involve multiple regions of the central nervous system. Severe hyperthermia can lead to direct thermal injury to brain tissue, resulting in widespread neuronal necrosis and gliosis. Additionally, the global physiological disturbances seen in heat stroke, such as impaired cerebral blood flow, oxidative stress, and metabolic derangements, can contribute to diffuse brain damage.

Histopathological examination of the brain in heat stroke cases often reveals acute ischemic changes, including neuronal shrinkage, cytoplasmic eosinophilia, and nuclear pyknosis. Cerebral edema and herniation may also be observed, reflecting the breakdown of the blood-brain barrier and the inability of the brain to effectively regulate its microenvironment.

In some cases, selective vulnerability of certain brain regions, such as the hippocampus, basal ganglia, and cerebellum, has been reported. These areas may be particularly susceptible to the metabolic and vascular derangements associated with heat stroke, leading to more pronounced neuropathological changes. Ultimately, the specific neuropathological findings in heat stroke cases can vary depending on the duration and severity of the hyperthermic insult, as well as the individual's underlying physiological and genetic factors that may influence the brain's response to extreme heat stress.

Toxicological Considerations in Heat Stroke Investigations

Toxicological assessments play a crucial role in the postmortem diagnosis and investigation of heat stroke cases. Comprehensive toxicological analyses can help identify any contributing factors, such as the presence of illicit drugs, medications, or other chemical substances that may have impaired the body's thermoregulatory mechanisms or exacerbated the physiological disturbances associated with heat stroke.

Role of Environmental Forensics in Heat Stroke Analysis

Environmental forensics plays a crucial role in the analysis of heat stroke cases. By investigating the environmental conditions and factors that contributed to the development of heat stroke, forensic experts can provide valuable insights to determine the cause and manner of death. This includes assessing ambient temperature, humidity, sun exposure, and other relevant environmental variables that may have exacerbated the heat-related illness⁷. Additionally, environmental forensics can help identify any potential contributing factors, such as the presence of urban heat islands, lack of shade or cooling infrastructure, or other environmental stressors that may have increased an individual's vulnerability to heat stroke. Integrating environmental forensic analysis with traditional medical and toxicological investigations is essential for a comprehensive understanding of heat stroke-related fatalities.

Establishing Cause and Manner of Death in Heat Stroke

The diagnosis of heat-related death be based on a history of exposure to high ambient temperature and the reasonable exclusion of other causes of hyperthermia. Postmortem examination, including complete autopsy, toxicological analysis, and environmental investigation, is crucial in establishing the cause and manner of death in heat stroke cases⁷. The presence of a core body temperature above 40°C (104°F), along with evidence of central nervous system dysfunction, such as delirium, seizures, or coma, is a key diagnostic criterion for heat stroke².

Careful consideration of the individual's medical history, any predisposing factors, and a thorough review of the circumstances surrounding the death are also essential for determining the cause and manner of death. In some cases, the cause of death may be attributed to heat stroke, while the manner of death could be classified as accidental, natural, or even homicide, depending on the specific circumstances and contributing factors involved.

Conclusion

Heat stroke is a life-threatening medical condition that requires prompt recognition and aggressive management to prevent severe complications and mortality. Multifactorial in nature, heat stroke can be challenging to diagnose and investigate, especially in forensic settings. A comprehensive approach, involving a thorough medical history, detailed physical examination, targeted laboratory testing, and a careful autopsy with supporting toxicological and environmental analyses, is crucial for establishing the cause and manner of death in heat stroke cases.

Financial Support and sponsorship: Nil

Conflicts of interest: There are no conflicts of interest.

Informed Consent: Not applicable as it is medicolegal autopsy-based case where subject confidentiality was maintained.

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Study of Pattern of Ligature Mark in Hanging Cases in Ahmedabad Region

¹Chandresh Tailor, ²Rakeshkumar Mori, ³Pratik Patel,

⁴Ni shith Chaudhari, ⁵Hardik Upadhyay, ⁶Yatin Sanghani

¹⁻³Associate Professor, Forensic Medicine Department, Govt. Medical College, Surat

⁴Associate Professor, Forensic Medicine Department, GMERS Medical College, Navsari

^{5,6}Resident doctor, Govt. Medical College, Surat

How to cite this article: Tailor C, Mori R, Patel P, Chaudhari N, Upadhyay H, Sanghani Y: Study of Pattern of Ligature Mark in Hanging Cases in Ahmedabad Region. 2025;25(2):6-9.

ABSTRACT

As a painless form of death hanging is the commonest method of committing suicide. A proper observation and study of ligature mark is characteristic hallmark of hanging. The ligature mark is a pressure abrasion on the neck at the site of the ligature which appears as a groove. This prospective study was conducted among victims of hanging brought to GMERS Medical College Sola, Ahmedabad during the period July 2018 to June 2022. 214 cases brought with a history of hanging out of total 2054 autopsy cases were selected for this prospective study. A typical ligature marks with partial hanging outnumbered typical ligature mark with complete hanging. Ligature mark was obliquely placed above the level of thyroid cartilage with a breadth of 1 to 2 cm is observed in the maximum number of cases. The colour of ligature mark was dark brown in one third of cases. Young adults of the age group 21 to 30 years accounted for the maximum cases 41.12% and the male: female ratio was 2:1. Churni (3%) was the most common ligature material used.

Keywords: Hanging, Ligature mark, Suicide, Thyroid cartilage

Introduction

Human suicidal behaviour has always been a source of dread and wonder to mankind. Hanging is the commonest method of committing suicide considered as a painless form of death. Hanging is a form of death produced by suspension of the body by a ligature around the neck, constricting force being the weight of the body (complete hanging), or part of the weight of the body (partial hanging).^[1] In India hanging is among the top 5 methods of choice for committing suicide.

In hanging the appreciation of external signs particularly ligature mark plays a vital role.

The ligature mark is a pressure abrasion on the neck at the site of the ligature which appears as a groove. The base of ligature mark is pale, hard leathery and parchment like and margins are red and congested. In typical hanging, the ligature mark is situated above the level of thyroid cartilage between the larynx and the chin. It is directed obliquely upwards along the line of the mandible and reaches the mastoid processes behind the ears. Character of the ligature mark depends on various factors like the nature of the ligature, body weight, length of time the body has remained suspended and the number of turns of the ligature round the neck. The course of the ligature mark depends

Corresponding author: Rakeshkumar Mori, Associate Professor, Forensic Medicine Department, Govt. Medical College, Surat

E-mail: rakeshmoree@gmail.com

Submitted: Nov 11, 2024

Accepted: Apr 2, 2025 **Published:** April-17-2025

on whether a fixed or running noose has been used.

In hanging, ligature material may be any substance that is available at the time of the impulse has been used by the suicides as a ligature. Knot is frequently in the form of a single knot to produce a running noose or fixed by a granny or reef knot, occasionally a simple loop is used.^[2] There may be more than one turn around the neck and/or more than one knot imparting corresponding complexity to the mark. A running noose can tighten at the time of suspension and may then produce a mark which takes a horizontal turn but it is likely to be above the thyroid cartilage. The mark will be circular and oblique if a ligature is passed round the neck more than once. Near the position of the knot, it is like an inverted 'V'. Ligature mark into consideration, there are a few points like (i) level (ii) continuous/ non-continuous (iii) oblique/ transverse of the ligature mark which differentiate hanging from ligature strangulation.

Materials and Methods

This prospective study was conducted among victims of hanging for the purpose of studying the pattern of ligature mark brought to GMERS

Medical College Sola, Ahmedabad during the period July 2018 to June 2022. Out Of 2054 cases brought to the department for medico-legal autopsy, 214 cases were identified and selected for this study in which death had resulted from hanging. A detailed history from police and relatives regarding age, sex, socio economical status, marital status, habits, illness (mental/ other disease/deformity), previous attempted suicides, suicide note if any etc. were taken. Detailed history from police regarding scene of crime, position of body etc. were taken.

Irrespective of information collected, both external and internal post mortem findings were observed meticulously especially the ligature mark. During observation of ligature mark, all the parameters like its site, size, level, number, discontinuity and obliquity were noted.

Results

Table 1: Type of Hanging - Depending on position of Knot

Type of Hanging	Cases	Percentage
Typical	41	19.16
Atypical	173	80.84
Total	214	100

Table 2: Type of Hanging- Depending on degree of Suspension

Type of Hanging	Cases	Percentage
Complete	138	64.48
Partial	76	35.52
Total	214	100

Table 3: According to the Level of Ligature Mark

Level of Ligature Mark	Cases	Percentage
Above the thyroid cartilage	180	84.12
Overriding the thyroid cartilage	22	10.28
Below the thyroid cartilage	12	05.60
Total	214	100

Table 4: Age and Sex Wise Distribution of death due to Hanging

Age group (Years)	Male	Female	Total	(Percentage)
0-10	3	1	4	01.87
11-20	28	16	44	20.56
21-30	58	30	88	41.12
31-40	26	16	42	19.63
41-50	14	9	23	10.75
51-60	5	3	8	03.74
>60	2	3	5	02.33
Total	136	78	214	100

Table 5: According to the Breadth of Ligature Mark

According to the Color of the Ligature Mark	Cases	Percentage
< 1cm	15	07.01
1-2 cm	142	66.36
2-3 cm	45	21.02
>3 cm	12	05.61
Total	214	100

Table 6: According to the Color of the Ligature Mark

Colour of ligature mark	Cases	Percentage
Dark brown	102	47.66
Red	35	16.36
Pale	26	12.15
Yellowish brown	51	23.83
Total	214	100

Table 7: According to the Characteristics of Ligature Mark

		Cases	Percentage
Depending on prominence	Prominent	177	82.71
	Faint	37	17.29
	Total	214	100
Depending on continuity	Continuous	12	05.60
	Interrupted	202	94.40
	Total	214	100
Depending on placement	Oblique	214	100
	Horizontal	00	00
	Total	214	100

Table 8: Distribution of type of Ligature material

Type of Ligature Material	No. of Cases	Percentage
Chunni	72	33.64
Nylon rope	56	26.16
Saree	40	18.69
Cotton rope	36	16.82
Shirt	04	01.87
Handkerchief	02	00.08
Metal wire	01	00.04
Not Known	03	01.40
Total (%)	214	100

Discussion

The hanging deaths are taking different types in their execution as typical/atypical and complete/partial. In this study, atypical hanging was seen in 173 (80.84%) cases and typical hanging in 41 (19.16%) cases. [Table 1] In this study, depending on degree of suspension, complete hangings were seen in 138 (64.48%) cases and partial hangings in 76 (35.52%) cases. [Table 2] Similar findings were observed in the studies conducted by other authors.^[1, 2]

In our study, it was observed that in 180 (84.12%) cases, the level of ligature mark was above the thyroid cartilage, overriding the thyroid cartilage in 22 (10.28%) cases and below the thyroid cartilage in 12 (05.60%) cases. [Table 3] This was also observed in various other authors studies.^[1, 5-11]

As per Table 4, maximum 88 cases (41.12%) of hanging deaths were reported in age group of 21-30 years, which is consistent with observations of Sheikh et al ^[12] (42.4%) and Joshi et al ^[13] (44.18%). Hanging deaths were reported in male were 136. 63.55%) cases and in female were 78 (36.44), which is consistent with observations of Sheikh et al ^[10] and Jani et al.^[12] The male: female ratio was approximately 1.75:1.

In present study the breadth of the ligature mark was between 1-2cms in 142 (66.36%) cases. [Table No.5] Similar findings are reported by others.^[7, 8, 14-16] The breadth of ligature mark depends on the width of the ligature material and also the multiplicity of the ligature material.

In this study, ligature mark was dark brown in 102 (47.66%) cases; yellowish brown in 51 (23.83%) cases; red colour in 35 (16.36%)

cases and pale in 26 (12.15%) cases. [Table 6] These are consistent with other author findings.

^[16] The reason being the colour of ligature mark depends on the duration of suspension and the complexion of the person.

The ligature mark was prominent in 177 (82.71%) cases and faint in 37 (17.29%) cases. [Table 7] Our findings were consistent with the findings observed in other studies ^[15-16]. The prominent mark is due to the type of the material being strong and also increased period of suspension. The ligature mark was interrupted in 202 (94.40%) cases and continuous in 12 (05.60%) cases.

As per Table 8, Chunni was used as a ligature material in maximum 72 cases (34.64%) of hanging followed by nylon rope in 56 cases (26.16%) cases. In study by Sharma B R et al ^[15] commonest ligature material was Chunni in 17 cases (30.90%)

Conclusion

Atypical ligature marks with complete hanging outnumbered typical ligature mark with partial hanging. From the medico-legal point of view, following measures and recommendations in cases of deaths due to hanging are very essential:

- Always bring the Ligature material along with the body for correlation with the Ligature mark.
- Photograph of the scene of occurrence should include point of suspension.
- In fatal cases not to disturb the ligature material and release only the suspension point or cut the ligature material away from the site of knot.
- Photograph of the scene of occurrence should include point of suspension.
- Radiograph of the neck plays a vital role to appreciate the fractures of hyoid bone and thyroid cartilage.
- Visit the scene of occurrence.

Source of Funding: None

Conflict of Interest: None

Ethical Approval: Obtained from institution ethical committee on 11/02/2018

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Body's Testimony: Injury Patterns in Sexual Assault Survivors of South-Central Mumbai

Harish M Pathak¹, Abhijith G Jayachandran^{2*}, N.S Dharmadhikari³,
Vikrant N Waghmare⁴

¹Academic dean, Prof & HOD, Department of Forensic Medicine, Seth GS Medical College & KEM Hospital Mumbai, ²Assistant Professor, Department of Forensic Medicine, Sree Gokulam Medical College & Research Foundation, Trivandrum, ³Medical Superintendent, Kalamb Sub-district Hospital, ⁴Assistant Professor, Department of Forensic Medicine, Grant Government Medical College & Sir JJ Hospital, Mumbai

How to cite this article: Pathak HM, Jayachandran AG, Dharmadhikari NS, Waghmare VN, Body's Testimony: Injury Patterns in Sexual Assault Survivors of South-Central Mumbai, 2025;25(2):10-17.

ABSTRACT

According to UNICEF, globally, an estimated 650 million girls and women, or approximately 1 in 5, have been subjected to sexual violence during their childhood¹. Both the physical healing and the legal pursuit of justice for survivors of sexual violence depend on the timely treatment and comprehensive documenting of extra-genital and genital injuries. This prospective observational study was conducted at a tertiary care facility in central-southern Mumbai from January 2016 to June 2017. A total of 160 survivors of alleged sexual assault, fulfilling inclusion criteria, were included in the study. The main aim of the study was to observe the injury patterns on survivors of alleged sexual assault presenting to the tertiary care facility, with a specific focus on extra-genital and genital trauma. In the study, we observed, majority of sexual assaults involved peno-vaginal penetration (84.37%), with genital injuries being the most common (69.38%). Hymenal tears at the 7 o'clock position was frequently observed (7.5%), often in conjunction with older tears (60.40%). Anal injuries were rare (6.25%). A significant proportion of survivors presented late for examination (25.62%).

Keywords: Sexual assault, Genital Injury, Forensic examination, Injury patterns

Introduction

According to the World Health Organization (WHO), nearly 1 in 3 women worldwide, about 35%, have experienced either extra-genital or sexual violence at some point in their lives². These numbers demonstrate the global and appalling prevalence of violence against women and girls. India reported 31,000 rapes in 2022, and the number unchanged since 2012. Mumbai ranks 5th in crime against women in metropolitan cities³. India's Health ministry,

recognizes the health sector's role in preventing and eliminating sexual assault. Laws like the Criminal Law Amendment Act and the POCSO Act of India 2013, aim to protect survivors and ensure justice.

Sexual assaults often occur without witnesses, making the survivor's account the primary source of evidence. This reality underscores the critical role of documentation and injury identification. In questionable situations proper documentation can help

Corresponding author: Abhijith G Jayachandran, Assistant Professor, Department of Forensic Medicine, College Building, Sree Gokulam Medical College & Research Foundation, Venjaramoodu, Trivandrum, pin 695607, Kerala.

E-mail: abhiget000@gmail.com

Submitted: Nov 20, 2024;

Accepted: Dec 21, 2024 **Published:** April-17-2025

overcome potential prosecutorial biases. The patterns, injuries, even subtle ones, can provide valuable insights into the nature of the assault. This study aims to detect and examine injury profiles, with a particular emphasis on genital and extra-genital injuries in cases of sexual assault.

Methodology

This prospective observational study was conducted by the department of forensic medicine at a tertiary care facility in Mumbai, over a period of one and half years, January 2016 to June 2017. All survivors of alleged sexual assault examined during the study period of one and a half years who fulfilled the inclusion criteria were included in this study, comprising a total of 160 cases.

Inclusion criteria:

- All the survivors of alleged sexual assault brought for medico-legal examination with request for examination by the police/magistrate;
- All the survivors of alleged sexual assault brought for examination directly to hospital without registering the crime.

Exclusion Criteria:

- Survivors of alleged sexual assault who refused to give consent for medico-legal examination;
- Dead bodies with alleged history of sexual assault.

A team of doctors from department of Forensic medicine and Obstetrics and gynecology were involved. Type of sexual assault, presence and distribution of extra-genital, genital and anal injuries, distribution of injuries to hymen, condition of clothes and duration of reporting were studied.

Objectives:

- To describe the types and distribution of both extra-genital and genital injuries in sexual assault cases.
- To assess the condition of the hymen, including the presence and specific positions

of injuries, in cases involving genital injury,

- To assess the patterns of clothing/changes and reporting delays among sexual assault survivors and their impact.

Data source: For a subsequent group assessment, all information gathered from various sources such as casualty records, medico-legal report of survivors, were entered into a pro-forma that was specifically created for each case.

Data analysis: The data was entered and analyzed by using MS-Excel SPSS software package. Frequency of all variables was derived to check completeness of data. Magnitude was expressed in percentages.

The study was conducted following approval from the Institutional Ethics Committee (IEC) II, of Seth GS Medical College & KEM Hospital, Mumbai, reference number: EC: 262/2015, dated: 03-March-2016. Confidentiality of subjects were strictly maintained throughout the study.

Results

Table 1: Distribution of cases by type of sexual assault

Type of act as per alleged history	No. of cases	Percentage
Peno-vaginal penetration	135	84.37
Peno-anal penetration	18	11.25
Digital- vaginal penetration	1	0.62
Peno-oral penetration	1	0.62
Molestation/fondling	5	3.125
Total	160	100

Table 2: Distribution based on presence of extra-genital*and genital injuries

Type	No. of Cases	Percentage
Extra-genital Injuries	26	16.25
Genital Injuries	111	69.375
Injuries absent	23	14.375
Total	160	100

(*extra-genital=general body excluding genitalia)

Table 3: Distribution of extra-genitalinjuries by location and type of injury

	Location	Type of Injury			
		Abrasion	Contusion	Laceration	Burns
Body	Head, Neck & Face	3	1	0	
	Upper Limb	5	1	1	
	Lower Limb	0	2	0	1
	Chest & Abdomen	2	1	0	
	Back	4	3	1	1
Total(%)		14 (8.75%)	8 (5%)	2 (1.25%)	2 (1.25%)

Table 04: Distribution of genital injuriesby location and type of injury

			Abrasion	Contusion	Tear
External Genitalia		Labia Majora	3	0	0
		Labia Minora	4	0	0
		Posterior Fourchette	2	0	0
	Hymen	2 O'CLOCK	0	0	3
		3 O'CLOCK	0	0	9
		4 O'CLOCK	0	0	5
		5O'CLOCK	0	0	5
		6O'CLOCK	0	0	10
		7O'CLOCK	0	0	12
		8O'CLOCK	0	0	4
		9O'CLOCK	0	0	7
		10O'CLOCK	0	0	1
		11O'CLOCK	0	0	2
		12O'CLOCK	0	0	1
Total (%)			9 (5.625%)		60 (37.5%)

(*No injuries found at urethral orifice, clitoris & perineum)

Table 5: Distribution of hymen condition among all the cases

Hymen findings		No. of cases	Percentage
Intact		52	34.89
Torn	Fresh tear	07	4.69
	Old tear	90	60.40

Table 6. Distribution bytype of anal injury

Type of Injury	No. of cases	Percentage
Tear	05	3.13
Fissure	04	2.5
Redness	01	0.6
Total	10	6.25

Table 7: Distribution by findings of survivor's clothing

Condition of clothes	No. of cases	Percentage
Stained by blood	1	0.62
Stained by semen	2	1.25
No stain	22	13.75
Clothing/apparels changed	135	84.38
Total	160	100

Discussion

Table 01.Type of Sexual Assault.

Peno-vaginal penetration was the predominant mode of assault in our study, based on the alleged

Table 8: Duration of reporting following assault

Duration of Reporting	No.of Cases	Percentage
0-12hrs	18	11.25
12-24hrs	25	15.62
24hours-72hrs	15	9.35
3days-7days	34	21.25
7days-1month	27	16.87
>1month	41	25.62



Fig. 1: Contusions over the upper arm

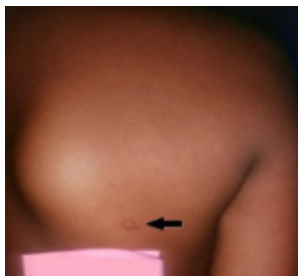


Fig. 2: Healed burns over the scapular region with puckering at the centre.

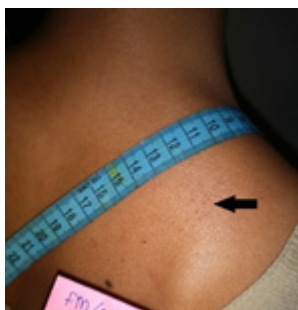


Fig. 3: Finger nail abrasions over the back.

history, accounting for 84.37% of cases. Similar findings were reported in studies from Mumbai by S. Tyagi et al⁴. (89.7%) and A.K. Jaiswani et al⁵. (58.2%), where peno-vaginal penetration was identified as the most common type of sexual assault.

Peno-vaginal penetration is a common form of sexual assault because it aligns with the assailant's intent to dominate and violate sexual boundaries in a way that often targets primary sexual organs.

Table 02. Distribution based on presence of extra-genital and genital injuries.

The table indicates that genital injuries were the most prevalent, occurring in 69.38% of cases, significantly higher than extra-genital (general body injuries other excluding genital injuries) injuries, which were observed in only 16.25% of cases. In 14.38% of cases, no injuries were reported.

These findings are consistent with those of A.K. Jaiswani et al.,⁵ who reported genital and extra-genital injuries in 30.9% and 24.9% of cases, respectively. S. Haridas et al.⁶ (Delhi) observed extra-genital injuries in only 1.91% of cases, while Sarkar et al.⁷ (Delhi) documented extra-genital injuries in 6.7% of survivors. S.K. Maring et al.⁸ (Manipur) reported genital injuries in 75.7% of cases, with extra-genital injuries noted in only 14.8% of cases, where survivors belonging to under 18 years.

A similar study from Tunisia (O. Brahim et al.⁹) reported that evidence of genital trauma was found in 87% of cases. A striking parallel can be drawn with a study conducted in Bahrain, located in West Asia (F.A.R. Alfadhel et al¹⁰.), where only one-third (30.9%) of the subjects exhibited extra-genital injuries. Comparable rates were reported in studies conducted in Egypt (30%) and Spain (30.6%).¹⁰

This suggests a strong relationship between the mode of assault and the occurrence of genital injuries. Given that peno-vaginal penetration typically involves direct contact with genital structures, it is reasonable to infer that this mode of assault significantly contributes to the high incidence of genital injuries.

Table 03. Distribution of extra-genital injuries by location and type

As discussed above extra-genital injuries were less prevalent than genital injuries. Abrasion was the most common injury type in extra-genital

examinations (8.75%), followed by contusion (5%). The least observed injuries included lacerations and burns, each at 1.25%. Abrasions were mostly seen in upper limbs followed by back.

Which is very much aligned with AK Jaiswani et al⁵. where the majority of the extra-genital injuries were abrasion (26.4%) followed by contusion (15.09%). Haridas et al.⁶ reported common body injuries seen are scratch abrasions and contusions mostly on face, neck and breast. FAR Alfadhel et al.¹⁰ and O Brahim et al.⁹ reported a slight variation, with contusions (24.6% and 70.8%, respectively) being the most common type of injury, followed by abrasions (11.4% and 51.7%, respectively).

Abrasions occur when the skin rubs or scrapes against a rough surface. During a sexual assault, defensive actions (e.g., pushing, shielding) often involve the upper limbs, explaining the high frequency of abrasions in these areas. The differences in injury patterns between your study and those of others may stem from, perpetrator behavior, or environmental factors (e.g., use of objects during the assault). For example, higher rates of contusions in FAR Alfadhel et al¹⁰. might suggest greater use of blunt-force trauma in their study population.

Table 04. Distribution of external genital Injuries by location and type

In this study, tears were the most commonly observed injury to external genitalia, seen in 60 cases (37.5%), primarily involving the hymen, followed by abrasions in 9 cases (5.625%), which were mostly present on the labia minora in 4 cases (2.5%).

Similarly in the study by AK Jaiswani et al⁵. the most frequent external genital injury was observed was laceration/tears (48%), majority at posterior fourchette; 38 (48%) cases followed by labia minora (<1%).

Brahim O et al.⁹ also reported majority of injuries especially laceration/tear at the posterior fourchette (82.5%). RR Zilkens et al.¹¹ (Australia) reported, the most frequent genital injuries in women reporting completed vaginal penetration were lacerations (13.1%), followed by abrasions

(11%). The posterior fourchette was the most common site of injury (7.4%), followed by the fossa navicularis (6.8%) and labia minora (6.1%).

The posterior fourchette is located at the junction where tension from penetration is concentrated, particularly during vaginal penetration. Probably why multiple studies, including ours, and by AK Jaiswani et al⁵. and Brahim O et al⁹., report the posterior fourchette being most common site for tears. Tears result from excessive stretching of the genital tissue during forceful penetration. This is likely to occur in non-consensual encounters where the tissue is not adequately prepared or lubricated, leading to increased friction and trauma. Abrasions are reported more frequently on areas like the labia minora due to their external location and exposure to rubbing or shearing forces during the assault. However, their overall incidence is lower compared to tears.

7 o'clock position was the most frequently observed location of hymenal tears, representing 12 cases (7.5%). This was followed by the 6 o'clock position with 10 cases (6.25%)

AK Jaiswani et al.⁵ reports most common site between 6 and 9 o'clock (35.7%) followed by 12 and 3 o'clock (28.6%) position. Almost similar observation by S Tyagi et al.⁴ who reported, maximum in 5 o'clock positions (24.39%) followed by 7 o'clock positions (19.51%). O Brahim et al⁹. reported most common position of tear between 3 and 9 o'clock, and between 5 and 7 o'clock.

The hymen is often described using a clockface, with 12 o'clock near the urethra (anterior) and 6 o'clock toward the anus (posterior). In this study, hymenal tears were most frequently observed at the 7 o'clock and 6 o'clock positions. The posterior hymen, being less elastic and poorly vascularized, may be more prone to tearing¹². Factors like the type of act, assailant's position, and assault duration further influence injury patterns, with the posterior region frequently affected. Studies by AK Jaiswani⁵, S Tyagi⁴, and O Brahim⁹ also highlight the prevalence of posterior tears.

Table 05. Distribution of cases based on the condition of hymen

In 97 (60.63%) cases hymenal tear was found. Among the tear majority were old tear 90 (60.40%) followed by 07(4.69%) of fresh tear. Haridas et al⁶. (91.44%), Sukul et al.¹³ (86.2%), S Bandyopadhyay et al¹⁴. (42%), UB Roy Choudhari et al¹⁵ (72%), all of them observed that in maximum cases, the hymen showed old tears. FAR Alfadhel et al.¹⁰ reported 45.6% of survivors had old hymen tears and only 6.6% of survivors had recent hymen tears.

The predominance of old hymenal tears in sexual assault cases, as reported in multiple studies above, can be attributed to various factors. Anatomically, the hymen's elasticity and susceptibility to tearing vary, and prior sexual activity or past trauma often results in old tears being more commonly observed during examinations. Additionally, delayed reporting by survivors is a significant factor, as fresh tears heal relatively quickly, making old injuries more likely to be documented. This pattern aligns with findings from studies like Haridas et al⁶. and FAR Alfadhel et al¹⁰., where old hymenal tears were prevalent due to delayed reporting and prior hymenal trauma.

Table 06. Anal Injuries

Anal injuries were noted only in 10 (6.25%) cases, primarily consisted of tears (3.13%) and fissures (2.5%). In one case redness was noted. AK Jaiswani et al⁵ reported 12 (2.72%) cases of anal injury, among that most common type of injury was laceration/tear (1.82%) followed by contusion (0.45%) and abrasion (0.45%).S. Tyagi et al⁴. observed anal injuries in 5.17% of cases, Alfadhel F.A. et al¹⁰. in 2.9%, and S.C. Sarkar et al⁷. in 7% of cases. Research indicates that injuries from anal penetration especially involving male survivors, are often underreported due to embarrassment or the stigma associated with anal trauma, impacting prevalence data¹³.

Table 07. Condition of clothes of the survivor at the time of examination

The majority of survivors had changed clothes by the time of examination (84.38%). Among those who retained their original clothing, only in least number cases, evidence of from clothes

are obtained, seminal stains (1.25%) and blood-stained clothes (0.62%) in present study.

The observation that the majority of survivors had changed clothes by the time of examination (84.38%) aligns with findings from other studies, such as Rahul Jain et al¹⁶. (41%), and AK Jaiswani et al⁵. (90%). Changing clothes and bathing are common behaviors post-assault due to psychological distress or the survivor's attempt to regain a sense of cleanliness and normalcy. However, these actions result in the loss of critical biological trace evidence, such as seminal stains, blood, or torn clothing, which are vital for forensic analysis and can significantly impact the trial outcome.

Table 08. Duration of reporting

The most common timeframe for reporting cases was over one month after the assault (25.62%), followed by 3-7 days (21.25%) and 7 days to 1 month (16.87%). The shortest reporting duration, 0-12 hours, accounted for only 11.25% of cases, making it the least frequent.

Similarly, a study by Rahul Jain et al¹⁶. reported that only 18% of cases were reported within 12 hours. Haridas et al⁶. found that the majority of cases (74.34%) were brought for medical examination more than 7 days after the alleged incident, while only 13.15% were examined within 24 hours. Comparable findings were observed by Sarkar et al.⁷

Delay in reporting can significantly affect the detectability of injuries in physical examination.

Delays often stem from psychological barriers like trauma, fear, and stigma, logistical challenges such as lack of access to medical or legal facilities, and distrust in the justice system.

Conclusion

Peno-vaginal penetration was linked to a high occurrence of genital injuries, with hymenal tears being the most common. Extra-genital injuries, though less prevalent, were observed on areas such as the head, neck, and upper limbs, indicating force or resistance during the assault.

Old hymenal tears were more common than fresh ones, likely due to delayed reporting.

The absence of injuries (14.38%) underscores the importance of understanding that sexual violence can occur without leaving visible extra-genital evidence. The absence of injuries and the lower prevalence of extra-genital injuries observed in this study may be attributed to delays in reporting by participants. This delay could potentially allow for the natural healing or resolution of injuries, thereby reducing the likelihood of their detection during the examination or assessment process. Additionally, the majority of survivors had changed clothes before assessment, potentially hindering the recovery of critical forensic evidence.

Recommendations

It is crucial to establish accessible, confidential, and survivor-friendly reporting mechanisms. Community education on the importance of legal and medical procedures can empower survivors to seek help. Many studies report that literature on sexual assault examinations shows disparities due to a lack of uniformity in examination protocols, injury classification, and examiner qualifications¹⁷. This should be addressed by implementing guidelines by apex health bodies. Strengthening and expanding laws defining rape and sexual assault, sensitizing and training police and judges about sexual violence, and improving the application of existing laws are crucial steps in addressing this issue.

Limitations

The study was conducted at a single tertiary care facility, limiting the generalizability of its findings. The study focused on physical injuries and did not address the psychological or social impacts of sexual assault, which are crucial for understanding the broader consequences for survivors. The absence of standardized protocols for examining and recording injuries across different examiners may have introduced inconsistencies. A significant proportion of survivors presented late for examination which may have affected the data.

Conflict of interest: Nil.

Source of Funding: Non funded.

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Multi-allelic Patterns Observed at Y-STR Loci: A Case Report

Sadhna Sahu^{1*}, Smita Taneja², Shriya Chourasia³, Apoorva Garg⁴, Pradeep K Mishra⁵

¹Senior Scientific Assistant, Biology Division, Central Forensic Science Laboratory (CFSL), Bhopal, ²⁻⁴Forensic Professional, Biology Division, CFSL Bhopal, ⁵Assistant Director, Biology Division, CFSL Bhopal

How to cite this article: Sahu S, Taneja S, Chourasia S, Garg A, Mishra PK, Multi-allelic Patterns Observed at Y-STR Loci: A Case Report, 2025;25(2):18-22.

ABSTRACT

The forensic relevance of Y-STR analysis is well understood and widely documented. In this report, the authors have described two unusual multi-allelic patterns in Y-STR profiles of individuals observed during forensic examination. The first is a case of a bi-allelic pattern at the locus DYS391 observed in a father/son duo in a paternity dispute. The second case is of a tri-allelic pattern at the locus DYS387S1a/b observed in the Y-STR profile of a suspected perpetrator in a case of sexual assault. In both cases, the individuals belong to the Himalayan state of Sikkim in Northeast India, and this is the first report of such abnormal Y-STR patterns observed in this population. Probable causes for these multi-allelic patterns and the relevance of these findings for forensic DNA analysis have been discussed.

Keywords: Y-STR analysis, Multi-allelic patterns, Y-STR duplication, Y-STR profile, Forensic examination, Forensic DNA Analysis

Introduction

Short Tandem Repeat (STR) is a form of genetic variation based on differences in the number of repeats of short DNA sequences (typically 2 to 6 nucleotides). Several repeats are possible at each locus and analysing multiple unlinked STR markers provides a high power of discrimination.¹ STR analysis has been the preferred method of forensic DNA analysis for the past three decades. In the case of autosomal STR, two alleles are present for each STR marker due to the diploid nature of inheritance. Gonosomal chromosomes follow unique inheritance patterns. For X chromosomal STR markers, two alleles are present in females and one in males. Y chromosome is passed down from father to

son and does not undergo recombination, except for the pseudoautosomal regions.² Therefore, STRs on Y chromosome or Y-STRs are ideal for tracing male lineage, comparing genetic diversity, and understanding the evolutionary history of the human population.³ In forensic examinations, Y-STRs cannot be used for individual identification, however, they can help in resolving mixed genetic profiles in complex cases of sexual assault and establish kinship. In cases of sexual assault where the male-to-female DNA quantity ratio is low, an autosomal profile of mixture of DNA can provide inconclusive results. In such cases, generating a Y-STR profile allows the identification of the paternal lineage of the male individual in the DNA mixture. In cases of disputed paternity

Corresponding author: Sadhna Sahu, Senior Scientific Assistant, Biology Division, Central Forensic Science Laboratory (CFSL), Bhopal

E-mail: sahu.sadhna@mha.gov.in,

Submitted: Oct 7, 2024

Accepted: Feb 15, 2025 **Published:** April-17-2025

where the alleged father is unavailable or unknown, a comparison of the male child's Y-STR profile with paternally related male individuals from the alleged father's family can help in establishing paternity.

Due to the Y-chromosome's uniparental presence and haplotype nature, each marker in an individual Y-STR profile appears as a single allele upon amplification with locus-specific primers. Examples of exceptions to this rule are DYF387S1a/b,⁴ DYS459,⁵ DYS527,⁶ and DYS385a/b⁷ which have double peaks in Y-STR profiles due to evolutionarily distant duplication events.⁸ Unusual STR profiles can lead to misinterpretation of results in forensic investigations. In *amelogenin* Y-allele (AMELY) null cases, male autosomal genetic profiles may be interpreted as female profile.⁹ A single male haplotype in Y-STR may be interpreted as female profile.⁹ A single male haplotype in Y-STR may be interpreted as female profile due to the presence of additional Y-alleles.^{10,11} Deletions, insertions, or duplications in STR allele scan also be misinterpreted as allele drop-outs or evidence of DNA contamination.¹²

This article reports two rare multi-allelic patterns at Y-STR loci in individuals from the Himalayan state of Sikkim in northeast India, observed during forensic examination in Central Forensic Science Laboratory, Bhopal. In case 1, a bi-allelic pattern was observed at the Y-STR locus DYS391 in two paternally related individuals. In case 2, a tri-allelic pattern was observed at the double

copy locus DYF387S1a/b in the Y-STR profile of an individual accused in a case of sexual assault. This is the first report of these unusual patterns of Y-STR peaks from the population of Sikkim.

Materials and Methods

For both cases, reference blood samples were collected by government medical officers and submitted by the investigating officer to Central Forensic Science Laboratory, Bhopal for forensic case examination. DNA was extracted using the EZ1 Advanced XL Automated DNA extraction machine (Qiagen). The extracted DNA samples were quantified using the Applied Biosystems Quantifiler™ Trio DNA Quantification Kit on the QuantStudio™ 5 Real-Time PCR machine. STR amplification was carried out with multiple STR kits using the Applied Biosystems Veriti™ thermal cyclers and amplified products were genotyped using Applied Biosystems 3500 genetic analyzer and GeneMapper™ ID-X Software v1.⁶ To exclude the possibility of contamination or allele artifacts, repeated DNA extraction of all samples was carried out using QIAamp Investigator Kit (Qiagen), amplified using the same STR kits, and analyzed along with necessary controls.

For case 1, STR amplification of extracted DNA was performed using Investigator® 26plex QS Kit (Qiagen) for 23 autosomal STR, Amelogenin, and one Y-STR locus DYS391 (Figure 1). Further, the

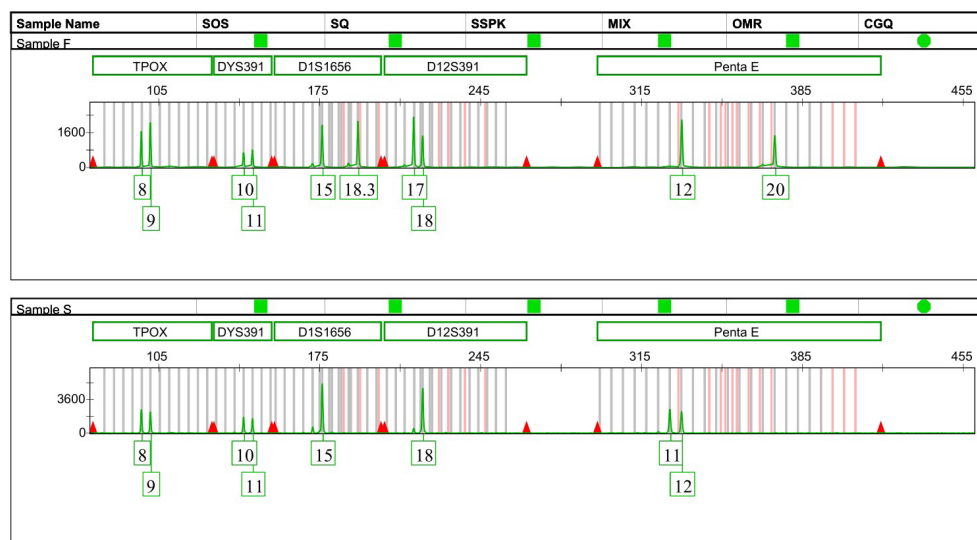


Fig. 1: Autosomal STR profile with bi-allelic peaks at the gender marker DYS391. Sample F – Alleged Father; Sample S- Son.

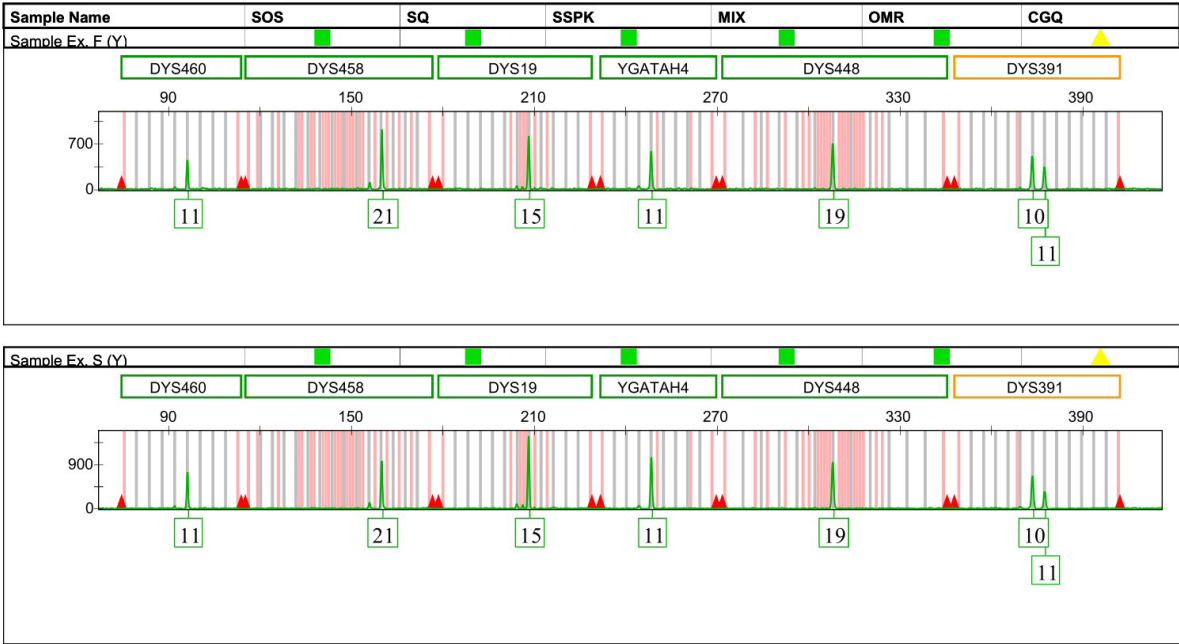


Fig 2: Y-STR profile with bi-allelic peaks at the marker DYS391. Sample Ex. F – Alleged Father; Sample Ex. S- Son.

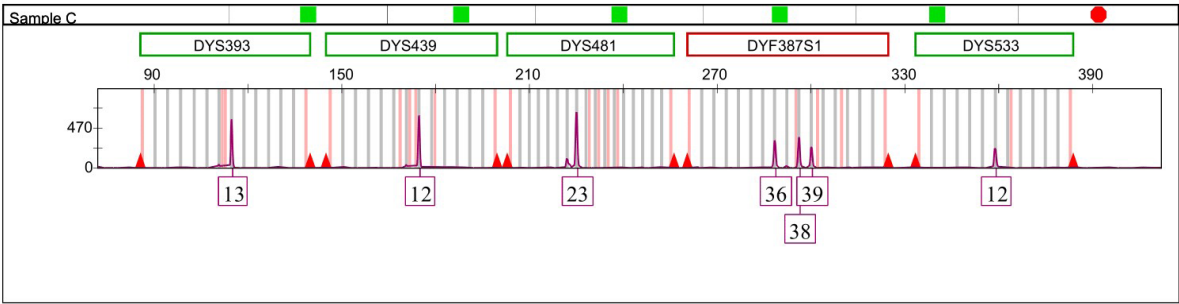


Fig. 3: Y-STR profile of Sample C with tri-allelic peaks at the marker DYF387S1.

DNA was amplified using the Applied Biosystems Yfiler™ Plus PCR Amplification Kit (Thermo Fisher Scientific) for 27 Y-STR markers. For case 2, extracted DNA was amplified using the Yfiler™ Plus PCR Amplification Kit (Thermo Fisher Scientific) for 27 Y-STR markers. All amplifications were carried out along with negative control and respective standard kit positive control DNA.

Results and Discussion

In case 1, the autosomal genetic profiles of an alleged father-son duo were generated for paternity determination. A bi-allelic pattern was observed at the Y-STR gender marker DYS391 in both genetic profiles (Fig. 1). To confirm this variant, the DNA was amplified using the Yfiler™

Plus PCR Amplification Kit. The double peaks at the DYS391 locus were consistent in both profiles (Fig. 2). The Y-STR haplotype database YHRD has no reports of multiple alleles at the locus DYS391.¹³ However, few cases of double peaks at DYS391 have been reported in the global literature¹⁴ as well as on the NIST STR Database.¹⁵ Such a bi-allelic pattern at this marker has not been reported in any Indian population.

In case 2, samples in a case of sexual assault, were amplified using the Yfiler™ Plus PCR Amplification Kit. A tri-allelic pattern was observed at the locus DYF387S1a/b in the Y-STR profile of the accused (Fig. 3). Such tri-allelic patterns at this double-copy locus have been observed in a few populations.^{16,17} Reports of

this pattern are also available on YHRD,¹⁸ and the NIST STR Database including a few reports from the states of Rajasthan and Madhya Pradesh in India.¹⁹

In both these cases, apart from the marker with additional peaks, the rest of the DNA profile was consistent with that of a single male individual, and negative controls were found to be contamination-free. Positive control profiles were consistent with expected results from respective kit control DNA. Repeated extraction of the blood samples produced genotyping results consistent with those of the first observations.

Variations in STR alleles are observed due to mutations leading to loss or gain of repeat units over generations. A possible explanation for additional peaks in STR loci is duplication of the chromosome region followed by independent mutation events altering the repeat number in one of the copy regions. The Y-chromosome contains many ampliconic repeats leading to rearrangements driven by non-allelic homologous recombination (NAHR), however, duplications due to rare non-homology-mediated processes have also been observed.²⁰ Due to NAHR between similar sequences, a high frequency of duplication has been reported in loci present in the palindromic regions of the Y-chromosome, such as the region containing DYF387S1a/b.¹⁶ Another possible explanation for additional Y-STR alleles could be the presence of a region of the Y-chromosome on a different chromosome.¹⁷ The duplicated alleles that have identical copy numbers are represented as a single peak on the electropherogram and therefore, data collected using capillary electrophoresis (CE) -based STR analysis underestimates the frequency of duplicated alleles.²¹ Further studies using advanced molecular biology techniques such as sequencing are required to understand the exact reason for abnormal STR patterns.

Conclusion

Globally, increased adoption of Massively Parallel Sequencing has improved the understanding of unusual STRs, however, in developing countries like India, conventional CE-based STR analysis is the primary method of DNA examination.

Knowledge of unusual patterns is necessary for accurately interpreting STR results, and to conclude with certainty in forensic examination reports. Abnormal Y-STR peaks as observed in these cases may not affect Kinship analysis, however, they could lead to misinterpretation of results in DNA mixtures often encountered in forensic DNA examinations. Careful analysis is required to estimate the number of contributors in a sample since additional peaks do not always imply additional contributors.

Compliance with ethical standards:

This study was performed in line with the principles of the Declaration of Helsinki. Data analysis was based on the samples referred by the investigating agencies wherein the courts of law authorized the submission of samples to the laboratory and subsequent DNA analysis.

Conflict of interest: The authors have no conflicts of interest to declare.

Funding Sources : No separate funding used for the study.

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