UNIT- III

Lipid Metabolism

- Lipids are a heterogeneous group of water-insoluble (hydrophobic) organic molecules that can be extracted from tissues by nonpolar solvents, because of their insolubility in aqueous solutions, body lipids are generally found compartmentalized, as in the case of membrane-associated lipids or droplets of triacylglycerol in adipocytes, or transported in plasma in association with protein, as in lipoprotein particles or on albumin.
- Lipids are a major source of energy for the body, and they provide the hydrophobic barrier.
- Lipids serve additional functions in the body, for example, some fat-soluble vitamins have regulatory or coenzyme functions, and the prostaglandins and steroid hormones play major roles in the control of the body's homeostasis.

Classification of lipids:

1. **Simple lipids:** Esters of fatty acids with various alcohols.
   a. *Fats:* Esters of fatty acids with glycerol. *Oils* are fats in the liquid state.
   b. *Waxes:* Esters of fatty acids with higher molecular weight monohydric alcohols.

2. **Complex lipids:** Esters of fatty acids containing groups in addition to an alcohol and a fatty acid.
   a. *Phospholipids:* Lipids containing, in addition to fatty acids and an
alcohol, a phosphoric acid residue. They frequently have nitrogen containing bases and other substituent’s, eg, in glycerophospholipids the alcohol is glycerol and in sphingophospholipids the alcohol is sphingosine.

b. **Glycolipids (glycosphingolipids):** Lipids containing a fatty acid, sphingosine, and carbohydrate.

c. **Other complex lipids:** Lipids such as sulfolipids and amino lipids. Lipoproteins may also be placed in this category.

3. **Precursor and derived lipids:** These include fatty acids, glycerol, steroids, other alcohols, fatty aldehydes, and ketone bodies, hydrocarbons, lipid-soluble vitamins and hormones.

Fatty acids occur mainly as esters in natural fats and oils but do occur in the unesterified form as **free fatty acids**, a transport form found in the plasma. Fatty acids that occur in natural fats are usually straight-chain derivatives containing an even number of carbon atoms. The chain may be **saturated** (containing no double bonds) or **unsaturated** (containing one or more double bonds).

- **Saturated Fatty Acids** may base on acetic acid (CH₃COOH) as the first member of the series in which -CH₂- is progressively added between the terminals -CH₃- and -COOH- groups.

- **Unsaturated Fatty Acids** contain one or more double bonds and it may be further subdivided as follows:

  (1) Monounsaturated (monoethenoid, monoenoic) acids, containing one double bond.
(2) Polyunsaturated (polyethenoid, polyenoic) acids, containing two or more double bonds.

(3) Eicosanoids: These compounds, derived from eicosa- (20-carbon) polyenoic fatty acids, comprise the prostanoids, leukotrienes (LTs), and lipoxins (LXs). Prostanoids include prostaglandins (PGs), prostacyclins (PGIs), and thromboxanes (TXs).

<table>
<thead>
<tr>
<th>Common Name</th>
<th>Number of C Atoms</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acetic</td>
<td>2</td>
<td>Major end product of carbohydrate fermentation by rumen organisms(^1)</td>
</tr>
<tr>
<td>Propionic</td>
<td>3</td>
<td>An end product of carbohydrate fermentation by rumen organisms(^1)</td>
</tr>
<tr>
<td>Butyric</td>
<td>4</td>
<td>In certain fats in small amounts (especially butter). An end product of carbohydrate fermentation by rumen organisms(^1)</td>
</tr>
<tr>
<td>Valeric</td>
<td>5</td>
<td></td>
</tr>
<tr>
<td>Caproic</td>
<td>6</td>
<td></td>
</tr>
<tr>
<td>Lauric</td>
<td>12</td>
<td>Spermaceti, cinnamon, palm kernel, coconut oils, laurels, butter</td>
</tr>
<tr>
<td>Myristic</td>
<td>14</td>
<td>Nutmeg, palm kernel, coconut oils, myrtles, butter</td>
</tr>
<tr>
<td>Palmitic</td>
<td>16</td>
<td>Common in all animal and plant fats</td>
</tr>
<tr>
<td>Stearic</td>
<td>18</td>
<td></td>
</tr>
</tbody>
</table>
Saturated fatty acids.

Unsaturated Fatty Acids

<table>
<thead>
<tr>
<th>Number of C Atoms and Number and Position of Double Bonds</th>
<th>Common Name</th>
<th>Occurrence</th>
</tr>
</thead>
<tbody>
<tr>
<td>16:1,9</td>
<td>Palmitoleic</td>
<td>in nearly all fats.</td>
</tr>
<tr>
<td>18:1,9</td>
<td>Oleic</td>
<td>Possibly the most common fatty acid in natural fats.</td>
</tr>
<tr>
<td>18:1,9</td>
<td>Elaidic</td>
<td>Hydrogenated and ruminant fats.</td>
</tr>
<tr>
<td>18:2,9,12</td>
<td>Linoleic</td>
<td>Corn, peanut, cottonseed, soybean, and many plant oils.</td>
</tr>
<tr>
<td>18:3,6,9,12</td>
<td>γ-Linolenic</td>
<td>Some plants, eg, oil of evening primrose, borage oil, minor fatty acid in animals.</td>
</tr>
<tr>
<td>18:3,9,12,15</td>
<td>α-Linolenic</td>
<td>Frequently found with linoleic acid but particularly in linseed oil.</td>
</tr>
<tr>
<td>20:4,5,8,11,14</td>
<td>Arachidonic</td>
<td>Found in animal fats and in peanut oil; important component of phospholipids in animals.</td>
</tr>
</tbody>
</table>

β-Oxidation of fatty acids

- The major pathway for catabolism of even-numbered saturated fatty acids is a mitochondrial pathway called β-oxidation. In which two-carbon fragments are successively removed from the carboxyl end of the fatty acyl CoA, producing acetyl CoA, NADH, and FADH$_2$.

- In β-oxidation, the fatty acid is broken down to release acetyl-CoA. The process involves 4 main steps:

  i. Dehydrogenation
Beta-oxidation of fatty acids takes place in the mitochondrial matrix for the most part. However, fatty acids have to be activated for degradation by coenzyme A by forming a fatty acyl-CoA thioester.

- The final fatty acid products are acetyl-CoA for the even numbered fatty acids (without double bonds)

**Beta-Oxidation of Fatty Acids (even chain)**

1. **Dehydrogenation** (Acyl-CoA Dehydrogenase): This first reaction is the oxidation of the $C_a$-$C_b$ bond. It is catalyzed by acyl-CoA dehydrogenases. This catalyst is a family of three soluble matrix enzymes. These enzymes carry noncovalently bound FAD that is reduced during the oxidation of the fatty acid.

   ![Dehydrogenation Reaction](image)

2. **Hydration** (Enoyl-CoA Hydratase): In this pathway is one in which water is added across the new double bond to make hydroacyl-CoA. The catalyst in this reaction is Enoyl-CoA hydratase. This is also called a crotonase and it converts trans-enoyl-CoA to L-B-Hydroxyacyl-CoA. This reaction would be classified as a hydration reaction because you are adding water.
3. **Oxidation (L-Hydroxyacyl-CoA Dehydrogenase):** Here the oxidation of the hydroxyl group at the beta position which forms a beta-ketoacyl-CoA derivative and it is catalyzed by *L-Hydroxyacyl-CoA Dehydrogenase*.

Mechanism of *L-Hydroxyacyl-CoA Dehydrogenase*

4. **Thiolysis:** This is the final reaction of this pathway and *thiolase* catalyzed this reaction. This reaction cleaves the beta-ketoacyl-CoA. The products of this reaction are an acetyl-CoA and a fatty acid that has been shortened by two carbons. So, this reaction is classified as a cleavage reaction.
Repetition of the Beta Oxidation Cycle: The shortened fatty acyl-CoA that was the product of the last reaction now goes through another beta-oxidation cycle. This keeps happening until eventually you wind up with two molecules of acetyl-CoA in the final step. This acetyl-CoA is then available to be further metabolized in the TCA cycle, or it can be used as a substrate in amino acid biosynthesis. It cannot be used as a substrate for gluconeogenesis.

Energy yield during β-oxidation of fatty acids

The ATP yield for every oxidation cycle is 14 ATP, broken down as follows:

1 FADH$_2$ x 2 ATP = 2 ATP
1 NADH x 3 ATP = 3ATP
1 acetyl-CoA x 12 ATP = 12ATP

the ATP yield of Palmitate ($C_{16}$, $n = 8$) is

Or

7 FADH$_2$ x 2 ATP = 14 ATP
7 NADH x 3 ATP = 21 ATP
8 acetyl-CoA x 12 ATP = 96 ATP

Total ATP = 131

ATP equivalent used during activation = -2
BIOSYNTHESIS OF FATTY ACIDS

- **Fatty acid synthesis** is the creating of fatty acids from acetyl-CoA and malonyl-CoA precursors through action of enzymes called *fatty acid synthases*. It is an important part of the lipogenesis process, which together with *glycolysis* stands behind creating fats from blood sugar in living organisms.

- Synthesis takes place in the cytosol.

- In humans, fatty acids are predominantly formed in the *liver* and *lactating mammary glands*, and, to a lesser extent, the *adipose tissue*.

- Most acetyl-CoA is formed from *pyruvate* by *pyruvate dehydrogenase* in the mitochondria. Acetyl-CoA produced in the mitochondria is condensed with oxaloacetate by *citrate synthase* to form *citrate*, which is then transported into the cytosol and broken down to yield acetyl-CoA and oxaloacetate by *ATP citrate lyase*. Oxaloacetate in the cytosol is reduced to *malate* by cytoplasmic *malate dehydrogenase*, and malate is transported back into the mitochondria to participate in the *Citric acid cycle*.

Acyl carrier protein (ACP): The acyl carrier protein (ACP) is an important component in both fatty acid and polyketide biosynthesis.
DIFFERENCES BETWEEN FATTY ACID DEGRADATION AND SYNTHESIS

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Degradation</th>
<th>Synthesis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Location</td>
<td>Mitochondrial Matrix</td>
<td>Cytosol</td>
</tr>
<tr>
<td>Activated intermediates</td>
<td>Thioesters of CoA</td>
<td>Thioesters of ACP</td>
</tr>
<tr>
<td>Process</td>
<td>2-Carbon fragments removed as acetyl CoA</td>
<td>2-Carbon elongation using malonyl CoA</td>
</tr>
<tr>
<td>Direction</td>
<td>Starts at carboxyl end</td>
<td>Starts at methyl end</td>
</tr>
<tr>
<td>Redox reaction cofactors</td>
<td>FAD/FADH₂ and NAD⁺/NADH</td>
<td>NADP⁺/NADPH</td>
</tr>
<tr>
<td>Major tissue site</td>
<td>Muscle and liver</td>
<td>Liver</td>
</tr>
<tr>
<td>Hormonal regulation</td>
<td>Low insulin/glucagon ratio</td>
<td>High insulin/glucagon ratio</td>
</tr>
<tr>
<td>Activator</td>
<td>FFA generated by hormone-sensitive lipase</td>
<td>Citrate</td>
</tr>
<tr>
<td>Inhibitor</td>
<td>Malonyl CoA (inhibits carnitine acyl transferase)</td>
<td>Fatty acyl CoA (inhibits acetyl CoA carboxylase)</td>
</tr>
</tbody>
</table>
KETOGENESIS

- **Ketogenesis** is the process by which ketone bodies are produced as a result of fatty acid breakdown.

- Ketone bodies are produced mainly in the mitochondria of liver cells. Its synthesis occurs in response to low glucose levels in the blood, and after exhaustion of cellular carbohydrate stores, such as glycogen. The production of ketone bodies is then initiated to make available energy that is stored as fatty acids.

- Besides its role in the synthesis of ketone bodies, HMG-CoA is also an intermediate in the synthesis of cholesterol.

- The three ketone bodies are:
  - *Acetoacetate*, which, if not oxidized to form usable energy, is the source of the two other ketone bodies below
  - *Acetone*, which is not used as an energy source, but is instead exhaled or excreted as waste
  - *β-hydroxybutyrate*, which is not, in the technical sense, a ketone according to IUPAC nomenclature.

 ✓ **Regulation:** Ketogenesis may or may not occur, depending on levels of available carbohydrates in the cell or body. This is closely related to the paths of acetyl-CoA:
  - When the body has ample carbohydrates available as energy source, glucose is completely oxidized to CO₂; acetyl-CoA is formed as an intermediate in this process, first entering the citric acid cycle followed by complete conversion of its chemical energy to ATP in oxidative phosphorylation.
  - When the body has excess carbohydrates available, some glucose
is fully metabolized, and some of it is stored by using acetyl-CoA to create fatty acids. (CoA is also recycled here.)

- When the body has no free carbohydrates available, fat must be broken down into acetyl-CoA in order to get energy.

✓ **Pathology**

- Ketone bodies are created at moderate levels in everyone's bodies, such as during sleep and other times when no carbohydrates are available.
- However, when ketogenesis is happening at higher-than-normal levels, the body is said to be in a state of ketosis.
- Both **acetoacetate** and **beta-hydroxybutyrate** are acidic, and, if levels of these ketone bodies are too high, the pH of the blood drops, resulting in **ketoacidosis**.
- **Ketoacidosis** is known to occur in untreated Type I diabetes (**diabetic ketoacidosis**) and in alcoholics after prolonged binging without intake of sufficient carbohydrates (**alcoholic ketoacidosis**).
Ketogenesis Pathway
KETOACIDOSIS

- **Ketoacidosis** is a metabolic state associated with high concentrations of ketone bodies, formed by the breakdown of fatty acids and the deamination of amino acids. The two common ketones produced in humans are acetoacetic acid and β-hydroxybutyrate.

- In ketoacidosis, the body fails to adequately regulate ketone production causing such a severe accumulation of ketone acids that the pH of the blood is substantially decreased. In extreme cases ketoacidosis can be fatal.

- Ketoacidosis occurs when the body is producing large quantities of ketone bodies via the metabolism of fatty acids (ketosis) and the body is producing insufficient insulin to slow this production.

- The excess ketone bodies can significantly acidify the blood.

- There are two common types of Ketoacidosis i.e. diabetic and alcoholic ketoacidosis.
  
  i. In diabetic patients, ketoacidosis is usually accompanied by insulin deficiency, hyperglycemia, and dehydration. Particularly in type 1 diabetics the lack of insulin in the bloodstream prevents glucose absorption and can cause unchecked ketone body production.

  ii. In alcoholic ketoacidosis, alcohol causes dehydration and blocks the first step of gluconeogenesis. The body is unable to synthesize enough glucose to meet its needs, thus creating an energy crisis resulting in fatty acid metabolism, and ketone body formation.
KETONE BODIES

- Ketone bodies are three water-soluble compounds that are produced as by-products when fatty acids are broken down for energy in the liver and kidney.
- They are used as a source of energy in the heart and brain. In the brain, they are a vital source of energy during fasting.
- The three endogenous ketone bodies are acetone, acetoacetic acid, and beta-hydroxybutyric acid, although beta-hydroxybutyric acid is not technically a ketone but a carboxylic acid.
- Ketone bodies can be used for energy. Ketone bodies are transported from the liver to other tissues, where acetoacetate and beta-hydroxybutyrate can be reconverted to acetyl-CoA to produce energy, via the citric acid cycle.
- Ketone bodies are produced from acetyl-CoA (ketogenesis) mainly in the mitochondrial matrix.
- When even larger amounts of ketone bodies accumulate such that the blood's pH is lowered to dangerously acidic levels, this state is called **ketoacidosis**.

**KETONURIA**

- Ketonuria is a medical condition in which ketone bodies are present in the urine.
- It is seen in conditions in which the body produces excess ketones as an alternative source of energy. It is seen during starvation or more commonly in type I diabetes mellitus. Production of ketone bodies is a normal response to a shortage of glucose, meant to provide an alternate source of fuel from fatty acids.
- *Causes of ketosis and ketonuria*
  
  i. Metabolic abnormalities such as diabetes, renal glycosuria, or glycogen storage disease
  ii. Dietary conditions such as starvation, fasting, high protein, or low carbohydrate diets, prolonged vomiting, and anorexia
  iii. Conditions in which metabolism is increased, such as hyperthyroidism, fever, pregnancy or lactation
- In nondiabetic persons, ketonuria may occur during acute illness or severe stress. Approximately 15% of hospitalized patients may have ketonuria, even though they do not have diabetes.
CHOLESTEROL SYNTHESIS, TRANSPORT & EXCRETION

- Cholesterol is present in tissues and in plasma either as free cholesterol or as a storage form, combined with a long-chain fatty acid as cholesteryl ester.

- Cholesterol is an amphipathic lipid and as such is an essential structural component of membranes and of the outer layer of plasma lipoproteins.

- It is synthesized in many tissues from acetyl-CoA and is the precursor of all other steroids in the body such as corticosteroids, sex hormones, bile acids, and vitamin D.

- Plasma low-density lipoprotein (LDL) is the vehicle of uptake of cholesterol and cholesteryl ester into many tissues. Free cholesterol is removed from tissues by plasma high-density lipoprotein (HDL) and transported to the liver, where it is eliminated from the body either unchanged or after conversion to bile acids in the process known as reverse cholesterol transport.

- Cholesterol is a major constituent of gallstones. However, its chief role in pathologic processes is as a factor in the genesis of atherosclerosis of vital arteries, causing cerebrovascular, coronary and peripheral vascular disease.

- **Biosynthesis of cholesterol:** Cholesterol synthesis occurs in the cytoplasm and microsomes from the two-carbon acetate group of acetyl-CoA.

- Biosynthesis of cholesterol in the liver accounts for approximately 10%, and in the intestines approximately 15%, of the amount produced each day. The process has five major steps:

1. Acetyl-CoAs are converted to 3-hydroxy-3-methylglutaryl-CoA
(HMG-CoA)

2. HMG-CoA is converted to mevalonate

3. Mevalonate is converted to the isoprene based molecule, isopentenyl pyrophosphate (IPP), with the concomitant loss of CO₂

4. IPP is converted to squalene and

5. Then Squalene is converted to cholesterol.
“Mevalonate” Pathway to IPP Synthesis

\[
\begin{align*}
\text{Acetyl CoA} & \quad \rightarrow \quad \text{CoASH} \quad \text{Acetoacetyl CoA} \\
\text{3-Hydroxy-3-methyl glutaryl CoA (HMG-CoA)} & \quad \rightarrow \quad \text{2 NADPH + 2H} \\
\text{Mevalonate} & \rightarrow \quad \text{ATP} \\
\text{5-Phosphomevalonate} & \rightarrow \quad \text{5-Pyrophosphomevalonate} \\
\text{Isopentenyl pyrophosphate [IPP]} & \rightarrow \quad \text{CO}_2
\end{align*}
\]
JAIPUR COLLEGE OF PHARMACY, JAIPUR
B.PHARMACY, FIRST YEAR, SECOND SEMESTER
BIOCHEMISTRY
Prepared by: Dr. Rakesh Kumar Gupta

Acetyl-CoA → 3-hydroxy-3-methylglutaryl-CoA (HMG-CoA)

Acetoacetyl-CoA → HMG-CoA synthase

HMG-CoA reductase

STATINS

Mevalonic acid → Mevalonate-5-phosphate

Mevalonate kinase

ATP

Phosphomevalonate kinase

Mevalonate-5-pyrophosphate

Isopentenyl-PP isomerase

Dimethylallyl-PP → Isopentenyl-5-pyrophosphate (PP)

Isopentenyl pyrophosphate (IPP)
Steroid hormone

Steroid hormone, are of a group of hormones that belong to the class of chemical compounds known as steroids; they are secreted by three “steroid glands”—the adrenal cortex, testes, and ovaries—and during pregnancy by the placenta. All steroid hormones are derived from cholesterol. They are transported through the bloodstream to the cells of various target organs where they carry out the regulation of a wide range of physiological functions.

Major pathways involved in the biosynthesis of steroid hormones.
These hormones often are classified according to the organs that synthesize them: the adrenal steroids are so called because they are secreted by the adrenal cortex, and the sex hormones are those produced by the ovaries and testes. This distinction is not exclusive, however, because the adrenal cortex also secretes sex hormones, albeit to a lesser extent than do the gonads, and the ovaries under abnormal conditions may produce adrenal steroids.

The adrenal cortex produces the adrenocortical hormones, which consist of the glucocorticoids and the mineralocorticoids. Glucocorticoids such as cortisol control or influence many metabolic processes, including the formation of glucose from amino acids and fatty acids and the deposition of glycogen in the liver. Glucocorticoids also help to maintain normal blood pressure, and their anti-inflammatory and immunosuppressive actions have rendered them useful in treating rheumatoid arthritis and preventing the rejection of transplanted organs. Mineralocorticoids such as aldosterone help maintain the balance between water and salts in the body, predominantly exerting their effects within the kidney.

The androgens are the male sex hormones. The principal androgen, testosterone, is produced primarily by the testes and in lesser amounts by the adrenal cortex and (in women) by the ovaries. Androgens are primarily responsible for the development and maintenance of reproductive function and stimulation of the secondary sex characteristics in the male. Androgens also have an anabolic (synthesizing and constructive, rather than degradative) function in stimulating the production of skeletal muscles and bone as well as red blood cells. To enhance the anabolic activity of androgens without increasing their masculinizing ability, anabolic steroids were developed. Though originally intended to combat diseases marked by wasting, these synthetic hormones have
been abused by individuals desiring to increase their muscle mass, such as athletes seeking to gain a competitive advantage. Overdosing has been linked to serious side effects, including infertility and coronary heart disease.

**Estrogens** are one of the two types of female sex hormones. They are secreted mainly by the ovaries and in smaller amounts by the adrenal glands and (in men) by the testes. Estradiol is the most potent of the estrogens. Functioning similarly to androgens, the estrogens promote the development of the primary and secondary female sex characteristics; they also stimulate linear growth and skeletal maturation. In other mammals these hormones have been shown to precipitate estrus (heat). The ovarian production of estrogen plummets during menopause.

**Progestins**, the most important of which is progesterone, are the other type of female sex hormone and are named for their role in maintaining pregnancy (progestation). Estrogens and progestins are secreted cyclically during menstruation. During the menstrual cycle, the ruptured ovarian follicle (the corpus luteum) of the ovary produces progesterone, which renders the uterine lining receptive to the implantation of a fertilized ovum. Should this occur, the placenta becomes the main source of progesterone, without which the pregnancy would terminate. As pregnancy progresses, placental production of progesterone increases, and these high doses suppress ovulation, preventing a second conception. The contraceptive quality of progesterone led to the development of structurally modified progestins and estrogens—the oral contraceptives known as birth-control pills, used by women to prevent unwanted pregnancy.
Steroidogenesis

- It is the process wherein desired forms of steroids are generated by transformation of other steroids. The pathways of human steroidogenesis are shown in the figure.

- Products of steroidogenesis include:
  
  a. androgens
  b. testosterone
  c. estrogens and progesterone
  d. corticoids
  e. cortisol
  f. aldosterone
Elimination of Steroids:

- Steroids are mainly oxidized by cytochrome P450 oxidase enzymes, such as CYP3A4.
- These reactions introduce oxygen into the steroid ring and allow the structure to be broken up by other enzymes, to form bile acids as final products.
- These bile acids can then be eliminated through secretion from the liver in the bile. The end products of cholesterol utilization are the bile acids, synthesized in the liver.
- Synthesis of bile acids is one of the predominant mechanisms for the excretion of excess cholesterol. However, the excretion of cholesterol in the form of bile acids is insufficient to compensate for an excess dietary intake of cholesterol.
- The most abundant bile acids in human bile are chenodeoxycholic acid (45%) and cholic acid (31%). These are referred to as the primary bile acids. Within the intestines the primary bile acids are acted upon by bacteria and converted to the secondary bile acids, identified as deoxycholate (from cholate) and lithocholate (from chenodeoxycholate). Both primary and secondary bile acids are reabsorbed by the intestines and delivered back to the liver via the portal circulation.
- Within the liver the carboxyl group of primary and secondary bile acids is conjugated via an amide bond to either glycine or taurine before their being re-secreted into the bile canaliculi.
- These conjugation reactions yield glycoconjugates and tauroconjugates, respectively.
- The bile canaliculi join with the bile ductules, which then form the bile...
ducts. Bile acids are carried from the liver through these ducts to the gallbladder, where they are stored for future use.

- The ultimate fate of bile acids is secretion into the intestine, where they aid in the emulsification of dietary lipids.
- In the gut the glycine and taurine residues are removed and the bile acids are either excreted (only a small percentage) or reabsorbed by the gut and returned to the liver. This process of secretion from the liver to the gallbladder, to the intestines and finally reabsorption is termed the **enterohepatic circulation**.
Vitamin D

Vitamin D exists in two forms, vitamin D2 and vitamin D3, which differ in the structure of their side chains. These are called ergocalciferol and cholecalciferol respectively. Both forms are equivalent as to their biological activity and equivalent in dosage. Both are metabolized by conversion to the 25-hydroxy form and then to the 1,25-dihydroxy metabolite in the kidney, which is the bioactive form. This has a structure which is similar to other steroid hormones produced in the body.

Vitamin D2 is found in a few plant sources, but is mostly produced on a commercial scale by the irradiation of yeast. This is the form used to fortify foods and to produce supplements. Vitamin D3 has several sources, being produced by ultraviolet radiation acting on the parent compound, or ingested in the form of deep sea fatty fish, egg yolks or liver, or supplements.

Vitamin D is a derivative of 7-dehydrocholesterol, also called ergosterol. This conversion is mediated by the action of ultraviolet radiation the parent compound, which is formed in the Malpighian layer of skin during a relatively minor route of cholesterol synthesis. Ultraviolet radiation with wavelengths between 290-315 nm causes the bond between the 9th and 10th position of the steroid ring to open, forming a compound called secosterol. This further undergoes cis-to-trans isomerization, by the formation of a trans bond between the 5th and 6th carbon atoms, leading to the formation of vitamin D3, or cholecalciferol. The involvement of ultraviolet radiation in the process has led to vitamin D being nicknamed the “sunshine vitamin.”

Cholecalciferol is then carried to the liver, where a mitochondrial hydroxylase enzyme introduces a hydroxyl group at the 25 position. This reaction requires
both energy in the form of NADPH and oxygen. The product, called 25-hydroxy cholecalciferol, is the inactive storage form of cholecalciferol, and is stored in the liver.

In case of need, 25-hydroxycholecalciferol is transported to the kidney where a second hydroxylation occurs at the 1 position, converting it to 1,25-dihydroxy cholecalciferol, the bioactive form of vitamin D. The production of this active form is regulated by an enzyme produced in the kidney, which is itself controlled by several factors. These include feedback from the level of the active form of the vitamin already in circulation, the secretion of parathyroid hormone, as well as calcium and phosphate levels which are the primary target of action of the vitamin.

1,25-dihydroxy cholecalciferol, also called calcitriol, is carried in the bloodstream to the intestinal mucosa. There it stimulates the absorption of calcium and phosphate, the mineral ions which are of prime importance in the building up of bone and other supportive tissue. It also promotes bone growth and remodeling by osteoblasts and osteoclasts.

**DISORDERS OF LIPID METABOLISM**

Lipids are large, water-insoluble molecules that have a variety of biological functions, including storing energy and serving as components of cellular membranes and lipoproteins. Cells that line the small intestine absorb dietary lipids and process them into lipoprotein particles that enter the circulation via the lymphatic system for eventual uptake by the liver. Triglycerides, cholesterol, and fat-soluble vitamins are transported through the blood by these lipoprotein particles.
Hypercholesterolemia

Hypercholesterolemia can be defined as the presence of high plasma cholesterol levels, with normal plasma triglycerides, as a consequence of the rise of cholesterol and apolipoprotein B (apoB)-rich lipoproteins, called low-density lipoprotein (LDL). According to the WHO definition (1970), hypercholesterolemia would be included in IIa phenotype (Ramasamy, 2016).

The limits to define hypercholesterolemia can be established according to plasma levels of total and LDL cholesterol (LDL-C) above the 95th percentile corrected for age and gender in each population.

- Hypercholesterolemia, or high cholesterol, occurs when there is too much cholesterol in the body.
- Cholesterol is a soft, waxy, fat-like substance that is a natural component of all the cells of the body.
- High cholesterol raises risk for heart disease, heart attack, and stroke. When there is too much cholesterol circulating in the blood, it can create sticky deposits (called plaque) along the artery walls. Plaque can eventually narrow or block the flow of blood to the brain, heart, and other organs. And blood cells that get caught on the plaque form clots, which can break loose and completely block blood flow through an artery, causing heart attack or stroke.
- There are two types of cholesterol -- HDL (high-density lipoproteins, or "good" cholesterol) and LDL (low-density lipoproteins, or "bad" cholesterol).
- The amount of HDL relative to LDL is considered a more important
indicator of heart disease risk.

- There is a third kind of fatty material, triglycerides, found in the blood. They also play a role (generally as triglyceride levels rise, "good" HDL cholesterol falls).
- The usual symptoms of high cholesterol, especially in early stages. The only way to determine cholesterol is high is through a blood test.
- The most important risk factors for high cholesterol are: Being overweight or obese, Eating a diet high in saturated fat and trans fatty acids (found in processed and fried foods), Not getting enough exercise, Family history of heart disease, High blood pressure, Smoking, Diabetes etc

**Treatment Approach:** Lowering your cholesterol level reduces your risk of heart disease and stroke. Changes in lifestyle -- better diet, more exercise and specific cholesterol-lowering medications are often prescribed like, Lovastatin, Pravastatin, Rosuvastatin, Simvastatin, Atorvastatin or Fluvastatin, etc

**Total cholesterol levels** (mg/dL):

- Desirable: Below 200
- Borderline high: 200 - 239
- High: Above 240

**LDL cholesterol level** (mg/dL):

- Optimal for people with heart disease or who are at high risk: Below 70
- Optimal for people at risk of heart disease: Below 100
- Optimal: 100 - 129
- Borderline high: 130 - 159
High: 160 - 189

**HDL cholesterol level (mg/dL):**

- Poor: Below 40
- Acceptable: 40 - 59
- Optimal: 60 or above

**Triglyceride levels (mg/dL):**

- Optimal: Below 150
- Borderline high: 150 - 199

High: Above 200

**ATHEROSCLEROSIS**

- Atherosclerosis is a disease in which plaque builds up on the insides of arteries.
- It is a syndrome affecting arterial blood vessels. It is a chronic inflammatory response in the walls of arteries, in large part due to the accumulation of macrophage white blood cells and promoted by low density (especially small particle) lipoproteins (plasma proteins that carry cholesterol and triglycerides) without adequate removal of fats and cholesterol from the macrophages by functional high density lipoproteins (HDL).
- It is commonly referred to as a "hardening" or "furring" of the arteries. It is caused by the formation of multiple plaques within the arteries.
The atheromatous plaque is divided into three distinct components:

1. The atheroma ("lump of porridge"), which is the nodular accumulation of a soft, flaky, yellowish material at the center of large plaques, composed of macrophages nearest the lumen of the artery
2. Underlying areas of cholesterol crystals
3. Calcification at the outer base of older/more advanced lesions.

Atherosclerosis can affect any artery in the body, including arteries in the heart, brain, arms, legs, and pelvis. As a result, different diseases may develop based on which arteries are affected.

1. **Coronary artery disease**: (CAD). This is when plaque builds up in the coronary arteries. These arteries supply oxygen-rich blood to your heart. When blood flow to your heart is reduced or blocked, it can lead to chest pain and heart attack. CAD also is called heart disease, and it's the leading cause of death in the United States.
2. **Carotid artery disease**: This happens when plaque builds up in the carotid arteries. These arteries supply oxygen-rich blood to your brain. When blood flow to your brain is reduced or blocked, it can lead to stroke.
3. **Peripheral arterial disease** (PAD): This occurs when plaque builds up in the major arteries that supply oxygen-rich blood to the legs, arms, and pelvis. When blood flow to these parts of your body is reduced or blocked, it can lead to numbness, pain, and sometimes dangerous infections.
Symptoms of Atherosclerosis

1. Unfortunately, atherosclerosis produces no symptoms until the damage to the arteries is severe enough to restrict blood flow.
2. Restriction of blood flow to the heart muscle due to atherosclerosis can cause angina pectoris or a myocardial infarction (a heart attack).
3. Restriction of blood flow to the muscles of the legs causes intermittent claudication (pains in the legs brought about by walking and relieved by rest).
4. Narrowing of the arteries supplying blood to the brain may cause transient ischemic attacks (symptoms and signs of a stroke lasting less than 24 hours) and episodes of dizziness, or ultimately, to a stroke itself.
Treatment of Atherosclerosis

1. Medication is unsatisfactory for treating atherosclerosis, since the damage has already been done.
2. Anticoagulant drugs have been used to try to minimize secondary clotting and embolus formation.
3. Vasodilator drugs are helpful in providing symptom relief, but are of no curative value.
4. Surgical treatment is available for those unresponsive to medical treatment or in certain high-risk situations.
5. Balloon angioplasty can open up narrowed vessels and promote an improved blood supply.
6. The blood supply to the heart can also be restored by coronary artery bypass surgery.
7. Medication is unsatisfactory for treating atherosclerosis, since the damage has already been done.
8. Anticoagulant drugs have been used to try to minimize secondary clotting and embolus formation.
9. Vasodilator drugs are helpful in providing symptom relief, but are of no curative value.
10. Surgical treatment is available for those unresponsive to medical treatment or in certain high-risk situations.
11. Balloon angioplasty can open up narrowed vessels and promote an improved blood supply.
12. The blood supply to the heart can also be restored by coronary artery bypass surgery.
FATTY LIVER:

1. It is also known as fatty liver disease (FLD), is a reversible condition where large vacuoles of triglyceride fat accumulate in liver cells via the process of steatosis (i.e. abnormal retention of lipids within a cell).

2. Causes: Fatty liver is commonly associated with alcohol or metabolic syndrome (diabetes, hypertension, obesity and dyslipidemia).

3. Diagnosis of Fatty Liver: in routine blood screening or images of the liver obtained by an ultrasound test, CT (computed tomography) scan, or MRI (magnetic resonance imaging) may suggest the presence of a fatty liver or liver biopsy, in which a small sample of liver tissue is obtained through the skin and analyzed under the microscope.

4. The treatment of fatty liver is related to the cause. It is important to remember that simple fatty liver may not require treatment. The benefit of weight loss, dietary fat restriction, and exercise in obese patients is inconsistent. Reducing or eliminating alcohol use can improve fatty liver due to alcohol toxicity. Controlling blood sugar may reduce the severity of fatty liver in patients with diabetes.

THE SPECTRUM OF NON-ALCOHOLIC FATTY LIVER DISEASE (NAFLD)
Obesity

Obesity is essentially an excessive accumulation of triacylglycerols in fatty tissue that is the net result of excessive energy intake compared to energy usage. Severe forms of the disease are most likely to have a predominantly genetic basis and this is probably polygenic. The 'thrifty gene' hypothesis also describes the disturbance that a modern environment, including higher energy intake and decreased physical activity, has on otherwise advantageous genetic variations. While the physical consequences of obesity, such as arthritis, are debilitating and costly, the metabolic consequences are the drivers behind the modern epidemics of insulin resistance, diabetes, fatty liver disease, coronary artery disease, hypertension and polycystic ovary syndrome. The pathophysiological mechanisms behind these diseases are probably a combination of the toxic metabolic effects of free fatty acids and adipokines - the numerous messengers that adipose tissue has been discovered to produce.

Causes

1. Insufficient sleep
2. Endocrine disruptors (environmental pollutants that interfere with lipid metabolism
3. Decreased variability in ambient temperature
4. Decreased rates of smoking, because smoking suppresses appetite
5. Increased use of medications that can cause weight gain (e.g., atypical antipsychotics)
6. Proportional increases in ethnic and age groups that tend to be heavier
7. Pregnancy at a later age (which may cause susceptibility to obesity in children)
8. Epigenetic risk factors passed on generationally
9. Natural selection for higher BMI
10. Assortative mating leading to increased concentration of obesity risk factors (this would increase the number of obese people by increasing population variance in weight).

According to the Endocrine Society, there is "growing evidence suggesting that obesity is a disorder of the energy homeostasis system, rather than simply arising from the passive accumulation of excess weight".

**Effect on health**

Excessive body weight is associated with various diseases and conditions, particularly cardiovascular diseases, diabetes mellitus type 2, obstructive sleep apnea, certain types of cancer, osteoarthritis, and asthma. As a result, obesity has been found to reduce life expectancy.
Classification of Amino Acids

[A]. Classification based on structure

A comprehensive classification of amino acids is based on their structure and chemical nature. The 20 amino acids found in proteins are divided into seven distinct groups:

1. Amino acids with aliphatic side chains

   These are monoamino monocarboxylic acids. This group consists of the most simple amino acids — glycine, alanine, valine, leucine, and isoleucine.

   - Glycine (Gly, G)  
   - Alanine (Ala, A)  
   - Valine (Val, V)  
   - Leucine (Leu, L)  
   - Isoleucine (Ile, I)

   The last three amino acids (Leu, Ile, Val) contain branched aliphatic side chains, hence they are referred to as branched chain amino acids.

2. Hydroxyl group containing amino acids

   Serine, threonine, and tyrosine are hydroxyl group containing amino acids. Tyrosine — being aromatic in nature — is usually considered under aromatic amino acids.

   - Serine (Ser, S)  
   - Threonine (Thr, T)  
   - Tyrosine (Tyr, Y)
Cysteine with sulphhydryl group and methionine with thioether group are the two amino acids containing sulphur.

- Cystine is another important sulfur containing amino acid formed by condensation of two molecules of cysteine.

\[
\begin{align*}
\text{Cysteine (Cys, C)} & \quad \text{(Sulphhydryl)} & \quad \text{Methionine (Met, M)} \\
\text{(Thioether)} & \\
\end{align*}
\]

4. **Acidic amino acids and their amides**

Aspartic acid and glutamic acid are dicarboxylic monoamino acid while asparagine and glutamine are their respective amide derivatives.

\[
\begin{align*}
\text{Aspartic acid (Asp, D)} & \quad \text{(\(\beta\)-Carboxyl)} \\
\text{Asparagine (Asn, N)} & \quad \text{(Amide)} \\
\text{Glutamic acid (Glu, E)} & \quad \text{(\(\gamma\)-Carboxyl)} \\
\text{Glutamine (Gln, Q)} & \quad \text{(Amide)} \\
\end{align*}
\]

5. **Basic amino acids**

The three amino acids lysine, arginine, and histidine are dibasic mono carboxylic acids. They are highly basic in character.
Classification based on Polarity

Amino acids are classified into following groups based on their Polarity.

**AMINO ACIDS**

Hydrophobic amino acids
- non-Polar
  - R = alkyl
    - Glycine
    - Alanine
    - Valine
    - Leucine
    - Isoleucine
    - Methionine
    - Proline

Hydrophilic amino acids
- Polar
  - R = aromatic
    - Phenylalanine
    - Tryptophan
  - Acidic
    - Glutamic acid
    - Aspartic acid
  - Basic
    - Lysine
    - Histidine
    - Arginine
  - Neutral
    - Tyrosine
    - Serine
    - Threonine
    - Cysteine
    - Glutamine
    - Asparagine

Chemical nature of Amino acids

(A) Reactions due to -COOH group:

1. Amino acids form salts (-COONa) with bases and esters (-COOR') with alcohols.

2. Decarboxylation: Amino acids undergo decarboxylation to produce corresponding amine.

   \[ R-CH-COO^- + NH_3 \xrightarrow{} R-CH_2^- + CO_2 + NH_4^+ \]

3. Reaction with ammonia: The carboxyl group of dicarboxylic amino acids reacts with NH_3 to form amide.

   - Aspartic acid + NH_3 \rightarrow Asparagine
   - Glutamic acid + NH_3 \rightarrow Glutamine
Reactions due to -NH₂ group:

1. The amino groups behave as bases and combine with acids (e.g., HCl) to form salts (-NH₃⁺Cl⁻).

2. Reaction with ninhydrin: The α-amino acids react with ninhydrin to form a purple, blue, or pink complex. Ninhydrin reaction is effectively used for the quantitative determination of amino acids and proteins.

3. Transamination: Transfer of an amino group from amino acid to a keto acid to form a new amino acid is a very important reaction in amino acid metabolism.

4. Oxidative deamination: The amino acids undergo oxidative deamination to liberate free ammonia.

5. Aromatic amino acids: Phenylalanine, tyrosine, tryptophan (C indole ring) are aromatic amino acids. Besides these, histidine may also be considered under this category.

   - Phenylalanine (Phe, F)
   - Tyrosine (Tyr, Y)
   - Tryptophan (Trp, W)

6. Imino acids: Proline contains pyrroolidine ring and a unique amino acid.

   It has an imino group (-NH) instead of an amino (NH₂) group found in other amino acids.

   Therefore, proline is an imino acid, not an amino acid.
Functions of Proteins

Structural
- Certain proteins perform brick and mortar roles and are primarily responsible for structure and strength of body. These include collagen and elastin found in bone matrix, vascular system and other organs and a-keratin present in epidermat tissues.

Dynamic
- Dynamic functions of proteins includes protein acting as enzymes, hormones, blood clotting factors, immunoglobulins, membrane receptors. Storage proteins etc.

They also helps in genetic control, muscle contraction, respiration etc.

Amino Acids

Amino acids are a group of organic compounds containing two functional groups — amino and carboxyl. The amino group (-NH₂) is basic while carboxyl group (-COOH) is acidic in nature.

Structure:

\[
\text{R-} \left \{ \begin{array}{c}
\text{C} \\
\text{H} \\
\text{N} \\
\text{C} \\
\text{H}_2 \\
\text{C-OH}
\end{array}\right 
\]

General Structure

In a-amino acids, both amino and carboxyl groups are attached to the same carbon atom. Alpha carbon (\(\alpha\)) is a chiral centre, that is to say, this carbon atom is attached to four different groups. So amino acids are optically active molecules.
The only exception is glycine, the simplest amino acid, in which, 
\[ R = H. \]

\[
\begin{align*}
\text{H} & \quad \text{C} \quad \text{COOH} \\
\text{NH}_2 & \\
\text{Glycine} \rightarrow \text{optically inactive.}
\end{align*}
\]

**Fischer Projection**

Commonly amino acids are represented as follows:

\[
\begin{align*}
\text{COOH} \\
\text{H}_2\text{N} & \quad \text{C} \quad \text{H} \\
\text{R} & \\
\end{align*}
\]

- **D and L amino acids** are mirror images of each other and are non-superimposable on each other. Pairs of amino acids like these are called enantiomers.

- **only L-amino acids** are constituents of proteins. Our body synthesizes most of its own L-amino acids; these then incorporated into proteins.

**Note:** Proteins are catalysts for most of the biochemical reactions that take place in our body.
Proteins are nitrogen-containing macro-molecules consisting of L-α-amino acids as repeating units. They perform a wide variety of static (structural) and dynamic (enzyme, hormone, clotting factors, receptors, etc.) functions.

- The proteins on degradation release individual amino acids; then these amino acids metabolized in the body.

![Amino acid metabolism diagram]

**General Aspects of Amino Acids Metabolism**

- The amino group of the amino acids is utilized for the formation of urea, which is an excretory end product of protein metabolism.
- The carbon skeleton of the amino acids is first converted to keto acids (by transamination) and meet one or more of the following fates:
  1. Utilized to generate energy.
  2. Used for the synthesis of glucose.
  3. Diverted for the formation of fat or ketone bodies.
  4. Involved in the production of non-essential amino acids.

**General reaction:** can be used to describe AA metabolism are—

**Transamination:** The transfer of amino \((\text{NH}_2)\) group from an amino acid to a keto acid is KGA transamination. This process involves the interconversion of a pair of amino acids and a pair of keto acids.

- This process is catalyzed by a group of enzymes called **transaminases (amino-transferases)**.
Transamination, a chemical reaction that transfers an amino group from an amino acid to keto acid to form new amino acid.

\[ \text{Amino acid-I} \xrightarrow{\text{NH}_3} \text{Keto acid-I} \]

\[ \text{Keto acid-II} \quad \text{Amino acid-II} \]

**Key Points:***

1. All transaminases require Pyridoxal Phosphate (PLP), a coenzyme derived from vitamin B6.
2. Specific transaminases exist for each pair of amino acids and keto acids.
3. There is no free \( \text{NH}_3 \) liberated, only the transfer of amino group occurs.
4. Transamination is reversible.
5. Transamination is very important for the redistribution of aminogroups and production of non-essential amino acids as per requirement of the cell.
6. Transamination diverts the excess amino acids towards energy generation.
7. The amino acids undergo transamination to finally concentrate nitrogen in glutamate. Glutamate is only AA that undergoes oxidative deamination to liberate free \( \text{NH}_3 \) for urea synthesis.
8. All amino acids except lysine, threonine, proline, and hydroxyproline participate in transamination.
Deamination - the removal of amino group from the amino acid as NH₃ by deamination. Deamination results in the liberation of NH₃ for urea synthesis.

- simultaneously, the carbon skeleton of AA is converted to keto acids.

Deamination may be either oxidative or non-oxidative.

I: Oxidative deamination - oxidative deamination is the liberation of free ammonia from the amingroup of amino group of amino acids coupled with oxidation. This takes place mostly in liver and kidney.

• The purpose of oxidative deamination is to provide NH₃ for urea synthesis and keto acids for a variety of reactions, including energy generation.

• The glutamate (glutamic acid), serves as a collection centre for amino groups in the biological system. Glutamate rapidly undergoes oxidative deamination, catalysed by glutamate dehydrogenase (GDH) to liberate ammonia.

\[
\begin{align*}
\text{COO}^- & \quad \text{NAD}^+ \quad \text{NADH}^{+H^+} \\
\text{CH} & \quad \text{CH} \\
\text{CH} & \quad \text{CH} \\
\text{CH}_2 & \quad \text{NH}_2 \\
\text{COO}^- & \quad \text{H}_2\text{O} \\
\end{align*}
\]

L-Glutamate \quad \xrightarrow{\text{NAD}^+} \quad \xrightarrow{\text{H}_2\text{O}} \quad \alpha\text{-amino glutarate}

\[
\begin{align*}
\text{COO}^- & \quad \text{NAD}^+ \quad \text{NADH}^{+H^+} \\
\text{CH} & \quad \text{CH} \\
\text{CH} & \quad \text{CH} \\
\text{CH}_2 & \quad \text{NH} \quad \text{H}_2\text{O} \\
\text{COO}^- & \quad \text{C} \quad \text{COO}^- \\
\end{align*}
\]

\( \xrightarrow{\text{H}_2\text{O}} \) \quad \alpha\text{-keto glutarate}

\[
\begin{align*}
\text{HOOC-CH}_2\text{-CH}_2\text{-CH} \quad \overset{6}{\text{HN}} \\
\text{C-COOH} & \quad \overset{\text{NADH}^{+H^+}}{\text{O}} \\
\text{NAD}^+ & \quad \text{NAD}^+ \\
\end{align*}
\]

\( \text{x-keto glutarate \rightarrow glutamic acid} \)

\[
\begin{align*}
\text{NH}_2 & \quad \text{O} \\
R-\text{CH} \quad \overset{\text{R-COOH}}{\text{COOH}} \\
\text{transamination} & \quad \text{any amino acid} \\
\end{align*}
\]

Fig: Glutamate and oxidative deamination

• Conversion of glutamate to x-keto glutarate occurs through the formation of an intermediate, α-amino glutarate.
Glutamate dehydrogenase (Gdh) is a zinc-containing mitochondrial enzyme and is controlled by allosteric regulation. GTP and ATP inhibit—whereas GDP and ADP activate—glutamate dehydrogenase.

II. Non-oxidative deamination—some of the amino acids can be deaminated to liberate NH₃ without undergoing oxidation.

- Serine, threonine, and homoserine are the hydroxy amino acids. They undergo non-oxidative deamination catalysed by dehydratases.

```
Serine                   Dehydratase  \---\ Respective α-ket acids
Theonine                 NH₃
Homoserine               
```

**Eg.**
```
\[
\begin{array}{c}
\text{NH}_2 \\
\text{H-C-COOH} \\
\text{CH}_3 \\
\text{OH}
\end{array}
\rightarrow
\begin{array}{c}
\text{Serine dehydratase} \\
\text{NH}_2 \\
\text{C-COOH} \\
\text{CH}_2 \\
\text{H}_2\text{O}
\end{array}
\rightarrow
\begin{array}{c}
\text{Aminoacylase} \\
\text{OH} \\
\text{C-COOH} \\
\text{CH}_3 \\
\text{NH}_3
\end{array}
\]
```

- The sulfur amino acids, namely cysteine and homocysteine, undergo deamination coupled to desulphhydration to give keto acids.

```
\text{Cysteine \rightarrow Desulphhydrase \rightarrow Pyruvate}
\rightarrow \text{NH}_3 + \text{H}_2\text{S}
```

- The enzyme histidase acts on histidine to liberate NH₃ by a non-oxidative deamination process.

```
\text{Histidine \rightarrow Histidase \rightarrow Urocanate}
\rightarrow \text{NH}_3
```

* The reactions involved in the catabolism of amino acids are decarboxylation, transulfuration, desulfuration, dehydroxylation etc.
Decarboxylation

* The decarboxylation process is important since the products of decarboxylation reactions give rise to physiologically active amines.

* The enzyme takes part in decarboxylation of amino acids are amino acid decarboxylase, pyridoxal phosphate dependent enzymes.

* The physiologically active amines, epinephrine, nor-epinephrine, dopamine, serotonin, γ-aminobutyrate and histamine are formed through decarboxylation of the corresponding Precursor amino acids.

\[
\text{R-CH-COONH}_2 \xrightarrow{\text{Decarboxylase}} \text{R-CH}_2\text{NH}_2
\]

<table>
<thead>
<tr>
<th>S.No.</th>
<th>Amino acid</th>
<th>Amine</th>
<th>Function(s)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Serine</td>
<td>Ethanolamine</td>
<td>Forms Choline</td>
</tr>
<tr>
<td>3.</td>
<td>Phenylalanine</td>
<td>Dopamine</td>
<td>For synthesis of nor-epinephrine and epinephrine</td>
</tr>
<tr>
<td>4.</td>
<td>Tyrosine</td>
<td>Tyramine</td>
<td>Vasodilator, increase blood pressure.</td>
</tr>
<tr>
<td>5.</td>
<td>Tryptophan</td>
<td>Tryptamine, serotonin, melatonin</td>
<td>Elevates blood pressure, stimulates cerebral activity, Circadian rhythms.</td>
</tr>
</tbody>
</table>

Transport and storage of NH₃: Despite a regular and constant production of NH₃ from various tissues, its concentration in the circulation is surprisingly low (10-20 mg/dl). This is mostly because the body has an efficient mechanism for NH₃ transport and its immediate utilization for urea synthesis.
**Urea cycle** (Krebs-Henseleit cycle)

Urea is the end product of protein metabolism (amino acid metabolism). Urea is synthesized in liver and transported to kidney for excretion in urine.

- Urea has two amino (-NH₂) groups, one derived from NH₃ and the other from aspartate. Carbon atom is supplied by CO₂.
- Five-step cyclic process, with five distinct enzymes. The first two enzymes are present in mitochondria while the rest are localized in cytosol.

---

**Figure 2:** Reactions of urea cycle
**Urea Cycle**

**Step 1:** Synthesis of carbamoyl phosphate. Carbamoyl phosphate synthase I (CPSI) of mitochondria catalyzes the condensation of NH₃ with CO₂ to form carbamoyl phosphate. This step consumes two ATP and is irreversible, and rate limiting.

**Step 2:** Formation of citrulline: citrulline is synthesized by carbamoyl phosphate and ornithine by ornithine transcarbamoylase. Citrulline produced in this reaction is transported to cytosol by a transporter system.

**Step 3:** Synthesis of arginosuccinate: arginosuccinate synthase condenses citrulline with aspartate to produce arginosuccinate. The second amino group of urea is incorporated in this reaction.

**Step 4:** Cleavage of arginosuccinate: arginosuccinase cleaves arginosuccinate to give arginine and fumarate. Arginine is the immediate precursor for urea.

Fumarate liberated here provides a connecting link with TCA cycle, gluconeogenesis etc.

**Step 5:** Formation of urea: Arginase is the fifth and final enzyme that cleaves arginine to yield urea and ornithine. Ornithine is regenerated, enters mitochondria for its reuse in the urea cycle.

### Metabolic Disorders of Urea Cycle

Metabolic defects associated with each of the five enzymes of urea cycle have been reported

<table>
<thead>
<tr>
<th>Defect</th>
<th>Enzyme Involved</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hyperammononemia type-I</td>
<td>Carbamoyl Phosphate Synthase I</td>
</tr>
<tr>
<td>Hyperammononemia type-II</td>
<td>Ornithine transcarbamoylase</td>
</tr>
<tr>
<td>Citrullinemia</td>
<td>Arginosuccinate Synthase</td>
</tr>
<tr>
<td>Arginosuccinic aciduria</td>
<td>Arginosuccinase</td>
</tr>
<tr>
<td>Hyperargininemia</td>
<td>Arginase</td>
</tr>
</tbody>
</table>
All disorders invariably lead to a build-up in blood ammonia (hyperammonemia), leading to toxicity. Other metabolites of urea cycle also accumulate enzyme defect.

The clinical symptoms associated with defect in urea cycle enzymes include vomiting, lethargy, irritability, ataxia, and mental retardation. The loss of full control of body movements (loss of body coordination).

Metabolism of Phenylalanine & Tyrosine

Phenylalanine (Phe, F) and tyrosine (Tyr, Y) are structurally related aromatic amino acids. Phenylalanine is an essential amino acid while tyrosine is non-essential. The predominant metabolism of Phenylalanine occurs through tyrosine. Tyrosine is involved in the synthesis of a variety of biologically important compounds—epinephrine, norepinephrine, dopamine, thyroid hormone, and the pigment melanin.

Conversion of Phenylalanine to Tyrosine

Phenylalanine is hydroxylated at para-position by phenylalanine hydroxylase to produce tyrosine (P-hydroxyphenylalanine). This reaction is irreversible and requires biotin as a coenzyme.

![Diagram showing the metabolism of Phenylalanine and Tyrosine]
The enzyme phenylalanine hydroxylase is present in the liver.

Degradation of Tyrosine (Phenylalanine) - The sequence of the reaction in the degradation of these amino acids, depicted as:

1. A Single pathway is responsible for the degradation of both these amino acids, and occurs mostly in liver.

2. Tyrosine first undergoes transamination to give P-hydroxyphenylpyruvate.
   This reaction is catalysed by tyrosine transaminase.

\[
\text{Phenylalanine} \xrightarrow{\text{Phenylalanine hydroxylase}} \text{Tyrosine} \\
\xrightarrow{\text{KGl}} \text{Glu} \\
\xrightarrow{\text{Tyrosine transaminase}} \text{P-hydroxyphenylpyruvate} \\
\xrightarrow{\text{Ascorbate}} \text{P-hydroxyphenylpyruvate dioxygenase (Cu^{2+})} \\
\xrightarrow{\text{O}_{2}} \text{Homogentisate oxidase (Fe^{2+})} \\
\xrightarrow{\text{O}} \text{4-Maleylacetoacetate} \\
\xrightarrow{\text{4-Fumaryl-acetoacetate hydrolase}} \text{Acetoacetate} \\
\xrightarrow{\text{Fat}} \text{Fumarate} \\
\xrightarrow{\text{TCA cycle}} \text{Glucose}
\]

Fig: Tyrosine Metabolism

(3) P-Hydroxyphenylpyruvate hydrolase (or dioxygenase) catalyses oxidative decarboxylation as well as hydroxylation of the phenyl ring to produce homogentisate.

- This reaction involves a shift in hydroxyl group from p-position to meta-position and incorporation of new hydroxyl group at p-position.

- This step in tyrosine metabolism requires ascorbic acid.

Contd. . . .
4) Homogenized citratecleaves to benzyl ring of Homogentisate to form 4-maleylacetoacetate. Molecular oxygen is required for this reaction to break the aromatic ring.

(5) Maleylacetoacetate undergoes isomerisation to form 4-fumaryl-acetoacetate and this reaction is catalysed by maleylacetoacetate isomerase.

(6) Fumaryl-acetoacetase (fumaryl acetoacetate hydrolase) brings about the hydrolysis of fumaryl acetoacetate to liberate fumarate and acetooacetate.

- Fumarate is intermediate of citric acid cycle – can serve as Precursor of glucose.
- Acetoacetate is a ketone body from c fat can be synthesized.

**Metabolic disorders:** The inborn errors of Phenylalanine and Tyrosine metabolism are indicated as –

**Phenylketonuria:** Most common metabolic disorder in amino acid metabolism (1 in 10,000 births). It is due to the deficiency of Hepatic enzyme Phenylalanine hydroxylase caused by an autosomal recessive gene.

- This enzyme deficiency impairs the synthesis of Tetrahydrobiopterin required for the action of Phenylalanine hydroxylase. The net outcome in PKU is that Phenylalanine is not converted to Tyrosine.

```
Phenylalanine \[\xrightarrow{\text{Tetrahydrobiopterin}}\] Tyrosine
```

**Fig:** metabolites that accumulate in Phenylketonuria

- Phenylketonuria Primarily causes the accumulation of Phenylalanine in tissues and blood and results in its increased excretion in urine.
- In Case of Phenylketonuria, Phenylalanine is diverted to alternative pathways, result in the excessive production of Phenylpyruvate, Phenyl lactate and Phenylacetyl-glutamine → All these excreted in urine.
The enzyme phenylalanine hydroxylase is found in the liver.

Degradation of Tyrosine (Phenylalanine) - The sequence of the reactions in the degradation of these amino acids, depicted as:

1. A single pathway is responsible for the degradation of both these amino acids, which occurs mostly in the liver.

2. Tyrosine first undergoes transamination to give P-hydroxyphenylpyruvate. This reaction is catalysed by tyrosine transaminase.

Fig: Tyrosine Metabolism

(3) P-hydroxyphenylpyruvate hydroxylase (or dioxygenase) catalyses oxidative decarboxylation as well as hydroxylation of the phenyl ring to produce homogentisate.

- This reaction involves a shift in the hydroxyl group from p-position to meta-position and incorporation of a new hydroxyl group at p-position.

- This step in tyrosine metabolism requires ascorbic acid.
(4) Homogentisate oxidase clears the benzenoid ring of homogentisate to form 4-maleylacetoacetate. Molecular oxygen is required for this reaction to break the aromatic ring.

(5) Maleylacetoacetate undergoes isomerisation to form 4-fumaryl-acetoacetate and this reaction is catalysed by maleylacetoacetate isomerase.

(6) Fumaryl-acetoacetase (fumaryl acetoacetate hydratase) brings about the hydrolysis of fumaryl acetoacetate to liberate fumarate and acetoacetate.

- Fumarate is intermediate of citric acid cycle → can serve as precursor of gluconeogenesis.
- Acetoacetate is a ketone body from C fat can be synthesized.

**Metabolic disorders:** The inborn errors of phenylalanine and tyrosine metabolism are indicated as:

**Phenylketonuria** - Most common metabolic disorder in amino acid metabolism (1 in 10,000 births). It is due to the deficiency of hepatic enzyme phenylalanine hydroxylase caused by an autosomal recessive gene.

- This enzyme deficiency impairs the synthesis of tetrahydrobiopterin required for the action of phenylalanine hydroxylase. The net outcome in PKU is that phenylalanine is not converted to tyrosine.

\[
\text{Phenylalanine} \xrightarrow{\text{Phenylalanine hydroxylase}} \text{Tyrosine}
\]

\[
\text{NAD}^+ \quad \text{Phenylpyruvate (keto acid)} \quad \text{Phenylpyruvate}
\]

\[
\text{Phenylacetate} \quad \text{Glutamine} \quad \text{Phenyllactate} \quad \text{Phenylacetyl-glutamine}
\]

\[
\text{H}_2\text{O} \quad \text{NADH} + \text{H}^+ \quad \text{Phenylacetate}
\]

\[
\text{NAD}^+ \quad \text{Phenylpyruvate (keto acid)} \quad \text{Phenylpyruvate}
\]

**Fig:** metabolites that accumulate in Phenylketonuria

- Phenylketonuria primarily causes the accumulation of phenylalanine in tissues and blood and results in its increased excretion in urine.
- In case of Phenylketonuria, phenylalanine is diverted to alternate pathways, resulting in the excessive production of phenylpyruvate, phenyl-lactate and phenylacetyl-glutamine → all these excreted in urine.
Classical manifestations of PKU

- Effect on CNS: mental retardation, failure to walk or talk, failure of growth, seizures and tremor. If untreated, the patient shows very low IQ.
- Effect on pigmentation: melanin is the pigment synthesized from tyrosine by tyrosinase. Accumulation of phenylalanine competitively inhibits tyrosinase and impairs melanin formation.
  - The result is hypopigmentation that causes light skin colour, fair hair, blue eyes, etc.

Treatment in PKU:

- Select foods with low phenylalanine content and/or feeding synthetic amino acid preparations, low in phenylalanine.
- Early diagnosis and treatment for 4-5 years can prevent the damage to brain. The restriction to protein diet should be continued for many more years.
- Since the amino acid tyrosine cannot be synthesized in PKU patients, it becomes essential and should be provided in the diet in sufficient quantity.
- In some seriously affected PKU patients, treatment includes administration of 5-hydroxytryptophan and dopa to restore the synthesis of serotonin and catecholamines.

ALKEPTONURIA (Black urine disease)

- Autosomal recessive disorder, 1 in 25,000.
- The defective enzyme in alkeptonuria is homogentisate oxidase in tyrosine metabolism.
- Homogentisate accumulates in tissues and blood, and is excreted into urine. Homogentisate, on standing gets oxidized to the corresponding quinones, which polymerize to give black or brown colour. Urin resembles coke in colour.
- Homogentisate $\xrightarrow{\text{oxidase}}$ 4-maleylacetacetate
- 4-Maleylacetacetate $\rightarrow$ Benzquinone acetate
- Polymerisation
- Alkalization
- Binds to tissues
Biochemical manifestations: Homogenized fats oxidized by polyphenol oxidase to benzoquinone acetate undergoes polymerization to produce pigments called alkapton. Alkapton deposition occurs in connective tissues, bones, and various organs (nose, ear etc.) resulting in a condition known as ochronosis.

- Many alkaptonuric patients suffer from arthritis (alkapton in the joints).

Treatment: Alkaptonuria is not a dangerous disorder and therefore, does not require any specific treatment. However, consumption of protein diet with relatively low phenylalanine content is recommended.

TYROSINEMIA

Tyrosinemia type-I: This is due to the deficiency of the enzymes fumarylacetoacetate hydroxylase and/or malaylacetoacetate isomerase. Also called tyrosinosis. It is rare but serious disorder.

- It causes liver failure, rickets, renal tubular dysfunction and polyneuropathy.
- Tyrosine, its metabolites and many other amino acids are excreted in urine.
- In acute tyrosinosis, the infant exhibits diarrhea, vomiting and 'cabbage-like' odor. Death may even occur due to liver failure within one year.
- Treatment: diet low in tyrosine, phenylalanine, and methionine are recommended.

Tyrosinemia type-II (Richner-Hanhart syndrome)

This is due to a defect in the enzyme tyrosine transaminase. The result is a blockade in the routine degradative pathway of tyrosine.

- Accumulation and excretion of tyrosine and its metabolites - p-hydroxyphenylpyruvate, p-hydroxyphenyl-lactate, hydroxy-phenylacetate, N-acetyl-tyrosine - and tyramine are observed.
- Characterized by skin (dermatitis) and eye lesions and early mental retardation. A disturbed self-coordination is seen in these patients.
ALBINISM

(Greek: albino - white) - Inborn error, due to the lack of synthesis of the pigment melanin. Autosomal recessive disorder with a frequency of 1 in 20,000. The melanin synthesis can be influenced by a variety of factors.

Many possible causes for albinism have been identified:

1. Deficiency or lack of the enzyme tyrosinase.
2. Decrease in melanosomes of melanocytes.
3. Impairment in melanin polymerization.
4. Lack of protein matrix in melanosomes.
5. Limitation of substrate (tyrosine) availability.
6. Presence of inhibitors of tyrosinase.

The most common cause of albinism is a defect in tyrosinase, the enzyme most responsible for the synthesis of melanin.

Clinical manifestations - The most important function of melanin is the protection of body from sun radiation. Lack of melanin in albinos makes them sensitive to sunlight. Increased susceptibility to skin cancer (Carcinoma) is observed. Photophobia (intolerance to light) is associated with lack of pigment in the eyesight of albinos.
Biosynthesis of Serotonin (5-HT) and Melatonin

Serotonin and melatonin are synthesized by the metabolism of tryptophan by the serotonin pathway:

\[ \begin{align*}
\text{Tryptophan (AA)} & \rightarrow 5-\text{Hydroxytryptophan} \\
& \rightarrow \text{Serotonin} \\
& \rightarrow \text{N-Acetylseryotonin} \\
& \rightarrow \text{Melatonin}
\end{align*} \]

- Serotonin (5-HT) is a neurotransmitter, synthesized from tryptophan. About 1% of the tryptophan is converted to serotonin in the target tissues.
- The largest amount of serotonin is synthesized in the intestinal cells.
- Tryptophan is first hydroxylated at the 5th position by tryptophan hydroxylase.
- 5-Hydroxytryptophan is decarboxylated by aromatic amino acid decarboxylase (PLP dependent) to give serotonin.

Functions of Serotonin:
- Serotonin is a neurotransmitter and performs a variety of functions:
  1. Powerful vasoconstrictor & results in smooth muscle contraction in bronchioles and arterioles.
(2) Closely involved in the regulation of cerebral activity (excitation).

(3) 5-HT controls the behavioural patterns, sleep, blood pressure and body temperature.

(4) Serotonin evokes the release of peptide hormones from gastrointestinal tract.

(5) It is also necessary for motility of GIT (Peristalsis).

**Serotonin and brain** - The brain itself synthesizes 5HT. The outside serotonin cannot enter the brain due to blood-brain barrier. Serotonin is a stimulator of brain activity, hence its deficiency causes depression.

**Melatonin**

Melatonin is a hormone mostly synthesized by the pineal gland.

- Serotonin produced from tryptophan is acted upon by serotonin N-acetylase to give N-acetylserotonin.
- The latter undergoes methylation to produce melatonin or N-acetyl-5-methoxyserotonin.

The synthesis and secretion of melatonin from pineal gland is controlled by light.

**Functions of melatonin**

(1) Melatonin is involved in circadian rhythms or diurnal variations (24 hrs cyclic process) of the body. It plays a significant role in sleep and wake process.

(2) Melatonin inhibits the production of melanocyte stimulating hormone (MSH) and adrenocorticotropic hormone (ACTH).

(3) It has some inhibitory effect on ovarian function.

(4) Melatonin also performs a neurotransmitter function.
Tyrosine is the precursor for the synthesis of catecholamines, namely dopamine, noradrenaline and adrenaline.

* The conversion of tyrosine to catecholamines occurs in adrenal medulla and CNS involving the following reactions:

- Tyrosine is hydroxylated to 3,4-dihydroxy-phenylalanine (DOPA) by tyrosine hydroxylase. In contrast to this enzyme, tyrosinase present in melanocytes converts tyrosine to DOPA. Hence two different enzyme systems exist to convert tyrosine to DOPA.
- DOPA undergoes PLP-dependent decarboxylation to give dopamine, in turn is hydroxylated to produce norepinephrine.
- Methylation of norepinephrine gives epinephrine by N-methyltransferase.
Function of Catecholamines: Norepinephrine and epinephrine regulate carbohydrate and lipid metabolism. They cause an increase in the blood pressure.

- Dopamine and norepinephrine serve as neurotransmitters in the brain and autonomous nervous system.
- Catecholamines can serve as neurotransmitters, transferring signals from neuron to neuron, as well as hormones, to regulate physiological functions such as your heartbeat and breathing rate.

Catabolism of Heme

Erythrocytes have a life span of 120 days. At the end of this period, they are taken up and degraded by the macrophages of the reticuloendothelial (RE) system in the spleen and liver.

- The hemoglobin is cleaved to the protein part globin and non-protein heme.
- About 6 g of hemoglobin per day is broken down, and resynthesized in an adult man (70 kg).
- It is estimated that about 90% of the heme that is subjected to degradation comes from erythrocytes, and the rest (20%) comes from immature RBC, myoglobin, and cytochromes.

Heme oxygenase: A complex microsomal enzyme namely oxygenase utilizes NADPH and O₂ and cleaves the methenyl bridge between the two pyrrole rings (A and B) to form biliverdin. Simultaneously, Fe⁴⁺ is oxidized to ferric form (Fe³⁺) and released.

- The products of heme oxygenase reaction are biliverdin (a green pigment) Fe³⁺ and carbon monoxide (CO).

Biliverdin is excreted in birds and amphibians while in mammals it is further degraded.

Biliverdin reductase: Biliverdin’s methenyl bridge (blw pyrrole ring C₄0) is reduced to methylene group to form bilirubin (yellow pigment). This reaction is catalyzed by enzyme bilirubin reductase.
One gram of hemoglobin on degradation (finally yields about 35 mg of bilirubin).

- Bilirubin is transported to liver in form of bilirubin–albumin complex.
  As the albumin–bilirubin complex enters the liver, bilirubin dissociates, and it is taken up by hepatocyte by a carrier-mediated active transport.
- Inside the hepatocytes, bilirubin binds to a specific intracellular protein, namely ligandin.

In liver, bilirubin is conjugated to two molecules of glucuronate supplied by UDP-glucuronate. This reaction, catalyzed by bilirubin glucuronyltransferase, results in the formation of water-soluble bilirubin diglucuronide. When bilirubin is in excess, bilirubin monoglucuronide also accumulates in the body.

**Diagram:**

- **Aged Erythrocyte** → Macrophage → Hemoglobin (α, β chain) → globin → Amino acids → Reutilized or degraded
  
  - **Heme** → NADPH + H⁺ + O₂ → NADP⁻⁺ → Heme Oxygenase → Fe²⁺, CO
  - **Biliverdin** → Biliverdin reductase → Bilirubin

  - **Bilirubin–albumin Complex** → Blood

  - **UDP-glucuronate** → **Bilirubin glucuronyltransferase** → Bilirubin diglucuronide (to bile) → Microbial enzymes (Intestine)

  - Urobiligen → Kidney
  - Urobilin → To Urin
  - Stercobilin → To Feces

  - **Liver**
Jaundice

The normal serum total bilirubin concentration is in the range of 0.2 to 1.0 mg/dl (Conjugated & unconjugated).

1. Jaundice (French: Jaune-Yellow) is a clinical condition characterized by yellow color of the white of the eyes and skin. It is caused by deposition of bilirubin due to its elevated levels in the serum.

2. The term hyperbilirubinemia is often used to represent the increased Concentration of serum bilirubin.

1. Hemolytic Jaundice: This condition is associated with increased hemolysis of erythrocytes (eg. malaria, sickle-cell anemia, incompatible blood transfusion).

2. Overproduction of bilirubin and excrete the same in urine.

3. Feces becomes dark brown due to high content of stercobilinogen.

2. Hepatic Jaundice: This type of Jaundice is caused by dysfunction of the liver due to damage to the parenchymal cells.

1. Due to viral infection (viral hepatitis, poisons and toxin), cirrhosis of liver, cardiac failure etc. Viral hepatitis is most common.

2. Increased levels of conjugated and unconjugated bilirubin in the serum.

3. Dark colored urine due to the excessive excretion of bilirubin and urobilinogen.

4. Increased activities of alanine transaminase (SGPT) and aspartate transaminase (SGOT) released into circulation due to damage to hepatocyte.

5. Pale, clay colored stools due to the absence of stercobilinogen.

6. Nausea & anorexia - Common Symptoms

3. Obstructive Jaundice: Due to a obstruction in the bile duct that prevents the passage of bile into intestine. The obstruction may be caused by gallstones, tumors etc. Due to the blockage in the bile duct, the conjugated bilirubin from liver enters the circulation.

4. Increased concentration of conjugated bilirubin in serum.

5. Serum alkaline phosphate is elevated (dark colored urine, clay colored feces)