COURSE INTRODUCTION

The objective of this course is to provide the basic introduction and concept to clinical biochemistry and its applications. The aim is to provide the basic concept of biochemistry, clinical enzymology, diagnostic enzymes and various disorders. The course is organized into following ten units as under:

The course is organized into following ten units as under:

Unit 1

It covers the basic concepts in clinical biochemistry and brief review of expressing concentration as well as standard solution.

Unit 2

It covers the role and regulation of electrolyte content in body fluids and maintenance of pH.

Unit 3

It covers the enzymes, hormones, plasma enzymes and isoenzymes with examples.

Unit 4

To know about diagnostic enzymes (enzymes in health and diseases) and enzyme assays such as SGPT, CPK and LDH.

Unit 5

To know about nutrition and drugs, special feeding methods, tube feeding, parenteral nutrition, drugs, alcohol and toxicants.

Unit 6

To know about regulation of blood sugar, glycogen storage diseases, diabetes mellitus and sugar levels in blood.

Unit 7

To know about low and high density lipoproteins, cholesterol, triglycerides, phospholipids in health, Gaucher's and Tay-Sach's disease.

Unit 8

It covers abnormalities in nitrogen metabolism (Uremia, hyperuricemia, porphyria) and factors affecting nitrogen balance.

Unit 9

It covers blood clotting: blood clotting mechanism-hemorrhagic disorders-hemophilia, blood groups, antigen, antibodies and circulating anticoagulants.

Unit 10

To know about types of cancer, multiple steps of tumor development, cell death, apoptosis, carcinogens and cancer therapy.

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SBSBCH-04 Clinical Biochemistry

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SBSBCH-04
Clinical Biochemistry

Unit-1

1.1 Introduction

Biochemistry or biological chemistry, is the study of chemical processes within and relating to living organisms. A sub-discipline of both chemistry and biology, biochemistry may be divided into three fields: structural biology, enzymology and metabolism. Over the last decades of the 20th century, biochemistry has become successful at explaining living processes through these three disciplines. Almost all areas of the life sciences are being uncovered and developed through biochemical methodology and research. Biochemistry focuses on understanding the chemical basis which allows biological molecules to give rise to the processes that occur within living cells and between cells, in turn relating greatly to the understanding of tissues and organs, as well as organism structure and function. Biochemistry is closely related to molecular biology, which is the study of the molecular mechanisms of biological phenomena.

Much of biochemistry deals with the structures, bonding, functions, and interactions of biological macromolecules, such as proteins, nucleic acids, carbohydrates, and lipids. They provide the structure of cells and perform many of the functions associated with life. The chemistry of the cell also depends upon the reactions of small molecules and ions. These can be inorganic (for example, water and metal ions) or organic (for example, the amino acids,

which are used to synthesize proteins). The mechanisms used by cells to harness energy from their environment via chemical reactions are known as metabolism. The findings of biochemistry are applied primarily in medicine, nutrition and agriculture. In medicine, biochemists investigate the causes and cures of diseases. Nutrition studies how to maintain health and wellness and also the effects of nutritional deficiencies. In agriculture, biochemists investigate soil and fertilizers. Improving crop cultivation, crop storage, and pest control are also goals.

Objectives

This is the first unit on clinical biochemistry. Under first unit we have following objectives. These are as under:

- To know about basic concept of clinical biochemistry.
- To know different types of concentrations and standard solution.
- To know different units, abbreviations and biomolecules .

1.1 What Is Clinical Biochemistry?

Clinical Biochemistry is the division of laboratory medicine that deals with the measurement of chemicals (both natural and unnatural) in blood, urine and other body fluids. These test results are useful for detecting health problems, determining prognosis and guiding the therapy of a patient. Clinical Biochemists ensure that consistent high quality, accurate and precise biochemical test results are provided so that high quality care can be provided to the patient. Clinical Biochemists lead the development and implementation of laboratory quality management systems that encompass all aspects of the testing process: pre-analytical, analytical and post-analytical.

1.2 The primary responsibilities of a Clinical Biochemist include:

- Interpretation of patient laboratory tests for screening, diagnosis, management and monitoring of disease processes.
- Development of interpretive guides for other professionals using the laboratory service, through the selection and validation of reference intervals, interpretive comments and critical values.

- In consultation with clinical colleagues, development, implementation and monitoring of testing algorithms, appropriate testing turnaround times, practice guidelines and care pathways.

- Provide oversight and guidance for point-of-care testing programs both in hospital and community settings.
- Development and implementation of policies and procedures to ensure the laboratory produces high quality information, and meets regulatory requirements and standards of practice.
- Selection of test methods and instrumentation.
- Assessment of the scientific and medical value of potential new tests, evaluation of the ongoing value of existing tests, in order to optimize patient care and the use of health care resources.
- Teaching and research.

Clinical Biochemists work in a variety of settings, including hospital, community and reference laboratories and in industry. The Clinical Biochemist interacts with many of other professionals, including physicians, nurses, technologists, administrators, government officials, students (medical and technical), and business personnel. The daily variety makes Clinical Biochemistry a stimulating, challenging and enjoyable profession.

1.3 Management of Clinical Biochemistry

Clinical biochemical tests are used to examine fluids in the body (blood, urine, cerebral spinal fluid, and collections in joints, abscesses, and body cavities). Besides establishing a diagnosis, they can also be used for tracking the progress of disease management. Below are some of the more common illnesses that depend on biochemical testing in management and treatment.

1.4 Diabetes

Diabetes relies completely on blood testing for its successful management and treatment. While a fasting blood glucose is an effective screen for narrowing down the more suspicious individuals in the general population, once diagnosed, both Type 1 DM and Type 2 DM are followed by interval measurements of the glycated hemoglobin A1c, which gives a months-long appraisal of glycemic control. A normal levels in non-diabetics is <6%, but in established diabetics, a target goal of 7% is ideal in preventing DM complications.

Diabetics are at increased risk for cardiovascular disease, lipid abnormalities, and kidney failure, so cardiac enzymes (if there has been chest pain or an abnormal ECG), cholesterol and triglyceride measurements that indicate the need for statin medication, and appraisals of kidney function are important parts of diabetic management.

1.5 Immunosuppression

Doctor-caused immunosuppression, e.g., chemotherapy, can be quantitated to adjust dosages and the timing of intravenous anti-cancer therapy. RF (rheumatoid factor) and ANA (antinuclear antibody), when positive, can make the difference in whether a person is treated for age-related osteoarthritis or immunological rheumatoid arthritis.

1.6 Thyroid Disease

When the thyroid function is low (hypothyroidism) or high (hyperthyroidism), metabolism is severely affected. The brain's pituitary gland acts accordingly by either lowering thyroid stimulating hormone (TSH) or making more. The TSH serves as a screen for thyroid dysfunction and its normal value can indicate successful management with medication used to stabilize thyroid function.

1.7 Liver Disease

The enzymes and proteins of the liver can be quantitated to guide the on-going therapy for liver disease and to predict the levels of liver failure that may have to be taken into account in long-term management. Bilirubin is an exact reflection of liver disease, and the severity of clinical jaundice is proportionate to elevated bilirubin in the blood and urine.

1.7 Kidney Disease

Electrolyte balance based on hydration can be followed exactly with blood tests, upon which is based medication choices for diuresis or kidney function. Dyslipidemia and Its Complications, Atherosclerosis and Cardiovascular Disease. Lipid testing is used to follow the therapeutic progress of statin drug therapy for dyslipidemia (abnormal cholesterol and triglyceride).

1.8 What Can I Prevent By Using Clinical Biochemistry?

Prevention of disease using clinical biochemistry is both by screens, to identify those at risk, and by interval assessments to prevent further disease progression. The following, among other blood and non-blood tests, are screens used for identifying those at risk:

- **Fasting glucose:** For diabetes and the hemoglobin A1c to judge how severe it has been over the previous months when the screen is positive.
- **Genetic screen for mutations:** Such as the BRCA I and II, predict those at risk for ovarian, uterine, breast, and gastrointestinal cancers; CA-125 for ovarian cancer,

carcinoma embryonic antigen (CEA) for other types of gynecological malignancies, and PSA for prostatic cancer.

- **Blood tests for amyloid beta:** Using spectrometry, to screen for those at risk for Alzheimer disease, even decades before any onset is suspected.
- **In pregnancy:** Amniotic fluid can be evaluated to determine lung maturity for a pre-term infant who may benefit from early delivery necessary due to maternal illness. Noninvasive prenatal testing (NIPT) can screen for Down syndrome and can even reveal the gender of a baby during pregnancy.
- **Testing for allergens:** Via ELISA and RAST testing is used to detect allergies, but can also screen those at risk for life-threatening anaphylactoid reactions.
- **Biochemical identification of HPV in Pap smears:** Can prevent pre-cancerous conditions (dysplasia) from progressing into cervical cancer.
- **Antibody screens:** Are useful both to prevent blood transfusion reactions and to indicate which patients may need immunizations to prevent specific contagious diseases. They are also used to confirm successful immunization or the need for boosters.
- **Titers of blood concentrations:** Can be used to screen for infectious diseases such as syphilis, but can also prevent further progress by alerting the physician when titers plateau or rise, indicating when treatment failure with one treatment modality calls for another.

Biochemistry revolves around reproduction processes that cause chemical reactions and combinations such as reproduction, metabolism and growth. Applicants who have passed high school or 10+2 with mathematics and biology as the main subjects are eligible for this course. People who choose biochemistry as a career path are generally known as Biochemists. The scope of biochemistry is so immense that even lucrative offers are provided by public sector companies like blood service, research departments, agriculture and fisheries, drug manufacturing firms, forensic science, hospitals, public health organizations.

1.9 Scope of Biochemistry: Quick Facts

- The development of life took us from the little microorganism to apes, to the people which we have now become. All that study falls under one of the parts of Biochemistry. Furthermore, it is a review that is supposed to be the language of nature.

Every one of the instruments that you find in the field of clinical science is biochemical.

- Biotechnology is utilizing hereditary designing, which is one of the most modern parts of science. The utilization of biotechnology can be found in the field of clinical science, animal cultivating, obsessive examination, and so forth.
- In conclusion, one more branch we might want to refer to is medical chemistry, and this is one of the fundamental parts of the entire science as it manages both the wellbeing and the illness of the human body.
- As a field Biochemistry has seen phenomenal development owing to its immense contribution towards the illumination and seizing of the DNA structure.
- Scope of biochemistry allows biochemists to examine structures and roles of proteins, enzymes, fats, processes of metabolism.

1.10 Scope of Biochemistry in Different Fields

As a biochemist, one can take the road of opening their laboratories. Apart from that, other fields are-

- Physician associate
- Analytical chemist
- Biotechnologist
- Healthcare scientist, clinical biochemistry
- Clinical research associate
- Research scientist (life sciences)
- Toxicologist
- Biomedical scientist
- Forensic scientist
- Nanotechnologist

1.11 Scope of Biochemistry in Agriculture

- Agriculture Biochemistry focuses mainly on the subject of biochemistry that makes it applicable to agricultural scientists specifically. Agriculture Biochemistry deals with the chemistry of animals and plants metabolism.
- Agritech is a 4-year undergraduate course. Ph.D. Chemists toil in the agricultural chemical fields. B.Sc with zoology, biochemistry, botany, B.Sc agriculture (55%marks).

- Agricultural chemists spadework at government agencies in the industry domain and as far as academia is concerned, teaching, research, outreach are other options available. Working with food producers is another field of interest to help with the production of fungicides, fertilizers, insecticides.

1.12 Scope of Biochemistry in Pharmacy

- All India Council for Technical Education has pushed the number of seats across the country for Biochemistry in Pharmacy looking at the growing demand for undergraduate, postgraduate and diploma degrees.
- Biochemist graduates in pharmacy are engaged with the innovative work of new therapeutics just to guarantee their assembling quality control. The interest for pharma graduates is high in areas like - medical care, research, fabricating, clinical promoting, pharmacovigilance and so on.
- Career options: Assistant Professor, Scientist, Quality Control, Regulatory Affairs, Research Associate. Other options are drugs monitor, drugs regulator, emergency clinic drug specialist.
- Salary differs from INR 3-15 LPA depending upon the experience.

1.13 Scope of Biochemistry in Nursing

- Biochemistry concentrates on different exercises that occur in the body of a living being and the substance measures that go on in the body. Nurses need to have decent information on this sort of science since it manages natural matter and its response to various types of synthetic substances.
- The medium salary package offered to graduates of BSc Nursing varies from INR 3.2 lakhs to INR 7.8 LPA.
- The freshers after completing BSc Nursing can get salaries varying from INR 7,000 to INR 15000. After 2 to 3 years, the salary increases and ranges between INR 20,000 to INR 30,000.
- You can continue higher studies in MSc Neuroscience, MSc Biochemistry, MSc Nursing, MSc Medical Microbiology and MSc Biotechnology.
- Medical Advisor, Nursing Executive, Nursing In-charge, Nursing Supervisor, Nursing Tutor, Occupational Health Nurse, Paramedic Nurse, Ward Nurse, Asst. Professor, etc. are various career options.

- Top recruiters - Nursing Homes, Military Hospitals, Medicine Industry, Medical Colleges and Universities, Healthcare Centres, Content Writing (Medical), etc.

1.14 Scope of Clinical Biochemistry

- Clinical biochemistry is concerned with biochemical features that are entangled in several conditions. Quantitative and qualitative analysis of body fluids aids the clinicians in the diagnosis and treatment of the disease along with organ transplantation, tissue, drug monitoring.
- Clinical biochemistry is used in clinical prognosis, manufacturing of different biological products, nutrition, treatment of diseases. Clinical biochemistry also deals with disclosing the abnormalities in the metabolism.
- This field plays a pivotal role in the health sector through curation and prevention of a nation's health delivery network.
- Clinical biochemists also take care of the development of any disease and treatment of the patients.
- Career options: Hospital Laboratories are generally one domain of work for clinical biochemists. Other areas can be clinics, operating theatres.
- Top recruiters: Ajanta Pharmaceuticals, Apollo Health Care, Aurobindo Pharma, Biocon, Cipla, Reddy's Laboratories, Ranbaxy

1.15 Units of concentration

1.16 What does concentration mean?

In the sciences, there are basically two different meanings for concentration; first, how close things are to each other, and second, the composition of substances within a mixture. In General Chemistry 1 we used concepts like density and molarity to describe the first concept, and mass percent and mole fraction to describe the second.

How dense (packed) things are

- **Density** (d=mass/volume)
- **Molarity** (M=moles/volume)

Note, the above can be used to describe a pure substance or a mixture. Molarity was very useful for identifying the number of solute particles in a solution as you could only measure the total solution (either its mass or volume), but could not directly measure its components.

- Relative composition of a mixture
- **Mass Percent** (Mass % A=100 (MassA/ MassTotal)

- **Mole Fraction** (XA = (molesA/ molesTotal)

Note, the above can be used to describe the relative composition of homogenous mixtures (solutions), but don't really tell you about how close things are to each other. For example, we used the concept of mole fraction to describe the partial pressure of a gas, where the partial pressure was the mole fraction times the total pressure, but that told you nothing about how close the molecules were to each other

It should also be noted that "percent" is simply the "fraction times 100", and so they are two different ways of stating the same relationship, and that it is a matter of convention to use percent for mass and fraction for the number of molecules, but you could also have "mass fraction" and "mole percent".

1.16 Units of Concentration

Solutions are homogeneous mixtures containing one or more **solutes** in a **solvent**. The solvent that makes up most of the solution, whereas a solute is the substance that is dissolved inside the solvent.

1.17 Relative Concentration Units

Concentrations are often expressed in terms of relative unites (e.g. percentages) with three different types of percentage concentrations commonly used:

1. **Mass Percent**: The mass percent is used to express the concentration of a solution when the mass of a solute and the mass of a solution is given:

$$\text{Mass Percent} = \text{Mass of Solute} / \text{Mass of Solution} \times 100\ \% \quad (1)$$

2. **Volume Percent**: The volume percent is used to express the concentration of a solution when the volume of a solute and the volume of a solution is given:

$$\text{Volume Percent} = \text{Volume of Solute} / \text{Volume of Solution} \times 100\% \quad (2)$$

Mass/Volume Percent: Another version of a percentage concentration is mass/volume percent, which measures the mass or weight of solute in grams (e.g., in grams) vs. the volume of solution (e.g., in mL). An example would be a 0.9% (w/v) NaCl. NaCl solution in medical saline solutions that contains 0.9 g of NaCl for every 100 mL of solution.

The mass/volume percent is used to express the concentration of a solution when the mass of the solute and volume of the solution is given. Since the numerator and denominator have different units, this concentration unit is not a true relative unit (e.g. percentage), however it is often used as an easy concentration unit since volumes of solvent and solutions are easier to

measure than weights. Moreover, since the density of dilute aqueous solutions are close to 1 g/mL, if the volume of a solution in measured in mL (as per definition), then this well approximates the mass of the solution in grams (making a true reletive unit (m/m)).

$$\text{Mass/Volume Percent} = \text{Mass of Solute (g)}/\text{Volume of Solution (mL)} \times 100\% \quad (3)$$

1.18 Dilute Concentrations Units

Sometimes when solutions are too dilute, their percentage concentrations are too low. So, instead of using really low percentage concentrations such as 0.00001% or 0.000000001%, we choose another way to express the concentrations. This next way of expressing concentrations is similar to cooking recipes. For example, a recipe may tell you to use 1 part sugar, 10 parts water. As you know, this allows you to use amounts such as 1 cup sugar + 10 cups water in your equation. However, instead of using the recipe's "1 part per ten" amount, chemists often use *parts per million*, *parts per billion* or *parts per trillion* to describe dilute concentrations.

Parts per Million: A concentration of a solution that contained 1 g solute and 1000000 mL solution (same as 1 mg solute and 1 L solution) would create a very small percentage concentration. Because a solution like this would be so dilute, the density of the solution is well approximated by the density of the solvent; for water that is 1 g/mL (but would be different for different solvents). So, after doing the math and converting the milliliters of solution into grams of solution (assuming water is the solvent):

$$1 \text{ g solute}/1000000 \text{ mL solution} \times 1 \text{ mL}1 \text{ g} = 1 \text{ g solute}/1000000 \text{ g solution}$$

We get (1 g solute)/(1000000 g solution). Because both the solute and the solution are both now expressed in terms of grams, it could now be said that the solute concentration is 1 part per million (ppm).

$$1 \text{ ppm} = 1 \text{ mg Solute}1/\text{ L Solution}$$

The ppm unit can also be used in terms of volume/volume (v/v) instead.

1.19 Parts per Billion: Parts per billion (ppb) is almost like ppm, except 1 ppb is 1000-fold more dilute than 1 ppm.

$$1 \text{ ppb} = 1\mu\text{g Solute}/1 \text{ L Solution}$$

1.20 Parts per Trillion: Just like ppb, the idea behind parts per trillion (ppt) is similar to that of ppm. However, 1 ppt is 1000-fold more dilute than 1 ppb and 1000000-fold more dilute than 1 ppm.

$$1 \text{ ppt} = 1 \text{ ng Solute}/1 \text{ L Solution}$$

1.21 Concentration Units based on moles

Mole Fraction: The mole fraction of a substance is the fraction of all of its molecules (or atoms) out of the total number of molecules (or atoms). It can also come in handy sometimes when dealing with the PV=nRTPV = nRT equation.

$$\chi A = \text{number of moles of substance A}/\text{total number of moles in solution}$$

Also, keep in mind that the sum of each of the solution's substances' mole fractions equals 1.

$$\chi A + \chi B + \chi C + ... = 1$$

1.22 Mole Percent: The mole percent (of substance A) is $\chi A \chi A$ in percent form.

$$\text{Mole percent (of substance A)} = \chi A \times 100\%$$

Molarity: The molarity (M) of a solution is used to represent the amount of moles of solute per liter of the solution.

$$M = \text{Moles of Solute}/\text{Liters of Solution}$$

Molality: The molality (m) of a solution is used to represent the amount of moles of solute per kilogram of the solvent.

$$m = \text{Moles of Solute}/\text{ Kilograms of Solvent}$$

Fig. 1 Different molarities of liquids in the laboratory. 50 ml of distilled water (0 M), Sodium Hydroxide solution of 0.1 M, and Hydrochloric acid solution of 0.1 M.

The molarity and molality equations differ only from their denominators. However, this is a huge difference. As you may remember, volume varies with different temperatures. At higher temperatures, volumes of liquids increase, while at lower temperatures, volumes of liquids decrease. Therefore, molarities of solutions also vary at different temperatures. This creates an advantage for using molality over molarity. Using molalities rather than molarities for lab experiments would best keep the results within a closer range. Because volume is not a part of its equation, it makes molality independent of temperature.

1.23 Standard solution

In analytical chemistry, a standard solution is a solution containing a precisely known concentration of an element or a substance. A known mass of solute is dissolved to make a specific volume. It is prepared using a standard substance, such as a primary standard. Standard solutions are used to determine the concentrations of other substances, such as solutions in titration.

The concentrations of standard solutions are normally expressed in units of moles per litre (mol/L, often abbreviated to M for molarity), moles per cubic decimetre (mol/dm^3), kilomoles per cubic metre ($kmol/m^3$) or in terms related to those used in particular titrations (such as titres A simple standard is obtained by the dilution of a single element or a substance in a soluble solvent with which it reacts. A primary standard is a reagent that is extremely pure, stable, has no waters of hydration, and has high molecular weight. Some primary standards of titration of acids include sodium carbonate.

Uses

A known volume of a solution of acid can be standardized by titrating it against a solution of alkali of known concentration. Standard solutions are also commonly used to determine the concentration of an analyte species. By comparing the absorbance of the sample solution at a specific wavelength to a series of standard solutions at differing known as concentrations of the analyse species, the concentration of the sample solution can be found via Beer's Law. Any form of spectroscopy can be used in this way so long as the analyte species has substantial absorbance in the spectra. The standard solution is a reference guide to discover

the molarity of unknown species. Titration methods can be used to acquire the concentration of a standard solution. These involve using equipment such as a burette.

1.24 Properties

The properties of a standard solution for titrations are:

1. Its concentration must remain constant all the time. This is so that there is no need for restandardization.
2. Its reaction with the analyte must be rapid in order to minimize the waiting period after addition of each reagent.
3. Its reaction must be reasonably complete.
4. It should be possible to describe the reaction by a balanced chemical reaction.
5. A method must exist for detecting the equilibrium point.

1.2.5 Summary

In this unit we summarize that the biochemistry is the branch of science that explores the chemical processes within and related to living organisms. It is a laboratory based science that brings together biology and chemistry. By using chemical knowledge and techniques, biochemists can understand and solve biological problems Biochemistry focuses on processes happening at a molecular level. It focuses on what's happening inside our cells, studying components like proteins, lipids and organelles. It also looks at how cells communicate with each other, for example during growth or fighting illness. Biochemists need to understand how the structure of a molecule relates to its function, allowing them to predict how molecules will interact. Biochemistry covers a range of scientific disciplines, including genetics, microbiology, forensics, plant science and medicine. Because of its breadth, biochemistry is very important and advances in this field of science over the past 100 years have been staggering. It's a very exciting time to be part of this fascinating area of study.

Biochemistry is the application of chemistry to the study of biological processes at the cellular and molecular level. It emerged as a distinct discipline around the beginning of the 20th century when scientists combined chemistry, physiology, and biology to investigate the chemistry of living systems. Biochemists are interested, for example, in mechanisms of brain function, cellular multiplication and differentiation, communication within and between cells and organs, and the chemical bases of inheritance and disease. The biochemist seeks to determine how specific molecules such as proteins, nucleic acids, lipids, vitamins, and hormones function in such processes. Particular emphasis is placed on the regulation of chemical reactions in living cells.

1.2.6 Terminal questions

Q. 1 What do you mean by clinical biochemistry? Describe it.

Answer:----------

Q.2 What are the scopes of biochemistry in different fields?

Answer:----------

Q.3 Describe the different units of concentration.

Answer:----------

Q.4 Explain relative concentration of units?

Answer:----------

Q.5 What are the differences between active and passive immunity?

Answer:----------

Q.6 Write a short note on following.

(a) Immunosuppression

(b) Thyroid Disease

Answer:----------

Q.6 Write a short note on following.

(a) Diabetes

(b) Liver disease

Answer:----------

Q.7 Explain scopes of biochemistry in nursing.

Answer:----------

Further readings

1. Biochemistry- Lehninger A.L.
2. Biochemistry –J.H.Weil.
3. Biochemistry fourth edition-David Hames and Nigel Hooper.
4. Textbook of Biochemistry for Undergraduates - Rafi, M.D.
5. Biochemistry and molecular biology- Wilson Walker.

Unit-2

2.1 Introduction

Electrolyte, in chemistry and physics, substance that conducts electric current as a result of a dissociation into positively and negatively charged particles called ions, which migrate toward and ordinarily are discharged at the negative and positive terminals (cathode and anode) of an electric circuit, respectively. The most familiar electrolytes are acids, bases, and salts, which ionize when dissolved in such solvents as water or alcohol. Many salts, such as sodium chloride, behave as electrolytes when melted in the absence of any solvent; and some, such as silver iodide, are electrolytes even in the solid state.

Electrolytes are essential for basic life functioning, such as maintaining electrical neutrality in cells, generating and conducting action potentials in the nerves and muscles. Sodium, potassium, and chloride are the significant electrolytes along with magnesium, calcium, phosphate, and bicarbonates. Electrolytes come from our food and fluids. These electrolytes can have an imbalance, leading to either high or low levels. High or low levels of electrolytes disrupt normal bodily functions and can lead to even life-threatening complications. This article reviews the basic physiology of electrolytes and their abnormalities, and the consequences of electrolyte imbalance.

Objectives

This is the second unit on clinical biochemistry. Under second unit we have following objectives. These are as under:

- To know about electrolytes and acid-base balance and buffer solution
- To know about role and regulation of electrolyte content.
- To know about pH and its importance.
- To discuss body fluids and fluid compartments

2.2 Electrolyte

An electrolyte is a medium containing ions that is electrically conducting through the movement of ions, but not conducting electrons. This includes most soluble salts, acids, and bases dissolved in a polar solvent, such as water. Upon dissolving, the substance separates into cations and anions, which disperse uniformly throughout the solvent. Solid-state electrolytes also exist. In medicine and sometimes in chemistry, the term electrolyte refers to the substance that is dissolved.

Electrically, such a solution is neutral. If an electric potential is applied to such a solution, the cations of the solution are drawn to the electrode that has an abundance of electrons, while the anions are drawn to the electrode that has a deficit of electrons. The movement of anions and cations in opposite directions within the solution amounts to a current. Some gases, such as hydrogen chloride (HCl), under conditions of high temperature or low pressure can also function as electrolytes. Electrolyte solutions can also result from the dissolution of some biological (e.g., DNA, polypeptides) or synthetic polymers (e.g., polystyrene sulfonate), termed "polyelectrolytes", which contain charged functional groups.

A substance that dissociates into ions in solution or in the melt acquires the capacity to conduct electricity. Sodium, potassium, chloride, calcium, magnesium, and phosphate in a liquid phase are examples of electrolytes. In medicine, electrolyte replacement is needed when

a person has prolonged vomiting or diarrhea, and as a response to sweating due to strenuous athletic activity. Commercial electrolyte solutions are available, particularly for sick children (such as oral rehydration solution, Suero Oral, or Pedialyte) and athletes (sports drinks). Electrolyte monitoring is important in the treatment of anorexia and bulimia. Electrolytes are minerals in your body that have an electric charge. They are in your blood, urine, tissues, and other body fluids. Electrolytes are important because they help:

- Balance the amount of water in your body
- Balance your body's acid/base (pH) level
- Move nutrients into your cells
- Move wastes out of your cells
- Make sure that your nerves, muscles, the heart, and the brain work the way they should Sodium, calcium, potassium, chloride, phosphate, and magnesium are all electrolytes.

You get them from the foods you eat and the fluids you drink. The levels of electrolytes in your body can become too low or too high. This can happen when the amount of water in your body changes. The amount of water that you take in should equal the amount you lose. If something upsets this balance, you may have too little water (dehydration) or too much water (overhydration). Some medicines, vomiting, diarrhea, sweating, and liver or kidney problems can all upset your water balance.

2.3 Examples of electrolytes

2.3.1 Sodium

Sodium, which is an osmotically active cation, is one of the most important electrolytes in the extracellular fluid. It is responsible for maintaining the extracellular fluid volume, and also for regulation of the membrane potential of cells. Sodium is exchanged along with potassium across cell membranes as part of active transport. Sodium regulation occurs in the kidneys. The proximal tubule is where the majority of sodium reabsorption takes place. In the distal convoluted tubule, sodium undergoes reabsorption. Sodium transport takes place via sodium-chloride symporters, which are by the action of the hormone aldosterone.

Among the electrolyte disorders, hyponatremia is the most frequent. Diagnosis is when the serum sodium level is less than 135 mmol/L. Hyponatremia has neurological manifestations. Patients may present with headaches, confusion, nausea, delirium. Hypernatremia presents when the serum sodium levels are greater than145 mmol/L. Symptoms of hypernatremia

include tachypnea, sleeping difficulty, and feeling restless. Rapid sodium corrections can have serious consequences like cerebral edema and osmotic demyelination syndrome.

2.3.2 Potassium

Potassium is mainly an intracellular ion. The sodium-potassium adenosine triphosphatase pump has the primary responsibility for regulating the homeostasis between sodium and potassium, which pumps out sodium in exchange for potassium, which moves into the cells. In the kidneys, the filtration of potassium takes place at the glomerulus. The reabsorption of potassium takes place at the proximal convoluted tubule and thick ascending loop of Henle. Potassium secretion occurs at the distal convoluted tubule. Aldosterone increases potassium secretion. Potassium channels and potassium-chloride cotransporters at the apical membrane also secrete potassium.

Potassium disorders are related to cardiac arrhythmias. Hypokalemia occurs when serum potassium levels under 3.6 mmol/L—weakness, fatigue, and muscle twitching present in hypokalemia. Hyperkalemia occurs when the serum potassium levels are above 5.5 mmol/L, which can result in arrhythmias. Muscle cramps, muscle weakness, rhabdomyolysis, myoglobinuria are presenting signs and symptoms in hyperkalemia.

2.3.3 Calcium

Calcium has a significant physiological role in the body. It is involved in skeletal mineralization, contraction of muscles, the transmission of nerve impulses, blood clotting, and secretion of hormones. The diet is the predominant source of calcium. It is mostly present in the extracellular fluid. Absorption of calcium in the intestine is primarily under the control of the hormonally active form of vitamin D, which is 1,25-dihydroxy vitamin D3. Parathyroid hormone also regulates calcium secretion in the distal tubule of kidneys. Calcitonin acts on bone cells to increase the calcium levels in the blood.

Hypocalcemia diagnosis requires checking the serum albumin level to correct for total calcium, and the diagnosis is when the corrected serum total calcium levels are less than 8.8 mg/dl, as in vitamin D deficiency or hypoparathyroidism. CHecking serum calcium levels is a recommended test in post-thyroidectomy patients. Hypercalcemia is when corrected serum total calcium levels exceed 10.7 mg/dl, as seen with primary hyperparathyroidism. Humoral hypercalcemia presents in malignancy, primarily due to PTHrP secretion.

2.3.4 Bicarbonate

The acid-base status of the blood drives bicarbonate levels. The kidneys predominantly regulate bicarbonate concentration and are responsible for maintaining the acid-base balance. Kidneys reabsorb the filtered bicarbonate and also generate new bicarbonate by net acid excretion, which occurs by excretion of both titrable acid and ammonia. Diarrhea usually results in loss of bicarbonate, thus causing an imbalance in acid-base regulation.

2.3.5 Magnesium

Magnesium is an intracellular cation. Magnesium is mainly involved in ATP metabolism, contraction and relaxation of muscles, proper neurological functioning, and neurotransmitter release. When muscle contracts, calcium re-uptake by the calcium-activated ATPase of the sarcoplasmic reticulum is brought about by magnesium. Hypomagnesemia occurs when the serum magnesium levels are less under 1.46 mg/dl. It can present with alcohol use disorder and gastrointestinal and renal losses—ventricular arrhythmias, which include torsades de pointes seen in hypomagnesemia.

2.3.6 Chloride

Chloride is an anion found predominantly in the extracellular fluid. The kidneys predominantly regulate serum chloride levels. Most of the chloride, which is filtered by the glomerulus, is reabsorbed by both proximal and distal tubules (majorly by proximal tubule) by both active and passive transport. Hyperchloremia can occur due to gastrointestinal bicarbonate loss. Hypochloremia presents in gastrointestinal losses like vomiting or excess water gain like congestive heart failure.

2.3.7 Phosphorus

Phosphorus is an extracellular fluid cation. Eighty-five percent of the total body phosphorus is in the bones and teeth in the form of hydroxyapatite; the soft tissues contain the remaining 15%. Phosphate plays a crucial role in metabolic pathways. It is a component of many metabolic intermediates and, most importantly of adenosine triphosphate (ATPs) and nucleotides. Phosphate is regulated simultaneously with calcium by Vitamin D3, PTH, and calcitonin. The kidneys are the primary avenue of phosphorus excretion. Phosphorus imbalance may result due to three processes: dietary intake, gastrointestinal disorders, and excretion by the kidneys.

2.3.8 Acid-base balance

To maintain homeostasis, the human body employs many physiological adaptations. One of these is maintaining an acid-base balance. In the absence of pathological states, the pH of the human body ranges between 7.35 to 7.45, with the average at 7.40. Why this number? Why

not a neutral number of 7.0 instead of a slightly alkaline 7.40? A pH at this level is ideal for many biological processes, one of the most important being the oxygenation of blood. Also, many of the intermediates of biochemical reactions in the body become ionized at a neutral pH, which causes the utilization of these intermediates to be more difficult.

A pH below 7.35 is an acidemia, and a pH above 7.45 is an alkalemia. Due to the importance of sustaining a pH level in the needed narrow range, the human body contains compensatory mechanisms. It intends to impart a basic understanding of acid-base balance in the body while providing a systematic way to approach patients who present with conditions causing alterations in pH.

The human body experiences four main types of acid-based disorders: metabolic acidosis, metabolic alkalosis, respiratory acidosis, and respiratory alkalosis. If one of these conditions occurs, the human body should induce a counterbalance in the form of an opposite condition. For example, if a person is experiencing a metabolic acidemia, their body will attempt to induce a respiratory alkalosis to compensate. It is rare for the compensation to make the pH completely normal at 7.4. When using the term acidemia or alkalemia, one is denoting that overall the pH is acidic or alkalotic, respectively. While not necessary, it can be useful to employ this terminology to distinguish between individual processes and the overall pH status of the patient since multiple imbalances can happen at the same time.

2.3.9 Cellular

A basic comprehension of respiration at the cellular level is important in understanding acid-base equilibrium in the human body. Aerobic cellular respiration is necessary for human life; humans are obligate aerobes. While individual cells can perform anaerobic respiration, in order to sustain life, oxygen must be present. One of the byproducts of aerobic cellular respiration is carbon dioxide. The simplified chemical equation denoting aerobic cellular respiration is:

$$C_6H_{12}O_6 \text{ (glucose)} + 6O_2 \text{ --> } 6CO_2 + 6H_20 + \text{energy (38 ATP molecules and heat)}$$

The first stage of cellular respiration is glycolysis, which takes a six-carbon glucose and breaks it down into two pyruvate molecules which contain three carbons each. Glycolysis uses two ATP and creates four ATP, meaning it generates two net ATP. This process does not need oxygen to occur. Since patients are often deficient, it is worth noting that magnesium is a cofactor for two reactions in glycolysis.

Eventually, the pyruvate molecules are oxidized and enter into the TCA Cycle. The TCA cycle generates NADH from NAD+, FADH2 from FAD, and two ATP molecules. It is an aerobic process and does demand oxygen. Pyruvate is brought into the mitochondria and forms acetyl-CoA with the loss of carbon dioxide. This excess carbon dioxide is then exhaled during the process of expiration. The last step in aerobic cellular respiration is the electron transport chain (ETC). The ETC produces the majority of the ATP created in cellular respiration with 34 ATP molecules being created. For the ETC reaction to occur, oxygen is needed. If there is not enough oxygen present, the products of glycolysis proceed to a reaction called fermentation to produce ATP. The byproduct of fermentation is lactic acid. During glycolysis and the TCA cycle, NAD+ is reduced to NADH and FAD is reduced to FADH2. Reduction is characterized by a gain of electrons. This is what drives the ETC. For every single molecule of glucose, ten NAD+ molecules are converted to NADH molecules, which produce three ATP molecules a piece in the ETC.

This process of aerobic cellular respiration characterizes why humans need oxygen. Anaerobic respiration allows the body to produce some ATP when there is not enough oxygen present; however, the process only generates two ATP as opposed to the 38 ATP produced with aerobic respiration. The two ATP molecules per reaction are not enough to sustain life. As noted above, carbon dioxide is produced as a byproduct of the TCA cycle. This carbon dioxide is instrumental to acid-base balance in the body which is demonstrated with the following reaction:

$$CO_2 + H_20 <-> H_2C0_3 <-> HCO_3- + H^+$$

The carbon dioxide formed during cellular respiration combines with water to create carbonic acid. Carbonic acid then dissociates into bicarbonate and a hydrogen ion. This reaction is one of the many buffer systems in the human body; it resists dramatic changes in pH to allow a person to remain within the narrow physiological pH range. This buffer system is in equilibrium, that is, all components of the reaction exist throughout the body and are shifted to the side of the equation appropriate for the environment. This reaction can and does occur without an enzyme; however, carbonic anhydrase is an enzyme that assists with this process. It catalyzes the first reaction above to form carbonic acid which can then freely dissociate into bicarbonate and a hydrogen ion. Carbonic anhydrase is located in red blood cells, renal tubules, gastric mucosa, and pancreatic cells.

Other buffer systems in the human body include the phosphate buffer system, proteins, and hemoglobin. All of these contain bases which accept hydrogen ions which keep the pH from plummeting. The phosphate buffer system, while present globally, is important for the regulation of urine pH. Proteins assist with intracellular pH regulation. Red blood cells use the reaction above to help hemoglobin buffer; carbon dioxide can diffuse across red blood cells and combine with water. This alone would cause an increase in hydrogen ions; however, hemoglobin can bind hydrogen ions. Hemoglobin also can bind carbon dioxide without this reaction. This depends on the amount of oxygen that is bound to hemoglobin. This is called the Haldane effect and the Bohr effect. When hemoglobin is saturated with oxygen, it has a lower affinity for CO_2 and hydrogen ions and is able to release it.

2.4 Importance of Electrolyte Balance

Electrolytes play a vital role in maintaining homeostasis within the body. They help regulate myocardial and neurological function, fluid balance, oxygen delivery, acid-base balance, and other biological processes. Electrolytes are important because they are what cells (especially those of the nerve, heart, and muscle) use to maintain voltages across their cell membranes and to carry electrical impulses (nerve impulses, muscle contractions) across themselves and to other cells. Electrolyte imbalances can develop from excessive or diminished ingestion and from the excessive or diminished elimination of an electrolyte. The most common cause of electrolyte disturbances is renal failure. The most serious electrolyte disturbances involve abnormalities in the levels of sodium, potassium, and/or calcium.

Other electrolyte imbalances are less common, and often occur in conjunction with major electrolyte changes. Chronic laxative abuse or severe diarrhea or vomiting (gastroenteritis) can lead to electrolyte disturbances combined with dehydration. People suffering from bulimia or anorexia nervosa are especially at high risk for an electrolyte imbalance. Kidneys work to keep the electrolyte concentrations in blood constant despite changes in your body. For example, during heavy exercise electrolytes are lost through sweating, particularly sodium and potassium, and sweating can increase the need for electrolyte (salt) replacement. It is necessary to replace these electrolytes to keep their concentrations in the body fluids constant.

2.5 Dehydration

There are three types of dehydration:

1. Hypotonic or hyponatremic (primarily a loss of electrolytes, sodium in particular).
2. Hypertonic or hypernatremic (primarily a loss of water).

3. Isotonic or isonatremic (an equal loss of water and electrolytes).

In humans, the most common type of dehydration by far is isotonic (isonatraemic) dehydration; which effectively equates with hypovolemia; but the distinction of isotonic from hypotonic or hypertonic dehydration may be important when treating people with dehydration.

Physiologically, and despite the name, dehydration does not simply mean loss of water, as both water and solutes (mainly sodium) are usually lost in roughly equal quantities as to how they exist in blood plasma. In hypotonic dehydration, intravascular water shifts to the extravascular space and exaggerates the intravascular volume depletion for a given amount of total body water loss.

2.6 What is pH?

The pH is a negative logarithmic scale, which measures the molar concentration of hydrogen ions from 1 to 10^{14} ions. It therefore has no unit of measurement. Numbers on a negative logarithmic scale decrease in magnitude by an order of 10 from the previous one. At a pH of 1, the hydrogen ion concentration is 10 times higher than that at 2.
The pH is thus expressed as

$$\mathbf{pH = \log 1/[H+] = -\log [H+].}$$

The square bracket is a symbol of the concentration of the substance.

Pure water has a pH of 7, which is considered perfectly neutral. This means that it has exactly equal numbers of hydrogen and hydroxyl ions. Any substance which is to the left of this point on the scale is considered acidic. All substances whose pH falls to the right of 7 on the scale are basic. Just as an example, a substance with a pH of 4 on the scale is 3 points down from water with a pH of 7. On the logarithmic scale, this means a difference of 10^3 or 1000. Thus, the hypothetical substance is 1000 times more acidic than pure water. The strongest possible acid has one hundred trillion times more hydrogen ions than the weakest does (the weakest acid being the strongest base).

Alkalinity measures the ability of a solution to neutralize an acid. It may also describe the ability of water to act as a buffer, keeping the pH stable despite small changes in the chemistry of the water by the addition of small amounts of acids or bases. In natural water, alkalinity is caused by the weak acid salts present in it such as bicarbonates.

2.7 Understanding acids and alkalis

In pure water, a small portion of the molecules lose one hydrogen from the H_2O structure, in a process called dissociation. The water thus contains a small number of hydrogen ions, H+, and residual hydroxyl ions, OH-. There is equilibrium between the constant formation and dissociation of a small percentage of water molecules. Hydrogen ions (OH^-) in water join with other water molecules to form hydronium ions, H_3O^+ ions, which are more commonly and simply called hydrogen ions. Since these hydroxyl and hydronium ions are in equilibrium, the solution is neither acidic nor alkaline. An acid is a substance which donates hydrogen ions into solution, while a base or alkali is one which takes up hydrogen ions.

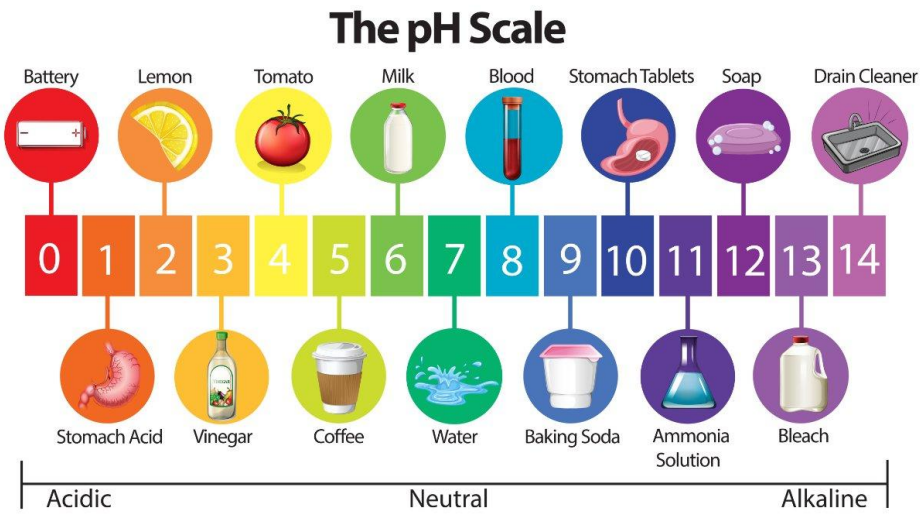
All substances that contain hydrogen are not acidic as the hydrogen must be present in a state that is easily released, unlike in most organic compounds which bind hydrogen to carbon atoms very tightly. The pH thus helps to quantify the strength of an acid by showing how many hydrogen ions it releases into solution.

Hydrochloric acid is a strong acid because the ionic bond between the hydrogen and the chloride ions is a polar one which is easily dissolved in water, generating many hydrogen ions and making the solution strongly acidic. This is why it has a very low pH. This kind of dissociation within water is also very favorable in terms of energetic gain, which is why it happens so easily. Weak acids are compounds which do donate hydrogen but not very readily, such as some organic acids. Acetic acid, found in vinegar, for instance, contains a lot of hydrogen but in a carboxylic acid grouping, which holds it in covalent or nonpolar bonds.

As a result, only one of the hydrogens is able to leave the molecule, and even so, there is not much stability gained by donating it. A base or alkali accepts hydrogen ions, and when added to water, it soaks up the hydrogen ions formed by the dissociation of water so that the balance shifts in favor of the hydroxyl ion concentration, making the solution alkaline or basic. An example of a common base is sodium hydroxide, or lye, used in making soap. When an acid and an alkali are present in exactly equal molar concentrations, the hydrogen and hydroxyl ions react readily with each other, producing a salt and water, in a reaction called neutralization.

2.8 pH in the Human Body

The pH of the human body lies in a tight range between 7.35-7.45, and any minor alterations from this range can have severe implications.



One of the most important indicators for water quality is its pH level. The pH scale runs from 0 to 14 and measures the acid or base quality of water. A pH of 7 is neutral, while a reading below 7 is acidic, and one above 7 is alkaline or basic. Water quality depends on proper pH levels. In acidic water, for example, toxic heavy metals dissolve easily and are more harmful to living things. The pH level also affects the availability of essential plant nutrients, with many nutrients being less available at a pH above 7

2.9 pH and Drinking Water

There is no legally enforceable standard for drinking water pH levels because pH is considered an aesthetic water quality. However, the U.S. Environmental Protection Agency (EPA) recommends a pH between 6.5 and 8.5 for drinking water. Since metals dissolve readily in acidic water, dissolved metals may be present in drinking water with a low pH level. Metals such as iron, manganese, copper, and lead can leach into drinking water from pipes or the local aquifer. In acidic water, iron causes a metallic taste as well as reddish stains on clothing and plumbing, while other metals such as lead are toxic. Alkaline or “hard” water contains excess calcium and other minerals that cause the familiar scaly deposits on cookware and a bitter taste in coffee.

2.10 pH and Groundwater

Groundwater flows through rocks and soil that can affect the water’s pH level. For example, contact with sandstone results in a nearly neutral pH between 6.5 and 7.5. Limestone, on the other hand, can result in an alkaline pH of 8.5. Soils contain minerals and other substances that affect groundwater pH. Decaying organic matter in soils causes pH levels in groundwater

to drop as low as 4.0. That pH level is well below the recommended 6.5 to 7.5 pH for drinking water. Human-induced pollution affects groundwater pH as well. For example, runoff from shale and coal mining contains iron sulfide, which can result in pH readings as low as 2.

2.11 pH and Streams and Lakes

The pH level in a lake or stream is crucial for the survival of fish and aquatic plants. Freshwater lakes and streams typically have pH levels between 6.0 and 8.0. Deeper lakes usually have a higher pH near the surface. Aquatic organisms are sensitive to pH changes. For example, the optimal pH level for fish ranges from 6.5 to 9.0. At levels outside this range, fish become susceptible to poisoning from toxic chemicals. Changes in pH can also cause an overload of available plant nutrients, resulting in excessive plant growth and depleted oxygen levels for fish. This condition, known eutrophication, threatens the survival of plant and animal life in the water.

2.12 Water Testing With a pH Meter

Scientists use a pH meter to measure pH levels in the water. The water testing takes place on-site using a relatively small, portable meter or in a lab using a larger benchtop meter. The benchtop meter has a cup that holds the water sample and a glass probe with two specialized electrodes. The pH electrode measures the acidity of the water sample, while the reference electrode is immersed in a liquid with a fixed acidity. After the reading from the pH electrode is compared with the reading from the reference electrode, the meter converts the voltage to a pH level. A pH meter provides a much more accurate reading than a test strip kit and prevents the mess associated with having to use droppers. Calibration before testing water samples ensures that the pH meter will provide accurate readings and test results.

2.13 Importance of pH in Water

Water quality is crucial for all living things, as well as for agriculture and recreation. Optimal pH levels are an essential factor in maintaining water quality and healthy ecosystems. Accurate pH testing helps keep drinking water and groundwater safe and helps protect aquatic plants and animals.

2.14 pH of Different Body Fluids

Although the pH of blood ranges from 7.35-7.45, the pH of other body fluids is different. pH indicates the level of H+ ions, where low pH indicates too many H+ ions and high pH indicates too many OH- ions. If the pH levels drop below 6.9, it can lead to coma. However,

different body fluids have different pH values. The pH of saliva is ranges from 6.5 to 7.5. After swallowing, the food reaches the stomach where upper and lower parts of stomach have different pH values. The upper part has a pH of 4−6.5, while the lower part is highly acidic with a pH of 1.5−4.0. It then enters the intestine which is slightly alkaline, with a pH of 7−8.5. Maintaining the pH values of different regions is critical for their function.

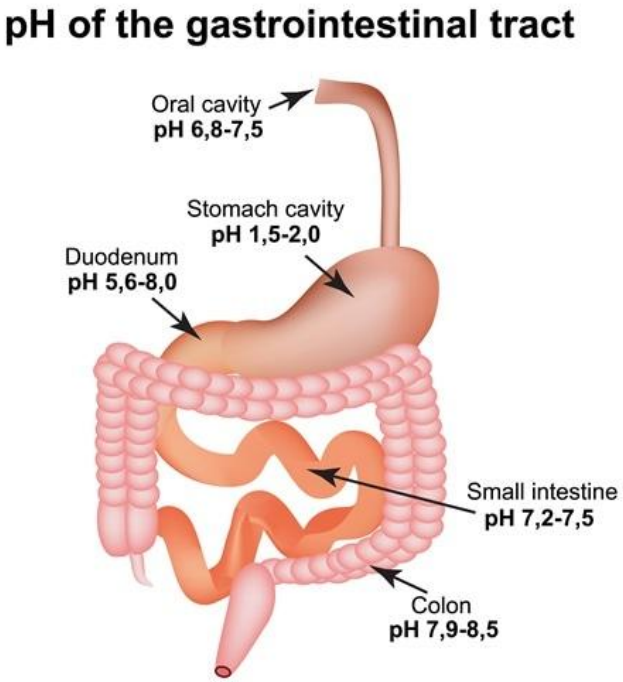


Fig. pH of the gastrointestinal tract. Esophagus, stomach, duodenum, small intestine, colon.

2.15 Impact of Altering the pH Balance

Different organs function at their optimal level of pH. For example, the enzyme pepsin requires low pH to act and break down food, while the enzymes in intestine require high pH or alkaline environment to function. Similarly, any increase or decrease in the blood pH can lead to several disorders.

2.16 Maintaining the Body pH

pH is maintained in the body using primarily three mechanisms: buffer systems, respiratory control, and renal control.

2.17 Buffer Systems

Proteins form a part of the buffer system to regulate the pH levels. These proteins can act as H+ acceptors or donors because of the presence of basic or acidic groups. Similarly phosphate buffers also help in moderating the levels of pH. Buffers may help in regulating pH during

minor physiological changes, such as during breath holding (which increases the CO2 in the blood), exercise (which increases lactic acid in the blood), or when gastric acid is secreted.

2.18 Respiratory Control

The pH of blood during normal conditions is 7.4. However, CO2 dissociates into carbonic acid in the tissues. Thus, presence of more CO2 makes the blood more acidic. That is the reason when we hold the breath for long durations, the CO2 levels increase in the blood lowering our pH leading to fainting. On the other hand during alkalosis or increased pH, the breathing may get slow in order to increase the CO2 levels and reduce the alkalinity. However, low breathing rate could also lead to low oxygen levels which could be detrimental. Thus, respiration provides an important control to regulate the pH levels.

2.19 Renal Control

The renal system regulates the pH of extracellular fluid. The changes in pH induced by the respiratory system are in minutes, while the changes induced by the renal system are in the order of days. If the acidity of the fluids is high, kidney secretes H+ ions, while if the carbonate ion levels are high it retains H+ ions and secretes HCO3 ions. Although this process is slow but it can prove an effective mode to regulate pH. One limitation of renal regulation is that the pH of urine cannot be below 4.4. Thus, strong acids can be removed by reacting with basic salts of phosphoric acid or by addition of base (NH3) to urine.

2.20 Abnormalities in Acid-Base Balance

The abnormalities in acid-base balance are of two types: acidosis and alkalosis. In acidosis, the blood pH is low or there is too much acid in the blood, while in alkalosis, the blood pH is high or there is too much base in the blood. Acidosis and alkalosis may be caused either due to imbalance of acid-base secretion by the kidneys or altered levels of CO_2 in the blood due to breathing disorders.

2.21 Body Fluid Compartments

There are three major fluid compartments; intravascular, interstitial, and intracellular. Fluid movement from the intravascular to interstitial and intracellular compartments occurs in the capillaries. A capillary “membrane,” which consists of the endothelial glycocalyx, endothelial cells, and the subendothelial cell matrix, separates the capillary intravascular space from the interstitial fluid compartment. This capillary “membrane” is freely permeable to water and small-molecular-weight particles such as electrolytes, glucose, acetate, lactate, gluconate, and

bicarbonate. Gases such as oxygen and carbon dioxide diffuse freely through this membrane, following their concentration gradient, to enter or exit the intravascular compartment.

The interstitial compartment is the space between the capillaries and the cells. Fluids support the matrix and cells within the interstitial space. The intracellular compartment is separated from the interstitial space by a cell membrane. This membrane is freely permeable to water but not to small- or large-molecular-weight particles. Any particle movement between the interstitium and the cell must occur through some transport mechanism (eg, channel, ion pump, carrier mechanism).

Fluids are in a constant state of flux across the capillary endothelial membrane, through the interstitium, and into and out of the cell. The amount of fluid that moves across the capillary "membrane" depends on a number of factors, including capillary colloid oncotic pressure (COP), hydrostatic pressure, and permeability, which is dictated by factors such as the endothelial glycocalyx layer (EGL) and pore sizes between the cells. The natural particles in blood that create COP are proteins: primarily albumin but also globulins, fibrinogen, and others. The hydrostatic pressure within the capillary is the pressure forcing outward on the capillary membrane generated by the blood pressure and cardiac output.

Fluid moves into the interstitial space when intravascular hydrostatic pressure is increased over COP, when membrane pore size increases, the EGL is disrupted, or when intravascular COP becomes lower than interstitial COP. The EGL is now known to play an important role in controlling fluid and other molecule (eg, albumin) transport across the capillary layer, and the oncotic pressure of the glycocalyx plays a larger role than the oncotic pressure of the interstitium; various disease processes and therapy (such as IV fluid administration) can significantly disrupt the EGL, resulting in altered transcapillary movement.

2.22 Summary

Under this unit we summarize the various electrolytes with applications, acid-base balance and pH, body fluids and different body fluid compartments. An electrolyte is a medium containing ions that is electrically conducting through the movement of ions, but not conducting electrons. This includes most soluble salts, acids, and bases dissolved in a polar solvent, such as water. Upon dissolving, the substance separates into cations and anions, which disperse uniformly throughout the solvent. Solid-state electrolytes also exist. In

medicine and sometimes in chemistry, the term electrolyte refers to the substance that is dissolved.

Electrically, such a solution is neutral. If an electric potential is applied to such a solution, the cations of the solution are drawn to the electrode that has an abundance of electrons, while the anions are drawn to the electrode that has a deficit of electrons. The movement of anions and cations in opposite directions within the solution amounts to a current. Some gases, such as hydrogen chloride (HCl), under conditions of high temperature or low pressure can also function as electrolytes. Electrolyte solutions can also result from the dissolution of some biological (e.g., DNA, polypeptides) or synthetic polymers (e.g., polystyrene sulfonate), termed "polyelectrolytes", which contain charged functional groups. A substance that dissociates into ions in solution or in the melt acquires the capacity to conduct electricity. Sodium, potassium, chloride, calcium, magnesium, and phosphate in a liquid phase are examples of electrolytes.

2.23 Terminal questions

Q. 1 What do you mean by electrolytes? Describe it with examples.

Answer:----------

Q.2 What do you mean by acid-base balance?

Answer:----------

Q.3 Describe the different role and regulation of electrolytes.

Answer:----------

Q.4 Explain about pH and its importance?

Answer:----------

Q.5 Describe body fluids and fluid compartments?

Answer:----------

Q.6 Write a short note on following.

(a) Buffer system

(b) pH of different body fluids

Answer:----------

Q.6 Write a short note on following.

(a) pH of groundwater

(b) pH of drinking water

Answer:----------

Q.7 Explain the importance of pH in water.

Answer:----------

Further readings

1. Biochemistry- Lehninger A.L.
2. Biochemistry –J.H.Weil.
3. Biochemistry fourth edition-David Hames and Nigel Hooper.
4. Textbook of Biochemistry for Undergraduates - Rafi, M.D.
5. Biochemistry and molecular biology- Wilson Walker.

Unit-3

3.1 Introduction

This unit discusses clinical enzymology. An enzyme in serum, to be valuable as a clinical diagnostic aid, must be readily assayable; its assay must be economically feasible and reasonably reflect pathological change in a specific organ or group of organs. An example of assay ability is the case of the three liver-specific enzymes, anginas (ARG), sorbitol dehydrogenase (SDH), and alanine aminotransferase (GPT). ARG and SDH are liver-specific enzymes in many animals and are excellent markers of hepatocellular damage. GPT is also liver specific in many species but is used in favor of the former two enzymes because the assay for GPT is simpler than that for ARG and SDH, and a large body of clinical background data for GPT is available.

Cells need not be necrotic to release their enzymes into plasma. Anoxia causes the cell membrane to lose its integrity and soluble enzyme from the cytosol leaks into plasma. This loss of integrity is often first observed microscopically as a swelling of the cell. An example of enzyme leak is in hepatic congestion, where the GPT activity in serum increases in the absence of frank hepatocytic necrosis. When a serum enzyme lacks sufficient specificity for an organ, a second enzyme may be combined with the first to increase its diagnostic value. Serum alkaline phosphatase activity increases in bone and liver disorders, and, to assist in identifying the source of the increase, GPT or γ-glutamyltransferase may be assayed as part of a hepatic profile.

Objectives

This is the third unit on clinical biochemistry. Under third unit we have following objectives. These are as under:

- To know about clinical enzymology
- To know about enzymes and hormones.
- To know about plasma enzymes and isoenzymes.
- To discuss liver damage and bone disorder.

3.2 Enzymes

Enzymology is the study of enzymes, their kinetics, structure, and function, as well as their relation to each other. The enzyme changes shape by induced fit upon substrate binding to

form an enzyme-substrate complex. Hexokinase has a large induced fit motion that closes over the substrates adenosine triphosphate and xylose. Binding sites in blue, substrates in black and Mg^{2+} cofactor in yellow.

The human body is composed of different types of cells, tissues and other complex organs. For efficient functioning, our body releases some chemicals to accelerate biological processes such as respiration, digestion, excretion and few other metabolic activities to sustain a healthy life. Hence, enzymes are pivotal in all living entities which govern all the biological processes. Let us understand what are enzymes, types, their structure, mechanism and various factors that affect its activity.

3.3 What Are Enzymes?

"Enzymes can be defined as biological polymers that catalyze biochemical reactions."

Majority of enzymes are proteins with catalytic capabilities crucial to perform different processes. Metabolic processes and other chemical reactions in the cell are carried out by a set of enzymes that are necessary to sustain life. The initial stage of metabolic process depends upon the enzymes, which react with a molecule and is called the substrate. Enzymes convert the substrates into other distinct molecules and are called the products.

The regulation of enzymes has been a key element in clinical diagnosis because of their role in maintaining life processes. The macromolecular components of all enzymes consist of protein, except in the class of RNA catalysts called ribozymes. The word ribozyme is derived from the ribonucleic acid enzyme. Many ribozymes are molecules of ribonucleic acid, which catalyze reactions in one of their own bonds or among other RNAs. Enzymes are found in all tissues and fluids of the body. Catalysis of all reactions taking place in metabolic pathways are carried out by intracellular enzymes. The enzymes in plasma membrane govern the catalysis in the cells as a response to cellular signals and enzymes in the circulatory system regulate clotting of blood. Most of the critical life processes are established on the functions of enzymes.

3.4 Enzyme Structure

Enzymes are a linear chain of amino acids, which give rise to a three-dimensional structure. The sequence of amino acids specifies the structure, which in turn identifies the catalytic activity of the enzyme. Upon heating, enzyme's structure denatures, resulting in a loss of enzyme activity, that typically is associated with temperature.

Compared to its substrates, enzymes are typically large with varying sizes, ranging from 62 amino acid residues to an average of 2500 residues found in fatty acid synthase. Only a small section of the structure is involved in catalysis and is situated next to the binding sites. The catalytic site and binding site together constitute the enzyme's active site. A small number of ribozymes exist which serve as an RNA-based biological catalyst. It reacts in complex with proteins.

3.5 Enzymes Classification

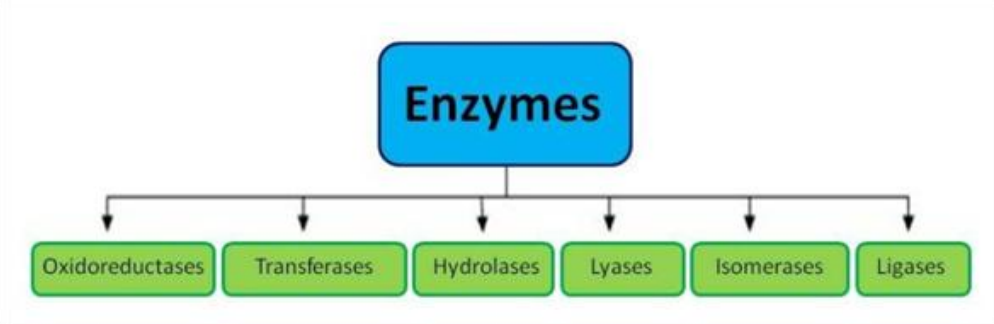


Fig. Enzymes

Earlier, enzymes were assigned names based on the one who discovered it. With further researches, classification became more comprehensive. According to the International Union of Biochemists (I U B), enzymes are divided into six functional classes and are classified based on the type of reaction in which they are used to catalyze. The six kinds of enzymes are hydrolases, oxidoreductases, lyases, transferases, ligases and isomerases. Listed below is the classification of enzymes discussed in detail:

Types	**Biochemical Property**
Oxidoreductases	The enzyme Oxidoreductase catalyzes the oxidation reaction where the electrons tend to travel from one form of a molecule to the other.
Transferases	The Transferases enzymes help in the transportation of the functional group among acceptors and donor molecules.
Hydrolases	Hydrolases are hydrolytic enzymes, which catalyze the hydrolysis reaction by adding water to cleave the bond and hydrolyze it.

Lyases	Adds water, carbon dioxide or ammonia across double bonds or eliminate these to create double bonds.
Isomerases	The Isomerases enzymes catalyze the structural shifts present in a molecule, thus causing the change in the shape of the molecule.
Ligases	The Ligases enzymes are known to charge the catalysis of a ligation process.

Table. Enzymes and biochemical property

3.6 Oxidoreductases

These catalyze oxidation and reduction reactions, e.g. pyruvate dehydrogenase, catalysing the oxidation of pyruvate to acetyl coenzyme A.

3.7 Transferases

These catalyze transferring of the chemical group from one to another compound. An example is a transaminase, which transfers an amino group from one molecule to another.

3.8 Hydrolases

They catalyze the hydrolysis of a bond. For example, the enzyme pepsin hydrolyzes peptide bonds in proteins.

3.9 Lyases

These catalyze the breakage of bonds without catalysis, e.g. aldolase (an enzyme in glycolysis) catalyzes the splitting of fructose-1, 6-bisphosphate to glyceraldehyde-3-phosphate and dihydroxyacetone phosphate.

3.10 Isomerases

They catalyze the formation of an isomer of a compound. Example: phosphoglucomutase catalyzes the conversion of glucose-1-phosphate to glucose-6-phosphate (phosphate group is transferred from one to another position in the same compound) in glycogenolysis (glycogen is converted to glucose for energy to be released quickly).

3.11 Ligases

Ligases catalyze the association of two molecules. For example, DNA ligase catalyzes the joining of two fragments of DNA by forming a phosphodiester bond.

3.12 Cofactors

Cofactors are non-proteinous substances that associate with enzymes. A cofactor is essential for the functioning of an enzyme. An enzyme without a cofactor is called an apoenzyme. An enzyme and its cofactor together constitute the holoenzyme.

There are three kinds of cofactors present in enzymes:

- **Prosthetic groups**: These are cofactors tightly bound to an enzyme at all times. A fad is a prosthetic group present in many enzymes.
- **Coenzyme**: A coenzyme binds to an enzyme only during catalysis. At all other times, it is detached from the enzyme. NAD^+ is a common coenzyme.
- **Metal ions**: For the catalysis of certain enzymes, a metal ion is required at the active site to form coordinate bonds. Zn^{2+} is a metal ion cofactor used by a number of enzymes.

3.13 Examples of Enzymes

Following are some of the examples of enzymes:

3.13.1 Beverages

Alcoholic beverages generated by fermentation vary a lot based on many factors. Based on the type of the plant's product, which is to be used and the type of the enzyme applied, the fermented product varies. For example, grapes, honey, hops, wheat, cassava roots, and potatoes depending upon the materials available. Beer, wines and other drinks are produced from plant fermentation.

3.13.2 Food Products

Bread can be considered as the finest example of fermentation in our everyday life.

A small proportion of yeast and sugar is mixed with the batter for making bread. Then one can observe that the bread gets puffed up as a result of fermentation of the sugar by the enzyme action in yeast, which leads to the formation of carbon dioxide gas. This process gives the texture to the bread, which would be missing in the absence of the fermentation process.

3.13.3 Drug Action

Enzyme action can be inhibited or promoted by the use of drugs which tend to work around the active sites of enzymes.

3.13.4 Mechanism of Enzyme Reaction

Any two molecules have to collide for the reaction to occur along with the right orientation and a sufficient amount of energy. The energy between these molecules needs to overcome the barrier in the reaction. This energy is called activation energy. Enzymes are said to possess an active site. The active site is a part of the molecule that has a definite shape and the functional group for the binding of reactant molecules. The molecule that binds to the enzyme is referred to as the substrate group. The substrate and the enzyme form an intermediate reaction with low activation energy without any catalysts.

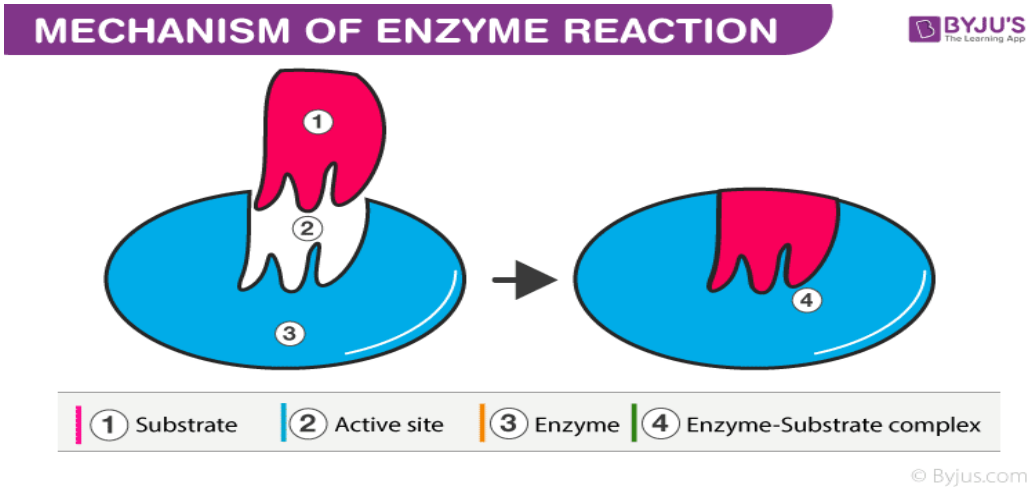


Fig. Mechanism of enzyme reaction

The basic mechanism of enzyme action is to catalyze the chemical reactions, which begins with the binding of the substrate with the active site of the enzyme. This active site is a specific area that combines with the substrate.

3.13.5 Enzyme-Substrate Interactions

Enzymes are the biocatalysts with high molecular weight proteinous compound. It enhances the reactions which occur in the body during various life processes. It helps the substrate by providing the surface for the reaction to occur. The enzyme comprises hollow spaces occupying groups such as -SH, -COOH, and others on the outer surface. The substrate which has an opposite charge of the enzyme fits into these spaces, just like a key fits into a lock. This substrate binding site is called the active site of an enzyme (E).

The favourable model of enzyme-substrate interaction is called the induced-fit model. This model states that the interaction between substrate and enzyme is weak, and these weak interactions induce conformational changes rapidly and strengthen binding and bring catalytic sites close enough to substrate bonds. There are four possible major mechanisms of catalysis:

3.13.6 Catalysis by Bond Strain

The induced structural rearrangements in this type of catalysis produce strained substrate bonds that attain transition state more easily. The new conformation forces substrate atoms and catalytic groups like aspartate into conformations that strain substrate bonds.

3.13.7 Covalent Catalysis

The substrate is oriented to active place on the enzymes in such a manner that a covalent intermediate develops between the enzyme and the substrate, in catalysis that occurs by covalent mechanisms. The best example of this involves proteolysis by serine proteases that have both digestive enzymes and various enzymes of the blood clotting cascade. These proteases possess an active site serine whose R group hydroxyl generates a covalent bond with a carbonyl carbon of a peptide bond and results in the hydrolysis of the peptide bond.

3.13.8 Catalysis Involving Acids and Bases

Other mechanisms add to the completion of catalytic events which are launched by strain mechanisms such as the usage of glutamate as a general acid catalyst.

3.13.9 Catalysis by Orientation and Proximity

Enzyme-substrate interactions induce reactive groups into proximity with one another. Also, groups like aspartate are chemically reactive, and their proximity towards the substrate favours their involvement in catalysis.

3.13.10 Action and Nature of Enzymes

Once substrate (S) binds to this active site, they form a complex (intermediate-ES) which then produces the product (P) and the enzyme (E). The substrate which gets attached to the enzyme has a specific structure and that can only fit in a particular enzyme. Hence, by providing a surface for the substrate, an enzyme slows down the activation energy of the reaction. The intermediate state where the substrate binds to the enzyme is called the transition state. By breaking and making the bonds, the substrate binds to the enzyme (remains unchanged), which converts into the product and later splits into product and enzyme. The free enzymes

then bind to other substrates and the catalytic cycle continues until the reaction completes. The enzyme action basically happens in two steps:

Step1: Combining of enzyme and the reactant/substrate.

$$E+S \rightarrow [ES]$$

Step 2: Disintegration of the complex molecule to give the product.

$$[ES] \rightarrow E+P$$

Thus, the whole catalyst action of enzymes is summarized as:

$$E + S \rightarrow [ES] \rightarrow [EP] \rightarrow E + P$$

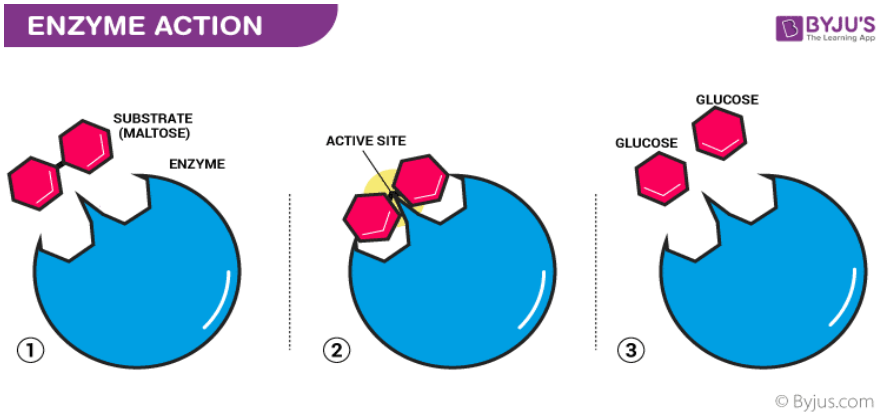


Fig. Enzyme action

3.13.11 Biological Catalysts

Catalysts are the substances which play a significant role in the chemical reaction. Catalysis is the phenomenon by which the rate of a chemical reaction is altered/ enhanced without changing themselves. During a chemical reaction, a catalyst remains unchanged, both in terms of quantity and chemical properties. An enzyme is one such catalyst which is commonly known as the biological catalyst. Enzymes present in the living organisms enhance the rate of reactions which take place within the body.

Biological catalysts, enzymes, are extremely specific that catalyze a single chemical reaction or some closely associated reactions. An enzyme's exact structure and its active site decide an enzyme's specificity. Substrate molecules attach themselves at the active site of an enzyme. Initially, substrates associate themselves by noncovalent interactions to the enzymes which include ionic, hydrogen bonds and hydrophobic interactions. Enzymes reduce the reactions and activation energy to progress towards equilibrium quicker than the reactions that are not catalyzed. Both eukaryotic and prokaryotic cells usually make use of allosteric regulation to respond to fluctuations in the state inside the cells.

The nature of enzyme action and factors affecting the enzyme activity are discussed below.

3.13.12 Factors Affecting Enzyme Activity

The conditions of the reaction have a great impact on the activity of the enzymes. Enzymes are particular about the optimum conditions provided for the reactions such as temperature, pH, alteration in substrate concentration, etc.

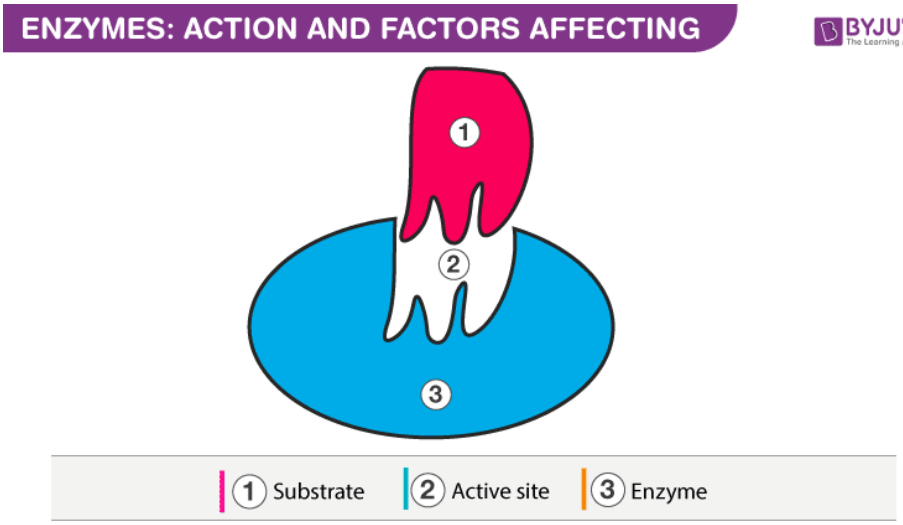


Fig. Enzymes and factors

Typically, enzyme activities are accelerated with increasing temperatures. As enzymes are functional in cells, the feasible conditions for nearly all enzymes are temperatures that are moderate. At higher temperatures, given a specific point, there is a drastic decrease in the activity with the denaturation of enzymes. In diluted solutions, purified enzymes denature quickly compared to enzymes in crude extracts. Denaturation of enzymes can also take place when enzymes are incubated for long durations. More appropriate is to utilize a shorter time duration when it comes to incubation time to gauge the starting velocities of such enzyme reactions.

The International Union of Biochemistry suggests the standard assay temperature to be 30 °C. Almost all enzymes are extremely sensitive to pH change. Just some enzymes feasibly operate with pH above 9 and below 5. Most enzymes have their pH – optimum near to neutrality. Any alteration of pH causes the ionic state of amino acid residues to change in the whole protein and in the active site. The modifications in the ionic state can modify catalysis and substrate binding. The preference of substrate concentration is critical as at lower concentrations, the rate is driven by concentration, however, at high concentrations, the rate does not depend on any increase in the concentration of the substrate.

3.13.14Active site

Enzymatic catalysis depends upon the activity of amino acid side chains assembled in the active centre. Enzymes bind the substrate into a region of the active site in an intermediate conformation. Often, the active site is a cleft or a pocket produced by the amino acids which take part in catalysis and substrate binding. Amino acids forming an enzyme's active site is not contiguous to the other along the sequence of primary amino acid.

The active site amino acids are assembled to the cluster in the right conformation by the 3-dimensional folding of the primary amino acid sequence. The most frequent active site amino acid residues out of the 20 amino acids forming the protein are polar amino acids, aspartate, cysteine, glutamate, histidine, Serine, and lysine. Typically, only 2-3 essential amino acid residues are involved directly in the bond causing the formation of the product. Glutamate, Aspartate, and histidine are the amino acid residues which also serve as a proton acceptor or donor.

3.14 Temperature and pH

Enzymes require an optimum temperature and pH for their action. The temperature or pH at which a compound shows its maximum activity is called optimum temperature or optimum pH, respectively. As mentioned earlier, enzymes are protein compounds. A temperature or pH more than optimum may alter the molecular structure of the enzymes. Generally, an optimum pH for enzymes is considered to be ranging between 5 and 7.

3.15 Hormones

Hormones are chemical messengers that are secreted directly into the blood, which carries them to organs and tissues of the body to exert their functions. There are many types of hormones that act on different aspects of bodily functions and processes. Some of these include:

- Development and growth
- Metabolism of food items
- Sexual function and reproductive growth and health
- Cognitive function and mood
- Maintenance of body temperature and thirst

A **hormone** is any member of a class of signaling molecules in multicellular organisms, that are transported by intricate biological processes to distant organs to regulate physiology and behavior. Hormones are required for the correct development

of animals, plants and fungi. The lax definition of a hormone (as a signalling molecule that acts distant from its site of production) means that many different classes of molecule can be defined as hormones. Among the substances that can be considered hormones, are eicosanoids (e.g. prostaglandins and thromboxanes), steroids (e.g. oestrogen and brassinost eroid), aminoacid derivatives(e.g. epinephrine and auxin), protein / peptides (e.g. insulin and CLE peptides) and gases (e.g ethylene and nitric oxide).

Hormones are used to communicate between organs and tissues. In vertebrates, hormones are responsible for the regulation of many physiological processes and behavioral activities such as digestion, metabolism, respiration, sensoryperception, sleep, excretion, lactation, stress ind uction, growth and development, movement, reproduction, and mood manipulation. In plants, hormones modulate almost all aspects of development, from germination to senescence.

Hormones affect distant cells by binding to specific receptor proteins in the target cell, resulting in a change in cell function. When a hormone binds to the receptor, it results in the activation of a signal transduction pathway that typically activates gene transcription, resulting in increased expression of target proteins.

Hormones can also act in rapid, non-genomic pathways that can be synergistic with genomic effects. Water-soluble hormones (such as peptides and amines) generally act on the surface of target cells via second messengers. Lipid soluble hormones, (such as steroids) generally pass through the plasma membranes of target cells (both cytoplasmic and nuclear) to act within their nuclei. A notable exception to this are brassinosteroids (Brassinosteroids are a class of polyhydroxysteroids that have been recognized as a sixth class of plant hormones and may have utility as an anticancer drug for endocrine-responsive cancers to induce apoptosis and inhibit growth in plants) which despite being lipid soluble, still bind to their receptor at the cell surface.

In vertebrates, endocrine glands are specialized organs that secrete hormones into the endocrine signaling system. Hormone secretion occurs in response to specific biochemical signals and is often subject to negative feedback regulation. For instance, high blood sugar (serum glucose concentration) promotes insulin synthesis. Insulin then acts to reduce glucose levels and maintain homeostasis, leading to reduced insulin levels. Upon secretion, water soluble hormones are readily transported through the circulatory system. Lipid-soluble hormones must bond to carrier plasma glycoproteins (e.g., thyroxine-binding globulin (TBG)) to form ligand-protein complexes.

Completely active hormones can be released into the bloodstream (as seen in insulin and growth hormones), but some travel as prohormones that must be activated in specific cells through a series of activation steps that are commonly highly regulated. The endocrine system secretes hormones directly into the bloodstream, typically via fenestrated capillaries, whereas the exocrine system secretes its hormones indirectly using ducts. Hormones with paracrine function diffuse through the interstitial spaces to nearby target tissue.

Plants lack specialized organs for the secretion of hormones, although there is spatial distribution of hormone production. For example, the hormone auxin is produced mainly at the tips of young leaves and in the shoot apical meristem. The lack of specialised glands means that the main site of hormone production can change throughout the life of a plant, and the site of production is dependent on the plant's age and environment.

3.15 Where are they secreted from?

Hormones are secreted from the endocrine glands in the body. The glands are ductless, so hormones are secreted directly into the blood stream rather than by way of ducts. Some of the major endocrine glands in the body include:

- Pituitary gland
- Pineal gland
- Thymus
- Thyroid
- Adrenal glands
- Pancreas
- Testes
- Ovaries

These organs secrete hormone in microscopic amounts and it takes only very small amounts to bring about major changes in the body. Even a very slight excess of hormone secretion can lead to disease states, as can the slightest deficiency in a hormone.

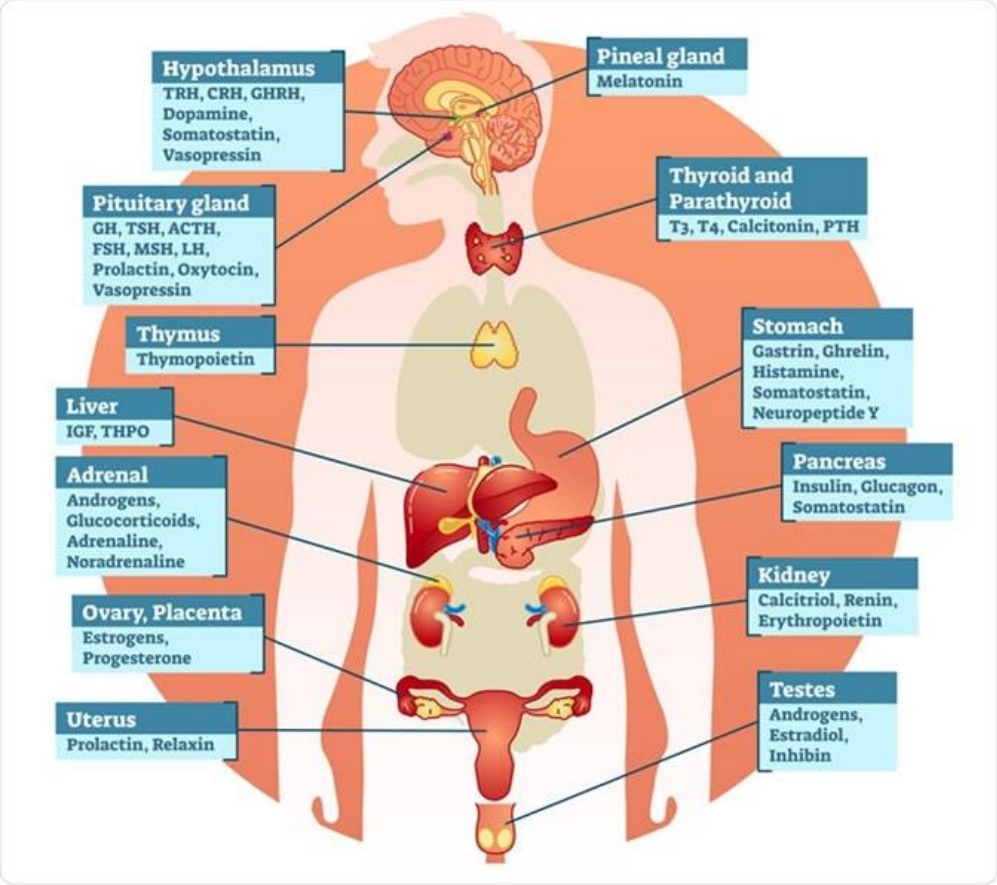


Fig. Human body hormones

3.16 Hormones and diseases

Hormone disorders are diagnosed in the laboratory as well as by clinical appearance and features. Laboratory tests can be used to test bodily fluids such as the blood, urine or saliva for hormone abnormalities. In the case of hormone deficiency, a synthetic hormone replacement therapy may be used and in cases of excess hormone production, medications may be used to curb the effects of the hormone. For example, a person with an underactive thyroid gland or hypothyroidism may be treated with synthetic thyroxine which can be taken in the form of a pill, while a person with an overactive thyroid may be administered a drug such as propranolol to counteract the effects of the excess thyroid hormone.

3.17 Plasma Enzymes

Blood plasma contains many enzymes, which are classified into functional and non-functional plasma enzymes. Differences between functional and non-functional plasma enzymes:-

Sources of non-functional plasma enzymes:

1. Increase in the rate of enzyme synthesis) e.g. bilirubin increases the rate of synthesis of alkaline phosphatase in obstructive liver diseases.

2. Obstruction of normal pathway e.g. obstruction of bile ducts increases alkaline phosphatase.

3. Increased permeability of cell membrane as in tissue hypoxia.

4. Cell damage with the release of its content of enzymes into the blood e.g. myocardial infarction and viral hepatitis.

3.18 Medical importance of non-functional plasma enzymes:

Measurement of non-functional plasma enzymes is important for:

1. Diagnosis of diseases as diseases of different organs cause elevation of different plasma enzymes.

2. Prognosis of the disease; we can follow up the effect of treatment by measuring plasma enzymes before and after treatment.

Examples of medically important non-functional plasma enzymes:

1. Amylase and lipase enzymes increase in diseases of the pancreas as acute pancreatitis. 2. Creatine kinase (CK) enzyme increases in heart, brain and skeletal muscle diseases.
2. Lactate dehydrogenase (LDH) enzyme increases in heart, liver and blood diseases.
3. Alanine transaminase (ALT) enzyme, it is also called serum glutamic pyruvic transaminase (SGPT). It increases in liver and heart diseases.
4. Aspartate transaminase (AST) enzyme, it is also called serum glutamic oxalacetic transaminase (SGOT). It increases in liver and heart diseases.
5. Acid phosphatase enzyme increases in cancer prostate.
6. Alkaline phosphatase enzyme increases in obstructive liver diseases, bone diseases and hyperparathyroidism.

3.19 Classification of Enzymes

Enzymes are classified according to the type of reaction they catalyze into six groups:

1. Oxido-reductases

These are enzymes that catalyze oxidation-reduction reactions. Oxido-reductases are further classified into five subgroups:

A- Oxidases

These are enzymes that catalyze direct transfer of hydrogen to oxygen and form water e.g. cytochrome oxidase and ascorbic acid oxidase.

B- Aerobic Dehydrogenases

These are enzymes that catalyze direct transfer of hydrogen to oxygen and form hydrogen peroxide (H_2O_2) e.g. L-amino oxidase and D-amino acid oxidase.

C- Anaerobic dehydrogenases

These are enzymes cannot transfer hydrogen directly to oxygen but hydrogen is indirectly transferred to oxygen through many hydrogen carriers e.g. glucose-6-phosphate dehydrogenase and succinate dehydrogenase.

D- **Hydroperoxidase**

These enzymes use hydrogen peroxide (H_2O_2) as substrate changing it into water (H_2O) e.g. peroxidases and catalases

E- Oxygenases

These enzymes catalyze direct incorporation of oxygen into substrate. e.g. i- Dioxygenases (True oxygenases): These enzymes catalyze incorporation (introduction) of two oxygen atoms into substrate e.g. tryptophan pyrrolase enzyme. ii- Monooxygenases (pseudo-oxygenases or hydroxy

3.20 Isoenzyme

Isozymes (also known as isoenzymes) are enzymes that differ in amino acid sequence but catalyze the same chemical reaction. These enzymes usually display different kinetic parameters (i.e. different KM values), or different regulatory properties. The existence of isozymes permits the fine-tuning of metabolism to meet the particular needs of a given tissue or developmental stage (for example lactate dehydrogenase (LDH)). In biochemistry, isozymes (or isoenzymes) are isoforms (closely related variants) of enzymes. In many cases, they are coded for by homologous genes that have diverged over time. Although, strictly speaking, allozymes represent different alleles of the same gene, and isozymes represent different genes whose products catalyse the same reaction, the two words are usually used interchangeably.

Isozymes were first described by hunter and Markert (1957) who defined them as different variants of the same enzyme having identical functions and present in the same individual. This definition encompasses

(1) Enzyme variants that are the product of different genes and thus represent different loci (described as isozymes)

(2) Enzymes that are the product of different alleles of the same gene (described as allozymes).

Isozymes are usually the result of gene duplication, but can also arise from polyploidisation or hybridization. Over evolutionary time, if the function of the new variant remains identical to the original, then it is likely that one or the other will be lost as mutations accumulate,

resulting in a pseudogene. However, if the mutations do not immediately prevent the enzyme from functioning, but instead modify either its function, or its pattern of gene Expression, then the two variants may both be favoured by natural selection and become specialised to different functions. For example, they may be expressed at different stages of development or in different tissues.

Allozymes may result from point mutations or from insertion-deletion (indel) events that affect the dna coding sequence of the gene. As with any other new mutation, there are three things that may happen to a new allozyme:
It is most likely that the new allele will be non-functional — in which case it will probably result in low fitness and be removed from the population by natural selection. Alternatively, if the amino acid residue that is changed is in a relatively unimportant part of the enzyme, for example a long way from the active site then the mutation may be selectively neutral and subject to genetic drift.

In rare cases the mutation may result in an enzyme that is more efficient, or one that can catalyse a slightly different chemical reaction, in which case the mutation may cause an increase in fitness, and be favoured by natural selection.

Example of isozyme:

An example of an isozyme is glucokinase, a variant of hexokinase which is not inhibited by glucose 6-phosphate. Its different regulatory features and lower affinity for glucose (compared to other hexokinases), allows it to serve different functions in cells of specific organs, such as control of insulin release by the beta cells of the pancreas, or initiation of glycogen synthesis by liver cells. Both of these processes must only occur when glucose is abundant, or problems occur.

3.21 Liver Damage and Their Causes

- Symptoms
- Common problems
- Risk factors
- Prevention
- Diagnosis
- Treatment

Your liver is a vital organ that performs hundreds of tasks related to metabolism, energy storage, and waste filtering. It helps you digest food, convert it to energy, and store the energy until you need it. It also helps filter toxic substances out of your bloodstream. Liver disease is a general term that refers to any condition affecting your liver. These conditions may develop for different reasons, but they can all damage your liver and affect its function.

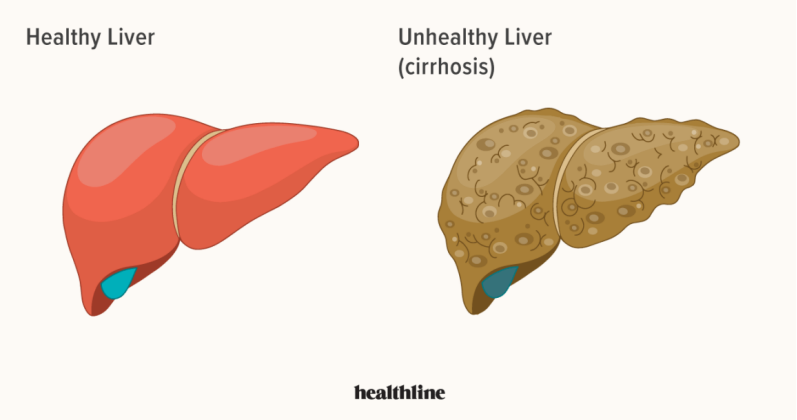


Fig. Stages in liver

What are the general symptoms?

Liver disease symptoms vary, depending on the underlying cause. It's also possible for someone to have liver disease and not have any symptoms at all. However, Hepatitis NSW says that a few general symptoms can indicate some kind of severe liver damage.

These include:

- Yellowish skin and eyes, known as jaundice
- Pale, bloody, or black stools
- Enlarged stomach due to ascites, which may make it uncomfortable to lie down or eat
- Encephalopathy, a brain issue resulting in marked changes in mood, sleep, and cognition

What are some common liver problems?

Many conditions can affect your liver. Here's a look at some of the main ones.

Hepatitis

Hepatitis is defined as an inflammation of the liver. When that inflammation is caused by a virus, it's referred to as viral hepatitis. Hepatitis can cause liver damage, making it difficult for your liver to function as it should. Most types of viral hepatitis are contagious, but you can reduce your risk by getting vaccinated for types A and B and by taking other preventive steps, including using a condom during sex and not sharing needles.

Five types of hepatitis include:

- **Hepatitis A.** Hepatitis A is typically spread through contact with contaminated food or water. Symptoms may clear up without treatment, but recovery can take a few weeks.
- **Hepatitis B.** This type of viral hepatitis can be acute (short-term) or chronic (long-term). It's spread through bodily fluids, such as blood and semen. While hepatitis B is treatable, there's no cure for it. Early treatment is key to avoiding complications, so it's best to get regular screenings if you're at risk.
- **Hepatitis C.** Hepatitis C can also be acute or chronic. It's often spread through contact with blood from someone with hepatitis C. While it often doesn't cause symptoms in its early stages, it can lead to permanent liver damage in its later stages.
- **Hepatitis D.** This is a serious form of hepatitis that only develops in people with hepatitis B — it can't be contracted on its own. It can also be either acute or chronic.
- **Hepatitis E.** Hepatitis E is usually caused by drinking contaminated water. Generally, it clears up on its own within a few weeks without any lasting complications.

3.22 Fatty liver disease

Fat buildup in the liver can lead to fatty liver disease. There are two types of fatty liver disease. These two types can manifest alone, or they can overlap:

- Alcoholic fatty liver disease, which is caused by heavy alcohol consumption
- Nonalcoholic fatty liver disease, which is caused by other factors experts are still trying to understand

Without management, both types of fatty liver disease can cause liver damage, leading to cirrhosis and liver failure. Diet and other lifestyle changes can often improve symptoms and lower your risk of complications.

Autoimmune conditions

Autoimmune conditions involve your immune system mistakenly attacking healthy cells in your body. Several autoimmune conditions involve your immune system attacking cells in your liver, including:

- **Autoimmune hepatitis.** This condition causes your immune system to attack your liver, resulting in inflammation. Without treatment, it can lead to cirrhosis and liver failure.

- **Primary biliary cirrhosis (PBC).** This results from damage to the bile ducts in your liver, causing a buildup of bile. PBC can eventually lead to cirrhosis and liver failure.
- **Primary sclerosing cholangitis.** This inflammatory condition causes gradual damage to your bile ducts. They eventually become blocked, causing bile to build up in your liver. This can lead to cirrhosis or liver failure.

3.23 Genetic conditions

Several genetic conditions, which you inherit from one of your parents, can also affect your liver:

- Hemochromatosis causes your body to store more iron than it needs. This iron remains in your organs, including your liver. This can lead to damage over a long period of time if not managed.
- Wilson's disease causes your liver to absorb copper instead of releasing it into your bile ducts. Eventually, your liver may become too damaged to store more copper, allowing it to travel through your bloodstream and damage other parts of your body, including your brain.
- Alpha-1 antitrypsin deficiency occurs when your liver can't make enough alpha-1 antitrypsin, a protein that helps prevent enzyme breakdowns throughout your body. This condition can cause lung disease as well as liver disease. There's no cure, but treatment can help.

3.24 Drug-induced liver disease

It's possible to damage your liver by overexposing it to certain drugs and supplements, as seen in a 2019 studyTrusted Source. Many times, this damage can be reversed once you stop taking the drug. But if it continues, the damage can become chronic.

3.24.1 Cancer

Liver cancer first develops in your liver. If cancer starts elsewhere in the body but spreads to the liver, it's called secondary liver cancer. The most common type of liver cancer is hepatocellular carcinoma. It tends to develop as several small spots of cancer in your liver, though it can also start as a single tumor. Complications of other liver diseases, especially those that aren't treated, may contribute to the development of liver cancer.

3.24.2 Cirrhosis

Cirrhosis refers to scarring that results from liver diseases and other causes of liver damage, such as alcohol use disorder. Cystic fibrosis and syphilis may also lead to liver damage and, eventually, cirrhosis — although these two causes are much less common. Your liver can regenerate in response to damage, but this process usually results in the development of scar tissue. The more scar tissue that develops, the harder it is for your liver to function properly. In its early stages, cirrhosis is often treatable by addressing the underlying cause. But without management, it can lead to other complications and become life threatening.

3.24.3 Liver failure

Chronic liver failure typically happens when a significant part of your liver is damaged and can't function properly. Generally, liver failure related to liver disease and cirrhosis happens slowly. You may not have any symptoms at first. But over time, you might start to notice:

- Jaundice
- Diarrhea
- Confusion
- Fatigue and weakness
- Nausea

It's a serious condition that requires ongoing management. Acute liver failure, on the other hand, happens suddenly, often in response to an overdose or poisoning.

3.25.3 Bone Disorders

Avascular Necrosis

Avascular necrosis develops when blood supply to a bone is cut off. This causes the bone to die and can cause pain and joint problems.

Understanding Bones

A typical bone in your body contains 3 types of tissue - a hard outer tissue, a sponge-like inner tissue, and smooth tissue at the ends.

Bone Cancers

Detailed information on bone cancers, including chondrosarcoma, ewings sarcoma, myeloma bone disease, multiple myeloma, and osteosarcoma.

Bone Disorders

Detailed information on bone disorders, including avascular necrosis, fibrous dysplasia, osteogenesis imperfecta, osteomyelitis, Paget's disease of the bone, and primary hyperparathyroidism.

3.26 Chondroblastoma

A chondroblastoma is a rare type of noncancerous bone tumor that begins in cartilage. This is the specialized, gristly connective tissue from which most bones develop. It plays an important role in the growth process. There are many different types of cartilage in the body. Chondroblastoma most often affects the ends of the long bones, near the growth plate, in the arms at the shoulder, and in the legs at the hip and knee. It is also called Codman's tumor.

3.27 **Chondrosarcoma**

Chondrosarcoma is the second most common type of primary bone cancer in adults. It mainly affects the cartilage cells of the femur, arm, pelvis, knee, and spine.

3.28 **Diagnosing Bone Disorders**

Detailed information on diagnostic procedures for bone disorders, including bone densitometry, radionuclide bone scan, and biopsy

3.29 **Enchondroma**

An enchondroma is a type of noncancerous bone tumor that begins in cartilage. An enchondroma most often affects the cartilage that lines the inside of the bones. It often affects the tiny long bones of the hands and feet. It may also affect other bones such as the femur (thighbone), humerus (upper arm bone), or tibia (one of the two lower leg bones).

3.30 **Fibrous Dysplasia**

Fibrous dysplasia is a chronic disorder in which an abnormal development of fibrous tissue causes bones to expand. Any bone can be affected. More than one bone can be affected at any one time, and, when multiple bones are affected, it is not unusual for them to all be on one side of the body. However, fibrous dysplasia does not spread from one bone to another.

3.31 **Giant Cell Tumor**

Giant cell tumor of bone is a rare, fast-growing noncancer tumor. It often grows near a joint at the end of a bone. Read on to learn about symptoms, diagnosis, and treatment.

3.32 **Primary Hyperparathyroidism**

Primary hyperparathyroidism is when one or more of the parathyroid glands produces too much parathyroid hormone. This can lead to bone tissue loss. Here's what you need to know.

3.33 Summary

Under this unit we summarize enzymology, isozymes and hormones. Enzymology is approaching an era where many problems can benefit from computational studies. While ample challenges remain in quantitatively predicting behavior for many enzyme systems, the insights that often come from computations are an important asset for the enzymology community. Here we provide a primer for enzymologists on the types of calculations that are most useful for mechanistic problems in enzymology. In particular, we emphasize the

integration of models that range from small active-site motifs to fully solvated enzyme systems for cross-validation and dissection of specific contributions from the enzyme environment. Enzymology is the branch of biochemistry aiming to understand how enzymes work through the relationship between structure and function and how they fold into their native state. To understand enzyme catalytic mechanisms in-depth, one must perform a series of steady-state and pre-steady-state kinetic experiments and determine the precise three-dimensional structure of the enzyme.

Enzymology is a multidisciplinary research field and integrates areas of biochemistry, microbiology, molecular biology, molecular genetics, and biophysics. The core of enzymology consists of the development of reliable activity assays, (over)expression and purification, steady-state kinetic characterization, and an initial basic structural characterization, which may include determination of subunit structure, molecular mass, prosthetic group content, cofactor requirement, and post-translational modifications. More detailed structural and mechanistic characterizations often require comparison of the wild-type with mutant enzymes and with specifically labeled enzymes. In addition to the academic interest, understanding enzyme catalytic mechanisms is essential for the successful application of enzymes in industrial processes.

3.34 Terminal questions

Q. 1 What do you mean by clinical enzymology? Describe it with examples.

Answer:----------

Q.2 What do you mean by enzymes? Explain it with examples.

Answer:----------

Q.3 What do you mean by hormones? Explain it with examples.

Answer:----------

Q.4 Explain biological catalysts with examples?

Answer:----------

Q.5 Describe liver damage and bone disorder.

Answer:----------

Q.6 Write a short note on following.

(a) Isoenzymes

(b) Active sites

Answer:----------

Q.6 Write a short note on following.

(a) Hormones and disease

(b) Fatty liver disease

Answer:----------

Q.7 Explain the following.

(a) Jaundice

(b) Diarrhea

(c) Nausea

Answer:----------

Further readings

1. Biochemistry- Lehninger A.L.
2. Biochemistry –J.H.Weil.
3. Biochemistry fourth edition-David Hames and Nigel Hooper.

4. Textbook of Biochemistry for Undergraduates - Rafi, M.D.
5. Biochemistry and molecular biology- Wilson Walker.

Unit-4

4.1 Introduction

Enzyme metabolism is a fundamental biological process that is vital for the survival of all species. Their specific function is to catalyze chemical reactions. Enzymes have found wide and diverse applications at which enzymes increase the rate of reactions which approach to equilibrium. Enzymes play critical role in the metabolic activities of all living organisms whether humans, animals, plants or microorganisms and are widely applied in microbialtechnology and their diagnosis processes. Abnormality of the enzyme metabolism system leads to a number of metabolic diseases. It is shown that many diseases associate with many components of the enzyme metabolism systems are now widely applied in clinical examinations as special markers for diseases. An interesting discovery suggesting that new roles of enzymes as a potential link that associates to prevent metabolic disorder. The aim of this review is to discuss the diverse diagnostic application of biotechnological enzymes for the purpose of teaching, education and research.

Rapid and accurate diagnosis of critical diseases and their appropriate treatment are the primary factors that promote optimal clinical outcomes and general public health. Enzymes have remarkable biocatalytic properties, and because of this, they are extensively used in diagnostics of various diseases. Many enzymes are preferred markers for the detection of various diseases such as jaundice, myocardial infarction, neurodegenerative disorders, cancer, and so forth. Enzymes provide insights into various diseases by diagnosis, prognosis, and assessment of response therapy.

Objectives

This is the fourth unit on clinical biochemistry. Under fourth unit we have following objectives. These are as under:

- To know about diagnostic enzymes
- To know about enzymes in health and diseases
- To know about various enzyme assay (SGPT, CPK, LDH)
- To discuss biochemical diagnosis of diseases

4.2 Applications of enzymes

Applications of enzymes in industrial chemical conversions, various forms of catalysis, and more recently in pharmaceutical manufacture are widely known. In medicine, enzymes are used in analytical tests, disease diagnosis, treatment of enzyme deficiencies and wound therapies. In clinical pathology, enzymes are invaluable tools in diagnosing tissue damage and cellular disorders from increased activity or concentration of specific tissue enzymes in body fluids. In this chapter the definition of "diagnostic enzymology" will be assumed to be the detection of biological malfunctions by changes in enzyme levels or activity in body fluids. Enzymes subject for detection are called diagnostic enzymes.

4.3 Enzymes in disease diagnosis

Enzymes are found in all body tissues and involved in most forms of metabolic activity. The regulation of enzymes allows metabolism to adapt rapidly to changing conditions. The activity of an enzyme in blood serum represents a balance between its rate of liberation into the extracellular space and its rate of uptake from the extracellular space. Factors such as age, sex and body weight affect this balance in individual subjects. Enzymes frequently appear in the serum following cellular injury or sometimes in smaller amounts from degraded cells or storage areas. Damage to or proliferation of cells from which enzyme originates leads to increased activity of enzyme in the plasma. Such increased or decreased activity levels tissue specific enzymes in the serum serve are references in the diagnosis of disease or cellular disorder.

Isoenzymes are proteins that possess similar catalytic activity but genetically determined differences in structure result in differing properties such as electrophoretic mobility, allowing identification. Isoenzyme distribution pattern also helps in diagnosis. For example, alkaline phosphatase (ALP) isoenzymes found in human serum originate from several sources with the greatest activity found in bone, liver, intestine, and the placenta. Tissue sources of elevated alkaline phosphatase in serum can be determined by identifying the isoenzyme. Diagnostic enzymology measures increase in plasma enzymes resulting from damage or increased turnover. Enzyme tests measure activity or concentration using fixed reaction conditions. An important point that has to be considered in selecting an appropriate enzyme test is that it should determine the extent of tissue damage and the type of tissues that have been damaged.

4.4 Diagnostically relevant enzymes

Along with the research advances in clinical biochemistry and diagnostic enzymology, large advances have been made in increased capability and refinement of the identification of disease-associated enzymes and factors. While fertile markets for clinical enzymes and test kits indeed prove that enzymes make powerful diagnostic tools, international efforts to standardize analyses are continually fostered. In pathology tests, enzymes in different kinds of tissues and biological fluids are the analytes while other diagnostically relevant applications include those in which enzymes are used as antigens, standard references, or labels.

Diagnostic enzymes are used to detect and quantify certain substances. As a marker in an enzyme immunoassay (EIA) system, clinical laboratories usually use many alternative techniques for diagnosis, including electrophoresis, chromatography, isoelectric focusing, etc.

4.5 Applications of diagnostic enzymes

4.5.1 For Blood Lipid Metabolism

Enzymes which are involved in the metabolism of lipids are being extensively studied. Creative Enzymes provide a wide range of blood lipids related enzyme products from different sources to help your research. We can provide you with cholesterol esterase, cholesterol oxidase, cholesterol dehydrogenase, glucose-6-phosphate dehydrogenase, hexokinase.

4.5.2 For Diabetes Diagnosis

Blood sugar test serves as the rapidest and most straightforward means to give a sign of diabetes. A number of enzymes involved in glucose metabolism were developed to react with glucose and subsequently to indicate glucose content in blood sample. A wide variety of diabetes related enzymes from different sources can be located in Creative Enzymes. For instance, glucose-6-phosphate dehydrogenase deficiency was found to be a risk factor for diabetes. Intensive studies are being carried out to explain this association. Glucose oxidase and glucose dehydrogenase are mostly commonly used for clinical blood sugar test. Hexokinase catalyzes the phosphorylation of glucose to yield glucose 6- phosphate, which initiates the utilization of glucose. Other relevant enzyme products are also available in our catalog.

4.5.3 Diagnostic Enzymes for Liver Function Test

Liver function test is vital for early diagnosis of liver diseases and management of hepatic dysfunction patients. A number of enzymatic reactions were developed to detect the presence of unusual substances which are associated with different types of liver disorder or liver damage. Creative Enzymes has been supporting the clinical and research use of liver function evaluation with our diagnostic enzyme products. To meet your specific requirements, we

include diverse liver function related enzymes from distinct natural sources or from biochemical synthesis. We can provide you with malate dehydrogenase, D-lactate dehydrogenase, alcohol dehydrogenase, and alkaline phosphatase in natural form or recombinant form.

4.5.4 Enzyme assays

Enzyme assays can be used for a variety of purposes, which include identifying the presence of an enzyme, investigation of specific enzyme kinetics or the activity of inhibition within a sample.

4.5.4.1 Measuring Enzyme Activity

When it comes to measuring enzyme activity, both qualitative and quantitative methodologies can be used. Qualitative assays are used to identify the presence (or absence) of a particular enzyme. On the other hand, a quantitative assay can be performed to determine the amount of the target enzyme that is present within a sample.

For qualitative assays, the use of coloured compounds is typically enough to confirm or deny the presence of the target enzyme. However, to ensure reproducibility and avoid operator error, using an appropriate instrument such as a colorimeter or photometer may be more appropriate than visual confirmation alone. Alternatively, quantitative assays are used in cases where instruments operating in the visual range are not suitable or determination of enzyme concentration of kinetic mechanisms are of interest.

4.5.4.2 Types of Enzyme Assay

There are two types of enzyme assay, which can be split into two; continuous and discontinuous assays.

4.5.4.3 Continuous Enzyme Assay

In continuous assays, the course of the reaction is continually followed until completion.

Sometimes referred to as ‘endpoint assays’, enzyme activity is measured via the quantity of substrate consumed, or the amount of product formed during the reaction over a fixed period of time. Both values are directly proportional to the concentration of enzymes present in the sample. Examples of continuous assays include spectrophotometry, calorimetry, chemiluminescence and fluorimetry. In these methods, the progress of reactions are measured by light or heat.

4.5.4.4 Discontinuous Enzyme Assay

In contrast to continuous enzyme assays, discontinuous assays are performed when samples are taken at set intervals. This form of enzyme assay directly or indirectly measures changes

in substrate or products over time, to understand how the reaction rate changes. Examples of instrumentation used during discontinuous enzyme assays include radiometric assays as well as chromatographic assays such as HPLC or TLC. Comparing the two methods, the continuous enzyme assay method is typically the easiest to perform and can give whilst discontinuous enzyme assays are used in cases where higher precision or complex sample matrices are present.

4.6 Factors That Affect Enzyme Assay

In order for an enzyme assay to remain accurate, controlling external factors so they do not influence the outcome of the assay is crucial.

4.7 pH

All enzymes have an optimum pH range where their rate of reaction is highest. Anything too far out of the optimum range will cause denaturation and a reduction in reaction rate.

4.8 Temperature

Generally speaking, as temperature increases, so does the reaction rate of an enzyme. However, once temperature exceeds a certain threshold, reaction rates can drastically drop. This is due to the effect that temperature has on the bonding of the reaction site.

4.9 Substrate Saturation

Increase in substrate concentration will increase the rate of reaction, but only up to a certain point. At the point of saturation, the reaction rate is at its upper limit and will not increase, regardless of how much substrate is added.

4.10 Salt Concentration

Enzymes typically work best in low salt concentration environments. Increase in salt concentration causes interference with bonds, which impact the structure of the bonding site and the resulting reaction rate.

4.11 Enzyme Assay Use Cases

Enzyme assays cover a wide range of real life use cases. A couple of examples of enzyme assays include the following:

4.12 Alcohol Concentration

The measurement of short chain alcohols (methanol, ethanol, propanol) can be useful to determine a range of properties, such as the level of alcohol in food and drink, or metabolism rates. Alcohol can be metabolized via many pathways, and is broken down by two enzymes – alcohol dehydrogenase (ADH) and aldehyde dehydrogenase (ALDH) – so measuring the properties of these enzymes can give a wealth of information.

4.13 Lactate Enzymatic Assay

Lactate is a metabolite produced by the breakdown of glucose in humans and animals via anaerobic metabolism. It forms when NADH is oxidized to NAD+, due to the reduction of pyruvate to lactate, which is catalyzed by the enzyme lactate dehydrogenase (LDH). Measuring the activity of LDH can help understand information about the process of the metabolism process.

4.14 Performing Enzyme Assays With Photopette Cell

In traditional enzyme assays, performing measurements via a spectrophotometer typically means obtaining samples in a cuvette and repeating. With **Photopette Cell**, our handheld device has been designed for a sampling free workflow to increase efficiency and decrease cross-contamination. Dedicated to measurements at 340 nm, 570 nm and 600 nm, Photopette Cell is the ideal instrument for enzyme assays.

4.15 What is SGPT Blood Test?

SGPT means Serum Glutamic Pyruvic Transaminase. This test is done to measure the amount of Glutamate Pyruvate Transaminase (GPT) in blood serum. GPT is an enzyme found in heart cells, kidney, muscles and liver. An SGPT test is needed on a regular basis to keep the liver in a healthy state. We know the following with a SGPT Blood Test:

- If there's any disease or damage to the liver
- Current status of liver's functioning
- The current level of SGPT
- Identify diseases like hepatic failure, liver disorder, hepatitis, and jaundice

The normal SGPT level is about 7 to 56 units per liter of serum, which might vary according to the techniques used by different laboratories. About 5-ml of blood sample is required to conduct this test.

4.16 What causes abnormal SGPT levels?

The health conditions given below can can cause a high SGPT level:

- Acute viral hepatitis A and B
- Hepatitis C
- Celiac disease
- Epstein-barr virus
- Diabetes

- Heart attack
- Obesity
- Gallbladder inflammation

SGPT:

Serum glutamic pyruvic transaminase, an enzyme that is normally present in liver and heart cells. SGPT is released into blood when the liver or heart are damaged. The blood SGPT levels are thus elevated with liver damage (for example, from viral hepatitis) or with an insult to the heart (for example, from a heart attack). Some medications can also raise SGPT levels. Also called alanine aminotransferase (ALT).

How to keep the SGPT level in control?

A good lifestyle with proper food and exercise can keep our liver healthy. Vitamin D can help in preventing liver diseases and also in reducing the SGPT levels.

How to get the SGPT test done?

Indus Health Plus' Master Health Checkup Package includes all essential tests as well as SGPT. It is imperative to check the status of the body's functions at least once in a year. Considering that an increase in the levels of SGPT doesn't show any symptoms, a regular health checkup is the easiest way to keep track of our health status. SGPT means Serum Glutamic Pyruvic Transaminase. This test is done to measure the amount of Glutamate Pyruvate Transaminase (GPT) in blood serum. GPT is an enzyme found in heart cells, kidney, muscles and liver.

The liver is located in the right upper portion of the abdomen just below the rib cage. Some of the important functions of the human liver are as follows:

- Detoxification or purification of blood
- Producing important clotting factors, albumin, and many other proteins
- Processing medications and nutrients
- Processing of waste products of haemoglobin and other cells of the body
- Storage of vitamins, fat, cholesterol, and bile
- Production of glucose for functions of body

4.17 Liver blood function tests

Liver function tests can be used to know about the liver functions or liver injury. The most common and widely used liver enzymes tests are the aminotransferases that include aspartate aminotransferase (AST or SGOT) and alanine aminotransferase (ALT or SGPT). These enzymes are normally present within liver cells and to a lesser extent in the muscle cells. If the liver is injured or damaged, the liver cells spill these enzymes into the blood, which causes the SGOT and SGPT enzyme blood levels to rise.

4.18 What are the normal levels of SGOT and SGPT?

- Normal levels of SGPT: **7-56 unit**s/liter of serum
- Normal levels of SGOT: **5-40 unit**s/liter of serum.

4.19 What are the elevated level of SGOT and SGPT?

SGPT is present predominantly in the liver and when it is produced in excess, it leaks into the bloodstream. The normal range of SGPT is about 7 to 56 units / liter of blood serum. A very high level of SGPT in the blood indicates some damage or issues related to the liver.

4.20 What are the causes of high SGPT levels?

Following are few conditions and diseases causing increased SGPT levels:

- Drinking excess of alcohol
- Acute viral hepatitis A and B
- Celiac disease (Autoimmune disorder)
- Heart attack
- Diabetes
- Obesity
- Hepatitis C
- Infectious mononucleosis Glandular fever caused by Epstein-Barr virus)
- Gallbladder inflammation (cholecystitis)
- Dermatomyositis (Inflammation of skin and muscles)

4.21 What are symptoms of high SGPT level?

The most common symptoms of high SGPT level include:

- Nausea and vomiting
- Weakness or fatigue

- Swelling in legs
- Shortness of breath
- Excessive bruising or bleeding
- Jaundice

If you find these symptoms, you should immediately consult your doctor for further investigations.

4.22 What are the causes of high SGOT level?

The high SGOT levels can indicate the following disorders:

- Pancreatitis
- heart damage, possibly from a heart attack
- muscle injuries
- kidney disease
- liver damage from toxins such as excess of alcohol
- acute hepatitis
- gallbladder disease

4.21 Which diseases Cause elevated SGOT and SGPT in the blood?

The most common diseases that causes abnormally high SGOT and SGPT are:

- Hepatitis A or B or C
- Chronic viral hepatitis,
- Cirrhosis of the liver (fibrosis of liver because of prolonged inflammation of the liver),
- Liver damage from alcohol,
- Hemochromatosis (a genetic condition caused due to long-standing liver damage), and
- Reduced blood flow to the liver (from shock or heart failure).

What are the Simple and Home measures to lower your SGPT and SGOT levels in the blood?

If the liver is healthy then our body is healthy too. Liver helps in many of body functions and also helps to flush out toxins by boosting the metabolism. One should take care of the liver by maintaining a healthy lifestyle or following healthy habits. Also, following a healthy lifestyle

can keep you strong and keeps your liver away from various diseases. Everything and anything we consume, such as medicines, food, alcohol, etc. is filtered by our liver.

Therefore, it is very important for the body to function properly and take care of the liver that can help avoiding harsh treatments and complications of the certain diseases on the body. SGPT and SGOT are certain enzymes that are produced by the liver and its cells. Elevated SGPT and SGOT levels are an indication of liver cell injury or damage and hence they should be detected through regular health check-ups. There are few lifestyle habits that can keep your liver and body in their healthier state.

Keep a watch on everything you eat.

- Increase Vitamin D intake in diet: Vitamin D helps in preventing any damage happening to the liver and decreases the SGPT levels. Oranges, mushrooms, soy milk, apples, fortified cereals, tofu, eggs, dairy products, oysters cod liver oil, and green leafy vegetables are very good sources of vitamin D.
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- Eat nutritious diet: Organic and plant-based diet is the best for your liver function. Avoid having too much salt and sodium-rich foods.

- Eat more vegetables and fruits: Fruits and vegetables are high in antioxidants, such as papaya, kiwi, pomegranate, carrot, spinach, bell pepper, etc. are good for liver.

- Avoid fried and junk foods: Try to avoid eating deep fried foods and junk food such as pizza, burgers, etc. Also, try to be away from chicken, meat, pork, cheese, carbonated drinks, bacon and butter.

- Limit alcohol and quit smoking: Alcohol is the most dangerous thing for the liver. Consuming excess of alcohol can produce toxins and damage the liver raising the SGOT and SGPT levels in your blood. Also quit smoking as it can also cause damage.

- Don’t try to self-medicate: Absorbing medicines is too much stress on the liver, therefore having medicines that are not prescribed by a qualified doctor can do more harm to the liver.

- Eat nutritious diet: Organic and plant-based diet is the best for your liver function. Avoid having too much salt and sodium-rich foods.
- Eat more vegetables and fruits: Fruits and vegetables are high in antioxidants, such as papaya, kiwi, pomegranate, carrot, spinach, bell pepper, etc. are good for liver.
- Avoid fried and junk foods: Try to avoid eating deep fried foods and junk food such as pizza, burgers, etc. Also, try to be away from chicken, meat, pork, cheese, carbonated drinks, bacon and butter.
- Limit alcohol and quit smoking: Alcohol is the most dangerous thing for the liver. Consuming excess of alcohol can produce toxins and damage the liver raising the SGOT and SGPT levels in your blood. Also quit smoking as it can also cause damage.

- Don’t try to self-medicate: Absorbing medicines is too much stress on the liver, therefore having medicines that are not prescribed by a qualified doctor can do more harm to the liver.

4.22 Regularly exercise

Doing regular exercise is important for maintaining a healthy liver. It decreases the stress on the liver and also increases the energy levels. Exercise helps in preventing obesity, which is one of the risk factors for liver diseases.

4.23 Regular health check-ups

Go for regular health check-ups, as liver dysfunctions do not always show symptoms. Hence, don’t wait for too long for symptoms to occur. Regular preventive screening is one way to know if the liver is functioning properly. High SGPT or SGOT is usually an indication of liver cell injury. Implementing these healthy lifestyle modifications with a healthy diet is the best resolution to lower SGPT and SGOT levels.

4.24 Creatine Phosphokinase

What Is Ck And Cpk?

Creatine Kinase (CK) also referred to as creatine phosphokinase (CPK) or phosphocreatine kinase is an enzyme in the body that causes the phosphorylation of creatine. Creatine kinase (CK) is found in the skeletal muscle, cardiac muscle, brain, bladder, stomach and colon. The CPK levels can be elevated by muscle diseases or muscle necrosis. CPK leaks into the blood when a muscles tissue is damaged, and as such high levels of CPK is indicative of stress or injury to the heart or other muscles. CK enzymes comprises of two subunits, which is CK (B) brain type and CK (M) muscles type.

What Is Cpk Test?

A Creatine Phosphokinase or CPK test determines the levels of enzymes in the blood stream. The CPK test usually administered in the following cases;

- To help diagnose a heart attack
- To ascertain the cause of chest pain
- To assess the extent of damage to heart or muscle tissue
- To determine muscular dystrophy.

The varied diseases diagnosed with the help of a CPK test are as follows;

- Dermatomyositis

- Polymyositis
- Malignant hyperthermia
- Other conditions related to muscle breakdown.

4.25 Introduction

Creatine phosphokinase (CPK), also known by the name creatine kinase (CK), is the enzyme that catalyzes the reaction of creatine and adenosine triphosphate (ATP) to phosphocreatine and adenosine diphosphate (ADP). The phosphocreatine created from this reaction is used to supply tissues and cells that require substantial amounts of ATP, like the brain, skeletal muscles, and the heart, with their required ATP. The normal CPK level is considered to be 20 to 200 IU/L. Many conditions can cause derangement in CPK levels, including rhabdomyolysis, heart disease, kidney disease, or even certain medications.

4.26 Pathophysiology

4.26.1 Molecular

There exist four major isoenzymes of CPK, with two in the cytosol and two in the mitochondria. The cytosol variants are the muscle type (M chain) and the brain type (B chain). The cytosol variants can produce the heterodimer CK-MB or homodimers CK-MM and CK-BB. CK-MB is mostly present in cardiac muscle, CK-MM in skeletal muscle, and CK-BB in smooth muscle and most non-muscle tissues such as the brain. The mitochondrial variants are an octamer of Mi-CK in the brain and a dimer of Mi-CK in muscle. CK-MB further subdivides into two types, CK-MB1 and CK-MB2, when it is in the bloodstream.

Normally, creatine phosphokinase occurs in heart tissue, skeletal muscles, the brain, etc. However, upon muscular injury, there is leakage of CPK into the bloodstream. Thus, CPK is indicative of muscular damage. CK-MB is a more specific indicator of myocardial muscle damage, while CK-MM is more indicative of skeletal muscle damage.

4.26.2 Clinical Significance

Creatine kinase activity is one of the oldest markers of acute myocardial infarction (AMI). Creatine kinase activity begins to rise within 12 hours of AMI symptoms, peaks at 24 to 36 hours, and normalizes after 48 to 72 hours. The issue with measuring creatine kinase activity for AMI is that it is not specific to the heart. CK activity can increase in several conditions such as rhabdomyolysis, chronic muscle diseases, burns, and even after strenuous exercise. Thus, the CK-MB isoenzyme started being used to aid in the diagnosis of AMI. Although the CK-MB measurement is an improvement over just CK, it can still increase in other conditions such as acute muscle injury, congestive cardiac failure, and arrhythmias.

Elevated levels of CK-MB have long been used to diagnose a case of AMI. Although many centers are now going by troponin levels instead of CK-MB, there is a newer, more specific CK-MB method. The new testing method involves measuring the values of the CK-MB1 and CK-MB2 isoforms. In a normal patient, the ratio of CK-MB2 to CK-MB1 should be at 1 to 1. In the case of AMI, the ratio will be at its peak within 4 hours of the infarction. However, some evidence of AMI can be detected as early as 1-2 hours after the infarction. To diagnose AMI, the ratio of CK-MB2 to CK-MB1 should be greater than 1.7 to 1. However, even a ratio of more than or equal to 1.5 to 1 points strongly to a diagnosis of AMI. Patients with Alzheimer disease and Pick disease may have decreased CPK activity in the brain. The BB-CK activity primarily decreased in these patients, resulting in an overall decrease in total CPK activity.

CPK levels also elevate in patients with rhabdomyolysis. Rhabdomyolysis may result from a crush injury, drug use, viral infections, and strenuous exercise. It typically presents with muscle pain and weakness alongside dark-colored urine. There is a breakdown of skeletal muscle, which leads to a release of CPK along with alanine aminotransferase (ALT), aspartate aminotransferase (AST), and electrolytes. The reason for the dark urine is due to myoglobinuria. A CPK level that increases to more than 1000 IU/L is indicative of rhabdomyolysis; values over 5000 IU/L indicate severe rhabdomyolysis. Patients with sickle cell trait who suddenly start a new strenuous exercise program such as spin class are also at an increased risk of rhabdomyolysis, with reported levels of creatine kinase higher than 70000 IU/L in some cases. The most common complication resulting from rhabdomyolysis is acute kidney injury. Therefore, any patient with suspected rhabdomyolysis should receive prompt treatment with intravenous fluids to preserve kidney function.

Patients on statins such as simvastatin may have an adverse effect of significantly elevated CPK levels, potentially leading to rhabdomyolysis. This adverse effect becomes amplified if the patient also receives a concurrent drug that inhibits cytochrome P450-3A4 (CYP3A4). Some common medications to avoid in patients on statin therapy include clarithromycin, erythromycin, verapamil, tamoxifen, and many antifungal agents.

4.26.3 Enhancing Healthcare Team Outcomes

Creatine phosphokinase is an important enzyme, more so in the diagnosis of rhabdomyolysis than AMI in the current medical setting. It is essential in patients with sickle cell anemia or sickle cell trait. It requires careful management with an interprofessional team consisting of a pediatrician and geneticist. The geneticist should assess the type of sickle cell disease during

newborn screening. The pediatrician should advise the parents of the child that there exist increased chances of rhabdomyolysis from strenuous exercise in children with sickle cell anemia and sickle cell trait.

In a patient who already presents with rhabdomyolysis after an increased creatine phosphokinase level, an interprofessional team consisting of a nephrologist, surgeon, and nurse may manage the condition. The nephrologist would be working to increase the kidney function in such patients as acute kidney injury is the most common complication of rhabdomyolysis. The surgeon may need to surgically repair any damaged muscle or tissue that leads to the condition. The nurse should teach the patient about managing their condition and how to avoid having an attack of rhabdomyolysis again. The healthcare team can also consult with the pharmacist to verify that any of the patient's medications are not potential sources for elevated CPK. Any hospital staff members working in the emergency department should be aware that intravenous fluid therapy should be started promptly to curb acute kidney injury in patients with suspected rhabdomyolysis.

4.26.4 Lactate dehydrogenase (LDH)

Lactate dehydrogenase (LDH) is an oxidoreductase enzyme that catalyses the interconversion of pyruvate and lactate. Cells release LDH into the bloodstream after tissue damage or red blood cell hemolysis. Since LDH is a fairly stable enzyme, it has been widely used to evaluate the presence of damage and toxicity of tissue and cells. LDH is also elevated in certain pathological conditions such as cancer. Quantification of LDH has a broad range of applications. LDH catalyzes the synthesis of lactate and pyruvate in a reversible reaction, and it is commonly used as a biomarker of cell damage or death.

Analysis of LDH and its isoenzymes in skeletal muscles of control rats revealed that LDH activity was highest in EDL followed by diaphragm, and it was lowest in soleus. Compared to muscles, the enzyme activity is low in serum. Diaphragm and serum contained all five electrophoretically distinct LDH isoenzymes, but with varying quantity. Diaphragm had predominantly LDH-5 and LDH-4 (40 and 27.7%, respectively). In control serum, isoenzyme LDH-5 comprises 87% of the total LDH activity (100%).

4.27 Application

Lactate dehydrogenase activity assay kit has been used for measuring the activity of lactate dehydrogenase (LDH) in the cell culture medium.

Lactate Dehydrogenase Activity Assay Kit has been used to determine the concentration of lactate dehydrogenase in samples. Lactate dehydrogenase (LDH) is a nonspecific marker of hepatocellular injury. Extremely elevated LDH signifies massive hepatocyte damage, usually from ischemia or drug-induced hepatotoxicity (such as acetaminophen overdose). These patients will also have extreme elevations in AST and ALT. Elevated LDH concomitant with elevated alkaline phosphatase (AP) suggests malignant infiltration of the liver. Extrahepatic disorders that cause LDH elevation include hemolysis, rhabdomyolysis, tumor necrosis, renal infarction, acute stroke, and myocardial infarction.

4.28 Suitability

Suitable for the detection of L (+)-Lactate in biological samples such as serum, plasma, cells, culture and fermentation media.

4.29 Principle

In this kit, LDH reduces NAD to NADH, which is specifically detected by colorimetric (450 nm) assay. The LDH Activity Assay kit quantifies LDH activity in variety of biological samples. The assay is quick, convenient, and sensitive.

4.30 Summary

Under this unit we summarize that diagnostic enzymes, enzyme mechanism and enzyme assays such as SGPT and LDH. Over the years, enzymes have emerged as critical regulators of human diseases. In addition, as biomarkers, some enzymes often present consequential cue about the disease state due to their corresponding altered pattern in tissues and serum. A vast embodiment of data from experimental research in model organisms and humans has ensured the clinical utilization of specific enzymes as an evidence-based strategy in disease prevention and diagnosis. These have been incorporated in the diagnostic premises as biomarkers, which are quantifiable laboratory measure of a disease specific biologically relevant molecule that can act as an indicator of a disease burden. This chapter provides a brief review of the current paradigm on harnessing the clinical utility of enzymes with potential impact in the diagnosis of cancer disease, myocardial disorders and gastrointestinal tract related impairments in humans.

Enzymes are biocatalysts and because of their remarkable properties, they are extensively used in medical diagnosis. Researches in the last two decades have concentrated more on enzymes such as creatine kinase-MB, alanine transaminase, aspartate transaminase, acid phosphatase, alkaline phosphatase etc. for clinical applications. Enzymes are the preferred

markers in various disease states such as myocardial infarction, jaundice, pancreatitis, cancer, neurodegenerative disorders, etc. They provide insight into the disease process by diagnosis, prognosis and assessment of response therapy.

4.31Terminal questions

Q. 1 What do you mean by diagnostic enzymes? Describe it.

Answer:----------

Q.1. What are the role of enzymes in health and diseases?

Answer:----------

Q.2. Describe the mechanism of enzyme action.

Answer:----------

Q.3. What are the enzyme assays? Explain LDH

Answer:----------

Q.4. What are applications of diagnostic enzymes?

Answer:----------

Q.5. Write a short note on SGPT.

Answer:----------

Q.6. Explain biochemical diagnosis of diseases.

Answer:----------

Further readings

1. Biochemistry- Lehninger A.L.
2. Biochemistry –J.H.Weil.
3. Biochemistry fourth edition-David Hames and Nigel Hooper.
4. Textbook of Biochemistry for Undergraduates - Rafi, M.D.
5. Biochemistry and molecular biology- Wilson Walker.

Unit-5

5.1 Introduction

Nutrition can affect the body's response to drugs; conversely, drugs can affect the body's nutrition. Foods can enhance, delay, or decrease drug absorption. Foods impair absorption of many antibiotics. They can alter metabolism of drugs; eg, high-protein diets can accelerate metabolism of certain drugs by stimulating cytochrome P-450. Eating grapefruit can inhibit cytochrome P-450 34A, slowing metabolism of some drugs (eg, amiodarone, carbamazepine, cyclosporine, certain calcium channel blockers). Diets that alter the bacterial flora may markedly affect the overall metabolism of certain drugs. Some foods affect the body's response to drugs. For example, tyramine, a component of cheese and a potent vasoconstrictor, can cause hypertensive crisis in some patients who take monoamine oxidase inhibitors and eat cheese.

Nutritional deficiencies can affect drug absorption and metabolism. Severe energy and protein deficiencies reduce enzyme tissue concentrations and may impair the response to drugs by reducing absorption or protein binding and causing liver dysfunction. Changes in the gastrointestinal tract can impair absorption and affect the response to a drug. Deficiency of calcium, magnesium, or zinc may impair drug metabolism. Vitamin C deficiency decreases activity of drug-metabolizing enzymes, especially in older people.

Many drugs affect appetite, food absorption, and tissue metabolism. Some drugs (eg, metoclopramide) increase gastrointestinal motility, decreasing food absorption. Other drugs (eg, opioids, anticholinergics) decrease gastrointestinal motility. Some drugs are better tolerated if taken with food. Certain drugs affect mineral metabolism. Certain antibiotics (eg, tetracyclines) reduce iron absorption, as can certain foods (eg, vegetables, tea, bran).

Objectives

This is the fifth unit on clinical biochemistry. Under fifth unit we have following objectives. These are as under:

- To know about nutrition and drugs
- To know about routine hospital diets and special feeding methods.
- To discuss drugs, alcohol and toxicants

Many drugs are capable of interacting with nutrients and, therefore, nutritional feeds. Pharmacists need to be aware of when such interactions may occur. This article outlines the key mechanisms and factors to be considered**.** Drugs can interact with nutritional compounds, and their pharmacokinetics can be affected by a patient's nutritional status. These effects can be clinically positive or negative and vary in significance. The interrelationships between drugs and nutrients are complex, so there are many ways in which drugs and nutrients, or nutritional therapy, can interact.

For example:

- A nutrient may bind to a drug and reduce its, or the drug's, absorption
- A drug may change the consistency of the feed formulation
- A nutrient may be involved in the metabolism of a drug, therefore promoting its clearance.
- A nutrient's excretion may be affected by a drug.

Pharmacists should have a working knowledge of drug-nutrient interactions. This allows them to offer counselling on the optimum timing of drug administration around meals. Medicines used to treat HIV, or to prevent organ rejection after trans-plant, are particularly susceptible to nutrient interactions. The British National Formulary or summary of product characteristics should be consulted for individual preparations.

5.2 Direct absorption effects

There are many reported interactions between drugs and individual dietary components.

5.2.1 Nutrient absorption

Drugs can affect nutrient absorption. Antacids containing aluminium or magnesium hydroxide bind to dietary phosphate to form insoluble phosphate salts that cannot be absorbed. Osteomalacia has been reported as a result of phosphate depletion due to antacid abuse.

5.2.3 Tetracyclines

Tetracyclines readily chelate with divalent and trivalent metal cations (eg, calcium, magnesium, iron), for which there are higher concentrations in enteral feed and milk-based diets than normal diets. The chelated drug molecule changes in size and solubility, hence experiences reduced absorption from the gastrointestinal (GI) tract. Tetracycline absorption can be reduced by 80 per cent if it is co-administered with a nutritional feed. However, unlike other tetracyclines, doxycycline absorption is not affected by simultaneous ingestion of food and milk.

5.2.4 Macrolides

Ciprofloxacin absorption is decreased by 50 per cent if it is given with enteral feed. This is thought to be because drug molecules bind to divalent ions. A similar reaction occurs with levofloxacin and ofloxacin, although it is less significant. There is no reaction with moxifloxacin. Ciprofloxacin also chelates with ions in tap or mineral water. However there is currently no research published on the significance of this interaction.

5.2.5 Protein binding

Reports of reduced absorption of phenytoin and warfarin highlight the potential for a drug to bind to dietary protein in the GI tract, consequently reducing its absorption. There a recurrently no reports comparing the inter-action between these drugs and whole dietary protein, to that with the amino acids found in enteral feeds.

5.2.6 Pectin and fibre

Paracetamol absorption is reduced by pectin, which is present in apples and pears, although the significance of this interaction is unknown. The absorption of digoxin is reduced if it is taken at the same time as fibre-rich foods. The significance of this is negligible for patients who eat a healthy, balanced diet, but because digoxin has a narrow therapeutic index, its blood concentration should be closely monitored if a patient changes his or her fibre intake considerably (eg, from a high fibre normal diet to a low fibre enteral feed).

5.2.7 Indirect absorption effects

Most drugs are absorbed by passive diffusion from the gut lumen into the splanchnic circulation, across the GI mucosa. The rate of diffusion depends on contact time (between the drug and the mucosa) and the drug’s lipophilicity.

5.2.8 Delayed gastric emptying

Hot food and fatty meals delay gastric emptying to a greater extent than high protein or carbohydrate meals. This has two effects:

- It increases the time the drug spends in the stomach, which can increase drug disintegration and dissolution (and possibly degradation of acid-sensitive drugs)
- It delays the time taken for peak blood concentrations to occur for drugs absorbed from the small intestine.

The combined effect usually increases the overall absorption of drugs, and the delay and reduction in peak plasma concentrations can be beneficial (eg, taking nifedipine with a meal delays and reduces the severity of the drug's flushing side effect).

5.2.9 Increased hepatic bioavailability

The presence of food or enteral feed in the small intestine increases splanchnic blood flow. This may enhance drug absorption, and may also increase bioavailability, because blood flow through the portal system (to the liver)is reduced. This is the suggested mechanism by which concentrations of propranolol and metoprolol are higher if taken with food compared with when they are taken during a period of fasting.

5.3 Other GI effects

Bile salts, released in response to fat ingestion, may promote the absorption of highly lipid-soluble drugs, such as griseofulvin. Drugs can reduce the absorptive capacity of the GI tract. For example, colchicine is associated with a malabsorptive diarrhoea, and long-term treatment with metformin is associated with vitamin B12 deficiency. Some drugs cross the GI mucosa using active transport systems that are usually used to transport nutrients (eg, methyldopa and levodopa). The absorption of such drugs is decreased by a high protein diet.

5.4 Changes in feed consistency

Alterations in feed consistency are caused by a physical interaction between the drug and a component of the feed or the feed formulation — resulting in a physical change in the consistency of the feed. This can block an enteral feeding tube, or in extreme cases, result in physical obstruction of the GI tract. The drug that has been most commonly reported to interact with enteral feed in this way is sucralfate. Adequate flushing between feeds and drug administration will minimise this effect. Drugs should never be added directly to a nutritional feed. Guidance for pharmacists dealing with enteral drug administration are be found in the British Pharmaceutical Nutrition Group handbook.

5.5 Metabolism interactions

Several foods can affect drug metabolism. The most well known is grapefruit juice, although interactions with pomegranate juice have become apparent more recently.

5.6 Grapefruit juice

Cytochrome P450 is a family of drug metabolising enzymes found predominantly in the small intestine and the liver. Grapefruit juice contains furanocoumarins, and other substances, that inhibit intestinal CYP3A, therefore reducing the metabolism of orally administered drugs. Intestinal drug transporters, such as p-glycoprotein, may also be inhibited. Research has identified many drugs affected by this interaction. These drugs tend to have a low oral bioavailability, indicating significant presystemic metabolism in the intestine or the liver. Examples of drugs affected by this reaction include:

- Antibiotics (eg, erythromycin,albendazole, saquinavir)
- Cardiovascular drugs (eg, amiodarone,felodipine, verapamil)
- Phosphodiesterase type-5 inhibitors
- Immunosuppressants (eg, cyclosporin)

5.7 Pomegranate juice

The effect of pomegranate juice on drug metabolism is the subject of current research. It may affect CYP2C9, an enzyme also inhibited by fluconazole. It is possible that drugs whose metabolism is affected by fluconazole may also be affected by pomegranate juice. The clinical importance of this is not yet known.

5.8 Nutrient excretion

The most common group of drugs that affect nutrient excretion are the diuretics. Their ability to increase the excretion of electrolytes have been widely documented. Other drugs that increase electrolyte excretion include amphotericin, and if cisplatin causes nephrotoxicity, this can increase the excretion of magnesium and zinc.

5.9 Pharmacist responsibilities

Pharmacists must have access to all the necessary information relating to:

- The drug and its formulation
- The patient’s condition
- The type of feeding tube, type of enteral feed and regimen being used

This information is used to recommend a suitable formulation for administration via a specified route. Pharmacists are responsible for informing the appropriate medical team if a

drug is being administered by an unlicensed route. When a drug formulation is changed, pharmacists must ensure that the dose of the new formulation is bioequivalent to the previous dose, to avoid treatment failure or toxicity.

In primary care, community pharmacists may not have access to information relating to the intended route of administration. Pharmacists supplying care homes should be aware of the route of administration for the medicines they supply. Information regarding drug administration via feeding tubes for care home patients should be communicated to the GP and community pharmacist when these patients are discharged from hospital. Every pharmacist has a role to play in minimising drug-nutrient interactions.

5.10 Rotine hospital diets

5.11 Clear Fluid Diet

This diet is made up of clear fluids that leave no residue, and it is non gas forming, non irritating and non stimulating to peristaltic action. This diet can meet the requirement of fluids and some minerals and can be given in 1 to 2 hour intervals. The foods which can be included are barley water, dhal water, tea and coffee without milk, etc.

5.12 Full Fluid Diet

This diet bridges the gap between the clear fluid and soft diet In this diet, foods which are liquid or which readily become liquid on reaching the stomach are given. It is used following operations, in acute gastritis, acute infections and in diarrhoea. This diet is given at 2 - 4 hours interval. The foods included are kanji, milk shakes, lassi, custards, etc.

5.13 Soft Diet

It may be used in acute infections, following surgery, and for patients who are unable to chew. The soft diet is made up of simple, easily digested food and contains no harsh fibre and no rich highly seasoned food. In this diet, three meals with intermediate feedings should be given.

5.14 Special feeding methods

5.14.1 Tube Feeding

This is done by passing a tube into the stomach or duodenum through the nose which is called nasogastric feeding or directly by surgical operation known as gastrostomy and jejunostomy feeding. The type of foods supplied through the tube may be.

5.14.2 Natural liquid foods

Solid foods blenderised to make liquid food.

Commercially supplied polymeric mixtures or elemental diet like Complan, Horlicks, etc.

5.14.2 The advantages of tube feeding are

Adequate nutrition could easily be given by this method.

Foods and drugs which may not be liked by the patients can be administered.

5.14.3 Parenteral Feeding

Here the nutrient preparations are given directly into a vein. This method may be used to supplement normal feeding by mouth but can provide all the nutrients necessary to meet a patient's requirements. Then it is known as total parenteral nutrition or TPN. The nutrients given in TPN are glucose, emulsified fat, crystalline aminoacids, Vitamins including B_{12}, folic acid and vitamin K, electrolytes like sodium, potassium, calcium and magnesium, trace elements like Zinc, Copper, Iodine and Water.

5.14.4 Enteral

The term, enteral, refers to nutrition administered via the gastrointestinal tract. It may be administered orally or via tube feeding.

5.14.5 Oral

Oral nutritional supplements (ONS) are nutrition support products that provide an effective and non-invasive way for people to meet their nutrition needs or increase their nutritional intake. People who take ONS may also be able to eat regular food but cannot meet all their nutritional requirements through a regular diet alone and thus require supplemental nutrition. In other instances, a patient can benefit from ONS if they require a liquid-based diet. ONS products are often prescribed or recommended by a physician or registered dietitian. In some cases, people rely on ONS as their sole source of nutrition.

5.14.6 Tube Feeding

If a person has a condition or illness which limits or impairs oral intake, enteral nutrition (EN) therapy can be administered directly into the gastrointestinal tract as a tube feeding.[1] Enteral nutrition via tube feeding provides life-sustaining nutrients and is often required as a first option feeding method when a person is unable to consume food orally and/or has an impaired digestive system. EN therapy includes specialized liquid feedings containing protein, carbohydrates, fats, vitamins, minerals, and other nutrients needed to live. These nutrition support products are formulated to meet individual needs for a variety of disease states and conditions.

5.15 Parenteral

Parenteral nutrition (PN) is the intravenous administration (feeding into a vein) of nutrients directly into the systemic circulation, bypassing the gastrointestinal tract.[2] It is a special liquid mixture containing protein, carbohydrates, fats, vitamins, minerals, and other nutrients needed

to live. PN represents an alternative or additional approach for nutrition intervention when nutrition needs cannot be met from the oral or enteral routes alone, or are contraindicated.

5.16 Special feeding methods

Feeding methods (hand-feeding, blower feeding, automatic feeders or demand feeders) may affect tilapia performance. Hand-feeding is recommended in small-scale tilapia farms because it allows the feeder to regulate the amount of feed required, prevent overfeeding and observe fish behaviour and feeding activity. However, automatic feeders, feed blowers and demand feeders are usually used with large commercial farms. Hargreaves et al. found no difference in the performance of *O. aureus* fed by automatic feeders and those fed fixed rations.

They recommended that automatic feeders be used because they reduced 88%–94% of the labour requirement. Rakocy et al. (2000) also recommended demand feeders for tilapia reared in cages suspended in watershed ponds. Similarly, demand feeders provide excellent growth and feed conversion of tilapia reared in marine cages, in addition to reducing labour. Endo et al. (2002) found that self-fed Nile tilapia had significantly lower blood cortisol, higher phagocytic activity of their macrophages, higher antibody production and higher number of blood lymphocytes.

Feeding methods can help control excess nutrient intake. Methods of feeding include free-choice feeding, time-restricted feeding, and food-restricted feeding. Free-choice feeding allows the pet to eat ad libitum, thereby increasing the risk for excess nutrient intake. Time-restricted feeding allows the owner to feed 2 to 3 times per day for a set period of time, which may encourage the pet to eat ravenously, past the normal satiety mechanism. Food-restricted feedings allow the owner to control caloric intake and maintain optimum growth rate and body condition. Determining a daily energy requirement (DER) allows the owner to feed the correct amount of food, which can be changed as the puppy increases in size and age.

The body condition should be evaluated every 2 weeks. Food can be adjusted as needed to decrease excess fat, which will decrease the growth rate. Puppies and kittens should be scored on a 9-point body-condition scale. The ideal body condition is an hourglass shape when viewed from above, with a definitive waist behind the ribs. Environment, genetics, and nutrient composition play key roles in skeletal development. We can minimize the effects of skeletal disease in large breed puppies by regulating nutrient and caloric intake. The goal is to regulate growth rate not maximize it. Feeding an AAFCO-approved commercial diet is recommended to help achieve this goal.

5.17 Tube feeding

A plastic feeding tube is a medical device used to feed a person who is unable to take food or drink safely by mouth. This problem may be due to difficulty swallowing, an altered mental status, or another issue that makes eating a challenge. There are many types of feeding tubes used for different reasons, some temporary and some permanent. If you need to make a decision about feeding tubes for yourself or a loved one, it's important to have good information about them.

5.18 What a Feeding Tube Is For

A feeding tube has uses beyond making sure that someone with dysphagia, who cannot swallow or chew, is fed. The most common uses of a feeding tube include:

- **Providing nutrition**: Food, in liquid form, can be given through a feeding tube. Tube feeding, or enteral nutrition, allows for needed carbohydrates, protein, and fats to support the body.
- **Providing fluids**: Water given through a feeding tube can keep a person hydrated without needing intravenous IV fluids.
- **Providing medication**: Needed drugs, including many pills and tablets, can be given through a feeding tube. Their contents can be powdered and blended with water to administer them.
- **Decompressing the stomach**: Some feeding tubes can be used to remove air in the stomach. Suction connected to the tubes can remove gas, reducing distention (enlargement) and bloating.
- **Removing stomach contents**: Undigested food sitting in the stomach can cause nausea, vomiting, stomach pain, and bloating. Suction can be used to remove fluids and food particles.

5.19 Reasons to Use a Feeding Tube

The body does better with food delivered to the gut rather than having artificial nutrition and fluids sent through an IV and into the blood vessels. It is safer and healthier for a person to receive food and fluids in the stomach for normal digestion. Trouble swallowing can cause a person to choke on food and fluids. They can “go down the wrong pipe” and be inhaled into the lungs through the trachea, rather than into the esophagus that leads to the stomach. This can lead to serious illness, including aspiration pneumonia.

Some people may be too sick to swallow. They may need a ventilator to keep them breathing, which is an endotracheal tube placed in the airway that keeps them from swallowing. Even

fully alert people may lose the ability to swallow. A disease like oral cancer may make a feeding tube necessary. People may need a feeding tube in quite a few health situations that may leave them with an inability to safely swallow food and water. There are also other uses for a feeding tube. They deliver fluids and food in liquid form, but medications too. The tubes also allow for suctioning out air and stomach contents when needed.

5.20 Decision to Place a Feeding Tube

The decision to place a feeding tube is based on more than medical factors alone. It is made more complicated when the person's medication or illness leaves them unable to voice their own wishes. They also may not have previously shared these wishes with you and a healthcare team, or left you with advance directives that explain the types of care they want in situations like this. This may leave families and healthcare providers to instead discuss the choice on a loved one's behalf. In some cases, a decision about placing a feeding tube really means two decisions. That's because a feeding tube is often placed at the same time a tracheostomy is done. This hole in the throat allows for long-term use of a ventilator to support breathing after initial intubation in the trachea.

5.21 Types of Feeding Tubes

The type of feeding tube used will depend on what's causing the problem. Some are intended to be temporary, and can only be used safely for about 14 days. If they are used longer, there is a risk of permanent damage to the larynx (voice box) and tissues in the throat or esophagus. Others are meant to be long-term or even permanent. They can be used for months or even years across a lifetime. They can be removed as needed but don't result in the same complications.

Short-Term Feeding Tubes

5.22 Nasogastric (NG) tube: This type of tube is inserted into the nose and down through the throat. It is threaded into the esophagus and rests in the stomach. It can stay in place for four to six weeks before it is removed entirely or replaced with a long-term feeding tube.[4]

5.23 Orogastric (OG) tube: This is the same type of tube as the NG tube, but it is inserted into the mouth instead. It then follows the same pathway through the throat and esophagus, and into the stomach. It can remain there for up to two weeks before it is removed or replaced.

Long Term Feeding Tubes

5.24 Gastric tube (G tube): The G tube offers direct access to the stomach through a surgical cut in the left upper side of your abdomen. This means it bypasses the mouth and throat entirely. It allows for food, fluids, and medication to be given without swallowing.

5.25 Jejunostomy tube (J tube): Like the G tube, the J tube is placed through an incision in the abdomen. But this cut is placed lower than the G tube so that the tube ends in the middle third of the small intestine, known as the jejunum. It tends to be smaller than the G tube, so only thin liquids and powdered drugs can pass through it. Temporary feeding tubes work much the same way, with tubes that end in either the stomach (G tubes) or further into the small intestine (J tubes).

Some G tubes are placed using the percutaneous endoscopic gastrostomy (PEG) technique. This means that a gastroenterologist specializing in digestive disorders will use a lighted instrument called an endoscope to place the G tube through a surgical hole in the stomach. It has a camera attached that allows them to see and confirm this placement.

5.26 Placement Procedure

It doesn't take long to place a feeding tube. Anesthesia is required for some people who would otherwise be conscious during the procedure, but that's often not the case for very ill patients who are sedated and need intensive care. The endoscope is threaded from the mouth into the stomach. The healthcare provider can see the lighted tip of the endoscope and it shows them where to make a small incision. This is about a half-inch long. Then the G tube is passed through it and secured in place.

A cap on the tube can be opened to administer food and fluids directly into the stomach. With good tube care, the incision will close tightly around the tube and prevent leaks in just a few days' time. If the skin is irritated by leaking fluid, an ointment can help to protect it. Washing the site with soap and water will usually be all the care it needs. There are two types of feeding tubes: Those meant for short-term use can stay in place for only a matter of weeks before they need to be removed or replaced. Long-term tubes are safe for permanent use but it's a little more complicated to put them in place.

5.27 Removal Procedure

The procedures for removal depend on whether it is a temporary or permanent feeding tube.

5.28 Temporary Feeding Tube Removal

It's a simple and quick procedure to remove a temporary feeding tube. Any irritation to the mouth, throat, and nose is typically minimal. A syringe is used to empty the tube of food and fluids. It then takes a matter of seconds to withdraw the tube and verify it has been done safely.

5.29 Permanent Feeding Tube Removal

Some people may recover enough ability to eat and drink well, even though their tube is considered permanent. The decision to do so is usually based on whether you've maintained your weight for a month while still on a feeding tube, though some healthcare providers may want more time. The withdrawal process is similar to the temporary tubes but it requires more force. It also may cause more pain and small amounts of blood are not uncommon. These issues resolve quickly. The incision used to place the tube will usually close within a week of its removal.

5.30 Parenteral nutrition (PN)

Parenteral nutrition (PN) is the feeding of nutritional products to a person intravenously, bypassing the usual process of eating and digestion. The products are made by pharmaceutical compounding companies. The person receives a nutritional mix according to a formula including glucose, salts, amino acids, lipids and vitamins and dietary minerals. It is called total parenteral nutrition (TPN) or total nutrient admixture (TNA) when no significant nutrition is obtained by other routes, and partial parenteral nutrition (PPN) when nutrition is also partially enteric. It is called peripheral parenteral nutrition (PPN) when administered through vein access in a limb rather than through a central vein as central venous nutrition (CVN).

5.31 Medical uses

Total parenteral nutrition (TPN) is provided when the gastrointestinal tract is nonfunctional because of an interruption in its continuity (it is blocked, or has a leak – a fistula) or because its absorptive capacity is impaired. It has been used for comatose patients, although enteral feeding is usually preferable, and less prone to complications. Parenteral nutrition is used to prevent malnutrition in patients who are unable to obtain adequate nutrients by oral or enteral routes. The Society of Critical Care Medicine (SCCM) and American Society for Parenteral and Enteral Nutrition recommends waiting until the seventh day of hospital care.

5.32 Absolute indications for TPN

Diseases that would require use of TPN include:

- Short bowel syndrome
- Small bowel obstruction
- Active gastrointestinal bleeding
- Pseudo-obstruction with complete intolerance to food
- High-output (defined as > 500ml/day) enteric-cutaneous fistulas (unless a feeding tube can be passed distal to the fistula)

5.33 Gastrointestinal disorders

TPN may be the only feasible option for providing nutrition to patients who do not have a functioning gastrointestinal tract or who have disorders requiring complete bowel rest, including bowel obstruction, short bowel syndrome, gastroschisis, prolonged diarrhea regardless of its cause, very severe Crohn's disease or ulcerative colitis, and certain pediatric GI disorders including congenital GI anomalies and necrotizing enterocolitis.

5.34 In geriatric population

There are physical, physiological, or mental differences in the geriatric population that could potentially lead to poor nutrient intake that would require them to have nutrition therapy. Geriatric patients are more inclined to have delayed muscle restoration compared to the younger population. Additionally, older patients are observed to have greater cardiac and renal impairment, insulin resistance, and to have deficiencies in vitamins and crucial elements. Patients who require nutrition therapy but have contraindications for or cannot tolerate enteral nutrition are appropriate candidates for parenteral nutrition. In the geriatric population, it is indicated if oral or enteral nutrition is impossible for 3 days or when oral or enteral nutrition is likely insufficient for more than 7 to 10 days. While there are no complications of parenteral nutrition specific to the geriatric population, complications are more prevalent in this population due to increased comorbidities.

5.35 In cancer

Patients who are diagnosed with cancer, whether as outpatient undergoing treatment or hospitalized, are at a greater risk of malnutrition and cachexia. Cancer-related malnutrition can be attributed to the decrease in food intake, increase in the need for energy, and the alteration of metabolism. Patients should be assessed early on in their cancer treatment for any nutritional risk, such as by taking routine weights and BMI. Parenteral nutrition is indicated in cancer patients when it is not possible to access the digestive tract or if the tract is ineffective. In advanced cancer patients, the use of PN should be discussed in context of the risks and benefits, such as if the approximate survival rate is longer than 3 months and if PN would be expected to greatly improve the patients' quality of life. It is uncertain whether home parenteral nutrition improves survival or quality of life in people with malignant bowel obstruction.

5.36 Duration

Short-term PN may be used if a person's digestive system has shut down (for instance by peritonitis), and they are at a low enough weight to cause concerns about nutrition during an extended hospital stay. Long-term PN is occasionally used to treat people suffering the extended consequences of an accident, surgery, or digestive disorder. PN has extended the life of children born with nonexistent or severely deformed organs.

5.37 Living with TPN

Approximately 40,000 people use TPN at home in the United States, and because TPN requires 10–16 hours to be administered, daily life can be affected. Although daily lifestyle can be changed, most patients agree that these changes are better than staying at the hospital. Many different types of pumps exist to limit the time the patient is "hooked up". Usually a backpack pump is used, allowing for mobility. The time required to be connected to the IV is dependent on the situation of each patient; some require once a day, or five days a week.

It is important for patients to avoid as much TPN-related change as possible in their lifestyles. This allows for the best possible mental health situation; constantly being held down can lead to resentment and depression. Physical activity is also highly encouraged, but patients must avoid contact sports (equipment damage) and swimming (infection). Many teens find it difficult to live with TPN due to issues regarding body image and not being able to participate in activities and events.

5.38 Complications

TPN fully bypasses the GI tract and normal methods of nutrient absorption. Possible complications, which may be significant, are listed below. Other than those listed below, common complications of TPN include hypophosphatemia, hypokalemia, hyperglycemia, hypercapnia, decreased copper and zinc levels, elevated prothrombin time (if associated with liver injury), hyperchloremic metabolic acidosis and decreased gastrointestinal motility.

5.39 Infection

TPN requires a chronic IV access for the solution to run through, and the most common complication is infection of this catheter. Infection is a common cause of death in these patients, with a mortality rate of approximately 15% per infection, and death usually results from septic shock. When using central venous access, the subclavian (or axillary) vein is preferred due to its ease of access and lowest infectious complications compared to the jugular and femoral vein insertions.

Catheter complications include pneumothorax, accidental arterial puncture, and catheter-related sepsis. The complication rate at the time of insertion should be less than 5%. Catheter-related infections may be minimised by appropriate choice of catheter and insertion technique.

5.40 Blood clots

Chronic IV access leaves a foreign body in the vascular system, and blood clots on this IV line are common. Death can result from pulmonary embolism wherein a clot that starts on the IV line breaks off and travels to the lungs, blocking blood flow. Patients on TPN who have such clots occluding their catheter may receive a thrombolytic flush to dissolve the clots and prevent further complications.

5.41 Fatty liver and liver failure

Fatty liver is usually a more long-term complication of TPN, though over a long enough course it is fairly common. The pathogenesis is due to using linoleic acid (an omega-6 fatty acid component of soybean oil) as a major source of calories. TPN-associated liver disease strikes up to 50% of patients within 5–7 years, correlated with a mortality rate of 2–50%. Onset of this liver disease is the major complication that leads TPN patients to requiring an intestinal transplant.

Intralipid (Fresenius-Kabi), the US standard lipid emulsion for TPN nutrition, contains a 7:1 ratio of n-6/n-3 ratio of polyunsaturated fatty acids (PUFA). By contrast, Omegaven has a 1:8 ratio and showed promise in multiple clinical studies. Therefore n-3-rich fat may alter the course of parenteral nutrition associated liver disease (PNALD).

5.42 Hunger

Because patients are being fed intravenously, the subject does not physically eat, resulting in intense hunger pangs (pains). The brain uses signals from the mouth (taste and smell), the stomach and gastrointestinal tract (fullness) and blood (nutrient levels) to determine conscious feelings of hunger. In cases of TPN, the taste, smell and physical fullness requirements are not met, and so the patient experiences hunger, although the body is being fully nourished. Patients who eat food despite the inability can experience a wide range of complications, such as refeeding syndrome.

5.43 Cholecystitis

Total parenteral nutrition increases the risk of acute cholecystitis due to complete disuse of the gastrointestinal tract, which may result in bile stasis in the gallbladder. Other

potential hepatobiliary dysfunctions include steatosis, steatohepatitis, cholestasis, and cholelithiasis. Six percent of patients on TPN longer than three weeks and 100% of patients on TPN longer than 13 weeks develop biliary sludge. The formation of sludge is the result of stasis due to lack of enteric stimulation and is not due to changes in bile composition. Gallbladder sludge disappears after four weeks of normal oral diet. Administration of exogenous cholecystokinin (CCK) or stimulation of endogenous CCK by periodic pulse of large amounts of amino acids has been shown to help prevent sludge formation. These therapies are not routinely recommended. Such complications are suggested to be the main reason for mortality in people requiring long-term total parenteral nutrition, such as in short bowel syndrome. In newborn infants with short bowel syndrome with less than 10% of expected intestinal length, thereby being dependent upon total parenteral nutrition, five-year survival is approximately 20%.

5.44 Gut atrophy

Infants who are sustained on TPN without food by mouth for prolonged periods are at risk for developing gut atrophy.

5.45 Hypersensitivity

Hypersensitivity is a rarely described but significant complication of parenteral nutrition therapy. First reported in 1965, the incidence of these reactions is speculated to be around one in 1.5 million patients who are provided parenteral nutrition. There is a wide range in how and when these reactions manifest. Cutaneous manifestations are the most common presentation. Hypersensitivity is thought to occur to the individual components of TPN, with the intravenous lipid emulsion being the most frequently implicated component, followed by the multivitamin solution and the amino acid solution.

5.46 Medications

Patients who are receiving intravenous parenteral nutrition may also need to receive intravenous medications as well using the same Y-site. It is important to assess the compatibility of the medications with the nutrition components. Incompatibilities can be observed physically through discoloration, phase separation, or precipitation.

5.47 Metabolic complications

Metabolic complications include the refeeding syndrome characterised by hypokalemia, hypophosphatemia and hypomagnesemia. Hyperglycemia is common at the start of therapy, but can be treated with insulin added to the TPN solution. Hypoglycaemia is likely to occur with abrupt cessation of TPN. Liver dysfunction can be limited to a reversible

cholestatic jaundice and to fatty infiltration (demonstrated by elevated transaminases). Severe hepatic dysfunction is a rare complication. Overall, patients receiving TPN have a higher rate of infectious complications. This can be related to hyperglycemia.

5.48 Pregnancy

Pregnancy can cause major complications when trying to properly dose the nutrient mixture. Because all of the baby's nourishment comes from the mother's blood stream, the doctor must properly calculate the dosage of nutrients to meet both recipients' needs and have them in usable forms. Incorrect dosage can lead to many adverse, hard-to-guess effects, such as death, and varying degrees of deformation or other developmental problems. It is recommended that parenteral nutrition administration begin after a period of natural nutrition so doctors can properly calculate the nutritional needs of the fetus. Otherwise, it should only be administered by a team of highly skilled doctors who can accurately assess the fetus' needs.

5.49 Drugs

A **drug** is any chemical substance that causes a change in an organism's physiology or psychology when consumed. Drugs are typically distinguished from food and substances that provide nutritional support. Consumption of drugs can be via inhalation, injection, smoking, ingestion, absorption via a patch on the skin, suppository, or dissolution under the tongue.

In pharmacology, a drug is a chemical substance, typically of known structure, which, when administered to a living organism, produces a biological effect. A pharmaceutical drug, also called a medication or medicine, is a chemical substance used to treat, cure, prevent, or diagnose a disease or to promote well-being. Traditionally drugs were obtained through extraction from medicinal plants, but more recently also by organic synthesis. Pharmaceutical drugs may be used for a limited duration, or on a regular basis for chronic disorders.

Pharmaceutical drugs are often classified into drug classes—groups of related drugs that have similar chemical structures, the same mechanism of action (binding to the same biological target), a related mode of action, and that are used to treat the same disease. The Anatomical Therapeutic Chemical Classification System (ATC), the most widely used drug classification system, assigns drugs a unique ATC code, which is an alphanumeric code that assigns it to specific drug classes within the ATC system. Another major classification system is the Biopharmaceutics Classification System. This classifies drugs according to their solubility and permeability or absorption properties. Psychoactive drugs are chemical substances that affect the function of the central nervous system, altering perception, mood or consciousness.

These drugs are divided into different groups like: stimulants, depressants, antidepressants, anxiolytics, antipsychotics,and hallucinogens.

These psychoactive drugs have been proven useful in treating wide range of medical conditions including mental disorders around the world. The most widely used drugs in the world include caffeine, nicotine and alcohol, which are also considered recreational drugs, since they are used for pleasure rather than medicinal purposes. All drugs can have potential side effects. Abuse of several psychoactive drugs can cause addiction and/or physical dependence. Excessive use of stimulants can promote stimulant psychosis. Many recreational drugs are illicit and international treaties such as the Single Convention on Narcotic Drugs exist for the purpose of their prohibition.

5.50 Control of drugs

There are numerous governmental offices in many countries that deal with the control and oversee of drug manufacture and use, and the implementation of various drug laws. The Single Convention on Narcotic Drugs is an international treaty brought about in 1961 to prohibit the use of narcotics save for those used in medical research and treatment. In 1971, a second treaty the Convention on Psychotropic Substances had to be introduced to deal with newer recreational psychoactive and psychedelic drugs. The legal status of Salvia divinorum varies in many countries and even in states within the United States. Where it is legislated against the degree of prohibition also varies.

The Food and Drug Administration (FDA) in the United States is a federal agency responsible for protecting and promoting public health through the regulation and supervision of food safety, tobacco products, dietary supplements, prescription and over-the-counter medications, vaccines, biopharmaceuticals, blood transfusions, medical devices, electromagnetic radiation emitting devices, cosmetics, animal foods and veterinary drugs.

In India, the Narcotics Control Bureau (abbr. **NCB**), an Indian federal law enforcement and intelligence agency under the Ministry of Home Affairs, Government of India is tasked with combating drug trafficking and assisting international use of illegal substances under the provisions of Narcotic Drugs and Psychotropic Substances Act.

5.5.1 Alcohol

An alcoholic drink (also called an alcoholic beverage, adult beverage, or simply a drink) is a drink that contains ethanol, a type of alcohol produced by fermentation of grains, fruits, or other sources of sugar that acts as a drug. The consumption of alcoholic drinks, often referred to as "drinking", plays an important social role in many cultures. Most countries have laws regulating the production, sale, and consumption of alcoholic beverages. Regulations may require the labeling of the percentage alcohol content (as ABV or proof) and the use of a warning label. Some countries ban such activities entirely, but alcoholic drinks are legal in most parts of the world. The global alcoholic drink industry exceeded $1 trillion in 2018.

Alcohol is a depressant, which in low doses causes euphoria, reduces anxiety, and increases sociability. In higher doses, it causes drunkenness, stupor, unconsciousness or death. Long-term use can lead to an alcohol use disorder, an increased risk of developing several types of cancer, and physical dependence.

Alcohol is one of the most widely used recreational drugs in the world, and about 33% of all humans currently drink alcohol. In 2015, among Americans, 86% of adults had consumed alcohol at some point, with 70% drinking it in the last year and 56% in the last month. Alcoholic drinks are typically divided into three classes—beers, wines, and spirits—and typically their alcohol content is between 3% and 50%.

Discovery of late Stone Age jugs suggest that intentionally fermented drinks existed at least as early as the Neolithic period (c. 10,000 BC). Several other animals are affected by alcohol similarly to humans and, once they consume it, will consume it again if given the opportunity, though humans are the only species known to produce alcoholic drinks intentionally.

5.52 Fermented drinks

Beer

Beer is a beverage fermented from grain mash. It is typically made from barley or a blend of several grains and flavored with hops. Most beer is naturally carbonated as part of the fermentation process. If the fermented mash is distilled, then the drink becomes a spirit. Beer is the most consumed alcoholic beverage in the world.

Cider

Cider or cyder is a fermented alcoholic drink made from any fruit juice; apple juice (traditional and most common), peaches, pears ("Perry" cider) or other fruit. Cider

alcohol content varies from 1.2% ABV to 8.5% or more in traditional English ciders. In some regions, cider may be called "apple wine".

Fermented tea

Fermented tea (also known as post-fermented tea or dark tea) is a class of tea that has undergone microbial fermentation, from several months to many years. The tea leaves and the liquor made from them become darker with oxidation. Thus, the various kinds of fermented teas produced across China are also referred to as dark tea, not be confused with black tea. The most famous fermented tea is kombucha which is often homebrewed, pu-erh, produced in Yunnan Province, and the Anhua dark tea produced in Anhua County of Hunan Province. The majority of kombucha on the market are under 0.5% ABV.

Fermented water

Fermented water is an ethanol-based water solution with approximately 15-17% ABV without sweet reserve. Fermented water is *exclusively* fermented with white sugar, yeast, and water. Fermented water is clarified after the fermentation to produce a colorless or off-white liquid with no discernible taste other than that of ethanol.

Fermented sugar water

Fermented sugar water is fermented water with added refined sugar.

Mead

Mead (/miːd/) is an alcoholic drink made by fermenting honey with water, sometimes with various fruits, spices, grains, or hops. The alcoholic content of mead may range from as low as 3% ABV to more than 20%. The defining characteristic of mead is that the majority of the drink's fermentable sugar is derived from honey. Mead can also be referred to as "honeywine."

Pulque

Pulque is the Mesoamerican fermented drink made from the "honey water" of maguey, *Agave americana*. The drink distilled from pulque is tequila or mescal Mezcal.

Rice wine

Sake, huangjiu, mijiu, and cheongju are popular examples of East Asian rice wine.

Wine

Wine is a fermented beverage produced from grapes and sometimes other fruits. Wine involves a longer fermentation process than beer and a long aging process (months or years), resulting in an alcohol content of 9%–16% ABV.

Others

Fruit wines are made from fruits other than grapes, such as plums, cherries, or apples. Sparkling wine like French Champagne, Catalan Cava or Italian Prosecco can be made by means of a secondary fermentation.

Toxicants

Toxins are poisons produced within living cells or organs of plants, animals, and bacteria. Toxicants are synthetic, human-made, toxic chemicals. The difference is not merely one of semantics. Until the late 19th century, nearly all toxic substances were called toxins and they were normally made up of animal and plant positions and naturally occurring minerals such as arsenic. But by the 1930s and 40s, a new term came to describe chemical-biological harm and its relations with living systems. The term "toxicant" replaced "toxin" in descriptions of pesticides in the scientific and engineering literature. Toxicant became the term of art for industrially-manufactured "poisons."

Toxicants are significantly different from toxins not only because of their synthetic origins, but also because of their mass tonnage, wide economic production and distribution processes, compositional heterogeneity, and increasing ubiquity in homes, bodies, and environments. These traits were remarked upon as early as the 1930s (Kallet & Schlink 1933), and have only intensified since that time. We have also come to realize that some toxicants, such as those listed in the Stockholm Convention on Persistent Organic Pollutants (POPs) including DDT, dioxins, and PCBs, are so long-lived that they will outlast the human species. POPs can cause harm for their entire life cycle.

These differences between toxins and toxicants is not a matter of degree, but of kind. They are two different kinds of things. Even minerals that occur underground like arsenic or are produced by plankton such as methyl mercury are now created at unprecedented scales and intensities via mining, hydroelectric dams, and other industrial processes that they are more aligned with industrial scales, processes, and politics than natural ones. The large-scale mercury poisoning in Minamata, Japan, or Grassy Narrows, Canada are impossible for naturally occurring toxins.

Furthermore, toxins and toxicants can cause biochemical harm differently. Toxins like haemotoxins in snakes destroy red blood cells. Necrotoxins like those produced in bacteria destroy cells in tissues. While there are many types of toxins, they tend to work by destroying or disrupting regular cell activity. Toxicants, on the other hand, often make things

work different than they normally would. Carcinogens like asbestos make cells multiply differently and cause cancer. Endocrine disrupting compounds like Bisphenol A interact with the hormone system and can cause early puberty, heart disease, infertility, and obesity. Rather than breaking or destroying bodily processes, many toxicants make then work differently, often to detrimental effect.

5.5.3 Toxic Agents in the Environment

Poison ivy contains a toxin that causes severe reactions in many people, so we tend to steer clear of it. Now think of doing your laundry. You probably use detergent in the wash to get your clothes clean, right? You may not put poison ivy and laundry soap in the same category, but environmental toxicologists do. They study toxicants, which are toxic substances in the environment. Toxicants come in all shapes and sizes, and while they can come from both natural and human-made sources, this lesson will focus on human-made toxicants and their effects on human and environmental health.

5.5.4 Types of Toxicants

As mentioned before, there's a wide variety of toxicants in the environment. To better understand them, we can put them into specific categories that are based on the types of problems they cause. Carcinogens are probably the best-known toxicant because these are cancer-causing chemicals. Cigarette smoke falls into this category as it contains over 4,000 chemicals, many of which cause cancer. Mutagens are mutation-causing chemicals. When organisms are exposed to a mutagen, it literally mutates their DNA, leading to cancer and other disorders. X-rays are well known mutagens. Teratogens are chemicals that cause harm to unborn babies. The name of this toxicant comes from the Greek word teras, which means monster.

These chemicals cause birth defects during development in the womb. Thalidomide was used in the 1950s as a sleeping pill and to prevent nausea during pregnancy, but turned out to be a very harmful teratogen. Even a single dose is powerful enough to cause severe birth defects in children.

Allergens are chemicals that stimulate overactivity in the immune system. When you are exposed to allergens, your body goes into overdrive, triggering an immune response to try and get rid of the allergen. This is why pollen and dust cause symptoms that are similar to being sick. Neurotoxins are chemicals that attack the nervous system. These include heavy metals,

like lead and mercury, as well as pesticides and chemical weapons. Neurotoxins can lead to symptoms like slurred speech, loss of muscle control and even death.

Endocrine disrupters are chemicals that disrupt the endocrine system in organisms and most often come from prescription drugs and chemicals in plastics. The endocrine system is also known as the hormone system, and this part of your body is what regulates growth, development, sexual maturity, brain function and even appetite. Toxicants that disrupt hormone functioning can lead to some serious problems because they so closely resemble real hormones in your body. Reptiles and amphibians are especially sensitive to endocrine disrupters and exposure often leads to feminization of male animals. This may seem like a lot to remember! But if you look closely, you'll see that the name of the toxicant describes the effect it has on organisms, which helps us understand these toxicants better.

5.5.5 Sources of Toxicants

We are surrounded by synthetic chemicals and encounter them countless times on a daily basis. Plastics, household cleaners, solvents, detergents, cosmetics and perfumes are all toxicants. So are antibiotics, prescription drugs, steroids, food additives, preservatives and other things we ingest. Pesticides, herbicides and fertilizers are also toxicants. Though toxicants come from many sources, they tend to move through the environment in certain ways. Toxicants may find their way into aquatic systems as they get carried away by runoff from large areas of land. Because the water systems are smaller than the land that supplied the contaminants, the toxicants tend to get concentrated in the water.

5.5.6 Summary

Under this unit we have discussed nutrition, drugs, diet and toxicants. Nutrition is the study of nutrients in food, how the body uses them, and the relationship between diet, health, and disease. Nutritionists use ideas from molecular biology, biochemistry, and genetics to understand how nutrients affect the human body. Nutrition also focuses on how people can use dietary choices to reduce the risk of disease, what happens if a person has too much or too little of a nutrient, and how allergies work.

Nutrients provide nourishment. Proteins, carbohydrates, fat, vitamins, minerals, fiber, and water are all nutrients. If people do not have the right balance of nutrients in their diet, their risk of developing certain health conditions increases. This article will explain the different nutrients a person needs and why. It will also look at the role of the dietitian and the nutritionist.

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5.57 Terminal questions

Q.1 What do you mean by nutrition. Describe with types.

Answer:----------

Q.7. What are the special feeding methods? Explain it.

Answer:----------

Q.8. Describe the routine hospital diets.

Answer:----------

Q.9. What are the toxicants? Explain it.

Answer:----------

Q.10. What do you mean by drugs? Describe it with examples.

Answer:----------

Q.11. Write a short note on drugs.

Answer:----------

Q.12. Write a short note on alcohols.

Answer:----------

Further readings

1. Biochemistry- Lehninger A.L.
2. Biochemistry –J.H.Weil.
3. Biochemistry fourth edition-David Hames and Nigel Hooper.
4. Textbook of Biochemistry for Undergraduates - Rafi, M.D.
5. Biochemistry and molecular biology- Wilson Walker.

Unit-6

6.1 Introduction

Many disorders of carbohydrate metabolism are characterized by hypoglycaemia and attacks of neuroglycopenia. Hypoglycaemia can also be caused by disorders affecting the use of other fuels, such as those producing fatty acids and ketone bodies which are important alternative sources of energy. Thus when investigating a patient with hypoglycaemia it is necessary to investigate not only pathways that provide glucose directly, but also those which spare glucose utilization and thus provide defence mechanisms when carbohydrate energy sources

become depleted. The defence mechanisms that are activated during fasting to preserve blood glucose are:

- Glycogenolysis—glucose liberation from glycogen degradation
- Gluconeogenesis—glucose production from pyruvate/lactate and from noncarbohydrate sources such as glucogenic amino acids and glycerol
- Fatty acid β-oxidation—catabolism of triglycerides to acetyl-CoA and ketone bodies

Although there is much overlap, the activation of these defence mechanisms during fasting is sequential. The first defence mechanism, glycogenolysis, is exhausted within 8–12 h of fasting. The second and third defence mechanisms provide glucose once glycogen stores have been depleted. In a patient with glycogen storage disease (GSD) where glycogenolysis is blocked, gluconeogenesis and fatty acid oxidation are activated immediately on fasting and can only maintain normoglycaemia for a few hours. In patients with defects affecting gluconeogenesis or fatty acid oxidation, hypoglycaemia does not occur until glycogen stores have been depleted. When more than one pathway is affected, as in GSD I, where neither glycogenolysis nor gluconeogenesis can release glucose into the circulation, patients can be entirely dependent on oral carbohydrate intake to maintain normoglycaemia.

These pathways are also susceptible to hormonal influences. Insulin in particular inhibits all three pathways and stimulates some enzymes of the reverse pathways: glycogen synthesis, glycolysis, and fatty acid synthesis. Therefore hyperinsulinaemia of whatever cause leads to severe hypoglycaemia which is resistant to treatment. Other hormones, such as glucagon, adrenaline, and growth hormone, also activate some enzymes of glucose homoeostasis, though less markedly.

The metabolism of the other monosaccharides, galactose and fructose, is connected with that of glucose. As well as causing hypoglycaemia, inherited defects that affect the metabolism of these sugars lead to the accumulation of toxic metabolites which also contribute to pathology.

Objectives

This is the sixth unit on clinical biochemistry. Under sixth unit we have following objectives. These are as under:

- To know about disorders of carbohydrate metabolism.
- To know about regulation of blood sugar, glycogen storage
- To know about diseases, diabetes mellitus, glucose and galactose tolerance tests

- To discuss about sugar levels in blood.

6.2 Carbohydrates

Carbohydrates are the main source of energy for the body. They are the sugars, starches, and dietary fiber that occur in plant foods and dairy products. Carbohydrates are mainly found in plant foods. They also occur in dairy products in the form of a milk sugar called lactose. Foods high in carbohydrates include bread, pasta, beans, potatoes, rice, and cereals. Carbohydrates play several roles in living organisms, including providing energy. Byproducts of carbohydrates are involved in the immune system, the development of disease, blood clotting, and reproduction. Carbohydrates, also known as saccharides or carbs, provide energy for the body. Each gram of carbohydrates provides 4 calories. The body breaks carbohydrates down into glucose, which is the primary energy source for the brain and muscles. Carbohydrates are one of three macronutrients, which are nutrients that the body needs in larger amounts. The other macronutrients are protein and fats. Proteins provide 4 calories per gram, and fats provide 9 calories per gram.

6.3 Disorders of carbohydrate metabolism

The metabolism of the carbohydrates galactose, fructose, and glucose is intricately linked through interactions between different enzymatic pathways, and disorders that affect these pathways may have symptoms ranging from mild to severe or even life-threatening. Clinical features include various combinations of hypoglycemia (low blood sugar), liver enlargement, and muscle pain. Most of these disorders can be treated, or at least controlled, with specific dietary interventions.

6.4 Galactose and fructose disorders

Galactosemia usually is caused by a defective component of the second major step in the metabolism of the sugar galactose. When galactose is ingested, as in milk, galactose-1-phosphate accumulates. Therefore, the clinical manifestations of galactosemia begin when milk feeding is started. If the feeding is not stopped, infants with the disorder will develop lethargy, jaundice, progressive liver dysfunction, kidney disease, and weight loss. They are also susceptible to severe bacterial infections, especially by *Escherichia coli*. Cataracts develop if the diet remains galactose-rich. Intellectual disability occurs in most infants with galactosemia if the disorder is left untreated or if treatment is delayed. Therapy is by exclusion of galactose from the diet and results in the reversal of most symptoms. Most children have normal intelligence, although they may have learning difficulties and a degree of intellectual disability despite early therapy.

Hereditary fructose intolerance (HFI) is caused by a deficiency of the liver enzyme fructose-1-phosphate aldolase. Symptoms of HFI appear after the ingestion of fructose and thus present later in life than do those of galactosemia. Fructose is present in fruits, table sugar (sucrose), and infant formulas containing sucrose. Symptoms may include failure to gain weight satisfactorily, vomiting, hypoglycemia, liver dysfunction, and kidney defects. Older children with HFI tend to avoid sweet foods and may have teeth notable for the absence of caries. Children with the disorder do very well if they avoid dietary fructose and sucrose.

Fructose 1,6-diphosphatase deficiency is associated with an impaired ability to form glucose from other substrates (a process called gluconeogenesis). Symptoms include severe hypoglycemia, intolerance to fasting, and enlargement of the liver. Rapid treatment of hypoglycemic episodes with intravenous fluids containing glucose and the avoidance of fasting are the mainstays of therapy. Some patients require continuous overnight drip feeds or a bedtime dose of cornstarch in order to control their tendency to develop hypoglycemia.

6.5 Glycogen storage disorders

The brain, red blood cells, and inner portion of the adrenal gland (adrenal medulla) depend on a constant supply of glucose for their metabolic functions. This supply begins in the small intestine, where transport proteins mediate the uptake of glucose into cells lining the gut. Glucose subsequently passes into the bloodstream and then the liver, where it is stored as glycogen. In times of starvation or fasting or when the body requires a sudden energy supply, glycogen is broken down into glucose, which is then released into the blood.

Muscle tissue also has its own glycogen stores, which may be degraded during exercise. If enzymes responsible for glycogen degradation are blocked so that glycogen remains in the liver or muscle, a number of conditions known as glycogen storage disorders (GSD) can arise. Depending upon which enzyme is affected, these conditions may affect the liver, muscles, or both. In GSD type I (von Gierke disease), the last step in glucose release from the liver is defective, leading to hypoglycemia. Therapy consists of supplying continuous glucose to the digestive tract (e.g., by continuous drip feedings) during infancy and early childhood. As the child grows, an improvement in symptoms tends to occur.

Adequate glucose is supplied by frequent feedings of carbohydrates and slow-release glucose (uncooked cornstarch) before bedtime. Liver transplantation may also be curative, but this drastic measure is reserved for the small percentage of patients who do not respond to the usual treatment or who develop liver cancer. For the muscular forms of the disease, avoidance

of strenuous exercise is the usual therapy. Defects in earlier steps in glycogen breakdown in the liver cause GSD types III, IV, VI, and IX, which usually lead to milder versions of type I disease. Pompe disease (GSD type II) is discussed in the section Lysosomal storage disorders.

In addition to glycogen degradation, glucose may be manufactured from amino acids and pyruvate in the process of gluconeogenesis. Key enzymes in the gluconeogenic pathway include carboxylase, phosphoenolpyruvate carboxykinase, and fructose-1,6-diphosphatase. Persons with defects in these enzymes develop conditions including fasting hypoglycemia, lactic acidemia, and liver enlargement. Thus, gluconeogenesis disorders may be difficult to distinguish from glycogen storage disorders at first presentation.

6.6 Congenital disorders of glycosylation

Congenital disorders of glycosylation (CDG; formerly known as carbohydrate-deficient glycoprotein syndrome) are recently described diseases that affect the brain and many other organs. The primary biochemical defects of CDG are in the N-glycosylation pathway that occurs in the cytoplasm and endoplasmic reticulum, cellular organelles involved in the synthesis of proteins and lipids. A defect in a mannose-processing enzyme, phosphomannomutase 2, causes the most common form of CDG (type I). Other enzymatic defects have been identified, but the biochemical bases of some CDG subtypes have not yet been determined.

The classic form of CDG is characterized by low muscle tone in infancy, severe developmental delay, and brain abnormalities. Children with type Ia also have inverted nipples and an unusual distribution of fat, especially in the suprapubic region and buttocks. Other features include hypoglycemia, seizures, stroke-like episodes, retinal damage, impaired heart contractility, vomiting, liver disease, diarrhea, and a bleeding tendency. No effective therapy exists for CDG, except for the rare type Ib disease (phosphomannose isomerase deficiency), in which oral administration of mannose may reverse symptoms in some cases.

Blood sugar regulation is the process by which the levels of blood sugar, primarily glucose, are maintained by the body within a narrow range. This tight regulation is referred to as glucose homeostasis. Insulin, which lowers blood sugar, and glucagon, which raises it, are the most well known of the hormones involved, but more recent discoveries of other glucoregulatory hormones have expanded the understanding of this process. The gland

called pancreas secrete two hormones and they are primarily responsible to regulate glucose levels in blood.

6.7 Mechanisms

Blood sugar levels are regulated by negative feedback in order to keep the body in balance. The levels of glucose in the blood are monitored by many tissues, but the cells in the pancreatic islets are among the most well understood and important. Granule docking is an important glucose-dependent step in human insulin secretion that does not work properly in type 2 diabetes.

6.8 Glucagon

If the blood glucose level falls to dangerously low levels (as during very heavy exercise or lack of food for extended periods), the alpha cells of the pancreas release glucagon, a hormone which travels through the blood to the liver, where it binds to glucagon receptors on the surface of liver cells and stimulates them to break down glycogen stored inside the cells into glucose (this process is called glycogenolysis). The cells release the glucose into the bloodstream, increasing blood sugar levels. Hypoglycemia, the state of having low blood sugar, is treated by restoring the blood glucose level to normal by the ingestion or administration of dextrose or carbohydrate foods. It is often self-diagnosed and self-medicated orally by the ingestion of balanced meals. In more severe circumstances, it is treated by injection or infusion of glucagon.

6.9 Insulin

When levels of blood sugar rise, whether as a result of glycogen conversion, or from digestion of a meal, a different hormone is released from beta cells found in the islets of Langerhans in the pancreas. This hormone, insulin, causes the liver to convert more glucose into glycogen (this process is called glycogenesis), and to force about 2/3 of body cells (primarily muscle and fat tissue cells) to take up glucose from the blood through the GLUT4 transporter, thus decreasing blood sugar. When insulin binds to the receptors on the cell surface, vesicles containing the GLUT4 transporters come to the plasma membrane and fuse together by the process of endocytosis, thus enabling a facilitated diffusion of glucose into the cell. As soon as the glucose enters the cell, it is phosphorylated into glucose-6-phosphate in order to preserve the concentration gradient so glucose will continue to enter the cell.

Insulin also provides signals to several other body systems, and is the chief regulator of metabolic control in humans. There are also several other causes for an increase in blood

sugar levels. Among them are the 'stress' hormones such as epinephrine (also known as adrenaline), several of the steroids, infections, trauma, and of course, the ingestion of food. Diabetes mellitus type 1 is caused by insufficient or non-existent production of insulin, while type 2 is primarily due to a decreased response to insulin in the tissues of the body (insulin resistance). Both types of diabetes, if untreated, result in too much glucose remaining in the blood (hyperglycemia) and many of the same complications. Also, too much insulin and/or exercise without enough corresponding food intake in diabetics can result in low blood sugar (hypoglycemia).

6.10 Glycogen storage disease (GSD)

What is glycogen storage disease in children?

Glycogen storage disease (GSD) is a rare condition that changes the way the body uses and stores glycogen, a form of sugar or glucose. Glycogen is a main source of energy for the body. Glycogen is stored in the liver. When the body needs more energy, certain proteins called enzymes break down glycogen into glucose. They send the glucose out into the body. When someone has GSD, they are missing one of the enzymes that breaks down glycogen. When an enzyme is missing, glycogen can build up in the liver. Or glycogen may not form properly. This can cause problems in the liver or muscles, or other parts of the body. GSD is passed down from parents to children (is hereditary). It is most often seen in babies or young children. But some forms of GSD may appear in adults.

6.11 Types of GSD

Types of GSD are grouped by the enzyme that is missing in each one. Each GSD has its own symptoms and needs different treatment. There are several types of GSD, but the most common types are types I, III, and IV. These types are also known by other names:

- **Type I or von Gierke disease.** This is the most common form of GSD. People with type I don’t have the enzyme needed to turn glycogen into glucose in the liver. Glycogen builds up in the liver. Symptoms often appear in babies around 3 to 4 months old. They may include low blood sugar (hypoglycemia) and a swollen belly because of an enlarged liver.
- **Type III, Cori disease, or Forbes disease.** People with type III don’t have enough of an enzyme called the debranching enzyme, which helps break down glycogen. The glycogen can’t fully break down. It collects in the liver and in muscle tissues. Symptoms include a swollen belly, delayed growth, and weak muscles.

- **Type IV or Andersen disease.** People with type IV form abnormal glycogen. Experts think the abnormal glycogen triggers the body's infection-fighting system (immune system). This creates scarring (cirrhosis) of the liver and other organs such as muscle and the heart.

6.12 What causes glycogen storage disease in a child?

Glycogen storage disease is passed down from parents to children (hereditary). It happens because both parents have an abnormal gene (gene mutation) that affects a specific way that glycogen is stored or used. Most GSDs occur because both parents pass on the same abnormal gene to their children. In most cases parents don't show any symptoms of the disease.

Which children are at risk for glycogen storage disease?

Glycogen storage disease is passed down from parents to children (inherited). Someone is more at risk for GSD if they have a family member with the disease.

What are the symptoms of glycogen storage disease in a child?

With many types of GSD, symptoms first appear in babies or in very young children. Symptoms will vary based on the type of GSD a child has, and on which enzyme he or she is missing. Because GSD most often affects the muscles and the liver, those areas show the most symptoms. General symptoms of GSD may include:

- Not growing fast enough
- Not feeling comfortable in hot weather (heat intolerance)
- Bruising too easily
- Low blood sugar (hypoglycemia)
- An enlarged liver
- A swollen belly
- Weak muscles (low muscle tone)
- Muscle pain and cramping during exercise

Symptoms for babies may include:

- Too much acid in the blood (acidosis)
- High blood cholesterol levels (hyperlipidemia)

The symptoms of GSD may look like other health problems. Always see your child's healthcare provider to be sure. Some types of GSD can appear in adults. See your healthcare provider if you think you may have GSD.

6.13 How is glycogen storage disease diagnosed in a child?

Your child's healthcare provider will ask about your child's symptoms and past health. The provider will do a physical exam to check for symptoms such as an enlarged liver or weak muscles. Your child's provider may do a few blood tests. He or she may also take a small tissue sample (biopsy) of your child's liver or muscle. The sample will be taken to a lab. It will be tested to see how much of a certain enzyme is in that part of the body. If you are pregnant and concerned about GSD, your healthcare provider may do some tests before your baby is born (prenatal tests) to check for GSD.

6.14 How is glycogen storage disease treated in a child?

Treatment will vary depending on what type of GSD your child has. For types I, III, and IV, your child's healthcare provider may suggest a special diet to help control symptoms. Your child may also have to take certain medicines. For other types of GSD, your child may need to limit exercise to avoid muscle cramps. He or she may need to have a medical treatment to replace the enzyme that is missing (enzyme replacement therapy).

6.15 What are possible complications of glycogen storage disease in a child?

Glycogen buildup can hurt the liver and muscles. This can create other problems if your child has certain types of GSD such as:

- **Type III.** This can cause harmless (benign) tumors in the liver.
- **Type IV.** Over time this can cause scarring (cirrhosis) of the liver. This disease leads to liver failure.

6.16 What can I do to prevent glycogen storage disease in my child?

There is no way to prevent glycogen storage disease. But early treatment can help control symptoms once a child has GSD. If you or your partner have GSD, or a family history of this disease, see a genetic counselor before you get pregnant. He or she can find out your chances of having a child with GSD.

6.17 How can I help my child live with glycogen storage disease?

A child with GSD may have special needs. Be sure that your child gets regular medical care. It is important that his or her healthcare provider checks your child's condition. Regular medical visits will also help you keep up with new treatment options. Online or in-person support groups may also be helpful for you and your family.

6.18 When should I call my child's healthcare provider?

Many forms of glycogen storage disease appear in babies and young children. Call your healthcare provider if your baby's behavior changes after you stop night feedings. Talk with your healthcare provider if your child:

- Is not growing fast enough
- Has constant (chronic) hunger
- Has a swollen belly

Teens and adults should watch for the following symptoms when they exercise:

- Muscle weakness
- Pain
- Cramps

Key points about glycogen storage disease in children

- Glycogen storage disease (GSD) is a rare condition that changes the way the body uses and stores glycogen, a form of sugar.
- It is passed down from parents to children (inherited). For most GSDs, each parent must pass on one abnormal copy of the same gene.
- Most parents do not show any signs of GSD.
- There are several types of GSD, but types I, III, and IV are most common. Each GSD has its own symptoms and needs different treatment.
- Symptoms often first appear in babies or young children. In some cases GSD can appear in adults.

Metabolism is the process your body uses to make energy from the food you eat. Food is made up of proteins, carbohydrates, and fats. Chemicals in your digestive system (enzymes) break the food parts down into sugars and acids, your body's fuel. Your body can use this fuel right away, or it can store the energy in your body tissues. If you have a metabolic disorder, something goes wrong with this process. Carbohydrate metabolism disorders are a group of metabolic disorders. Normally your enzymes break carbohydrates down into glucose (a type of sugar). If you have one of these disorders, you may not have enough enzymes to break down the carbohydrates. Or the enzymes may not work properly. This causes a harmful amount of sugar to build up in your body. That can lead to health problems, some of which can be serious. Some of the disorders are fatal.

These disorders are inherited. Newborn babies get screened for many of them, using blood tests. If there is a family history of one of these disorders, parents can get genetic testing to see whether they carry the gene. Other genetic tests can tell whether the fetus has the disorder or carries the gene for the disorder. Diabetes mellitus refers to a group of diseases that affect how your body uses blood sugar (glucose). Glucose is vital to your health because it's an important source of energy for the cells that make up your muscles and tissues. It's also your brain's main source of fuel. The underlying cause of diabetes varies by type. But, no matter what type of diabetes you have, it can lead to excess sugar in your blood. Too much sugar in your blood can lead to serious health problems.

Chronic diabetes conditions include type 1 diabetes and type 2 diabetes. Potentially reversible diabetes conditions include prediabetes and gestational diabetes. Prediabetes occurs when your blood sugar levels are higher than normal, but not high enough to be classified as diabetes. And prediabetes is often the precursor of diabetes unless appropriate measures are taken to prevent progression. Gestational diabetes occurs during pregnancy but may resolve after the baby is delivered.

6.19 What is diabetes?

Diabetes happens when your body isn't able to take up sugar (glucose) into its cells and use it for energy. This results in a build up of extra sugar in your bloodstream. Poorly controlled diabetes can lead to serious consequences, causing damage to a wide range of your body's organs and tissues – including your heart, kidneys, eyes and nerves.

6.20 Why is my blood glucose level high? How does this happen?

The process of digestion includes breaking down the food you eat into various different nutrient sources. When you eat carbohydrates (for example, bread, rice, pasta), your body breaks this down into sugar (glucose). When glucose is in your bloodstream, it needs help – a "key" – to get into its final destination where it's used, which is inside your body's cells (cells make up your body's tissues and organs). This help or "key" is insulin.

Insulin is a hormone made by your pancreas, an organ located behind your stomach. Your pancreas releases insulin into your bloodstream. Insulin acts as the “key” that unlocks the cell wall “door,” which allows glucose to enter your body’s cells. Glucose provides the “fuel” or energy tissues and organs need to properly function. If you have diabetes:

- Your pancreas doesn’t make any insulin or enough insulin.

Or

- Your pancreas makes insulin but your body's cells don't respond to it and can't use it as it normally should.

If glucose can't get into your body's cells, it stays in your bloodstream and your blood glucose level rises.

6.21 What are the different types of diabetes?

The types of diabetes are:

- **Type 1 diabetes:** This type is an autoimmune disease, meaning your body attacks itself. In this case, the insulin-producing cells in your pancreas are destroyed. Up to 10% of people who have diabetes have Type 1. It's usually diagnosed in children and young adults (but can develop at any age). It was once better known as "juvenile" diabetes. People with Type 1 diabetes need to take insulin every day. This is why it is also called insulin-dependent diabetes.
- **Type 2 diabetes:** With this type, your body either doesn't make enough insulin or your body's cells don't respond normally to the insulin. This is the most common type of diabetes. Up to 95% of people with diabetes have Type 2. It usually occurs in middle-aged and older people. Other common names for Type 2 include adult-onset diabetes and insulin-resistant diabetes. Your parents or grandparents may have called it "having a touch of sugar."
- **Prediabetes:** This type is the stage before Type 2 diabetes. Your blood glucose levels are higher than normal but not high enough to be officially diagnosed with Type 2 diabetes.
- **Gestational diabetes:** This type develops in some women during their pregnancy. Gestational diabetes usually goes away after pregnancy. However, if you have gestational diabetes you're at higher risk of developing Type 2 diabetes later on in life.

Less common types of diabetes include:

- **Monogenic diabetes syndromes:** These are rare inherited forms of diabetes accounting for up to 4% of all cases. Examples are neonatal diabetes and maturity-onset diabetes of the young.
- **Cystic fibrosis-related diabetes:** This is a form of diabetes specific to people with this disease.
- **Drug or chemical-induced diabetes:** Examples of this type happen after organ transplant, following HIV/AIDS treatment or are associated with glucocorticoid steroid use.

Diabetes insipidus is a distinct rare condition that causes your kidneys to produce a large amount of urine.

How common is diabetes?

Some 34.2 million people of all ages – about 1 in 10 – have diabetes in the U.S. Some 7.3 million adults aged 18 and older (about 1 in 5) are unaware that they have diabetes (just under 3% of all U.S. adults). The number of people who are diagnosed with diabetes increases with age. More than 26% of adults age 65 and older (about 1 in 4) have diabetes.

Who gets diabetes? What are the risk factors?

Factors that increase your risk differ depending on the type of diabetes you ultimately develop.

Risk factors for Type 1 diabetes include:

- Having a family history (parent or sibling) of Type 1 diabetes.
- Injury to the pancreas (such as by infection, tumor, surgery or accident).
- Presence of autoantibodies (antibodies that mistakenly attack your own body's tissues or organs).
- Physical stress (such as surgery or illness).
- Exposure to illnesses caused by viruses.

Risk factors for prediabetes and Type 2 diabetes include:

- Family history (parent or sibling) of prediabetes or Type 2 diabetes.
- Being African-American, Hispanic, Native American, Asian-American race or Pacific Islander.
- Being overweight.
- Having high blood pressure.
- Having low HDL cholesterol (the "good" cholesterol) and high triglyceride level.
- Being physically inactive.
- Being age 45 or older.
- Having gestational diabetes or giving birth to a baby weighing more than 9 pounds.
- Having polycystic ovary syndrome.
- Having a history of heart disease or stroke.
- Being a smoker.

Risk factors for gestational diabetes include:

- Family history (parent or sibling) of prediabetes or Type 2 diabetes.
- Being African-American, Hispanic, Native American or Asian-American.

- Being overweight before your pregnancy.
- Being over 25 years of age.

6.22 Symptoms and causes?

What causes diabetes?

The cause of diabetes, regardless of the type, is having too much glucose circulating in your bloodstream. However, the reason why your blood glucose levels are high differs depending on the type of diabetes.

- **Causes of Type 1 diabetes:** This is an immune system disease. Your body attacks and destroys insulin-producing cells in your pancreas. Without insulin to allow glucose to enter your cells, glucose builds up in your bloodstream. Genes may also play a role in some patients. Also, a virus may trigger the immune system attack.
- **Cause of Type 2 diabetes and prediabetes:** Your body's cells don't allow insulin to work as it should to let glucose into its cells. Your body's cells have become resistant to insulin. Your pancreas can't keep up and make enough insulin to overcome this resistance. Glucose levels rise in your bloodstream.
- **Gestational diabetes:** Hormones produced by the placenta during your pregnancy make your body's cells more resistant to insulin. Your pancreas can't make enough insulin to overcome this resistance. Too much glucose remains in your bloodstream.

6.23 What are the symptoms of diabetes?

Symptoms of diabetes include:

- Increased thirst.
- Weak, tired feeling.
- Blurred vision.
- Numbness or tingling in the hands or feet.
- Slow-healing sores or cuts.
- Unplanned weight loss.
- Frequent urination.
- Frequent unexplained infections.
- Dry mouth.

Other symptoms

- In women: Dry and itchy skin, and frequent yeast infections or urinary tract infections.
- In men: Decreased sex drive, erectile dysfunction, decreased muscle strength.

Type 1 diabetes symptoms: Symptoms can develop quickly – over a few weeks or months. Symptoms begin when you're young – as a child, teen or young adult. Additional symptoms include nausea, vomiting or stomach pains and yeast infections or urinary tract infections.

Type 2 diabetes and prediabetes symptoms: You may not have any symptoms at all or may not notice them since they develop slowly over several years. Symptoms usually begin to develop when you're an adult, but prediabetes and Type 2 diabetes is on the rise in all age groups.

Gestational diabetes: You typically will not notice symptoms. Your obstetrician will test you for gestational diabetes between 24 and 28 weeks of your pregnancy.

What are the complications of diabetes?

If your blood glucose level remains high over a long period of time, your body's tissues and organs can be seriously damaged. Some complications can be life-threatening over time. Complications include:

- Cardiovascular issues including coronary artery disease, chest pain, heart attack, stroke, high blood pressure, high cholesterol, atherosclerosis (narrowing of the arteries).
- Nerve damage (neuropathy) that causes numbing and tingling that starts at toes or fingers then spreads.
- Kidney damage (nephropathy) that can lead to kidney failure or the need for dialysis or transplant.
- Eye damage (retinopathy) that can lead to blindness; cataracts, glaucoma.
- Foot damage including nerve damage, poor blood flow and poor healing of cuts and sores.
- Skin infections.
- Erectile dysfunction.
- Hearing loss.
- Depression.
- Dementia.
- Dental problems.

6.24 Complications of gestational diabetes:

In the mother: Preeclampsia (high blood pressure, excess protein in urine, leg/feet swelling), risk of gestational diabetes during future pregnancies and risk of diabetes later in life.

In the newborn: Higher-than-normal birth weight, low blood sugar (hypoglycemia), higher risk of developing Type 2 diabetes over time and death shortly after birth.

6.25 Glucose tolerance test

The glucose tolerance test is a medical test in which glucose is given and blood samples taken afterward to determine how quickly it is cleared from the blood. The test is usually used to test for diabetes, insulin resistance, impaired beta cell function, and sometimes reactive hypoglycemia and acromegaly, or rarer disorders of carbohydrate metabolism. In the most commonly performed version of the test, an *oral glucose tolerance test* (OGTT), a standard dose of glucose is ingested by mouth and blood levels are checked two hours later. Many variations of the GTT have been devised over the years for various purposes, with different standard doses of glucose, different routes of administration, different intervals and durations of sampling, and various substances measured in addition to blood glucose.

6.26 History

The glucose tolerance test was first described in 1923 by Jerome W. Conn. The test was based on the previous work in 1913 by A. T. B. Jacobson in determining that carbohydrate ingestion results in blood glucose fluctuations, and the premise (named the Staub-Traugott Phenomenon after its first observers that a normal patient fed glucose will rapidly return to normal levels of blood glucose after an initial spike, and will see improved reaction to subsequent glucose feedings.

6.27 Testing

Since the 1970s, the World Health Organization and other organizations interested in diabetes agreed on a standard dose and duration.

Preparation

The patient is instructed not to restrict carbohydrate intake in the days or weeks before the test. The test should not be done during an illness, as results may not reflect the patient's glucose metabolism when healthy. A full adult dose should not be given to a person weighing less than 42.6 kg (94 lb), or the excessive glucose may produce a false positive result. Usually the OGTT is performed in the morning as glucose tolerance can exhibit a diurnal rhythm with a significant decrease in the afternoon. The patient is instructed to fast (water is allowed) for 8–12 hours prior to the tests. Medication such as large doses of salicylates, diuretics, anticonvulsants, and oral contraceptives affect the glucose tolerance test.

Procedure

1. A zero time (baseline) blood sample is drawn.
2. The patient is then given a measured dose (below) of glucose solution to drink within a 5-minute time frame.
3. Blood is drawn at intervals for measurement of glucose (blood sugar), and sometimes insulin levels. The intervals and number of samples vary according to the purpose of the test. For simple diabetes screening, the most important sample is the 2 hour sample and the 0 and 2 hour samples may be the only ones collected. A laboratory may continue to collect blood for up to 6 hours depending on the protocol requested by the physician.

Dose of glucose and variations

- 75 g of oral dose is the recommendation of the WHO to be used in all adults, and is the main dosage used in the United States. The dose is adjusted for weight only in children. The dose should be drunk within 5 minutes.
- A variant is often used in pregnancy to screen for gestational diabetes, with a screening test of 50 g over one hour. If elevated, this is followed with a test of 100 g over three hours.
- In UK general practice, the standard glucose load was provided by 394 ml of the energy drink Lucozade with original carbonated flavour, but this is being superseded by purpose-made drinks.

6.28 Substances measured and variations

If renal glycosuria (sugar excreted in the urine despite normal levels in the blood) is suspected, urine samples may also be collected for testing along with the fasting and 2 hour blood tests.

Galactose

Galactose sometimes abbreviated **Gal**, is a monosaccharide sugar that is about as sweet as glucose, and about 65% as sweet as sucrose. It is an aldohexose and a C-4 epimer of glucose. A galactose molecule linked with a glucose molecule forms a lactose molecule. Galactan is a polymeric form of galactose found in hemicellulose, and forming the core of the galactans, a class of natural polymeric carbohydrates.

Etymology

The word *galactose* was coined by Charles Weissman in the mid-19th century and is derived from Greek *galaktos* (of milk) and the generic chemical suffix for sugars -*ose*. The etymology

is comparable to that of the word *lactose* in that both contain roots meaning "milk sugar". Lactose is a disaccharide of galactose plus glucose.

6.29 Structure and isomerism

Galactose exists in both open-chain and cyclic form. The open-chain form has a carbonyl at the end of the chain. Four isomers are cyclic, two of them with a pyranose (six-membered) ring, two with a furanose (five-membered) ring. Galactofuranose occurs in bacteria, fungi and protozoa, and is recognized by a putative chordate immune lectin intelectin through its exocyclic 1, 2-diol. In the cyclic form there are two anomers, named alpha and beta, since the transition from the open-chain form to the cyclic form involves the creation of a new stereocenter at the site of the open-chain carbonyl. The IR spectra for galactose shows a broad, strong stretch from roughly wavenumber 2500 cm^{-1} to wavenumber 3700 cm^{-1}.

lipid. Many speculate that it is for this reason that a pathway for rapid conversion from galactose to glucose has been highly conserved among many species. The main pathway of galactose metabolism is the Leloir pathway; humans and other species, however, have been noted to contain several alternate pathways, such as the De Ley Doudoroff Pathway. The Leloir pathway consists of the latter stage of a two-part process that converts β-D-galactose to UDP-glucose.

The initial stage is the conversion of β-D-galactose to α-D-galactose by the enzyme, mutarotase (GALM). The Leloir pathway then carries out the conversion of α-D-galactose to UDP-glucose via three principal enzymes: Galactokinase (GALK) phosphorylates α-D-galactose to galactose-1-phosphate, or Gal-1-P; Galactose-1-phosphate uridyltransferase (GALT) transfers a UMP group from UDP-glucose to Gal-1-P to form UDP-galactose; and finally, UDP galactose-4'-epimerase (GALE) interconverts UDP-galactose and UDP-glucose, thereby completing the pathway.

The above mechanisms for galactose metabolism are necessary because the human body cannot directly convert galactose into energy, and must first go through one of these processes in order to utilize the sugar. Galactosemia is an inability to properly break down galactose due to a genetically inherited mutation in one of the enzymes in the Leloir pathway. As a result, the consumption of even small quantities is harmful to galactosemics.

6.29 Sources

Galactose is found in dairy products, avocados, sugar beets, other gums and mucilages. It is also synthesized by the body, where it forms part of glycolipids and glycoproteins in several tissues; and is a by-product from the third-generation ethanol production process (from macroalgae).

6.30 Clinical significance

Chronic systemic exposure of mice, rats, and *Drosophila* to D-galactose causes the acceleration of senescence (aging). It has been reported that high dose exposure of D-galactose (120 mg/kg) can cause reduced sperm concentration and sperm motility in rodent and has been extensively used as an aging model when administered subcutaneous. Two studies have suggested a possible link between galactose in milk and ovarian cancer. Other studies show no correlation, even in the presence of defective galactose metabolism. More recently, pooled analysis done by the Harvard School of Public Health showed no specific correlation between lactose-containing foods and ovarian cancer, and showed statistically insignificant increases in risk for consumption of lactose at 30 g/day.

Some ongoing studies suggest galactose may have a role in treatment of focal segmental glomerulosclerosis (a kidney disease resulting in kidney failure and proteinuria). This effect is likely to be a result of binding of galactose to FSGS factor. Galactose is a component of the antigens present on blood cells that determine blood type within the ABO blood group system. In O and A antigens, there are two monomers of galactose on the antigens, whereas in the B antigens there are three monomers of galactose.

A disaccharide composed of two units of galactose, galactose-alpha-1,3-galactose (alpha-gal), has been recognized as a potential allergen present in mammal meat. Alpha-gal allergy may be triggered by lone star tick bites. Galactose in sodium saccharin solution has also been found to cause conditioned flavor avoidance in adult female rats within a laboratory setting when combined with intragastric injections. The reason for this flavor avoidance is still unknown, however it is possible that a decrease in the levels of the enzymes required to convert galactose to glucose in the liver of the rats could be responsible.

6.31 Sugar levels in blood

Blood sugar, or glucose, is the main sugar found in your blood. It comes from the food you eat, and is your body's main source of energy. Your blood carries glucose to all of your body's cells to use for energy. Diabetes is a disease in which your blood sugar levels are too high. Over time, having too much glucose in your blood can cause serious problems. Even if you don't have diabetes, sometimes you may have problems with blood sugar that is too low or

too high. Keeping a regular schedule of eating, activity, and taking any medicines you need can help. If you do have diabetes, it is very important to keep your blood sugar numbers in your target range. You may need to check your blood sugar several times each day. Your health care provider will also do a blood test called an A1C. It checks your average blood sugar level over the past three months. If your blood sugar is too high, you may need to take medicines and/or follow a special diet.

6.32 Summary

Under this unit we summarize that **Carbohydrate metabolism** is a fundamental biochemical process that ensures a constant supply of energy to living cells. The most important carbohydrate is glucose, which can be broken down via glycolysis, enter into the Kreb's cycle and oxidative phosphorylation to generate ATP. Further important pathways in carbohydrate metabolism include the pentose phosphate pathway (conversion of hexose sugars into pentoses), glycogenesis (conversion of excess glucose into glycogen, stimulated by insulin), glycogenolysis (conversion of glycogen polymers into glucose, stimulated by glucagon) and gluconeogenesis (*de novo* glucose synthesis).

There are multiple diseases that arise from improper carbohydrate metabolism. Diabetes mellitus is caused by a lack of, or a resistance to, insulin leading to hypo- or hyperglycemia. Lactose intolerance is a common allergy in adults and results from a lack of the enzyme lactase, which converts lactose disaccharides (found in dairy products) into glucose monosaccharides. Much rarer diseases such as galactosemia and von Gierke's diseases are caused by congenital mutations in enzymes involved in glucose metabolic pathways.

Regulation of glucose in the body is done autonomically and constantly throughout each minute of the day. Normal BG levels should be between 60 and 140 mg/dL in order to supply cells of the body with its required energy. Brain cells don’t require insulin to drive glucose into neurons; however, there must still be normal amounts available. Too little glucose, called **hypoglycemia**, starves cells, and too much glucose (**hyperglycemia**) creates a sticky, paralyzing effect on cells. Euglycemia, or blood sugar within the normal range, is naturally ideal for the body’s functions.

6.33 Terminal questions

Q.13. What do you mean by disorder of carbohydrate metabolism? Describe it.

Answer:----------

Q.14. Describe the regulation of blood sugar.

Answer:----------

Q.16 What is glycogen storage disease in children?

Answer:----------

Q.15. Write a short note on diabetes mellitus.

Answer:----------

Q.16. Explain sugar levels in blood.

Answer:----------

Further readings

6. Biochemistry- Lehninger A.L.
7. Biochemistry –J.H.Weil.
8. Biochemistry fourth edition-David Hames and Nigel Hooper.
9. Textbook of Biochemistry for Undergraduates - Rafi, M.D.
10. Biochemistry and molecular biology- Wilson Walker.

Unit-7

7.1 Introduction

Lipid disorders are highly prevalent in both treated and untreated HIV infection. The introduction of cART has resulted in improvement in long-term survival and in individuals with HIV infection now facing the same causes of major morbidity and mortality as the uninfected population; this increases the importance of appropriate intervention for lipid disorders and cardiovascular risk reduction.

Lipid disorders in untreated HIV infection are well described. In advanced HIV infection, increased levels of serum triglycerides and reduced levels of total low-density lipoprotein (LDL) and high-density lipoprotein (HDL) are seen. Hypertriglyceridemia is also associated with greater immune system activation. Additionally, lipoprotein particles may be more atherogenic, with a higher number of small, dense LDL particles that are able to access the blood vessel wall.

In addition, some antiretroviral medications may also cause lipid abnormalities, and each medication may have its own unique effect, making it difficult to assign a "class effect" to any particular drug class. Hypertriglyceridemia is common with certain PIs, in particular those regimens where ritonavir is used to boost other PIs such as tipranavir, lopinavir, and fosamprenavir Combinations of these PIs with ritonavir are known to have the greatest effect on lipids. Saquinavir–ritonavir and indinavir–ritonavir as well as nelfinavir (a PI that is used without ritonavir have more of an intermediate effect. Newer PIs, including atazanavir and darunavir–ritonavir, appear to have little impact on serum lipids.

Certain NRTIs have also been associated with lipid disorders. Regimens containing stavudine result in higher serum lipid levels compared with those without it. Tenofovir has fewer lipid effects compared with stavudine and may be considered preferable to other NRTIs in those with lipid disorders. The use of non-nucleoside reverse transcriptase inhibitors (NNRTIs) are generally not associated with a more artherogenic lipid profile and have not been associated with an increased risk of myocardial infarction.

Objectives

This is the seventh unit on clinical biochemistry. Under seventh unit we have following objectives. These are as under:

- To know about lipids and disorders of lipids
- To know about low and high density lipoproteins
- To know about cholesterol, triglycerides
- To discuss about gaucher's and tay-sach's disease

7.2 Disorder in lipids

The term “lipid disorder” covers a range of conditions that can cause abnormal levels of lipids, or fats, in the blood. These fats include low-density lipoproteins (LDLs), also known as “bad” cholesterol, and fatty acids called triglycerides. High-density lipoproteins (HDLs), known as “good” cholesterol, are also present in the blood. This unit will look at the symptoms of lipid disorders, their causes, and what treatments are available, including lifestyle changes people can make to prevent or reduce high cholesterol.

7.3 What is a lipid disorder?

A lipid disorder will typically increase levels of LDLs, triglycerides, or both. HDLs are known as good cholesterol because they help to transport bad cholesterol out of the body. A buildup of LDLs and triglycerides can cause fatty materials to accumulate in the body’s tissues, including in the arteries. This can have serious consequences for cardiovascular health and increase the risk of problems such as heart disease.

7.4 Symptoms of lipid disorder

It is important to point out that, most of the time, a person will have no symptoms of a lipid disorder until they experience a significant health problem, such as a stroke or heart attack. However, the following symptoms have been occasionally observed in some people with very high lipid levels:

- Yellowish, fatty bumps or yellow creases on the skin, formed by an accumulation of fatty deposits around tendons and joints (xanthomas)
- White arcs around the cornea of the eye (arcus senilis), which sometimes occur in younger people with high cholesterol
- Raised, yellow lumps at the inner corners of the eyes (xanthelasma)

7.5 Causes of lipid disorder

A range of factors can cause lipid disorders, including genetics, lifestyle, and other underlying health conditions.

7.6 Genetic factors

Many lipid disorders are inherited. Familial hypercholesterolemia (FH) for example, is when high cholesterol runs in a family. FH affects an estimated 1 in 200–500 people worldwide but is more common in people of French Canadian, Ashkenazi Jewish, Lebanese, or Afrikaner (a South African ethnic group) descent. Knowing their family history of cholesterol and heart disease may help people decide if they want to get a blood test to check if they have high cholesterol.

7.7 Diet and lifestyle factors

Foods that are high in saturated fats can also cause high blood cholesterol and high levels of triglycerides. Foods that contain saturated fats include:

- Dairy products, such as cheese, cream, and butter
- Sugary treats, such as cakes, cookies, and ice cream
- Fatty or cured meats, such as sausages, bacon, and salami
- Foods containing coconut oil, palm oil, or lard

Foods that contain trans fats are particularly dangerous, as they raise bad cholesterol and lower good cholesterol. Foods that contain trans fats include:

- Baked goods such as cookies, pies, and pastries
- Fried, fatty foods, such as fried chicken, french fries, and doughnuts
- Shortening, certain margarines, and vegetable oils
- Non-dairy coffee creamers

Other dietary and lifestyle factors that can elevate cholesterol include:

- Smoking or exposure to tobacco smoke
- Being physically inactive
- Having overweight or obesity
- Drinking too much alcohol
- Taking certain medications, such as diuretics

7.8 Medical conditions

Underlying health conditions may increase cholesterol levels, including:

- Diabetes
- Hypothyroidism
- Kidney disease
- Liver disease

7.9 Diagnosis of lipid disorder

A doctor can run a blood test called a lipid profile or lipid panel to initially diagnose a lipid disorder. This measures levels of total cholesterol, LDLs, HDLs, and triglycerides, among other things. If blood test results show elevated cholesterol, a doctor may order other tests to rule out underlying problems that may be causing it, such as thyroid or liver tests. Occasionally, if a doctor suspects a patient needs intensive treatment, they may order

advanced lipid testing. This checks the concentration of lipoproteins in the blood and helps to predict the risk of cardiovascular disease more accurately.

7.10 Treatments for lipid disorder

Treatment for a lipid disorder may depend on cholesterol levels, age, and what underlying health conditions are also present. A healthcare professional will usually recommend lifestyle changes as a primary intervention to tackle high cholesterol. These changes may include:

- Reducing or cutting out saturated and trans fats
- Eating more healthily and following a diet with more oily fish, brown rice and pasta, and fruit and vegetables
- Taking up moderate exercise to lose weight — at least 150 minutesTrusted Source a week
- Quitting smoking or vaping
- Cutting down on alcohol

Medication may also be necessary to manage cholesterol levels, so many healthcare professionals may recommend a combination of medication and lifestyle changes. Medications for cholesterol management may include:

7.11 LDL and HDL Cholesterol: "Bad" and "Good" Cholesterol

Cholesterol travels through the blood on proteins called “lipoproteins.” Two types of lipoproteins carry cholesterol throughout the body:

7.12 LDL (low-density lipoprotein), **sometimes called “bad” cholesterol,** makes up most of your body’s cholesterol. High levels of LDL cholesterol raise your risk for heart disease and stroke. Low-density lipoproteins (LDL) carry cholesterol from the liver to the rest of the body. Cells latch onto these particles and extract fat and cholesterol from them. When there is too much LDL cholesterol in the blood, these particles can form deposits in the walls of the coronary arteries and other arteries throughout the body. Such deposits, called plaque, can narrow arteries and limit blood flow. When plaque breaks apart, it can cause a heart attack or stroke. Because of this, LDL cholesterol is often referred to as bad, or harmful, cholesterol.

7.13 HDL (high-density lipoprotein), or “good” cholesterol, absorbs cholesterol and carries it back to the liver. The liver then flushes it from the body. High levels of HDL cholesterol can lower your risk for heart disease and stroke. When your body has too much LDL cholesterol, the LDL cholesterol can build up on the walls of your blood vessels. This buildup is called “plaque.” As your blood vessels build up plaque over time, the insides of the vessels

narrow. This narrowing blocks blood flow to and from your heart and other organs. When blood flow to the heart is blocked, it can cause angina (chest pain) or a heart attack. High-density lipoproteins (HDL) scavenge cholesterol from the bloodstream, from LDL, and from artery walls and ferry it back to the liver for disposal. Think of HDL as the garbage trucks of the bloodstream. HDL cholesterol is often referred to as good, or protective, cholesterol.

7.14 What is cholesterol?

Cholesterol is a waxy, fat-like substance that's found in all the cells in your body. Your liver makes cholesterol, and it is also in some foods, such as meat and dairy products. Your body needs some cholesterol to work properly. But having too much cholesterol in your blood raises your risk of coronary artery disease.

7.15 What are HDL and LDL?

HDL and LDL are two types of lipoproteins. They are a combination of fat (lipid) and protein. The lipids need to be attached to the proteins so they can move through the blood. HDL and LDL have different purposes:

- **HDL stands for high-density lipoproteins.** It is sometimes called the "good" cholesterol because it carries cholesterol from other parts of your body back to your liver. Your liver then removes the cholesterol from your body.
- **LDL stands for low-density lipoproteins.** It is sometimes called the "bad" cholesterol because a high LDL level leads to a buildup of cholesterol in your arteries.

7.16 How do I know what my HDL level is?

A blood test can measure your cholesterol levels, including HDL. When and how often you should get this test depends on your age, risk factors, and family history. The general recommendations are:

For people who are age 19 or younger:

- The first test should be between ages 9 to 11
- Children should have the test again every 5 years
- Some children may have this test starting at age 2 if there is a family history of high blood cholesterol, heart attack, or stroke.

For people who are age 20 or older:

- Younger adults should have the test every 5 years
- Men ages 45 to 65 and women ages 55 to 65 should have it every 1 to 2 years

7.16 What should my HDL level be?

With HDL cholesterol, higher numbers are better, because a high HDL level can lower your risk for coronary artery disease and stroke. How high your HDL should be depends on your age and sex:

Group	Healthy HDL Level
Age 19 or younger	More than 45mg/dl
Men age 20 or older	More than 40mg/dl
Women age 20 or older	More than 50mg/dl

7.17 How can I raise my HDL level?

If your HDL level is too low, lifestyle changes may help. These changes may also help prevent other diseases, and make you feel better overall:

7.18 Cholesterol

Cholesterol is a waxy substance found in your blood. Your body needs cholesterol to build healthy cells, but high levels of cholesterol can increase your risk of heart disease. With high cholesterol, you can develop fatty deposits in your blood vessels. Eventually, these deposits grow, making it difficult for enough blood to flow through your arteries. Sometimes, those deposits can break suddenly and form a clot that causes a heart attack or stroke. High cholesterol can be inherited, but it's often the result of unhealthy lifestyle choices, which make it preventable and treatable. A healthy diet, regular exercise and sometimes medication can help reduce high cholesterol.

- The biggest influence on blood cholesterol level is the mix of fats and carbohydrates in your diet—not the amount of cholesterol you eat from food.
- Although it remains important to limit the amount of cholesterol you eat, especially if you have diabetes, for most people dietary cholesterol is not as problematic as once believed.
- The body uses cholesterol as the starting point to make estrogen, testosterone, vitamin D, and other vital compounds.
- Cholesterol in the bloodstream, specifically the bad LDL cholesterol, is what’s most important in determining health risk.

7.19 How Fat Moves from Food to the Bloodstream

Fat and cholesterol can't dissolve in water or blood. Instead, the body packages fat and cholesterol into tiny, protein-covered particles called lipoproteins. Lipoproteins can transport a lot of fat; they mix easily with blood and flow with it. Some of these particles are big and fluffy, while others are small and dense. The most important ones are low-density lipoproteins (LDL), high-density lipoproteins (HDL), and triglycerides.

- **Triglycerides**

Triglycerides make up most of the fat that you eat and that travels through the bloodstream. As the body's main vehicle for transporting fats to cells, triglycerides are important for good health, though high levels of triglycerides can be unhealthy.

In general, the lower your LDL and the higher your HDL, the better your chances of preventing heart disease and other chronic conditions.

How Fat and Cholesterol in Food Affect Blood Cholesterol Levels

The types of fat in the diet help determine the amount of total, HDL, and LDL cholesterol in the bloodstream. The types and amount of carbohydrate in the diet also play a role. Cholesterol in food matters, too, but not nearly as much.

- The discovery half a century ago that high blood cholesterol levels were strongly associated with an increased risk for heart disease triggered numerous warnings to avoid foods that contain cholesterol, especially eggs and liver. However, scientific studies show a weak relationship between the amount of cholesterol a person consumes and his or her blood cholesterol levels.

- In studies of more than 80,000 female nurses, Harvard researchers found that consuming about an egg a day was not associated with higher risk of heart disease. However, people who have heart disease or diabetes should monitor egg consumption.

For most people, the amount of cholesterol eaten has only a modest impact on the amount of cholesterol circulating in the blood. (24) For some people, though, blood cholesterol levels rise and fall very strongly in relation to the amount of cholesterol eaten. For these "responders," avoiding cholesterol-rich foods can have a substantial effect on blood cholesterol levels. Unfortunately, at this point there is no way other than by trial and error to identify responders from non-responders to dietary cholesterol.

7.20 Triglyceride (TG)

A triglyceride (TG, triacylglycerol, TAG, or triacylglyceride) is an ester derived from glycerol and three fatty acids (from *tri-* and *glyceride*). Triglycerides are the main constituents of body fat in humans and other vertebrates, as well as vegetable fat. They are also present in the blood to enable the bidirectional transference of adipose fat and blood glucose from the liver, and are a major component of human skin oils. Many types of triglycerides exist. One specific classification focuses on saturated and unsaturated types. Saturated fats have *no* C=C groups; unsaturated fats feature one or more C=C groups. Unsaturated fats tend to have a lower melting point than saturated analogues; as a result, they are often liquid at room temperature.

7.21 Homo- and heterotriglycerides

The simplest triglycerides are those where the three fatty acids are identical. Their names indicate the fatty acid: stearin derived from stearic acid, palmitin derived from palmitic acid, etc. These compounds can be obtained in three crystalline forms (polymorphs): α, β, and β′, the three forms differing in their melting points. If the first and third fatty acids on the glycerol differ, then the triglyceride is chiral.

7.22 Conformation

The shape of fat and fatty acid molecules is usually not well-defined. Any two parts of a molecule that are connected by just one single bond are free to rotate about that bond. Thus a fatty acid molecule with *n* simple bonds can be deformed in *n*-1 independent ways (counting also rotation of the terminal methyl group). Such rotation cannot happen across a double bond, except by breaking and then reforming it with one of the halves of the molecule rotated by 180 degrees, which requires crossing a significant energy barrier.

Thus a fat or fatty acid molecule with double bonds (excluding at the very end of the chain) can have multiple cis–trans isomers with significantly different chemical and biological properties. Each double bond reduces the number of conformational degrees of freedom by one. Each triple bond forces the four nearest carbons to lie in a straight line, removing two degrees of freedom.

It follows that depictions of "saturated" fatty acids with no double bonds (like stearic) having a "straight zig-zag" shape, and those with one *cis* bond (like oleic) being bent in an "elbow" shape are somewhat misleading. While the latter are a little less flexible, both can be twisted to assume similar straight or elbow shapes. In fact, outside of some specific contexts like crystals or bilayer membranes, both are more likely to be found in randomly contorted configurations than in either of those two shapes.

7.23 Phospholipids

Phospholipids are ubiquitous molecules that participate in innumerous cellular events. The biochemical and physiological properties of phospholipids are largely related to their amphipathic feature. Mammalian cell membranes contain more than 1000 different phospholipids. Phospholipids exert structural functions in cellular membranes, which vary in phospholipid composition, according to cell and organelle functions. Phospholipid regulatory functions are both direct and indirect, through their metabolites. It is well established that phospholipids participate in a broad range of cellular events such as apoptosis and regulation of mitochondrial physiology and that phospholipid-derived messenger molecules are crucial for the transduction of extracellular signals to intracellular events.

Phospholipids are an important class of membrane lipids that contain two categories of lipids, glycerophospholipids and sphingolipids. Glycerophospholipids are similar to triglycerides except that one hydroxyl group of glycerol is replaced by the ester of phosphoric acid and an amino alcohol, bonded through a phosphodiester bond. They form a very important part of cell membranes, which could be the possible reason behind the success of liposomes (which are made up of phospholipids). Several kinds of phospholipids exist in the body, including the brain. Lecithins are a kind of phospholipids that can be synthesized by the body, are found in the body cells, and also participate in the digestion.

Sphingolipids (or brain lipids) are another kind of phospholipids that are esters of an 18-carbon alcohol called sphingosine. They are similar to phospholipids, but the glycerol backbone (in phospholipids) is replaced by sphingosine. They are essential to the structure of cell membranes and are abundant in brain tissues and nerve cell membranes. Sphingolipids include the sphingomyelins and cerebrosides that are based on the molecule sphingosine. As the name suggests, these lipids are affiliated with the myelin sheath surrounding the cells of the neurons. Sphingomyelins comprise about 25% of the lipids in the myelin sheath, and their key role is to transmit electric signals. Cerebrosides are not phospholipids, but contain sphingosine.

7.24 Applications

Phospholipids have been widely used to prepare liposomal, ethosomal and other nanoformulations of topical, oral and parenteral drugs for differing reasons like improved bio-availability, reduced toxicity and increased permeability across membranes. Liposomes are often composed of phosphatidylcholine-enriched phospholipids and may also contain mixed phospholipid chains with surfactant properties. The ethosomal formulation

of ketoconazole using phospholipids is a promising option for transdermal delivery in fungal infections. Advances in phospholipid research lead to exploring these biomolecules and their conformations using lipidomics.

7.25 Simulations

Computational simulations of phospholipids are often performed using molecular dynamics with force fields such as GROMOS, CHARMM, or AMBER.

7.26 Characterization

Phospholipids are optically highly birefringent, i.e. their refractive index is different along their axis as opposed to perpendicular to it. Measurement of birefringence can be achieved using cross polarisers in a microscope to obtain an image of e.g. vesicle walls or using techniques such as dual polarisation interferometry to quantify lipid order or disruption in supported bilayers.

7.27 Analysis

There are no simple methods available for analysis of phospholipids, since the close range of polarity between different phospholipid species makes detection difficult. Oil chemists often use spectroscopy to determine total phosphorus abundance and then calculate approximate mass of phospholipids based on molecular weight of expected fatty acid species. Modern lipid profiling employs more absolute methods of analysis, with NMR spectroscopy, particularly P-NMR, while HPLC-ELSD provides relative values.

7.28 Phospholipid synthesis

Phospholipid synthesis occurs in the cytosolic side of ER membrane that is studded with proteins that act in synthesis (GPAT and LPAAT acyl transferases, phosphatase and choline phosphotransferase) and allocation (flippase and floppase). Eventually a vesicle will bud off from the ER containing phospholipids destined for the cytoplasmic cellular membrane on its exterior leaflet and phospholipids destined for the exoplasmic cellular membrane on its inner leaflet.

7.29 Sources

Common sources of industrially produced phospholipids are soya, rapeseed, sunflower, chicken eggs, bovine milk, fish eggs etc. Phospholipids for gene delivery such as distearoylphosphatidylcholine, dioleoyl-3-trimethylammonium propane etc. are produced synthetically. Each source has a unique profile of individual phospholipid species, as well as fatty acids, and consequently differing applications in food, nutrition, pharmaceuticals, cosmetics and drug delivery.

7.30 In signal transduction

Some types of phospholipid can be split to produce products that function as second messengers in signal transduction. Examples include phosphatidylinositol (4,5)-bisphosphate (PIP_2), that can be split by the enzyme phospholipase C into inositol triphosphate (IP_3) and diacylglycerol (DAG), which both carry out the functions of the G_q type of G protein in response to various stimuli and intervene in various processes from long term depression in neurons to leukocyte signal pathways started by chemokine receptors.

Phospholipids also intervene in prostaglandin signal pathways as the raw material used by lipase enzymes to produce the prostaglandin precursors. In plants they serve as the raw material to produce jasmonic acid, a plant hormone similar in structure to prostaglandins that mediates defensive responses against pathogens.

7.31 Food technology

Phospholipids can act as emulsifiers, enabling oils to form a colloid with water. Phospholipids are one of the components of lecithin, which is found in egg yolks, as well as being extracted from soybeans, and is used as a food additive in many products and can be purchased as a dietary supplement. Lysolecithins are typically used for water–oil emulsions like margarine, due to their higher HLB ratio.

7.32 Gaucher's disease

Gaucher's disease or Gaucher disease (GD) is a genetic disorder in which glucocerebroside (a sphingolipid, also known as glucosylceramide) accumulates in cells and certain organs. The disorder is characterized by bruising, fatigue, anemia, low blood platelet count and enlargement of the liver and spleen, and is caused by a hereditary deficiency of the enzyme glucocerebrosidase (also known as glucosylceramidase), which acts on glucocerebroside. When the enzyme is defective, glucocerebroside accumulates, particularly in white blood cells and especially in macrophages (mononuclear leukocytes). Glucocerebroside can collect in the spleen, liver, kidneys, lungs, brain, and bone marrow.

Manifestations may include enlarged spleen and liver, liver malfunction, skeletal disorders or bone lesions that may be painful, severe neurological complications, swelling of lymph nodes and (occasionally) adjacent joints, distended abdomen, a brownish tint to the skin, anemia, low blood platelet count, and yellow fatty deposits on the white of the eye (sclera). Persons seriously affected may also be more susceptible to infection. Some forms of Gaucher's disease may be treated with enzyme replacement therapy.

The disease is caused by a recessive mutation in the GBA gene located on chromosome 1 and affects both males and females. About one in 100 people in the United States are carriers of the most common type of Gaucher disease. The carrier rate among Ashkenazi Jews is 8.9% while the birth incidence is one in 450. Gaucher's disease is the most common of the lysosomal storage diseases. It is a form of sphingolipidosis (a subgroup of lysosomal storage diseases), as it involves dysfunctional metabolism of sphingolipids. The disease is named after the French physician Philippe Gaucher, who originally described it in 1882.

7.33 Signs and symptoms

- Painless hepatomegaly and splenomegaly: the size of the spleen can be 1500–3000 g, as opposed to the normal size of 50–200 g. Splenomegaly may decrease the affected individual's capacity for eating by exerting pressure on the stomach. While painless, enlargement of spleen increases the risk of splenic rupture.
- Hypersplenism and pancytopenia, the rapid and premature destruction of blood cells, leads to anemia, neutropenia, leukopenia, and thrombocytopenia (with an increased risk of infection and bleeding).
- Cirrhosis of the liver is rare.
- Severe pain associated with joints and bones occurs, frequently presenting in hips and knees.
- Neurological symptoms occur only in some types of Gaucher's (see below):

7.34 Diagnosis

Gaucher disease is suggested based on the overall clinical picture. Initial laboratory testing may include enzyme testing. As a result, lower than 15% of mean normal activity is considered to be diagnostic. Decreased enzyme levels will often be confirmed by genetic testing. Numerous different mutations occur; sequencing of the beta-glucosidase gene is sometimes necessary to confirm the diagnosis. Prenatal diagnosis is available and is useful when a known genetic risk factor is present.

A diagnosis can also be implied by biochemical abnormalities such as high alkaline phosphatase, angiotensin-converting enzyme, and immunoglobulin levels, or by cell analysis showing "crinkled paper" cytoplasm and glycolipid-laden macrophages. Some lysosomal enzymes are elevated, including tartrate-resistant acid phosphatase, hexosaminidase, and a human chitinase, chitotriosidase. This latter enzyme has proved to be very useful for monitoring Gaucher's disease activity in response to treatment, and may reflect the severity of the disease.

7.35 Classification

Gaucher's disease (GD) has three common clinical subtypes. These subtypes have come under some criticism for not taking account of the full spectrum of observable symptoms. Also, compound heterozygous variations occur which considerably increase the complexity of predicting disease course.

GD type I (non-neuropathic) is the most common and least severe form of the disease. Symptoms may begin early in life or in adulthood and mainly affect the liver, spleen, and bone. Enlarged liver and grossly enlarged spleen (together hepatosplenomegaly) are common; the spleen can rupture and cause additional complications. Skeletal weakness and bone disease may be extensive. Spleen enlargement and bone marrow replacement cause anemia, thrombocytopenia, and leukopenia.

The brain and nervous system are not affected pathologically, but lung and, rarely, kidney impairment may occur. Patients in this group usually bruise easily (due to low levels of platelets) and experience fatigue due to low numbers of red blood cells. Depending on disease onset and severity, type I patients may live well into adulthood. The range and severity of symptoms can vary dramatically between patients.

GD type II (acute infantile neuropathic) typically begins within 6 months of birth and has an incidence rate around one 1 in 100,000 live births. Symptoms include an enlarged liver and spleen, extensive and progressive brain damage, eye movement disorders, spasticity, seizures, limb rigidity, and a poor ability to suck and swallow. Affected children usually die by age two.

GD type III (chronic neuropathic) can begin at any time in childhood or even in adulthood, and occurs in about one in 100,000 live births. It is characterized by slowly progressive, but milder neurologic symptoms compared to the acute or type II version. Major symptoms include an enlarged spleen and/or liver, seizures, poor coordination, skeletal irregularities, eye movement disorders, blood disorders including anemia, and respiratory problems. Patients often live into their early teen years and adulthood.

Treatment

For those with type-I and most type-III, enzyme replacement treatment with intravenous recombinant glucocerebrosidase can decrease liver and spleen size, reduce skeletal abnormalities, and reverse other manifestations. This treatment costs about US$200,000 annually for a single person and should be continued for life. The rarity of the disease means dose-finding studies have been difficult to conduct, so controversy remains

over the optimal dose and dosing frequency. Due to the low incidence, this has become an orphan drug in many countries, meaning a government recognizes and accommodates the financial constraints that limit research into drugs that address a small population.

The first drug for Gaucher's was alglucerase (Ceredase), which was a version of glucocerebrosidase that was harvested from human placental tissue and then modified with enzymes. It was approved by the FDA in 1991 and has been withdrawn from the market due to the approval of similar drugs made with recombinant DNA technology instead of being harvested from tissue; drugs made recombinantly are preferable, since there is no concern about diseases being transmitted from the tissue used in harvesting, there are fewer risks of variations in enzyme structure from batch to batch, and they are less expensive to manufacture.

7.36 Tay-Sachs Disease

Tay-Sachs disease is a genetic condition. Tay-Sachs is caused by a baby receiving two defective HEXA genes, one from each parent. Tay-Sachs disease symptoms include failing to meet motor milestones, such as sitting and standing. Babies born with Tay-Sachs often die at a young age. Genetic testing can help you make family planning decisions.

7.37 What is Tay-Sachs disease?

Tay-Sachs disease affects the nerve cells in the brain and spinal cord. Babies with Tay-Sachs lack a particular enzyme, which is a protein that triggers chemical reactions in cells. The lack of the enzyme, hexosaminidase A, causes a fatty substance to collect. The buildup of this substance, GM2 ganglioside, leads to Tay-Sachs symptoms such as muscle weakness. Tay-Sachs is a genetic condition. It's caused by changes in a pair of genes inherited from parents. It's a progressive disease, meaning it gets worse over time. Children born with Tay-Sachs often die by age 4, usually from complications of pneumonia. There's no cure, with treatment aimed at supporting the child and keeping them comfortable.

Genetic testing is available for couples who may face a higher risk for having a baby with Tay-Sachs. Genetic testing and counseling can help parents-to-be make informed decisions about family planning.

7.38 Who is at risk for Tay-Sachs?

People across racial and ethnic groups can carry a genetic change tied to Tay-Sachs disease. But it's much more common among people of Ashkenazi (Eastern European) Jewish descent.

Other populations with higher numbers of people carrying the disease-causing genetic change include:

- French Canadians.
- Cajuns (from Louisiana).
- Old Order Amish (in Pennsylvania).
- Those with Irish ancestry. Some studies have shown that they have a 1 in 50 chance of carrying such a gene.

How common is Tay-Sachs disease?

For people not from high-risk backgrounds, around 1 in 300 people carry the genetic change (or variant gene) for Tay-Sachs. For people of Ashkenazi Jewish descent:

- About 1 in 30 people carry the variant gene.
- About 1 in 3,600 newborns is affected.

7.39 What are the forms of Tay-Sachs?

There are several forms of Tay-Sachs disease. The type a child has depends on when symptoms develop. Families usually only have one form of the disease. So if a child has infantile Tay-Sachs, it's not likely that older siblings will develop juvenile or late-onset Tay-Sachs:

- **Classic infantile Tay-Sachs:** This is the most common form of Tay-Sachs. Children develop symptoms around 6 months.
- **Juvenile Tay-Sachs:** Children develop symptoms between ages 2 and 5. This form is very rare.
- **Chronic Tay-Sachs:** Children develop symptoms before age 10.
- **Late-onset Tay-Sachs:** Symptoms can appear during the teen years or early adulthood. They can also develop later as well. This type of the disease may not affect life expectancy. It's also a very rare form of Tay-Sachs.

7.40 Are there similar diseases?

The symptoms and progression of Tay Sachs are like those of Sandhoff disease, another inherited condition. Sandhoff disease involves hexosaminidase A and a second enzyme, hexosaminidase B.

7.41 SYMPTOMS AND CAUSES

What causes Tay-Sachs disease?

Tay-Sachs comes from a disease-causing variant (change) to the HEXA gene. Each baby has two copies of the HEXA gene, one from their biological father and one from their biological mother. Tay-Sachs happens when both parents had a variant HEXA gene and passed it on. That means neither copy of the baby's HEXA gene works well. Healthcare providers call this hexosaminidase A deficiency, or hex A deficiency.

7.42 What does it mean to be a Tay-Sachs carrier?

A carrier has one working copy of the HEXA gene and one copy with a disease-causing variant. Carriers don't have the disease or show symptoms, since their bodies can rely on the working gene. But if two carriers have children together, there's a:

- 25% (1 in 4) chance that the child won't inherit any variant HEXA genes. The child won't have Tay-Sachs or be a carrier.
- 50% (1 in 2) chance that the child gets a variant gene from one parent. When this happens, the child will be a carrier but won't have Tay-Sachs. Since they're a carrier, they may pass it to their children.
- 25% (1 in 4) chance that the child gets a variant gene from both parents. In this case, the child has Tay-Sachs disease.

7.43 What are typical symptoms of Tay-Sachs disease?

Symptoms of the most common form of Tay-Sachs start developing when babies are around 3 to 6 months old. The symptoms continue to progress as the child gets older. Children often don't meet their developmental milestones.

7.44 Summary

Under this unit we summarize that; metabolism is the process your body uses to make energy from the food you eat. Food is made up of proteins, carbohydrates, and fats. Chemicals in your digestive system (enzymes) break the food parts down into sugars and acids, your body's fuel. Your body can use this fuel right away, or it can store the energy in your body tissues. If you have a metabolic disorder, something goes wrong with this process. Lipid metabolism disorders, such as Gaucher disease and Tay-Sachs disease, involve lipids. Lipids are fats or fat-like substances. They include oils, fatty acids, waxes, and cholesterol. If you have one of these disorders, you may not have enough enzymes to break down lipids. Or the enzymes may not work properly and your body can't convert the fats into energy.

They cause a harmful amount of lipids to build up in your body. Over time, that can damage your cells and tissues, especially in the brain, peripheral nervous system, liver, spleen, and bone marrow. Many of these disorders can be very serious, or sometimes even fatal. Cholesterol travels through the blood on proteins called "lipoproteins." Two types of lipoproteins carry cholesterol throughout the body. LDL (low-density lipoprotein), sometimes called "bad" cholesterol, makes up most of your body's cholesterol.

High levels of LDL cholesterol raise your risk for heart disease and stroke. HDL (high-density lipoprotein), or "good" cholesterol, absorbs cholesterol and carries it back to the liver. The liver then flushes it from the body. High levels of HDL cholesterol can lower your risk for heart disease and stroke. When your body has too much LDL cholesterol, the LDL cholesterol can build up on the walls of your blood vessels. This buildup is called "plaque." As your blood vessels build up plaque over time, the insides of the vessels narrow. This narrowing blocks blood flow to and from your heart and other organs. When blood flow to the heart is blocked, it can cause angina (chest pain) or a heart attack.

7.45 Terminal questions

Q.17. What do you mean by disorders of lipids? Explain it.

Answer:----------

Q.18. What are the low density lipoproteins?

Answer:----------

Q.19. What are the high density lipoproteins?

Answer:----------

Q.20. What are the phospholipids in health and disease?

Answer:----------

Q.21. What are the differences between low and high density lipoproteins?

Answer:----------

Q.22. Write a short note on cholesterol.

Answer:----------

Q.23. Write a short note on triglycerides.

Answer:----------

Further readings

11. Biochemistry- Lehninger A.L.

12. Biochemistry –J.H.Weil.

13. Biochemistry fourth edition-David Hames and Nigel Hooper.

14. Textbook of Biochemistry for Undergraduates - Rafi, M.D.

15. Biochemistry and molecular biology- Wilson Walker.

Unit-8

8.1 Introduction

About four-fifths of Earth's atmosphere is nitrogen, which was isolated and recognized as a specific substance during early investigations of the air. Carl Wilhelm Scheele, a Swedish chemist, showed in 1772 that air is a mixture of two gases, one of which he called "fire air," because it supported combustion, and the other "foul air," because it was left after the "fire air" had been used up. The "fire air" was, of course, oxygen and the "foul air" nitrogen. At about the same time, nitrogen also was recognized by a Scottish botanist, Daniel Rutherford, by the British chemist Henry Cavendish, and by the British clergyman and scientist Joseph Priestley, who, with Scheele, is given credit for the discovery of oxygen.

Later work showed the new gas to be a constituent of nitre, a common name for potassium nitrate (KNO_3), and, accordingly, it was named nitrogen by the French chemist Jean-Antoine-Claude Chaptal in 1790. Nitrogen first was considered a chemical element by Antoine-Laurent Lavoisier, whose explanation of the role of oxygen in combustion eventually overthrew the phlogiston theory, an erroneous view of combustion that became popular in the early 18th century. The inability of nitrogen to support life led Lavoisier to name it *azote*, still the French equivalent of *nitrogen*.

This unit provides concept of nitrogen metabolism. It describes the biosynthesis of amino acids in plants and microorganisms, and in mammals and humans. Degradation of amino acids in microorganisms and plants is a rare event, but is of major importance in animals and humans. In man and mammals, the ammonium ions are converted into urea in the liver. The form in which amino nitrogen is excreted from the organism is dependent upon the adaptation of the organism to its habitat. In terms of amino nitrogen excretion, the animal kingdom can be classified into ammonotelic (ammonia), ureotelic (urea), and uricotelic (uric acid) organisms depending on the nature of the discharged substance. Urea is produced as a nontoxic soluble vehicle for the elimination of nitrogen originating from the catabolism of amino acids. A major function of the liver is urea biosynthesis, but enzymes of the pathway also occur in kidney, skin, brain plus some other cells where their primary purpose is to synthesize arginine. The chapter also discusses the biosynthesis of heterocyclic compounds, the hybridoma technique, and the degradation of heterocyclic compounds.

Objectives

This is the eighth unit on clinical biochemistry. Under eighth unit we have following objectives. These are as under:

- To know about nitrogen and nitrogen cycle
- To know about nitrogen metabolism
- To know about uremia, hyperuricemia and porphyria

8.2 The Nitrogen Cycle

Fig. Nitrogen cycle

Nitrogen is essential to life because it is a key component of proteins and nucleic acids. Nitrogen occurs in many forms and is continuously cycled among these forms by a variety of bacteria. Although nitrogen is abundant in the atmosphere as diatomic nitrogen gas (N_2), it is extremely stable, and conversion to other forms requires a great deal of energy.

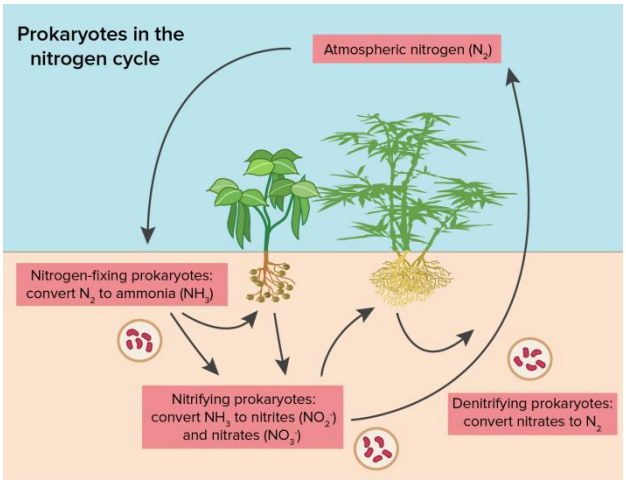

Fig. Prokaryotes in nitrogen cycle

Historically, the biologically available forms NO_3- and NH_3 have often been limited; however, current anthropogenic processes, such as fertilizer production, have greatly increased the availability of nitrogen to living organisms. The cycling of nitrogen among its many forms is a complex process that involves numerous types of bacteria and environmental conditions.

In general, the nitrogen cycle has five steps:

1. Nitrogen fixation (N_2 to NH_3/ NH^{4+} or $NO^{3-)}$
2. Nitrification (NH_3 to $NO^{3-)}$
3. Assimilation (Incorporation of NH_3 and NO^{3-} into biological tissues)
4. Ammonification (organic nitrogen compounds to NH_3)
5. Denitrification (NO^{3-} to N_2)

8.3 Nitrogen Fixation

Nitrogen fixation is the process by which gaseous nitrogen (N2) is converted to ammonia (NH3 or NH4+) via biological fixation or nitrate (NO3-) through high-energy physical processes. N2 is extremely stable and a great deal of energy is required to break the bonds that join the two N atoms. N2 can be converted directly into NO3- through processes that exert a tremendous amount of heat, pressure, and energy. Such processes include combustion, volcanic action, lightning discharges, and industrial means. However, a greater amount of biologically available nitrogen is naturally generated via the biological conversion of N_2 to NH_3/ NH^{4+}. A small group of bacteria and cyanobacteria are capable using the enzyme nitrogenase to break the bonds among the molecular nitrogen and combine it with hydrogen.

Nitrogenase only functions in the absence of oxygen. The exclusion of oxygen is accomplished by many means. Some bacteria live beneath layers of oxygen-excluding slime on the roots of certain plants. The most important soil dwelling bacteria, Rhizobium, live in oxygen-free zones in nodules on the roots of legumes and some other woody plants. Aquatic filamentous *cyanobacteria* utilize oxygen-excluding cells called *heterocysts*.

8.4 Nitrification

Nitrification is a two-step process in which NH3/ NH4+ is converted to NO3-. First, the soil bacteria *Nitrosomonas* and *Nitrococcus* convert NH3 to NO2-, and then another soil bacterium, *Nitrobacter*, oxidizes NO^{2-} to NO^{3-}. These bacteria gain energy through these conversions, both of which require oxygen to occur.

8.5 Assimilation

Assimilation is the process by which plants and animals incorporate the NO3- and ammonia formed through nitrogen fixation and nitrification. Plants take up these forms of nitrogen through their roots, and incorporate them into plant proteins and nucleic acids. Animals are then able to utilize nitrogen from the plant tissues.

8.6 Ammonification

Assimilation produces large quantities of organic nitrogen, including proteins, amino acids, and nucleic acids. *Ammonification* is the conversion of organic nitrogen into ammonia. The ammonia produced by this process is excreted into the environment and is then available for either nitrification or assimilation.

8.7 Denitrification

Denitrification is the reduction of NO^{3-} to gaseous N_2 by anaerobic bacteria. This process only occurs where there is little to no oxygen, such as deep in the soil near the water table. Hence,

areas such as wetlands provide a valuable place for reducing excess nitrogen levels via denitrification processes.

8.8 Common Forms of Nitrogen

The most common forms of inorganic nitrogen in the environment are diatomic nitrogen gas (N_2), nitrate (NO^{3-}), nitrite (NO^{2-}), ammonia (NH_3), and ammonium (NH^{4+}). The species that predominate depend on the chemical, physical, and biological environment. In aquatic environments, the presence of nitrogen as unionized ammonia (NH_3) or ammonium (NH^{4+}) is dependent on the pH and temperature. When the pH is below 8.75, NH^{4+} predominates. Increases in pH signify increases in the hydroxyl ion (OH-) concentration of the water, meaning the above reaction will shift to the left in order to reach equilibrium. Above a pH of 9.75, NH_3 predominates (Hem, 1985). NH_3 is a more toxic to aquatic life. If biological assimilation of NH_3 is not occurring at a sufficient rate, NH_3 may accumulate and cause detrimental effects to aquatic life.

In soils, $NH4^{+}$ ions are strongly sorbed by clay particles and organic matter, which have a net negative surface charge. In alkaline soils, NH^{4+} will be converted to NH3 gas, and lost to the atmosphere. Under warm growing conditions, NH^{4+} in the soil will be transformed to NO^{3-} via nitrification. NO^{3-} is very soluble, and can easily be leached from soils under wet conditions.

8.9 Nitrogen Monitoring

Monitoring nitrogen levels is necessary for many reasons, including detecting baseline nutrient levels and trends, preventing eutrophication, maximizing soil productivity, and minimizing toxic effects of ammonia or nitrite poisoning.

8.10 Nitrogen metabolism

Nitrogen metabolism is not only one of the basic processes of plant physiology, but also one of the important parts of global chemical cycle. Plant nitrogen assimilation directly takes part in the synthesis and conversion of amino acid through the reduction of nitrate. During this stage, some key enzymes, e.g., nitrate reductase (NR), glutamine synthetase (GS), glutamate dehydrogenase (GDH), glutamine synthase (GOGAT), aspargine synthetase (AS), and asparate aminotransferase (AspAT) participate these processes. The protein is assimilated in plant cell through amino acid, and becomes a part of plant organism through modifying, classifying, transporting and storing processes, etc.

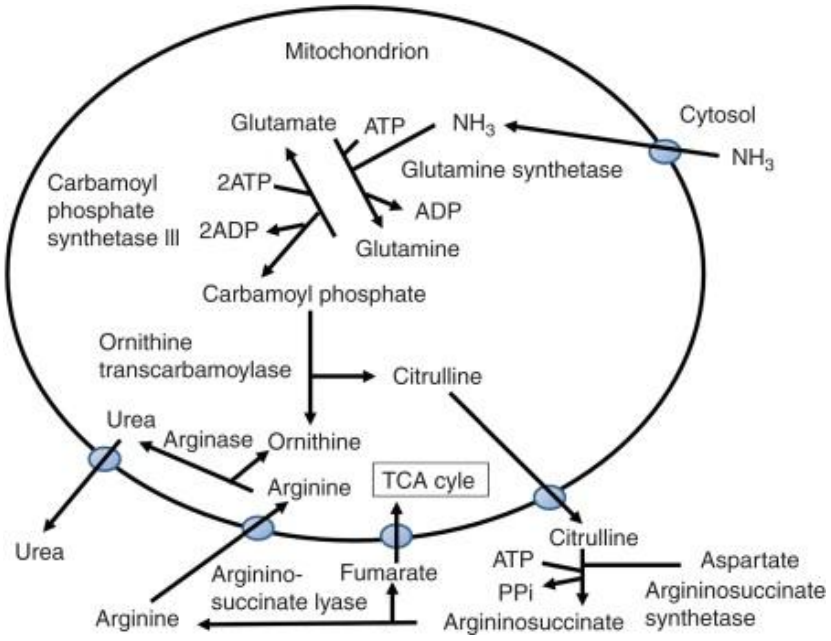

Fig. Nitrogen metabolism

The nitrogen metabolism is associated with carbonic metabolism through key enzyme regulations and the conversion of products, which consists of basic life process. Among these amino acids in plant cell, glutamic acid (Glu), glutamine (Gln), aspartic acid (Asp) and asparagines (Asn), etc., play a key role, which regulates their conversion each other and their contents in the plant cell through regulating formation and activity of those key enzymes. Environmental factors also affect the conversion and recycle of the key amino acids through regulating gene expression of the key enzymes and their activities. Nitrate and light intensity positively regulate the gene transcription of NR, but ammonium ions and Glu, Gln do the negative way. Water deficit is a very serious constraint on N_2 fixation rate and soybean grain yield, in which, ureide accumulation and degradation under water deficit appear to be the key issues of feedback mechanism on nitrogen fixation.

Water stress decreases NR activity, but increases proteinase activity, and thus, they regulate plant nitrogen metabolism, although there are some different effects among species and cultivars. Water stress also decreases plant tissue protein content, ratio of protein and amino acid, and reduces the absorption of amino acid by plant. On the contrary, soil flooding decreases the content and accumulation amount of root nitrogen in winter wheat by 11.9% from booting to flowering stages and 39.1% during grain filling stage, and reduces the ratio of carbon and nitrogen by 79.6%. The results misadjust the metabolism between carbon and nitrogen, and result in the end of the root growth. Elevated CO_2 level could decrease plant leaf

nitrogen content under well-watered condition, but almost maintain stable under water deficit condition.

The radiation of UV-B significantly reduces the partitioning coefficient and synthetic rate of Rubisco, which significantly decreases the photosynthetic rate. This unit explains the pathway of plant nitrogen assimilation, characteristics of key enzymes and their regulating mechanisms with picturing the regulating mode of NR, and described the signal sensing and conduct of plant nitrogen metabolism and the formation, transportation, storage and degradation of plant cell protein with picturing the schedule of protein transport of membrane system in plant cell. Seven key tasks are emphasized in this paper in terms of the review on the effects and mechanisms of key ecological factors including water stress on plant nitrogen metabolism. They are:

1) The absorption mechanism of plant based on different nitrogen sources and environmental regulations,
2) The localization and compartmentalization of the key enzymes of nitrogen mechanism in plant cell,
3) The gene and environmental regulating model and their relationships in various key enzymes of nitrogen metabolism,
4) The function of main cell organs and their responses to environmental factors in nitrogen metabolism process,
5) Physiological and chemical mechanism of nitrogen and the relationship between the mechanism and protein formation during crop grain filling,
6) Improving gene structure of special species or cultivars using gene engineering methods to enhance the resistance to environmental factor stress and the efficiency of absorption and transportation of nitrogen, and
7) The mechanism of natural nitrogen cycle and its response to human activity disturbance.

8.11 Disorders of amino acid metabolism

Twenty amino acids, including nine that cannot be synthesized in humans and must be obtained through food, are involved in metabolism. Amino acids are the building blocks of proteins; some also function as or are synthesized into important molecules in the body such as neurotransmitters, hormones, pigments, and oxygen-carrying molecules. Each amino acid is further broken down into ammonia, carbon dioxide, and water. Disorders that affect the metabolism of amino acids include phenylketonuria, tyrosinemia, homocystinuria, non-ketotic

hyperglycinemia, and maple syrup urine disease. These disorders are autosomal recessive, and all may be diagnosed by analyzing amino acid concentrations in body fluids. (Maple syrup urine disease also features the production of organic acids and is discussed in the section Organic acidemias.)

Phenylketonuria (PKU) is caused by decreased activity of phenylalanine hydroxylase (PAH), an enzyme that converts the amino acid phenylalanine to tyrosine, a precursor of several important hormones and skin, hair, and eye pigments. Decreased PAH activity results in accumulation of phenylalanine and a decreased amount of tyrosine and other metabolites. Persistent high levels of phenylalanine in the blood in turn result in progressive developmental delay, a small head circumference, behaviour disturbances, and seizures. Due to a decreased amount of the pigment melanin, persons with PKU tend to have lighter features, such as blond hair and blue eyes, than other family members who do not have the disease. Treatment with special formulas and with foods low in phenylalanine and protein can reduce phenylalanine levels to normal and maintain normal intelligence. However, rare cases of PKU that result from impaired metabolism of biopterin, an essential cofactor in the phenylalanine hydroxylase reaction, may not consistently respond to therapy.

812 Uremia

Uremia is a buildup of toxins in your blood. It occurs when the kidneys stop filtering toxins out through your urine. Uremia is often a sign of end-stage renal (kidney) disease. Treatments include medication, dialysis and kidney transplant surgery. Left untreated, uremia can lead to serious health problems or death.

8.13 What is uremia?

Uremia is a dangerous condition that occurs when waste products associated with decreased kidney function build up in your blood. Uremia means “urine in the blood” and refers to the effects of the waste product accumulation. It affects the entire body. Uremia most often occurs due to chronic kidney disease (CKD) that may lead to end-stage renal (kidney) disease (ESKD), but can also occur quickly leading to acute kidney injury and failure (AKI) that is potentially reversible. Uremia may cause serious health complications such as fluid accumulation, electrolyte, hormone and metabolic problems. Left untreated, uremia is usually fatal and was always so before dialysis and transplantation were available.

8.14 Who is at risk for uremia?

People with CKD are at the highest risk for uremia. CKD may be due to disease within the kidneys or more generalized disease. In the United States CKD is most often a result of:

- Diabetes mellitus.
- High blood pressure.
- Glomerulonephritis (GN) (damage to the filters in the kidneys).
- Polycystic kidney disease (PKD) (caused by cysts, or fluid-filled sacs, in or around the kidneys).

8.15 Symptoms and causes

8.16 What causes uremia?

Healthy kidneys filter waste and fluids from your body through the urine. Kidneys help maintain normal levels of acids, electrolytes and hormones such as Vitamin D and erythropoietin (EPO). Damaged kidneys don't work well, allowing multiple toxins to build up in your blood. Most people feel sick when kidney function is less than 15% (15 ml/min) of normal and need to start dialysis when function is less than 10% (10 ml/min) of normal. Lab studies monitor chemicals that are elevated in uremia but not necessarily causative:

- Creatinine (waste product produced in muscle and in dietary protein).
- Urea (waste product formed in the liver as protein is broken down).
- Estimation of overall kidney function using a formula (eGFR - ml/min).
- Stage of CKD from 1 to 5 based on eGFR data from the National Kidney Foundation with dialysis usually starting at Stage 5.

8.17 What are the symptoms of uremia?

Symptoms of uremia include:

- Cognitive dysfunction (problems with thinking and remembering).
- Fatigue.
- Shortness of breath from fluid accumulation.
- Loss of appetite.
- Muscle cramps.
- Nausea and vomiting.
- Itching.
- Unexplained weight loss.

In very severe instances, symptoms may include uremic fetor (a urine-like odor on the breath or metallic taste in the mouth) and uremic frost (yellow-white crystals on the skin due to urea in sweat).

8.18 What are the complications of uremia?

Uremia can cause serious complications if it's not treated. Your body may accumulate excess acid, or hormone and electrolyte imbalances –especially for potassium - that can affect the heart. These problems can affect your metabolism, or your body's process of converting food to energy. The buildup of toxins in your blood can also cause blood vessels to calcify (harden). Calcification leads to bone, muscle, and heart and blood vessel problems. Other complications of uremia may include:

- Acidosis (too much acid in your blood).
- Anemia (too few healthy red blood cells).
- High blood pressure.
- Hyperkalemia (too much potassium in your blood).
- Hyperparathyroidism (too much calcium and phosphorus in your blood leading to elevated parathyroid hormone levels and bone abnormalities).
- Hypothyroidism (underactive thyroid).
- Infertility (inability to get pregnant).
- Malnutrition (lack of nutrients in your body).

Additional complications of uremia may include:

- Pulmonary edema (fluid in your lungs).
- Defective platelet function and blood clotting leading to bleeding
- Uremic encephalopathy (decreased brain function due to toxin buildup).
- Angina (chest pain).
- Atherosclerosis (hardened arteries).
- Heart failure.
- Heart valve disease.
- Pericardial effusion (fluid around your heart).
- Stroke.

8.19 Diagnosis and tests

How is uremia diagnosed?

Your healthcare provider:

- Evaluates your symptoms.

- Performs a physical exam.
- Reviews your health history, especially your kidney health and your family history.
- Reviews the lab tests mentioned above. Creatinine and BUN blood tests help your provider confirm a diagnosis of uremia. These tests check your blood for high levels of waste products. They also used to estimate your glomerular filtration rate (eGFR). This rate measures your kidney function.

A kidney ultrasound checks the shape and size of your kidneys and looks for scarring. An ultrasound can also detect kidney blockages, such as kidney stones, or injuries. Additional tests may be needed in certain situations

8.20 Management and treatment

8.20.1 How is uremia treated?

Dialysis (a procedure to clean the blood) is the most common treatment for uremia. There are two kinds of dialysis. Hemodialysis uses a machine to filter blood outside the body. Peritoneal dialysis uses the lining of your belly and a special fluid to filter blood.

You may need a kidney transplant if uremia is the result of end-stage renal (kidney) failure. A transplant replaces the failing kidney with a donor kidney from either a living or deceased donor.

Your healthcare provider may recommend iron supplementation for anemia, replacement of EPO, calcium and Vitamin D supplements, phosphorus binders taken with meals to prevent bone loss due to hyperparathyroidism. Blood pressure needs to be controlled and any risks for heart disease need to be addressed. Other underlying medical problems must also be managed.

8.21 Are there foods or medications I should avoid?

Talk to your healthcare provider or dietician before changing your diet or taking medication or supplements. Certain medications need to be adjusted or avoided and your healthcare provider can assist you in making safe choices. A generally good diet with low sodium and potassium intake is most often used. People with uremia need to be careful about their intake of potassium, phosphate, sodium and protein.

8.22 Prevention

8.23 How can I prevent uremia?

People with end-stage kidney disease should have regular dialysis to keep toxins under control. If you have chronic kidney disease, you may be able to prevent or delay disease progression by:

- Controlling your blood pressure, diabetes or other medical problems.
- Taking your prescribed medications.
- Avoiding medications that may further damage your kidneys.
- Eating a heart healthy diet.
- Exercising.
- Maintaining a healthy weight.
- Quitting smoking.

8.24 What is the outlook for people with uremia?

Uremia usually requires dialysis and careful monitoring. Severe uremia can lead to coma or death. The most common health complication in people with uremia is heart disease.

8.25 When should I contact my healthcare provider about uremia?

Uremia can be a medical emergency. Seek help if you or someone else with kidney issues shows signs of:

- Abnormal behavior
- Chest pain.
- Cognitive dysfunction.
- Confusion.
- Difficulty breathing.
- Disorientation (not knowing where you are).
- Drowsiness.
- Extreme fatigue.
- Nausea and vomiting.

8.26 Hyperuricemia:

8.26.1 Symptoms and treatment

8.26.2 Is hyperuricemia common?

Hyperuricemia occurs when there's too much uric acid in your blood. High uric acid levels can lead to several diseases, including a painful type of arthritis called gout. Elevated uric acid levels are also associated with health conditions such as heart disease, diabetes, and kidney disease. Rates of hyperuricemia have risen sharply since 1960. The most recent significant study of hyperuricemia and gout found that 43.3 millionTrusted Source Americans have the condition.

8.27 Why hyperuricemia occurs

Uric acid is formed when purines break down in your body. Purines are chemicals found in certain foods. This typically includes:

- Red meat
- Organ meat
- Seafood
- Beans

Normally, your body rids itself of uric acid when you urinate. Hyperuricemia occurs when your body either makes too much uric acid or is unable to excrete enough of it. It usually happens because your kidneys aren't eliminating it quickly enough. Excess uric acid levels in your blood can lead to the formation of crystals. Although these can form anywhere in the body, they tend to form in and around your joints and in your kidneys. Your body's defensive white blood cells may attack the crystals, causing inflammation and pain.

8.28 Hyperuricemia symptoms

Only about one-third of people with hyperuricemia experience symptoms. This is known as asymptomatic hyperuricemia. Although hyperuricemia isn't a disease, if uric acid levels remain high, over time they can lead to several diseases.

8.28.1 Gout

Gout, sometimes called gouty arthritis, occurs in about 20 percent of people with hyperuricemia. A rapid drop in uric acid levels can also trigger gout. Gout can appear as isolated attacks, or flares. Some people experience chronic gout, which involves a number attacks occurring over short periods of time. Gout can affect any joint in your body, but flares often first appear in your large toe. Feet, ankles, knees, and elbows are also common sites of gout. Gout attacks tend to occur suddenly, often at night. The attacks peak in intensity in about 12 to 14 hours. Even untreated, attacks of gout usually subside within two weeks.

Symptoms of gout may include:

- Severe pain in your joints
- Joint stiffness
- Difficulty moving affected joints
- Redness and swelling
- Misshapen joints

8.28.2 Tophaceous gout

If you've had hyperuricemia for several years, uric acid crystals can form clumps called tophi. These hard lumps are found under your skin, around your joints, and in the curve at the top of your ear. Tophi can worsen joint pain and over time damage your joints or compress your nerves. They're often visible to the eye and can become disfiguring.

8.28.3 Kidney stones

Uric acid crystals can cause a buildup of stones in your kidneys. Often, the stones are small and are passed in your urine. Sometimes, they can become too large to pass and block parts of your urinary tract. Symptoms of kidney stones include:

- Pain or aching in your lower back, side, abdomen, or groin
- Nausea
- Increased urge to urinate
- Pain when urinating
- Difficulty urinating
- Blood in your urine
- Foul-smelling urine

If you also have a kidney infection, you may experience fever or chills. This buildup of urine is an ideal breeding zone for bacteria. As a result, urinary tract infections are common when you have kidney stones.

8.28.4 Who is at risk for hyperuricemia

Anyone can have hyperuricemia, but it's more common in men than women and your risk increases with age. You're also more likely to get it if you are of Pacific Island heritage or African-American. Several risk factors are associated with hyperuricemia:

- Alcohol use
- Some medications, particularly medications for heart disease
- Lead exposure
- Pesticide exposure
- Kidney disease
- High blood pressure
- High blood glucose levels
- Hypothyroidism
- Obesity

- Extreme levels of physical activity

8.29 Porphyria

Porphyria is a group of liver disorders in which substances called porphyrins build up in the body, negatively affecting the skin or nervous system. The types that affect the nervous system are also known as acute porphyria, as symptoms are rapid in onset and short in duration. Symptoms of an attack include abdominal pain, chest pain, vomiting, confusion, constipation, fever, high blood pressure, and high heart rate. The attacks usually last for days to weeks. Complications may include paralysis, low blood sodium levels, and seizures. Attacks may be triggered by alcohol, smoking, hormonal changes, fasting, stress, or certain medications. If the skin is affected, blisters or itching may occur with sunlight exposure.

8.30 What are porphyrias?

Porphyrias are rare disorders that mainly affect the skin or nervous system. These disorders are usually inherited, meaning they are caused by gene mutations *NIH external link* passed from parents to children. If you have porphyria, cells fail to change chemicals in your body—called porphyrins and porphyrin precursors—into heme, the substance that gives blood its red color. When these chemicals build up in your body, they cause illness. Depending on the type of porphyria you have, porphyrins or porphyrin precursors may build up in the liver or the bone marrow. Bone marrow is the spongy tissue inside most of your bones.

Most types of porphyria are inherited from one or both of a person's parents and are due to a mutation in one of the genes that make heme. They may be inherited in an autosomal dominant, autosomal recessive, or X-linked dominant manner. One type, *porphyria cutanea tarda*, may also be due to hemochromatosis (increased iron in the liver), hepatitis C, alcohol, or HIV/AIDS. The underlying mechanism results in a decrease in the amount of heme produced and a build-up of substances involved in making heme. Porphyrias may also be classified by whether the liver or bone marrow is affected. Diagnosis is typically made by blood, urine, and stool tests. Genetic testing may be done to determine the specific mutation.

Treatment depends on the type of porphyria and the person's symptoms. Treatment of porphyria of the skin generally involves the avoidance of sunlight, while treatment for acute porphyria may involve giving intravenous heme or a glucose solution. Rarely, a liver transplant may be carried out. The precise prevalence of porphyria is unclear, but it is estimated to affect between 1 and 100 per 50,000 people. Rates are different around the

world. Porphyria cutanea tarda is believed to be the most common type. The disease was described as early as 370 BC by Hippocrates. The underlying mechanism was first described by German physiologist and chemist Felix Hoppe-Seyler in 1871.

8.31 What are the types of porphyria?

Experts often divide porphyrias into two groups—acute porphyrias and cutaneous porphyrias—based on whether they primarily affect the nervous system or the skin.

8.32 Acute porphyrias

Four types of acute porphyrias affect the nervous system. Two of those types can also affect the skin. Symptoms for acute porphyrias develop over hours or days and last for days or weeks.

8.33 Types of acute porphyria

Type of Acute Porphyria	Parts of the Body Affected	Where Porphyrins or Porphyrin Precursors Build Up
acute intermittent porphyria	nervous system	liver
variegate porphyria	nervous system and skin	liver
hereditary coproporphyria	nervous system and skin	liver
delta-aminolevulinic acid (ALA) dehydratase deficiency porphyria	nervous system	liver

8.34 Cutaneous porphyrias

Four types of cutaneous porphyrias affect only the skin and cause chronic, or long lasting, symptoms. People with cutaneous porphyria may develop skin symptoms—such as blistering or pain—after their skin is exposed to sunlight.

8.35 Types of cuteaneous porphyria

Type of Cutaneous Porphyria	**Parts of the Body Affected**	**Where Porphyrins Build Up**
porphyria cutanea tarda	skin	liver
protoporphyrias: erythropoietic protoporphyria and x-linked protoporphyria	skin	bone marrow

Type of Cutaneous Porphyria	Parts of the Body Affected	Where Porphyrins Build Up
congenital erythropoietic porphyria	skin	bone marrow
hepatoerythropoietic porphyria	skin	liver

8.36 How common are porphyrias?

Porphyrias are rare diseases. Studies suggest that all types of porphyrias combined affect fewer than 200,000 people in the United States. The most common type of acute porphyria is acute intermittent porphyria. The most common type of cutaneous porphyria—and the most common type of porphyria overall—is porphyria cutanea tarda, which affects about 5 to 10 out of every 100,000 people. The most common type of porphyria in children is a cutaneous porphyria called erythropoietic protoporphyria.

8.37 Who is more likely to get porphyria?

Acute porphyria is more common in females than in males and often begins when people are between the ages of 15 and 45. Among types of cutaneous porphyria, porphyria cutanea tarda most often develops in people older than age 40, usually men. For other types of cutaneous porphyria, symptoms often appear in early childhood.

8.38 What are the complications of porphyrias?

Different types of porphyrias may lead to different complications.

8.39 Liver problems

Several types of porphyrias can cause liver problems. Acute porphyria increases the chance of developing liver cancer. Porphyria cutanea tarda can damage the liver and increase the chance of developing cirrhosis and liver cancer. Some people with protoporphyria also develop liver damage and cirrhosis, and up to 5 percent of people with protoporphyria develop liver failure. In people with protoporphyria, bile carries extra porphyrins from the liver to the gallbladder, which may lead to gallstones that are made of porphyrins.

8.40 Anemia

Two types of cutaneous porphyria, congenital erythropoietic porphyria and, less commonly, hepatoerythropoietic porphyria, may cause severe anemia. These diseases may also cause the spleen to become enlarged, which can make anemia worse.

8.41 High blood pressure and kidney problems

People with acute porphyria have an increased chance of developing high blood pressure and chronic kidney disease, which can lead to kidney failure.

8.42 What are the symptoms of porphyrias?

8.42.1 Acute porphyrias

Symptoms of acute porphyria can be mild or severe, lasting days or weeks. Times when symptoms occur are called attacks. Without early treatment, symptoms of an attack may become more severe and even life-threatening. Symptoms may include

- Pain in the abdomen, back, or arms and legs
- Digestive symptoms, such as constipation, nausea, and vomiting
- Mental changes, such as anxiety, confusion, hallucinations, and seizures.
- Problems with nerves that control movement, which may cause muscle weakness, paralysis and breathing problems
- Urinary symptoms, such as dark or reddish-brown urine, urinary retention, or incontinence
- Skin blisters when skin is exposed to sunlight, for people with variegate porphyria or hereditary coproporphyria

Symptoms of acute porphyria may include severe pain in the abdomen that lasts for hours to days. Most people with acute porphyria only have one or a few attacks throughout their lives. Among people diagnosed with acute porphyria after one attack, about 3 to 5 percent will have four or more attacks in a year. Factors that may increase the chance of getting acute porphyria attacks or make attacks worse include female sex hormones, especially progesterone; certain medicines; a lowered intake of carbohydrates; drinking alcohol; and smoking.

8.42.2 Cutaneous porphyrias

In people with porphyria cutanea tarda, congenital erythropoietic porphyria, or hepato erythropoietic porphyria, areas of skin exposed to sunlight may develop symptoms such as

- Blisters
- Fragile skin that is easily wounded and slow to heal
- Infection of blisters or wounds
- Scarring or changes in skin color

Protoporphyrias—erythropoietic protoporphyria and x-linked protoporphyria—typically do not cause blisters. Instead, skin exposed to sunlight may develop symptoms such as

- Pain, burning, stinging, or tingling
- Redness
- Swelling

8.43 What causes porphyrias?

Most types of porphyrias are caused by gene mutations. Some types of porphyrias result from inheriting a gene mutation from one parent, while other types result from inheriting two gene mutations, one from each parent. Many people with gene mutations for acute porphyrias never develop the disease. In people who have these gene mutations, factors that increase the chance of developing acute porphyria attacks or make attacks worse include

- Female sex hormones, especially progesterone.
- Some medicines, including hormonal types of birth control and certain types of antibiotics anesthetics and anticonvulsants—medicines designed to treat seizures.
- Lowered intake of carbohydrates, due to fasting, dieting, illness, or bariatric surgery.
- Drinking alcohol, especially binge drinking, which the National Institute on Alcohol Abuse and Alcoholism defines as having four or more drinks within about 2 hours for women and having five or more drinks within about 2 hours for men.
- Smoking

The most common type of porphyria, porphyria cutanea tarda, is most often acquired, meaning that factors other than inherited genes may cause this condition. These factors may include

- A buildup of iron in the body, which may be caused by gene mutations that can lead to hemochromatosis
- Heavy alcohol drinking, which the National Institute on Alcohol Abuse and Alcoholism defines as more than 14 drinks per week for men and more than 7 drinks per week for women.
- Smoking
- Viral infections, such as hepatitis C and HIV infections.
- Taking estrogen, which may be found in medicines such as birth control pills and hormonal replacement therapy.

In some cases, an inherited gene mutation plays a role in causing porphyria cutanea tarda, along with one or more of the factors listed above.

8.44 How do doctors diagnose porphyrias?

The doctor will ask about your medical history and symptoms and perform a physical exam. If a doctor suspects you may have porphyria, he or she will order tests to diagnose the disease.

8.45 Tests for porphyria

Tests for porphyria measure the amounts of porphyrins and porphyrin precursors in your blood, urine, or stool and are used to detect porphyria and to monitor the disease. Additional testing may be needed to determine what type of porphyria you have.

8.46 Genetic tests

Genetic tests check for the gene mutations that cause porphyrias. The test may help confirm the diagnosis and determine which specific gene mutation you have. If you have a mutation, your doctor may recommend testing for the same mutation in your family members. If you or family members are considering genetic testing, you may want to consider genetic counseling. Genetic counseling can help you and your families understand how test results may affect your lives.

8.47 Factors affecting nitrogen balance

A dietary regimen employing a protein-poor diet (2.7–3 g N/day) supplemented by essential amino acids, given intravenously or orally, changed the nitrogen balance in severe uremic patients from a negative to a positive one. The addition of histidine to the essential amino acids further improved the nitrogen balance. No difference in the nitrogen balance was observed when intravenous (i.v.) and oral administration were compared. ^{15}N studies indicate that protein synthesis takes place preferably in the muscle cells when the amino acids are infused intravenously, whereas oral treatment resulted in preferential synthesis of plasma protein. Insufficient caloric intake, lack of non-essential nitrogen, potassium depletion, corticosteroid administration, infection or cardiac insufficiency have been found to cause a deterioration of the nitrogen balance and an increase of plasma urea or concentration. In preliminary studies, administration of human growth hormone (HGH) to uremic patients maintained on the protein-poor diet was found to further improve the nitrogen balance. In other preliminary experiments, administration of extra tryptophan with the diet amino acids to uremic patients was also found to improve the nitrogen balance.

8.48 Summary

Under this unit we have summarize the nitrogen metabolism in mammals and humans. Degradation of amino acids in microorganisms and plants is a rare event, but is of major importance in animals and humans. In man and mammals, the ammonium ions are converted into urea in the liver. The form in which amino nitrogen is excreted from the organism is dependent upon the adaptation of the organism to its habitat. In terms of amino nitrogen excretion, the animal kingdom can be classified into ammonotelic (ammonia), ureotelic (urea), and uricotelic (uric acid) organisms depending on the nature of the discharged substance. Urea is produced as a nontoxic soluble vehicle for the elimination of nitrogen originating from the catabolism of amino acids. A major function of the liver is urea biosynthesis, but enzymes of the pathway also occur in kidney, skin, brain plus some other cells where their primary purpose is to synthesize arginine. The chapter also discusses the biosynthesis of heterocyclic compounds, the hybridoma technique, and the degradation of heterocyclic compounds.

Nitrogen is a fundamental component of amino acids, which are the molecular building blocks of protein. Therefore, measuring nitrogen inputs and losses can be used to study protein metabolism. Positive nitrogen balance is associated with periods of growth, hypothyroidism, tissue repair, and pregnancy. This means that the intake of nitrogen into the body is greater than the loss of nitrogen from the body, so there is an increase in the total body pool of protein.

Negative nitrogen balance is associated with burns, serious tissue injuries, fevers, hyperthyroidism, wasting diseases, and during periods of fasting. This means that the amount of nitrogen excreted from the body is greater than the amount of nitrogen ingested. A negative nitrogen balance can be used as part of a clinical evaluation of malnutrition.

Nitrogen balance is the traditional method of determining dietary protein requirements. Determining dietary protein requirements using nitrogen balance requires that all nitrogen inputs and losses are carefully collected, to ensure that all nitrogen exchange is accounted for. In order to control nitrogen inputs and losses, nitrogen balance studies usually require participants to eat very specific diets (so total nitrogen intake is known) and stay in the study location for the duration of the study (to collect all nitrogen losses). Because of these conditions, it can be difficult to study the dietary protein requirements of certain populations using the nitrogen balance technique (e.g. children).

8.49 Terminal questions

Q.24. What do you mean by nitrogen cycle? Describe it.

Answer:----------

Q.25. What do you mean by uremia?

Answer:----------

Q.26. Describe the mechanism of nitrogen metabolism.

Answer:----------

Q.27. What do you mean by hyperuricemia?

Answer:----------

Q.28. What are the factors which affect nitrogen balance?

Answer:----------

Q.29. Write a short note on porphyria.

Answer:----------

8.38 What are the complications of porphyrias?

Answer:----------

Further readings

16. Biochemistry- Lehninger A.L.

17. Biochemistry –J.H.Weil.

18. Biochemistry fourth edition-David Hames and Nigel Hooper.

19. Textbook of Biochemistry for Undergraduates - Rafi, M.D.

20. Biochemistry and molecular biology- Wilson Walker.

Unit-9

9.1 Introduction

Blood clots are gel-like collections of blood that form in your veins or arteries when blood changes from liquid to partially solid. Clotting is a normal function that stops your body from bleeding too much when you get hurt. However, blood clots that form in some places and don't dissolve on their own can be dangerous to your health. Normally, a blood clots start as a response to injury of a blood vessel. At first, the blood stays in one place. Two substances — platelets (a type of blood cell) and fibrin (a firm string-like substance) — combine to form what is called a platelet plug to stop up the cut or hole.

When a blood clot forms where it should not have developed, it is called a thrombus. A blood clot is also called a thrombus. The clot may stay in one spot (called thrombosis) or move through the body (called embolism or thromboembolism). The clots that move are especially dangerous. Blood clots can form in arteries (arterial clots) or veins (venous clots). The symptoms of a blood clot, and the recommended treatment, depend on where a clot forms in your body and how much damage it could cause. Knowing the most common blood clot signs and risk factors can help you spot or even prevent this potentially life-threatening condition.

9.1 Which blood clots pose the most health risk?

Any blood clots that form in arteries (arterial clots) or veins (venous clots) can be serious. You should call your healthcare provider immediately if you suspect a blood clot. A clot that forms in one of your body's larger veins is called a deep vein thrombosis (DVT). A stationary blood clot, or one that stays in place, may not hurt you. A blood clot that dislodges and begins moving through the bloodstream can be harmful.

One of the most pressing blood clot concerns is when a DVT makes its way to your lungs and gets stuck. This condition, called pulmonary embolism (PE), can stop blood from flowing and the results can be very serious, even fatal. In fact, as many as 100,000 people in the United States die from DVTs and PEs every year. Arterial clots in the brain are called strokes. Clots can form in the heart arteries, causing heart attacks. Blood clots can also form in the abdominal blood vessels, causing pain and/or nausea and vomiting.

Objectives

This is the ninth unit on clinical biochemistry. Under ninth unit we have following objectives. These are as under:

- To know about blood clotting and blood clotting mechanism.
- To know about disorders-hemophilia and thrombocytopenic purpura
- To know about blood groups, antigen, antibodies and anticogulants

9.2 Blood Clots

Blood clots are gel-like collections of blood that form in your veins or arteries when blood changes from liquid to partially solid. Clotting is normal, but clots can be dangerous when they do not dissolve on their own. Treatments range from medications to surgery.

9.3 What is a blood clot?

Blood clots are gel-like collections of blood that form in your veins or arteries when blood changes from liquid to partially solid. Clotting is a normal function that stops your body from bleeding too much when you get hurt. However, blood clots that form in some places and don't dissolve on their own can be dangerous to your health. Normally, a blood clots start as a response to injury of a blood vessel. At first, the blood stays in one place. Two substances — platelets (a type of blood cell) and fibrin (a firm string-like substance) — combine to form what is called a platelet plug to stop up the cut or hole.

When a blood clot forms where it should not have developed, it is called a thrombus. A blood clot is also called a thrombus. The clot may stay in one spot (called thrombosis) or move through the body (called embolism or thromboembolism). The clots that move are especially dangerous. Blood clots can form in arteries (arterial clots) or veins (venous clots). The symptoms of a blood clot, and the recommended treatment, depend on where a clot forms in your body and how much damage it could cause. Knowing the most common blood clot signs and risk factors can help you spot or even prevent this potentially life-threatening condition.

9.4 Which blood clots pose the most health risk?

Any blood clots that form in arteries (arterial clots) or veins (venous clots) can be serious. You should call your healthcare provider immediately if you suspect a blood clot. A clot that forms in one of your body's larger veins is called a deep vein thrombosis (DVT). A stationary blood clot, or one that stays in place, may not hurt you. A blood clot that dislodges and begins moving through the bloodstream can be harmful.

One of the most pressing blood clot concerns is when a DVT makes its way to your lungs and gets stuck. This condition, called pulmonary embolism (PE), can stop blood from flowing and the results can be very serious, even fatal. In fact, as many as 100,000 people in the United States die from DVTs and PEs every year. Arterial clots in the brain are called strokes. Clots can form in the heart arteries, causing heart attacks. Blood clots can also form in the abdominal blood vessels, causing pain and/or nausea and vomiting. You don't need to be worried about blood clots that you might see during your period causing these kinds of symptoms or effects.

9.5 Who is most at risk for blood clots?

Some risk factors put certain people at higher risk for developing a blood clot. Blood clots become more common as people get older, especially when they are over age 65. Long hospital stays, surgeries and trauma may significantly increase your risk of blood clots. Other factors can increase your risk to a lesser degree. You might be more at risk if you:

- Take birth control pills or hormone replacement therapy.
- Are pregnant.
- Have cancer, or have been treated for cancer.
- Have a family history of blood clots, or a specific condition, such as Factor V Leiden disease, antiphospholipid syndrome or polycythemia vera, that makes clots more likely.
- Have coronavirus disease 2019 (COVID-19).

Some factors are based on lifestyle choices. Risks might be higher if you:

- Are overweight or obese.
- Live a sedentary (or inactive) lifestyle.
- Smoke cigarettes.

9.6 Symptoms and causes

9.1 What are the most common symptoms of a blood clot?

Blood clot symptoms will depend on where a clot forms in your body. Some people may experience no symptoms at all. Blood clots can occur in the:

- **Abdomen:** Blood clots in the belly area can cause pain or nausea and vomiting.
- **Arms or legs:** A blood clot in the leg or arm may feel painful or tender to the touch. Swelling, redness and warmth are other common signs of blood clots.
- **Brain:** Blood clots in the brain (strokes) can cause a range of symptoms, depending which part of the brain they affect. These clots may cause problems speaking or seeing, inability to move or feel one side of your body and sometimes seizure.
- **Heart or lungs:** A blood clot in the heart will cause symptoms of a heart attack such as crushing chest pain, sweating, pain that travels down the left arm, and/or shortness of breath. A blood clot in the lungs can cause chest pain, difficulty breathing, and sometimes can lead to coughing up blood.

9.2 Diagnosis and tests

9.3 How are blood clots diagnosed?

Blood clot symptoms can mimic other health conditions. Doctors use a variety of tests to detect blood clots and/or rule out other causes. If your doctor suspects a blood clot, he or she may recommend:

- Blood tests can, in some cases, be used to rule out a blood clot.
- Ultrasound provides a clear view of your veins and blood flow.
- CT scan of the head, abdomen, or chest, may be used to confirm that you have a blood clot. This imaging test can help rule out other potential causes of your symptoms.
- Magnetic resonance angiography (MRA) is an imaging test similar to a magnetic resonance imaging (MRI) test. An MRA looks specifically at blood vessels.
- **V/Q scans** test circulation of air and blood in the lungs.

9.4 Management and treatment

9.5 How are blood clots treated?

The goal in treating blood clots, especially DVTs, is to prevent the blood clot from getting larger or breaking loose. Treatment can reduce your chances of developing more blood clots in the future. Treatment depends on where the blood clot is and how likely it is to harm you. Your doctor might recommend:

- **Medication:** Anticoagulants, also called blood thinners, help prevent blood clots from forming. For life-threatening blood clots, drugs called thrombolytics can dissolve clots that are already formed.
- **Compression stockings:** These tight-fitting stockings provide pressure to help reduce leg swelling or prevent blood clots from forming.
- **Surgery:** In a catheter-directed thrombolysis procedure, specialists direct a catheter (a long tube) to the blood clot. The catheter delivers medication directly to the clot to help it dissolve. In thrombectomy surgery, doctors use special instruments to carefully remove a blood clot.
- **Stents:** Doctors may decide if a stent is necessary to keep a blood vessel open.
- **Vena cava filters:** In some cases, a person might be unable to take blood thinners, and a filter is put into the inferior vena cava (the body's largest vein) to catch blood clots before they can travel to the lungs.

9.6 Prevention

How can you prevent blood clots?

You can reduce your risk of blood clots by:

- Enjoying regular physical activity.
- Do not smoke.
- Eating a healthy diet and making sure that you stay hydrated.
- Maintaining a healthy weight.
- Controlling medical problems such as high blood pressure and diabetes.

9.7 Mechanism of Blood Coagulation

Blood coagulation or clotting is an important phenomenon to prevent excess loss of blood in case of injury or trauma. The blood stops flowing from a wound in case of injury. The blood clot or 'coagulum' is formed by a network of fibrin threads. In this network, deformed and dead formed elements (erythrocytes, leukocytes and platelets) get trapped. The enzyme thrombin converts fibrinogen present in the plasma to fibrin. It is a cascade process of a series of enzyme catalysed reactions. Fibrinogen and various inactive blood clotting factors are present in the plasma. An injury stimulates platelets or thrombocytes to release various factors that initiate the blood clotting cascade. Calcium ions play an important role in blood coagulation. Let's learn more in detail about the blood coagulation pathway.

9.8 Blood Coagulation Pathway

The process of blood coagulation leads to haemostasis, i.e. prevention of bleeding or haemorrhage. Blood clotting involves activation and aggregation of platelets at the exposed

endothelial cells, followed by deposition and stabilisation of cross-linked fibrin mesh. Primary haemostasis involves platelet aggregation and formation of a plug at the site of injury, and secondary haemostasis involves strengthening and stabilisation of platelet plug by the formation of a network of fibrin threads. The secondary haemostasis involves two coagulation pathways, the intrinsic pathway and the extrinsic pathway. Both pathways merge at a point and lead to the activation of fibrin, and the formation of the fibrin network.

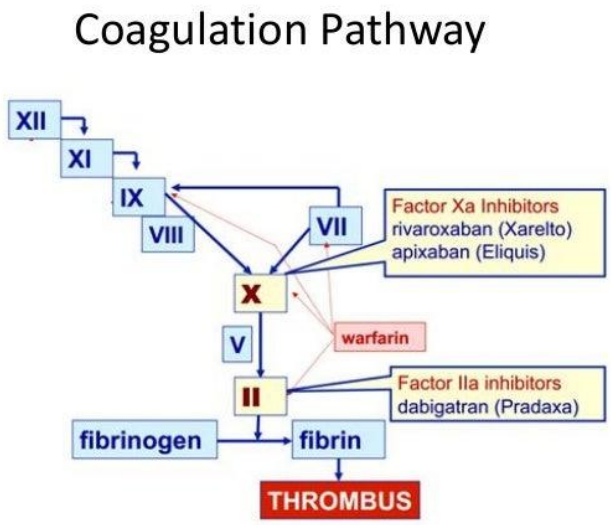

Fig. Coagulation cascade showing the factors involved in clot formation.

9.9 Platelet Activation

The blood circulating in the blood vessel does not clot under normal circumstances. The blood coagulation process is stimulated when there is any damage to the endothelium of blood vessels. It leads to platelet activation and aggregation. When collagen is exposed to the platelets due to injury, the platelets bind to collagen by surface receptors. This adhesion is stimulated by the von Willebrand factor released from endothelial cells and platelets. This forms additional cross-linking and activation of platelet integrins, which facilitate tight binding and aggregation of platelets at the site of injury. This leads to primary haemostasis.

9.10 Blood Coagulation Cascade

The process of coagulation is a cascade of enzyme catalysed reactions wherein the activation of one factor leads to the activation of another factor and so on. The three main steps of the blood coagulation cascade are as follows:

1. Formation of prothrombin activator
2. Conversion of prothrombin to thrombin
3. Conversion of fibrinogen into fibrin

1. Formation of prothrombin activator

The formation of a prothrombin activator is the first step in the blood coagulation cascade of secondary haemostasis. It is done by two pathways, viz. extrinsic pathway and intrinsic pathway.

Extrinsic Pathway

It is also known as the tissue factor pathway. It is a shorter pathway. The tissue factors or tissue thromboplastins are released from the damaged vascular wall. The tissue factor activates the factor VII to VIIa. Then the factor VIIa activates the factor X to Xa in the presence of Ca

Intrinsic Pathway

It is the longer pathway of secondary haemostasis. It begins with the exposure of blood to the collagen from the underlying damaged endothelium. This activates the plasma factor XII to XIIa. XIIa is a serine protease, it activates the factor XI to XIa. The XIa then activates the factor IX to IXa in the presence of Ca^{2+} ions. The factor IXa in the presence of factor VIIIa, Ca^{2+} and phospholipids activate the factor X to Xa.

Common Pathway

The factor Xa, factor V, phospholipids and calcium ions form the prothrombin activator. This is the start of the common pathway of both extrinsic and intrinsic pathways leading to coagulation.

2. Conversion of prothrombin to thrombin

Prothrombin or factor II is a plasma protein and is the inactive form of the enzyme thrombin. Vitamin K is required for the synthesis of prothrombin in the liver. The prothrombin activator formed above converts prothrombin to thrombin. Thrombin is a proteolytic enzyme. It also stimulates its own formation, i.e. the conversion of prothrombin to thrombin. It promotes the formation of a prothrombin activator by activating factors VIII, V and XIII.

3. Conversion of fibrinogen into fibrin

Fibrinogen or factor I is converted to fibrin by thrombin. Thrombin forms fibrin monomers that polymerise to form long fibrin threads. These are stabilised by the factor XIII or fibrin stabilising factor. The fibrin stabilising factor is activated by thrombin to form factor XIIIa. The activated fibrin stabilising factor (XIIIa) forms cross-linking between fibrin threads in the presence of Ca^{2+} and stabilises the fibrin meshwork. The fibrin mesh traps the formed elements to form a solid mass called a clot.

9.11 Blood Clotting Disorders

Haemophilia is the main blood clotting disorder. Haemophilia is characterised by excessive bleeding. It is due to the absence of some of the factors required in the blood clotting cascade. The three main forms of haemophilia are as follows:

- Haemophilia A – Factor VIII deficiency
- Haemophilia B – Factor IX deficiency or "Christmas disease"
- Haemophilia C – Factor XI deficiency

Thrombosis is the formation of a blood clot inside the blood vessel. It blocks the flow of blood. Thrombosis can occur in arteries as well as veins. Arterial thrombosis affects the blood supply and leads to the damage of tissue, i.e. ischemia or necrosis. The clot may sometimes break free and circulate in the body and lead to embolism.

9.12 Hemorrhage

Hemorrhage is bleeding from a damaged blood vessel. Many things can cause hemorrhage inside and outside the body. Types of hemorrhage range from minor, such as a bruise, to major, such as bleeding in the brain. If you can't stop external bleeding or suspect internal bleeding, seek immediate medical attention.

9.13 What is hemorrhage?

Hemorrhage is loss of blood from a damaged blood vessel. The bleeding can be inside or outside the body, and blood loss can be minor or major.

9.14 Possible causes

What are the most common causes of hemorrhage?

There are many possible causes of hemorrhage, including:

- Alcohol, drug or tobacco use that is heavy or long-term (bleeding in the brain).
- Blood clotting disorders.
- Cancer.
- Complications from medical procedures, such as surgery or childbirth.
- Damage to an internal organ.
- Hereditary (inherited) disorders, such as hemophilia and hereditary hemorrhagic telangiectasia.
- Injuries, such as cuts or puncture wounds, bone fracture or traumatic brain injury.
- Violence, such as a gunshot or knife wound, or physical abuse.
- Viruses that attack the blood vessels, such as viral hemorrhagic fever.

Depending on the location or cause, a hemorrhage might be called:

- Bruise or hematoma (a particularly bad bruise). Both involve bleeding just under the skin.
- Hemothorax, blood collecting between the chest wall and lungs.
- Intracranial hemorrhage, bleeding in the brain.
- Nosebleed.
- *Petechiae,* tiny spots on the skin that may be purple, red or brown.
- Postpartum hemorrhage, more bleeding than normal after childbirth.
- Subarachnoid hemorrhage, a type of stroke that can be caused by head trauma.
- Subconjunctival hemorrhage_, _broken blood vessels in the eye.
- Subdural hematoma, blood leaking into the dura mater, the membrane between the brain and skull.

9.15 How might bleeding make me feel?

The way a hemorrhage makes you feel varies a lot, depending on where it is and how severe it is. For example, with a bruise, you may have only mild discomfort compared to head injury. Another example: Hemorrhage in the brain may cause headache, but in the chest it may cause trouble breathing. Serious blood loss may make you feel:

- Cool when someone touches your skin.
- Dizzy.
- Tired.
- Nauseous.
- Short of breath.
- Weak.

If severe hemorrhage is left untreated, you may experience:

- Chest pain.
- Confusion.
- Faster breathing or heart rate.
- Organ failure.
- Seizures.
- Shock.
- Coma or death.

9.16 Care and treatment

9.17 How is bleeding treated?

Treatment for hemorrhage depends on:

- Where it is in the body.
- How serious the hemorrhage is.
- How much blood may have been lost.
- How the person is feeling overall (for example, symptoms or other injuries).

Sometimes, external bleeding can be stopped with first aid:

- Apply pressure to the wound with your hands.
- Find a dressing (clean cloth) and press on the wound.
- Tie a tourniquet near the wound, but toward the heart. You can make a tourniquet from something tied very tightly, such as a stretchy band, cloth or belt.

Seek immediate medical attention for external bleeding that won't stop, or for suspected internal bleeding. It should be treated in an emergency room. Seek immediate medical attention if you or someone else is bleeding externally or may be bleeding internally and:

- Can't breathe normally.
- Coughs or spits up blood.
- Faints.
- Has bleeding that can't be stopped.
- Has severe chest or belly pain.
- Has cold or "clammy" skin.
- Is dizzy, light-headed or confused.

9.18 Hemophilia

Hemophilia is a rare disorder in which the blood doesn't clot in the typical way because it doesn't have enough blood-clotting proteins (clotting factors). If you have hemophilia, you might bleed for a longer time after an injury than you would if your blood clotted properly. Small cuts usually aren't much of a problem. If you have a severe form of the condition, the main concern is bleeding inside your body, especially in your knees, ankles and elbows. Internal bleeding can damage your organs and tissues and be life-threatening. Hemophilia is almost always a genetic disorder. Treatment includes regular replacement of the specific clotting factor that is reduced. Newer therapies that don't contain clotting factors also are being used.

9.19 Symptoms

Signs and symptoms of hemophilia vary, depending on your level of clotting factors. If your clotting-factor level is mildly reduced, you might bleed only after surgery or trauma. If your

deficiency is severe, you can bleed easily for seemingly no reason. Signs and symptoms of spontaneous bleeding include:

- Unexplained and excessive bleeding from cuts or injuries, or after surgery or dental work
- Many large or deep bruises
- Unusual bleeding after vaccinations
- Pain, swelling or tightness in your joints
- Blood in your urine or stool
- Nosebleeds without a known cause
- In infants, unexplained irritability

9.20 Bleeding into the brain

A simple bump on the head can cause bleeding into the brain for some people who have severe hemophilia. This rarely happens, but it's one of the most serious complications that can occur. Signs and symptoms include:

- Painful, prolonged headache
- Repeated vomiting
- Sleepiness or lethargy
- Double vision
- Sudden weakness or clumsiness
- Convulsions or seizures

9. 21When to see a doctor

Seek emergency care if you or your child has:

- Signs or symptoms of bleeding into the brain
- An injury in which the bleeding won't stop
- Swollen joints that are hot to the touch and painful to bend

9.22 Causes

When a person bleeds, the body typically pools blood cells together to form a clot to stop the bleeding. Clotting factors are proteins in the blood that work with cells known as platelets to form clots. Hemophilia occurs when a clotting factor is missing or levels of the clotting factor are low.

9.23 Congenital hemophilia

Hemophilia is usually inherited, meaning a person is born with the disorder (congenital). Congenital hemophilia is classified by the type of clotting factor that's low.

The most common type is hemophilia A, associated with a low level of factor 8 The next most common type is hemophilia B, associated with a low level of factor 9.

9.24 Acquired hemophilia

Some people develop hemophilia with no family history of the disorder. This is called acquired hemophilia.

Acquired hemophilia is a variety of the condition that occurs when a person's immune system attacks clotting factor 8 or 9 in the blood. It can be associated with:

- Pregnancy
- Autoimmune conditions
- Cancer
- Multiple sclerosis
- Drug reactions

9.25 Hemophilia inheritance

In the most common types of hemophilia, the faulty gene is located on the X chromosome. Everyone has two sex chromosomes, one from each parent. Females inherit an X chromosome from the mother and an X chromosome from the father. Males inherit an X chromosome from the mother and a Y chromosome from the father.

This means that hemophilia almost always occurs in boys and is passed from mother to son through one of the mother's genes. Most women with the defective gene are carriers who have no signs or symptoms of hemophilia. But some carriers can have bleeding symptoms if their clotting factors are moderately decreased.

9.26 Risk factors

The biggest risk factor for hemophilia is to have family members who also have the disorder. Males are much more likely to have hemophilia than are females.

9.27 Complications

Complications of hemophilia can include:

- **Deep internal bleeding.** Bleeding that occurs in deep muscle can cause the limbs to swell. The swelling can press on nerves and lead to numbness or pain. Depending on where the bleeding occurs, it could be life-threatening.
- **Bleeding into the throat or neck.** This can affect a person's ability to breathe.
- **Damage to joints.** Internal bleeding can put pressure on the joints, causing severe pain. Left untreated, frequent internal bleeding can cause arthritis or destruction of the joint.

- **Infection.** If the clotting factors used to treat hemophilia come from human blood, there's an increased risk of viral infections such as hepatitis C. Because of donor screening techniques, the risk is low.
- **Adverse reaction to clotting factor treatment.** In some people with severe hemophilia, the immune system has a negative reaction to the clotting factors used to treat bleeding. When this happens, the immune system develops proteins that keep the clotting factors from working, making treatment less effective.

9.28 Thrombotic thrombocytopenic purpura

Thrombotic thrombocytopenic purpura is a rare disorder that causes blood clots (thrombi) to form in small blood vessels throughout the body. These clots can cause serious medical problems if they block vessels and restrict blood flow to organs such as the brain, kidneys, and heart. Complications resulting from these clots can include neurological problems (such as personality changes, headaches, confusion, and slurred speech), fever, abnormal kidney function, abdominal pain, and heart problems. Blood clots normally form to stop blood loss at the sites of blood vessel injury.

9.29 Thrombotic thrombocytopenic purpura (**TTP**) is a blood disorder that results in blood clots forming in small blood vessels throughout the body. This results in a low platelet count, low red blood cells due to their breakdown, and often kidney, heart, and brain dysfunction. Symptoms may include large bruises, fever, weakness, shortness of breath, confusion, and headache. Repeated episodes may occur.

In about half of cases a trigger is identified, while in the remainder the cause remains unknown. Known triggers include bacterial infections, certain medications, autoimmune diseases such as lupus, and pregnancy. The underlying mechanism typically involves antibodies inhibiting the enzyme ADAMTS13. This results in decreased break down of large multimers of von Willebrand factor (vWF) into smaller units. Less commonly TTP is inherited from a person's parents, known as Upshaw–Schulman syndrome, such that ADAMTS13 dysfunction is present from birth. Diagnosis is typically based on symptoms and blood tests. It may be supported by measuring activity of or antibodies against ADAMTS13. With plasma exchange the risk of death has decreased from more than 90% to less than 20%. Immunosuppressants, such as glucocorticoids, and rituximab may also be used. Platelet transfusions are generally not recommended.

In people with thrombotic thrombocytopenic purpura, clots develop even in the absence of apparent injury. Blood clots are formed from clumps of cells called platelets that circulate in the blood and assist with clotting. Because a large number of platelets are used to make clots in people with thrombotic thrombocytopenic purpura, fewer platelets are available in the bloodstream. A reduced level of circulating platelets is known as thrombocytopenia. Thrombocytopenia can lead to small areas of bleeding just under the surface of the skin, resulting in purplish spots called purpura.

This disorder also causes red blood cells to break down (undergo hemolysis) prematurely. As blood squeezes past clots within blood vessels, red blood cells can break apart. A condition called hemolytic anemia occurs when red blood cells are destroyed faster than the body can replace them. This type of anemia leads to paleness, yellowing of the eyes and skin (jaundice), fatigue, shortness of breath, and a rapid heart rate.

There are two major forms of thrombotic thrombocytopenic purpura, an acquired (noninherited) form and a familial (inherited) form. The acquired form usually appears in late childhood or adulthood. Affected individuals may have a single episode of signs and symptoms, or, more commonly, they may experience multiple recurrences over time. The familial form of this disorder is much rarer and typically appears in infancy or early childhood, although it can appear later in life. In people with the familial form, signs and symptoms often recur on a regular basis and may return during times of stress, such as during illness or pregnancy.

9.29 Causes

TTP, as with other microangiopathic hemolytic anemias (MAHAs), is caused by spontaneous aggregation of platelets and activation of coagulation in the small blood vessels. Platelets are consumed in the aggregation process and bind vWF. These platelet-vWF complexes form small blood clots which circulate in the blood vessels and cause shearing of red blood cells, resulting in their rupture and formation of schistocytes. The two best understood causes of TTP are due to autoimmunity and an inherited deficiency of ADAMTS13. The majority of the remaining cases are secondary to some other factor.

9.30 Autoimmune

TTP of unknown cause was long known as idiopathic TTP but in 1998 the majority of cases were shown to be caused by the inhibition of the enzyme ADAMTS13 by antibodies. The relationship of reduced ADAMTS13 to the pathogenesis of TTP is known as the Furlan-Tsai

hypothesis, after the two independent groups of researchers who published their research in the same issue of the *New England Journal of Medicine*. These cases are now classed as an autoimmune disease and are known as autoimmune TTP (not to be confused with immune/idiopathic thrombocytopenic purpura).

ADAMTS13 is a metalloproteinase responsible for the breakdown of von Willebrand factor (vWF), a protein that links platelets, blood clots, and the blood vessel wall in the process of blood coagulation. Very large vWF multimers are more prone to lead to coagulation. Hence, without proper cleavage of vWF by ADAMTS13, coagulation occurs at a higher rate, especially in the microvasculature, part of the blood vessel system where vWF is most active due to high shear stress. In idiopathic TTP, severely decreased (<5% of normal) ADAMTS13 activity can be detected in most (80%) people, and inhibitors are often found in this subgroup (44–56%).

9.31 Genetic

This condition may also be congenital. Such cases may be caused by mutations in the ADAMTS13 gene.[15] This hereditary form of TTP is called the Upshaw–Schulman syndrome.[16][17][18] People with this inherited ADAMTS13 deficiency have a surprisingly mild phenotype, but develop TTP in clinical situations with increased von Willebrand factor levels, e.g. infection. Reportedly, less than 1% of all TTP cases are due to Upshaw–Schulman syndrome. People with this syndrome generally have 5–10% of normal ADAMTS-13 activity.

9.32 Treatment

Due to the high mortality of untreated TTP, a presumptive diagnosis of TTP is made even when only microangiopathic hemolytic anemia and thrombocytopenia are seen, and therapy is started. Transfusion is contraindicated in thrombotic TTP, as it fuels the coagulopathy. Since the early 1990s, plasmapheresis has become the treatment of choice for TTP. This is an exchange transfusion involving removal of the person's blood plasma through apheresis and replacement with donor plasma (fresh frozen plasma or cryosupernatant); the procedure must be repeated daily to eliminate the inhibitor and abate the symptoms.

If apheresis is not available, fresh frozen plasma can be infused, but the volume that can be given safely is limited due to the danger of fluid overload. Plasma infusion alone is not as beneficial as plasma exchange. Corticosteroids (prednisone or prednisolone) are usually given. Rituximab, a monoclonal antibody aimed at the CD20 molecule on B lymphocytes,

may be used on diagnosis; this is thought to kill the B cells and thereby reduce the production of the inhibitor. A stronger recommendation for rituximab exists where TTP does not respond to corticosteroids and plasmapheresis.

9.33 Blood group

A blood type (also known as a blood group) is a classification of blood, based on the presence and absence of antibodies and inherited antigenic substances on the surface of red blood cells (RBCs). These antigens may be proteins, carbohydrates, glycoproteins, or glycolipids, depending on the blood group system. Some of these antigens are also present on the surface of other types of cells of various tissues. Several of these red blood cell surface antigens can stem from one allele (or an alternative version of a gene) and collectively form a blood group system. Blood types are inherited and represent contributions from both parents of an individual. As of 2021, a total of 43 human blood group systems are recognized by the International Society of Blood Transfusion (ISBT). The two most important blood group systems are ABO and Rh; they determine someone's blood type (A, B, AB, and O, with + or − denoting RhD status) for suitability in blood.

9.34 Blood group systems

A complete blood type would describe each of the 43 blood groups, and an individual's blood type is one of many possible combinations of blood-group antigens. Almost always, an individual has the same blood group for life, but very rarely an individual's blood type changes through addition or suppression of an antigen in infection, malignancy, or autoimmune disease. Another more common cause of blood type change is a bone marrow transplant. Bone-marrow transplants are performed for many leukemias and lymphomas, among other diseases.

If a person receives bone marrow from someone of a different ABO type (e.g., a type A patient receives a type O bone marrow), the patient's blood type should eventually becomes the donor's type, as the patient's hematopoietic stem cells (HSCs) are destroyed, either by ablation of the bone marrow or by the donor's T-cells. Once all the patient's original red blood cells have died, they will have been fully replaced by new cells derived from the donor HSCs. Provided the donor had a different ABO type, the new cells' surface antigens will be different from those on the surface of the patient's original red blood cells.

Some blood types are associated with inheritance of other diseases; for example, the Kell antigen is sometimes associated with McLeod syndrome. Certain blood types may affect

susceptibility to infections, an example being the resistance to specific malaria species seen in individuals lacking the Duffy antigen. The Duffy antigen, presumably as a result of natural selection, is less common in population groups from areas having a high incidence of malaria.

9.35 ABO blood group system

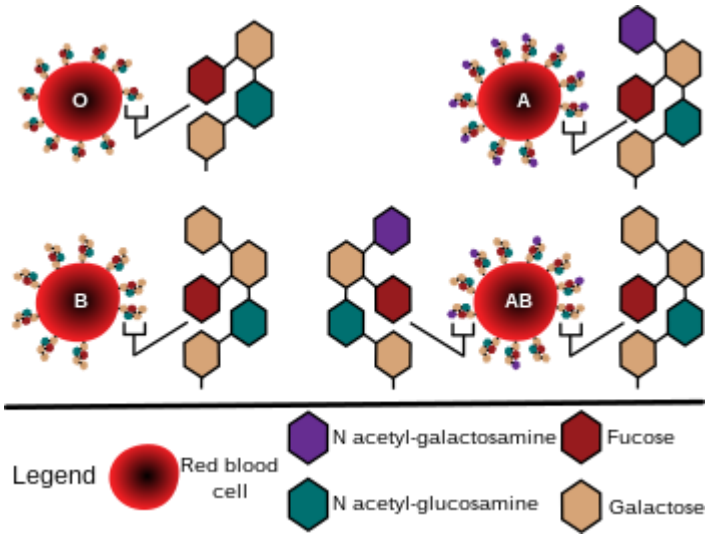

Fig. ABO blood group system: diagram showing the carbohydrate chains that determine the ABO blood group

The ABO blood group system involves two antigens and two antibodies found in human blood. The two antigens are antigen A and antigen B. The two antibodies are antibody A and antibody B. The antigens are present on the red blood cells and the antibodies in the serum. Regarding the antigen property of the blood all human beings can be classified into four groups, those with antigen A (group A), those with antigen B (group B), those with both antigen A and B (group AB) and those with neither antigen (group O). The antibodies present together with the antigens are found as follows:

1. Antigen A with antibody B
2. Antigen B with antibody A
3. Antigen AB with neither antibody A nor B
4. Antigen null (group O) with both antibody A and B

There is an agglutination reaction between similar antigen and antibody (for example, antigen A agglutinates the antibody A and antigen B agglutinates the antibody B). Thus, transfusion can be considered safe as long as the serum of the recipient does not contain antibodies for the blood cell antigens of the donor. The ABO system is the most important blood-group system in human-blood transfusion. The associated anti-A and anti-B antibodies are

usually immunoglobulin M, abbreviated IgM, antibodies. It has been hypothesized that ABO IgM antibodies are produced in the first years of life by sensitization to environmental substances such as food, bacteria, and viruses, although blood group compatibility rules are applied to newborn and infants as a matter of practice. The original terminology used by Karl Landsteiner in 1901 for the classification was A/B/C; in later publications "C" became "O". Type O is often called *0* (*zero*, or *null*) in other languages.

9.36 Rh blood group system

The Rh system (Rh meaning *Rhesus*) is the second most significant blood-group system in human-blood transfusion with currently 50 antigens. The most significant Rh antigen is the D antigen, because it is the most likely to provoke an immune system response of the five main Rh antigens. It is common for D-negative individuals not to have any anti-D IgG or IgM antibodies, because anti-D antibodies are not usually produced by sensitization against environmental substances. However, D-negative individuals can produce IgG anti-D antibodies following a sensitizing event: possibly a fetomaternal transfusion of blood from a fetus in pregnancy or occasionally a blood transfusion with D positive RBCs. Rh disease can develop in these cases. Rh negative blood types are much less common in Asian populations (0.3%) than they are in European populations (15%). The presence or absence of the Rh (D) antigen is signified by the + or − sign, so that, for example, the A− group is ABO type A and does not have the Rh (D) antigen.

9.37 What is an antigen?

In general, antigens are composed of proteins, peptides, and polysaccharides. Any portion of bacteria or viruses, such as surface protein, coat, capsule, toxins, and cell wall, can serve as antigens. Moreover, a combination of lipid or nucleic acid with proteins or polysaccharides can form more complex antigens, such as lipopolysaccharides. Lipopolysaccharides are major ingredients of endotoxins produced by gram-negative bacteria. An antigen contains distinct sites on its surface, which is called an epitope or antigenic determinant. Antibodies generated against an antigen recognize and interact with specific epitopes via antigen-binding sites (paratopes) to trigger immune responses.

9.38 What are the types of antigens?

Antigens are mainly categorized based on their origins. For example, antigens that enter the body from outside via ingestion, inhalation, or injection are termed as exogenous antigens. These include pathogens, chemicals, toxins, allergens, pollens, etc. Autoantigens or self-antigens are normal cellular proteins or a complex of proteins that are mistakenly attacked by

the immune system, leading to autoimmune diseases. A normal self-protein becomes a self-antigen because of impaired immunological tolerance, which can be caused by genetic or environmental factors.

Tumor antigens are produced due to tumor-specific mutations that occur during the neoplastic transformation of normal cells into cancerous cells. These antigens are expressed on the cancer cell surface to be recognized by the immune system. However, despite expressing cell surface antigens, the majority of cancer cells gain the ability to escape immune system-mediated elimination.

9.39 How do antigens trigger an immune response?

The specificity of the immune response depends on the epitope – paratope interaction. An epitope can be of two types: conformational (discontinuous amino acid sequence of the antigen) and linear epitopes (continuous amino acid sequence of the antigen). Upon entering the body, an antigen triggers the adaptive immune system that comprises specialized immune cells, such as B and T lymphocytes (B cells and T cells). There are two types of adaptive immune responses: antibody-mediated and cell-mediated immune responses. The antibody-mediated immunity is triggered when antibodies expressed on the B cell surface recognize specific epitopes of an antigen and subsequently internalize the antigen. The antigen is then presented on the B cell surface to be recognized by helper T cells, which subsequently activate the B cell. Activated B cells rapidly divide to produce two types of cells:

1) Plasma cells that produce antigen-specific antibodies, and

2) Memory B cells that store antigen-specific information for future protection. In the cell-mediated immune system, antigen-presenting cells, such as dendritic cells, macrophages, and B cells, internalize and digest the antigen, and subsequently present the antigenic fragments on their cell surface through major histocompatibility complex (MHC).

9.40 What is an Antibody (Ab)?

An antibody is simply the component produced by the immune system in response to antigens. So basically antigens are the generator of antibodies. They interact with each other to induce an immune response.

– Also called immunoglobulins **(**Ig**)**

– Y-shaped

– Glycoproteins

– Produced by plasma B-cells

– Antigen binding site is called paratope**.**

– Types: IgG**,** IgA**,** IgM**,** IgE**,** IgD (Pneumonics: "GAMED**")**

A**ntibody** (**Ab**), also known as an immunoglobulin (Ig), is a large, Y-shaped protein used by the immune system to identify and neutralize foreign objects such as pathogenic bacteria and viruses. The antibody recognizes a unique molecule of the pathogen, called an antigen. Each tip of the "Y" of an antibody contains a paratope (analogous to a lock) that is specific for one particular epitope (analogous to a key) on an antigen, allowing these two structures to bind together with precision. Using this binding mechanism, an antibody can *tag* a microbe or an infected cell for attack by other parts of the immune system, or can neutralize it directly (for example, by blocking a part of a virus that is essential for its invasion).

To allow the immune system to recognize millions of different antigens, the antigen-binding sites at both tips of the antibody come in an equally wide variety. In contrast, the remainder of the antibody is relatively constant. It only occurs in a few variants, which define the antibody's *class* or *isotype*: IgA, IgD, IgE, IgG, or IgM. The constant region at the trunk of the antibody includes sites involved in interactions with other components of the immune system. The class hence determines the function triggered by an antibody after binding to an antigen, in addition to some structural features. Antibodies from different classes also differ in where they are released in the body and at what stage of an immune response.

Together with B and T cells, antibodies comprise the most important part of the adaptive immune system. They occur in two forms: one that is attached to a B cell, and the other, a soluble form, that is unattached and found in extracellular fluids such as blood plasma. Initially, all antibodies are of the first form, attached to the surface of a B cell – these are then referred to as B-cell receptors (BCR). After an antigen binds to a BCR, the B cell activates to proliferate and differentiate into either plasma cells, which secrete soluble antibodies with the same paratope, or memory B cells, which survive in the body to enable long-lasting immunity to the antigen. Soluble antibodies are released into the blood and tissue fluids, as well as many secretions. Because these fluids were traditionally known as humors, antibody-mediated immunity is sometimes known as, or considered a part of, humoral immunity. The soluble Y-shaped units can occur individually as monomers, or in complexes of two to five units.

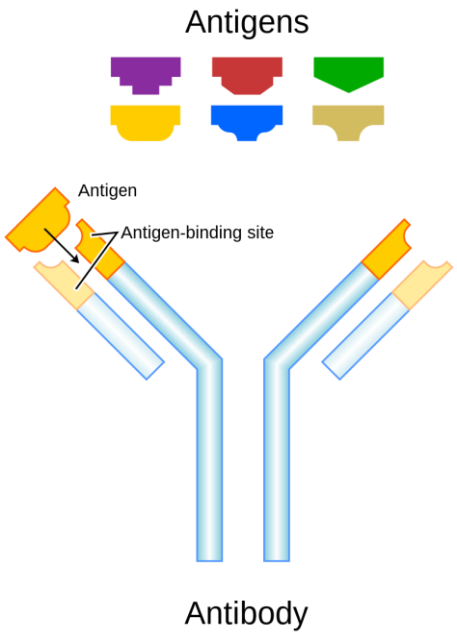

Fig. Each antibody binds to a specific antigen; an interaction similar to a lock and key.

Antibodies are glycoproteins belonging to the immunoglobulin superfamily. The terms antibody and immunoglobulin are often used interchangeably, though the term 'antibody' is sometimes reserved for the secreted, soluble form, i.e. excluding B-cell receptors.

9.41 Structure

Antibodies are heavy (~150 kDa) proteins of about 10 nm in size, arranged in three globular regions that roughly form a Y shape. In humans and most mammals, an antibody unit consists of four polypeptide chains; two identical *heavy chains* and two identical *light chains* connected by disulfide bonds. Each chain is a series of domains: somewhat similar sequences of about 110 amino acids each. These domains are usually represented in simplified schematics as rectangles. Light chains consist of one variable domain V_L and one constant domain C_L, while heavy chains contain one variable domain V_H and three to four constant domains C_H1, C_H2,.

Structurally an antibody is also partitioned into two antigen-binding fragments (Fab), containing one V_L, V_H, C_L, and C_H1 domain each, as well as the crystallisable fragment (Fc), forming the trunk of the Y shape. In between them is a hinge region of the heavy chains, whose flexibility allows antibodies to bind to pairs of epitopes at various distances, to form complexes (dimers, trimers, etc.), and to bind effector molecules more easily.

9.42 Classes

Antibodies can come in different varieties known as *isotypes* or *classes*. In placental mammals there are five antibody classes known as IgA, IgD, IgE, IgG, and IgM, which are further subdivided into subclasses such as IgA1, IgA2. The prefix "Ig" stands for *immunoglobulin*, while the suffix denotes the type of heavy chain the antibody contains: the heavy chain types α (alpha), γ (gamma), δ (delta), ε (epsilon), μ (mu) give rise to IgA, IgG, IgD, IgE, IgM, respectively. The distinctive features of each class are determined by the part of the heavy chain within the hinge and Fc region.

The classes differ in their biological properties, functional locations and ability to deal with different antigens, as depicted in the table.[8] For example, IgE antibodies are responsible for an allergic response consisting of histamine release from mast cells, often a sole contributor to asthma (though other pathways exist as do exist symptoms very similar to yet not technically asthma). The antibody's variable region binds to allergic antigen, for example house dust mite particles, while its Fc region (in the ε heavy chains) binds to Fc receptor ε on a mast cell, triggering its degranulation: the release of molecules stored in its granules.

9.43 Antibody–antigen interactions

The antibody's paratope interacts with the antigen's epitope. An antigen usually contains different epitopes along its surface arranged discontinuously, and dominant epitopes on a given antigen are called determinants. Antibody and antigen interact by spatial complementarity (lock and key). The molecular forces involved in the Fab-epitope interaction are weak and non-specific – for example electrostatic forces, hydrogen bonds, hydrophobic interactions, and van der Waals forces. This means binding between antibody and antigen is reversible, and the antibody's affinity towards an antigen is relative rather than absolute. Relatively weak binding also means it is possible for an antibody to cross-react with different antigens of different relative affinities.

9.44 Summary

Under this unit we summarize that the Blood clotting, or coagulation, is an important process that prevents excessive bleeding when a blood vessel is injured. Platelets (a type of blood cell) and proteins in your plasma (the liquid part of blood) work together to stop the bleeding by forming a clot over the injury. Typically, your body will naturally dissolve the blood clot after the injury has healed. Sometimes, however, clots form on the inside of vessels without an obvious injury or do not dissolve naturally. These situations can be dangerous and require accurate diagnosis and appropriate treatment.

Clots can occur in veins or arteries, which are vessels that are part of the body's circulatory system. While both types of vessels help transport blood throughout the body, they each function differently. Veins are low-pressure vessels that carry deoxygenated blood away from the body's organs and back to the heart. An abnormal clot that forms in a vein may restrict the return of blood to the heart and can result in pain and swelling as the blood gathers behind the clot. Deep vein thrombosis (DVT) is a type of clot that forms in a major vein of the leg or, less commonly, in the arms, pelvis, or other large veins in the body. In some cases, a clot in a vein may detach from its point of origin and travel through the heart to the lungs where it becomes wedged, preventing adequate blood flow. This is called a pulmonary (lung) embolism (PE) and can be extremely dangerous.

Hemophilia is usually an inherited bleeding disorder in which the blood does not clot properly. This can lead to spontaneous bleeding as well as bleeding following injuries or surgery. Blood contains many proteins called clotting factors that can help to stop bleeding. People with hemophilia have low levels of either factor VIII (8) or factor IX (9). The severity of hemophilia that a person has is determined by the amount of factor in the blood. The lower the amount of the factor, the more likely it is that bleeding will occur which can lead to serious health problems. middle-aged or elderly people, or young women who have recently given birth or are in the later stages of pregnancy. This condition often resolves with appropriate treatment.

9.45 Terminal questions

Q.30. What do you mean by blood clotting? Describe it.

Answer:----------

Q.31. What is the mechanism of blood clotting?

Answer:----------

Q.32. Describe the thrombocytopenic purpura.

Answer:----------

Q.33. What are the blood groups? Describe it.

Answer:----------

Q.34. What are the differences between antigen and antibodies?

Answer:----------

Q.35. Write a short note on anticoagulants.

Answer:----------

Q.36. Write a short note on antigens.

Answer:----------

Further readings

21. Biochemistry- Lehninger A.L.

22. Biochemistry –J.H.Weil.

23. Biochemistry fourth edition-David Hames and Nigel Hooper.

24. Textbook of Biochemistry for Undergraduates - Rafi, M.D.

25. Biochemistry and molecular biology- Wilson Walker.

Unit-10

10.1 Introduction

Cancer is a group of diseases involving abnormal cell growth with the potential to invade or spread to other parts of the body. These contrast with benign tumors, which do not spread. Possible signs and symptoms include a lump, abnormal bleeding, prolonged cough, unexplained weight loss, and a change in bowel movements. While these symptoms may indicate cancer, they can also have other causes. Over 100 types of cancers affect humans.

Tobacco use is the cause of about 22% of cancer deaths. Another 10% are due to obesity, poor diet, lack of physical activity or excessive drinking of alcohol. Other factors include certain infections, exposure to ionizing radiation, and environmental pollutants. In the developing world, 15% of cancers are due to infections such as *Helicobacter pylori*, hepatitis B, hepatitis C, human papillomavirus infection, Epstein–Barr virus and human immunodeficiency virus (HIV). These factors act, at least partly, by changing the genes of a cell. Typically, many genetic changes are required before cancer develops. Approximately 5–10% of cancers are due to inherited genetic defects. Cancer can be detected by certain signs and symptoms or screening tests. It is then typically further investigated by medical imaging and confirmed by biopsy.

The risk of developing certain cancers can be reduced by not smoking, maintaining a healthy weight, limiting alcohol intake, eating plenty of vegetables, fruits, and whole grains, vaccination against certain infectious diseases, limiting consumption of processed meat and red meat, and limiting exposure to direct sunlight. Early detection through screening is useful for cervical and colorectal cancer. The benefits of screening for breast cancer are controversial. Cancer is often treated with some combination of radiation therapy, surgery, chemotherapy and targeted therapy. Pain and symptom management are an important part of care.[2] Palliative care is particularly important in people with advanced disease. The chance of survival depends on the type of cancer and extent of disease at the start of treatment. In children under 15 at diagnosis, the five-year survival rate in the developed world is on average 80%. For cancer in the United States, the average five-year survival rate is 66%. In 2015, about 90.5 million people worldwide had cancer. In 2019, annual cancer cases grew to 23.6 million people and 10 million deaths worldwide, representing over the previous decade increases of 26% and 21%, respectively.

The most common types of cancer in males are lung cancer, prostate cancer, colorectal cancer, and stomach cancer. In females, the most common types are breast cancer, colorectal cancer, lung cancer, and cervical cancer. If skin cancer other than melanoma were included in total

new cancer cases each year, it would account for around 40% of cases. In children, acute lymphoblastic leukemia and brain tumors are most common, except in Africa, where non-Hodgkin lymphoma occurs more often. In 2012, about 165,000 children under 15 years of age were diagnosed with cancer. The risk of cancer increases significantly with age, and many cancers occur more commonly in developed countries. Rates are increasing as more people live to an old age and as lifestyle changes occur in the developing world. The financial costs of cancer were estimated at 1.16 trillion USD per year as of 2010.

Objectives

This is the tenth unit of clinical biochemistry. Under tenth unit we have following objectives. These are as under:

- To know about cancer and its types
- To know about multiple steps of tumor development
- To know about death and apoptosis
- To discuss carcinogens and cancer therapy

Cancer is a type of disease where cells grow out of control, divide and invade other tissues. In a person without cancer, cell division is under control. In most tissues, healthy cells divide in a controlled way and copy themselves to create new healthy cells. With cancer, this normal cell division goes out of control. Cells change their nature because mutations have occurred in their genes. All the daughter cells of cancer cells are also cancerous. If the abnormal cells do not invade, but just divide and swell up their original tissue, this is not called "cancer". It is called a tumour. Tumours are usually not a threat to life because they can be cut out. However, some tumours occur in places where they cannot be cut out, and they can be fatal. Some brain tumours are of this type.

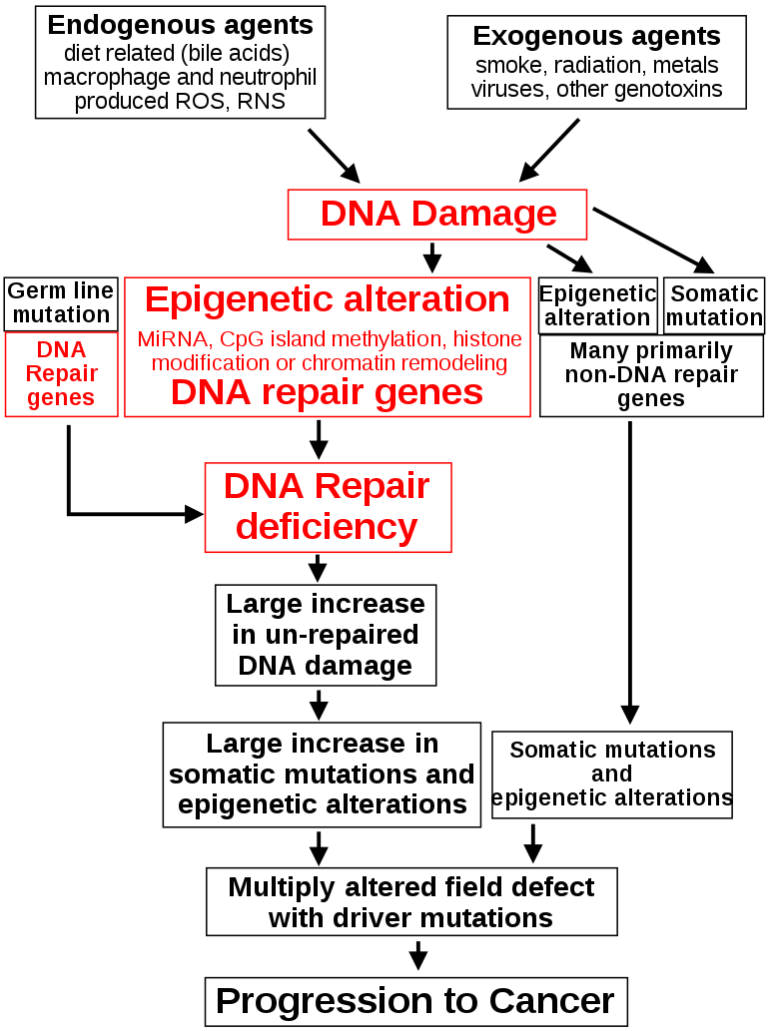

Fig. 2 Progression to cancer

The symptoms of cancer are caused by the cancerous cells invading other tissues. This is called metastasis. Metastasis is how which cancer cells move through the bloodstream or lymphatic system. When this happens, a person's cancer can be spread throughout his or her body. Eventually those other tissues cannot work as well, and the whole body begins to get worse, and may die. Cancer can affect anybody at any age. Most types of cancer are more likely to affect people as they get older. This is because as a person's DNA gets older, their DNA may become damaged, or damage that happened in the past may get worse. One type of cancer that is more common in young men, rather than older people, is testicular cancer (cancer of the testicles). Cancer is one of the biggest and most researched causes of death in developed countries. Studying cancer and its treatment is called oncology.

10.2 Causes

Cancer is one of the most common causes of death around the world. It causes about 12.5% (or one out of every eight) of all deaths worldwide, according to the World Health Organization. Different types of cancer have different causes. Some things are known to cause cancer in a specific body part; other things are known to be able to cause many different types of cancer. For example, using tobacco (smoked or smokeless) can cause many types of cancers, such as lung, mouth, tongue, and throat cancers. Other things that are known to be able to cause cancer - or make a person more likely to get cancer - include: radiation including sunlight and X-rays in large or many doses, and exposure to radiation (for example in a nuclear power plant); chemicals and materials used in building and manufacturing (for example, asbestos and benzene); high-fat or low-fiber diets; air and water pollution; eating very little fruits and vegetables; obesity; not enough physical activity; drinking too much alcohol; and certain chemicals commonly used at home. Some cancers can also be caused by viruses. Many people who are exposed to these things do get cancer - but some do not.

10.3 Kinds

The following is a **list of cancer types**. Cancer is a group of diseases that involve abnormal increases in the number of cells, with the potential to invade or spread to other parts of the body. Not all tumors or lumps are cancerous; benign tumors are not classified as being cancer because they do not spread to other parts of the body. There are over 100 different known cancers that affect humans. Cancers are often described by the body part that they originated in. However, some body parts contain multiple types of tissue, so for greater precision, cancers are additionally classified by the type of cell that the tumor cells originated from. These types include:

- *Carcinoma*: Cancers derived from epithelial cells. This group includes many of the most common cancers that occur in older adults. Nearly all cancers developing in the breast, prostate, lung, pancreas, and colon are carcinomas.
- *Sarcoma*: Cancers arising from connective tissue (i.e. bone, cartilage, fat, nerve), each of which develop from cells originating in mesenchymal cells outside of the bone marrow.
- *Lymphoma* and *leukemia*: These two classes of cancer arise from immature cells that originate in the bone marrow, and are intended to fully differentiate and mature into normal components of the immune system and the blood, respectively. Acute lymphoblastic leukemia is the most common type of cancer in children, accounting for ~30% of cases. However, far more adults than children develop lymphoma and leukemia.

- *Germ cell tumor*: Cancers derived from pluripotent cells, most often presenting in the testicle or the ovary (seminoma and dysgerminoma, respectively).
- *Blastoma*: Cancers derived from immature "precursor" cells or embryonic tissue. Blastomas are *generally* more common in children (e.g. neuroblastoma, retinoblastoma, nephroblastoma, hepatoblastoma, medulloblastoma, etc.) than in older adults.

Cancers are usually named using -carcinoma, -sarcoma or -blastoma as a suffix, with the Latin or Greek word for the organ or tissue of origin as the root. For example, the most common cancer of the liver parenchyma ("hepato-" = liver), arising from malignant epithelial cells ("carcinoma"), would be called a hepatocarcinoma, while a malignancy arising from primitive liver precursor cells is called a hepatoblastoma. Similarly, a cancer arising from malignant fat cells would be termed a liposarcoma. For some common cancers, the English organ name is used. For example, the most common type of breast cancer is called ductal carcinoma of the breast.

Benign tumors (which are not cancers) are usually named using *-oma* as a suffix with the organ name as the root. For example, a benign tumor of smooth muscle cells is called a *leiomyoma* (the common name of this frequently occurring benign tumor in the uterus is *fibroid*). Confusingly, some types of cancer use the *-noma* suffix, examples including melanoma and seminoma. Some types of cancer are named for the size and shape of the cells under a microscope, such as giant cell carcinoma, spindle cell carcinoma, and small-cell carcinoma. There are many different kinds of cancers. Some of the most common are:

- Breast cancer
- Brain cancer
- Leukemia (a blood cancer)
- Testicular cancer
- Mesothelioma (which starts in the lungs, and is usually caused by breathing in asbestos for long periods of time)
- Lung cancer

10.4 Treatment of cancer

There is no sure cure for cancer. It can only be cured if all of the cancerous cells are cut out or killed in place. This means that the earlier the cancer is treated, the better the chances are for a cure (because the cancer cells may not have had enough time to copy themselves and spread

so much that the person cannot be cured). There are a few different types of treatments that may kill cancer cells. These treatments are:

- Radiotherapy (radiation therapy), which uses radiation to kill cancer cells
- Chemotherapy, which uses strong medications to kill cancer cells
- Immunotherapy works by "inducing, enhancing, or suppressing an immune response".
- Surgery to take out part or all of a tumor
- After surgery, many patients may need radiotherapy or chemotherapy to keep the tumor from growing again

10.5 Treating cancer is complicated

There are a few reasons why treating cancer is complicated. For example:

- Most things that kill cancer cells also kill normal, healthy cells. This can cause many side effects, like hair loss and vomiting.
- The body's immune system usually will not attack cancer cells, even though they could easily kill the body. This is because the cancer has actually become a part of the body by invading cells and tissues. So the immune system sees the cancer as part of the body it is trying to protect, not as a threat to be attacked.
- There are many different types of cancer, and each has its own symptoms and causes. Even with the same type of cancer, different people may have different symptoms, and may react to treatments differently; their cancer also may grow or spread at different speeds. Treatment has to be a good fit to both the type of cancer and the individual patient who has the cancer.

Many, many people in many countries study cancer and work on finding treatments. There has been some good progress in finding treatments, and many cancers are treated with success. Along with looking for different medical treatments to treat cancer, some studies also look for things that people with cancer can do themselves to try to make themselves healthier. For example, one study showed that if a person with lymphedema (a swelling of the arm linked to breast cancer) lifts weights, he may be able to fight his cancer better than somebody who does not lift weights.

10.6 History

Cancer has been around for many thousands of years. Today, a lot of the medical terms used to describe cancer come from ancient Greek and Latin. For example, the Latinized Greek

word carcinoma is used to describe a malignant tumor - a tumor made up of cancer cells. The Greeks also used the word "karkinos", which would be translated by Aulus Cornelius Celsus into the Latin word *cancer*. The prefix 'carcino' is still used in medical words like carcinoma and carcinogenic. A famous Greek doctor, Galen, helped create another word that is very important to medicine today by using the word "*onkos*" to describe *all* tumours. This is where the word oncology, the branch of medicine that deals with cancer, comes from.

10.7 Ancient history

Hippocrates (a very famous ancient doctor who is often called the father of modern medicine) named many kinds of cancer. He called benign tumours (tumors that are not made up of cancer cells) *oncos*. In Greek, *onkos* means 'swelling'. He called malignant tumours *karkinos*. This means crab or crayfish in Greek. He used this term because he thought that if a solid malignant tumor was cut into, its veins looked like a crab: "the veins stretched on all sides as the animal the crab has its feet, whence it derives (gets) its name". Hippocrates later added -*oma* (Greek for 'swelling') after the word 'carcinos'. This is how the word *carcinoma* came about.

Because the ancient Greeks did not believe in opening up dead bodies to study them, Hippocrates was only able to describe and make drawings of tumors he saw from the outside of the body. He drew tumors that had been on the skin, nose, and breasts.

Hippocrates and other doctors at that time treated people based on the humor theory. This theory said that there were four types of fluid in the body (black, yellow bile, blood, and phlegm). Doctors tried to figure out whether these four "humors" (or body fluids) were in balance. They would then use treatments like blood-letting (cutting the patient and letting him bleed so that he would lose blood); laxatives (giving the patient foods or herbs to make him go to the bathroom), and/or changing the patient's diet. The doctors thought that these treatments would work to get the patient's four humors back into the right balance. The humor theory treatment was popular until the 19th century (the 1800s), when cells were discovered. By this time, people had realized that cancer can happen anywhere in the body.

10.8 What are the four stages of cancer?

Most cancers have four stages. The specific stage is determined by a few different factors, including the size and location of the tumor:

- **Stage I: C**ancer is localized to a small area and hasn’t spread to lymph nodes or other tissues.
- **Stage II:** Cancer has grown, but it hasn’t spread.

- **Stage III:** Cancer has grown larger and has possibly spread to lymph nodes or other tissues.
- **Stage IV:** Cancer has spread to other organs or areas of your body. This stage is also referred to as metastatic or advanced cancer.

Though stages I through IV are the most common, there is also stage zero. This earliest phase describes cancer that is still localized to the area in which it started. Cancers that are still in stage zero are usually easily treatable and are considered pre-cancerous by most healthcare providers.

10.9 What are the 5 types of cancer?

There are five main types of cancer. These include:

- **Carcinoma.** This type of cancer affects organs and glands, such as the lungs, breasts, pancreas and skin. Carcinoma is the most common type of cancer.
- **Sarcoma.** This cancer affects soft or connective tissues, such as muscle, fat, bone, cartilage or blood vessels.
- **Melanoma.** Sometimes cancer can develop in the cells that pigment your skin. These cancers are called melanoma.
- **Lymphoma.** This cancer affects your lymphocytes or white blood cells.
- **Leukemia.** This type of cancer affects blood.

10.10 How common is cancer?

Cancer is a common disease that can affect almost every part of your body. About 39.5% of all people will be diagnosed with cancer at some point in their lives.

10.11 How does cancer start in your body?

Cancer occurs when your genes stop controlling the way your cells divide. For example, instead of old cells dying, they grow and form abnormal cells.

10.12 How dangerous is cancer?

Cancer is potentially fatal. Currently, it’s the leading cause of death worldwide. However, fatality rates largely depend on the type of cancer and how far it has spread. Many types of cancer are successfully treated with prompt care.

10.13 Why is cancer so deadly?

When cancer cells develop, they can disturb proper organ function. This can result in reduced oxygen supply and a buildup of waste products. If vital organ function is impaired, it can lead to death.

10.14 What causes cancer?

Several factors contribute to the development of cancer in your body. Smoking and using tobacco products is one of the main causes of:

- Lung cancer.
- Oral cancer.
- Laryngeal cancer.
- Esophageal cancer.

Other causes of cancer include:

- **An unhealthy lifestyle**. Eating high-fat or high-sugar foods can increase your risk for many types of cancer. You're also more vulnerable to disease if you don't get enough exercise.
- **A toxic environment**. Exposure to toxins in your environment, such as asbestos, pesticides and radon, can eventually lead to cancer.
- **Radiation exposure**. Ultraviolet radiation from the sun significantly increases your risk for skin cancer. Over-exposure to radiation treatment can also be a risk factor.
- **Hormone therapy**. Women who are taking hormone replacement therapy may have an increased risk for breast cancer and endometrial cancer.

10.15 What is the first sign of cancer?

Cancer symptoms can vary significantly for each person. However, there are a few things that could indicate the early signs of disease. Schedule an appointment with your healthcare provider if you experience:

- Unexplained weight loss.
- Chronic tiredness.
- Persistent pain.
- Fever that occurs mostly at night.
- Skin changes.

10.16 What are common signs of cancer?

As time goes on, you may notice other cancer symptoms surfacing. These may include:

- An unusual lump.
- A sore that doesn't go away.
- Hoarseness.
- Dysphagia (difficulty swallowing).
- A mole or wart that changes in appearance.

One can note that these symptoms do not mean that you definitely have cancer. However, if any of these symptoms appear, you should see your healthcare provider right away.

10.17 How does cancer spread?

When cancer spreads, the cancer cells break away from the original tumor, travel through the body via your bloodstream or lymphatic system, then form new tumors in other areas. This process is called metastasis.

10.18 How is cancer diagnosed?

In order to treat your cancer, your healthcare provider needs to know the location of the tumor, the stage (whether it has spread) and whether you are strong enough to handle the treatment. They will perform a comprehensive examination and ask you about your symptoms. They may also order certain tests, including:

- Blood tests.
- Urine tests.
- X-rays.
- CT (computed tomography) scans.
- Magnetic resonance imaging (MRI).
- Ultrasonography.
- Biopsy.

10.19 How is cancer treated?

Once your medical team has given you a diagnosis, they'll design a personalized treatment plan based on their findings. Cancer treatment may include:

- **Chemotherapy.** One of the most common cancer treatments, chemotherapy uses powerful drugs to destroy cancer cells. Chemotherapy may be given through an IV or in pill form.
- **Radiation therapy.** This treatment kills cancer cells with high dosages of radiation. In some instances, radiation may be given at the same time as chemotherapy.
- **Surgery.** In some cases, your surgeon can surgically remove the tumor.
- **Hormone therapy.** Sometimes hormones can block other cancer-causing hormones. For example, men with prostate cancer might be given hormones to keep testosterone (which contributes to prostate cancer) at bay.
- **Biological response modifier therapy.** This treatment stimulates your immune system and helps it perform more effectively. It does this by changing your body's natural processes.
- **Immunotherapy.** Sometimes called biological therapy, immunotherapy treats disease by using the power of your body's immune system. It can target cancer cells while leaving healthy cells intact.
- **Bone marrow transplant.** Also called stem cell transplantation, this treatment replaces damaged stem cells with healthy ones. Prior to transplantation, you'll undergo chemotherapy to prepare your body for the process.

10.20 What are the side effects of cancer treatment?

People who undergo cancer treatment may experience a wide range of side effects. The exact side effects that you experience will depend on the type of cancer treatment you receive. Listed below are common side effects for various types of cancer treatment:

10.21 Chemotherapy

- Hair loss.
- Fatigue.
- Nausea and vomiting.

Radiation

- Fatigue.
- Hair loss.
- Skin problems.

Surgery

- Pain.

- Weakness.
- Infection.
- Blood clots.
- Allergy to anesthesia.

Hormone therapy

- Fatigue.
- Water retention (bloating).
- Hot flashes.
- Erectile dysfunction
- Blood clots.

10.22 Biological response modifier therapy/immunotherapy

- Fever.
- Chills.
- Muscle aches.
- Skin rash.
- Swelling.
- Increased bruising or bleeding.

Stem cell transplantation

- Nausea.
- Vomiting.
- Flu-like symptoms.
- Greater risk of infection.

10.23 How can I manage side effects of cancer treatment?

If you're undergoing cancer treatment, talking with your healthcare provider can help you manage your side effects. Many people find that maintaining a healthy diet helps them feel better and stay stronger. You may also benefit from incorporating exercise into your daily routine. Be sure to clear any dietary changes and activities with your healthcare provider first.

10.24 Cell death

Cell death is the event of a biological cell ceasing to carry out its functions. This may be the result of the natural process of old cells dying and being replaced by new ones, or may result from such factors as disease, localized injury, or the death of the organism of which the cells are part. Apoptosis or Type I cell-death, and autophagy or Type II cell-death are both forms of

programmed cell death, while necrosis is a non-physiological process that occurs as a result of infection or injury.

10.25 Programmed cell death

Programmed cell death (or PCD) is cell death mediated by an intracellular program. PCD is carried out in a regulated process, which usually confers advantage during an organism's life-cycle. For example, the differentiation of fingers and toes in a developing human embryo occurs because cells between the fingers apoptose; the result is that the digits separate. PCD serves fundamental functions during both plant and metazoa (multicellular animals) tissue development.

Apoptosis

Apoptosis is the process of programmed cell death (PCD) that may occur in multicellular organisms. Biochemical events lead to characteristic cell changes (morphology) and death. These changes include blebbing, cell shrinkage, nuclear fragmentation, chromatincondensation,and chromosomal DNA fragmentati on. It is now thought that – in a developmental context – cells are induced to positively commit suicide whilst in a homeostatic context; the absence of certain survival factors may provide the impetus for suicide. There appears to be some variation in the morphology and indeed the biochemistry of these suicide pathways; some treading the path of "apoptosis", others following a more generalized pathway to deletion, but both usually being genetically and synthetically motivated. There is some evidence that certain symptoms of "apoptosis" such as endonuclease activation can be spuriously induced without engaging a genetic cascade, however, presumably true apoptosis and programmed cell death must be genetically mediated. It is also becoming clear that mitosis and apoptosis are toggled or linked in some way and that the balance achieved depends on signals received from appropriate growth or survival factors.

10.26 Autophagy

Autophagy is *cytoplasmic*, characterized by the formation of large vacuoles that eat away organelles in a specific sequence prior to the destruction of the nucleus. Macroautophagy, often referred to as autophagy, is a catabolic process that results in the autophagosomic-lysosomal degradation of bulk cytoplasmic contents, abnormal protein aggregates, and excess or damaged organelles. Autophagy is generally activated by conditions of nutrient deprivation but has also been associated with physiological as well

as pathological processes such as development, differentiation, neurodegenerative diseases, stress, infection and cancer.

10.27 Other variations of PCD

Other pathways of programmed cell death have been discovered. Called "non-apoptotic programmed cell-death" (or "caspase-independent programmed cell-death"), these alternative routes to death are as efficient as apoptosis and can function as either backup mechanisms or the main type of PCD. Some such forms of programmed cell death are anoikis, almost identical to apoptosis except in its induction; cornification, a form of cell death exclusive to the eyes; excitotoxicity; ferroptosis, an iron-dependent form of cell death and Wallerian degeneration. Plant cells undergo particular processes of PCD similar to autophagic cell death. However, some common features of PCD are highly conserved in both plants and metazoa. Activation-induced cell death (AICD) is a programmed cell death caused by the interaction of Fas receptor and Fas ligand.

It occurs as a result of repeated stimulation of specific T-cell receptors (TCR) and it helps to maintain the periphery immune tolerance. Therefore, an alteration of the process may lead to autoimmune diseases. In the other words AICD is the negative regulator of activated T-lymphocytes.

Ischemic cell death, or oncosis, is a form of accidental, or passive cell death that is often considered a lethal injury. The process is characterized by mitochondrial swelling, cytoplasm vacuolization, and swelling of the nucleus and cytoplasm. Mitotic catastrophe is a mode of cell death that is due to premature or inappropriate entry of cells into mitosis.

It is the most common mode of cell death in cancer cells exposed to ionizing radiation and many other anti-cancer treatments. Immunogenic cell death or immunogenic apoptosis is a form of cell death caused by some cytostatic agents such as anthracyclines, oxaliplatin and bortezomib, or radiotherapy and photodynamic therapy (PDT). Pyroptosis is a highly inflammatory form of programmed cell death that occurs most frequently upon infection with intracellular pathogens and is likely to form part of the antimicrobial response in myeloid cells.

10.28 Necrotic cell death

Necrosis is cell death where a cell has been badly damaged through external forces such as trauma or infection and occurs in several different forms. In necrosis, a cell undergoes swelling, followed by uncontrolled rupture of the cell membrane with cell contents being

expelled. These cell contents often then go on to cause inflammation in nearby cells. A form of programmed necrosis, called necroptosis, has been recognized as an alternative form of programmed cell death. It is hypothesized that necroptosis can serve as a cell-death backup to apoptosis when the apoptosis signaling is blocked by endogenous or exogenous factors such as viruses or mutations. Necroptotic pathways are associated with death receptors such as the tumor necrosis factor receptor 1.

10.29 Field of study and etymology

The term "cell necrobiology" has been used to describe the life processes associated with morphological, biochemical, and molecular changes which predispose, precede, and accompany cell death, as well as the consequences and tissue response to cell death. The word is derived from the Greek νεκρό meaning "death", βìο meaning "life", and meaning "the study of". The term was initially coined to broadly define investigations of the changes that accompany cell death, detected and measured by multiparameter flow- and laser scanning-cytometry. It has been used to describe the real-time changes during cell death, detected by flow cytometry.

10.30 Carcinogen

A carcinogen is any substance, radionuclide, or radiation that promotes carcinogenesis, the formation of cancer. This may be due to the ability to damage the genome or to the disruption of cellular metabolic processes. Several radioactive substances are considered carcinogens, but their carcinogenic activity is attributed to the radiation, for example gamma rays and alpha particles, which they emit. Common examples of non-radioactive carcinogens are inhaled asbestos, certain dioxins, and tobacco smoke. Although the public generally associates carcinogenicity with synthetic chemicals, it is equally likely to arise from both natural and synthetic substances.[11] Carcinogens are not necessarily immediately toxic; thus, their effect can be insidious.

Cancer is any disease in which normal cells are damaged and do not undergo programmed cell death as fast as they divide via mitosis. Carcinogens may increase the risk of cancer by altering cellular metabolism or damaging DNA directly in cells, which interferes with biological processes, and induces the uncontrolled, malignant division, ultimately leading to the formation of tumors. Usually, severe DNA damage leads to programmed cell death, but if the programmed cell death pathway is damaged, then the cell cannot prevent itself from becoming a cancer cell.

There are many natural carcinogens. Aflatoxin B_1, which is produced by the fungus *Aspergillus flavus* growing on stored grains, nuts and peanut butter, is an example of a potent, naturally occurring microbial carcinogen. Certain viruses such as hepatitis B and human papilloma virus have been found to cause cancer in humans. The first one shown to cause cancer in animals is Rous sarcoma virus, discovered in 1910 by Peyton Rous. Other infectious organisms which cause cancer in humans include some bacteria (e.g. *Helicobacter pylori*) and helminths (e.g. *Opisthorchis viverrini* and *Clonorchis sinensis*).

Dioxins and dioxin-like compounds, benzene, kepone, EDB, and asbestos have all been classified as carcinogenic.[6] As far back as the 1930s, industrial smoke and tobacco smoke were identified as sources of dozens of carcinogens, including benzo[*a*]pyrene, tobacco-specific nitrosamines such as nitrosonornicotine, and reactive aldehydes such as formaldehyde, which is also a hazard in embalming and making plastics. Vinyl chloride, from which PVC is manufactured, is a carcinogen and thus a hazard in PVC production.

Co-carcinogens are chemicals that do not necessarily cause cancer on their own, but promote the activity of other carcinogens in causing cancer. After the carcinogen enters the body, the body makes an attempt to eliminate it through a process called biotransformation. The purpose of these reactions is to make the carcinogen more water-soluble so that it can be removed from the body. However, in some cases, these reactions can also convert a less toxic carcinogen into a more toxic carcinogen.

DNA is nucleophilic; therefore, soluble carbon electrophiles are carcinogenic, because DNA attacks them. For example, some alkenes are toxicated by human enzymes to produce an electrophilic epoxide. DNA attacks the epoxide, and is bound permanently to it. This is the mechanism behind the carcinogenicity of benzo[*a*]pyrene in tobacco smoke, other aromatics, aflatoxin and mustard gas.

10.31 Types of Cancer Treatment

There are many types of cancer treatment. The types of treatment that you receive will depend on the type of cancer you have and how advanced it is. Some people with cancer will have only one treatment. But most people have a combination of treatments, such as surgery with chemotherapy and radiation therapy. When you need treatment for cancer, you have a lot to learn and think about. It is normal to feel overwhelmed and confused. But, talking with your doctor and learning about the types of treatment you may have can help you feel more in control.

10.32 Biomarker Testing for Cancer Treatment

Biomarker testing is a way to look for genes, proteins, and other substances (called biomarkers or tumor markers) that can provide information about cancer. Biomarker testing can help you and your doctor choose a cancer treatment.

10.33 Chemotherapy

Chemotherapy is a type of cancer treatment that uses drugs to kill cancer cells. Learn how chemotherapy works against cancer, why it causes side effects, and how it is used with other cancer treatments.

10.34 Hormone Therapy

Hormone therapy is a treatment that slows or stops the growth of breast and prostate cancers that use hormones to grow. Learn about the types of hormone therapy and side effects that may happen.

10.35 Hyperthermia

Hyperthermia is a type of treatment in which body tissue is heated to as high as 113 °F to help damage and kill cancer cells with little or no harm to normal tissue. Learn about the types of cancer and precancers that hyperthermia is used to treat, how it is given, and the benefits and drawbacks of using hyperthermia.

10.36 Immunotherapy

Immunotherapy is a type of cancer treatment that helps your immune system fight cancer. This page covers the types of immunotherapy, how it is used against cancer, and what you can expect during treatment.

10.37 Photodynamic Therapy

Photodynamic therapy uses a drug activated by light to kill cancer and other abnormal cells. Learn how photodynamic therapy works, about the types of cancer and precancers it is used to treat, and the benefits and drawbacks of this treatment.

10.38 Radiation Therapy

Radiation therapy is a type of cancer treatment that uses high doses of radiation to kill cancer cells and shrink tumors. Learn about the types of radiation, why side effects happen, which side effects you might have, and more.

10.39 Stem cell transplant

Stem cell transplants are procedures that restore stem cells that grow into blood cells in people who have had theirs destroyed by high doses of chemotherapy or radiation therapy. Learn

about the types of transplants, side effects that may occur, and how stem cell transplants are used in cancer treatment.

10.40 Surgery

When used to treat cancer, surgery is a procedure in which a surgeon removes cancer from your body. Learn the different ways that surgery is used against cancer and what you can expect before, during, and after surgery.

10.41 Targeted Therapy

Targeted therapy is a type of cancer treatment that targets the changes in cancer cells that help them grow, divide, and spread. Learn how targeted therapy works against cancer and about common side effects that may occur.

10.42 Summary

Under this unit we have summarized cancer, its types, tumor development and cancer therapy etc. In simple terms, cancer is an abnormal growth of body cells. Each one of us is born with a potential for cancer. One cannot "Catch" it as one would an infection or a cold. When the programming of a cell or a group of cells is affected, growth may become uncontrolled. Some of the factors that can alter the code are chronic irritation, tobacco, smoke and dust, radioactive substances, age, sex, race and heredity. While one cannot control many of these factors , we need to be aware of the ones we can control of. Prevention is definitely better than treatment of cancer. Normal cells grow in a well regulated pattern. When cancer sets in, a group of cells suddenly starts multiplying in a haphazard and uncontrolled way, forming lumps or tumors. a malignant tumor never stops growing and can spread to other parts of the body.

Cancer is a group of diseases that involve abnormal increases in the number of cells, with the potential to invade or spread to other parts of the body. Not all tumors or lumps are cancerous; benign tumors are not classified as being cancer because they do not spread to other parts of the body. There is no sure cure for cancer. It can only be cured if all of the cancerous cells are cut out or killed in place. This means that the earlier the cancer is treated, the better the chances are for a cure (because the cancer cells may not have had enough time to copy themselves and spread so much that the person cannot be cured). There are a few different types of treatments that may kill cancer cells.

10.43 Terminal questions

Q. 1 What do you mean by cancer? Describe it.

Answer:----------

Q. 2 Describe endocytosis, phagocytosis and macrophages.

Answer:----------

Q. 3 Describe the multiple steps of tumor development.

Answer:----------

Q. 4. Explain cell death?

Answer:----------

Q. 5 Explain cell apoptosis?

Answer:----------

Q. 6 Write a short note on carcinogens.

Answer:----------

Further readings

1. Biochemistry- Lehninger A.L.
2. Biochemistry –J.H.Weil.
3. Biochemistry fourth edition-David Hames and Nigel Hooper.
4. Textbook of Biochemistry for Undergraduates - Rafi, M.D.
5. Biochemistry and molecular biology- Wilson Walker.