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SCHOOL OF BIO & CHEMICAL ENGINEERING

DEPARTMENT OF BIOMEDICAL ENGINEERING

UNIT – I – ANIMAL CELL STRUCTURE – SBMA4007

UNIT I

Forensic science is the application of science to criminal and civil laws. Forensic scientists collect, preserve, and analyze scientific evidence during the course of an investigation.

Forensic investigation - The term forensic investigation refers to the use of science or technology in the investigation and establishment of facts or evidence to be used in criminal justice or other proceedings. Forensic investigation is a rather broad field with many different subdivisions.

Forensic Data Analysis (FDA) is a branch of Digital forensics. It examines structured data with regard to incidents of financial crime. The aim is to discover and analyse patterns of fraudulent activities.

Origins of forensic science

Ambroise Paré's surgical work laid the groundwork for the development of forensic techniques in the following centuries.

In 16th-century Europe, medical practitioners in army and university settings began to gather information on the cause and manner of death. Ambroise Paré, a French army surgeon, systematically studied the effects of violent death on internal organs. Two Italian surgeons, Fortunato Fidelis and Paolo Zacchia, laid the foundation of modern pathology by studying changes that occurred in the structure of the body as the result of disease. In the late 18th century, writings on these topics began to appear. These included *A Treatise on Forensic Medicine and Public Health* by the French physician Francois Immanuele Fodéré and *The Complete System of Police Medicine* by the German medical expert Johann Peter Frank.

Two examples of English forensic science in individual legal proceedings demonstrate the increasing use of logic and procedure in criminal investigations at the time. In 1784, in Lancaster, John Toms was tried and convicted for murdering Edward Culshaw with a pistol. When the dead body of Culshaw was examined, a pistol wad (crushed paper used to secure powder and balls in the muzzle) found in his head wound matched perfectly with a torn newspaper found in Toms's pocket, leading to the conviction.

Fingerprints

Sir William Herschel was one of the first to advocate the use of fingerprinting in the identification of criminal suspects. While working for the Indian Civil Service, he began to use thumbprints on

documents as a security measure to prevent the then-rampant repudiation of signatures in 1858.

Fig 1.1: Fingerprints taken by William Herschel 1859/60.

In 1877 at Hooghly (near Calcutta), Herschel instituted the use of fingerprints on contracts and deeds, and he registered government pensioners' fingerprints to prevent the collection of money by relatives after a pensioner's death.

In 1880, Dr. Henry Faulds, a Scottish surgeon in a Tokyo hospital, published his first paper on the subject in the scientific journal *Nature*, discussing the usefulness of fingerprints for identification and proposing a method to record them with printing ink. He established their first classification and was also the first to identify fingerprints left on a vial. Returning to the UK in 1886, he offered the concept to the Metropolitan Police in London, but it was dismissed at that time.

Faulds wrote to Charles Darwin with a description of his method, but, too old and ill to work on it, Darwin gave the information to his cousin, Francis Galton, who was interested in anthropology. Having been thus inspired to study fingerprints for ten years, Galton published a detailed statistical model of fingerprint analysis and identification and encouraged its use in forensic science in his book *Finger Prints*. He had calculated that the chance of a "false positive" (two different individuals having the same fingerprints) was about 1 in 64 billion.

Juan Vucetich, an Argentine chief police officer, created the first method of recording the fingerprints of individuals on file. In 1892, after studying Galton's pattern types, Vucetich set up the world's first fingerprint bureau. In that same year, Francisca Rojas of Necochea was found in a house with neck injuries whilst her two sons were found dead with their throats cut. Rojas accused a neighbour, but despite brutal interrogation, this neighbour would not confess to the crimes. Inspector Alvarez, a colleague of Vucetich, went to the scene and found a bloody thumb mark on a door. When it was compared with Rojas' prints, it was found to be identical with her right thumb. She then confessed to the murder of her sons.

A Fingerprint Bureau was established in Calcutta (Kolkata), India, in 1897, after the Council of the Governor General approved a committee report that fingerprints should be used for the classification of criminal records.

Bureau, before it became the Fingerprint Bureau, were Azizul Haque and Hem Chandra Bose. Haque and Bose were Indian fingerprint experts who have been credited with the primary development of a fingerprint classification system eventually named after their supervisor, Sir Edward Richard Henry. The Henry Classification System, co-devised by Haque and Bose, was accepted in England and Wales when the first United Kingdom Fingerprint Bureau was founded in Scotland Yard, the Metropolitan Police headquarters, London, in 1901. Sir Edward Richard Henry subsequently achieved improvements in dactyloscopy.

In the United States, Dr. Henry P. DeForrest used fingerprinting in the New York Civil Service in 1902, and by December 1905, New York City Police Department Deputy Commissioner Joseph A. Faurot, an expert in the Bertillon system and a fingerprint advocate at Police Headquarters, introduced the fingerprinting of criminals to the United States.

DNA fingerprinting was first used in 1984. It was discovered by Sir Alec Jefferys who realized that variation in the genetic code could be used to identify individuals and to tell individuals apart from one another. The first application of DNA profiles was used by Jefferys in a double murder mystery in a small England town called Narborough, Leicestershire in 1983. A 15-year-old school girl by the name of Lynda Mann was raped and murdered in Carlton Hayes psychiatric hospital. The police did not find a suspect but were able to obtain a semen sample.

In 1986, Dawn Ashworth, 15 years old, was also raped and strangled in a nearby village of Enderby. Forensic evidence showed that both killers had the same blood type. Richard Buckland became the suspect because he worked at Carlton Hayes psychiatric hospital, had been spotted near Dawn Ashworth's murder scene and knew unreleased details about the body. He later confessed to Dawn's murder but not Lynda's. Sir Alec Jefferys was brought into case to analyze the semen samples. He concluded that there was no match between the samples and Buckland, who became the first person to be exonerated using DNA. Jefferys confirmed that the DNA profiles were identical for the two murder semen samples. To find the perpetrator, DNA from entire male population, more than 4,000 aged from 17 to 34, in town was collected. They all were compared to semen samples from the crime.

A friend of Colin Pitchfork was heard saying that he had given his sample to the police claiming to be Colin. Colin Pitchfork was arrested in 1987 and it was found that his DNA profile matched the semen samples from the murder.

Because of this case, DNA databases came into being. There is the national (FBI) and international database as well as the European countries (ENFSI). These searchable databases are used to match crime scene DNA profiles to those already in database.

20th century

Alec Jeffreys invented the DNA profiling technique in 1984. Later in the 20th century several British pathologists, Mikey Rochman, Francis Camps, Sydney Smith and Keith Simpson pioneered new forensic science methods. Alec Jeffreys pioneered the use of DNA profiling in forensic science in 1984. He realized the scope of DNA fingerprinting, which uses variations in the genetic code to identify individuals. The method has since become important in forensic science to assist police detective work, and it has also proved useful in resolving paternity and immigration disputes. DNA fingerprinting was first used as a police forensic test to identify the rapist and killer of two teenagers, Lynda Mann and Dawn Ashworth, who were both murdered in Narborough, Leicestershire, in 1983 and 1986 respectively. Colin Pitchfork was identified and convicted of murder after samples taken from him matched semen samples taken from the two dead girls.

Forensic science has been fostered by a number of national forensic science learned bodies including the Chartered Society of Forensic Sciences, (founded in 1959), then known as the Forensic Science Society, publisher of *Science & Justice*;. American Academy of Forensic Sciences (founded 1948), publishers of the *Journal of Forensic Sciences*; the Canadian Society of Forensic Science (founded 1953), publishers of the *Journal of the Canadian Society of Forensic Science*; the British Academy of Forensic Sciences (founded 1960), publishers of *Medicine, science and the law*,[53] and the Australian Academy of Forensic Sciences (founded 1967), publishers of the *Australian Journal of Forensic Sciences*.

CELL

The cell was discovered by Robert Hooke in 1665, who named the biological unit for its resemblance to cells inhabited by Christian monks in a monastery. cells are the fundamental unit of structure and function in all living organisms, that all cells come from preexisting cells, and that all cells contain the hereditary information necessary for regulating cell functions and for transmitting information to the next generation of cells.

Structure of cells

Cells are minute structures which can only be studied using specialist viewing equipment called microscopes. There are two types of microscope: light microscopes use light beams and are useful for viewing tissues and larger cells; electron microscopes use beams of electrons and provide much greater magnification in order to study the interior of human cells.

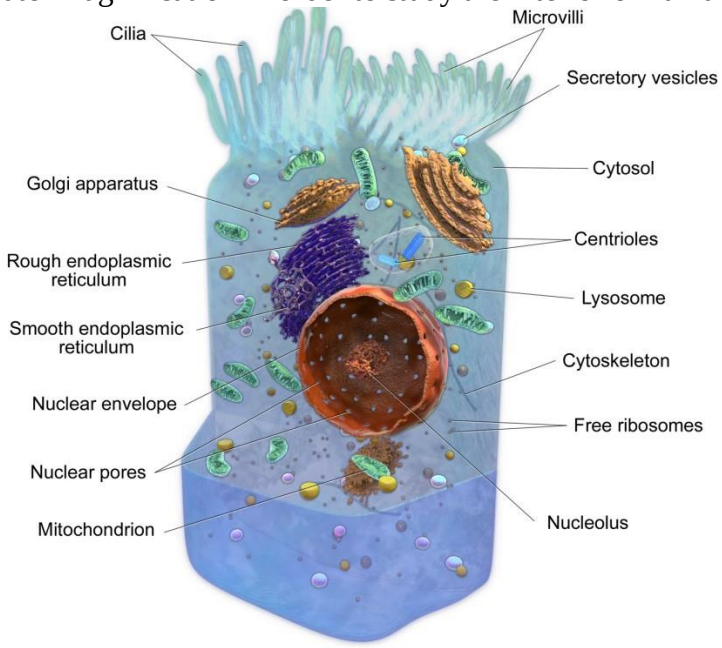

Fig. 1.2. Structure of a Cell

Plants, animals, fungi, slime moulds, protozoa, and algae are all eukaryotic. These cells are about fifteen times wider than a typical prokaryote and can be as much as a thousand times greater in volume. The main distinguishing feature of eukaryotes as compared to prokaryotes is compartmentalization: the presence of membrane-bound organelles (compartments) in which

specific metabolic activities take place. Most important among these is a cell nucleus, an organelle that houses the cell's DNA. This nucleus gives the eukaryote its name, which means "true kernel (nucleus)". Other differences include:

- The plasma membrane resembles that of prokaryotes in function, with minor differences in the setup. Cell walls may or may not be present.

- The eukaryotic DNA is organized in one or more linear molecules, called chromosomes, which are associated with histone proteins. All chromosomal DNA is stored in the *cell nucleus*, separated from the cytoplasm by a membrane. Some eukaryotic organelles such as mitochondria also contain some DNA.
- Many eukaryotic cells are ciliated with *primary cilia*. Primary cilia play important roles in chemosensation, mechanosensation, and thermosensation. Cilia may thus be "viewed as a sensory cellular antennae that coordinates a large number of cellular signaling pathways, sometimes coupling the signaling to ciliary motility or alternatively to cell division and differentiation."
- Motile cells of eukaryotes can move using *motile cilia* or *flagella*. Motile cells are absent in conifers and flowering plants.[15] Eukaryotic flagella are less complex than those of prokaryotes.

Sub-cellular components

All cells, whether prokaryotic or eukaryotic, have a membrane that envelops the cell, regulates what moves in and out (selectively permeable), and maintains the electric potential of the cell. Inside the membrane, the cytoplasm takes up most of the cell's volume. All cells (except red blood cells which lack a cell nucleus and most organelles to accommodate maximum space for hemoglobin) possess DNA, the hereditary material of genes, and RNA, containing the information necessary to build various proteins such as enzymes, the cell's primary machinery. There are also other kinds of biomolecules in cells.

Membrane

The cell membrane, or plasma membrane, is a biological membrane that surrounds the cytoplasm of a cell. In animals, the plasma membrane is the outer boundary of the cell, while in plants and prokaryotes it is usually covered by a cell wall. This membrane serves to separate and protect a cell from its surrounding environment and is made mostly from a double layer of phospholipids, which are amphiphilic (partly hydrophobic and partly hydrophilic). Hence, the layer is called a phospholipid bilayer, or sometimes a fluid mosaic membrane. Embedded within this membrane is a variety of protein molecules that act as channels and pumps that move different molecules into and out of the cell. The membrane is said to be 'semi-permeable', in that it can either let a substance (molecule or ion) pass through freely, pass through to a limited extent or not pass through at all. Cell surface membranes also contain receptor proteins that allow cells to detect external signaling molecules such as hormones.

Cytoskeleton

The cytoskeleton acts to organize and maintain the cell's shape; anchors organelles in place; helps during endocytosis, the uptake of external materials by a cell, and cytokinesis, the separation of daughter cells after cell division; and moves parts of the cell in processes of growth and mobility. The eukaryotic cytoskeleton is composed of microfilaments, intermediate filaments

and microtubules. There are a great number of proteins associated with them, each controlling a cell's structure by directing, bundling, and aligning filaments. The prokaryotic cytoskeleton is less well-studied but is involved. in the maintenance of cell shape, polarity and cytokinesis. The subunit protein of microfilaments is a small, monomeric protein called actin. The subunit of microtubules is a dimeric molecule called tubulin. Intermediate filaments are heteropolymers whose subunits vary among the cell types in different tissues. But some of the subunit protein of intermediate filaments include vimentin, desmin, lamin (lamins A, B and C), keratin (multiple acidic and basic keratins), neurofilament proteins (NF - L, NF - M).

Genetic material

Two different kinds of genetic material exist: deoxyribonucleic acid (DNA) and ribonucleic acid (RNA). Cells use DNA for their long-term information storage. The biological information contained in an organism is encoded in its DNA sequence. RNA is used for information transport (e.g., mRNA) and enzymatic functions (e.g., ribosomal RNA). Transfer RNA (tRNA) molecules are used to add amino acids during protein translation.

Prokaryotic genetic material is organized in a simple circular DNA molecule (the bacterial chromosome) in the nucleoid region of the cytoplasm. Eukaryotic genetic material is divided into different, linear molecules called chromosomes inside a discrete nucleus, usually with additional genetic material in some organelles like mitochondria and chloroplasts (see endosymbiotic theory).

A human cell has genetic material contained in the cell nucleus (the nuclear genome) and in the mitochondria (the mitochondrial genome). In humans the nuclear genome is divided into 46 linear DNA molecules called chromosomes, including 22 homologous chromosome pairs and a pair of sex chromosomes. The mitochondrial genome is a circular DNA molecule distinct from the nuclear DNA. Although the mitochondrial DNA is very small compared to nuclear chromosomes, it codes for 13 proteins involved in mitochondrial energy production and specific tRNAs.

Foreign genetic material (most commonly DNA) can also be artificially introduced into the cell by a process called transfection. This can be transient, if the DNA is not inserted into the cell's genome, or stable, if it is. Certain viruses also insert their genetic material into the genome.

Organelles

Organelles are parts of the cell which are adapted and/or specialized for carrying out one or more vital functions, analogous to the organs of the human body (such as the heart, lung, and kidney, with each organ performing a different function). Both eukaryotic and prokaryotic cells have organelles, but prokaryotic organelles are generally simpler and are not membrane-bound.

There are several types of organelles in a cell. Some (such as the nucleus and golgi apparatus) are typically solitary, while others (such as mitochondria, chloroplasts, peroxisomes and lysosomes) can be numerous (hundreds to thousands). The cytosol is the gelatinous fluid that fills the cell and surrounds the organelles.

Eukaryotic

Human cancer cells with nuclei (specifically the DNA) stained blue. The central and rightmost cell are in interphase, so the entire nuclei are labeled. The cell on the left is going through mitosis and its DNA has condensed.

- Cell nucleus: A cell's information center, the cell nucleus is the most conspicuous organelle found in a eukaryotic cell. It houses the cell's chromosomes, and is the place where almost all DNA replication and RNA synthesis (transcription) occur. The nucleus is spherical and separated from the cytoplasm by a double membrane called the nuclear envelope. The nuclear envelope isolates and protects a cell's DNA from various molecules that could accidentally damage its structure or interfere with its processing. During processing, DNA is transcribed, or copied into a special RNA, called messenger RNA (mRNA). This mRNA is then transported out of the nucleus, where it is translated into a specific protein molecule. The nucleolus is a specialized region within the nucleus where ribosome subunits are assembled. In prokaryotes, DNA processing takes place in the cytoplasm.
- Mitochondria and Chloroplasts: generate energy for the cell. Mitochondria are self-replicating organelles that occur in various numbers, shapes, and sizes in the cytoplasm of all eukaryotic cells. Respiration occurs in the cell mitochondria, which generate the cell's energy by oxidative phosphorylation, using oxygen to release energy stored in cellular nutrients (typically pertaining to glucose) to generate ATP. Mitochondria multiply by binary fission, like prokaryotes. Chloroplasts can only be found in plants and algae, and they capture the sun's energy to make carbohydrates through photosynthesis.
- Endoplasmic reticulum: The endoplasmic reticulum (ER) is a transport network for molecules targeted for certain modifications and specific destinations, as compared to molecules that float freely in the cytoplasm. The ER has two forms: the rough ER, which has ribosomes on its surface that secrete proteins into the ER, and the smooth ER, which lacks ribosomes. The smooth ER plays a role in calcium sequestration and release.
- Golgi apparatus: The primary function of the Golgi apparatus is to process and package the macromolecules such as proteins and lipids that are synthesized by the cell.
- Lysosomes and Peroxisomes: Lysosomes contain digestive enzymes (acid hydrolases). They digest excess or worn-out organelles, food particles, and engulfed viruses or bacteria. Peroxisomes have enzymes that rid the cell of toxic peroxides. The cell could not house these destructive enzymes if they were not contained in a membrane-bound system.
- Centrosome: the cytoskeleton organiser: The centrosome produces the microtubules of a cell – a key component of the cytoskeleton. It directs the transport through the ER and the Golgi apparatus. Centrosomes are composed of two centrioles, which separate during cell division and help in the formation of the mitotic spindle. A single centrosome is present in the animal cells. They are also found in some fungi and algae cells.
- Vacuoles: Vacuoles sequester waste products and in plant cells store water. They are often described as liquid filled space and are surrounded by a membrane. Some cells, most notably Amoeba, have contractile vacuoles, which can pump water out of the cell if there is too much water. The vacuoles of plant cells and fungal cells are usually larger than those of animal cells.

Eukaryotic and prokaryotic

- Ribosomes: The ribosome is a large complex of RNA and protein molecules. They each consist of two subunits, and act as an assembly line where RNA from the nucleus is used to synthesise proteins from amino acids. Ribosomes can be found either floating freely or bound to a membrane (the rough endoplasmatic reticulum in eukaryotes, or the cell membrane in prokaryotes).

Types of Cells in the Human Body

Human body has a number of cells and related tissues. Since the Cell is the basic unit of any tissue, it decides the type, nature and function of the tissues.

A group of different types of cells form tissues in the human body and one or more types of tissues form organs. These body organs working in mutual manner form the organ systems.

If any tissue is damaged, it actually means the cells and cell structure in the organ is damaged. These damaged cells decides the fate of entire tissue i.e. either to recover or die. Many cells have internal tendency to multiply and help in repair while few don't have this property. Let's see list of cells of human body with their functions.

Different Types of cells in the human body (based on tissue formation)

1. Bone cells (*Osteocytes*) They are the toughest body Cell as they are bound together by calcium and phosphate. As you knows they give strength, support and frame-work to the body by enclosing organs in skeletal system i.e bones.

2. Cartilage cells (*chondrocytes*)– These cells are similar to bone cells but the surrounding material is just loose and flexible compared to those of bone cells. Hence they are freely bendable. They are present in ear bone (hence ears are fold-able), in between large bones to help them bend and move freely like in between two ribs, spinal bones, joints etc.

3. Nerve cells: These cells are very long and have many branching at either ends. Their specialty is they never multiply in one's life time. They are present all over the body and are sometimes as long as few meters long. They are human brain cells and are found in plenty in brain and the spinal cord and form the nervous tissue.

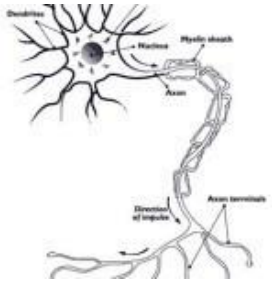

Fig 1.3: Neural cell

4. Epithelial cell: These cells are very simple cells which form covering of other cells. These cells form covering layers of all the organs and hence are preset in skin, scalp, respiratory tract, in the buccal cavity surface etc. Ex: Skin cells, mucous cells.

5. Muscle cells: These cells are of muscle tissue mostly long, large and have ability to contract and relax providing movements. They are three types as skeletal, cardiac and smooth muscles.

- Skeletal muscles cells are attached to long bones and assist in their movement (by muscle contraction).
- Cardiac muscles cells are present only in heart muscle and responsible for heart beats.

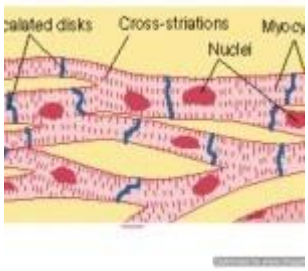

Fig 1.4: Cardiac cell

- Smooth muscle cells are flexible yet, can contract and relax and are present in stomach, intestine, blood vessel walls (vascular tissue) etc. helping in movement of food through the gut.

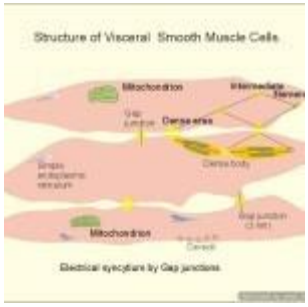

Fig 1.5: Smooth cell

6 Secretory cell: These cells as name indicates are secretory in nature. They form glands and secrete something important ex; pancreatic cells which secrete insulin, glucagon, salivary gland secrete salivary amylase, sebaceous gland secrete oil on the skin etc.They are found in all secretory organs.

7. Adipose cells: These are fat cells and are storage in nature to store fat. Especially seen in the soles, palms, bums etc. They reduce friction to the body.

8. Blood cells: These cells include RBC's, WBC, Thormbocytes etc. They are always motile and never stay in one place. They have limited life span and they never multiply to form new cells. Instead new cells are formed from other cells.

Types of cells in human body (based on their function)

Conductive cells: Nerve cells, muscle cells come under this category. They have internal ability to conduct an electric impulse from region to other distant region in the body.

Connective cells: Bone cells, blood cells come under this category. They help connect other cells and tissues.

Glandular cells: These cells secretory cells. They form glands like pancreas, salivary glands etc and help in production of enzymes, hormones etc.

Storage cells: Adipose cells, some liver cells etc act to store materials like fat for later use. This fat

is consumed in time of starvation and also in excess cold temperatures.

Supportive cells: These are the cells which are present as support to adjacent cells. Ex: Glial cells in the brain and spinal cord help provide nourishment to the nerve cells and also protect them from shocks and trauma.

Special type of cells: These are specialized cells with important functions in the body.
They are

a) Sperms: These cells unlike others are haploid (i.e. have only one set of chromosome). They are present only in the males after puberty. These cells have a tail which enables them to swim and move in the female uterus. They have an enzyme namely hayaluronidase which helps them penetrate through uterine tissue and reach oocytes.

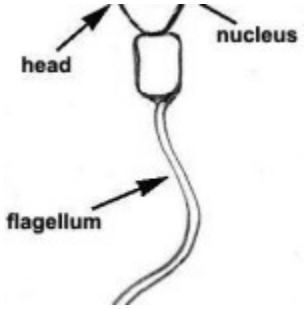

Fig 1.6: Structure of a Sperm

b) Oocytes: Cells are haploid and present in adult female genital system. They are also haploid like sperms. They start to form after puberty and continue so till the stage of menopause. They accept sperm cells to form zygote (fertilized egg). This further grows in the uterus to form a baby.

c) Stem cells: These are basic cells or parent cells which can differentiate into any cell based on the requirement. These stem cells in human body are given so importance due to their promising role in treatment of disorders in future. See the article on stem cell types.

d) Rods & cones: These cells are in eye and have capacity to capture image color and light.

e) Ciliated cells: These cells are present as lining of respiratory tract, esophagus etc. and have a pointed thread like cilia which move in one particular direction to pass material.

f) Blood cells: These are quite interesting cells and they are never attached to one another. Blood cells freely flow in the liquid blood. Some of them are not alive (RBC‘s) while others have varied shapes like WBC, platelets (spindle shape). Further these WBC’s are of different types. Of them macrophages have ability to eat (gulp) any foreign particle like bacteria in the body. Hence they are body defense cells.

Blood

Blood is a constantly circulating fluid providing the body with nutrition, oxygen, and waste removal. Blood is mostly liquid, with numerous cells and proteins suspended in it, making blood "thicker" than pure water. The average person has about 5 liters (more than a gallon) of blood.

A liquid called plasma makes up about half of the content of blood. Plasma contains proteins that help blood to clot, transport substances through the blood, and perform other functions. Blood plasma also contains glucose and other dissolved nutrients.

About half of blood volume is composed of blood cells:
• Red blood cells, which carry oxygen to the tissues
• White blood cells, which fight infections
• Platelets, smaller cells that help blood to clot

Blood is conducted through blood vessels (arteries and veins). Blood is prevented from clotting in the blood vessels by their smoothness, and the finely tuned balance of clotting factors.

Fig 1.7: Structure of a blood vessel

Body fluids

Body fluids are liquids originating from inside the bodies of living humans. They include fluids that are excreted or secreted from the body. Human blood, body fluids, and other body tissues are widely recognised as vehicles for the transmission of human disease.

List of body fluids - Amniotic fluid, Blood serum, Breast milk, Feces , Female ejaculate, Gastric acid, Gastric juice, Lymph, Mucus , Sebum (skin oil), Semen, Sputum, Synovial fluid, Sweat, Tears, Urine, Vaginal secretion, Vomit etc.,

Body fluid is the term most often used in medical and health contexts.
Modern medical, public health, and personal hygiene practices treat body fluids as potentially unclean. This is because they can be vectors for infectious diseases, such as sexually transmitted diseases or blood-borne diseases. Universal precautions and safer sex practices try to avoid exchanges of body fluids. Body fluids can be analyzed in medical laboratory in order to find microbes, inflammation, cancers, etc. Clinical samples are generally defined as non-infectious human or animal materials including blood, saliva, excreta, body tissue and tissue fluids, and also FDA-approved pharmaceuticals that are blood products. In medical contexts, it is a specimen taken for diagnostic examination or evaluation, and for identification of disease or condition.

There are many different types of bodily fluid that are secreted by the body and are also present within the body at any given time. These fluids may be useful in helping forensic scientists and pathologists put together a detailed picture of how an individual died and likewise may also present

means of identifying the perpetrator.

Types of Bodily Fluid

Bodily fluids are broken down into two categories: excreted and secreted. Within these categories you will find the following:

- Excreted: Sweat, Breast Milk, Cerumen (Earwax), Faeces (included because faeces are often covered in a mucus membrane to enable travelling through the bowel), Chyme (found in the stomach), Bile, Vomit, Aqueous Humour (a watery substance that covers the eye), Sebum (Skin Oil)
- Secreted: Cowper's Fluid (Pre-ejaculatory fluid), Blood or Plasma, Semen, Saliva, Female Ejaculate, Serum, or Urine.

Bodily Fluids as Evidence

These varying fluids are to be found contained within the human body and those that are classed as being secreted can be found on or about the body of an individual who has been the victim of a crime.

The most common bodily fluids to be found in these instances are blood, semen, serum, saliva and sometimes - given the severity of the crime - urine or faeces. It is the job of the forensic scientist or Scene of Crime Officer (SOCO) to establish if any of these bodily fluids are present at a crime scene and take the necessary steps to ensure that they are collected. These fluids - if detected at a crime scene - are swabbed, bagged and collected in vials, which are air tight and at low risk of cross contamination.

In addition, the use of chemicals and ultra violet light can be deployed to uncover the existence of any of these fluids in circumstances where they might be overlooked. Particularly if the crime scene is a darkly lit area or an outdoor area which covers considerable distance.

Also useful to note is the fact that not all bodily fluids contain sufficient information to gain a DNA comparison. This happens when the individual is what is described as a 'non-secretor'. A 'non-secretor' will not have sufficient levels of protein in their bodily fluids to determine a match between blood and bodily fluids found at a crime scene. Ofcourse the percentage of 'non-secretors' among the populous as opposed to 'secretors' is very small indeed.

Another important factor when dealing with a crime scene that may have bodily fluids secreted is that all personnel must be fully clothed in protective clothing to avoid both contamination of the crime scene and potential exposure to blood-borne diseases that may be passed to them via cuts, scrapes and/or ingestion.

Again it is worth considering the risks of infection to all concerned and also looking at how crime scene evidence is collected; without proper quarantine procedures evidence that might be vital to the success of any criminal investigation could - and has on occasion - been inadvertently cross

contaminated by the person or persons handling its collection and transportation. Of course these instances are rare but it is vital that every care is taken to ensure that this is not the case.

As well as bodily fluids the forensic scientist - and/or Scenes of Crime Officer (SOCO) will look for trace evidence such as skin particles, hairs, fingernails and anything else that might have been in contact with an attacker or assailant.

Determining Cause of Death

Determining cause of death is the most important task a pathologist can perform during the course of an autopsy. In terms of legality it is a necessity for law enforcement to be able to prove beyond any doubt that the deceased has died of means other than natural causes.

Indeed such are the statistics of murder in parts of the world that it is a basic requirement for any individual who has died unexpectedly to have an autopsy performed on them in order to prove definitely the cause of death and rule out the chance of foul play.

Factors to Consider

Determining the cause of death is done by taking a number of factors into consideration.

Firstly was the deceased found at a location where he or she would not normally have been? Where they in a state of undress or have visible injuries not normally present in a natural death? If injuries were present were they the result of a knife or gun attack?

All of these questions have to be answered and answered satisfactorily in the eyes of a pathologist. In some deaths an inquest is necessary to decide on how an individual died

- be it death by misadventure, accidental death or natural causes and a pathologist will be called upon to give expert testimony that will prove beyond any doubt what the cause of death actually was.

Human skeletal system

The skeletal system includes all of the bones and joints in the body. Each bone is a complex living organ that is made up of many cells, protein fibers, and minerals. The skeleton acts as a scaffold by providing support and protection for the soft tissues that make up the rest of the body. The skeletal system also provides attachment points for muscles to allow movements at the joints. New blood cells are produced by the red bone marrow inside of our bones. Bones act as the body's warehouse for calcium, iron, and energy in the form of fat. Finally, the skeleton grows throughout childhood and provides a framework for the rest of the body to grow along with it.

The skeletal system in an adult body is made up of 206 individual bones. These bones are arranged into two major divisions: the *axial skeleton* and the *appendicular skeleton*. The axial skeleton runs along the body's midline axis and is made up of 80 bones in the following regions:

- Skull
- Hyoid
- Auditory ossicles
- Ribs

- Sternum
- Vertebral column

The appendicular skeleton is made up of 126 bones in the folowing regions:

- Upper limbs
- Lower limbs
- Pelvic girdle
- Pectoral (shoulder) girdle

Skull

The skull is composed of 22 bones that are fused together except for the mandible. These 21 fused bones are separate in children to allow the skull and brain to grow, but fuse to give added strength and protection as an adult. The mandible remains as a movable jaw bone and forms the only movable joint in the skull with the temporal bone. The bones of the superior portion of the skull are known as the cranium and protect the brain from damage. The bones of the inferior and anterior portion of the skull are known as facial bones and support the eyes, nose, and mouth.

Vertebrae
Twenty-six vertebrae form the vertebral column of the human body. They are named by region:

- Cervical (neck) - 7 vertebrae
- Thoracic (chest) - 12 vertebrae
- Lumbar (lower back) - 5 vertebrae
- Sacrum - 1 vertebra
- Coccyx (tailbone) - 1 vertebra

With the exception of the singular sacrum and coccyx, each vertebra is named for the first letter of its region and its position along the superior-inferior axis. For example, the most superior thoracic vertebra is called T1 and the most inferior is called T12.

Ribs and Sternum

The sternum, or breastbone, is a thin, knife-shaped bone located along the midline of the anterior side of the thoracic region of the skeleton. The sternum connects to the ribs by thin bands of cartilage called the costal cartilage.
There are 12 pairs of ribs that together with the sternum form the ribcage of the thoracic region. The first seven ribs are known as “true ribs” because they connect the thoracic vertebrae directly to the sternum through their own band of costal cartilage. Ribs 8, 9, and 10 all connect to the sternum through cartilage that is connected to the cartilage of the seventh rib, so we consider these to be “false ribs.” Ribs 11 and 12 are also false ribs, but are also considered to be “floating ribs” because they do not have any cartilage attachment to the sternum at all.

Pectoral Girdle and Upper Limb

The pectoral girdle connects the upper limb (arm) bones to the axial skeleton and consists of the

left and right clavicles and left and right scapulae.
The humerus is the bone of the upper arm. It forms the ball and socket joint of the shoulder with the scapula and forms the elbow joint with the lower arm bones. The radius and ulna are the two bones of the forearm. The ulna is on the medial side of the forearm and forms a hinge joint with the humerus at the elbow. The radius allows the forearm and hand to turn over at the wrist joint.
The lower arm bones form the wrist joint with the carpals, a group of eight small bones that give added flexibility to the wrist. The carpals are connected to the five metacarpals that form the bones of the hand and connect to each of the fingers. Each finger has three bones known as phalanges, except for the thumb, which only has two phalanges. Pelvic Girdle and Lower Limb Formed by the left and right hip bones, the pelvic girdle connects the lower limb (leg) bones to the axial skeleton.
The femur is the largest bone in the body and the only bone of the thigh (femoral) region. The femur forms the ball and socket hip joint with the hip bone and forms the knee joint with the tibia and patella. Commonly called the kneecap, the patella is special because it is one of the few bones that are not present at birth. The patella forms in early childhood to support the knee for walking and crawling.
The tibia and fibula are the bones of the lower leg. The tibia is much larger than the fibula and bears almost all of the body's weight. The fibula is mainly a muscle attachment point and is used to help maintain balance. The tibia and fibula form the ankle joint with the talus, one of the seven tarsal bones in the foot.
The tarsals are a group of seven small bones that form the posterior end of the foot and heel. The tarsals form joints with the five long metatarsals of the foot. Then each of the metatarsals forms a joint with one of the set of phalanges in the toes. Each toe has three phalanges, except for the big toe, which only has two phalanges.

Microscopic Structure of Bones

The skeleton makes up about 30-40% of an adult's body mass. The skeleton's mass is made up of nonliving bone matrix and many tiny bone cells. Roughly half of the bone matrix's mass is water, while the other half is collagen protein and solid crystals of calcium carbonate and calcium phosphate.

Living bone cells are found on the edges of bones and in small cavities inside of the bone matrix. Although these cells make up very little of the total bone mass, they have several very important roles in the functions of the skeletal system. The bone cells allow bones to:

- Grow and develop
- Be repaired following an injury or daily wear
- Be broken down to release their stored minerals

Types of Bones

All of the bones of the body can be broken down into five types: long, short, flat, irregular, and sesamoid.

- *Long.* Long bones are longer than they are wide and are the major bones of the limbs. Long bones grow more than the other classes of bone throughout childhood and so are responsible for the bulk of our height as adults. A hollow medullary cavity is found in the center of long bones and serves as a storage area for bone marrow. Examples of long bones include the femur, tibia, fibula,

metatarsals, and phalanges.

- *Short.* Short bones are about as long as they are wide and are often cubed or round in shape. The carpal bones of the wrist and the tarsal bones of the foot are examples of short bones.

- *Flat.* Flat bones vary greatly in size and shape, but have the common feature of being very thin in one direction. Because they are thin, flat bones do not have a medullary cavity like the long bones. The frontal, parietal, and occipital bones of the cranium—along with the ribs and hip bones—are all examples of flat bones.

- *Irregular.* Irregular bones have a shape that does not fit the pattern of the long, short, or flat bones. The vertebrae, sacrum, and coccyx of the spine—as well as the sphenoid, ethmoid, and zygomatic bones of the skull—are all irregular bones.

- *Sesamoid*. The sesamoid bones are formed after birth inside of tendons that run across joints. Sesamoid bones grow to protect the tendon from stresses and strains at the joint and can help to give a mechanical advantage to muscles pulling on the tendon. The patella and the pisiform bone of the carpals are the only sesamoid bones that are counted as part of the 206 bones of the body. Other sesamoid bones can form in the joints of the hands and feet, but are not present in all people.

Parts of Bones

The long bones of the body contain many distinct regions due to the way in which they develop. At birth, each long bone is made of three individual bones separated by hyaline cartilage. Each end bone is called an epiphysis (epi = on; physis = to grow) while the middle bone is called a diaphysis (dia = passing through). The epiphyses and diaphysis grow towards one another and eventually fuse into one bone. The region of growth and eventual fusion in between the epiphysis and diaphysis is called the metaphysis (meta = after). Once the long bone parts have fused together, the only hyaline cartilage left in the bone is found as articular cartilage on the ends of the bone that form joints with other bones. The articular cartilage acts as a shock absorber and gliding surface between the bones to facilitate movement at the joint.

Deep to the periosteum is the compact bone that makes up the hard, mineralized portion of the bone. Compact bone is made of a matrix of hard mineral salts reinforced with tough collagen fibers. Many tiny cells called osteocytes live in small spaces in the matrix and help to maintain the strength and integrity of the compact bone.

Deep to the compact bone layer is a region of spongy bone where the bone tissue grows in thin columns called trabeculae with spaces for red bone marrow in between. The trabeculae grow in a specific pattern to resist outside stresses with the least amount of mass possible, keeping bones light but strong. Long bones have a spongy bone on their ends but have a hollow medullary cavity in the middle of the diaphysis. The medullary cavity contains red bone marrow during childhood, eventually turning into yellow bone marrow after puberty.

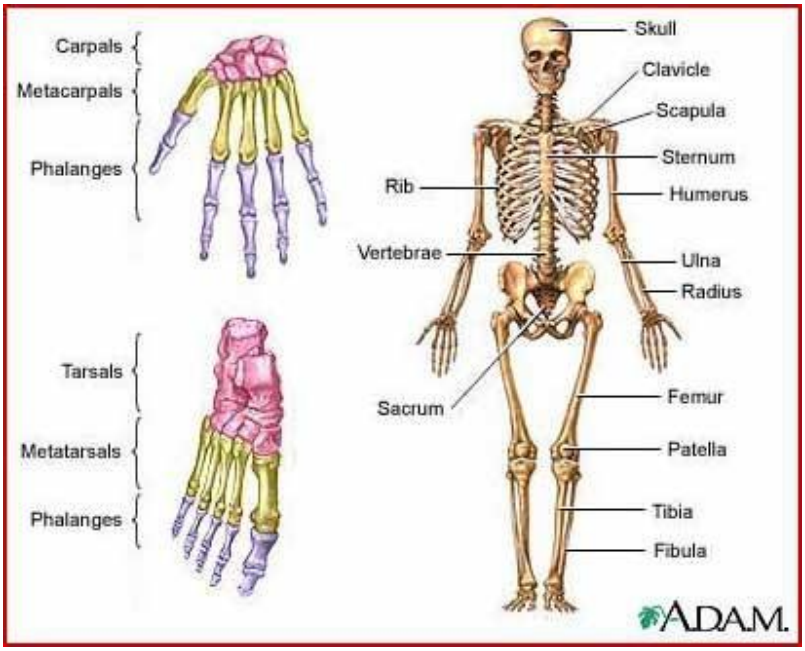

Fig 1.8: Medico Legal Importance of Bones

Bones are defined to be rigid organs that constitute part of endoskeleton of vertebrates. Bones are important medico-legally in various aspects. Before bones are to be used for any medico-legal examination, there are certain things to take care of. Many a times, the bones are fragmented, burnt, mixed with wood, or decolorized to such extents that a direct examination of the bone without establishing the following facts would result in wrong interpretation.

1. Whether the specimens are bone or not. This may be identified by physical examination, chemical examination or microscopy.
2. Whether the bones are human in origin or not. Many a times, bones are recovered in such degraded states that even experts of osteology find it difficult to confirm the human origin with mere physical examination. A chemical test such as precipitin test would come handy in such cases. Bones of great apes are not very different from that of humans and it becomes virtually indistinguishable in the case of the distal carpals, meta-carpals, tarsals or meta-tarsals.
3. Whether the bones are from a single individual or not. This is achieved by making a skeleton chart of the available bones. Any disproportion in size or count would point in the direction of multi individual origin. This becomes important in bones that are obtained while digging unknown areas as it might even be from an old graveyard.

Once these characteristics of the available bones are established, the bones may be considered for further forensic analysis. Bones are helpful in forensic medicine in establishing the following facts.

Establishing identity

Bones are extremely useful in establishing the identity of the individual of origin. Bones help in this regard because they may be used to establish the following characteristics of the individual of origin:

1. Race: This had been observed that there are differences in the anatomy of different races at the skeletal level. For example, the Negro populations of Africa tend to have long femurs compared to the rest of the word. This would mean that a skeleton with an abnormally long femur should remind us to think in that direction.
2. Gender: There exist anatomical, physiological and mental differences between the male and the female. It is safe to assume that these differences reflect in the bones as well. The most important of it is the pelvic bone or the hip bone. In females, the pelvic bone should be adapted to serve the additional function of child bearing and delivery and hence will be different from the male pelvic bone. Forensic experts claim that they can state with 95% certainty the gender of the person if the hip bone is provided and with 98% certainty if the hip bone and skull are provided.
3. Age at Death: In children, bones serve the additional function of providing a scope for growth. These centers, also called growth centers, close down, or ossify once a specific age is achieved. This age is different for different bones. If there is a full set of skeleton, there opens an option to compare them to determine the approximate age of the person at the time of death. There are other changes in the skeleton that are age dependent, such as fusion of sutures, ossification of fibrous joints, ossification of cartilages, etc. These add to the level of certainty of the age estimation. Advanced microscopy can determine the age at death by looking at the cross section of the bone and comparing calcification levels and patters.
4. Dimensions: Modern scientists had succeeded in applying this knowledge to the levels of bones to successfully derive relations between the length of the bones and the dimensions

of the person, such as height. There are formulae that can predict the height of a person to astonishing degrees of accuracies from lengths of certain bones. This will add to the knowledge for identification.

5. Direct Identification: Very rarely, bones provide direct clues to the identity of the person. Consider an example of a person who has undergone a hip replacement surgery. The head of his corresponding femur would always contain the replaced metallic part and would give strong indication to identity, if the skeleton is obtained for identification.

6. Source for DNA: We owe a lot to the developments in the field of molecular science. DNA being the basic molecule of life work as the unique unmistakable source of identity. If a body is found in burnt stage, it will be very difficult to identify physically. DNA fingerprinting comes to the rescue in such scenarios, but from where will we collect DNA on a burnt body? Depending on the level of burn, teeth or bones act as sources for DNA. Teeth have got pulp that may be protected from fire by the intrinsic properties of teeth. Similarly, bone can provide bone marrow from which DNA extraction is possible.

Establishing Time since Death - It is very important to establish the time since death of a body, as this stand as a strong evidence in court. In very old dead bodies, the bones are sometimes the only things left from decomposition and the time since death of the body should be extracted from the bones, in such cases. Carbon dating, being the usual method of estimating age of biological specimens is not very useful here, because it had been observed that there is no considerable reduction in the levels of C^{14} for bodies that are less than 100 years old. Scientists had discovered alternatives such as estimation of levels of certain isomers of nitrogen, estimation of levels of certain amino acids, etc. However, these details give only a rough estimate of the time since death and should only be considered with caution because the levels of biological parameters on a decomposing body are mostly dependent on the surroundings of decomposition.

Establishing Cause of Death -This is ultimately the reason why we do a bone examination. This is most important in cases of exhumations and re-autopsies. In countries such as India, there is no limit to the time after which one cannot do exhumation. This implies that the jurisdiction can always ask for exhumation of a body that was dead hundreds of years ago. In such scenarios, what one will encounter will be mostly bones, and a re-autopsy will consist mostly of bone examination. The details that bones can provide may not be many. It can provide the following information, to start with:

7. *Toxicological Profile:* Levels of various toxins. It is important to compare this with the levels of the same toxin in the surroundings to rule out environmental influences.

8. *Fracture Profile:* Any fracture in the bones that would have been missed in the previous

autopsy. Care should be taken to separate this from the natural disintegration of bones.

9. *Wound Profile:* Wounds such as those produced from gun shots are often deep enough to produce marks on the bones. These marks can provide useful insights into the cause of death.

10. *Organic Profile:* Scientists had succeeded in recovering the microorganisms that caused the death, from old fossilized bodies and mummies. Due to the ability of bones to provide a micro environment for the organism to fossilize, bones provide an important reservoir to investigate for the presence of causative organism.

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EXERCISES

PART A

1. Define identification
2. What are human remains? Give some examples.
3. What technique is adopted for identification of human remains?
4. List out the different types of blotting techniques.
5. What is Southern Blotting?
6. What is Northern Blotting?
7. What is Western Blotting?
8. Define primers.
9. What is biological scissors?
10. What is RFLP?
11. What is PCR?

PART B

12. What medico legal information can we have from examination of body fragments?
13. What is exhumation? What are the procedure and precaution recommended for exhumation?
14. What is identification? What are the different data for identification of a person?

15. How do you determine gender in a highly decomposed or mutilated and dismembered dead body?
16. How identification is established in an extremely decomposed or mutilated dead body?
17. What is rigor mortis? How is it formed, add a note on its forensic importance?
18. Explain steps involved in PCR analysis. Add a note on forensic importance of the same.
19. Elaborate in detail technique used to get multiple copies of DNA molecules in forensic science.
20. Explain blotting technique used to detect the DNA. Add a note on its forensic importance.
21. Explain in detail about northern blotting technique. Why named as northern blotting. Add a note on its forensic importance.
22. Explain blotting technique used to detect the antibody. Add a note on its forensic importance.

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DEPARTMENT OF BIOMEDICAL ENGINEERING

UNIT – II – BLOTTING TECHNIQUES – SBMA4007

UNIT II

IDENTIFICATION OF HUMAN REMAINS

The identification of human remains is important for both legal and humanitarian reasons. This document provides a brief overview for non experts of the scientific process by which human remains are identified. It does not set out guidelines or standard operating procedures for carrying out forensic investigations, and must not be understood as doing so. The ICRC recommends the use of standardized policies and procedures in all contexts, which should be developed by the pertinent authorities (courts, investigators, medico-legal institutes, etc.). Ed Ou/ICRC When a missing person is believed to be dead, two complementary lines of investigation must be pursued: • tracing the whereabouts of that person once he or she has been reported missing by his or her family or other reliable sources • forensic identification of human remains. The forensic identification of human remains is a legal determination (sealed by the jurisdictional authority's signature on a death certificate) based on the scientific matching of information on missing persons with unidentified human remains. Identification requires a holistic approach that takes into consideration all available scientific and contextual evidence. Each line of evidence must be weighed and treated on its merits. The scale of the identification project must also be considered and the identification strategy adjusted accordingly.

Forensic identification of human remains generally involves three main stages of investigation:

- background research
- recovery of remains
- laboratory analysis and reconciliation.

RECOVERY OF REMAINS

Proper recovery and management of remains and associated evidence (e.g. clothes, personal belongings, and other pieces of evidence) are vitally important in forensic identification and can help clarify the fate of the person in question. Ideally, forensic archaeologists should conduct this

process, especially when the circumstances of recovery are complex (clandestine burials, mass graves, extensive surface scatter as can occur in plane crashes, etc.). Their specialized skills and knowledge help ensure the proper recovery of remains and the gathering of as much additional information as possible for use in identification and other related activities. Improper recovery of remains and disrespectful handling can result in the loss of important data and may be extremely traumatic for families, and thus seriously undermine the humanitarian aims of forensic investigations.

Proper recovery makes possible, among other things: • the recovery of all biological and associated physical evidence for assistance in identification • less mixing when the remains of several individuals are found together, at the same site ("commingling") • identification of disturbed graves and differentiation between primary and secondary burials2 • less post-mortem damage to the remains • proper documentation of findings.

Broadly speaking, the recovery of remains involves three main phases: • locating the remains • mapping the remains and the entire site and documenting all relevant information • retrieving the remains properly, labelling them, and securing them for transport, which can be particularly difficult if the remains are of many different individuals. Human remains can be found in any setting, indoors (e.g. within buildings or amidst the rubble of destroyed structures) and outdoors (e.g. burial sites, on the ground, in watery surroundings, wells or caves). There are many methods and tools for finding them. However, there is, as yet, no device for detecting bones. Often, the best information about the location of remains comes from witness statements. Proper mapping and documentation make it possible to re-create the site, should it be necessary to return for any reason, and to create a physical or 'hard-copy' map (and/or an electronic one) for evidentiary, archival and analytical purposes. Mapping and documenting data, and ensuring their proper handling and management, are essential in the recovery process and should be carried out by trained individuals.

LABORATORY ANALYSIS AND RECONCILIATION

After the remains have been properly recovered, they should be sent for laboratory analysis and reconciliation, which involves answering five main questions: • Are the remains human or non-human? • Are the remains related to the conflict/ disaster/situation in question? • How many

individuals do the recovered remains represent? • Who are they? What are their IDENTITIES? • What is the cause of death? The first step in laboratory analysis and reconciliation is preparing and examining the remains. This must be done by specially trained experts (forensic pathologists, anthropologists, odontologists, etc.) who gather information – post-mortem data (PMD) – about the remains themselves and any circumstantial evidence.

PMD may include the following types of information: • general information about the remains (age range, sex, height, etc.) • medical and dental facts including unique characteristics of the remains (signs of old bone fractures or evidence of surgery, condition of the teeth and presence of any dental work such as fillings, etc.) • trauma and post-mortem damage to the remains (both intentional and accidental) • fingerprint information • DNA data • clothes and personal items found with the remains • circumstantial information about the remains (where they were found and how they came to be in that location, including witness testimony, etc.).

Methods of reconstruction

Facial reconstruction is a method used in forensic anthropology to aid in the identification of skeletal remains. The reproduction of the facial features of an individual is based upon the average soft tissue thicknesses over various anatomical sites of the skull and jaws (Krogman and Iscan 1986) and is duplicated by means of modeling clay.

There are significant differences in the thicknesses of the soft tissues of males and females of different races (Gatliff and Snow 1979; Lebedinskaya et al.1993; Phillips and Smuts 1996; Rhine and Campbell 1980; Rhine et al.1982; Steward 1954; Suzuki 1948). Various techniques have been employed to measure the thickness of the facial tissues of adults, children, and young adults (Altemus 1963; George 1987; Heglar 1980; Lebedinskaya et al. 1993; Phillips and Smuts 1996). The methods used to “flesh out” a face may vary, but each method incorporates a harmonious balance between science and art that eventually results in a reproduction of a face that may lead to identification.

Forensic facial reconstruction (or forensic facial approximation) is the process of recreating the face of an individual (whose identity is often not known) from their skeletal remains

through an amalgamation of artistry, forensic science, anthropology, osteology, and anatomy. It is easily the most subjective—as well as one of the most controversial—techniques in the field of forensic anthropology. Despite this controversy, facial reconstruction has proved successful frequently enough that research and methodological developments continue to be advanced.

Types of reconstruction

Two-dimensional facial reconstructions are based on ante mortem photographs, and the skull. Occasionally skull radiographs are used but this is not ideal since many cranial structures are not visible or at the correct scale. This method usually requires the collaboration of an artist and a forensic anthropologist. A commonly used method of 2D facial reconstruction was pioneered by Karen T. Taylor of Austin, Texas during the 1980s. Taylor's method involves adhering tissue depth markers on an unidentified skull at various anthropological landmarks, then photographing the skull. Life-size or one-to- one frontal and lateral photographic prints are then used as a foundation for facial drawings done on transparent vellum. Recently developed, the F.A.C.E. and C.A.R.E.S. computer software programs quickly produce two-dimensional facial approximations that can be edited and manipulated with relative ease. These programs may help speed the reconstruction process and allow subtle variations to be applied to the drawing, though they may produce more generic images than hand-drawn art work.

Three-dimensional reconstructions

Three-dimensional facial reconstructions are either: 1) sculptures (made from casts of cranial remains) created with modeling clay and other materials or 2) high-resolution, three-dimensional computer images. Like two-dimensional reconstructions, three-dimensional reconstructions usually require both an artist and a forensic anthropologist. Computer programs create three-dimensional reconstructions by manipulating scanned photographs of the unidentified cranial remains, stock photographs of facial features, and other available reconstructions. These computer approximations are usually most effective in victim identification because they do not appear too artificial.[5] This method has been adapted by the National Center for Missing & Exploited Children, which uses this method often to show approximations of an unidentified decedent to release to the public in hopes to identify the subject.

Superimposition

Superimposition is a technique that is sometimes included among the methods of forensic facial reconstruction. It is not always included as a technique because investigators must already have some kind of knowledge about the identity of the skeletal remains with which they are dealing (as opposed to 2D and 3D reconstructions, when the identity of the skeletal remains is generally completely unknown). Forensic superimpositions are created by superimposing a photograph of an individual suspected of belonging to the unidentified skeletal remains over an X-ray of the unidentified skull. If the skull and the photograph are of the same individual, then the anatomical features of the face should align accurate testes. Cellular sexing using barr bodies in buccal mucosal cells, drumsticks in WBC of female.

Identification of the living and the dead

1. IDENTIFICATION OF THE LIVING AND THE DEAD

State the medico-legal significance of identification 2. List the general and specific identity criteria 3. Describe and discuss each criterion used for human identification 4. State the circumstances of an exhumation.5. Briefly describe the procedure of exhumation.

2. Why identify the living? Cases of amnesia, unconscious, imposters, issue of identity cards, passports etc. Specific ages in penal code – infanticide, criminal responsibility, rape, consent, domestic employment Why identify the dead? To give information to surviving relatives, statistical and legal purposes, registration of death, for burial or cremation, discharge property, claim life insurance, hold inquests, facilitate police investigations etc. Establishment of identity may be required upon; Intact fresh corpses, decomposed corpses, mutilated and dismembered corpses and skeletalized material.

3. General identity (broad group) 1. Sex 2. Age 3. Stature 4. race

SEX male/female. But 100% incorrect if inaccurate .1.1 Presumptive evidence Outward appearance – but imposters! 1.2 Highly probable evidence External sexual characteristics Female – breasts, body hair, labia and vagina Male – body hair distribution, external genitalia 1.3 Certain evidence Female ovaries and male

6. Putrefied body / SkeletonUterus and prostate last to putrefy.Secondary sex characteristics of bonesAccuracyEntire skeleton 100%Skull 90%Pelvis 95%Long bones 80%Skull + Pelvis 98%Prepubertal material muscle ridges, larger foramina / depressions / processes / articular surfaces in males. Pelvis and Skull. 75%All bones larger, robust,

7. AGE Different periods –1. Embryo – full term2. Child who has survived birth 3. Adult Embryo – full term· Diameter, weight of ovum (1/3” in one month, 4” in three months)· Appearance, formation of limbs, sex organs, placenta· Appearance of ossific centers· Length of fetus

8. 5 - Crown rump length· Circumference of head, abdomen.· Foot length Length in centimetres 6 months to 10 months - Length in centimeters Gestational age in months Upto 5 months

9. Child who has survived birthChild recently born Changes in the umbilical cord Ring of inflammation around cord – 24 – 48 hoursDrying of cord – 4-6 dSeparation of cord from the naval – 6 – 8 d Complete healing – 10 – 12 d

10. Child who has survived birth for longer Ductus arteriosus, fetal Hb, Fontanelles Height and weightEruption of teeth Appearance of other ossification centersBone / epiphyseal unionPubertal features

11. Adults Uncertain after 25 years Premature ageing – illness, malnutrition, familial.Calcified

arteries / arcus senilis / calcification of cartilages – larynx, ribs – after 40 yearsOsteoporosis / lipping of bones / loss of teeth, attrition, root resorption – after 50 – 60 years.
12. Skeletal remainsAppearance of ossific centersUnion of epiphyses with the bonesFusion of skull sutures
13. STATURE Span = height = UL + 6" + 1" x 2 = head to pubis x 2Lengths of bones – osteometric board
14. 4. Race Caucaseans, negroes, mongoloids
16. Specific identity1. Facial features2. Clothing, personal effects3. Scars, Marks, Tattoos, deformities.4. Hair and eyes5. Finger/Palm/Foot/Lip prints6. Forensic Odontology7. Forensic Radiology8. Blood grouping, DNA fingerprinting.9. Super- impostion10.Facial reconstruction<
17. 1. change of facial contours.b) Look alikes / twinsc) Putrefaction – all look the same. Facial Features Most useful. Routinely used in post mortems – 2 persons.Problems – a) Loss of muscle tone after death
18. 2. thereafter match. Clothing, personal effects Resist decay / heat / putrefaction – useful in mass graves / exhumation / tsunamis.Especially, jewellery. – also, name/initials engraved.Problems – a) Common clothingb) Borrowed / stolenc) Wrongly believe if similar. - Therefore, relatives are first asked to describe in detail

20. 3. Scars, Marks, Tattoos, deformities. ScarsSurgical, injury, diseaseSignificance more if lot of scars / keloid formation.Multiple, parallel, fine scars over the front of wrist are consistent with previous …… - commonly seen in ..... Healed bone fractures useful in skeletonized bodies.Problemsa) Uninfected abrasions, clean cuts do not leave scars. Some scars fade away.b) Common surgical scars – appendicectomy, caesarianc) Cannot date once scar tissue is formed.
21. Marks / Stigmata Occupational -Indentation of front incisor teeth in tailors, callosities of fingers in clerks, marks in hands in violinists, guitarists. Habits -Betel chewer's stained teeth, nicotine smoker's stained teeth / fingers.
22. Tattoos More significant if rare picture / initials of name. Woman – lover's name, sexual deviants – in private parts. Persist even when putrefied. Visualized when skin peeled off (dye in dermis).Attempts to destroy, makes it more obvious. Problems Common tattoos – 'Amma', 'face of tiger'.
26. Deformities / diseases Congenital – cleft lip/palate, kyphoscoliosis Acquired – polio limb, shortened leg, healed scars of chicken pox, fungal infection under nails. Internally – scars in the brain in 'strokes', myocardial fibrosis, scars of chronic pyelonephritis, TB cavities in lungs, cirrhotic liver
27. 4. Hair and eyes Hair Examination of weapons in assaults, vehicles in hit and run RTA, scene or body of victim or assailant in sexual abuse – Locard's principle Straight, curly, wavy / coloured / length / naturally fallen or forcibly removed. Eyes Colour of iris
28. 5. Finger/Palm/Foot/Lip prints Even identical twins have different prints. Print will not alter even if tried to scar by a criminal. Decomposed body – desquamated skin can be used. 6. Forensic Odontology Dental records – decays, fillings, crown and bridge work, prostheses Bite mark - Correct description, photograph, cast preparation and comparison by forensic odontologist.
30. 7. Forensic Radiology Previous fractures or orthopaedic maneuvers.Frontal sinuspattern Problem - Previous X-rays should be available. 8. Blood grouping, DNA fingerprinting. Blood grouping - Good tool to exclude a person rather than positive identification. Red cell antigens, serum proteins, red cell enzymes, HLA system DNA – nuclei are used – WBC, sperms, hair bulbs, bone marrow Comparison technique. Expensive. Used in private lab in Sri Lanka.

33. 9. Superimposition Photo / video, Comparison 10. Facial reconstruction Soft tissue thickness 36. EXHUMATIONS Disinterment after burial. Circumstances1. Criminal Suspicious death, but buried without inquest Inquest / PM findings - wrong / inadequate Surreptitious disposal 2. Civil Identify person buried as 'unidentified'Re-interment in place of choice Burial ground to be used for other purpose Authority Magistrate written order – Section 373 (2) of Criminal P. C. Procedure a. History b. Team c. Best in morning d. Note time of arrival, those present e. Identify grave f. Photograph, sketch, describe the grave g. Map out area of digging, dig, measurements h. Soil samples i. Coffin lifted up j. Coffin identified by undertaker, sample of embalming fluid k. Body identified l. Transport to mortuary m. PM by experienced, qualified Forensic Pathologist.

DNA fingerprinting

DNA fingerprinting is a laboratory technique used to establish a link between biological evidence and a suspect in a criminal investigation. A DNA sample taken from a crime scene is compared with a DNA sample from a suspect. If the two DNA profiles are a match, then the evidence came from that suspect.

Why Use DNA Fingerprinting?

This process is used as one means of identification when an attacker or assailant has left some kind of bodily fluid or blood at the scene of a crime and when no visual identification is possible.

DNA - or genetic - fingerprinting relies heavily on the principle that no two individuals share the same genetic code - except for identical twins and statistically those elements of DNA that are examined and used to obtain a match will be unique.

The process of DNA fingerprinting was first used during the 1980's and its application was quickly to become that of identification of suspects involved in serious crimes including murder. The premise that most attackers or killers will leave some measure of bodily fluid at a crime scene - be it saliva, blood, semen or other such fluid - was quickly accepted as common place and it became a staple of many criminal investigations.

Collecting Evidence

DNA fingerprinting works on the basis that each individual's DNA structure - or genetic make-up - is unique and therefore cannot be forged, faked or altered in any way. Just like normal fingerprints taken from a suspect they are so unique that only in the case of identical twins - as we have already mentioned - could they be the same.

Developed by Professor of Genetics Sir Alec Jeffreys, the process begins with a sample of an individual's DNA (typically called a "reference sample"). A common method of collecting a reference sample is the use of a buccal swab, which is easy, non-invasive and cheap. When this is not available (e.g. because a court order is needed but not obtainable) other methods may need to be used to collect a sample of blood, saliva, semen, or other appropriate fluid or tissue

from personal items (e.g. a toothbrush, razor) or from stored samples (e.g. banked sperm or biopsy tissue). Samples obtained from blood relatives (related by birth, not marriage) can provide an indication of an individual's profile, as could human remains that had been previously profiled.

A reference sample is then analyzed to create the individual's DNA profile using one of a number of techniques, discussed below. The DNA profile is then compared against another sample to determine whether there is a genetic match.

RFLP analysis

The first methods for finding out genetics used for DNA profiling involved RFLP analysis. DNA is collected from cells, such as a blood sample, and cut into small pieces using a restriction enzyme (a restriction digest). This generates thousands of DNA fragments of differing sizes as a consequence of variations between DNA sequences of different individuals. The fragments are then separated on the basis of size using gel electrophoresis.

The separated fragments are then transferred to a nitrocellulose or nylon filter; this procedure is called a Southern blot. The DNA fragments within the blot are permanently fixed to the filter, and the DNA strands are denatured. Radiolabeled probe molecules are then added that are complementary to sequences in the genome that contain repeat sequences. These repeat sequences tend to vary in length among different individuals and are called variable number tandem repeat sequences or VNTRs. The probe molecules hybridize to DNA fragments containing the repeat sequences and excess probe molecules are washed away. The blot is then exposed to an X-ray film. Fragments of DNA that have bound to the probe molecules appear as dark bands on the film.

The Southern blot technique is laborious, and requires large amounts of undegraded sample DNA. Also, Karl Brown's original technique looked at many minisatellite loci at the same time, increasing the observed variability, but making it hard to discern individual alleles (and thereby precluding paternity testing). These early techniques have been supplanted by PCR-based assays.

PCR analysis

Developed by Kary Mullis in 1983, a process was reported by which specific portions of the sample DNA can be amplified almost indefinitely (Saiki et al. 1985, 1988). This has revolutionized the whole field of DNA study. The process, the polymerase chain reaction (PCR), mimics the biological process of DNA replication, but confines it to specific DNA sequences of interest. With the invention of the PCR technique, DNA profiling took huge strides forward in both discriminating power and the ability to recover information from very small (or degraded) starting samples.

PCR greatly amplifies the amounts of a specific region of DNA. In the PCR process, the DNA sample is denatured into the separate individual polynucleotide strands through heating. Two oligonucleotide DNA primers are used to hybridize to two corresponding nearby sites on opposite DNA strands in such a fashion that the normal enzymatic extension of the active terminal of each primer (that is, the 3’ end) leads toward the other primer. PCR uses replication enzymes that are

tolerant of high temperatures, such as the thermostable Taq polymerase. In this fashion, two new copies of the sequence of interest are generated. Repeated denaturation, hybridization, and extension in this fashion produce an exponentially growing number of copies of the DNA of interest. Instruments that perform thermal cycling are now readily available from commercial sources. This process can produce a million-fold or greater amplification of the desired region in 2 hours or less.

Early assays such as the HLA-DQ alpha reverse dot blot strips grew to be very popular due to their ease of use, and the speed with which a result could be obtained. However, they were not as discriminating as RFLP analysis. It was also difficult to determine a DNA profile for mixed samples, such as a vaginal swab from a sexual assault victim.

However, the PCR method was readily adaptable for analyzing VNTR, in particular STR loci. In recent years, research in human DNA quantitation has focused on new "real-time" quantitative PCR (qPCR) techniques. Quantitative PCR methods enable automated, precise, and high-throughput measurements. Interlaboratory studies have demonstrated the importance of human DNA quantitation on achieving reliable interpretation of STR typing and obtaining consistent results across laboratories.

Blotting techniques

A blotting techniques, in molecular biology and genetics, is a method of transferring proteins, DNA or RNA, onto a carrier (for example, a nitrocellulose PVDF or nylon membrane). In many instances, this is done after a gel electrophoresis, transferring the molecules from the gel onto the blotting membrane, and other times adding the samples directly onto the membrane. After the blotting, the transferred proteins, DNA or RNA are then visualized by colorant staining (for example, silver staining of proteins), autoradiographic visualization of radioactive labelled molecules (performed before the blot), or specific labelling of some proteins or nucleic acids. The latter is done with antibodies or hybridization probes that bind only to some molecules of the blot and have an enzyme joined to them. After proper washing, this enzymatic activity (and so, the molecules we search in the blot) is visualized by incubation with proper reactive, rendering either a colored deposit on the blot or a chemiluminiscent reaction which is registered by photographic film.

Southern blot

A Southern blot is a method routinely used in molecular biology for detection of a specific DNA sequence in DNA samples. Southern blotting combines transfer of electrophoresis-separated DNA fragments to a filter membrane and subsequent fragment detection by probe hybridization.

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The method is named after its inventor, the British biologist Edwin Southern.

Other blotting methods (i.e., western blot, northern blot, eastern blot, southwestern blot) that employ similar principles, but using RNA or protein, have later been named in reference to Edwin Southern's name. As the label is eponymous, Southern is capitalised, as is conventional for proper nouns. The names for other blotting methods may follow this convention, by analogy

Method

1. Restriction endonucleases are used to cut high-molecular-weight DNA strands into smaller fragments.

2. The DNA fragments are then electrophoresed on an agarose gel to separate them by size.

3. If some of the DNA fragments are larger than 15 kb, then prior to blotting, the gel may be treated with an acid, such as dilute HCl. This depurinates the DNA fragments, breaking the DNA into smaller pieces, thereby allowing more efficient transfer from the gel to membrane.

4. If alkaline transfer methods are used, the DNA gel is placed into an alkaline solution (typically containing sodium hydroxide) to denature the double- stranded DNA. The denaturation in an alkaline environment may improve binding of the negatively charged thymine residues of DNA to a positively charged amino groups of membrane, separating it into single DNA strands for later hybridization to the probe (see below), and destroys any residual RNA that may still be present in the DNA. The choice of alkaline over neutral transfer methods, however, is often empirical and may result in equivalent results.

5. A sheet of nitrocellulose (or, alternatively, nylon) membrane is placed on top of (or below, depending on the direction of the transfer) the gel. Pressure is applied evenly to the gel (either using suction, or by placing a stack of paper towels and a weight on top of the membrane and gel), to ensure good and even contact between gel and membrane. If transferring by suction, 20X SSC buffer is used to ensure a seal and prevent drying of the gel. Buffer transfer by capillary action from a region of high water potential to a region of low water potential (usually filter paper and paper tissues) is then used to move the DNA from the gel onto the membrane; ion exchange interactions bind the DNA to the membrane due to the negative charge of the DNA and positive charge of the membrane.

6. The membrane is then baked in a vacuum or regular oven at 80 °C for 2 hours (standard conditions; nitrocellulose or nylon membrane) or exposed to ultraviolet radiation (nylon membrane) to permanently attach the transferred DNA to the membrane.

7. The membrane is then exposed to a hybridization probe—a single DNA fragment with a specific sequence whose presence in the target DNA is to be determined. The probe DNA is labelled so that it can be detected, usually by incorporating radioactivity or tagging

the molecule with a fluorescent or chromogenic dye. In some cases, the hybridization probe may be made from RNA, rather than DNA. To ensure the specificity of the binding of the probe to the sample DNA, most common hybridization methods use salmon or herring sperm DNA for blocking of the membrane surface and target DNA, deionized formamide, and detergents such as SDS to reduce non-specific binding of the probe.

8. After hybridization, excess probe is washed from the membrane (typically using SSC buffer), and the pattern of hybridization is visualized on X-ray film by autoradiography in the case of a radioactive or fluorescent probe, or by development of colour on the membrane if a chromogenic detection method is used.

Hybridization of the probe to a specific DNA fragment on the filter membrane indicates that this fragment contains DNA sequence that is complementary to the probe. The transfer step of the DNA from the electrophoresis gel to a membrane permits easy binding of the labeled hybridization probe to the size-fractionated DNA. It also allows for the fixation of the target-probe hybrids, required for analysis by autoradiography or other detection methods. Southern blots performed with restriction enzyme-digested genomic DNA may be used to determine the number of sequences (e.g., gene copies) in a genome. A probe that hybridizes only to a single DNA segment that has not been cut by the restriction enzyme will produce a single band on a Southern blot, whereas multiple bands will likely be observed when the probe hybridizes to several highly similar sequences (e.g., those that may be the result of sequence duplication). Modification of the hybridization conditions (for example, increasing the hybridization temperature or decreasing salt concentration) may be used to increase specificity and decrease hybridization of the probe to sequences that are less than 100% similar.

Applications

Southern transfer may be used for homology-based cloning on the basis of amino acid sequence of the protein product of the target gene. Oligonucleotides are designed that are similar to the target sequence. The oligonucleotides are chemically synthesised, radiolabeled, and used to screen a DNA library, or other collections of cloned DNA fragments. Sequences that hybridize with the hybridization probe are further analysed, for example, to obtain the full length sequence of the targeted gene.

Southern blotting can also be used to identify methylated sites in particular genes. Particularly useful are the restriction nucleases *MspI* and *HpaII*, both of which recognize and cleave within the same sequence. However, *HpaII* requires that a C within that site be methylated, whereas *MspI* cleaves only DNA unmethylated at that site. Therefore, any methylated sites within a sequence analyzed with a particular probe will be cleaved by the former, but not the latter, enzyme.

Western blot

The western blot (sometimes called the protein immunoblot) is a widely used analytical technique used to detect specific proteins in a sample of tissue homogenate or extract. It uses gel

electrophoresis to separate native proteins by 3-D structure or denatured proteins by the length of the polypeptide. The proteins are then transferred to a membrane (typicallynitrocellulose or PVDF), where they are stained with antibodies specific to the target protein.[1][2] The gel electrophoresis step is included in western blot analysis to resolve the issue of the cross-reactivity of antibodies.

There are many reagent companies that specialize in providing antibodies (both monoclonal and polyclonal antibodies) against tens of thousands of different proteins.[3] Commercial antibodies can be expensive, although the unbound antibody can be reused between experiments. This method is used in the fields of molecular biology, immunogenetics and other molecular biology disciplines. A number of search engines, such as CiteAb, Antibodypedia, and SeekProducts, are available that can help researchers find suitable antibodies for use in western blotting.

Gel electrophoresis

The proteins of the sample are separated using gel electrophoresis. Separation of proteins may be by isoelectric point (pI), molecular weight, electric charge, or a combination of these factors. The nature of the separation depends on the treatment of the sample and the nature of the gel. This is a very useful way to identify a protein.

By far the most common type of gel electrophoresis employs polyacrylamide gels and buffers loaded with sodium dodecyl sulfate (SDS). SDS-PAGE (SDS polyacrylamide gel electrophoresis) maintains polypeptides in a denatured state once they have been treated with strong reducing agents to remove secondary and tertiary structure (e.g. disulfide bonds [S-S] to sulfhydryl groups [SH and SH]) and thus allows separation of proteins by their molecular weight. Sampled proteins become covered in the negatively charged SDS and move to the positively charged electrode through the acrylamide mesh of the gel. Smaller proteins migrate faster through this mesh and the proteins are thus separated according to size (usually measured in kilodaltons, kDa). The concentration of acrylamide determines the resolution of the gel - the greater the acrylamide concentration, the better the resolution of lower molecular weight proteins. The lower the acrylamide concentration, the better the resolution of higher molecular weight proteins. Proteins travel only in one dimension along the gel for most blots.

Samples are loaded into *wells* in the gel. One lane is usually reserved for a *marker* or *ladder*, a commercially available mixture of proteins having defined molecular weights, typically stained so as to form visible, coloured bands. When voltage is applied along the gel, proteins migrate through it at different speeds dependent on their size. These different rates of advancement (different electrophoretic mobilities) separate into *bands* within each *lane*.

It is also possible to use a two-dimensional (2-D) gel which spreads the proteins from a single sample out in two dimensions. Proteins are separated according to isoelectric point (pH at which they have neutral net charge) in the first dimension, and according to their molecular weight in the

second dimension.

Transfer

In order to make the proteins accessible to antibody detection, they are moved from within the gel onto a membrane made of nitrocellulose *or* polyvinylidene difluoride *(PVDF*). The primary method for transferring the proteins is called electroblotting and uses an electric current to pull proteins from the gel into the PVDF or nitrocellulose membrane. The proteins move from within the gel onto the membrane while maintaining the organization they had within the gel. An older method of transfer involves placing a membrane on top of the gel, and a stack of filter papers on top of that. The entire stack is placed in a buffer solution which moves up the paper by capillary action, bringing the proteins with it. In practice this method is not used as it takes too much time; electroblotting is preferred. As a result of either "blotting" process, the proteins are exposed on a thin surface layer for detection. Both varieties of membrane are chosen for their non-specific protein binding properties (i.e. binds all proteins equally well). Protein binding is based upon hydrophobic interactions, as well as charged interactions between the membrane and protein. Nitrocellulose membranes are cheaper than PVDF, but are far more fragile and do not stand up well to repeated probings.

Northern blot - The northern blot is a technique used in molecular biology research to study gene expression by detection of RNA (or isolated mRNA) in a sample.

A northern blot is a laboratory method used to detect specific RNA molecules among a mixture of RNA. Northern blotting can be used to analyze a sample of RNA from a particular tissue or cell type in order to measure the RNA expression of particular genes. This method was named for its similarity to the technique known as a Southern blot.

The first step in a northern blot is to denature, or separate, the RNA within the sample into single strands, which ensures that the strands are unfolded and that there is no bonding between strands. The RNA molecules are then separated according to their sizes using a method called gel electrophoresis. Following separation, the RNA is transferred from the gel onto a blotting membrane. (Although this step is what gives the technique the name "northern blotting," the term is typically used to describe the entire procedure.) Once the transfer is complete, the blotting membrane carries all of the RNA bands originally on the gel. Next, the membrane is treated with a small piece of DNA or RNA called a probe, which has been designed to have a sequence that is complementary to a particular RNA sequence in the sample; this allows the probe to hybridize, or bind, to a specific RNA fragment on the membrane. In addition, the probe has a label, which is typically a radioactive atom or a fluorescent dye. Thus, following hybridization, the probe permits the RNA molecule of interest to be detected from among the many different RNA molecules on the membrane.

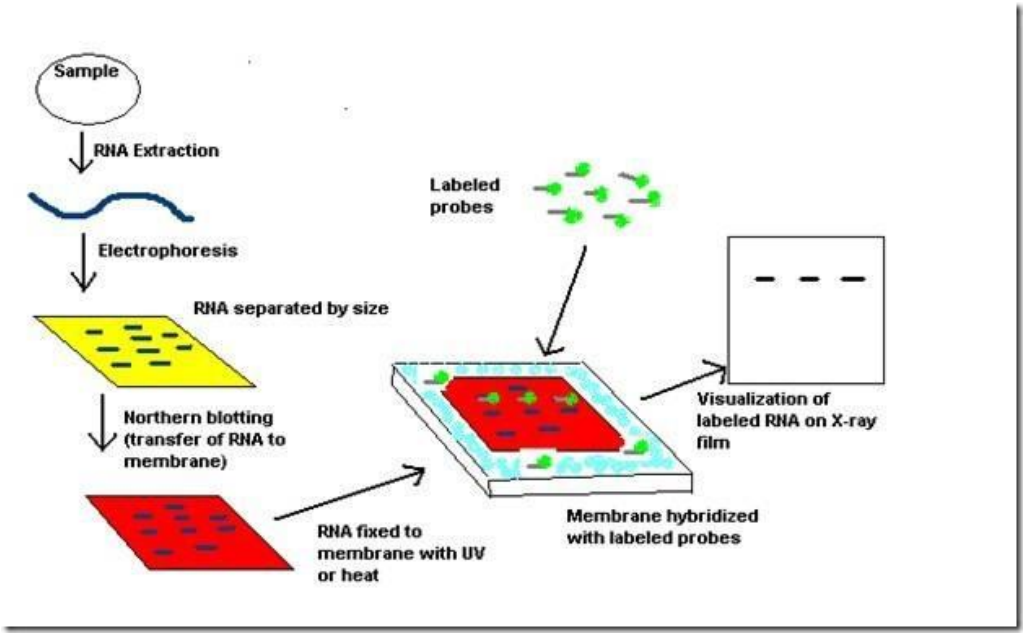

Fig. 2.1: Northern Blot

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EXERCISES

PART A

1. Define identification
2. What are human remains? Give some examples.
3. What technique is adopted for identification of human remains?
4. List out the different types of blotting techniques.
5. What is Southern Blotting?
6. What is Northern Blotting?
7. What is Western Blotting?
8. Define primers.
9. What is biological scissors?
10. What is RFLP?
11. What is PCR?

PART B

12. What medico legal information can we have from examination of body fragments?
13. What is exhumation? What are the procedure and precaution recommended for exhumation?
14. What is identification? What are the different data for identification of a person?
15. How do you determine gender in a highly decomposed or mutilated and dismembered dead body?
16. How identification is established in an extremely decomposed or mutilated dead body?
17. What is rigor mortis? How is it formed, add a note on its forensic importance?
18. Explain steps involved in PCR analysis. Add a note on forensic importance of the same.
19. Elaborate in detail technique used to get multiple copies of DNA molecules in forensic science.
20. Explain blotting technique used to detect the DNA. Add a note on its forensic importance.
21. Explain in detail about northern blotting technique. Why named as northern blotting. Add a note on its forensic importance.
22. Explain blotting technique used to detect the antibody. Add a note on its forensic importance.

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UNIT – III – FINGER PRINTS – SBMA4007

Unit - III

Fingerprints are the tiny ridges, whorls and valley patterns on the tip of each finger. They form from pressure on a baby's tiny, developing fingers in the womb. No two people have been found to have the same fingerprints -- they are totally unique. There's a one in 64 billion chance that your fingerprint will match up exactly with someone else's.

Fingerprints are even more unique than DNA, the genetic material in each of our cells. Although identical twins can share the same DNA -- or at least most of it -- they can't have the same fingerprints.

A fingerprint in its narrow sense is an impression left by the friction ridges of a human finger. The recovery of fingerprints from a crime scene is an important method of forensic science. Fingerprints are easily deposited on suitable surfaces (such as glass or metal or polished stone) by the natural secretions of sweat from the eccrine glands that are present in epidermal ridges. These are sometimes referred to as "Chanced Impressions".

Finger print analysis

Where Fingerprints May be Found

Fingerprints can be found on practically any solid surface, including the human body. Analysts classify fingerprints into three categories according to the type of surface on which they are found and whether they are visible or not: Fingerprints on soft surfaces (such as soap, wax, wet paint, fresh caulk, etc.) are likely to be three-dimensional plastic prints; those on hard surfaces are either patent (visible) or latent (invisible) prints. Visible prints are formed when blood, dirt, ink, paint, etc., is transferred from a finger or thumb to a surface. Patent prints can be found on a wide variety of surfaces: smooth or rough, porous (such as paper, cloth or wood) or nonporous (such as metal, glass or plastic).

Latent prints are formed when the body’s natural oils and sweat on the skin are deposited onto another surface. Latent prints can be found on a variety of surfaces; however, they are not readily visible and detection often requires the use of fingerprint powders, chemical reagents or alternate light sources. Generally speaking, the smoother and less porous a surface is, the greater the potential that any latent prints present can be found and developed.

How Fingerprints are Collected

Patent prints are collected using a fairly straightforward method: photography. These prints are photographed in high resolution with a forensic measurement scale in the image for reference. Investigators can improve the quality of the images by using low- angle or alternate light sources and/or certain chemicals or dyes during photography, but this is usually not necessary

However, fingerprint powders can contaminate the evidence and ruin the opportunity to perform other techniques that could turn up a hidden print or additional information. Therefore, investigators may examine the area with an alternate light source or apply cyanoacrylate (super glue) before using powders Alternate Light Source (ALS): It is becoming more commonplace for investigators to examine any likely surfaces (doors, doorknobs, windows, railings, etc.) with an alternate light source. These are laser or LED devices that emit a particular wavelength, or spectrum, of light. Some devices have different filters to provide a variety of spectra that can be photographed or further processed with powders or dye stains. For example, investigators may use a blue light with an orange filter to find latent prints on desks, chairs, computer equipment or other objects at the scene of a break-in

Cyanoacrylate: Investigators often perform cyanoacrylate (superglue) processing, or fuming, of a surface before applying powders or dye stains. This process, typically performed on non-porous surfaces, involves exposing the object to cyanoacrylate vapors. The vapors (fumes) will adhere to any prints present on the object allowing them to be viewed with oblique ambient light or a white light source.

Chemical Developers: Porous surfaces such as paper are typically processed with chemicals, including ninhydrin and physical developer, to reveal latent fingerprints. These chemicals react with specific components of latent print residue, such as amino acids and inorganic salts. Ninhydrin causes prints to turn a purple color, which makes them easily photographed. DFO (1,2-diazafluoren-9-one) is another chemical used to locate latent fingerprints on porous surfaces; it causes fingerprints to fluoresce, or glow, when they are illuminated by blue-green light.

Other Collection Methods: In addition to the methods identified above, there are special techniques for capturing prints from skin, clothing and other difficult surfaces. Amido Black, a non-specific protein stain that reacts with any protein present, is typically used for developing or enhancing bloody impressions on human skin. To reveal prints on clothing, high-tech methods such as vacuum metal deposition using gold and zinc

are showing promise for the investigator. AccuTrans®, a liquid casting compound, can be used to lift powdered latent prints from rough, textured or curved surfaces. AccuTrans® is basically a very thick liquid that fills in the nooks and crannies of rough or textured areas where conventional print lifting tape encounters difficulty.

Like fingerprint powders, chemical processing can reduce the investigator's ability to perform other techniques that could reveal valuable information. Therefore, any nondestructive investigations are performed before the evidence is treated with chemicals. For example, a ransom or hold-up note will be examined by a questioned documents expert before being treated with ninhydrin, since some formulations of ninhydrin will cause certain inks to run, thus destroying the writing.

Who Conducts the Analysis

In criminal justice cases, computerized systems are used to search various local, state and national fingerprint databases for potential matches. Many of these systems provide a value indicating how close the match is, based on the algorithm used to perform the search. Fingerprint examiners then review the potential matches and make a final determination.

Fingerprint examinations may be conducted by forensic scientists, technicians or police officers; however, the examiner should have the proper training and experience to perform the task. Currently many agencies require new examiners to have a four-year degree in science (biology, chemistry or physics). In addition, agencies may require examiners to become certified by the International Association for Identification (IAI).

How and Where the Analysis is Performed

Fingerprint analysis is usually performed by law enforcement agencies or crime laboratories; however, casework may be sent to private companies if there is a need, such as to reduce backlogs, verify results, or handle high-profile cases.

Fingerprint examination involves looking at the quality and quantity of information in order to find agreement or disagreement between the unknown print (from the crime scene) and known prints on file. To conduct the examination, fingerprint examiners use a small magnifier called a loupe to view minute details (minutiae) of a print. A pointer called a ridge counter is used to count the friction ridges.

Analysis involves assessing a print to determine if it can be used for a comparison. If the print is not suitable for comparison because of inadequate quality or quantity of features, the examination ends and the print is reported as not suitable. If the print is suitable, the analysis indicates the features to be used in the comparison and their tolerances (the amount of variation that will be accepted). The analysis may also uncover physical features such as recurves, deltas, creases and scars that help indicate where to begin the comparison.

Comparisons are performed by an analyst who views the known and suspect prints side-by-side. The analyst compares minutiae characteristics and locations to determine if they match. Known prints are often collected from persons of interest, victims, others present at the scene or through a search of one or more fingerprint databases such as the FBI's Integrated Automated Fingerprint.

Evaluation is where the examiner ultimately decides if the prints are from the same source (identification or individualization), different sources (exclusion) or is inconclusive. Inconclusive results may be due to poor quality samples, lack of comparable areas, or insufficient number of corresponding or dissimilar features to be certain.

Verification is when another examiner independently analyzes, compares and evaluates the prints to either support or refute the conclusions of the original examiner. The examiner may also verify the suitability of determinations made in the analysis phase.

Types of Fingerprints

Over the past sixty years, television has always been a reflection of our society. The programs are an indication of the norms, values and interests that society holds dear at any given time. Crime shows, detective stories and police tales have historically been a staple of the country's television viewing habit, and the country has always had a fascination with these kinds of shows. Today's programs, such as the most popular TV program over the last five years, C.S.I., bring a level of sophistication to the viewing audience that producers of the early crime shows, such as "Peter Gunn" and "Seventy- Seven Sunset Strip," never dreamed was possible. America has always had a fascination with the solving of crimes, and fingerprints are one of the most common types of evidence that investigators search for at crime scenes.

One of the main tasks of the crime scene investigator is to recover fingerprint impressions in order that a positive identification can be ascertained. Since no two individuals have the same fingerprint pattern and these remain unaltered during the course of a person's lifetime, the main type of physical evidence that can be extracted from a crime scene are fingerprints.

There are three distinct types of fingerprint impressions that can be recovered from a crime scene or a scene of interest for investigators looking for some clues as to a missing person, or for other identification purposes. These categories are as follows:

PATENT PRINTS - are visible prints that occur when a foreign substance on the skin of a finger comes in contact with the smooth surface of another object. These prints leave a distinct ridge impression that is visible with the naked eye without technological enhancement of any kind. The tried and true "blood on his hands" evidence is an example of patent prints recovered from a crime scene or scene of interest to investigators. These foreign substances contain dust particles which adhere to the ridges of the fingers and are easily identifiable when left on an object.

PLASTIC PRINTS - are visible, impressed prints that occur when a finger touches a soft, malleable surface resulting in an indentation. Some surfaces that may contain this type of fingerprint are those that are freshly painted or coated, or those that contain wax, gum, blood or any other substance that will soften when hand held and then retain the finger ridge impressions. These prints require no enhancement in order to be viewed, because they are impressed onto an object and are easily observable.

LATENT PRINTS - are fingerprint impressions secreted in a surface or an object and are usually invisible to the naked eye. These prints are the result of perspiration which is derived from sweat pores found in the ridges of fingers. When fingers touch other body parts, moisture, oil and grease adhere to the ridges so that when the fingers touch an object, such as a lamp, a film of these substances may be transferred to that object. The impression left on the object leaves a distinct outline of the ridges of that finger. These fingerprints must be enhanced upon collection and, because they serve as a means of identifying the source of the print, they have proven to be extremely valuable over the years in the identification of its source.

Now that we have categorized the various types of fingerprints, let's determine if we were crime scene investigators, could we differentiate among the fingerprint types? If you were a crime scene investigator or an investigator of a scene of interest, what type of fingerprints would you have discovered in these cases?

1. A Hershey's chocolate bar
2. A bloody print on a knife
3. A baseball helmet

1. Plastic prints because the chocolate bar softens when held and the ridges of the finger are present and visible to the naked eye.
2. Patent prints because a foreign substance, namely blood, has left a visible impression on an

object, namely the knife, which is visible to the naked eye.

3. Latent prints because the helmet must be examined and the surface of the helmet technologically enhanced in order for the fingerprints to be viewed. Some techniques available to provide for identification of these types of prints are lasers, powders and various light sources.

The knowledge of the different types of fingerprints is invaluable to investigators in their quest to identify the source of the fingerprints, and the science of fingerprints is fascinating to the lay person. For investigators, fingerprints can provide invaluable clues as they serve as a means of identifying the source of the print. Because the seasoned investigator has a thorough knowledge of the different types of fingerprints, he is able to recover them for use as evidence or for other purposes.

Techniques for collecting and analyzing finger prints

Fingerprint evidence left behind by a suspect or victim may identify who was at a crime scene and what he or she touched. However, it is important for defense attorneys to know, and to inform the jury, that the techniques used to locate and identify fingerprints are far from a perfect science. An understanding of how fingerprints are located and lifted can help attorneys recognize if a flawed analysis was performed by investigators or lab technicians. Further, knowledge of the various fingerprint collection techniques is essential to successful cross-examination of crime scene technicians and fingerprint examiners. This post attempts to provide an overview of the techniques used to locate, lift, and identify a fingerprint.

Step 1: Locating the fingerprint

Locating a fingerprint often requires a vigilant and calculated search. However, in circumstances where the print is visible to the naked eye, finding a fingerprint is relatively easy. The more intricate searches take place when the print is present on a surface but not visible. The type of fingerprint left behind usually determines the amount of time and effort investigators must put into locating the print.

According to *Forensic Science*, there are three types of fingerprints.

Patent prints are easy to locate since they are visible to the naked eye. Patent prints occur when someone has a substance on their fingers such as grease, paint, blood, or ink that leaves a visible print on a surface.

- Plastic prints are also easy to locate but are less common than patent prints since they occur when someone touches an object such as wax, butter, or soap and leaves a three-dimensional impression of the finger on the object.

- Latent prints are the most common type of print and take the most effort to locate since they are invisible. Latent prints occur when someone touches any porous or nonporous surface. The natural oils and residue on fingers leave a deposit on surfaces which mirror the ridges and furrows that are present on the individual's finger.

Investigators often follow a two-phase process when searching for fingerprints. The first phase involves looking for patent and plastic prints since they are visible. Often times, a flashlight is used during this phase. To narrow the search, investigators usually focus on the entry and exits points that the suspect used and any items that appear to have been disturbed, such as overturned lamps or possible weapons.

The type of surface being searched for fingerprints often determines the technique employed by investigators.

Nonporous Surfaces:

A powder technique is usually used to identify latent prints on nonporous surfaces such as glass, marble, metal, plastic, and finished wood. *Id.* When powder is distributed on the surface, it adheres to the residue deposited from the finger's touch, allowing investigators to find the print. Often times, to avoid smudging the print, a magnetic powder technique is used in which the powder is poured on the surface and then spread evenly over the surface using a magnetic force instead of spreading the powder with a brush. The color of the powder should contrast with the surface that is being searched to allow better visibility. For example, the investigator should use a white or grey powder if searching a black marble countertop for prints.

Attorneys should find out whether the crime scene technician who collected prints using fingerprint powder used a disposable brush. If a brush is reused in different locations at a crime scene or reused at another crime scene, the brush can transfer trace amounts of DNA evidence.

Another popular technique for fingerprint location and identification used by both lab technicians and investigators at the crime scene is superglue fuming. Superglue fuming is a chemical process that exposes and fixes fingerprints on a nonporous surface. In the lab, the process works by using an airtight tank, known as a fuming chamber, to heat up superglue (liquid cyanoacrylate) which releases gases that adhere to the oily residue of print, thereby creating an image of the fingerprint. Superglue fuming can also be performed at the crime scene. Rather than using a fuming chamber, crime scene investigators may use a handheld wand that heats up superglue and a florescent dye. Superglue fuming performed at the crime scene can be vital to preserve prints on items that are being sent to the lab via mail. One of the drawbacks is that if the evidence is fumed too long, it can distort the print, rendering it useless. To read the procedure used by North Carolina State Crime

Lab to conduct superglue fuming in a fuming chamber. To read the procedure used by the North Carolina State Crime Lab to conduct superglue fuming using a portable wand.

Porous Surfaces:

The powder technique is not as effective on porous surfaces such as fabric, unfinished wood, and paper. Instead, investigators often use chemical methods to locate the print such as iodine fuming, silver nitrate, or ninhydrin. When one of these chemicals comes into contact with the chemicals present in the fingerprint residue (natural oils, fats), the print become visual.

Iodine fuming takes place in a fuming chamber. The process works by heating up solid crystal iodine which creates vapors that adhere to the oily residue of print, producing a brown colored print. One of the drawbacks of using iodine fuming is that the print fades quickly after the fuming takes place and therefore must be photographed quickly. Alternatively, if the print is sprayed with a starch and water solution, it can be preserved for several weeks.

Silver nitrate, when exposed to latent prints, reacts with the chloride of the salt molecules found in print residue, forming silver chloride. When exposed to ultraviolet light, silver chloride turns black or brown, making the print visible *Id.* This method works particularly well on impressions left in cardboard and paper-like surfaces. Ninhydrin is more commonly used than iodine fuming and silver nitrate techniques to locate a latent print. *Id.* The object on which the print is located can be dipped in or sprayed with a ninhydrin solution, which reacts with the oils in the print's residue to create a bluish print. One of the drawbacks of using ninhydrin is that the reaction is very slow, often taking several hours for the print to become visible. *Id.* To accelerate the reaction, the object containing the print can be heated to 80 to 100 degrees Fahrenheit. *Id.* To read the North Carolina State Crime Lab's procedures for ninhydrin, A variety of other techniques are sometimes used. For example, laser illumination creates a contrast between the print and the surface which exposes the print.

Human Skin:

Locating and identifying fingerprints left on human skin is incredibly difficult. According to *Scientific Evidence*, the first major obstacle is finding the print since the oily residue left by fingers that creates the fingerprint itself is often present on human skin, making it difficult to create a contrast between the surface (skin) and the print. Further, after a print is left on human skin, the oily residue often disperses and is absorbed into the skin, blurring the print. Two hours is the maximum amount of time that a print on skin may be viable.

Textured Surfaces:

Surfaces that are not flat or have a rough surface, such as a painting with brush strokes or a golf ball will make the process of identifying and collecting fingerprints more difficult, but not impossible.

Step 2: Photographing the fingerprint

After the print is located, it is vital that it is photographed before it is lifted. A photograph captures where the print was located in comparison to other objects and captures the orientation of the print. Further, a photograph can serve as a key piece of identification of a patent or plastic print and can be used to compare and possibly match the print to its source. Photographing the print's location at the crime scene also guards against tampering of evidence.

Step 3: Lifting the fingerprint

"Lifting a fingerprint" means to make a permanent impression of the fingerprint. Lifting a print can be accomplished on either flat surfaces or round surfaces. Lifting a print usually involves a rubber tape with an adhesive surface which is applied to the fingerprint, leaving an imprint on the tape. Often times, a flat object, such as a ruler, will be slowly swiped across the top of the tape to ensure that there are no bubbles or ripples in the tape that will affect the imprint. Next, the tape is carefully peeled off the surface and a plastic cover is placed on the adhesive side of the tape to prevent disruption of the print. Identification information and a description of the location of the print should be written on the back of the tape or card. After the print is lifted, it is converted into digital data that can be modified to create a clearer image.

Step 4: Comparing the fingerprint

The final step involves a close examination of the characteristics of the fingerprints. The fingerprint examination process utilizes the ACE-V method which stands for Analysis, Comparison, Evaluation and Verification to compare a print collected from a crime scene to a set of known prints. This post will not address critiques of the ACE-V method, but additional information that can be used to challenge this technique in court can be found here.

A system called the Automated Fingerprint Identification System (AFIS) was created to find a match to the print using a computer database.

Fingerprint classification

Definition Fingerprint classification is a procedure in which fingerprints are grouped in a consistent and reliable way, such that different impressions of a same finger fall into a same group. It can be

viewed as a coarse-level pre-matching procedure so that a query fingerprint needs to be further compared with only a smaller subset of fingerprints in the database belonging to the same group. It is often necessary to integrate a classification module into a fingerprint identification system to speed up the database search. A database can be partitioned into ▸ human-interpretable fingerprint classes based on Galton–Henry scheme or into ▸ machine-generated fingerprint classes.

The identification of a person requires the comparison of his/her fingerprint with all the fingerprints in a database, which in large scale applications may be very large (several million fingerprints). A common strategy to reduce the number of comparisons during fingerprint retrieval and, consequently, to improve the response time of the identification process, is to divide the fingerprints into some predefined classes.

Fingerprint classification means assigning each fingerprint to a class in a consistent and reliable way, such that an unknown fingerprint to be searched, needs to be compared only with the subset of fingerprints in the database belonging to the same class. While fingerprint matching is usually performed according to fingerprint micro-features, such as ridge terminations and bifurcations (minutiae), fingerprint classification is usually based on macro-features, such as global ridge structure.

All the classification schemes currently used by police agencies are variants of the so- called Henry's classification scheme. Five classes (Arch, Tented arch, Left loop, Right loop and Whorl) are commonly used by today's fingerprint classification techniques. In reality, fingerprints are not uniformly distributed among these five classes: the proportions have been estimated as 3.7%, 2.9%, 33.8%, 31.7% and 27.9% for Arch, Tented arch, Left loop, Right loop and Whorl, respectively.

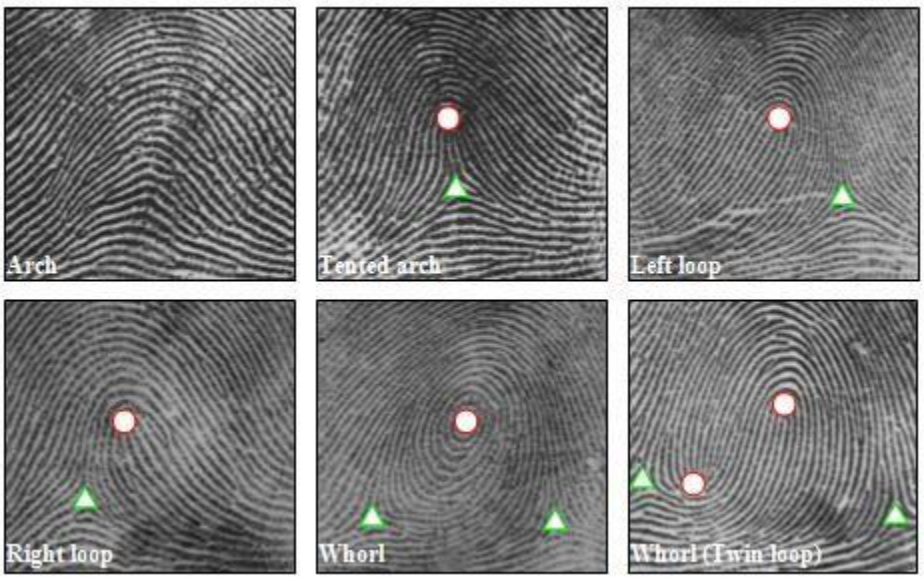


Fig 3.1: Fingerprint classification techniques

The Unidentified Persons (UP) Database provides a place to enter the 10-finger fingerprint classification. Over the years, several classification systems have been used, including Henry, NCIC, and most recently, IAFIS. This document provides a basic description of each of these systems and offers guidance on how to enter the fingerprint classification into the UP database.

The Henry Fingerprint Classification System

The Henry fingerprint classification system is seldom used today, but it may have been used to classify fingerprints in cases of unidentified persons in years past. Because it might still be encountered, familiarity with it is prudent.

As with all fingerprint classification systems, the fingers are numbered 1 through 10, with 1 being the right thumb and 6 being the left thumb.

In the Henry system, fingers that have a whorl pattern are used to define a primary grouping for the set of fingerprints. A number is assigned to those fingers that have a whorl pattern. The numbers are assigned for each finger as follows:

Finger	1	2	3	4	5	6	7	8	9	10
Number	16	16	8	8	4	4	2	2	1	1

Finally, a primary grouping is established using the following formula: 1 +
(The sum of finger values for the even-numbered fingers) Divided by:
1 + (The sum of finger values for the odd-numbered fingers) Thus,
in applying the formula to the example above:

The numerator is 1 + 16 + 4 = 21 and the denominator is 1 + 0 = 1. Therefore, the primary grouping is 21/1.

If the Henry classification were used to report results in the Unidentified Persons system, it could be done as follows for the example above:

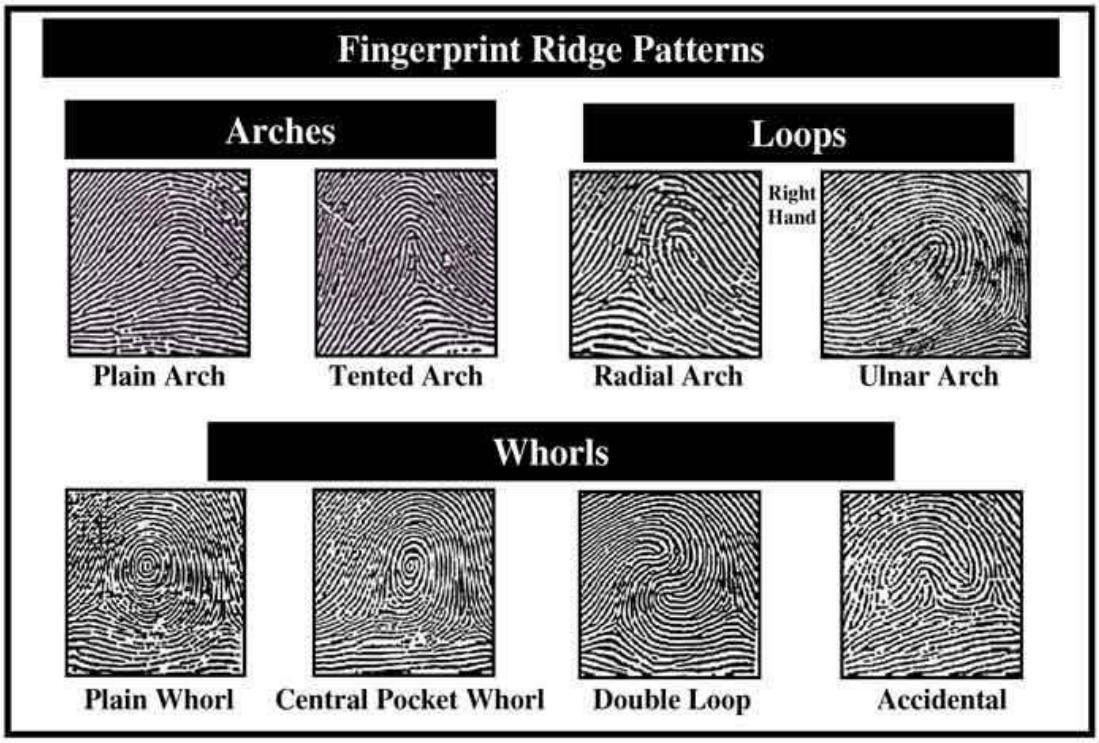
The fingerprint Comments box could include a notation that the Henry system was used and the primary grouping is 21/1.

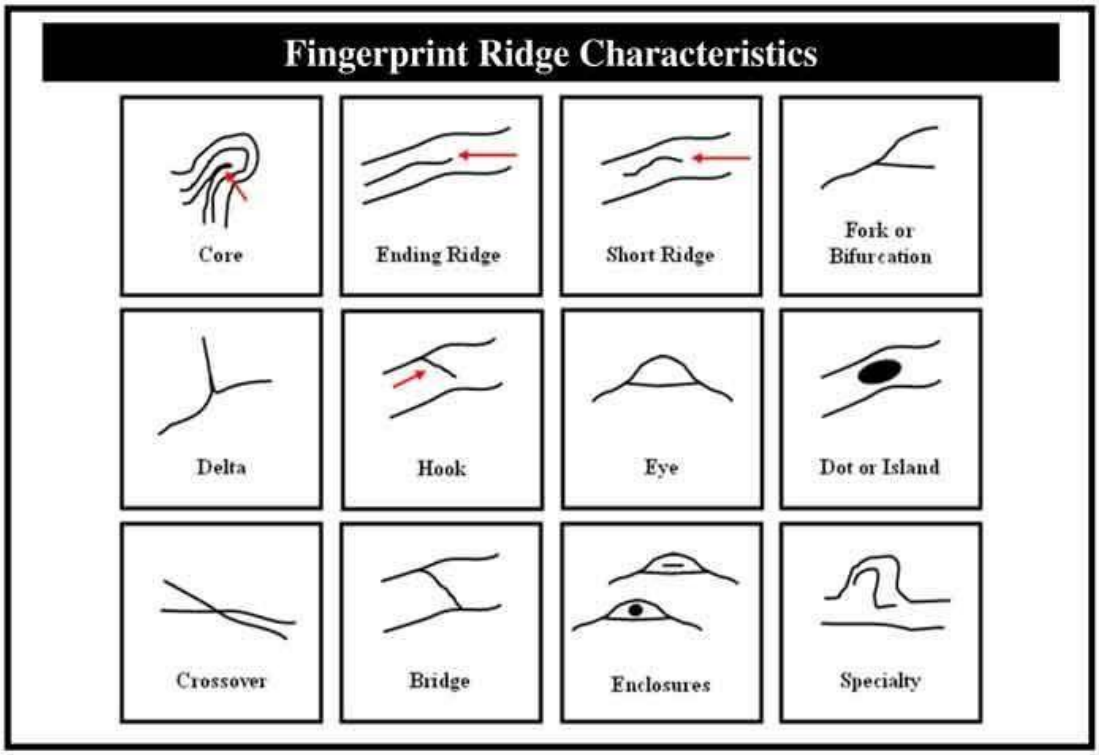
The text for the boxes is:

Classification box: A W A L L W L A A L.

Comments box: The classification at the left is the Henry Classification and the primary group is 21/1.

Fig. 3.2: Finger prints ridge patterns and their characteristics





Foot prints

Forensic footwear evidence can be used in legal proceedings to help prove that a shoe was at a crime scene. Footwear evidence is often the most abundant form of evidence at a crime scene and in some cases can prove to be as specific as a fingerprint. Initially investigators will look to identify the make and model of the shoe or trainer which made an impression.

ROLE OF FORENSIC PODIATRY

Footprint and footwear evidence is commonly present at a crime scene and must be discovered, recorded, and collected for further examination. When footprint analysis is required, the forensic podiatrist may act as an adjunct or a primary participant in the case. The foot is a complicated structure, and it requires years of experience to be able to distinguish all the intricacies—including soft tissue and skeletal pathologies—involved in its makeup and consequent evaluation.

THE CRIME SCENE

Physical evidence can be defined as articles and materials found during an investigation that may establish the identity of suspects and the circumstances under which the crime was committed. Footprints are known as physical evidence, as are fingerprints. It is evidence that speaks for itself and requires no explanation, only identification. Fingerprints are often discovered at the crime scene—but not always, because it is possible that nothing was touched or precautionary measures were taken (i.e., gloves may have been worn to preclude identification). However, it is unlikely that an individual can enter and leave the crime scene without using his or her feet. Discovering pedal evidence can be difficult, however, and a conscious effort must be made to do so. The initial officer(s) must recognize the importance of footprint evidence and try to preserve the integrity of the scene. This task can be quite difficult when medical personnel or other persons inadvertently destroy potential evidence. Foot impression evidence is most commonly discovered on ground surfaces, such as dirt, tile, concrete, and carpeting, but at times on counter-tops or other less common locations (6). Prints that are transient in nature, such as in snow, must be addressed and processed immediately. A print that is latent or invisible means it can be overlooked. The importance of this evidence to crime scene personnel needs to be stressed. If one footprint is discovered, then logically there may be more. For example, if there is a homicide scene with copious amounts of blood, then the expert should anticipate a good number of prints; if not, one would need to determine why not. Perhaps the scenario was manufactured or was altered or cleaned to conceal the presence of pedal evidence. General protocols regarding crime scenes are fairly universal. The main purpose is to discover evidence and recover it for scrutiny in the laboratory.

SECURE THE SCENE.

The first step is to secure the scene. This may seem basic, but at times it is difficult to enforce because there are often extraneous individuals who try to enter. Because our interest is in pedal evidence, foot traffic should be limited. RECORD THE SCENE. The most common methods of recording the scene are photography, sketching, and note taking. The use of video taping with commentary can be helpful and may negate the need for more time-consuming methods. The scene should be recorded as promptly as possible while it is in a relatively untouched state, especially when footprint evidence is being considered.

SEARCH THE SCENE (DISCOVER)

A systematic approach is necessary when footprint evidence is suspected. Depending on the type of crime, certain approaches and paths through the scene may vary. For example, where was the point of entry? Is there blood or a substrate that might be efficacious in exhibiting footprints or foot impressions? Is there a major crime area or several different sites? Where is the point of exit? An examination of the immediate exterior may yield many impressions in dirt or foot/shoe prints on a concrete walkway.

COLLECT (RECORD) AND PACKAGE EVIDENCE.

If footprints are visible, they must be photographed. This process includes proper positioning of the camera using a tripod, with the film parallel to the plane of the print or impression and directly over it, i.e., perpendicular to the impression. A scale should always be included so the photograph can be enlarged to reveal the natural size of the evidence, more commonly called 1:1, wherein 1 mm on the scale equals 1 mm. It is usually a good idea to take a similar photograph without a scale. As many photographs should be taken as possible, especially macro-views that will be used later for comparisons. It cannot be stressed enough how important accurate photographs are for a proper evaluation. Pedal impression evidence is often latent or poorly visible; therefore, various types of enhancement techniques must be used. Oblique lighting techniques using a strong white light are 378 DiMaggio. The Role of Feet and Footwear in Medicolegal Investigations 379 implemented to highlight or detect footprints that may not be clearly visible to the naked eye. If there is a suspicion that there might be bloody footprints but they are not visible, then Luminol or some other method can be used. Luminol causes the heme portion of the erythrocyte to luminesce; the technique must be performed in complete darkness. The luminescent effect is usually very short lived; therefore, a chemical agent such as amido black is used to enhance and stabilize the erythrocyte in a blue-black color and the footprints can then be photographed. An alternate light source, also known as a forensic light source, is an instrument that emits specific bands or wavelengths of light that are useful in detecting physical evidence.

Depending on the device, the range can start at 365 nm (which is in the ultraviolet [UV] range) and extend through the visible spectrum to infrared capabilities in the 700-nm range and higher. This instrument can supply bright white light for the oblique technique and has capabilities for footprints often in the UV range. It is usually used to detect biological fluids, hairs, and fibers. Three-dimensional impressions of footprints are often discovered in dirt, mud, or some other impressible substrate and should be photographed first, then casts made, if possible. One recommended material for casting is dental stone, because it is more rigid and durable than plaster of Paris. It is not uncommon for plaster of Paris casts to break when in transit or while being examined by different individuals. A broken cast is not an adequate exhibit. Lifting techniques can be used for certain types of footprint evidence, using adhesive and gelatin lifters as are used for fingerprints (7). If dust impressions are suspected, an electrostatic dust-lifting device can be used. It uses an electric charge to actually lift the dust print onto a foil surface that can be photographed and used for later evaluation. General protocol is used for packaging the evidence; most importantly, items must be kept separate to prevent cross-contamination. Shoes should be individually wrapped in separate paper bags, as should plaster or dental stone casts.

SUBMIT EVIDENCE TO THE LABORATORY. The modern laboratory is equipped to handle most types of evidence. It is advantageous to acquire as much evidence as possible from the scene for transport to the laboratory for processing. This may involve removing a door, flooring, or plasterboard if it has foot or shoe prints. Evidence can be enhanced both photographically and chemically. For instance, the sock liner of a shoe may be viewed with an alternate light source, using laser or bright white light, to give the most accurate depiction of the foot image (Fig. 1). In this instance, the sock liner was treated in a fuming chamber of cyanoacrylate ester (superglue) for 30 min at 80% humidity. Basic yellow-40 solution was applied with a soft brush; the liner was then rinsed with water for 2 min and dried. Excitation was accomplished using a Crimescope- 16 (SPEX Industries) at 455-nm. The camera used was a Crimescope VRM (SPEX Industries) with an orange long-pass filter or a 550-nm band-pass filter. In many cases, it takes experimentation to determine the best wavelength to get the best image because of the variability of the color of the sock-liner covering, which can be black, green, blue, white, or any of several other assorted colors. Many departments are using digital photography, which has certainly made the task much simpler and less time- consuming for obvious reasons. Footwear evidence that is recovered should be photographed and then, at a minimum, examined for trace evidence. Blood on footwear may be collected for DNA analysis.

The Forensic Team

The forensic team may include a police officer or detective, a crime scene specialist or criminalist, a footwear examiner, and an attorney. A professional tracker may be of value in certain situations. If the team is working for the defense, it may include a private investigator, a criminalist from a private laboratory, other forensic specialists, and the attorney. The footwear examiner specializes in footwear evidence and is trained to make an identification that involves class characteristics or individual (random or accidental) characteristics on the outsole. This individual must have expert knowledge of manufacturing techniques for different shoes, which can often be an important part of the evaluation.

Certification in the field is now available for those specializing in footwear evidence. The footwear examiner should be responsible for footwear-related evidence when the outsole evaluation is required and seeking appropriate podiatric medical consultation, if it is foot related.

Gait Pattern Significance in Forensics

Individuality of the Feet:

The human foot is highly individual in its form, size, and shape. These features originate from and are initially influenced by an individual's genetic code. However, environmental and physiological influences, as well as other acquired characteristics, injuries, and pathologies that occur throughout an individual's life, contribute to considerable and significant changes in the foot and result in the adult foot's uniqueness. Podiatrists' involvement in barefoot identification is both descriptive and interpretative. The particular emphasis is on the recognition and utilization of foot-related conditions and foot dimensions in this process. As podiatrists recognize a condition, state or pathology in an unknown barefoot print, this would then be described and compared with the recognized presence or absence of such a condition, state or pathology in a known barefoot print. Nature of Footprints: Footprints are produced under the following circumstances: - The impressions of the foot may be cause in mud, dust, sand, and snow or like substances. Such impressions will be depressed or three-dimensional type and referred to as sunken foot impressions. - If the footprint is produced by deposition of material like dirt, oil, blood, coloured powdery substance, etc on smooth and hard surfaces, giving rise to two-dimensional print, it is known as surface footprint. They are generally found indoors. - Footprint may also be produced by lifting dust or liquid material in which case a negative print will be left on the surface. Location of Footprints: Following places should be carefully searched: - At the crime scene - Around the place of occurrence - Along the route taken by the culprit, both at the time of ingress and egress, - At the places where the culprits gathered together for planning before action and for sharing the booty, - Besides the above, footprints may be found in the fields, courtyards, floors of rooms, walls, staircase, roofs, tables and chairs, papers, boxes, drain pipes etc, - If the culprit has visited a garage, he is likely to leave prints on the greasy surface of the garage floor, - If the culprit gets his foot or footwear smeared in blood he is likely to leave prints in blood. Identification characteristics: It is well established that every footprint has an individuality which cannot be duplicated. The individuality in a given print is established by studying the following characteristics: (a) Dimension: It should be noted. If the dimensions vary to a considerable extent then it can be concluded that the suspect could not have left the questioned print. (b) General Shape: The shape of the foot may have remarkable variations. It may be normal, flat, club shaped, bow shaped, broken bridge or abnormal. (c) Pattern: Sometimes ridge patterns may be noticed on the prints and impressions. Their study may reveal identifying characteristics. (d) Margins: Sometimes margin of the bare footprints are highly characteristic. The margins consist of outline of the heel and the inner and the outer boundary line of the foot. The heel may be oval of round; the outer margin may be straight, bulging outward or inward. (e) Toe marks: The shapes and sizes of the toes, their inter spaces and alignment vary greatly. Long and short toes, missing toes, an extra toe etc. may be highly characteristic. (f) Crease, phalanges, cut marks etc: The shape, size, position of the phalanges of the

toes, crease marks or cuts may be highly characteristic. Forensic Gait Analysis is defined as the identification of a person or persons by their gait or features of their gait, usually from CCTV footage and in comparison to footage of a known individual. Where the perpetrators of crime have been captured on CCTV, the persons gait and/or features of gait can assist in the subsequent identification process. Such gaits and features are identified on CCTV footage and compared to video or other recorded footage of a known person. This work is currently the exclusive domain of forensic podiatrists. Although electronic/ computing engineers are working on biometric systems in this same area, these are still under development and are not in practical use to date. The work is usually carried out at the request of police agencies or lawyers. (Bodziak) A single sequence of functions of one limb is called a gait cycle. It is essentially the functional unit of gait. The gait cycle has two basic components, the swing phase and the stance phase • Stance: phase in which the limb is in contact with the ground • Swing: phase in which the foot is in the air for limb advancement. A gait cycle is also referred to as a stride. Stride (Stride length): Linear distance between corresponding successive points of contact of the same foot (e.g., distance measured from heel strike to heel strike of the same foot) Step (Step Length): Linear distance in the plane of progression between corresponding successive contact points of opposite feet (e.g., distance measured from heel strike of one foot to heel strike of the other foot). Normally, the step length is approximately 15–20 inches.

Each stride comprises two steps.

Stance phase can be subdivided into: 1. Initial contact

. Loading response

1. Midstance
2. Terminal stance
3. Pre swing Swing phase subdivisions:

Initial swing

Midswing

Terminal swing

Gait pattern Analysis At least four consecutive foot or footwear marks are essential to determine gait pattern Discussion: When a person while walking or running makes a series of footprints or impressions, it is called a gait pattern or walking picture or walking ensemble. It is possible to deduce some of the characteristics of the owner's manner of walking. Walking pattern is found to be highly individualistic. It may therefore be of great value to study an individual.

The gait pattern is analyzed as follows:

i) The direction line: It is an imaginary line which indicates direction in which the person is walking or moving. It is a straight line.

ii) The gait line: In normal persons walk the gait line coincides with direction line and runs along the inner sides of both heel prints. It is more broken in stout persons and in pregnant women because they walk with their feet wide apart to maintain equilibrium. Broken walking line is suggestive of sluggish persons, drunkards and a person carrying heavy load on their backs.

iii) The foot line: This is a straight line running through the longitudinal axis of the footprint. It is a line which passes through the second toe to the center of the heel.

iv) The foot angle: The angle between the foot line and the direction line is known as the foot angle. It can be highly characteristic and does not change much. The normal foot angle is 30-32 degrees.

v) The principal angle: This is the angle between the foot lines of the two feet and is thus sum of the two foot angles.

vi) The step length or stride: This is the distance between the centers of two successive heel prints. It depends on the size of the walker, his habits and his speed.

vii) The step width: This is the distance between the parallel drawn in the direction of direction line touching the inner side of right and left foot. Other deductions from Gait pattern: - Age of the person: It may be possible to know whether the track marks left are of a young or old person. A young person may show twisting action of the toes and the ball of the foot. Old persons during their walking drag earth by their toes. Their step length will be comparatively shorter. Marks of stick may also be found by the side of the foot impressions. - The gender: Step length of an adult woman is between 25- 28 inches. The ball of the foot is comparatively narrower than that of a man. The unimpressed part in a woman's foot is more pronounced. Women press their feet without an inclination towards the outside and the first two toes kick the earth forward, whereas the other three leave stationary marks. Some Indian women wear rings on the toes, and if they walk barefooted are likely to leave their marks. - The Height of the person: The length of a footprint is not always suggestive of the person's height, but generally a longer footprint say, 10-12 inches and a step length exceeding 30 inches may suggest it to be that of a tall person (near about 5'- 10" to 6 feet in height). A short person will not only have a smaller foot (say, 6 to 8 inches in length), but would also register a shorter step length. Thus, a step length of about 20 or 24 inches with a foot whose length is about 6 to 8 inches suggest a person who is not more than 5'- 3" or 5'- 4 " in height. Footprint is 15% a person's height. Analysis indicates quick estimate of height may be made by presuming foot length to be 15.346% in men, 14.926% in women. The height of a man with a foot length of 28 cm would be estimated as 28 divided by .15346 = 182.5 cm. Likewise, a woman's foot length of 9 inches would be calculated in similar manner, 9 divided by .14926 = 60.29 inch. Conclusion: Walking pattern is highly individualistic. . It may therefore be of great value to study an individual. It may be possible to know whether the track marks left are of a young or old person. Step length of an adult woman is between 25-28 inches. The length of a footprint is not always suggestive of the person's height, but generally a longer footprint say, 10-12 inches and a step length exceeding 30 inches may suggest it to be that of a tall person

(near about 5'- 10" to 6 feet in height). A short person will not only have a smaller foot (say, 6 to 8 inches in length), but would also register a shorter step length.

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EXERCISES

PART A

1. What is Poroscopy?
2. What is dactylography?
3. What is anthropometry?
4. What are the advantages of Fingerprints?
5. What are the types of fingerprints?
6. What are the different methods of fingerprints?
7. What is visible and invisible finger prints?
8. How will you record and preserve foot prints?
9. What are the uses of foot prints?
10. What is FINDER?

PART B

11. What are the types of finger prints? How fingerprints are used for identification?
12. Briefly explain the different methods of development of latent finger prints.
13. What are the types of foot prints? How fingerprints are used for identification?
14. How will you record and preserve foot prints? What are the uses of foot prints?
15. What are the modern scientific techniques of detection and analysis of gait patterns? What is its importance?

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UNIT IV

POST-MORTEM EXAMINATION

An autopsy—also known as a post-mortem examination, necropsy, or autopsy

a cadaverum — is a highly specialized surgical procedure that consists of a thorough examination of a corpse by dissection to determine the cause and manner of death and to evaluate any disease or injury that may be present. It is usually performed by a specialized medical doctor called a pathologist.

Types

There are four main types of autopsies.

- *Medico-Legal Autopsy or Forensic* or *coroner's autopsies* seek to find the cause and manner of death and to identify the decedent. They are generally performed, as prescribed by applicable law, in cases of violent, suspicious or sudden deaths, deaths without medical assistance or during surgical procedures.
- *Clinical* or *Pathological autopsies* are performed to diagnose a particular disease or for research purposes. They aim to determine, clarify, or confirm medical diagnoses that remained unknown or unclear prior to the patient's death.
- *Anatomical* or *academic autopsies* are performed by students of anatomy for study purpose only.
- *Virtual* or *medical imaging autopsies* are performed utilizing imaging technology only, primarily magnetic resonance imaging (MRI) and computed tomography (CT)

Forensic autopsy

A forensic autopsy is used to determine the cause and manner of death. Forensic science involves the application of the sciences to answer questions of interest to the legal system. For example, in United States law, each death is classified into one of five headings:

- Natural
- Accident
- Homicide
- Suicide
- Undetermined

In some jurisdictions, the Undetermined category may include deaths in absentia, such as deaths at sea and missing persons declared dead in a court of law; in others, such deaths are classified under "Other". Medical examiners also attempt to determine the time of death, the exact cause of death, and what, if anything, preceded the death, such as a struggle. A forensic autopsy may include obtaining biological specimens from the deceased for toxicological testing, including stomach contents.

Toxicology tests may reveal the presence of one or more chemical "poisons" (all chemicals, in sufficient quantities, can be classified as a poison) and their quantity. Because post-mortem deterioration of the body, together with the gravitational pooling of bodily fluids, will necessarily alter the bodily environment, toxicology tests may overestimate, rather than underestimate, the quantity of the suspected chemical. Most states require the state medical examiner to complete an autopsy report, and many mandate that the autopsy be videotaped.

Following an in-depth examination of all the evidence, a medical examiner or coroner will assign one of the five manners of death listed above, and detail the evidence on the mechanism of the death.

Clinical autopsy - Clinical autopsies serve two major purposes. They are performed to gain more insight into pathological processes and determine what factors contributed to a patient's death. Autopsies are also performed to ensure the standard of care at hospitals. Autopsies can yield insight into how patient deaths can be prevented in the future.

Within the United Kingdom, clinical autopsies can be carried out only with the consent of the family of the deceased person as opposed to a medico-legal autopsy instructed by a Coroner (England & Wales) or Procurator Fiscal (Scotland) to which the family cannot object.

Post-mortem examination

What is a post-mortem examination?
A post-mortem examination is a medical procedure where a body is examined to find out what caused the death. It is not a common procedure and is only conducted in some circumstances, for example when the cause of death is unnatural or unknown.

The procedure involves extensive cutting of the body and removal of the internal organs for examination before replacing them in the abdominal cavity, not where they would have originally been located. Families should be aware that if they view a body after a post-mortem there may be marks which look like bruises which will have been caused by the post-mortem procedure. There can also be marks caused if there was an attempt to resuscitate the person prior to their death.

In some parts of the country and in some circumstances it may be possible to ask for a post-mortem to be conducted by using a MRI scan, so that the body does not have to be physically damaged. This is a very new method of undertaking post-mortems and there are mixed views about whether it is always a good idea. If you feel strongly about this, for example, for religious reasons, you can ask the coroner if it is possible but you will be expected to pay for it.

When does it happen?
A post-mortem is carried out as soon as possible after the death on behalf of the coroner as part of the investigation to establish the cause of death. It usually takes place within one or two days of the death.

The coroner is only required to inform the relatives of the deceased of the time and place at which the examination is to be made, if the relatives have told the coroner that they wish to be informed. The relatives have the right to be represented at the post- mortem by a medical practitioner or other representative (see below). In reality this very rarely happens and many families are not aware of their rights until it is too late and the post-mortem has already taken place. You should be aware that if you do have a representative attend the post-mortem on your behalf, you may be responsible for any of their related costs. In some exceptional circumstances these costs could be met by legal aid funding.

You will need to discuss this with a specialist solicitor to see if it would apply to your situation.
Where the police are investigating the death because they think a crime may have been committed, and when someone has been or may be charged with causing the death, the coroner may delay giving permission to bury the body so that the lawyers acting for the accused can arrange their own post-mortem. This can be very distressing for the family, but the coroner has a duty to protect the rights of the accused in this way.

If someone is charged with an offence and there is to be a criminal trial there will not usually be a full inquest as well. The coroner can hold an inquest after a trial but in practice this is very rare. If you think that there should be an inquest as well as a criminal trial, you will need to make sure the coroner knows your views and you may need to take legal advice.

Who may be present at the post-mortem examination?
The following people are entitled by law to be present at the post-mortem examination:

• A relative or their medically-qualified representative; a lawyer and anyone else representing the family.

• A pathologist representing the family if they have instructed one early enough. In many cases this will not occur at the initial examination but the family's pathologist may hold a second examination at a later date.

• The GP of the person who has died.

• A representative from the hospital where the person died.

• A representative of the Health and Safety Executive if the death was caused by an accident or disease following the issuing of a 'warning notice' as set out in the relevant legislation.
• Any government department which has notified the coroner that it wishes to attend.
• The chief officer of police.

• Any legally-qualified doctor representing any of the above.

• Any other person invited by the coroner. Can I

stop it?
Bereaved relatives do not have to give consent for a coroner's post-mortem examination to take place. However, if you feel strongly about this you should speak to the coroner.

If the coroner decides a post-mortem examination is necessary, the only means of stopping it is by a complicated legal procedure called a judicial review. It would be necessary to demonstrate that the coroner's decision was unreasonable, i.e. because the grounds for wanting a post-mortem were in some way insubstantial. In reality the post-mortem is often carried out very soon after the death and the family may not have sought advice that quickly, or they may not have been informed that it is taking place. The coroner will not release the body for the funeral until the procedures required have been carried out.

Who carries out a post-mortem examination?
A pathologist, who is a doctor specialising in this particular area. The coroner will appoint the pathologist.

Where will it take place?
Usually in the hospital where the person died or was taken immediately after death or at the public mortuary linked to the coroner's court.

Will any samples be kept?

At the post-mortem examination the pathologist may need to carry out some specialist tests in order to discover the cause of death, or be asked to carry out specific examinations by the coroner.

Sometimes very small pieces of tissue are kept for the pathologist to examine under the microscope at a later date. In some cases it may be necessary to keep the whole or parts of the organs to undertake particular tests. You can ask for information about this from the coroner or pathologist. You can also discuss delaying the funeral until the tests are completed so that the organs or tissue can be returned.

The coroner must tell you if organs or other samples are being kept after the post- mortem. You should also be told how long they will be kept and you will need to decide whether you want to have them returned to you, whether you wish to donate them for research, or whether they should be disposed of when the coroner decides they are no longer needed for the investigation. Further information about this is available online from the Human Tissue Authority.

Can I see the post-mortem report?

Under rule 57 of the Coroners Rules 1984, the relatives of the deceased may apply to the coroner for a copy of the post-mortem (pathology) report, for which you may have to pay a small charge.

There is no right to see the report before the inquest actually takes place, but it is the practice of most coroners to release it. This is sometimes made more difficult when there are controversial circumstances surrounding the death. It may take several weeks, or even longer, for the post-mortem report to be completed, especially where extra tests are taking place. Some coroners prefer to send the post-mortem report to the family GP so that the GP can explain the contents to the family. Some families find it too distressing to read the post-mortem report, but if you have instructed a solicitor this is something they can do on your behalf.

Can I have a second post-mortem examination?

Yes, with the consent of the coroner. In practice this is very rarely denied. If it is denied it can be challenged by judicial review.

If you consider having a second post-mortem you must be aware of the need to have a solicitor to instruct the pathologist and also of the cost involved. It will probably delay the funeral. But if you are not happy with the information you have received about the cause of death and you have worries about the circumstances of the death, you should consider a second post-mortem. It is important to use a pathologist with specialist expertise and you will need to take advice on this. You also need to ensure that the body is properly preserved. After a few days a body will start to deteriorate and if it needs to be kept for longer, the body will be frozen in order to preserve it.

What is a toxicology report?

A toxicology report is a report done on blood and tissue to establish whether there were any toxic substances in the body which may have contributed to the death. The toxicology report will detail prescription drugs, illegal drugs, alcohol and any other chemical substances which the toxicologist has been instructed to test for. It will usually take 6-8 weeks for the toxicology report to be completed. You can ask for a copy of this with the post-mortem report.

What will the body look like after a post-mortem?

A body will begin to deteriorate and this will become obvious if it has to be kept for any time. This can be very distressing for family and friends particularly if they want to have an open casket at the funeral or perform other rituals with the body. You will need to talk to your funeral director and take their advice about whether it is going to be possible to have an open casket.

POISONING

Poisoning occurs when any substance interferes with normal body functions after it is swallowed,

inhaled, injected, or absorbed. The branch of medicine that deals with the detection and treatment of poisons is known as toxicology.

Description

Poisonings are a common occurrence. About 10 million cases of poisoning occur in the United States each year. In 80%of the cases, the victim is a child under the age of five. About 50 children die each year from poisonings. Curiosity, inability to read warning labels, a desire to imitate adults, and inadequate supervision lead to childhood poisonings.

The elderly are the second most likely group to be poisoned. Mental confusion, poor eye sight, and the use of multiple drugs are the leading reasons why this group has a high r ate of accidental poisoning. A substantial number of poisonings also occur as suicide attempts or drug overdoses.

Poisons are common in the home and workplace, yet there are basically two major type. One group consists of products that were never meant to be ingested or inhaled, such as shampoo, paint thinner, pesticides, houseplant leaves, and carbon monoxide. The other group contains products that can be ingested in small quantities, but which are harmful if taken in large amounts, such as pharmaceuticals, medicinal herbs, or alcohol. Other types of poisons include the bacterial toxins that cause food poisoning, such as *Escherichia coli*; heavy metals, such as the lead found in the paint on older houses; and the venom found in the bites and stings of some animals and insects. The staff at a poison control center and emergency room doctors have the most experience diagnosing and treating poisoning cases.

Causes and symptoms

The effects of poisons are as varied as the poisons themselves; however, the exact mechanisms of only a few are understood. Some poisons interfere with the metabolism. Others destroy the liver or kidneys, such as heavy metals and some pain relief medications, including acetaminophen (Tylenol) and nonsteroidal anti-inflammatory drugs (Advil, Ibuprofen). A poison may severely depress the central nervous system, leading to coma and eventual respiratory and circulatory failure. Potential poisons in this category include anesthetics (e.g. ether and chloroform), opiates (e.g.,morphine and codeine), and barbiturates. Some poisons directly affect the respiratory and circulatory system. Carbonmonoxide causes death by binding with hemoglobin that would normally transport oxygen through out the body. Certain corrosive vapors trigger the body to flood the lungs with fluids, effectively drowning the person. Cyanide interferes with respiration at the cellular level. Another group of poisons interferes with the electrochemical impulses that travel between neurons in the nervous system. Yet another group, including cocaine, ergot, strychnine, and some snake venoms, causes potentially fatal seizures.

Severity of symptoms can range from headache and nausea to convulsions and death. The type of poison, the amount and time of exposure, and the age, size, and health of the victim are all factors which determine the severity of symptoms and the chances for recovery.

MEDICO-LEGAL PROCEDURES

Introduction

Medico-legal aspects of medical practice cover a wide spectrum and involve both the living and the dead. Areas concerning the living include certification of births, certification of mental illness, disputed

paternity, abortion, confidentiality of patients' medical records, compensation for injuries, non-lethal poisoning and various types of sexual offences. This lecture series deals with medico-legal procedures pertaining to the dead.

This spectrum includes verification and certification of death, estimation of the time of death, identification of the deceased, determining which deaths need autopsies and which autopsies are "coroner's cases", performing autopsies to determine the cause of death, and giving evidence in court as to the relevant autopsy findings, cause of death and manner of death where possible.

When called to see a "dead" person, a doctor's duties may entail any or all of the following:

- Making sure that death has actually occurred
- Making a superficial examination to exclude medical grounds for suspicion of foul play
- Forming an opinion as to the approximate time of death
- Issuing a death certificate (once satisfied that the cause of death is a natural one).

If death is thought to fall under the ambit of those deemed to be "Coroner's cases" (see below), then the office of the Coroner is informed and brought into play.

Who is the coroner? - In many countries the coroner is usually either a lawyer or a doctor employed by a city or county to enquire into certain types of deaths, these deaths being referred to him by the police, the public, doctors or by local Registrars of Deaths. In Jamaica (and many other Caribbean countries with a British-derived legal system) the coroner is usually a Resident Magistrate. The Coroner's Act of Jamaica states: "The officer for the time is being discharging the duties of *Resident Magistrate* for any parish shall ex officio is the Coroner of such parish ". The office of the coroner is a uniquely English institution with a very interesting and colourful history.

The Coroners Act states that any duly qualified medical practitioner may be directed by the coroner to make a post-mortem examination of a dead body in cases where *" there is reasonable cause to suspect that such person has died, either a violent, or an unnatural death, or has died a sudden death, of which the cause is unknown, or that a medical certificate of cause of death ...will not be forthcoming ".*

Different jurisdictions worldwide have different explicit indications for deaths that should be referred to the Coroner. The relative vagueness of our Coroners act has resulted in the following administrative guidelines being stipulated as indications for our Coroners cases:

1) All deaths due to violence – including all deaths due to accidents, suicide, homicide

2) All deaths due to drowning

2) All deaths from known or suspected poisoning

3) All deaths resulting from criminal abortion

4) All deaths during or soon after (24 hours) surgical operation or anaesthesia

5) All deaths that occur within 24 hours of admission to hospital

6) All deaths from sudden unexpected natural causes

7) All deaths of inmates of a government custodial institution or place of detention, e.g. prison or mental asylum

The stipulation for a coroner's autopsy in deaths that occur within 24 hours of admission is sometimes contentious. It is obvious that some deaths that occur within 24 hours of admission will truly be coroner's cases while others will not be. In some of these deaths the patient may have recently been seen by a doctor who, being aware of an illness that might have led to the patient's death, might be in a position to issue a death certificate, thus obviating the need for a post-mortem examination.

The following is a synopsis of the procedure involved in Coroner's cases as stipulated in the Coroners Act :

- The Coroner (or his/her representatives - the police force) is informed that a person has died (or a dead body found) under conditions that appear to fit the indications for a Coroner's post-mortem examination.

- The police investigate the circumstances relating to the death (visit to the scene of death, collect statements from witnesses/relatives etc.) and report to the Coroner.

- The Coroner directs a medical practitioner (any medical practitioner who holds public medical office may be so directed) to make a post-mortem examination of the body with a view to determining the cause of death and the circumstances connected with the death. This examination may extend to dissection of the body as far as is thought requisite for the purpose.

- After the autopsy, the report is delivered to the Coroner (usually via the police) and the police officer may then, unless otherwise contraindicated, authorize the burial of the body.

- If upon receipt of the medical and police reports the Coroner is satisfied that the deceased was not a victim of murder or manslaughter and that nothing further needs to be done in this case, he/she may abstain from holding an inquest and submit the findings to the Director of Public Prosecutions (DPP) for ratification and issuing of the death certificate. Note, however, that the DPP has the power to overrule the Coroner's decision not to hold an inquest if he/she deems it necessary.

- If upon the receipt of the medical and police reports the Coroner deems it necessary to hold an inquest, a jury is convened, the relevant witnesses are summoned and the facts touching on the case are examined. The medical practitioner who did the autopsy may be required to attend the inquest and maybe questioned concerning the postmortem findings.

- After hearing the evidence given in the inquest, the jury gives a verdict as to:

- The identity of the deceased
- How, when and where the deceased came by his/her death
- Whether the circumstances indicated death by natural causes, accident, suicide, murder, manslaughter etc.
- If murder/manslaughter is indicated, who is/are the person(s) to be charged with said murder/manslaughter

➢ If the inquisition charges a person(s) with murder or manslaughter, the Coroner issues a warrant for the arrest of said person(s) in order to bring the accused to trial.

The Coroner's (Medico-legal/Forensic) autopsy

Coroner's autopsies are performed for the well-being of the public, which demands answers and reasons as to causes of death among the citizenry.

The objectives of the coroner's (medico-legal/forensic) post-mortem examination include:

a) Establishing the identity of the deceased if in doubt

b) Determining the time of death

c) Determining the cause of death and stating if such cause is natural or unnatural. If unnatural, then a decision as to the manner of death is made, i.e. accidental, homicidal or suicidal?

The medico-legal autopsy must be performed by a registered medical practitioner, who should:

- Make relevant notes of the crime scene if it is visited, taking note of important data such as the position of the body, its relationship to surrounding objects, the state of the clothing, presence of drugs or weapons etc.
- Ensure that the body is identified to him/her before the autopsy is performed. This is done by the next of kin in the presence of the police officer who is in charge of the investigation.
- Make precise records of when and where the autopsy was performed, and in whose presence.
- Take special care to make a detailed and meticulous external examination of the body (preferably in a well-lit and equipped autopsy room/morgue) with particular attention being paid to any marks of injury or violence especially on the neck, hands, and genitalia. (Remember to examine the back of the body and the scalp)
- Take photographs of relevant findings if this facility is available.
- Carefully document all pertinent internal findings and record the direction and depth of injuries before removing organs.
- Collect all samples which are deemed to have a bearing on the case e.g. gastric contents, blood and urine samples, hair, nail scrapings, tissues or organs, bullets etc.
- Keep all samples collected separately in clean glass containers, clearly labelled, and hand the appropriate ones to the police and keep a record of this transaction i.e. obtain a receipt.
- Submit a report of the autopsy to the Coroner including a clear statement as to the cause of death. The report should be couched in clear and simple language avoiding confusing medical jargon as it will have to be read and understood by various laymen at different levels of the investigative process. Remember that the report is the property of the Coroner and neither its contents nor actual copies of the report may be given to anyone else without his/her permission.
- Testify at the inquest and/or trial if summoned.

What is the "cause of death"?

When conveying this information to the coroner on the report, one must be careful to make a distinction between cause and manner of death. The cause of death is the condition that was directly responsible for the patient's demise, e.g. stab wound to the heart with massive blood loss, or ischaemic heart disease with acute myocardial infarction etc. The manner of death refers to the circumstances of the death, i.e. homicidal, suicidal, accidental, natural cause etc.

Who may be present at an autopsy?

The pathologist/doctor performing the autopsy usually determines who may be present. The general aim is to have as few people present as possible, especially in cases of criminal or suspicious death, in view of the risk of loss of confidentiality. Too many people being present might hamper and distract the pathologist and increases the risk of contamination of evidence.

Naturally the autopsy room attendant and the coroner's representative(s) – usually in the form of the police – are entitled to attend, as is any relevant technical staff who might have to collect evidence, take photographs etc. Doctors in training and medical students may be allowed at the discretion of the pathologist. Casual observers (irrespective of calling or rank) who have no official connection to the case should be discouraged from attending.

In recent years here in Jamaica, relatives have lobbied to be represented at coroner's autopsies and provision has now been made for a medical practitioner to attend these autopsies as an observer on behalf of the family of the deceased. This intention has to be communicated in writing to the doctor performing the autopsy, and should be done expeditiously so that the scheduling of the autopsy is not disrupted.

Identification of the deceased

In most cases the identity of the deceased can be obtained simply by the viewing of the body and confirmation by the next of kin (or other appropriate relative or close friend). In some cases however the identification of a dead body may depend on data derived from any or all or the following sources, some of which may be provided by the police, others by the pathologist, and some by other experts:

- Documents or items of identification found on the body e.g. I.D. card, I.D. bracelet, wallet etc.
- Physical data/stigmata - height, weight, sex, race, skin and hair colour, scars, deformities, tattoos, etc
- Published photographs
- Fingerprints
- X-rays
- Blood type
- Dental records
- DNA testing

Forensic anthropologists, dentists and radiologists can examine skeletal remains and often give a good opinion as to:- (1) whether the source is human or animal, (2) whether the bones belong to one or more

individuals, (3) age, (4) sex, (5) stature, (6) race, (7) identity, (8) cause of death, and (9) time since death.

Timing of Death

The use of stomach emptying as a measure of time since death is fraught with controversy. The assumption that an average meal will be digested and pass out of the stomach in about 2 – 4 hours can be used as an aid in timing death if one has an idea when the deceased last ate.

However, some confounding factors that obtain are:

(a) digestion may continue for some time after death

(b) meals that are relatively more liquid exit the stomach faster than those that are more solid

(c) fatty foods take longer to exit the stomach.

These variables make timing death by the use of gastric contents and gastric emptying relatively inaccurate. The bodily changes that occur after death provide a better estimate.

Bodily changes after death

Certain bodily changes occur after death and may be used to help to time the death. These include:

- Cooling
- Hypostasis (lividity)
- Rigor mortis
- Putrefaction

Cooling continues for about 18-24 hours after death until body temperature reaches that of the surroundings (assuming that the environment is cooler than normal body temperature!). The rate of fall depends on the difference between body temperature at death and that of the surroundings, how well clad the body is, the amount of body fat, and if the body is immersed in water. Formulae exist by which the time of death can be calculated when the relevant data – temperature of the body, ambient temperature etc.

Temperature must be recorded in the rectum, not on the body surface. Naturally this parameter will be useless if the body had been previously refrigerated.

Hypostasis (lividity; livor mortis) is the process by which blood drains by gravity to the most dependent parts of the body when blood circulation ceases and the blood vessels relax and dilate after death. Timing of hypostasis is very difficult as the process is quite variable, but as a rough guide, it starts about 1-6 hours after death and is fully established in about 6-12 hours. After this, the blood tends to coagulate in the vessels and the hypostasis is said to have become “fixed”. If the body is moved after this time, the hypostasis will not alter its distribution – a useful pointer in some cases in determining if a body has been moved after death.

Rigor mortis is the stiffening of the muscles caused by the gelling of actin and myosin due to

exhaustion of glycolysis and depletion of ATP. It starts with the smaller muscles of the face and jaw and extremities before the larger muscle masses. It tends to progress from head to foot and reverses in the opposite direction. It begins in about 3-6 hours and will have affected the whole body in 12 hours. The stiffness lasts about 36- 48 hours and disappears as putrefaction begins to dissolve muscle protein. Therefore the speed with which rigor dissipates will be related to the speed of onset of putrefaction. Once rigor is fully developed in a muscle, if the muscle is stretched and the rigor is "broken", stiffening will not recur.

Putrefaction, or decomposition of a dead body, results from postmortem bacterial proliferation with gas formation. It begins its gross manifestation with a greenish discoloration of the skin of the anterior abdominal wall, which appears about 48 hours after death. Subsequently the body swells and discoloration appears along the lines of the superficial veins, with blistering of the skin, rupture of body cavities and liquefaction of organs. Flies are attracted to the putrefying body and lay eggs in open wounds and natural orifices. Maggots hatch in about 24 hours and develop into adult flies in about 4-5 days. Forensic entomologists can study the insect eggs, maggots or pupa taken from putrefying bodies and use their stage of development to help to determine time of death.

Factors modifying putrefaction include warmth, moisture and air, and both the speed of onset and the progression of putrefaction are more rapid in hotter climates.

Adipocere is a type of post mortem change that usually becomes apparent about 3 or more months after death. It is the name given to the white, greasy, waxy, musty-smelling material that replaces the fatty tissue of a part of, or the whole of the body. It results from hydrolysis and hydrogenation of body fats to fatty acids and soaps (saponification). It is most commonly found in bodies lying in relatively warm, moist, anaerobic conditions.

Mummification is the process by which the body becomes dry and brittle after lying for some time in warm dry environments. Mummification of an entire body occurs over a period of several weeks to months.

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EXERCISES

PART A

1. Define Poisoning. What are the types of Poisons?
2. What is post mortem?
3. What is ante mortem?
4. What is rigor mortis?
5. What is the purpose of autopsy?
6. What is exhumation?
7. List out few questions what autopsy surgeon usually faces in connection with examination of skeletal remains.
8. Define death.
9. How do you determine time passed after death?
10. What are the changes occurred due to immediate decomposition?
11. What are the changes after death?
12. What is banned drugs?
13. What are the sources of poisons?

PART B

14. Describe the procedure of autopsy examination.
15. Elaborate in detail about reconstruction of postmortem examined body.
16. What is Poison? What are its types, sign-symptoms, diagnosis and treatment of poison case?
17. What is suicide? What are the factors to be considered during post mortem findings of poison case and medico legal aspects of poisons?
18. What are the modern scientific techniques of detection and analysis of poisons and different trace elements?

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UNIT – V – Serological and Chromosomal Techniques – SBMA4007

UNIT V

Serology

Serology is the scientific study of serum and other bodily fluids. In practice, the term usually refers to the diagnostic identification of antibodies in the serum. Serological tests may be performed for diagnostic purposes when an infection is suspected, in rheumatic illnesses, and in many other situations, such as checking an individual's blood type.

Serology is the examination and analysis of body fluids. A forensic serologist may analyze a variety of body fluids including saliva, semen, urine, and blood. From 1950 to the late 1980's, forensic serology was a most important part of lab procedures. With the development of DNA techniques, more time, money, and significance was placed in developing DNA labs. However, with limited funds and the time required for DNA testing, most labs still use many of the basic serology testing procedures.

There are several serology techniques that can be used depending on the antibodies being studied. These include: ELISA, agglutination, precipitation, complement-fixation, and fluorescent antibodies.

Some serological tests are not limited to blood serum, but can also be performed on other bodily fluids such as semen and saliva, which have (roughly) similar properties to serum.

Serological tests may also be used in forensic serology, specifically for a piece of evidence (e.g., linking a rapist to semen sample).

Blood Terminology:

- ABO blood groups—based on having an A, B, both or no antigens on red blood cells
- Rh factor—may be present on red blood cells; positive if present and negative if not
- Antigen—a substance that can stimulate the body to make antibodies. Certain antigens (proteins) found in the plasma of the red blood cell's membrane account for blood type.
- Antibody—a substance that reacts with an antigen

Agglutination—clumping of red blood cells; will result if blood types with different antigens are mixed.

Presumptive Tests for Blood Determination:

- Kastle-Meyer color test—a mixture of phenolphthalein and hydrogen peroxide; the hemoglobin will cause the formation of a deep pink color if blood is present
- Hematest tablet—reacts with the heme group in blood causing a blue-green color
- Luminol test—reaction with blood to produce light
- Microscopic observation
- Precipitin test—blood is injected into a rabbit; antibodies are formed; the rabbit's blood is extracted as an antiserum; the antiserum is placed on sample blood. The sample will react with human proteins if human blood is present. This test is very sensitive and requires only a small amount of blood.

Human Blood:

- ✓ Red blood cells are most numerous; 5 to 6 million per mm^3
- ✓ White blood cells are larger and less numerous; 5 to 10,000 per mm^3
- ✓ Platelets are tiny, cellular fragments; 350 to 500,00 per mm^3

A. Blood Group Antigens and Antibodies

1. Blood group antigens are structures embedded in or protruding from RBCs, WBCs and Platelets.

2. Exposure to blood group antigens is usually the result of a prior transfusion or pregnancy causing exposure of the individual to antigens they do not possess.

3. An individual may form antibodies to the antigens on these types of cells which they lack.

a. Immunohematology is concerned with the detection of IgG and IgM class antibodies.

b. IgM antibodies are usually naturally occurring, RT agglutinins, easily detected by routine procedures.

c. IgG are immune antibodies produced as a result of exposure to the antigen and require a special serological test (the Coomb's procedure) to detect their presence.

4. Immunohematology uses some basic serological tests to detect the presence of antibodies in an individual's serum and/or antigens on an individual's cells.

5. The detection of antigens is routinely used to determine a person's blood group and D type.

6. Antibody detection tests are used to confirm the ABO antigen typing and to detect "unexpected antibodies" or immune antibodies in the serum which may cause the destruction or hemolysis of transfused cells containing the antigen.

B. Dynamics of Antigen-Antibody Reactions

1. Union of antigen with its specific antibody depends upon structure and charge of the molecule. 2. Physical forces hold antigen and antibody together.

a. Physical forces are weak.

b. The attractive forces vary in strength with changes in pH, ionic strength, temperature, and nature of the solvent.

c. Blood group antigens and antibodies bind to each other until a dynamic equilibrium is reached in which as many bonds are formed as are disrupted.

d. Represented by a bell shaped curve based on concentration of the reactants: 1) Pro- zone (antibody excess) 2) Zone of equivalence, optimal proportion of antigen and antibody.

3) Post zone (antigen excess).

C. Detection of Antigen-Antibody Reactions in Immunohematology

1. Agglutination is the most common procedure used in immunohematology.

a. RBCs have antigenic determinants which combine with antibodies present in the test serum added.

b. Antibody bridges form with antigenic determinants on adjacent cells resulting in agglutination 2. Hemolysis represents destruction of RBC membrane and when detected, hemolysis is considered a positive result.

3. Solid Phase Adherence is used to detect and identify antigens or antibodies.

a. Methodology primarily used in donor screening.

b. Uses microplates coated with RBC or antibody to which serum or RBCs are added.

c. If antigen-antibody reaction occurs, the cells adhere to the sides of the well, if no reaction occurs the cells settle to the bottom of the well.

D. Principle and Variables Involved In Agglutination Reactions

1. Two stages involved in agglutination reaction.

a. The first stage of agglutination is sensitization, attachment of antibody on to the corresponding antigen site on the RBC membrane.

b. The second stage is the formation of bridges between the sensitized cells.

1) IgG antibodies have two antigen binding sites, one site will bind to antigen on an RBC and the other site will bind to antigen on a different RBC resulting in the formation of a lattice.

2) Lattice formation results in hemagglutination.

2. Variables affecting first stage of agglutination. a. Antigen-antibody ratio (serum to cell ratio) is very critical.

1) Optimal amount of serum must be determined.

2) Increasing amount of serum increases number of antibody molecules coating the cell.

3) Albumin unless used under low ionic conditions does little to affect antibody uptake on to the cell, rather it influences the second stage of hemagglutination.

Concept of Antibody and antigen

Antibodies, also called immunoglobulins, are proteins manufactured by the body that help fight against foreign substances calledantigens. When an antigen enters the body, it stimulates the immune system to produce antibodies.

Antigens are any substance that stimulates the immune system to produce antibodies. Antigens can be bacteria, viruses, or fungi that cause infection and disease. They can also be substances, called allergens,that bring on an allergic reaction. Common allergens include dust, pollen, animal dander, bee stings, or certain foods. Blood transfusions containing antigens incompatible with those in the body's own blood will stimulate the production of antibodies, which can cause serious, potentially life- threatening reactions.

Classes of antibodies and their functions

There are five classes of antibodies, each having a different function. They are IgG, IgA, IgM, IgD, and IgE. Ig is the abbreviation for immunoglobulin, or antibody.

IgG antibodies are the most common and the most important. They circulate in the blood and other body fluids, defending against invading bacteria and viruses. The binding of IgG antibodies with bacterial or viral antigens activates other immune cells that engulf and destroy the antigens. The smallest of the antibodies, IgG moves easily across cell membranes. In humans, this mobility allows the IgG in a pregnant woman to pass through the placenta to her fetus, providing a temporary defense to her unborn child.

IgA antibodies are present in tears, saliva, and mucus, as well as in secretions of the respiratory, reproductive, digestive, and urinary tracts. IgA functions to neutralize bacteria and viruses and prevent them from entering the body or reaching the internal organs.

IgM is present in the blood and is the largest of the antibodies, combining five Y-shaped units. It functions similarly to IgG in defending against antigens but cannot cross membranes because of its size. IgM is the main antibody produced in an initial attack by a specific bacterial or viral antigen, while IgG is usually produced in later infections caused by the same agent.

Antigen - antibody reaction:

Antigen-antibody interaction, or antigen-antibody reaction, is a specific chemical interaction between antibodies produced by B cells of the white blood cells and antigens during immune reaction. It is the fundamental reaction in the body by which the body is protected from complex foreign molecules, such as pathogens and their chemical toxins. In the blood, the antigens are specifically and with high affinity bound by antibodies to form an antigen-antibody complex. The immune complex is then transported to cellular systems where it can be destroyed or deactivated.

There are several types of antibodies and antigens, and each antibody is capable of binding only to a specific antigen. The specificity of the binding is due to specific chemical constitution of each antibody. The antigenic determinant or epitope is recognized by the paratope of antibody, situated at the variable region of the polypeptide chain. The variable region in turn has hyper-variable regions which are unique amino acid sequences in each antibody. Antigens are bound to antibodies through weak and noncovalent bonds such as electrostatic interactions, hydrogen bonds, Van der Waals forces, and hydrophobic interactions.

Chemical bond

Antibodies bind antigens through weak chemical interactions, and bonding is essentially non-covalent. Electrostatic interactions, hydrogen bonds, van der Waals forces, and hydrophobic interactions are all known to be involved depending on the interaction sites.

Affinity

Antigen and antibody interact through a high affinity binding much like lock and key. A dynamic equilibrium exists for the binding. For example, the reaction is a reversible one, and can be expressed as:

$$[Ab] + [Ag] \leftrightharpoons [AbAg]$$

where [Ab] is the antibody concentration and [Ag] is the antigen concentration, either in free ([Ab],[Ag]) or bound ([AbAg]) state.

The equilibrium association constant can therefore be represented as: $K_a = \frac{k_{on}}{k_{off}} = \frac{[AbAg]}{[Ab][Ag]}$

where K is the equilibrium constant.

Reciprocally the dissociation constant will be: $K_d = \frac{k_{off}}{k_{on}} = \frac{[Ab][Ag]}{[AbAg]}$

where ratio of k_a and k_d describes the binding affinity:

$$K = \frac{K_a}{K_d} = \frac{[AbAg]}{[Ab][Ag]}$$

Cross-reactivity applies to the reaction between two different species as opposed to the self-reactivity. In immunology, the cross-reactivity has a more narrow meaning of the reaction between an antibody and an antigen that differs from the immunogen.

APPLICATION OF SEROLOGY IN FORENSIC SCIENCE

According to forensic serologist Marcella Jones, forensic serology is the analysis of body fluids as they relate to forensic cases, including DNA analysis. Accordingly, the role of the forensic serologist involves:

Examining evidence for the presence of body fluids e.g. blood, semen, hair, tissue, saliva, feces, and urine
Evaluating evidence for potential DNA analysis
Evaluating species of body fluids
Perform PCR (polymerase chain reaction) based typing of STR (short tandem repeat sequences) of genomic DNA, or mitochondrial sequence analysis of mitochondrial DNA
Statistically evaluating evidence relevant in the case to impart weight on the evidence
Performing all analysis with objectivity and unbiased scientific methods in court.

Karyotyping - A karyotype is the number and appearance of chromosomes in the

nucleus of a eukaryotic cell. The term is also used for the complete set of chromosomes in a species, or an individual organism.

Karyotypes describe the chromosome count of an organism and what these chromosomes look like under a light microscope. Attention is paid to their length, the position of the centromeres, banding pattern, any differences between the sex chromosomes, and any other physical characteristics. The preparation and study of karyotypes is part of cytogenetics.

The study of whole sets of chromosomes is sometimes known as *karyology*. The chromosomes are depicted in a standard format known as a *karyogram* or *idiogram*: in pairs, ordered by size and position of centromere for chromosomes of the same size.

The basic number of chromosomes in the somatic cells of an individual or a species is called the *somatic number* and is designated *2n*. Thus, in humans 2n = 46. In the germ- line (the sex cells) the chromosome number is *n* (humans: n = 23).

So, in normal diploid organisms, autosomal chromosomes are present in two copies. There may, or may not, be sex chromosomes. Polyploid cells have multiple copies of chromosomes and haploid cells have single copies.

The study of karyotypes is important for cell biology and genetics, and the results may be used in evolutionary biology (*karyosystematics*) and medicine. Karyotypes can be used for many purposes; such as to study chromosomal aberrations, cellular function, taxonomic relationships, and to gather information about past evolutionary events.

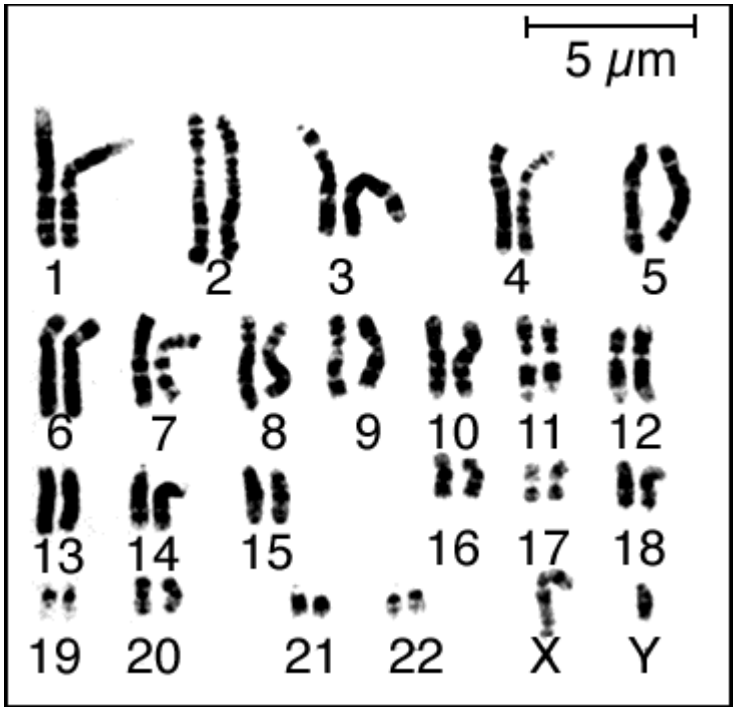

Fig. 5.1: Karyotype patterns

A karyotype is a picture of chromosomes. The normal human karyotype has a total of 46 chromosomes.

Homologous pairs 1-22 are called the autosomes. X and Y are the sex chromosomes.

CHROMOSOMAL ABNORMALITIES

Normally, humans have 23 pairs of chromosomes - making 46 in total. This includes one pair of chromosomes which are the sex chromosomes. The ova and the sperm each carry 23 chromosomes.

- Numerical Aberrations
- Structural Aberrations

Chromosomal abnormalities occur when there is a defect in a chromosome, or in the arrangement of the genetic material on the chromosome. Very often, chromosome abnormalities give rise to specific physical symptoms, however, the severity of these can vary from individual to individual.

Abnormalities can be in the form of additional material which may be attached to a chromosome, or where part or a whole chromosome is missing, or even in defective formation of a chromosome. Any increases or decreases in chromosomal material interfere with normal development and function.

There are two main types of chromosomal abnormality which can occur during meiosis and fertilization: numerical aberrations and structural aberrations.

Numerical Aberrations

These are usually caused by a failure of chromosome division, which results in cells with an extra chromosome or a deficiency in chromosomes.
Gametes with these anomalies can result in conditions such as Down syndrome (who have 47 chromosomes instead of 46), or Turner syndrome (45 chromosomes).
Common types of numerical aberrations are: triploidy, trisomy, monosomy and mosaicism.

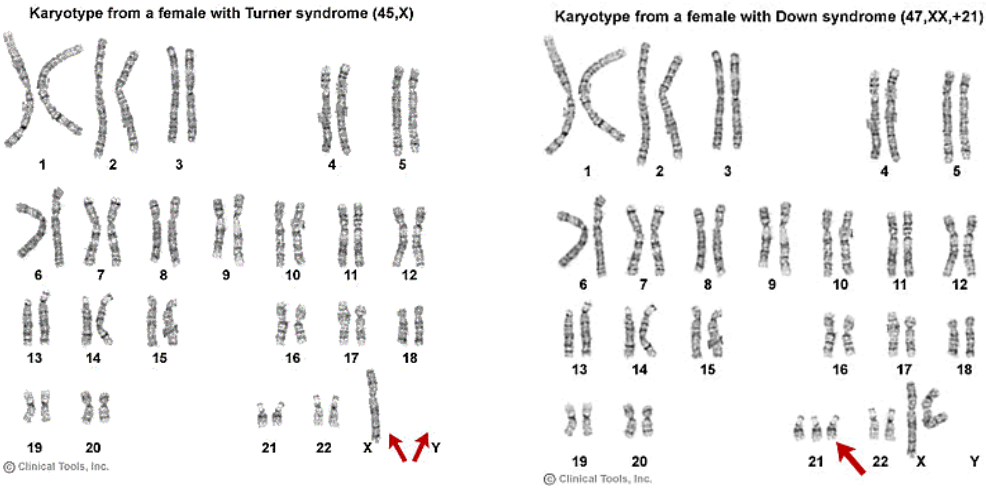

Fig 5.2 Karyotype of Turner syndrome (45chromosomes instead of 46) Karyotype of Down Syndrome (47 chromosomes instead of 46)

STRUCTURAL ABERRATIONS

These occur due to a loss or genetic material, or a rearrangement in the location of the genetic material. They include: deletions, duplications, inversions, ring formations, and translocations.

- Deletions: A portion of the chromosome is missing or deleted. Known disorders include Wolf-Hirschhorn syndrome, which is caused by partial deletion of the short arm of chromosome 4; and Jacobsen syndrome, also called the terminal 11q deletion disorder.

- Duplications: A portion of the chromosome is duplicated, resulting in extra genetic material. Known disorders include Charcot-Marie-Tooth disease type 1A which may be caused by duplication of the gene encoding peripheral myelin protein 22 (PMP22) on chromosome 17.

- Translocations: When a portion of one chromosome is transferred to another chromosome. There are two main types of translocations. In a reciprocal translocation, segments from two different chromosomes have been exchanged. In a Robertsonian translocation, an entire chromosome has attached to another at the centromere; these only occur with chromosomes 13, 14, 15, 21 and 22.

- Inversions: A portion of the chromosome has broken off, turned upside down and reattached, therefore the genetic material is inverted.

- Rings: A portion of a chromosome has broken off and formed a circle or ring. This can happen with or without loss of genetic material.

- Isochromosome: Formed by the mirror image copy of a chromosome segment including the centromere.

Structural aberrations also include some disorders which are characterized by chromosomal instability and breakage. One example, is the creation of a fragile site on the X Chromosome - Fragile X syndrome. Boys are worse affected by this because they only have one X-Chromosome but even in girls, Fragile X syndrome can cause learning difficulties.

Most chromosome anomalies occur as an accident in the egg or sperm, and are therefore not inherited. The anomaly is present in every cell of the body. Some anomalies, however, can happen after conception, resulting in mosaicism (where some cells have the anomaly and some do not). Chromosome anomalies can be inherited from a parent or be "de novo". This is why chromosome studies are often performed on parents when a child is found to have an anomaly.

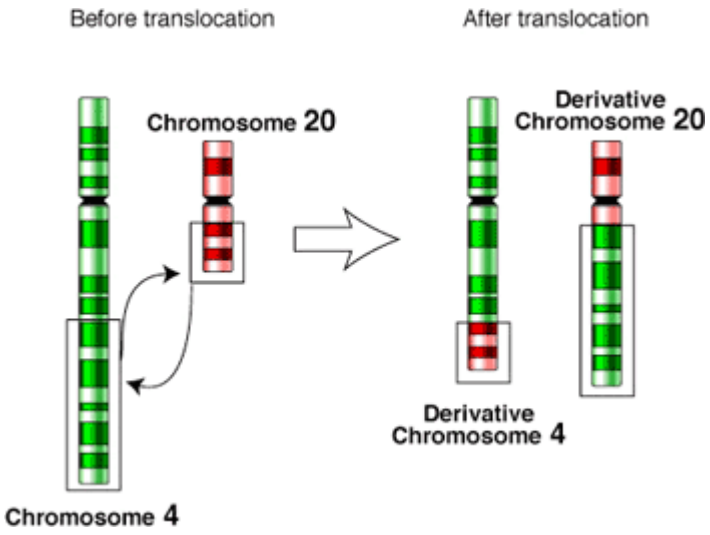

Fig. 5.3 Translocation - showing a portion of one chromosome transferred to another chromosome

SEX DETERMINATION IN MAN

In humans, sex is determined by a specific set of chromosomes. Females have two X chromosomes (XX), whereas males have an X and a Y chromosome (XY). A mature female will produce eggs, each with one X chromosome, a mature male will produce sperm with either an X chromosome or a Y chromosome.

Human Sex Differentiation

Sex differentiation, which is when a fetus gains characteristics of either males or females during development, depends on gonadal steroid hormones. These hormones perform 'organizing functions' to permanently differentiate sex organs during development. This process starts before the developing child is even old enough to be considered a fetus, and is instead still an embryo.

By the sixth week of development, all embryos have both Wolffian ducts and Müllerian ducts. In this stage, the internal organs are bipotential, meaning they have the potential to develop into both male and female sex organs. The way they develop is influenced by hormones, and each fetus will only have one of pair of these ducts by the end of differentiation.

The differentiation process is started for males by a gene on the Y chromosome known as the SRY-gene (Sex determining Region Y). When expressed, this gene initiates the correct biochemistry inside of a male fetus for him to develop male sex organs. The Wolffian ducts develop into the vas deferens and the seminal vesicle, and the Müllerian ducts degenerate. The undifferentiated gonads develop into testes, and other structures such as the prostate gland and scrotum develop. The illustration shows the male fetal sex organ development at four months.

Females have two X chromosomes, so they do not have the initiation of sex differentiation signaled by the SRY-gene; instead, their sex organs develop in the absence of these cues. The Müllerian ducts. The undifferentiated gonads develop into ovaries, and other structures such as the labia and vagina develop. The illustration shows the female fetal sex organ development at five months.

This is the sex differentiation process for mammals as a whole, but not all animals undergo this process. For example, sex differentiation in birds is seen as the reverse as in mammals. It depends on the presence of hormones to begin differentiation in the female, and a lack of those hormones to begin differentiation in the male.

Development of the External Sex Organs

The external genitalia of males and females are not as radically different as one might expect. The external genitalia are undifferentiated at four and six weeks, and then start to differentiate at ten weeks for males and twenty weeks for females. The final illustrations are the external genitalia at birth. It is interesting to realize that the same initial tissue makes up different structures in males and females, such as how the genital tubercle

develops into the glans in males and the clitoris in females.

Special Circumstances

Some people have different circumstances that prevent them from developing along the same path that others do. The results of these circumstances may be intersex individuals who have sex characteristics of both males and females. The following are three examples of hormonal circumstances that change the way that the sex organs develop in males and females.

Androgen Insensitivity Syndrome (AIS) can occur in both males and females, but really only has a large impact on males. In this syndrome, males have tissues that lack sensitivity to androgens, meaning they do not respond normally to the hormones that cause masculinization. There are different extents to which a male is insensitive, so development may vary from person to person with AIS. In the most extreme case, males will develop external genitalia that resembles the genitalia of a female. Because the testes secrete anti-Müllerian hormone and the Wolffian ducts do not receive the proper signals to develop, both duct systems degenerate.

Congenital Adrenal Hyperplasia

Congenital adrenal hyperplasia (CAH) is an affliction in females who have overactive adrenal glands. This produces extra cortisol, a hormone that is structurally and functionally similar to testosterone. These recessive genes can also be found in males, but they do not have a large impact. Females will develop masculinized genitalia to different degrees, such as enlargement of the clitoris or possibly labial fusion. If it is found early and treated, females are said to grow up to be completely normal in her 'female behaviors' and have no conflict with her sexual identity.

Sex balance theory or genic balance theory states that the X chromosome determines the sex of the individual and that sex is a dosage phenomena, where the ratio of the amount of the X relative to the autosomes determines the sex. In addition, environmental effects can influence the development of the intersex flies.

Further studies have shown that sex is ultimately determined by the locus *sex-lethal* on the X chromosome, though several other loci on the X chromosome and the autosomes are also needed for sex determination.

The sex balance theory was assumed to apply to other XY systems, including humans. However, cytologic evidence (chromosomal studies) of mice and humans showed that

1) XO were female (Turner)

2) XXY were male (klinefelter)
which is opposite of what the sex balance theory would predict. All males have at least one Y and all females have no Y's, regardless of the number of X's. The reason is that on the Y chromosome, there is a gene that causes an undifferentiated gonad to become a testis. This gene is called the sex determining region Y (*sry*). Its mode of action is basically to control a number of other genes that effect the development of the sexual characteristics.

Sex linked inheritance or X-linked Inheritance

In animals with XY sex determining mechanism, the X chromosome has many loci, many that have nothing to do with sex as such. The Y is usually smaller and possesses fewer loci that are not the same loci as that on the X chromosome. Thus females that have the same allele at a locus on the X chromosome are homozygous. Different alleles would be heterozygous. Males, because they have only one X, are hemizygous and can have only one allele at a locus. Because of this, one copy of a recessive allele will be expressed in the phenotype in males.

In sex-linked inheritance, crosses are not reciprocal. The X-linked pattern is called the criss-cross pattern of inheritance because fathers pass the trait to daughters who pass it on to sons.

Sex-limited traits are traits that are autosomally inherited, and they are expressed in one sex, but not in the other. Some examples include sexually dimorphic plumage in birds, milk yield in mammals, antlers in deer, beards in humans.

Sex-influenced traits appear in both sexes but more so in one sex than another. Male pattern baldness in humans is an example. The male hormone testosterone is needed for full expression of baldness. Because of this hormone difference, the allele for baldness behaves as a dominant trait in males (expressed when heterozygous), but behaves as a recessive allele in females (must be homozygous to be expressed).

Pedigree Analysis

Pleiotropy - a gene that causes or affects the phenotypic development in two or more characteristics. For example, the many symptoms that are seen in individuals who are homozygous for cystic fibrosis - one trait is the build-up of thick mucus in the lungs of affected individuals and a second trait is the abnormal development of the vas deferens in males, which often leads to sterility.

Penetrance - an allele that produces the same effect in every individual of the proper genotype is said to have complete penetrance. If this is not the case, the allele is said to be incompletely penetrant.

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EXERCISES

PART A

1. Define Serology.
2. What is antigen antibody reaction?
3. What are the basic principles of Serology?
4. What is Forensic Serology?
5. What is the application of Serology in Forensic Science?
6. What is karyotying?
7. What are the chromosomal abnormalities present?
8. What are banding patterns?
9. Define Sex linked inheritance.
10. What is sex determination?
11. What is agglutination?
12. What is cross reaction?
13. Depict antigen antibody reaction.
14. What is zone of equivalence?
15. What does karyotyping test is done?
16. What are allosomes?
17. Comment briefly agglutination and precipitation reactions.
18. What are serological techniques? Give some examples.
19. What is precipitation reaction?

PART B

20. Discuss the various precipitation reactions with suitable illustrations.
21. How can we quantify the concentration of antigen and antibody reactions?
22. Explain the principles and applications of different types of immunodiagnostic techniques.
23. Explain Autosomal dominant disorders.
24. Explain inherited disorders in human.
25. Explain the chromosomal abnormalities with examples.
26. Explain various disorders of Sex linked inheritance.
27. Explain karyotyping methods and its forensic significance.