SEMESTER V

17BTU511B BASICS OF FORENSIC SCIENCE PRACTICAL 3H –1 C

Total hours/week: L:0 T:0 P:3 Marks: Internal: 40 External: 60 Total: 100

Practical

1. Documentation of crime scene by photography, sketching and field notes.
2. a. Simulation of a crime scene for training.
   b. To lift footprints from crime scene.
3. Case studies to depict different types of injuries and death.
4. Separation of nitro compounds (explosives)/ ink samples by thin layer chromatography.
5. Investigate method for developing fingerprints by Iodine crystals.
6. PCR amplification on target DNA and DNA profiling,
7. E-Mail Investigation, E-Mail Tracking, IP Tracking, E-Mail Recovery, Recovering deleted evidences, Password Cracking.

References

1. Tilstone, W.J., Hastrup, M.L., & Hald, C. (2013). *Fisher's Techniques of Crime Scene Investigation*. CRC Press.
2. Bernard J. Glick, Jack J. Pasternak , & Cheryl L. Patten. (2010). *Molecular Biotechnology-Principles and Applications of recombinant DNA* (4th ed.). Washington: ASM Press.
3. Nanda, B.B., & Tiwari, R.K. (2001). *Forensic Science in India: A Vision for the Twenty First Century*. New Delhi :Select Publishers.
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5. James, S.H., & Nordby J.J. (2005). *Forensic Science: An Introduction to Scientific and Investigative Techniques* (2nd ed.). CRC Press, Boca Raton.
6. Eckert, W.G., & Wright, R.K. (1997). *An Introduction to Forensic Sciences* (2nd ed.). CRC Press, Boca Raton (1997).
7. Saferstein R. (2004). *Criminalistics: An Introduction to Forensic Science* (8th ed.). New Jersey: Prentice Hall.

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EXP. 01: Documentation of crime scene by photography, sketching and field notes.

AIM:

To comprehend the techniques of photography, sketching and field notes in the documentation of crime scene.

PRINCIPLE:

The function of an experienced forensic expert at the scene of death is generally to assess the environment, the local circumstances and the position of the body in relation to the nearby objects, the condition of the body. In many instances crime can rapidly be excluded in favor of accidents, suicides or even natural causes. This is most useful and cost effective function as a spurious murder investigation involving expensive public facilities like police force, vehicles in investigation process. The Forensic Expert should always have appropriate equipment like camera, hand lens, papers, pencils, pens. A sketch or photo graph is sometimes useful for instant recording of scene of crime.

A. Crime Scene Photography:

At crime scenes, individuals will take pictures to document the scene. The photographs can be used later in the case to refresh the memory of the investigator or as demonstrative evidence to show the jury the relation between the victim and the evidence. The different types include:

Example of a midrange photograph.

Example of a close-up photograph.

- Overview or Establishment; shows the entire location of the incident and a broad perspective of the scene, typically the direction of travel of the suspect from the exterior of the scene through the interior.
- Midrange; shows the evidence and/or victim in relation to the scene. The evidence and/or body must be captured with a fixed object in a room.
- Close-Up: A close-up image shows the evidence and fills the frame of the camera and always should contain a scale to depict the actual or true size of the image.

B. Crime Scene Visit - A Case Study:

A case where, information by I.O, to be strange, a person hanging over the middle of a window.

- **External Findings:** Aged about 30 yrs. Height of the person - 159 cm, scalp Hair – Black, Easily Detachable wearing, blue coloured Half Shorts with some stains & Discharge. Body was in a stage Advanced putrefactive changes. Face bloated, eyes protruded & collapsed. Maggots - 1cm Size. Lividity present in Lower Limbs.
- **Injuries:**
  1. Transverse pressure abrasion (Horizontal) - Front of the Neck above the level of thyroid cartilage (6 X 2. 5 cm and 4 X 2.5 cm).
  2. Two Transverse (Horizontal) pressure abrasions - over the back of the chest (Left side of 10 X 2.5 cm, Right side is 9 X 2.5cm).
  3. Pressure Abrasion over the inner side of the left wrist of 2 X 2.5 cm.
- **Internal Findings:** Skull intact & Thyroid cartilages are intact, Ribs & Chest cage -No Fracture, Lungs - Softened became Black mass, Heart – Flabby, Intestines - Distended due to putrefactive gases, Liver - Softened became black mass. Pancreas, Genital Organs, spleen, Kidneys – Softened

Observation Findings in Crime:

1. As the crime scene helped the forensic personal for coming to a conclusion in the cause of death.
2. He is a scrap lifter on the roads.
3. He tried to lift the scrap present inside the room where the waste material is placed in a room door locked from outside.
4. He tried for safe landing through window, his body, lower limbs easily passed into the room and feet failed to touch the hard ground, where the floor inside is lower than outside.
5. Injuries correspond with the patterned pressure abrasions of the window rods.
6. Clothes, bag, foot wear of the person present outside the window in situ.
7. The expert came to a conclusion as An Accidental Hanging.

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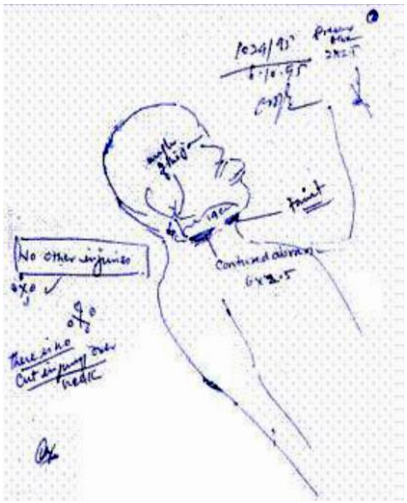
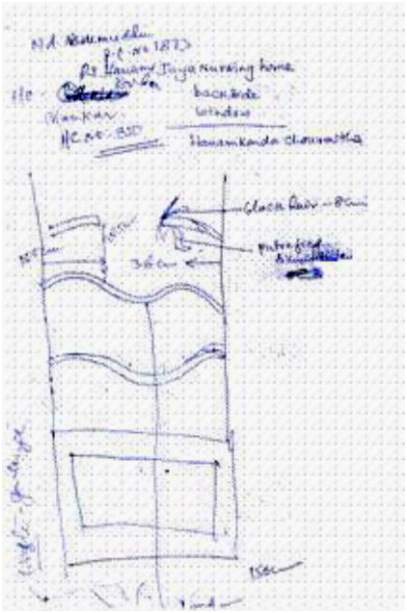
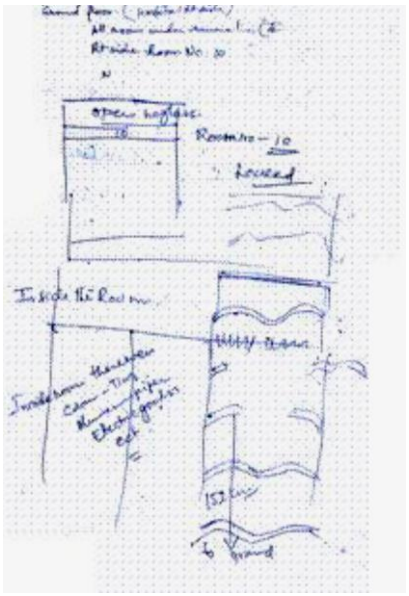
View From outside room

View From inside room







Opinion:

- Approximate time of death - about 3 to 4 days prior to the post mortem.
- The cause of death – **ACCIDENTAL HANGING.**

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Exp. 02: a. Simulation of a crime scene for training. b. To lift footprints from crime scene.

AIM:

A. To simulate a crime scene for training.
B. To lift footprints from crime scene.

A. To simulate a crime scene for training.

PRINCIPLE:

As with any discipline, the opportunity to practice, learn and apply such learning to a real-life scenario instills confidence within oneself. It allows mistakes to be made, actions to be evaluated and tasks to be set. In the sometimes high-pressure environment of a crime scene search involving complex physical and digital evidence, one incorrect action can be detrimental to any future judicial proceedings.

The field of digital forensic investigation works by providing considerations for crime scene investigation training that includes digital evidence; providing a 3D virtual environment in which law enforcement can practice learned crime scene search and seizure procedure; and evaluating such systems with law enforcement officers during classroom training.

3D Technology and Crime Scene Simulation: There are multiple examples of 3D technology been employed by law enforcement, military and educational institutes. Crime Scene Virtual Tour (CSVT) allows the user to reconstruct a 3D crime scene using photos, provides a 3D measurement feature and thereafter walks through the scene. By uploading photos of the crime scene the user is able to zoom, pan, tilt and rotate the scene and relive the crime scene. Unlike the Virtual Crime Scene Simulator developed as part of this project, however, it does not offer the ability to interact with devices found at the scene. Another solution is offered by AI2-3D, a Canadian company, specialising in the reconstruction of crime scenes using 3D forensic visualization for court presentations. Moreover in comparison with physical crime scene simulations, virtual environments have a number of distinct advantages:

- Virtual crime scenes are less expensive and faster to setup;
- Virtual crime scenes could be big, complex, and/or highly unusual;
- Virtual crime scenes can be used to perform joint training sessions where team members are geographically far apart;
- Virtual crime scene could allow straightforward simulation of live triage and crime scene processing using virtual machines.

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PROCEDURE:

1. Observe scene do not be hasty to handle it.
2. Photographer, Sketch preparer, Evidence recovery personal should be there.
3. Draw a simple sketch or Diagram of the Position of the body with location of blood stains, which gives much information. Measure the distances between body and the surrounding article or objects. Forms part of the experts original notes; he can refresh his memory after seeing original records.
4. Protect the scene.
5. Photographs in all angles, close, and near pictures should be taken by a professional photographer or by forensic expert itself.
6. Conduct final survey.
7. Release crime scene document. **(Sample format given below)**

Agency Name:

CRIME SCENE FIRST CONTACT:

Contacted by: _____ at: ☐ Home ☐ Work Date:_____ Time:_____

Location of Scene: County:_____

Address:_____

_____

_____

Crime Type: _____ # of Victims_____ ☐ Outdoor ☐ Indoor Body at Scene ☐ Yes ☐ No

Time of Incident: _____ ☐ am ☐ pm Special Precautions: _____

_____

Name of Victim(s)	DOB or Age	Location	Dead		Apparent Cause of Death	# of Wounds	Location Of Wounds
			Yes	No			

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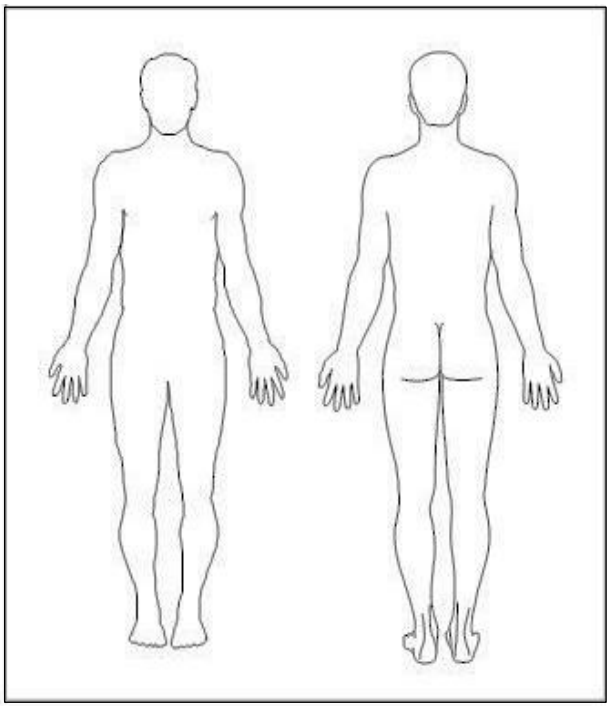
Marks and Wounds:



Name of Suspect(s)	DOB or Age	In Custody		Relationship to Victim(s)
		Yes	No	

Description of Crime:

_____
_____
_____
_____
_____
_____
_____
_____
_____
_____

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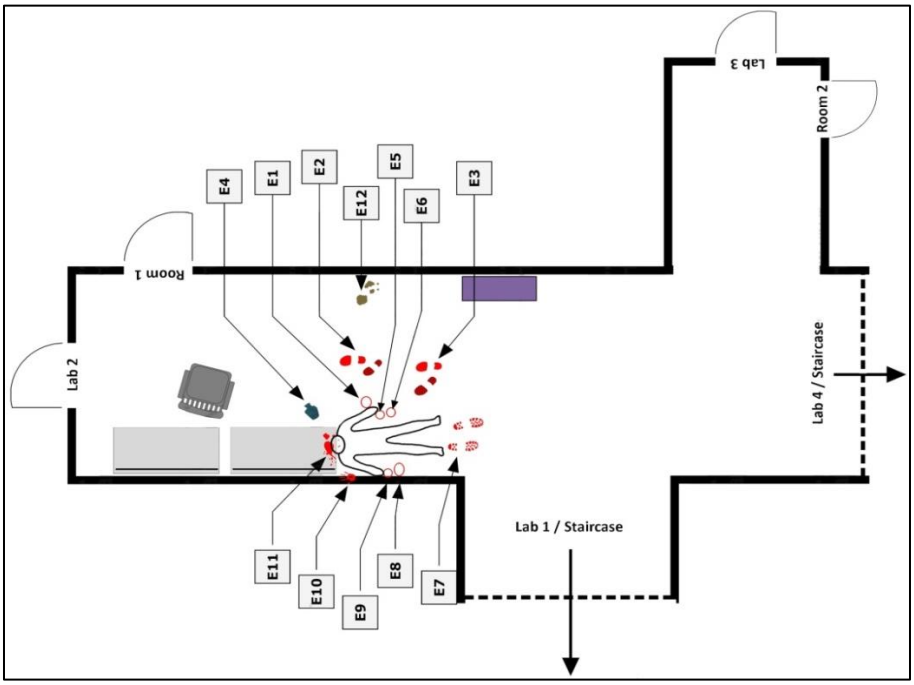
CRIME SCENE PHOTOGRAPHY	
1. (Image)	2. (Image)
3. (Image)	4. (Image)
5. (Image)	6. (Image)

CRIME SCENE EVIDENCE LOG:

LAB NUMBER _____ AGENCY CASE NUMBER _____

MARKER #	ITEM #	Description or Location Recovered	DATE	TIME	Collected By

CRIME SCENE RECONSTRUCTION:



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B. To lift footprints from crime scene.

PRINCIPLE:

A line of evidence that can establish a time frame for the presence of an individual is the interpretation of footwear impression evidence (Hamm, 1989; Bodziak, 1995; DiMaggio, 1995; Eckert, 1997). The gait of each individual is different and the unique features of each person's gait sculpt their footwear in individual and repeatable patterns.

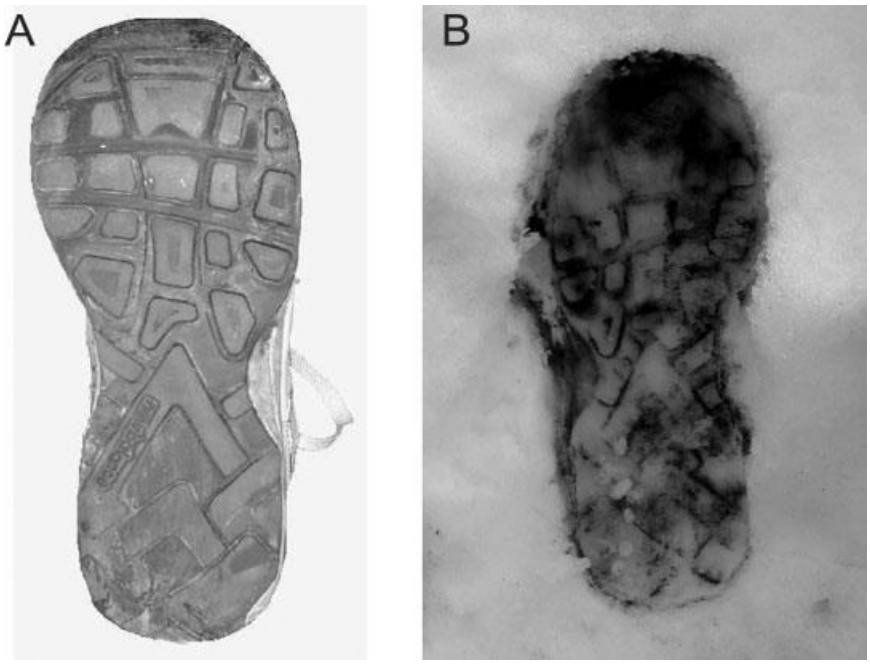

Figure.: *Sole of footwear (**A**) that made the print in snow shown in **B**.*

Footwear impressions reflect the real activities of individuals and can reveal patterns of activity in the order in which they occurred despite lack of other documentation of the crime scene. Impression evidence can be of two kinds;

The first is class, which refers to the general type of a footwear impression, including type of tread on the bottom of a piece of footwear. This is usually specific to a brand of manufacturer. Other kinds of information include the movement pattern of a person at a walk, trot, or run. Sizing of footwear also provides class information.

The second kind of information is more specific and is often possible to discern in addition to class characteristics. Examples of specific identifiers for footgear would include unique wear patterns, cuts or defects in sole impressions, and the contents of scrapings from treads or other parts. The number of individuals present at a crime scene is often difficult to determine, unless separate footwear impressions allow their number to be counted. The direction in which an

individual was moving, the speed at which they were moving, and whether they were carrying anything heavy, evidence of scuffs or dragging of feet can give unique habitual or temporary patterns of the gait at that time.

PROCEDURE:

1. Representatives (subjects) from each group step into plain water and make an impression on the brown wrapping paper and any other substrate they intend to test in this activity. Record the number of prints in a copy of the data sheet.
2. Students mix paint in the trays, starting with a mixture viscous enough to coat the soles of the footwear being tested. Subjects then step into the pans of paint and make prints repeatedly on the same substrate until no visible impression results. Record the number of prints in a copy of the data sheet.
3. Subjects make prints on substrates of different textures with the paint from the first mixture. Students then compare these impressions with those made on the brown wrapping paper.

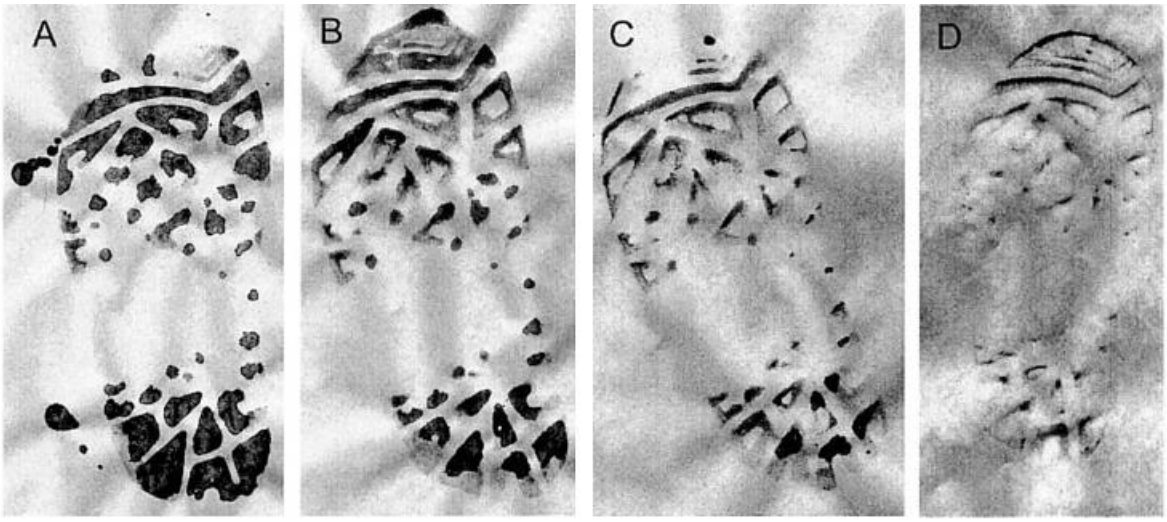

Figure: *A series of prints made by a left running shoe dipped into a moderately viscous paint mixture. The former include the tread pattern of small squares and the latter the extensively worn outer side of the heel.*

4. Subjects make footwear impressions starting with at least one additional dilution of the initial paint mixture and again determine how many prints they can make prior to them becoming invisible. Record the number of prints in a copy of the data sheet.
5. Students then perform a detailed analysis of the footwear impressions. By measuring the footwear impressions, students will be able to determine if there is variation among the prints even though they were made by the same person wearing the same item of footgear. Record the measurements in a copy of the data sheet.

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DATA SHEET FOR RECORDING FOOTWEAR IMPRESSION INFORMATION.

Experiment title:_____ Date: _____ Time: _____

Observer'snames:_____

Weather conditions: _____

Temperature: _____ Wind strength and direction: _____

Relative humidity: _____ Most recent precipitation: _____

Substrate: _____ Number of tracks identified: _____

Impression measurements: Length _____ Width (widest, generally ball of foot) _____

Position in track series _____ Direction of track way: _____

Class characters: _____

_____

_____

Individual characters: _____

_____

_____

Observations of track interactions at scene: _____

_____

_____

Conclusions: _____

_____

_____

RESULT:

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EXP. 03: Case studies to depict different types of injuries and death.

AIM:

To analyse case studies to depict different types of injuries and death.

PRINCIPLE:

An injury is define as any harm, whatever illegally caused to any person in body, mind, reputation or property as per Indian Panel Code (Sec. 44). Unintentional injuries consist of that subset of injuries for which there is no evidence of predetermined intent. Trauma is explained as an injury to the body caused by physical, mechanical or chemical factors, which may result in wounds or possible complications. The medical purposes, violence refers to either behaviour that result in injury or to the injury itself, resulting in both psychological and physical trauma.

Risk Factors for Road Traffic Injuries: The increasing volume of traffic is one of the main factors contributing to the increase in RTIs in LMICs (Low - and Middle-Income Countries). Several case-control studies in HICs (high-income Countries) have confirmed the role of alcohol in the increasing risk of road crashes; including fatigue, use of hand-held mobile telephones, and inadequate visibility of vulnerable road users (Peden and others 2004).

Risk Factors for Poisonings: Risk factors for fall related injuries in older people are generally considered in terms of risk factors for falling, risk factors associated with the severity of the impact following the fall, and risks factors associated with low levels of bone mineral density.

Risk Factors for Burn-Related Injuries: Country-specific surveys conducted in medical facilities suggest that scalds from hot water may be equally important or more important causes of burn-related injuries. However, in some countries, including China and particularly India, fire related injuries clearly outweigh scald-related injuries (Ahuja and Bhattacharya 2002; Jie and Ren 1992).

Risk Factors for Drowning: Most drowning incidents in LMICs are not associated with recreation or leisure, as is commonly the case in HICs, but instead are associated with everyday activities near bodies of water, including rivers, wells, and buckets. A number of studies find that most adult drowning incidents appear to be associated with positive blood alcohol tests (Celis 1997; Hyder and others 2003; Kobusingye, Guwatudde, and Lett 2001).

OBSERVATION AND INTERPRETATION:

The physical character of the injuries/wounds and Deaths where explored:

Homicide: Someone else caused the victim's death, whether by intention (robber shoots convenience store clerk) or by criminal negligence.

Suicide: The victim causing his/her own death on purpose.

Accidental: In this manner of death, the individual falls victim to a hostile environment. Some degree of human negligence may be involved in accidental deaths.

Natural causes: Here, the victim dies in the absence of an environment reasonably considered hostile to human life.

Different Stages of Death:

A. Rigor mortis: Rigor mortis passes as muscle decomposition begins and is usually gone in 36 hours. It can also be mechanically "broken" by stretching the rigid muscles by force.
B. Livor mortis, or hypostasis, a purplish discoloration of the body and organ surfaces, results when blood settles to dependent parts of the body (seen 1.5 hrs and 2hrs after death).
C. Desiccation occurs most prominently on the mucous membranes, which during life are kept moist (by blinking, lip licking, etc). The membranes may look "burned," and the conjunctiva may actually be black ("tache noire").
D. Putrefaction is the sequence of physicochemical events that begins with death and ends with dissolution of the nondurable parts of the body. It begins with a greenish discoloration of the skin and mucous membranes. The epidermis becomes detached; the soft tissues may also puff up and appear swollen, also as a result of gas release. Finally, autolysis and bacterial lysis hydrolyze proteins and fats, to produce frank liquefaction of the soft tissues.
E. Alternatives to putrefaction include mummification, in which the body dries out faster than decomposition takes place, and adiopocere formation.

Types of Injuries and Wounds:

A. Blunt Force Injury: On the body due to blunt forces or instruments, these injures are on skin and scratches, grazing, bruising are observed. These injuries are sub-categories as;

1. *Abrasions:* In this type of injuries the skin in which the outer layer of the skin is scraped off.
2. *Contusions/Bruises:* This type of injuries occurs when blood vessels in the skin or internal organ are ruptured.
3. *Lacerations:* Lacerations are tears or splits of skin, mucous membranes, muscle or Sharp Force Blunt Force Mech.

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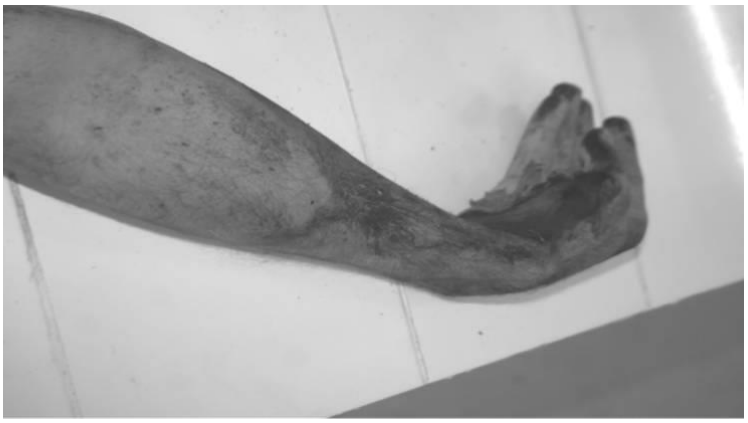

Fig.: *Abrasions*

B. Sharp Force Injury: these are caused by cutting or stabbing the skin with sharp instruments/weapons:

1. Incised/cuts Injury: This type of wound is a superficial injury, generally made of razor blade, axe and swords.
2. Stab/Penetrating/Puncture Injury: This type of injury is produced from the penetration of pointed / sharp instruments/ weapons, generally knives, broken glass bottles and tools.

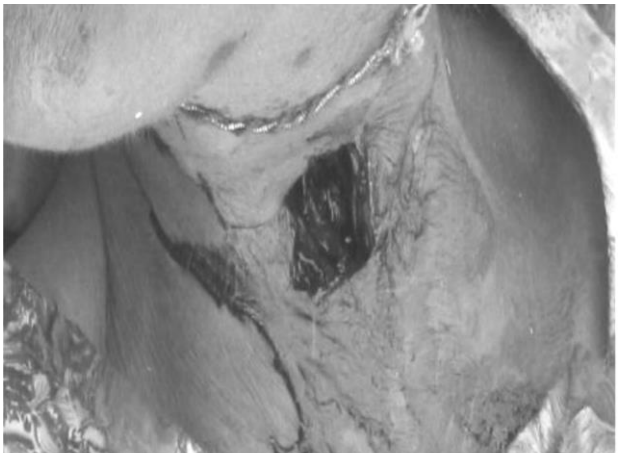

Fig.: *Single stab/penetrating injury.*

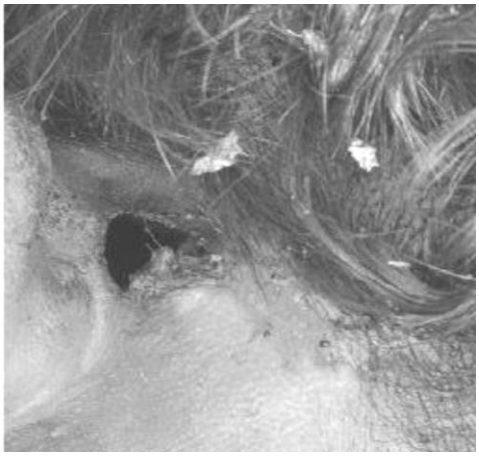

Fig.: Bullet Wound

C. Firearms Injury: The injuries produced by fire arms vary depending on the projectile, the muzzle velocity, distance, angle of firing and part of the body involved.

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EXP. 04: Separation of nitro compounds (explosives)/ ink samples by thin layer chromatography.

AIM:

To separate nitro-compounds (explosives) or ink samples by thin layer chromatography.

PRINCIPLE:

1,3,5-trinitroperhydro-1,3,5-triazine (RDX), octahydro-1,3,5,7-tetranitro-1,3,5,7-tetrazocine (HMX), and pentaerythritol tetranitrate (PETN), the major components in plastic explosives.
It is observed that explosive filling in shells and ammunitions etc. deteriorate on long storage under adverse climatic condition. These deteriorated products usually catalyse further decomposition, which effect the stability and performance of explosives. Therefore it is very essential to analyse the explosive filling from time to time in order to have a check on its deterioration under adverse condition of temperature and humidity. In most of the field areas, normal laboratory facilities do not exist and thus some simple and convenient method is required to check the stability of explosives. Considering the safety aspect such analyses are essential to minimise the explosive hazard. Although some work on identification of explosive mixture have been reported'-6, yet many explosives and their mixtures require further study. The present work aims to 6nd out new developing systems which could provide an efficient separation of components of explosive mixtures.

MATERIAL AND SAMPLES:

(a) Samples of various explosive mixtures:
p-mono nitrotoluene (MNT); 2,4, dinitrotoluene (DNT); 2,4,6 trinitrotoluene (TNT); 1,3 dinitrobenzene (DNB), 1,3,5 trinitrobenzene (TNB), trinitro phenylmethyl-N-nitramine (CE);
2,4,6 trinitrophenol (PA); 2,2', 4,4', 6,6', hexanitrostilbene (HNS); Hexa hydro-1,3,5 trinitro-s-triazine (RDX), and Octa hydro-1,3,5,7 tetranitro-s-tetrazine (HMX).

(b) Developing solvents:
Petroleum ether, Benzene, Cyclohexane, Dichloromethane, Chloroform, Ether, Ethyl acetate, and Acetone.

(c) Visulizers: Ethylene diamine, Iodine chamber and Greiss reagent.

(d) Ahorbents: Silica gel G, Oxalic acid and Zinc powder.

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PROCEDURE:

(a) Preparation of layers:

Glass plates of 5 x 20 cms were well cleaned with chromic acid, washed with water several times and dried. 25 g silica gel G was mixed with 5O ml of distilled water by shaking. This slurry was then spreaded over plates with the help of spreader within 3 minutes keeping layer thickness 0.25 mm. The plates were left for overnight for air drying. On the next day, the plates were kept in oven for 1.5 hrs at 110°C for activation. The plates were stored in a closed chamber to avoid any damage to layers due to exposure to dust or humid atmosphere.

(b) Preparation of samples:

(i) Different combinations of explosive mixture were formed, which were dissolved in non-polar and low boiling solvent as far as possible.
(ii) Pure compounds were also dissolved in the same solvent for reference sake.

(C) Spotting:

With the help of Hamilton micro syringe, fine spot of sample and pure compounds were applied to the starting line on the silica gel layer at a height of 1.5 cm from the base.

(D) Development:

The plates were air dried and put into the tank filled with developing solvent upto the 0.5 cm to 1 cm height. Prior to putting the plates, the atmosphere in the tank was saturated with the developing solvent by wrapping it from inside with filter paper sheet. The tank was closed, with lid and developing solvent was allowed to rise in the plates upto 10 cms height from-the starting line.

(E) Detection:

After the development, the plates were removed and dried to- make them free from developing solvent. Visulising agent was then sprayed over them which produce the coloured spots.

(F) Identification:

The coloured spots were compared with the reference coloured spots which helped to identify the component of mixture. Distance of coloured spots and distance of solvent from starting point were measured and Rf values for each component in the mixture were found out (Table 1).

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Table: *hRf* values of the components of explosive mixtures.

Mixture No.	Components of mixture	*hRf* values of components	Colour of spots	Developing solvent in ratios (viv)	Visualizer
01.	DNT TNT	53 45	Blue Brown	Pet ether/ dichloromethane **(40:60)**	Ethylene diamine
02.	CE TNT	21 46	Orange Brown	Pet ether/ dichloromethane **(40:60)**	do
03.	CE TNT	34 53	Orange Brown	Benzene pure	do
04.	CE TNT DNT	20 45 54	Orang Brown Blue	Pet ether/ dichloromethane **(40:60)**	do
05.	DNB TNT	52 46	Violet Brown	do	do
06.	PA TNT	30 52	Yellow Brown	do	do
07.	PA TNT	27 49	Yellow Brown	Pet ether/ acetone**(70:30)**	do
08.	PA TNT	39 65	Yellow Brown	Pet ether/ether **(60:40)**	do
09.	PA DNB	33 63	Yellow Violet	Pet ether/ dichloromethane **(40:60)**	do
10.	DNB TNB	52 34	Violet Red	Pet ether/ dichloromethane **(40:60)**	Ethylene diamine

RESULT:

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EXP.05: Investigate method for developing fingerprints by Iodine crystals.

AIM:

To investigate methods for developing fingerprints by Iodine crystals.

PRINCIPLE:

Latent fingermark detection and visualization is an important aspect of scientific investigation of scene of occurrence. Powder (1) dusting, (2) ninhydrin, (3) iodine fuming and (4) silver nitrate soaking have been the most commonly used methods of latent mark development. Iodine fuming is one of the oldest known techniques for the development of latent fingermarks. When iodine crystals are warmed, they produce a violet iodine vapor by sublimation. The iodine fumes get adsorbed on the fingerprint residues (sebaceous material) present on the latent finger mark deposit to give yellowish brown latent prints. The technique is very simple, rapid, efficient as well as economical and can be applied to a wide range of porous and nonporous surfaces (such as paper, wood, plastic, and glass). Latent fingermarks developed with iodine vapors pose a problem in recording photographically as they disappear within a very short time due to sublimation again. Therefore, fingermarks may require refuming with iodine to visualize or immediate post treatment to make them stable for a considerable time period. To prolong the visible life of iodine fumed latent prints involved post-treatment of iodine-fumed prints with starch, steam or water, 7,8-benzoflavone fixation reagent. A new method has been suggested in which brucine, an alkaloid, is used for fixing the iodine developed latent fingermarks on porous as well as non-porous surfaces.

MATERIALS:

1. *Porous and non-porous substrates:* Porous substrates included variety of commonly used papers(ordinary paper, bond paper, paper marker and non-porous surfaces (polythene high density and -low density), plastic sheets, aluminum foil, glass).
2. *Reagent preparation: the reagent was prepared in two ways as following:*

   A. 1% aqueous solution of the brucine was prepared by adding 0.250 g of in 25 ml of the distilled water. Brucine was dissolved thoroughly with the help of magnetic stirrer for about 15 min. pH of the reagent was measured with the help of digital pH meter (Eutech, U.S.A.).

   B. 1% solution was prepared by adding 0.250 g of brucine in 25 ml of distilled water. 0.25 g of potassium persulfate and 0.25 g of sodium chloride were added to the brucine solution. The

prepared solution was heated for about 5 min. There was a change in the color of the solution from milky white to red. pH of this solution was also measured.

PROCEDURE:

1. Latent fingermarks of the same individuals were collected on the substrates under the ambient environmental conditions.
2. Two sets of latent fingermarks were developed by Iodine fumes in a fuming chamber and treated separately with each of the reagent A and B.
3. The iodine developed prints were dipped in the brucine reagent A for about 20 – 40 s.
4. The reagent B was kept on the hot plate at 90 8C until it started evaporating. The latent fingermarks taken on the various substrates mentioned above and developed with iodine fumes were treated with vapors of the brucine. This process was named as double fuming method as the prints were fumed first with iodine followed by the vapors from reagent B.
5. Both porous as well as non-porous surfaces were treated with the reagents of different pH to see the possible effect of the same on fixing process.
6. Subsequent developing of the prints with ninhydrin was also examined.

Fig: *High density polythene with dipping method (pH 2–4).*

Fig: *Paper marker with double fuming.*

Fig: *Inked fingerprint.*

RESULT:

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COURSE NAME : Basics Of Forensic Science
BATCH : 2017 – 2020
COURSE CODE : 17BTU511B

EXP. 06: PCR amplification on target DNA and DNA profiling.

AIM:

To study the methods of PCR amplification on target DNA and DNA profiling.

PRINCIPLE:

Forensic DNA (deoxyribonucleic acid) analysis or DNA profiling has played a major role in the criminal justice system. DNA is the chemical code that is found in every cell of an individual's body. Although approximately 99.9 percent of human DNA sequences are the same in every person, forensic scientists are only interested in the 0.1 percent of the DNA that is unique to each individual. As a matter of fact, the likelihood of two unrelated individuals having the exact same DNA profile is ~ 10-15, or about 1 in 594 trillion individuals. Typically, the following steps are performed during forensic DNA analysis:

1. ***Sample preparation:*** crime-scene evidence is collected, stored, and transported to an accredited DNA laboratory;
2. ***DNA extraction:*** DNA is isolated from the unknown crime-scene evidence (and/or any bodily fluids from the suspect);
3. ***DNA amplification:*** certain regions of DNA are replicated exponentially in order to generate detectable amounts of DNA samples for subsequent analysis;
4. ***DNA quantitation:*** DNA fragments of different sizes are separated and detected spectrophotometrically; and
5. ***DNA profile matching:*** the profile obtained from the crime-scene evidence is either entered into a DNA database for comparison to locate a possible person of interest, or is compared directly with that from the suspect to determine whether the suspect contributed the DNA at the crime scene.

PROCEDURE:

A. Sample preparation:

1. One approach for determining if the biological origin of an unknown sample could be blood, saliva, semen, vaginal fluid, feces or urine is the use of mass spectrometry.
2. Before the sample collection process is started, it is valuable to know if even the DNA is intact.
3. Trypsin can be used to digest the sample to obtain the peptides that are present and then the peptides be injected into a mass spectrometer.
4. Biomarkers can be used to identify what type of sample is being analyzed.
5. In particular, FTIR can be used as indirect screening for DNA integrity when dealing with bone samples.

6. One approach is to use sterile water (or Triton X-100 and sodium dodecyl sulfate, SDS) which is used to moisten the tip of the swab which is then wiped across the specimen area, followed by a dry sterile swab that collects the water and cells.
7. In the process of storage DNA can be dehydrated via spray drying, spray freeze drying, air drying or lyophilization. Additives, such as trehalose (a disaccharide), can also be added to a dry DNA sample.

B. Polymerase chain reaction (PCR) amplification:

1. In forensics, repetitive DNA regions, which are located outside the coding regions of DNA, are used to further analyze DNA. These regions are different for each individual.
2. Developed by Kary Mullis in 1983, PCR continues to be a valuable tool in forensic DNA analysis.
3. PCR is able to replicate specific nucleotide sequences from low levels of DNA or degraded DNA.
4. The primers in PCR are specific to human DNA and results are not affected by bacterial DNA if it is present.
5. The DNA sequence is amplified after it is denatured and the single strands are separated.
6. Amplification involves the addition of DNA primers, nucleotides, and DNA polymerases, which are then taken through a series of temperature changes. The amplification process is able to amplify just a sequence-specific region or a whole genome.
7. Products of amplification, or amplicons, are then separated using electrophoresis.
8. The detection of DNA is often further evaluated through the use of fluorescence, which uses fluorescent dyes that attach to PCR primers in the amplicons.
9. Reagents used in the PCR process often consist of dimethyl sulfoxide (DMSO), glycerol, formamide, single stranded DNA binding proteins and betaine.
10. The development of real-time PCR (also known as quantitative PCR or qPCR) human DNA quantification kits has contributed to the popular use of real-time PCR in forensic genetics.
11. One of the positive features of a qPCR quantification assay is that it is able to establish the presence and quantity of male DNA in a mixed sample, usually from sexual assault cases.
12. Knowing the quality and quantity of male DNA can determine which STR amplification kit to use in further analysis of the DNA, saving time and decreasing costs.
13. A Real-Time degenerate oligonucleotide primed PCR (DOP-PCR) was designed, which can amplify the whole genome regardless of DNA size.
14. It is also independent of DNA sequence and can be used for many different species, giving it a universal property.
15. The primers of DOP-PCR are placed at the 3’ end, randomly in the middle, and at the 5’ end.
16. This method has been successfully demonstrated in the determination of the human placental DNA ranging from 80fg to 8ng.

C. DNA Profiling:

I. Short tandem repeat (STR) analysis –

STR loci are considered polymorphic (repeating 2-7 base pairs) due to being unique to each individual. In particular, 5-10 alleles of particular STRs are often the focus of forensic profiling. The amplification of STR, via PCR, starts with targeting loci by sequence-specific primers. The STR markers used exhibit the highest variability among individuals and are measured by the lengths of the different alleles. PCR of STRs also allows for multiplexing, which enables the analysis of several different loci at the same time. STR loci can even be obtained from maggots removed from a corpse. STR detection involves the use of fluorescence with a gel or capillary electrophoresis (CE) and ABI gel-based DNA sequences. STRs are most informative with samples that involve well-preserved soft tissue and bone. In ABO blood group recognition the ABO amplicons can be used with already existing STR kits. More specifically PCR with sequence-specific primers (PCR-SSP) has been paired with 15 STR genotypes to reduce time and costs in identifying DNA in forensic investigations.

In fact conventional STR kits have been found to be ineffective for the analysis of highly degraded or low copy number DNA samples (degraded ancient DNA/aDNA), and also Inconsistency of number of STR markers in different DNA databases.

II. Single nucleotide polymorphism (SNP) analysis –

SNPs offer an advantage over STRs due the fact that heavily degraded DNA fragments can be analyzed with SNPs. The SNPs are *base substitutions, insertions or deletions* and occur only at one position of a genome, and have a low mutation rate making them more stable as genetic markers.

To develop a way for an individual identification, a universal panel of 92 SNPs was created. Of the 86 SNPs that have no significant pairwise linkage disequilibrium, 45 can be used to determine individual identification. ABO genotyping used with SNP can be used to determine the ABH antigen expression that can be found on the surface of red blood cells. ABO genotyping has been used as a rapid screening tool before STR profiling.

The amelogenin gene has been most widely studied in humans, where it is a single copy gene, located on the X and Y chromosomes at Xp22.1-Xp22.3 and Yp 11.2. Variations of the SNP detection have included the use of four ABO loci and an amelogenin gender marker, so that individual identification and paternity testing can be done simultaneously. SNPs are preferred due to their ability to resist common degradation processes. However, it can also be limited by sensitivity and the presence of inhibitors and unsuitable for DNA mixtures.

III. Analysis of degraded or low-template (LT) DNA –

LT or degraded DNA has been successfully amplified for STR genetic profiling using whole genome amplification (WGA). In particular, WGA can be used to amplify highly degraded or LT DNA so that it can be further analyzed in the PCR method. Among the variations of WGA, there are degenerate oligonucleotide-primed PCR (DOP-PCR), primer extension pre-amplification (PEP), multiple displacement amplification (MDA), blunt-end ligation-mediated WGA (BL-WGA), rolling circle amplification (RCA), and restriction and circularization-aided rolling circle amplification (RCA-RCA). PEP is found to work well with highly-degraded DNA or low copy number DNA. It can amplify less than 200 picogram of DNA found in a sample.

Ease of contamination and amplification of contaminants; and there can be chances of mixed profiles being produced and wrongful accusations.

IV. Mitochondrial DNA (mtDNA) analysis –

Aside from the advances for LT DNA that SNPs and WGA have made, alternative sources and methods continue to be looked into including mtDNA and singe cell analysis. The hypervariable (HV) regions of mtDNA are used for analysis due to their polymorphic characteristics and are valuable sources for analysis of degraded DNA including bone samples.

In order to analyze more than just fragments of mtDNA, modified multiplex PCR systems can be used to produce small overlapping amplicons that can be used to determine the sequence of mtDNA. The analysis of mtDNA can be paired with the PCR to be further amplified. The traditional Sanger method is often used to sequence the amplicons, which involves fluorescent dideoxynucleotide and cycle sequencing. An alternative to the Sanger method is the use of electrospray ionization mass spectrometry (ESI-MS). However, both methods are limited in analyzing mixed samples due to chimeric mtDNA products forming during analysis.

V. DNA methylation analysis –

DNA methylation appears to be the best suited for body fluid identification at this time, due to its high specificity and compatibility with current STR typing protocols. In CpG dinucleotides there is a DNA methylation at C5 of cytosine residues. This is a modification that occurs in mammals and is involved in cellular differentiation. In particular, there are tissue-specific differential methylated regions (tDMRs) that occur in the genome of mammals. PCR methods that are methylation specific are able to identify body fluid identification and could possibly be multiplexed with existing STR typing protocols. DNA methylation also has the potential to be used to help determine an estimated age.

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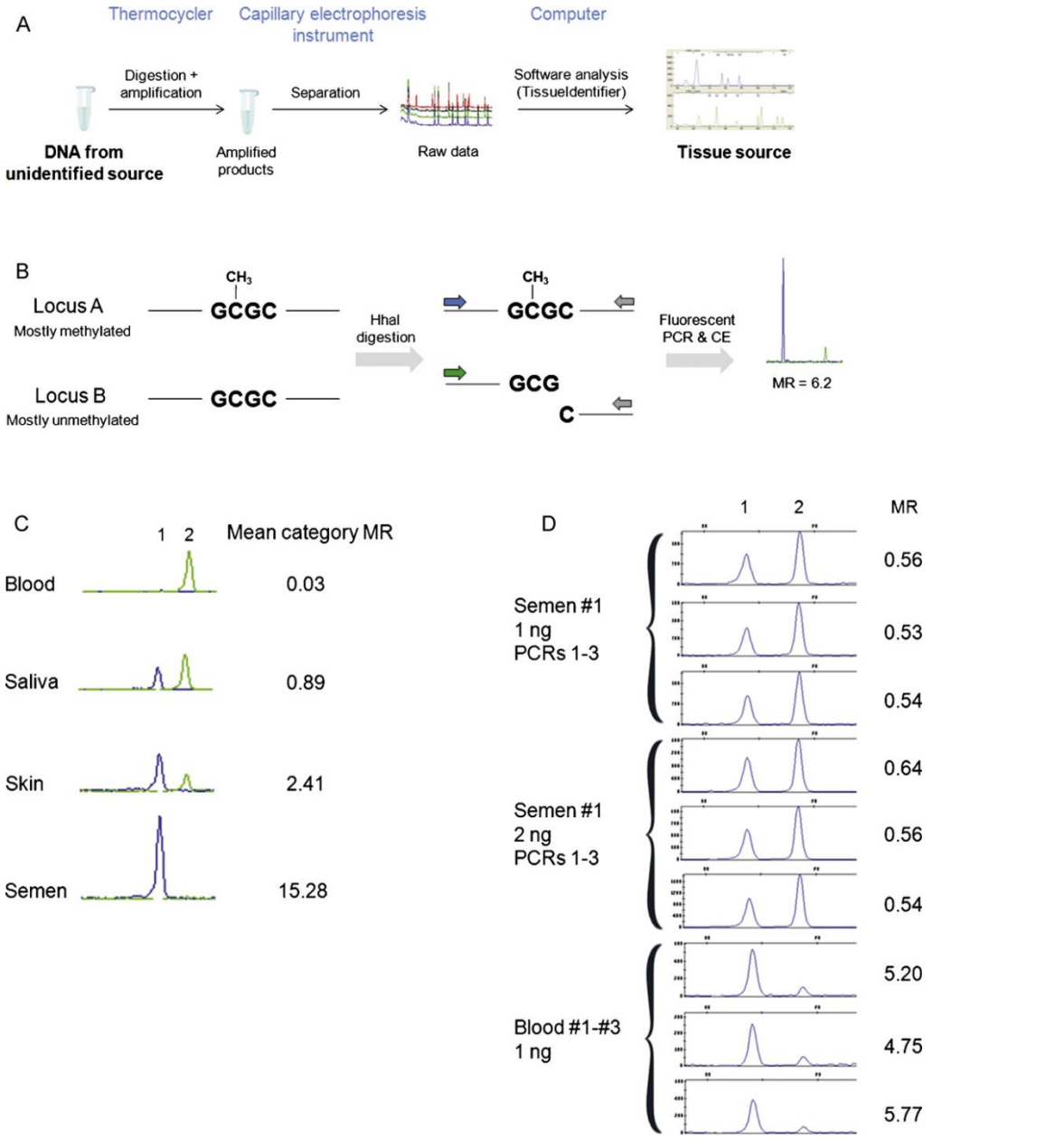

Fig.: *Tissue identification assay.* (A) Schematic overview of the assay. (B) Biochemical procedure—methylated loci remain intact during digestion and subsequently are amplified efficiently in the PCR, producing a strong signal (locus A) while unmethylated loci are digested and subsequently amplify inefficiently in the PCR, producing a weak signal (locus B). The methylation ratio (MR; rfu of locus A/rfu of locus B) reflects the differential methylation level between loci A and B. (C) Methylation ratios between locus 1 and locus 2 are different in blood, saliva, skin, and semen, reflecting the differential methylation patterns in these tissues. (D) The observed differences in methylation ratios between blood and semen (i.e. the signal) are more than a magnitude greater than the observed differences in methylation ratios obtained from different PCRs and from different amounts of input DNA (i.e. the noise).

RESULT:

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EXP.7: E-Mail Investigation, E-Mail Tracking, IP Tracking, E-Mail Recovery, Recovering deleted evidences, Password Cracking.

AIM:

To understand the methods of Forensic Examination of Cyber Crime in Social Networking Sites.

PRINCIPLE:

The use of the internet is very essential because they can gather and share information with other individuals as well as the decreasing cost and size of computers no matter where individuals are located on the globe. Thus facilitates business at domestic to global level. However this new technology has brought with it much advancement which makes our lives easier but unfortunately it has also led to advancements in crime. The growth of the internet has also resulted in the creation and growth of cybercrime due to ease of availability and connections through world web. Cybercrime is a major issue facing society today, requiring law makers and law enforcement agencies to take action. This issue can have a major impact on governments, businesses, and individuals and thus deserves the attention of researchers.

Some examples of cybercrime are: financial theft through E-banking, pornography, data stealing, data manipulation, hacking, cracking etc. The hacker and crackers are the individuals who get access into a system or into a network without any authorization. Mainly used techniques by the scammers involve reconnaissance in other word one can say foot printing (gathering information about the targeted person is called foot printing). It involves three techniques dumpster diving in this technique scammer uses to go through the victim's garbage and tries to gather useful information about the victim. The second technique is social engineering it is an art of convincing people to reveal sensitive information. The third is shoulder surfing it is a technique in which attacker spies over the victim's shoulder and try to steal sensitive data of the victim which is displayed on the screen of the victim.

Case study: 1

An officer of a steel plant named Akash Shrivastava of Jabalpur was browsing on his personal computer in his office a popup of 'Facebook notification' came. He eagerly clicked on that link to join the chat or see the new post in his profile but he found that it is an advertisement of another social networking site he refused it and went to lunch room to take his lunch. When he came back he found that his all deals and all data related to company working and marketing strategies are deleted and in this way he was in loss of Rs.5, 00,000. While investigation it is found that the popup

of 'Facebook chat' was containing an infected link that have a patch file hidden in it of a software called net bus through which the employee of the same company offended this crime, with the help of this software he hacked his boss's computer and committed this crime.

Elements of this case: fake link 'Facebook', netbus tool. In this case the interest of Mr. Akash Shrivastava in Facebook which is a social networking site made him a victim, and so many social networking sites and their fake pages with popup are available on internet which may helpful to the crackers or hackers.

"Be aware from subordinates and friends in financial matter."

Case study: 2

A business man activated internet banking on his account, after some days he found that all his account balance Rs.7,50,000 has been transferred to an account through internet banking. After the investigation offender confessed his offence when appeared in the court and told to the court that he was actually a friend of the victim. One day the victim's maid was dumping some paper pieces at dump yard through dumpster diving he found an envelope having information regarding the confirmation of activation of internet banking also mention that the new user ID and password has been sent to your registered e-mail account. That envelope was stolen by the accused. Through shoulder surfing he noticed that the victim use to save his all ID and passwords in his opera browser. In his first attempt to get the ID and password he found that the victim's laptop was protected with a password and a hint statement for that password, which was "add jay after your elder brother's best friend's name". Here he had taken advantage of social networking site called Facebook; firstly he opened victim's profile then he gone to his elder brother's profile where he checked the list of close friends and found only one name 'Surya'. He attempted many times with different password like 'Suryajay, Surajjay, Prabhakarjay etc." at last he got the password as Sunjay from there he had stolen the mail id password and from there he transferred money by login in internet banking site of the bank and transferred all the money to another account.

Elements of this case: Dumpster diving, shoulder surfing, Facebook profiles, Hit and trial method. As in above case social networking sites are also helpful to gather sensitive information like phone no., address, photos, friends etc. especially when victim is a female.

Case study: 3

One day a MMS came to a girl namely Divya Kapoor, containing her vulgar photograph followed by a SMS within a minute. The accused was blackmailing the girl for money in lieu of publically

displaying the photograph through web. The investigating team found that the photo was edited by using tool trick photography and the photograph of that girl was downloaded from her unsecured Facebook profile.

Elements of this case: Facebook profile, Photo editor software, Mobile phone. Thus uploading her photograph on the internet especially in social networking sites made her victim.

RESULT: