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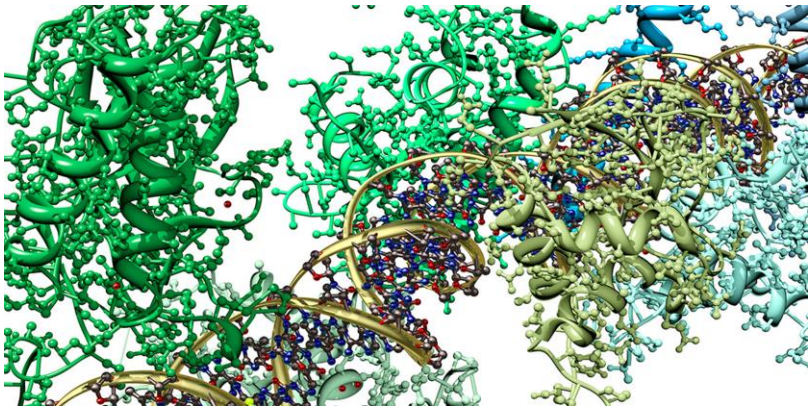

Department of Biochemistry

Syllabus for

Ph.D. Entrance Test

in Biochemistry

Dr. Babasaheb Ambedkar Marathwada University, Aurangabad

Entrance Test Syllabus for Ph.D. in Biochemistry
Syllabus for PET in Biochemistry

Paper II

Unit I: ***Research Methodology in Biochemistry***

Separation & Purification Techniques:

Chromatography: Paper, TLC, Adsorption, partition, ion-exchange, reverse phase, gel filtration, affinity, gas chromatography, HPLC (High Pressure Liquid Chromatography).

Electrophoresis: Moving boundary and zonal electrophoresis, paper and gel electrophoresis, isoelectric focusing.

Characterization and purification of proteins
Methods of protein characterization, methods of protein purification.
Centrifugation, Dialysis, Lyophilization, Ultrafilteration, Chromatography, Electrophoresis,

Techniques for manipulation of nucleic acids:
Isolation and purification of RNA , DNA (genomic and plasmid) and proteins, different separation methods. Analysis of RNA, DNA and proteins by one and two dimensional gel electrophoresis, Isoelectric focusing gels.

Isolation of nucleic acids, radioactive labeling of nucleic acids, restriction endonucleases, plasmids, purification of complementary DNA strands, hybridization by blotting, determining base sequence of DNA, preparation of DNA complementary to RNA.

Analytical Techniques

Biophysical Method: Molecular analysis using UV/visible, fluorescence, circular dichroism, NMR and ESR IR spectroscopy Molecular structure determination using X-ray diffraction and NMR, Molecular analysis using light scattering, different types of mass spectrometry and surface plasma resonance methods.

Statisitcal Methods: Measures of central tendency and dispersal; probability distributions (Binomial, Poisson and normal); Sampling distribution; Difference between parametric and non-parametric statistics; Confidence Interval; Errors; Levels of significance; Regression and Correlation; t-test; Analysis of variance; X_2 test;; Basic introduction to Muetrovariate statistics, etc.

Radiolabeling techniques: Detection and measurement of different types of radioisotopes normally used in biology, incorporation of radioisotopes in biological tissues and cells, molecular imaging of radioactive material, safety guidelines.

Microscopic techniques: Visualization of cells and subcellular components by light microscopy, resolving powers of different microscopes, microscopy of living cells, scanning and transmission electron microscopes, fluorescence microscopy, freeze-fracture techniques, specific staining of organelles or marker enzymes.

Molecular tools in phylogeny: classification and identification of species; Protein and nucleotide sequence analysis; origin of new genes and proteins; Gene duplication and divergence.

Unit II ***Biomolecules and Molecular Flux***

Carbohydrates and their derivatives: Monosaccharides and related compounds, glycosidic bond, disaccharides, polysaccharides, heteropolysaccharides.
Lipids: Fatty acids, Phospholipids, Cholesterol and related steroids, other biological lipids. Glycerolipids, sphingolipids, the eicosanoids.
Amino acids, peptides and polypeptides: Amino acids, peptides and polypeptides, determination of amino acid composition of proteins, determination of amino acid sequence of proteins, chemical synthesis of peptides and polypeptides.
3-D structure of proteins: Information for folding, forces that determine folding, hierarchy of structural organization, functional diversification of proteins.
Nucleotides and nucleic acids: Structural properties of DNA, chemical synthesis of DNA, conformational behavior of RNA, nucleoproteins.

Enzymes: Classification of enzymes. Review of unisubstrate enzyme kinetics and factors affecting the rates of enzyme catalyzed reactions. Michaelis-Menten equation, pH functions and their significance. Kinetics of multisubstrate reactions. Use of initial velocity, inhibition and exchange studies to differentiate between multisubstrate reaction mechanisms. Ligand binding, including measurement, analysis of binding isotherms. Cooperativity phenomenon. Hill and Scatchard Plots. Allosteric enzymes, Sigmoidal kinetics and their physiological significance. Symmetric and sequential modes for action of allosteric enzymes and their significance.
Structure and function of coenzymes: Thiamine pyrophosphate, Pyridoxal phosphate, Nicotinamide, flavins, phosphopentetheine, alfa-lipoic acid, biotin, folate, vitamin B12, Iron containing coenzymes, coenzymes in methanogenesis.
Mechanisms of enzyme catalysis: Enzyme catalyst and other chemical catalyst, unique features of enzyme catalysts, trypsin family of enzymes, chymotrypsin catalytic mechanism, carboxipeptidase A, pancreatic RNAase A, lysozyme, lactate dehydrogenase.

Molecules in flux

Carbohydrates: Glycolysis, citric acid cycle its function in energy generation and biosynthesis of energy rich bonds, pentose phosphate pathway and its regulation. Alternate pathways of carbohydrate metabolism. Gluconeogensis, interconversions of sugars. Biosynthesis of glycogen, starch and oligosaccharides. Regulation of blood glucose homeostasis. Hormonal regulation of carbohydrate metabolism.

Lipids: Fatty acid biosynthesis: Acetyl CoA carboxylase, Fatty acid synthase, desaturase and elongase. Fatty acid oxidation: (x, 0, w oxidation and lipoxidation. Lipid Biosynthesis: Ketone bodies: Formation and utilisation. Metabolism of Circulating lipids: chylomicrons, LDL, HDL and VLDL. Free fatty acids.

Amino Acids: Biosynthesis and degradation of amino acids and their regulation. Specific aspects of amino acid metabolism. Urea cycle and its regulation, In-born errors of amino acid metabolism.

Nucleic Acids: Biosynthesis or purines and pyrimidines, Degradation if purines and pyrimidines Regulation of purine and pyrimidine biosynthesis, Structure and regulation of ribonucleotide reductase. Biosynthesis of ribonucleotides, deoxyribonucleotides and polynucleotides Inhibitors of nucleic acid biosynthesis

Generation of Energy Currency: The mitochondrial respiratory chain, proton gradient, cytochromes and their characterization. The Q cycle and the stoichiometry of proton extrusion and uptake; P/0 and H/P ratios. Reversed electron transfer, respiratory controls and oxidative phosphorylation, uncouplers and inhibitors of energy transfer. Fractionation and reconstitution of respiratory chain complexes. ATP - synthetase complex. Microsomal electron transport, partial reduction of oxygen, superoxides.

Unit III *Cellular Basis of Life* and Cell-Cell Interactions in Immunity

Structural organization:
Unicellular, colonial and multicellular forms. Structural Organisation of prokaryotic and eukaryotic cells. Plant and animal cells: variation in structure and function.

The cytoskeleton - microtubules and microfilaments. Types of tissues, epithelium - types, epithelial apices - glycocalyx, microvilli. The basement membrane - structural features and characteristics. The extracellular matrix-collagen, elastin, fibrillin, fibronectin, laminin and proteoglycans.

Membrane structure and function
(Structure of model membrane, lipid bilayer and membrane protein diffusion, osmosis, ion channels, active transport, membrane pumps, mechanism of sorting and regulation of intracellular transport, electrical properties of membranes).

Structural organization and function of intracellular organelles (Cell wall, nucleus, mitochondria, Golgi bodies, lysosomes, endoplasmic reticulum, peroxisomes, plastids, vacuoles, chloroplast, structure & function of cytoskeleton and its role in motility).

Cell division and cell cycle (Mitosis and meiosis, their regulation, steps in cell cycle, regulation and control of cell cycle, cell cycle checkpoints, cyclins and cyclin-dependent kinases). Programmed cell death, aging and senescence

Cellular communication general principles of cell communication, cell adhesion and roles of different adhesion molecules, gap junctions, extracellular matrix, integrins, neurotransmission and its regulation.

Prokaryotic and eukaryotic living forms: bacteria, fungi, protozoa, Common parasites and pathogens of humans, domestic animals and crops.

Microbial Physiology (Growth yield and characteristics, strategies of cell division, stress response)

Single cell interactions for Immunity White blood cells, neutrophils, monocytes, tissue macrophages, lymphocytes: T and B cells. Structure of lymph node,

Phagocytosis by neutrophils and monocytes, intracellular events in phagocytosis leading to reactive oxygen species production, microbial killing post phagocytosis, NADPH-dehydrogenase complex, granuloma formation, NADPH-dehydrogenase complex deficiency, PAMP and Toll-like receptors

Humoral & Cellular Immunity Immunogen and antigen, B cell stimulation and antibody production, Immunoglobulin classes, T cell receptor (TCR), major histocompatibility complex and antigen presentation, T cell stimulation and cytokine production, T and B cell interaction, production of immunological synapse, molecules involved, role of cell adhesion molecules. Maturation of humoral and cellular immune responses, Clonal cells and role of clonal populations in immune responses. Generation of antibody diversity, hypersensitivity, autoimmunity.

Unit IV ***Principles of Central Dogma, Functionalities & Organ Systems***

Mendelian principles: Dominance, segregation, independent assortment.
Concept of gene : Allele, multiple alleles, pseudoallele, complementation tests
Gene mapping methods: mapping with molecular markers, mapping by using somatic cell hybrids, development of mapping population in plants.
Extra chromosomal inheritance: Inheritance of Mitochondrial and chloroplast genes, maternal inheritance. Plasmids, latent viral infections
Microbial genetics: transformation, conjugation, transduc-tion and sex-duction, mapping genes by interrupted mating, fine structure analysis of genes.
Human genetics: Pedigree analysis, karyotypes, genetic disorders.

SYSTEM PHYSIOLOGY - ANIMAL
Blood and circulation - Blood corpuscles, haemopoiesis and formed elements, plasma function, blood volume, blood volume regulation, blood groups, haemostasis.
Excretory system - Comparative physiology of excretion, kidney, urine formation, urine concentration, waste elimination, micturition, regulation of water balance, blood volume, blood pressure, electrolyte balance, acid-base balance.
Inborn errors of metabolism: carbohydrate, lipid, amino acid metabolism
Digestive system - Digestion, absorption, energy balance, BMR.
Endocrinology and reproduction - Endocrine glands, basic mechanism of hormone action, hormones and diseases; gametogenesis, ovulation, neuroendocrine regulation

SYSTEM PHYSIOLOGY - PLANT
Photosynthesis: Light harvesting complexes; mechanisms of electron transport; photoprote-ctive mechanisms; CO2 fixation-C3, C4 and CAM pathways.
Respiration and photorespiration: Citric acid cycle; plant mitochondrial electron transport and ATP synthesis; alternate oxidase; photorespiratory pathway.
Plant hormones: Biosynthesis, storage, breakdown and transport; physiological effects and mechanisms of action.
Sensory photobiology - Structure, function and mechanisms of action of phytochromes, cryptochromes and phototropins; stomatal movement; photoperiodism and biological clocks

Stem Cell Technology
Stem cell, definition, types of stem cells, scientific terms
Manipulations of stem cells, factors governing manipulations of stem cells
The future of stem cell technology using pluripotent stem cells
Culture of stem cells; Study of microenvironmental factors governing stem cell propogation,
Tissue engineering using stem cell technology
Reprogramming of genome function through epigenic inheritance
Ethical, social considerations of stem cell technology.

Functional Proteomics
What is proteome, Mass spectroscopy of various protein complexes, Organization of proteome in an organism and its systematic study, Protein chips

Microarray chips,
icroarray probes / chips, array fabrication, targets, assays, read out, image analysis, uses and examples.

SNPs
Identification and uses, DNA variations, SNP detection, data bases, study design, uses, genotyping.

Unit V ***Technical Advances and Derivatives***

Recombinant DNA Technology

Methods of creating recombinant DNA molecule, splicing, properties of restriction endonucleases and their mode of action, selection/screening, construction of DNA library, genomic *vs* cDNA library, chemical synthesis of gene, cloning vectors (X-phage, plasmid, M-13 phage, cosmid) shuttle vectors, yeast and viral vectors, expression vectors, uses of cloned gene, subcloning, sequencing by Sanger's method, proteins production in bacteria, site directed mutagenesis, RAPD, RFLP, PCR, qPCR, DNA finger printing, molecular diagnostics, antisense-RNA (micro RNA) technology, chromosomal walking.
In vitro mutagenesis and deletion techniques, gene knock out in bacterial and eukaryotic organisms. Methods for analysis of gene expression at RNA and protein level, large scale expression. Transgenic animals and plants, molecular approaches to diagnosis and strain identification. Genomics and its application to health and agriculture, including gene therapy.

Animal Cell and Tissue Culture
Culture techniques to study cell division - cell division by mitosis and meiosis. Cell cycle. Cell differentiation - organogenesis, morphological, functional and biochemical maturation of tissues. Biochemistry of cancer - carcinogenesis, characteristics of cancer cell, agents promoting carcinogenesis.

Hybridoma Technology

Monoclonal antibodies, mycelium cell fusion, selection of hybrids, hybridomas, protoplast fusion and HAT-medium, screening assays, purification and application of monoclonal antibodies.

Plant Tissue Culture

Micropropagation, somatic cell culture, somaclonal variations, somatic cell hybridization, protoplast isolation, protoplast fusion, protoplast culture, genetic transformation, various methods of gene transfer (all vector and vectorless methods), production of transgenic plants and animals, production of secondary metabolites, primary and transferred cell culture, differentiated cells in culture, applications.

Fermentation Technology

Primary and secondary metabolites in biotechnology, continuous and batch type culture techniques, principle types of fermentors, general design of fermentor, fermentation processes-brewing, manufacture of penicillin, production of single cell proteins, production strategies for other antibiotics and other organic compounds.

Applications of Immobilized Enzymes
Immobilized enzymes and their industrial applications. Effect of partition on kinetics and performance with particular emphasis on changes in pH and hydrophobicity. Biosensors, Bioremediation and phytoremediation

Histochemistry and Immunotechniques
Antibody generation, Detection of molecules using ELISA, RIA, western blot, immunoprecipitation, fluocytometry and immunofluorescence microscopy, detection of molecules in living cells, in situ localization by techniques such as FISH and GISH.

Reference Books and literature

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