Journal of Forensic Research

Harel et al., J Forensic Res 2015, 6:4
DOI: 10.4172/2157-7145.1000290

Research Article Open Access

# Detection of Semen Stains in Rape Cases by a Very High Powered UV-VIS Light Source, Facilitated Conviction of Accused Person

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**Rec date:** May 15, 2015 **Acc date:** July 25, 2015 **Pub date:** July 30, 2015



**Abstract**

Detection of semen in forensic cases mostly sexual assault or abuse is critical for evidence collection. A portable forensic light source, which emits wavelength from 430 nm to 700 nm (UV to VIS) and filters were used. The very high intensity light source detected semen in darkness and daylight using wavelength between 415 nm to 490 nm. ABO blood grouping method is an oldest method used in forensic biology for grouping purpose. But now a days, DNA fingerprinting is an advanced technique used to solve crimes like gang rape. Even though in this case, simplest and oldest ABO blood grouping method helped to solve gang rape case. In present case, victim was 5 years old girl who was raped by a person. Screening the semen stains using portable forensic light source and primary testing of semen stains from different places on victim's undergarments was done. ABO blood grouping of each selected semen stains were done by absorption-elution method. Those selected stains showed blood group which matched with the accused person's blood group. Thus the accused person was convicted in this heinous crime.

**Keywords:** ABO grouping; DNA fingerprinting; Portable forensic light source (Crime lite 80S)

## Introduction

As the number of Rape cases are increasing day by day there is a need of detection of sperm and semen even if present in small quantity, no matter how old the stains are, we have to detect these stains precisely. 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. This paper provides a brief review and knowhow of the need for detection of sperms and semen and various methods applied for the detection with special emphasis on the current chemical tests with accurate procedures for performing these tests.

### Semen consists of the following

1. Spermatozoa (10%)
2. Seminal Plasma (90%)
3. Epithelial Cell (<1%)

The Spermatozoa is produced in the testis by the process of spermatogenesis. Spermatozoa contain lipid, proteins like protamine and histone etc. and enzymes like dehydrogenises and transaminases. The total length of spermatozoa is about 50 μ it consist of head and tail. The head is flat, oval shaped: 4.6 × 2.6 × 1.6 μ in Length Width Thickness. The nucleus occupies major portion of the head. The tail portion is responsible for the movement of sperm [1,2].

The seminal plasma is a mixture of secretion derived from the male accessory reproductive organs like epididymis, seminal vesicles, the prostate, vasa-deferantia, bulbourethral and urethral glands. The seminal plasma contains Citric Acid, Ascorbic Acid, Lactic Acid, Fructose, potassium Choline Phosphate, Proteases, free Amino Acids, Ergothioniene, Zinc, Calcium, Spermine, Lipids, Enzymes like Fibrinogenase, Diastase, Acid and Alkaline Phosphatase, Glysidases, a and ß Mannosidases a and ß Glucosidases, ß Givcouridases.

## Medico Legal Significance of Detection of Sperm and Semen

Rape, Sodomy (Anal intercourse), 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 [3].

### Where to look for seminal stains

- Clothes: Underwear, Bed sheet, Carpet, Towel, Pillow cover.
- Body: Perineum, thigh, Vagina and pubic hair.
- Seen of crime: On the floor or grass etc.

## Method

The semen responds to illumination by longer wavelength frequencies of UV light (~350 nm) which is invisible to the human eye. When the substance is illuminated, it absorbs the energy and exhibits luminescence at a lower energy (longer wavelength) frequency of visible blue light.

The advantage of this is that you can make invisible semen stains appear visible to the human eye. In addition, excitation and emission spectra of untreated dry semen indicate clearly that there is an alternative to using UV radiation when searching for semen stains [4,5].

### The primary features of these data are the following

- Under standard conditions of visible light (500 nm) illumination, untreated dry semen has a broad band of emission from 350-400 nm, just below the range of visibility to the naked eye.
- Long wavelength UV (350 nm) illumination of untreated dry semen produces a more narrow band of emissions centered near the blue visible region.
- Illuminating dried semen with a band of visible (450 nm) light produces strong visible fluorescence in a broad region with a maximum around 520 nm (orange).

### The primary screening of semen stains using light source

To test the exhibits of the victim we used a newly developed very high intensity light source manufactured by "Crime lite 80S". A mercury lamp inside the unit produces high intensity light of UV (320-400 nm) and visible light (400-700 nm) which detects biological stains even in day. Wavelengths can be chosen by adjustable filter positions. In combination with settings white (>400 nm), Orange (>500 nm) and red (>590 nm) goggles were used to block excitation light and to visualize the fluorescence more precisely [6].

Chemical test for acid phosphotase was done by using above four chemicals. Enzyme acid phosphotase liberates phosphate group from disodium phenyl phosphate-substrate (Figure 1). Then phenol reacts with 4-aminoantipyrine in presence of potassium ferricyanide to give dark reddish brown colored antipyrine dye which indicates presence of semen (Figure 2).

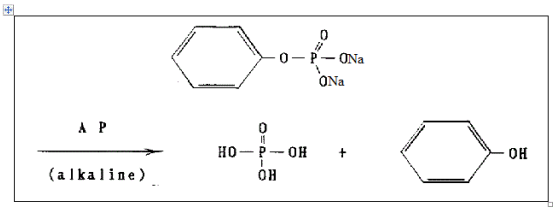


**Figure 1:** Enzyme acid phosphotase (from Semen) liberates phosphate group from disodium phenyl phosphate-substrate.

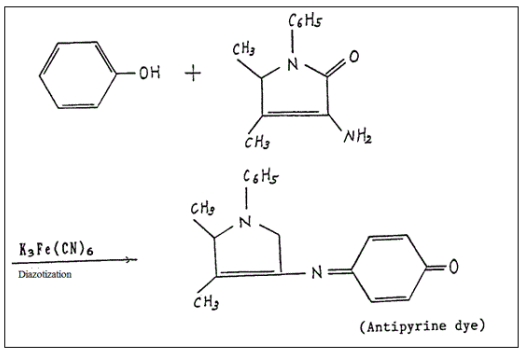


**Figure 2:** Phenol reacts with 4-aminoantipyrine in presence of potassium ferricyanide to give dark reddish brown colored Antipyrine dye.

Confirmatory test for enzyme seminal acid phosphotase was done by using agarose gel electrophoresis where extract of semen stains in Tris buffer (pH 4.9) was prepared and spotted on agarose gel. The plate was subjected to electrophoresis for 30 minutes at 20 mAmp. Staining was done by 4-methyl umbellyferryl phosphate. Precipitin bands were visualized under UV source at 254 nm wavelength. Blue bands were observed to confirm the presence of the semen [7,8].

ABO grouping by absorption elution method was done after confirmation of presence of semen where ABO grouping of selected semen stains was performed. Fibers of dried semen stains were incubated with known antisera for overnight at 10°C. On next day, fibers were thoroughly washed by cold saline to remove unbound antibodies. Absorbed antibodies were eluted by keeping fibers at 56°C temperatures. Eluted antibodies were treated with known freshly prepared cells. Thus the blood groups were obtained [9,10].

## Results of Analysis

ABO grouping of selected semen stains detected on victim's undergarments gives blood group found to be matching with control blood of accused persons.

## Conclusion

Due to its high intensity the portable forensic light source detects biological stains in darkness and daylight and can therefore not only be used in laboratory but also at the crime scene that can't be darkened. Visualization is optimum when filters are chosen that blocks the bright excitation and ambient light. Careful detection of semen stains on undergarments of gang rape victim and their grouping by simplest and oldest ABO grouping method besides using DNA finger printing solved the case. It proved that accused person was involved in rape case.

## Acknowledgement

The author expresses their sincere thanks and gratitude to Dr. M.K Malve, Director, Directorate of Forensic Science Laboratory, Mumbai and Mr. B.B. Daundkar, Joint Director, Directorate of Forensic Science Laboratory, Mumbai for valuable guidance and able support.

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