## Syllabus

## For

# $P.G.\ C.E.T.\ (Bio-Chemistry-2018)$

Title

I Biomolecules
II Enzymes
III Biochemical techniques
IV Metabolism
V Molecular Biology
VI Nutritional Biochemistry
VII Clinical Biochemistry

Unit

Suggested readings

#### **BIOMOLECULES**

- I. History and Development of Biochemistry.
- II. Composition of living matter, importance of carbon, origin of life.
- III. Prokaryotic and Eukaryotic cells, Structure of Orangelles, Structureand Functions.
- IV. Carbohydrates: Definition, biological role and Classification of carbohydrates, Structure of monosaccharides, Stereoisomerism and anomerism of sugars, ring structures and anomeric forms, mutarotation, Reactions of aldehyde and Ketone groups reactions due to hydroxyl groups. Structure, occurrence and biologicalimportance of monosaccharides, (glucose, fructose, galactose) disaccharides (Sucrose, maltose, lactose) and polysaccharides (Cellulose, glycogen, starch) and chitin, agar, pectin, proteoglycans, sialic acids, blood group polysaccharides.
- V. Lipids :Definition, fatty acids nomenclature, structure.Classification of lipids; simple, complex & derived lipids. Physicalproperties and Chemical Properties hydrolysis, saponification, rancidity of fats, reaction of glycerol. Biological significance of fats. Properties and functions of sterols.
- VI. Amino Acids:- Common Structural features, Sterio-Isomerism, Classification and Structures of standard amino acids, amino acidsas zwitterions, physical and chemical properties, titration of aminoacids.
- VII. Peptides; Structure of peptide bond, Structure & functions of Oxytocin & Vasopressin.
- VIII. Proteins: Introduction, Composition. Protein structure, Levels of Structure; Primary, Secondary-helix and pleated sheets, Tertiary forcesstabilizing tertiary structure and quaternary structure of protein. Classification: Simple, conjugated & derived proteins, Physicochemical properties-IpH, denaturation, salting out & salting in, hydrolysis & color reactions of proteins due to side-chain. Biological functions of proteins.
  - IX. Nucleic Acids: Composition of DNA and RNA, occurrence & types of nucleic acids, generalized structural plan of nucleic acids, Chemistry of nucleotides features of DNA double helix. Denaturation of DNA, structure and roles of different types of RNA(m-RNA, t-RNA & r-RNA) DNA & RNA Viruses.

#### **ENZYMES**

- I. Introduction: History general characteristics, nomenclature, IUBenzyme classification with examples. Minimum 2 examples
- II. Holoenzyme, apo-enzymes, coenzymes, cofactor, activators, inhibitors, active site (identification of groups excluded)metallo-enzymes, units of enzyme activity, specific enzymes.
- III. Isoenzymes, monomeric enzymes, oligomeric enzymes and multi-enzymecomplexes, Enzyme specificity.
- IV. Measurement and expression of enzyme activity-enzyme assays.Definitionof IU, enzyme turn over number and specific activity.Coenzyme, prosthetic groups.
- V. Enzyme catalysis &Coenzymes: Role of cofactors in enzymecatalysis. NAD/NADP FMN/FAD, coenzyme A, biocytin, cobamide,lipoamide, TPP, pyridoxal phosphate, tetrahydrofolate and metalions with special emphasis on coenzyme functions.
- VI. Enzyme Kinetics: Factors affecting enzyme activity: enzymeconcentration, substrate concentration, pH and temperature.
- VII. Derivation of Michaelis-Menten equation for uni-substratereactions, Km and its significance, Line weaver-Burk plot and itslimitations. Importance of Kcat/Km. Bi-substrate reactions-briefintroduction to sequential and ping-pong mechanisms withexamples. Kinetics of Zero and first order reactions.
- VIII. Significance and evaluation of energy of activation and free energy.
  - IX. Reversible and irreversible inhibition, competitive, non-competitive and uncompetitive ihibilitions, determination of Km and Vmax inpresence and absence of inhibitor.
  - X. Allosteric enzymes.
    - 1. Isolations & purification of enzymes-selection of sources, isolation, stabilization, methods of solubilization, extraction, precipitation, filtration, centrifugation and purification.
    - 2. Isolation of amylase, papain, trypsm, chymotrypsin.
    - 3. Mechanism of action of chymotrypsin, carboxypeptidase, ribonuclease and Lysozyme.

- Immobilization-Advantages & disadvantages, Immobilization, Techniques, Enzyme Stabilization, Bioreactors, Properties of immobilized enzymes, experimental procedures. Enzymes in industry.
- 5. Enzymes in industry.
- 6. Stabilization of enzymes by immobilization

## **BIOCHEMICAL TECHNIQUES**

IChromatography: General Principles working and applications of

- a) Adsorption chromatography
- b) Ion exchange chromatography
- c) Thin layer chromatography
- d) Molecular sieve chromatography
- e) Gas liquid chromatography
- f) Affinity chromatography
- g) Paper chromatography

## II) Electrophoresis:

Principle, factors, Tiselius apparatus, Continuous and discontinuous, paper, gel, immunolelectrophoresis, PAGE and SDS-PAGE, Two dimensionalelectrophoresis, its importance, isoelectrfocussing.

#### III) Spectroscopic Techniques

Beer-Lambert law, light absorption and its transmittance, determination and application of extinction coefficient, application of visible and UV spectroscopic techniques, Fluorescent and emissionspectroscopy techniques, Fluorescent and emission spectroscopy

## I) Centrifugation technique:

Principles of sedimentation, cell fractionation, preparativecentrifugation, analytical ultacentrifugation, determination of molecular weight,

## II) Radio isotopic Technique:

Types of radioisotopes used in Biochemistry, Units of radioactivity measurements, techniques used to measureradioactivity (gas ionization and liquid scintillation counting), isotopes

commonly used in biochemical studies-(P32, S35, C14 and H3), Autoradiography, Biological hazardsof radiation and safety measures in hadling radioisotopes. Biological applications.

III) Immunological Techniques:

Antigen, Antibody, Haptan, Antigen-Antibody reactions,

Immunodiffusion, Immunoelectrophoresis, Radioimmunoassay. ELISA, Immunofluorescence.

#### **METABOLISIM**

- 1. Concept of Bioenergetic:Concept of free energy, determination of G, Energy richcompounds, coupling of reactions, G and oxidationreduction.
- 2. Carbohydrate Metabolism:Reactions and energetics of glycolysis Alcoholicfermentations. Entry of fructose, galactose, mannose, etc.Reactions and energetic of TCA cycle.Gluconeogenesis,glycogenesis and glycogenosis.Reactions and physiologicalsignificance of pentose phosphate pathway, Regulation ofglycolysis and TCA cycle.
- 3. Photosynthesis:

Light & dark reactions, Chloroplast, Photosystems, Cyclic &non cyclic photo phosphorylation calvincyle.

4. Electron Transfer chain and Oxidative Phosphorylation:

Structure of mitochondria, sequence of electron carriers, sites of ATP production, inhibitors of electron transportchain. Hypothesis of mitochondrial oxidative phosphorylation (basis concepts) Inhibitors and uncouplersof oxidative phosphorylation. Transport of reducing potentials into mitochondria.

#### Lipid Metabolism:

Introduction, hydrolysis of triacyiglycerols, transport of fattyacids into mitochondria, oxidation of saturated fatty acids,ATP yield from fatty acid oxidation, Biosynthesis of saturated and unsaturated fatty acids. Metabolism of Ketone bodies, oxidation of unsaturated and odd chain fattyacids, Biosynthesis of triglycerides and important phospholipids, glycolipids, sphingolipids, and cholesterol. Regulation of cholesterol metabolism.

#### Amino acid Metabolism:

Transamination, decarboxylation, urea cycle,r-gamma glutamyl cycle. Degradation og glycine. Threonine, arginine, cystine. Biosynthesis of serence, Isoleucine, glycine, arginine. Cysteine, histidine.

Nucleotide Metabolism:

Sources of purine & pyrimidine, Salvage pathway of purine synthesis Synthesis of pyrimidine, Degradation of purines & pyrimidences.

#### MOLECULAR BIOLOGY

## 1) Basic Concepts of Genetic information

Nucleic acids as genetic information carries, experimental evidence eg. Bacterial genetic transformation, TMV reconstitution experiment. Central dogma in molecular biology reverse transcription and retroviruses, salient features in eukaryotic, prokaryotic and viral genomes, highly repetitive, moderately repetitive and unique DNA sequence.

#### 2) Structural levels of Nucleic acids:

Primary Structure of nucleic acids, Basic concepts about secondary structures of nucleic acid, 5'—3' direction secondary and tertiary structure of DNA: Watson and Crick model; anti-parallel strands, base composition, base equivalence base-paring and base-stacking in DNA molecule, Tm and its relationship G-C content in DNA, A, B and Z types of DNA major and minor grooves, Structures and functions of RNAs, t-RNA, r-RNA, M-RNA,

## 3) DNA replication:

DNA replication in prokaryotes: conservative, Semi conservative and dispersive types, Experimental evidences for semi conservative replication, DNA polymerases, other enzymes and protein factors involved in replication. Mechanism of replication.

## 4) Transcription:

Transcription in prokaryotes, DNA polymerase, promoters, initiation, elongation and termination of RNA synthesis, post-transcriptional processing of RNA in eukaryotes.

## 5) Translation:

Genetic Code: Basic features, biological significance of degeneracy, Wobble hypothesis, Mechanism of translation: Ribosome structure: A and F sites, charged, t-RNA, f-met-tRNA, initiator codon, formation of 70-S initiator complex, role of EF-tu, EF-Ts, EF,-G and GTF, Non-sense codons and release factors RF I & RF II concept of

## 6) Regulation:

Homoeostatis, Regulation by covalent modification, fed back inhibition: operon model (Lac-His-operon, Tryp-operon)

## 7) Mutation:

Molecular basis of mutation, types of mutation-transition transversion, frame shift, insertion, deletion, DNA repair: UV repair systems in E-coli.

8) Recombinant DNA Technology:

Restriction endonucleases, brief discussion of steps in DNA cloning, Application of recombinant DNA technology, human genome project.

#### **NUTRITIONAL BIOCHEMISTRY**

- 1) Introduction and definition of food and nutrition. The diet and its components, carbohydrates, fats, proteins, vitamins, minerals and water.
- 2) Composition of balanced diet, the nutritional value of carbohydrates, proteinsprotein requirement nitrogen balance, essential and non-essential amino acids Biologist value of proteins, supplementary value of proteins, supplementary value of proteins, nutritional value of lipids-essential fatty acids,
- 3) Vitamins A,D,E,K, Vit. B-complex and minerals (Ca, Fe Cu, P, CI, K, Mg, Zn, Se, and lodine) and their biological functions.
- 4) Basic concepts of energy expenditure, units of energy, Calorimetry, Measurement of energy value of foodstuffs by calorimetry, RQ of food, determination of heat production of the diet.

- 5) The basal metabolism, measurement of basal metabolic rate (BMR) factors influencing BMR, energy requirements and recommended dietary allowances (RDA) for an average Indian, SDA of foods.
- 6) Malnutrition-Kwashiorkor and Marasmus.
- 7) Food adulteration, common adulterants and their ill effects, simple physical tests for detection of food adulterants, simple chemical tests.

#### **CLINICAL BIOCHEMISTRY**

- 1) Definition and scope of clinical biochemistry in diagnosis, a brief review of units and collection and preservation of biological fluids (blood, serum, plasma, urine and CSF Normal values of important constituents (in SI units) in CSF and urine.
- 2) Blood-general composition-palsma and erythorcyes, leukocytes and platelets composition of plasma-plasma proteins, normal value of important constituents (in SI units) in Blood (Plasma / serum) Structure and functions of hemoglobin. Abnormal hemoglobins sickle cell anemia, thalessemia.
- 3) Definition functional and non-functional plasma enzymes, enzyme pattern in health and diseases with special mention of plasma lipase, amylase, cholinesterase, alkaline and acid phosphatasc, SGOT, SGPT, LDH and CPK.
- 4) Fucntional tests of kidney, liver and gastric fluids.
- 5) Genetic diseases (Inborn errors) of amino acid metabolism (alkaptonuria, phenylketonuria, Tyrosinemia, Cystinosis albinism)-Gout and hyperuricemia, crotic aciduria.
  - Carbohydrate metabolism (Diabetes, pentosuria, glycogen storage disease, glycosuria) Lipid metabolism (Libpoproteinaemia, Fatty liver, gangliosdosis, Atherosterosis, streatorrhoeas)