B. Pharmacy 2nd Semester - Biochemistry (Unit 3)

POINTS TO BE COVERED IN THIS TOPIC

- ➤ **LIPID METABOLISM** 🧬
- ➤ **AMINO ACID METABOLISM** 🔬

LIPID METABOLISM 🧬

Lipid metabolism encompasses the complex biochemical processes involved in the synthesis, breakdown, and utilization of lipids in living organisms. These processes are essential for energy production, membrane formation, and synthesis of important biological molecules.

β-OXIDATION OF SATURATED FATTY ACID (PALMITIC ACID) ⚡

β-Oxidation is the catabolic process by which fatty acid molecules are broken down in the mitochondria to generate acetyl-CoA, NADH, and FADH2 for energy production.

PROCESS OF β-OXIDATION:

The β-oxidation of palmitic acid (C16:0) involves the systematic removal of two-carbon units as acetyl-CoA through a cyclic process of four enzymatic reactions.

ACTIVATION PHASE:

- Fatty acid activation occurs in the cytoplasm
- Palmitic acid is converted to palmitoyl-CoA by acyl-CoA synthetase
- ATP is consumed in this activation step
- The activated fatty acid is transported into mitochondria via carnitine shuttle system

OXIDATION CYCLE: Each cycle of β-oxidation involves four sequential reactions:

1. **Oxidation:** Acyl-CoA dehydrogenase removes two hydrogen atoms, forming trans-enoyl-CoA and FADH2
2. **Hydration:** Enoyl-CoA hydratase adds water across the double bond, forming 3-hydroxyacyl-CoA
3. **Oxidation:** 3-hydroxyacyl-CoA dehydrogenase oxidizes the hydroxyl group, forming 3-ketoacyl-CoA and NADH
4. **Thiolysis:** Thiolase cleaves the 3-ketoacyl-CoA, releasing acetyl-CoA and shortened acyl-CoA

ENERGY YIELD:

- Palmitic acid undergoes 7 cycles of β-oxidation
- Produces 8 molecules of acetyl-CoA
- Generates 7 FADH2 and 7 NADH molecules
- Total ATP yield: approximately 129 ATP molecules

FORMATION AND UTILIZATION OF KETONE BODIES; KETOACIDOSIS 🔥

Ketone bodies are water-soluble molecules produced in the liver during periods of prolonged fasting, low carbohydrate intake, or metabolic stress.

KETONE BODY FORMATION (KETOGENESIS):

Primary Ketone Bodies:

- Acetoacetate
- β-hydroxybutyrate
- Acetone (minor component)

Formation Process: The synthesis occurs primarily in liver mitochondria through the following pathway:

- Two acetyl-CoA molecules condense to form acetoacetyl-CoA
- Third acetyl-CoA is added by HMG-CoA synthase to form 3-hydroxy-3-methylglutaryl-CoA (HMG-CoA)
- HMG-CoA lyase cleaves HMG-CoA to produce acetoacetate
- Acetoacetate can be reduced to β-hydroxybutyrate by β-hydroxybutyrate dehydrogenase
- Spontaneous decarboxylation of acetoacetate produces acetone

UTILIZATION OF KETONE BODIES: Extrahepatic tissues utilize ketone bodies as an alternative energy source:

- β-hydroxybutyrate is oxidized back to acetoacetate

- Acetoacetate is activated to acetoacetyl-CoA by succinyl-CoA transferase
- Acetoacetyl-CoA is cleaved by thiolase to produce two acetyl-CoA molecules
- Acetyl-CoA enters the citric acid cycle for ATP generation

KETOACIDOSIS: Ketoacidosis is a pathological condition characterized by excessive ketone body production, leading to metabolic acidosis.

Causes:

- Uncontrolled diabetes mellitus
- Prolonged starvation
- Alcoholism
- Certain metabolic disorders

Clinical Manifestations:

- Decreased blood pH (acidosis)
- Fruity breath odor (acetone)
- Dehydration and electrolyte imbalance
- Altered mental status
- Potential coma in severe cases

DE NOVO SYNTHESIS OF FATTY ACIDS (PALMITIC ACID)

De novo fatty acid synthesis is the anabolic process of constructing fatty acids from simpler precursor molecules, primarily occurring in the liver,

adipose tissue, and mammary glands.

LOCATION AND REQUIREMENTS:

- Primarily occurs in the cytoplasm
- Requires acetyl-CoA as starting material
- Needs NADPH as reducing agent
- Utilizes ATP for energy
- Requires biotin as cofactor

FATTY ACID SYNTHASE COMPLEX: The multienzyme complex responsible for fatty acid synthesis contains seven enzymatic activities:

1. Acetyl-CoA carboxylase (rate-limiting enzyme)
2. Fatty acid synthase complex with multiple domains
3. Acyl carrier protein (ACP) domain
4. Various reductase and synthase activities

SYNTHESIS PROCESS:

Initiation:

- Acetyl-CoA is carboxylated to malonyl-CoA by acetyl-CoA carboxylase
- This reaction requires ATP and biotin as cofactor
- Malonyl-CoA serves as the two-carbon donor for chain elongation

Elongation Cycles: Each elongation cycle involves four reactions:

1. **Condensation:** Malonyl-CoA condenses with the growing fatty acid chain

2. **Reduction:** First reduction using NADPH
3. **Dehydration:** Removal of water molecule
4. **Reduction:** Second reduction using NADPH

Completion:

- Seven cycles of elongation produce palmitic acid (16:0)
- Palmitic acid is released from the fatty acid synthase complex
- Further modifications can occur to produce other fatty acids

REGULATION:

- Acetyl-CoA carboxylase is the rate-limiting enzyme
- Regulated by allosteric effectors and covalent modification
- Citrate activates, while palmitoyl-CoA inhibits
- Hormonal regulation involves insulin (activation) and glucagon/epinephrine (inhibition)

BIOLOGICAL SIGNIFICANCE OF CHOLESTEROL 💎

Cholesterol is a crucial sterol molecule that serves multiple essential functions in cellular physiology and serves as a precursor for various biologically active compounds.

STRUCTURAL FUNCTIONS:

- Essential component of cell membranes
- Maintains membrane fluidity and stability
- Critical for membrane raft formation

- Important for cellular signaling processes

PRECURSOR FUNCTIONS: Cholesterol serves as the precursor for several important biological molecules:

CONVERSION TO BILE ACIDS:

- Primary bile acids: cholic acid and chenodeoxycholic acid
- Synthesized in hepatocytes through 7α-hydroxylase pathway
- Facilitate lipid digestion and absorption
- Aid in cholesterol elimination from the body

STEROID HORMONE SYNTHESIS: Cholesterol is the precursor for all steroid hormones:

- **Mineralocorticoids:** Aldosterone (regulates sodium and potassium balance)
- **Glucocorticoids:** Cortisol (stress response and metabolism)
- **Sex Hormones:**
  - Androgens (testosterone)
  - Estrogens (estradiol)
  - Progestogens (progesterone)

VITAMIN D SYNTHESIS:

- 7-dehydrocholesterol (cholesterol derivative) in skin
- Converted to vitamin D3 upon UV exposure
- Further hydroxylated in liver and kidneys
- Active form: 1,25-dihydroxyvitamin D3 (calcitriol)

- Regulates calcium homeostasis and bone metabolism

DISORDERS OF LIPID METABOLISM ⚠️

Various pathological conditions arise from disruptions in normal lipid metabolism, leading to significant health consequences.

Disorder	Cause	Consequences	Management
Hypercholesterolemia	Elevated blood cholesterol	Atherosclerosis, CVD risk	Statins, dietary changes
Atherosclerosis	Arterial plaque formation	Heart attack, stroke	Lifestyle modification, medications
Fatty Liver	Excess lipid accumulation	Liver dysfunction, cirrhosis	Weight loss, alcohol cessation

HYPERCHOLESTEROLEMIA: Condition characterized by elevated cholesterol levels in blood.

Types:

- **Primary:** Genetic disorders (familial hypercholesterolemia)
- **Secondary:** Diet, lifestyle, or other medical conditions

Risk Factors:

- High saturated fat intake
- Sedentary lifestyle
- Genetic predisposition

- Age and gender factors
- Certain medications

ATHEROSCLEROSIS: Progressive disease involving the buildup of plaques in arterial walls.

Pathogenesis:

- Endothelial dysfunction and injury
- Infiltration of lipoproteins into arterial wall
- Oxidation of LDL cholesterol
- Inflammatory response and foam cell formation
- Plaque formation and potential rupture

FATTY LIVER: Accumulation of excess fat in hepatocytes, leading to liver dysfunction.

Types:

- **Alcoholic Fatty Liver Disease (AFLD)**
- **Non-Alcoholic Fatty Liver Disease (NAFLD)**

Progression:

- Simple steatosis → steatohepatitis → fibrosis → cirrhosis

OBESITY: Excessive accumulation of body fat, often associated with metabolic dysfunction.

Metabolic Consequences:

- Insulin resistance
- Dyslipidemia
- Increased cardiovascular risk
- Fatty liver disease
- Metabolic syndrome

AMINO ACID METABOLISM 🔬

Amino acid metabolism encompasses the complex biochemical processes involved in the synthesis, degradation, and interconversion of amino acids, which are the building blocks of proteins and precursors of many important biomolecules.

GENERAL REACTIONS OF AMINO ACID METABOLISM ⚗️

TRANSAMINATION 🔄

Transamination is the transfer of an amino group from an amino acid to a keto acid, catalyzed by aminotransferases (transaminases).

Mechanism:

- Requires pyridoxal phosphate (PLP) as cofactor
- Forms Schiff base intermediate with amino acid substrate
- Transfers amino group to α-ketoglutarate or other keto acids
- Produces corresponding keto acid and new amino acid

Important Transaminases:

- **ALT (Alanine Aminotransferase):** Alanine + α-ketoglutarate ⇌ Pyruvate + Glutamate
- **AST (Aspartate Aminotransferase):** Aspartate + α-ketoglutarate ⇌ Oxaloacetate + Glutamate

Clinical Significance:

- Elevated ALT and AST levels indicate liver damage
- Used as diagnostic markers for hepatic disorders
- Essential for amino acid interconversion and nitrogen metabolism

DEAMINATION

Deamination involves the removal of amino groups from amino acids, producing ammonia and corresponding keto acids.

Types of Deamination:

Oxidative Deamination:

- Primarily occurs with glutamate
- Catalyzed by glutamate dehydrogenase
- Produces α-ketoglutarate, NH_3, and NADH
- Reversible reaction that can also fix ammonia

Non-oxidative Deamination:

- Occurs with specific amino acids
- Produces ammonia and unsaturated compounds
- Examples include deamination of serine and threonine

DECARBOXYLATION — CO_2

Decarboxylation is the removal of carboxyl groups from amino acids, producing corresponding amines and CO2.

Process:

- Catalyzed by amino acid decarboxylases
- Requires pyridoxal phosphate (PLP) as cofactor
- Produces biologically active amines
- Important for neurotransmitter synthesis

Examples:

- Histidine → Histamine (involved in allergic reactions)
- DOPA → Dopamine (neurotransmitter precursor)
- 5-Hydroxytryptophan → Serotonin (neurotransmitter)
- Glutamate → GABA (inhibitory neurotransmitter)

UREA CYCLE AND ITS DISORDERS 🔄

The urea cycle is the primary mechanism for disposing of toxic ammonia generated from amino acid catabolism by converting it to less toxic urea.

LOCATION:

- Partially in mitochondria (first two steps)
- Partially in cytoplasm (remaining steps)
- Primarily occurs in liver hepatocytes

UREA CYCLE STEPS:

Step	Enzyme	Location	Products
1	Carbamoyl phosphate synthetase I	Mitochondria	Carbamoyl phosphate
2	Ornithine transcarbamylase	Mitochondria	Citrulline
3	Argininosuccinate synthetase	Cytoplasm	Argininosuccinate
4	Argininosuccinate lyase	Cytoplasm	Arginine + Fumarate
5	Arginase	Cytoplasm	Urea + Ornithine

DETAILED MECHANISM:

Step 1: Carbamoyl Phosphate Formation

- $NH_3 + CO_2 + 2ATP \rightarrow$ Carbamoyl phosphate + $2ADP + Pi$
- Rate-limiting step
- Activated by N-acetylglutamate

Step 2: Citrulline Formation

- Carbamoyl phosphate + Ornithine → Citrulline + Pi
- Occurs in mitochondrial matrix
- Citrulline is transported to cytoplasm

Step 3: Argininosuccinate Formation

- Citrulline + Aspartate + ATP → Argininosuccinate + AMP + PPi
- Aspartate provides the second nitrogen for urea

Step 4: Arginine Formation

- Argininosuccinate → Arginine + Fumarate
- Fumarate can be converted to oxaloacetate and then aspartate

Step 5: Urea Formation

- Arginine → Urea + Ornithine
- Ornithine returns to mitochondria to restart the cycle

ENERGY REQUIREMENTS:

- 4 ATP molecules consumed per urea molecule produced
- 2 ATP in step 1, 1 ATP in step 3 (converted to AMP), 1 ATP for AMP conversion

DISORDERS OF UREA CYCLE:

Types and Characteristics:

- **Carbamoyl Phosphate Synthetase I Deficiency:** Severe hyperammonemia, early onset
- **Ornithine Transcarbamylase Deficiency:** X-linked, most common urea cycle disorder
- **Argininosuccinate Synthetase Deficiency:** Citrullinemia, elevated citrulline levels
- **Argininosuccinate Lyase Deficiency:** Argininosuccinic aciduria
- **Arginase Deficiency:** Hyperargininemia, developmental delays

Clinical Manifestations:

- Hyperammonemia (elevated blood ammonia)

- Neurological symptoms (lethargy, seizures, coma)
- Developmental delays
- Intellectual disability
- Potential death in severe cases

Management:

- Protein restriction
- Ammonia-lowering medications
- Arginine supplementation (in some deficiencies)
- Liver transplantation in severe cases

CATABOLISM OF PHENYLALANINE AND TYROSINE

The catabolism of phenylalanine and tyrosine involves a series of enzymatic reactions that can be disrupted, leading to various metabolic disorders.

NORMAL CATABOLIC PATHWAY:

Phenylalanine → Tyrosine:

- Catalyzed by phenylalanine hydroxylase
- Requires tetrahydrobiopterin (BH4) as cofactor
- Rate-limiting step in phenylalanine catabolism

Tyrosine Catabolism:

- Tyrosine → DOPA → Dopamine → Norepinephrine → Epinephrine

- Alternative pathway: Tyrosine → p-Hydroxyphenylpyruvate → Homogentisate → Fumarate + Acetoacetate

METABOLIC DISORDERS:

PHENYLKETONURIA (PKU) 🚫

Cause:

- Deficiency of phenylalanine hydroxylase enzyme
- Inability to convert phenylalanine to tyrosine
- Autosomal recessive inheritance

Biochemical Changes:

- Elevated phenylalanine levels in blood and urine
- Increased production of phenylpyruvate, phenyllactate, and phenylacetate
- Tyrosine becomes essential amino acid

Clinical Manifestations:

- Intellectual disability (if untreated)
- Seizures and behavioral problems
- Light skin pigmentation and blue eyes
- Musty odor due to phenylacetic acid excretion

Management:

- Phenylalanine-restricted diet from birth
- Regular monitoring of blood phenylalanine levels

- Tyrosine supplementation
- Early detection through newborn screening

ALBINISM

Cause:

- Deficiency of tyrosinase enzyme
- Inability to produce melanin from tyrosine
- Various genetic subtypes exist

Types:

- **Oculocutaneous Albinism:** Affects skin, hair, and eyes
- **Ocular Albinism:** Primarily affects eyes
- **Partial Albinism:** Localized pigmentation defects

Clinical Features:

- Lack of pigmentation in skin, hair, and eyes
- Increased sensitivity to sunlight
- Vision problems (nystagmus, photophobia)
- Increased risk of skin cancer

ALKAPTONURIA

Cause:

- Deficiency of homogentisate oxidase
- Accumulation of homogentisic acid

- Autosomal recessive inheritance

Clinical Features:

- Dark urine upon standing (homogentisic acid oxidation)
- Ochronosis (blue-black pigmentation of connective tissues)
- Arthritis in later life
- Cardiac valve involvement

TYROSINEMIA 🔴

Types:

- **Type I:** Fumarylacetoacetase deficiency (most severe)
- **Type II:** Tyrosine aminotransferase deficiency
- **Type III:** 4-Hydroxyphenylpyruvate dioxygenase deficiency

Clinical Features:

- Hepatic dysfunction
- Renal tubular defects
- Neurological crises (Type I)
- Skin and eye lesions (Type II)

SYNTHESIS AND SIGNIFICANCE OF BIOLOGICAL SUBSTANCES 🧪

Various amino acids serve as precursors for important biological molecules that have significant physiological functions.

5-HYDROXYTRYPTAMINE (SEROTONIN) 😊

Synthesis:

- Tryptophan → 5-Hydroxytryptophan (via tryptophan hydroxylase)
- 5-Hydroxytryptophan → Serotonin (via aromatic L-amino acid decarboxylase)

Functions:

- Neurotransmitter in central nervous system
- Regulates mood, sleep, and appetite
- Involved in gastrointestinal motility
- Platelet aggregation and vasoconstriction

Clinical Significance:

- Deficiency associated with depression
- Target for antidepressant medications (SSRIs)
- Elevated levels in carcinoid syndrome

MELATONIN 🌙

Synthesis:

- Serotonin → N-Acetylserotonin (via serotonin N-acetyltransferase)
- N-Acetylserotonin → Melatonin (via hydroxyindole-O-methyltransferase)

Functions:

- Regulates circadian rhythms

- Sleep-wake cycle control
- Antioxidant properties
- Immune system modulation

Clinical Applications:

- Treatment of sleep disorders
- Jet lag management
- Seasonal affective disorder

DOPAMINE 🧠

Synthesis:

- Tyrosine → DOPA (via tyrosine hydroxylase)
- DOPA → Dopamine (via aromatic L-amino acid decarboxylase)

Functions:

- Neurotransmitter in brain reward pathways
- Motor control and coordination
- Prolactin regulation
- Cognitive functions

Clinical Significance:

- Deficiency in Parkinson's disease
- Involved in addiction and reward mechanisms
- Target for antipsychotic medications

NORADRENALINE (NOREPINEPHRINE) ⚡

Synthesis:

- Dopamine → Noradrenaline (via dopamine β-hydroxylase)

Functions:

- Sympathetic nervous system neurotransmitter
- Fight-or-flight response
- Blood pressure and heart rate regulation
- Attention and arousal

ADRENALINE (EPINEPHRINE) 🔥

Synthesis:

- Noradrenaline → Adrenaline (via phenylethanolamine N-methyltransferase)

Functions:

- Hormone of adrenal medulla
- Emergency response ("fight or flight")
- Metabolic effects (glycogenolysis, lipolysis)
- Cardiovascular stimulation

CATABOLISM OF HEME 🔴

Heme catabolism is the process by which heme groups from hemoglobin and other heme proteins are broken down and disposed of.

HEME BREAKDOWN PATHWAY:

Step 1: Heme Oxygenase

- Heme → Biliverdin + CO + Fe^{2+}
- Rate-limiting enzyme
- Requires NADPH and O2
- Produces carbon monoxide as byproduct

Step 2: Biliverdin Reductase

- Biliverdin → Bilirubin (unconjugated)
- Requires NADPH
- Bilirubin is lipophilic and potentially toxic

Step 3: Conjugation

- Bilirubin + Glucuronic acid → Bilirubin glucuronide (conjugated)
- Catalyzed by UDP-glucuronyl transferase
- Occurs in hepatocytes
- Makes bilirubin water-soluble

Step 4: Excretion

- Conjugated bilirubin secreted into bile
- Transported to intestine
- Bacterial conversion to urobilinogen and stercobilinogen
- Partial reabsorption and renal excretion

HYPERBILIRUBINEMIA AND JAUNDICE

Types of Hyperbilirubinemia:

Type	Cause	Bilirubin Type	Examples
Pre-hepatic	Excessive hemolysis	Unconjugated	Hemolytic anemia
Hepatic	Liver dysfunction	Both types	Hepatitis, cirrhosis
Post-hepatic	Bile duct obstruction	Conjugated	Gallstones, tumors

UNCONJUGATED HYPERBILIRUBINEMIA:

- **Causes:** Hemolytic disorders, Gilbert's syndrome, Crigler-Najjar syndrome
- **Characteristics:** Elevated unconjugated bilirubin, normal liver enzymes
- **Complications:** Kernicterus in neonates (bilirubin encephalopathy)

CONJUGATED HYPERBILIRUBINEMIA:

- **Causes:** Biliary obstruction, Dubin-Johnson syndrome, Rotor syndrome
- **Characteristics:** Elevated conjugated bilirubin, dark urine
- **Associated:** Pale stools, pruritus

JAUNDICE (ICTERUS): Clinical manifestation of hyperbilirubinemia characterized by yellowish discoloration of skin, sclera, and mucous membranes.

Clinical Classification:

- **Physiological Jaundice:** Normal in neonates due to immature liver
- **Pathological Jaundice:** Requires investigation and treatment

- **Kernicterus:** Bilirubin deposition in brain nuclei (neurological damage)

Diagnostic Approach:

- Total and direct bilirubin levels
- Liver function tests
- Complete blood count
- Imaging studies (ultrasound, CT, MRCP)
- Specific tests based on suspected etiology