



A PRELIMINARY STUDY ON THE FACTORS AFFECTING THE DETECTION OF SEMEN AND SPERMATOZOA IN SEXUAL ASSAULT CASES RECEIVED FROM ANDHRA PRADESH & TELANGANA STATES AT TSFSL, HYDERABAD, INDIA.

Forensic Science

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ABSTRACT

Evidence of semen plays a crucial role in corroborating the victim's allegations. Biological samples collected confirm the presence of sperm and convict an assailant. An abnormally high concentration of acid phosphatase is a reliable indication of the presence of seminal fluid. Microscopic identification of sperm cells provides unambiguous proof that stain under scrutiny contains semen. Delayed collection involves delay in collecting the evidences/samples from victim or the objects found in crime scene due to delay in reporting in p.s., victims in some cases were found to hesitate to report/reveal the assault to police officials and the reasons included were found to be apprehension of society.

KEYWORDS

Sexual Assault, Acid Phosphatase, Swabs&smears, Spermatozoa

INTRODUCTION

Sexual assault is a broad term that encompasses many types of assault and it includes all forms of unwanted sexual occurrences from lesser/smaller crimes to more heinous crimes involving unwilling sexual contact or behavior experienced by a victim that occurs without the consent of the victim, where in physical contact between perpetrator and victim involves, consequent transfer of material, specifically semen. Assault need not be penetrative; it can include fondling and molestation. Sexual assault is considered Rape when it involves oral, Anal or vaginal penetration without the consent of the victim. (1, 2, 14)

Rape is unique(3,13) and is usually an unwitnessed crime(2) and no other violent crime is so filled with debate, so trapped in dispute and in the affairs of state of gender and sexuality, And within the province of rape, the most highly charged area of debate concerns the issue of false allegations(3,10).

Evidence of semen plays a crucial role in corroborating the victim's allegations. Biological samples collected confirm the presence of sperm and convict an assailant (2, 4, and 13). Swabs taken routinely provide evidence of semen recovered in the genital tracts even if the victim is so emotionally traumatized that facts were not disclosed during questioning. The presence of spermatozoa in the current criterion confirms sexual intercourse. This biological evidence is accepted both by the medical forensic teams, and by the judicial authorities (2).

The Spermatozoa in human semen were described by Antonie van Leeuwenhoek. For many years, and even today finding sperm is the only confirmatory method for the forensic identification of sexual assault.(4,11,15) Rape is an unlawful sexual intercourse with a female without her consent. Successful rape prosecutions are increasingly utilizing scientific evidence to investigate and prove the elements of rape. Presence of spermatozoa has traditionally served as major evidence that intercourse took place. The presence of spermatozoa can only be detected for a limited period and its presence in the vagina is not conclusive evidence that intercourse has occurred. Furthermore, the lack of spermatozoa in the vagina is far from conclusive that intercourse did not occur, especially in a society where vasectomies and azoospermic are not uncommon.(16,17,19)

The present study aims at recognizing the probable reasons for the detection of semen and spermatozoa in the cases which are received at FSL during the year 2017.

Composition Of Semen:

Seminal fluid which is a mixture of semen cells; amino acids, sugars, salts; ions& other organic and inorganic materials consists of the following 1. Spermatozoa (10%), 2. Seminal plasma (90%). Epithelial cells (<1%). The seminal plasma contains citric acid, Ascorbic acid, lactic acid, fructose, potassium choline phosphate, proteases, free

amino acids, Ergothionine, zinc calcium, lipids, spermine, Enzyme fibrinogenase, Diastase, Acid and Alkaline phosphatase, Glysidases, α & β -mannosidases, α & β -glucosidases, β -Givcouridases. All these are found in much smaller or negligible quantities in other body fluids as compared to semen. (5)

Acid Phosphatase which is classes of enzymes catalyses the hydrolysis of certain organic phosphatase. They are ubiquitous in nature. Kutscher and wolbergs in 1935 identified uniquely high levels of this enzyme in semen i.e. seminal Acid phosphatase. No correlation exist between levels of seminal Acid phosphatase and number of sperm cells present in a ejaculate.(1)

Acid Phosphatase Another substance tested for in medical examinations of rape victims is acid phosphatase. Research indicates that the normal acid phosphatase content of body fluid is twenty units per cubic centimeter. Semen, however, has an acid phosphatase content of 400-8000 units per cubic centimeter. Therefore, an abnormally high concentration of acid phosphatase is a reliable indication of the presence of seminal fluid. The total length of spermatozoa is about 50 μ it consist of head and tail. The head is flat, oval shaped: $4.6 \times 2.6 \times 1.6 \mu$ in Length Width Thickness (5). The nucleus occupies major portion of the head. Rape, Sodomy, Bestiality (Sexual intercourse by a human being with a lower animal like dogs, calves, sheep etc.), In case of false Accusation by a women, Incest (Sexual intercourse in blood relation) and Sexual Murders

MATERIALS AND METHODS:

FSL receives cases like girl missing, cheating, elopement, kidnapping, rape, gang rape, rape and murder irrespective of age and gender. These cases are registered under section 376, 375,417,366, 363, 420, 504,506, 451 IPC. Medico legally, vaginal, cervical, anal, buccal, perineum swabs, smears, pubic hair, Nail clippings, Scalp hair, Auxiliary hair and semen are collected and sent to the laboratory for chemical analysis. The clothing of the victim and suspect, along with scene of offence exhibits like bed sheets, carpets, towels, cloth pieces, pillow covers, weapons if used, condoms, semen are also forwarded to FSL for further confirmation. In material objects with dark background Seminal stains appear as translucent or opaque spots with a yellowish tint, where as in material objects with lighter background the stains are detected either by UV light or feeling the stains with bear hands which are stiff and starchy to touch. Seminal stains are first examined physically, then chemically tested by Acid phosphatase reagent and are confirmed microscopically by spermatozoa detection. Acid phosphatase test is also called as Brentamine fast blue-B /walkers test.

Chemicals:

Sodium Acetate (Merck), Fast Blue-B salt (SDFCL), Sodium chloride (SRL), sodium/potassium α -naphthyl phosphate (lobal chemie), Eosin Blue (lobal chemie), malachite green (SDFCL), Glacial acetic acid (Ranbaxy), Ethanol.

Instruments:

MEIJI Techno jenoptik Trinocular Microscope (japan make), Hot Plate (Tempo Equipments pvt. Ltd.Mumbai)

Principle of Acid Phosphatase

The seminal fluid contains the enzyme acid phosphatase and hence α -naphthyl phosphate being a preferred substrate complex with Brentamine fast blue –B the colour developer thus imparting intense purple colour within the stipulated time when semen is diluted 500 folds.(1)

Preparation Of The Sample For Staining

The microscopic slide is heat fixed, followed by acid wash and water wash. Two stains Eosin and malachite green are used for staining the microscopic slide. The anterior portion of the sperm head takes the pink stain and the posterior part purple, the neck and tail takes green colour. Since sperm head are rich in sulphur containing amino acids takes pink stain, indicating the difference in the PH of the head and tail of the spermatozoa. The stained slide is then observed under microscope for the detection of spermatozoa in 100X.(6, 9, 12)

Confirmatory Test For Semen

Microscopic identification of sperm cells provides unambiguous proof that stain under scrutiny contains semen. Staining techniques greatly assist the trained eye to easily distinguish sperm cells from extraneous material such as epithelial cells.(1)

Results and Discussion

The delay in reporting of sexual assault cases in the P.S. were due to the reasons that included fear of society, fear of honour of the family, Some cases were even reported after attaining pregnancy without marriage or illegal marriage. The cases that were forwarded contained the clothing along with the material objects collected by the gynecologist; the clothing did not give positive result as they appeared washed.

Table. I: Reasons Vs number Of Cases Pertaining To That Reason

S. No.	Reasons	No. of cases
1	Delay in reporting hence delay in collection	81
2	Immediate collection but in periods	24
3	Immediate collection but washed the private parts and took bath	05
4	Delay and in periods	03
5	One day delay	23
6	Two days delay	04
7	Immediate collection but with fungus	01
8	Dead (traced the body after certain period of time)	01

The minimum cases were those associated with fungal infection and dead victim where in the body was traced after certain time period. 2% of the cases were combination of both the delaying report and victim being in monthly cycle. 15.86% of the cases were reported after the one day of offence and around 2.75% the cases were reported after two days of the crime, where in the victim took bath and hence a negative report. 16.5%cases included the crime when the victim was in her monthly cycle. 3.44% of the victims washed private parts immediately after the offence due to ignorance. Immediate collection was considered when the sample was collected within a span of 2 to 6 hours. All the above factors affect the detection of semen and spermatozoa.

The cases which were registered under section 376, and were found to be negative were taken for study. The standard test, in which a trained eye and experienced mind uses microscope to look for sperm in vaginal, oral or anal swabs, is very sensitive. But it can fail if the swabs are taken more than two days after the rape, if the assailant has a low sperm count or does not ejaculate, or if the woman is menstruating or using spermicide. It can also fail if the rape involved oral or anal sex, because salivary and bacterial enzymes can rapidly destroy sperm. (7, 18)

Like spermatozoa, acid phosphatase can only be detected soon after intercourse (20). One study indicated that seminal acid phosphatase sometimes remained detectable up to 3 days after sexual intercourse. The test was most useful on swabs taken within 1 day and rarely useful after 2 days. Another study revealed a positive acid phosphatase reaction for periods longer than one day. Yet another experiment

concluded that acid phosphatase activity will only be useful on anal and rectal swabs when semen is abundant and will not be helpful on vaginal swabs more than two days after the offense.(1)

Time since intercourse a review of literature reveals a variation in maximum survival times of spermatozoa in body orifices. The studies have shown that the max. times the sperm heads to be detected in living persons are 7 days in vaginal cavity; 2-3 days in anus & rectum and 24 hours in the mouth. Intact spermatozoa are observed in the vaginal swabs taken between 0 to 26 hours after intercourse. Time since intercourse estimate is made on the assumption that levels of the seminal constituents drop over the time in a predictable manner. The actions of victim host coitus, and whether the victim was menstruating, and defecated or voided may all render a time since intercourse approximation.(1)There is a limit to how long spermatozoa are detectable after intercourse. Studies indicated that smears without spermatozoa were obtained from swabs as early as 28 hours, but remained rare until 2 days after intercourse.

The first barrier in detecting semen & spermatozoa is the time dimension and delay in reporting. An important aspect of the chronological aspect is the sequencing of events from assault to disclosure including information about the time lapse from the incident to a physical examination where forensic evidence is collected which is necessary for adequate evidence for the detection of semen & spermatozoa. Delayed collection involves delay in collecting the evidences/samples from victim or the objects found in crime scene due to delay in reporting in p.s., victims in some cases were found to hesitate to report/reveal the assault to police officials and the reasons included were found to be apprehension of society, fear of the honour of the family, and some cases were reported after attaining pregnancy due to live in physical relationship in before marriage and accused denies relationship/marriage, and such cases were forwarded with the clothing that were washed along with the material objects collected by the gynecologist giving no sign of positive result.(8)

The time lapse in some cases is due to impaired expressive or receptive communication of the elder that delays the recognition of the crime by providers. Providers may not recognize the physical or mental injuries as a consequence of a sexual assault. The other barriers which lead to a negative result include: lack of knowledge, i.e. washing of clothes, taking of bath after the incident. Accused age, Vasectomised, Aspermic, Azoospermic are the various factors which decide the presence of spermatozoa. Samples collected by gynecologist (swabs) are acted on by anaerobic bacteria and fungus when they are wet and it being a suitable medium for the microorganisms to grow and further degrade the biological samples leading to a negative result.(4)

CONCLUSIONS:

Teens and young adults, elderly people, people with disabilities, children especially below the age group of 6 years, individuals who have been drunken and drugged are at risk for becoming victims of sexual assault. Therefore, it is safe to conclude that the test for acid phosphatase should also be conducted within twenty-four hours after intercourse. Another tremendous advantage of the acid phosphatase test is that it does not rely on the presence of spermatozoa for positive results. In fact, the acid phosphatase activity is not in the spermatozoon itself, but rather in the seminal fluid. For detection of sperm and semen are the most reliable marker for the investigation in cases of rape, sodomy, Bestiality, sexual murder etc. detection of sperm is an important factor in confirming sexual assaults. Microscopic observation is the only source for spermatozoa identification. The intensity of detection of spermatozoa on the cotton swab from vagina and cervix decreases with Increase of period of offence. The present study conclude that time delay plays a major role in the detection of spermatozoa.

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