ANDHRA PRADESH

RECRUITMENT OF ASSISTANT PROFESSORS IN THE UNIVERSITY
SYLLABUS FOR THE SCREENING TEST

BIOCHEMISTRY

SUBJECT CODE – 5

Unit-I:- Chemistry of Biomolecules

- Amino acids – classification, structure and physiochemical properties, chemical synthesis of peptides – solid phase peptide synthesis. Proteins – classification, purification, and criteria of homogeneneity. Structural organization, sequence determination and characterization of proteins. Conformation of proteins – Ramachandran plots. Denaturation of proteins.

- Classification, chemical properties of carbohydrates, Chemistry and biological roles of homo and heteropolysaccharides, peptidoglycan, glycosaminoglycans, glycoconjugates, glycoproteins, Structural elucidation of polysaccharides; Oligosaccharides – lectin interaction in biochemical processes.

- Classification of Lipids, Fatty acids and their physiochemical properties. Structure and properties of Prostaglandins. Fats and waces, physicochemical properties and characterization of fats and oil. Structure, properties and biological roles of phospholipids and Sphingolipids. Chemistry and properties of Sterols and Steroids. Salient features of bacterial and plant lipids.

- Nucleic acids – bases, nucleosides, nucleotides, physicochemical properties of nucleicacids, cleavage of nucleic acids by enzymatic methods, non – enzymatic transformation of nucleotides and nucleic acids, methylation, Sequencing, chemical synthesis of DNA. Three dimensional structure of DNA. Different forms of DNA – circular DNA and Supercoiling. Types of RNA. Strucure of t-RNA, mRNA and rRNA. Nucleotids as regulatory molecules, enzyme cofactors and mediatros of chemical energy in cells.

- Porphyrins – Structure and properties of porphyrins – heme , Chlorophyll and Cytochromes.

UNIT-II:- Bioanalytical Techniques

- Electrode and Indicator dye Techniques**:** Water and its properties. Handerson - Hesselbalch equation, standard hydrogen electrode, reference electrodes - calomel electrode and silver-silver chloride electrode. pH measurement using organic

indicators. PH meter - glass electrode, measurement of PH, selection of buffer, Ion specific electrode (oxygen electrode).

- Tissue homogenization. Disruption of tissues and cells, Centrifuges – Principle, applications and types. Differential and density gradient centrifugation. Preparative and analytical ultracentrifuge. Principles and applications of manometry and Principle and applications of microscopy, types of microscopes, phase contrast, fluorescent and electron microscopes.

- Separation Techniques: Principles, methods and applications of chromatography – Paper, thin layer, ion exchange, ge filtration and affinity chromatography, GLC, HPLC and chromatofocussing.

- Basic Principles of spectroscopy, basic laws of light absorption; instrumentation and applications of UV_visible, IR,ESR,NMR, aatomic absorption and Mass spectroscopy, flourimetry, flame photometry, bephelometry, ORD, CD, X-ray diffraction.

- Radioactivity: - detection and measurements of radioactivity, Radio isotopic techniques, incorporation of radioisotopes in biological tissues and cells. Biochemical uses of isotopes. Radiation hazards and methods of radioactive disposal.

- Principles, methods and applications of electrophoresis, moving boundary electrophoresis, zone electrophoresis, paper, starch, agarose, SDS-PAGE, High voltage and Capillary electrophoresis, Isoelectric focusing, two dimensional electrophoresis, Pulse field Gel Electrophoresis (PFGE).

- Blotting techniques: Introduction, significance, southern, northern and western blotting techniques (Principle, technique, methodology and applications).

- Biostatistics: Collection of data – Classification, Tabulation, Diagrammatic representation of data. Measures ofcentral tendency: Mean, median, mode, measures of dispersion - Range, interquartile Range. Average deviation, standard deviation, coefficient of variation. Correlation- Rank correlation, Regression lines, properties & examples.

UNIT-III Enzymology

- Historical aspects of enzymes. Remarkable properties of enzymes: catalytic power, specificity, regulation, transformation of different forms of energy. Holoenzyme, Apoenzyme, Coenzymes and cofactors. Nomenclature and classification of enzymes as per enzyme commission. Active site- Fisher and Koshland models. Formation of enzyme substrate complex – evidences.; Kinetic and chemical mechanisms of enzyme catalyzed reactions – Basic concepts of bioenergetics. The collision theory, activation energy and transition state theory. Basic concepts of kinetics of chemical reactions. Law of mass action and order of reactions. Initial velocity studies, progressive curves. Rapid reaction kinetics

(Continuous flow and stopped flow techniques).; Assay of enzymes: By kinetics, determination of catalytic activity, coupled kinetic assays, RIA of enzymes. Immunoradiometric assay. Units of enzyme activity IU, Katal, turnover. Investigation of sub cellular compartmentation of enzymes. Enzyme histochemistry, the use of centrifugation.

- Kinetics of single substrate enzyme catalyzed reaction, equilibrium, steady state assumption -Michaelis-Menten (Briggs- Haldane) equation. Transformation of Michaelis- Menten equation:- Lineweaver Burk, Eadie-Hofstee and Hanes plots. Determination of Vmax, Km,Kcat, Specificity constant (Kcat/Km) and their significance. Effect of pH temperature, enzyme and substrate concentrations on enzyme activity, Kinetics of enzyme reactions having two or more substrates. Single displacement and double displacement reactions. Enzyme inhibition: Reversible inhibition-Competitive, Noncompetitive (pure, mixed) inhibition, partial inhibition. Substrate inhibition, allosteric and irreversible inhibition. Feedback inhibition.
- Mapping of active site: conformation and nature of active site. Models identification of functional groups essential for catalysis. Trapping the enzyme substrate complex use of substrate analogs, Enzyme modification by chemical procedures affecting amino acid side chain, treatment with proteases, applications of site directed mutagenesis for mapping of active site. The effect of changing pH. Factors contributing to the catalytic efficiency-proximity and orientation, covalent catalysis, acid-base catalysis, metal ion catalysis, factor of strain in enzyme catalysis. Mechanism of enzyme action- Lysozyme, Carboxy peptidase, Chymotrypsin and Ribonuclease.

- Vitamin coenzymes, structure and functions. Enzyme regulation, allosteric enzymes, feed-back inhibition. Allosteric kinetics (ATcase), cooperativity, symmetry and sequential models. Hill equation-Plot. Covalent modification (Glycogen phosphorylase, Glutamine synthetase). Zymogen activation (Chymotrypsin). Isozymes (LDH). Multi-enzyme complexes (Pyruvate dehydrogenase complex). Multifunctional enzymes. Modern concepts of evolution of catalysis – catalytic RNA (Ribozymes), abzymes (catalytic antibodies), Immobilized enzymes.

UNIT-IV Cell Biology, Bioenergetic and Human nutrition

- Organization and Structure of Cells: Molecule - organelle - cell - organism; Oparin's chemical evolution. Cell structure and organization in bacteria, yeast, higher plant and animal cells: comparison of prokaryotic and eukaryotic cells. Organization, structure and functions of cell wall, plasma membrane, lysosomes, ribosomes, golgi complex, peroxisomes, glyoxysomes, mitochondria, plastids, endoplasmic

reticulum, vacuoles, centrioles, cytoskeleton, composition and structure and function of microtubules, microfilaments and intermediate filaments.

- Cell cycle – (i) Bacterial cell cycle (ii) Eukaryotic cell cycle-karyokinesis and cytokinesis – Growth coordination – partition and cytokinesis;, different phases, Check points, Molecular basis of cell cycle regulation. Cyclin dependent kinases, CDK-Cyclin diversity in yeast and animal cell cycles, regulation of CDK-Cyclin activity, progress through cell cycle – Apoptosis.

- Bioenergetics : Thermodynamic principles – Chemical equilibrium; free energy, enthalpy (H0 entropy(S). Free energy changes in biological transformations in living systems; High energy compounds. Energy change, oxidation-reduction reactions. Organization of electron carriers and enzymes in mitochondria. Classes of electron-transferring enzymes, inhibotrs of electon transport. Oxidative phosphorylation. Mitochondrial transport system. Microsomal electron transport; photorespiration, cyclic and non-cyclic reactions; photo-chemical events associated with pigment system – I and II. Utilization of oxygen by oxygenases, superoxide dismutase and catalase.
- Composition of food, criteria of energy value, measurement of energy value of food – direct and indirect methods. RQ of foods and its significance. Basal metabolism and measurement of BMR. Factors affecting BMR. Specific dynamic action of foods, sources and physiological functions of carbohydrates, proteins, fats. Dietary fiber and its role. Complete and incomplete proteins. Essential fatty acids. Nitrogen balance, methods employed to evaluate nutritive value of dietary proteins.

- Vitamins& Minerals - Fat and water soluble vitamins – structure, sources, daily allowances, physiological role, deficiency and toxicity symptoms, balanced diets and caloric requirements for people of various ages, sex and physiological state.
  Minerals and essential ultra trace elements – calcium, copper, iron, iodine, zinc, cobalt, selenium, fluorine, manganese and molybdenum

UNIT-V Intermediary Metabolism

- Approaches for studying intermediary metabolism. Glucose as fuel, glucose transporters, Glycolysis and its regulation. Substrate cycling, TCA cycle – function and regulation, Glyoxylate cycle, Gluconeogenesis and its regulation, HMP shunt and its significance, Uronic acid pathway, Glycogen metabolism and its regulation with special reference to phosphorylase and glycogen synthase, Metabolism of fructose, galactose and lactose, Biogenesis of amino sugars, peptidoglycans, glycosyl aminoglycans and glycoproteins. In born errors of carbohydrate metabolism.

- Proteins turn over – Role of ubiquitin. General metabolic reactions of amino acids. Metabolic breakdown of individual amino acids. Ketogenic and glycogenic amino acids. Formation of ammonia and urea. Regulation of urea cycle. Essential and

nonessential amino acids. Biosynthesis and regulation of branched chain amino acids, aromatic amino acids, histidine and methionine. Amino acids as biosynthetic precursors – formation of Creatine and creatinine, serotonin, melatonin, histamine, anserine, carnosine. Polyamines: GABA, melanin, catecholamine.

- Fats as energy stores, Oxidation of fatty acids, Formation and utilization of ketone bodies. Biosynthesis of fatty acids and regulation. Metabolism of arachidonic acid – formation of prostaglandins, thromboxanes, leucotrienes. Biosynthesis of triglycerides. Metabolism of phospholipids, sphingolipids. Biosynthesis and catabolism of cholesterol and its regulation, Formation of bile acids. Role of liver and adipose tissue in lipid metabolism. In born errors of lipid metabolism

- Biosynthesis, degradation and regulation of purine and pyrimidine nucleotides, chemical inhibition of biosynthesis of nucleotides. Biosynthesis of deoxyribonocleotides, regulation of ribonucleotide reductase. Salvage pathway. Disorders of purine and pyrimidines nucleotide metabolism.

- Biosynthesis and degradation of porphyrin (Heme). Porphyrias In born errors of amino acid metabolism.

Unit-VI Microbiology and Microbial Genetics

- Scope of microbiology, Spontaneous generation theory, germ theory of disease. Isolation of microorganisms- Direct and indirect, methods of maintenance of cultures.; General characteristics of bacteria, protozoa, fungi, algae, cyanobacteria, rickettsias. Spirochetes and archaebacteria.;Outline classification of bacteria as per Bergey's Manual of Systematic Bacteriology. Ecological grouping of bacteria.;Ultra structure of bacterium. An account of variant (Capsule, flagellum, pilus, endospore) and invariant (cell wall, cell Membrane, Mesosomes, Nucleoid, ribosomes) components of bacterial cell.; Nature and properties of Viruses.

- Viruses of Plants, vertebrates and microorganisms. Morphology, size, ultra structure and life cycles of some representative viruses - TMV, T4, Lambda and SV40, Prions, Viroids.

- Cell growth and kinetics of bacterial growth, Normal and biphasic growth curve, batch and continuous cultures, chemostats. Nutritional requirement of bacteria - phototroph, chemotrophs, organotrophs, methylotrophs, mixotrophs. Saprophytes, symbiotic and parasitic modes of nutrition. Fermentative and sulphur reducing bacteria.;Anaerobic and aerobic Growth. A brief note on animal, plant and human diseases caused by microorganisms. Introduction to Dairy microorganisms. Fundamentals of control- by physical agents, chemical agents, antibiotics and other chemotherapeutic agents. Assay of antimicrobial action

- Genome organization, Genome diversity, Viroids, Viruses, Genome of prokaryotes and Eukaryotes. Nucleic acids as genetic material - Transformation in Pneumococcus. The Hershey-Chase experiment - RNA as genetic material in small viruses. Classical and modern theories of gene - structures and function of plasmids. Episomes, transposons, retroposons, retroelement, Mitochondrial and chloroplast DNA. Colinearity of gene protein. Organization of Eukaryotic chromosomes.Nucleosomes - structure of chromatin fibre, scaffolds, euchromatin and heterochromatin, single copy genes in prokaryotes and Eukaryotes. Ig genes, overlapping genes, split genes, Lampbrush and Polytene Chromosomes.

- Recombination in Bacteria – Transformation, Conjugation, F - Mediated sexduction. Generalized and specialized Transduction. Mechanism of recombination and Recombination frequencies.; Mapping of bacterial chromosome, Use of interrupted mating technique- Transduction and recombination frequencies. Data elucidation. Construction of gene map. Benzer's classical studies on the r-II region of T4 phage, complementation test, Elucidation of T4 phage genetic map.

Unit-VII Molecular biology

- Replication and Repair:Semi-conservative mode of replication, experimental evidence of Meselson – Stahl and Cairns autoradiography experiments. Replication fork, continuous and discontinuous DNA synthesis. Evidence for Okazaki fragments – RNA primers, Enzymes and protein in replication ; Single Strand DNA binding proteins (SSB), Helicases, Topoisomerases, DNA ligases, DNA polymerases. E.coli DNA polymerase I, II and III, Eukaryotic DNA polymerases. Rolling circle replication. Replication of ϕ×174 and *E. coli* NDA. Eukaryotic DNA replication. Autonomous replication sequences (ARS) and regulation of plasmid DNA replication. Mitochondrial DNA replication. Termination and fidelity of DNA replication. Nearest neighbour base pair analysis. Replicons and termination signals. Inhibitors of DNA replication. Reserve transcriptase. DNA damage and repair: Photo reactivation, Direct reversal of damage, excision repair, Recombination repair. The SOS response.

- Transcription- Polynucleotide phosphorylase - RNA polymerases, structure of *E. coli* – RNA polymerase, Eukaryotic RNA polymerases – Template binding, promoters and enhancer sequences. Initiation, elongation and termination of RNA synthesis. Monocistronic and polycistronic RNAs. Post-transcriptional modifications of different RNA molecules – Eukaryotic mRNA – capping, methylation and poly adenylation. RNA splicing. Splicing mechanisms. Splicing of nuclear pre- tRNA, Introns, group I and group II and pre- mRNA splicing. Excision of multiple Introns, catalytic RNA.

- Translation -(Protein synthesis): The genetic code elucidation, experimental studies of Nirenburg and Khorona. General features of genetic code, codon degeneracy and universality. Mitochondrial genetic code, tRNArole in protein synthesis. Amino acyl-tRNAsynthetases, wobble hypothesis. Mechanism of initiation, elongation and termination of protein synthesis. Translational factors. Inhibitors of protein synthesis; antibiotics and other inhibitors. Post translational modifications. Protein sorting and targeting. The signal hypothesis, signal sequences and signal recognition particle, molecular chaperones, protein degradation, Lysosomal degradation. The ubiquitin pathway-protein stability and N-end rule.
- Mutations- Mutagens, transitions, transversions, frame shift mutations, deletion, transposon, mutagensis ; suppressor mutations.

- Regulation of gene expression : House keeping genes, constitutive genes, induction and repression. Regulatory proteins – DNA binding motif of regulatory proteins, role of Zinc fingers, leucine zippers, helix- turn – helix. Regulation of gene expression in prokaryotes operons, fine structure of lac operon, Regulatory protein. Repressors and the catabolic activator proteins, Negative regulation ; Positive regulation, Dual functions of the repressor, the arabinose operon. Transcriptional control by attenuation, the trp and his operons.
- Regulation of gene expression in Eukaryotes – Eukaryotic promoters, positive regulation, gene amplification, gene rearrangement, translational control, hormonal regulation.- Homeotic genes and their regulation.

UNIT-VIII Immunology

- Types of immunity – Innate and adaptive. Antigens, Super antigens, Adjuvants. Cells and organs of the immune system -Thymus, bone-marrow, spleen, lymph node. T and B lymphocytes – Origin, activation, differentiation, characteristics and functions. Nature of T and B cell surface receptors. Major Histocompatibility Complex- H-2, HLA, Polymorphism of MHC molecules. Congenic and inbred strains of mice. MHC restriction and its role in immune response, Antigen presenting cells, Processing and presentation of antigens.

- Structure of immunoglobulins, Immunoglobulin classes and biological activities. Isotypes, Allotypes, Idiotypes. Immunoglobulin genes and antibody diversity, Class switching, Humoral and cell-mediated immune responses, Cytokines, Interleukins, Interferons, The Complement components and biological consequences of complement activation.

- Antigen-antibody interactions: Antibody affinity and avidity, Precipitation reactions – Immunodiffusion, Radial immunodiffusion, double immunodiffusion, immunoelectrophoresis, Rocket immunoelectrophoresis, Agglutination reactions- Hemeagglutination and complement fixation, Immunoflourescence, FACS, RIA, ELISA, Immunoblotting, Hybridoma technology - production of monoclonal antibodies and their applications, humanized antibodies.

- Immune effector mechanisms – Hypersensitivity: immediate (type I, type II, type III) and delayed hypersensitivity reactions, Immunodeficiencies - SCID and AIDS. Autoimmunity - organ specific (Hashimoto's thyroiditis) and systemic (Rheumatoid arthritis) diseases. Tissue transplantation - auto, allo, iso and xenograft, tissue matching, transplantation rejection, mechanism and control, immunosuppressive agents. Cancer immunology – Tumor associated antigens, Immunological surveillance of cancer.

UNIT-IX Molecular Physiology

- Composition of blood, erythrocytes, leucocytes, thrombocytes, Coagulation of blood and fibrinolysis. Respiratory organs and mechanism of respiration. Hemoglobin and transport of gases, Physiology of heart, Digestion and absorption of foods. Structure of kidney and nephron. Physiology of kidney. Regulation of electrolyte, water and acid base balance in the body.

- Structure and organization of muscle cell, types of muscles. Molecular organization of contractile systems nd molecular mechanisms of contraction and relaxation of muscle. Biochemical changes associated with muscle contraction and relaxation. Structure of nerve cell, origin of membrane potential, mechanism of propagation of nerve impulse in unmyelinated and myelinated nerve fibers. Synapse – types of synapses, transmission at adrenergic and cholinergic nerve endings. Blood brain barrier, Neurotransmitters. Physiology of vision.

- Composition and structure of cell membranes, Molecular constituents of membranes, asymmetric organization of lipids and proteins, fluidity of membranes, different membrane models,. Membrane channels and pups, ligand gated ion channels, Ionic channels. Molecular models of transport mechanism, Membrane biogenesis, cell- cell interactions, ionophores, gap junctions, artificial membranes and liposomes.

- Cell signaling process (Autocrine, paracrine, Telecrineactions). Mechanism of action of steroid and peptide hormones Molecular mechanism of signal transduction - nature and types of receptors, ligand-receptor interactions, Scatchard plot, up anb down regulation of receptors, concept of second Messengers with focus on cAMP, cGMP, Calcium, IP3 Nitric oxide.

Unit-X Clinical Biochemistry and Endocrinology

- Gastrointestinal hormones - Gastrin, secretin and cholecystokinin. Disorders of gastric function, methods of evaluation. Pancreatic exocrine secretions, pancreatic diseases, steatorrhoea. Malabsorption syndrome – tests for their evaluation and significance. Plasma proteins – Properties, functions and their variations in diseases, Plasma lipids and lipoproteins, Interrelationship of lipids, lipoproteins and apolipoproteins. Erythropoiesis, abnormalities in blood formation. Anemias. Heamoglobinopathies. Cerebrospinal fluid – composition in health and diseases.

- Liver function tests, their significance, Liver diseases – Jaundice, hepatitis, gall stones, cirrhosis and fatty liver. Free radical mechanism and role of reactive oxygen species in diseases. Role of liver in metabolic regulation and drug metabolism.

- Clinical enzymology - Plasma enzymes in diagnosis and prognosis, Isoenzymes in health and diseases (Liver, cardiac and skeletal muscle enzymes)

- Kidney – Renal hormones –Renin, erythropoietin and angiotensin. Investigations of renal functions, biochemical investigation of renal disorders. Nephritis, nephritic syndrome and urolithiasis. Compensatory mechanism for acidosis and alkalosis.

- Pancreatic hormones – Biosynthesis of insulin, regulation of secretion of insulin and glucagon, their role in carbohydrate, lipid and protein metabolism. Endocrine disorders of pancreas – Diabetes mellitus, melliturias, hypoglycemia. Glucose tolerance test.

- Thyroidal hormones – Chemistry, function and metabolism. Hypo and hyper thyroidism, tests for thyroid function. Parathyroid hormones – Parathormone and calcitonin, their role in calcium and phosphate metabolism, abnormalities of parathyroid functions and methods of evaluation.

- Adrenals - Chemistry and biosynthesis of adrenal medullary and adrenal cortical hormones. Disorders of adrenal cortex and adrenal medulla, tests for the evaluation of adrenal functions. Biochemical effects of tumours.

- Synthesis, secretion, transport and biological actions of hypothalamic, adenohypophysial and neurohypophysial hormones. Hypothalamic disorders. Pituitary Clinical syndromes and their evaluation. Penial hormones – Melatonin and serotonin.

- Chemistry, biosynthesis and role of androgens, estrogens and progesterone.
  Hormonal regulation of menstrual cycle, Hormonal contraception. Placental hormones. Biochemistry of reproductive disorders, pregnancy toxemia, pregnancy tests.

UNIT-XI Genetic Engineering

- Introduction to genetic engineering – Tools for genetic engineering – isolation of genes from prokaryotic and eukaryotic organisms – mechanical shearing – restriction endonucleases - classification of restriction and modification systems – mechanism of action of restriction and modification enzymes – Type II restriction endonucleases – classification and nomenclature of Type II restriction endonucleases – application of restriction endonucleases in genetic engineering – determination of restriction map – RFLP – application of RFLP in forensic science – agriculture and medicine.

- Polymerase chain reaction (PCR) – theory and practice of PCR , RT-PCR, Inverse PCR, Nested PCR and their significance – designing of primers – degenerate primers – application of PCR technology in Molecular Cloning – uses of PCR

in Biology, Agriculture and Medicine. In-vitro synthesis of oligos – using PCR technology – chemical synthesis of genes and for performing in-vitro mutagenesis. RAPD and its significance. cDNA synthesis : Biochemistry and mechanism of action of Reverse transcriptase- isolation of eukaryotic mRNA – synthesis of cDNA.

- Cloning vectors – properties of cloning vectors - vectors of plasmid origin. Types, properties and functions. construction of plasmid vectors(pUC18/19, pBR322, Blue Script Vectors). Shuttle vectors – structure, function and heir significance in cloning genes other than E.coli hosts. Expression Vectors- use of lac, trp, trc, tac and bacterio phage lambda pL, pR promoters, bacteriophage T7 promoter and their significance in constructing expression vectors. Vectors for cloning genes in Plants – construction of Binary vectors, vectors of viral origin-cosmids, phagemids, vectors derived from bacculoviruses and their significance. Bacterial artifical chromosomes (BAC). Yeast Artificial Chromosomes (YACs), yeast expression vectors: Vectors for cloning in mammalian cells.

- Organisms and tissue culture systems commonly employed in genetic engineering experiments E.coli hosts – HB101, JM109, JM109(de3), BL21 de3 pLysis, E.coli DH5 alpha – Brief description about genotype and significance of the E.coli hosts in a cloning and expression of heterologous genes.

- Cloning strategies: Joining of DNA fragments to vectors- homopolymer tailing, cohesive and blunt end ligation and using linker DNA in cloning. Calf alkaline phosphatase (CIP) – mechanism of action – its significance in Molecular Cloning – Shrimp alkaline phosphatase – significance and mechanism of action.
  Construction of cDNA and genomic libraries. Partial digestion of DNA – use of cosmids, BAC and YACs in generation of genomic libraries.

- Introduction of recombinant DNA molecules into selected host cells. Biological and non-biological methods –preparation of competent cells of E.coli, Yeast for Tansformation – Transfection – Particle bombardment. *Agrobacterium* mediated transformation and its significance in delivering foreign genes into plant cells. Screening of genomic libraries for detection of clones- genetic techniques – complementation techniques – molecular hybridization methods – colony hybridization.

- Characterization of cloned genes. Determination of restriction map and sequencing of cloned DNA using Chemical method (Maxam and Gillberts), Sanger's dideoxy chain termination method and Automation of DNA sequencing use of Fluorescent probes in determination of DNA sequence. Strategies involved in generation of total genome sequence – primer walking and alignment of DNA sequences – identification of regulatory elements – promoter

test vectors, use of reporter genes – Lac Z, GUS, GFP in characterization of regulatory elements. Primer extension studies – mapping of transcriptional start sites (TSP) – prediction of Open Reading Frames (ORFs).

- Protein engineering: Site-directed mutagensis; Adding disulfide bonds – Changing asparagines to other amino acids – Modification of metal cofactor requirements. Increasing of specific activity – Stability to thermal and salinity condition.

- Protein engineering: Site-directed mutagensis; Adding disulfide bonds – Changing asparagines to other amino acids – Modification of metal cofactor requirements. Increasing of specific activity – Stability to thermal and salinity condition.

- Principles and preparation of DNA and RNA probes and their applications: Study and expression of cloned genes in Prokaryotes and Eukaryotic systems. Microbial production of Interferon, Human growth hormone, Insulin in *E.coli.*

UNIT-XII Applied **Biochemistry and Bioinformatics**

- Bioprocessing **:** Importance of fermenters – significance of scale up, design of basic fermenter, optimization of Large-Scale fermentation, Types – continuous fermentation batch fermentation – fed-batch fermentation – Solid-state fermentation. Types of reactors - Stirred Tank, Bubble Column, Internal-Loop Airlift, External-Loop Airlift. Downstream processing – Principle, various stages of downstream processing, and methods.
- DNA fingerprinting, SNPS, Mapping Genes – Somatic cell hybridization mapping, FISH, Transposon tagging. RNA silencing – siRNAs and anti- sense RNAs their design and applications; shRNA, epigenetic gene silencing.
- Gene Therapy – Ex-vivo gene therapy – In-vitro gene therapy – viral and non-viral gene therapy.
- Generation of anti-sense RNA – Anti-sense RNA technology and its significance in agriculture and medicine.Animal cell and tissue culture: Composition and preparation of culture media, Primary cultures, established/continuous cell lines. Tissue and organ culture. Stem cells – Sources- embryonic stem cells, adult stem cells, cord blood stem cells. Generation of stem cells by cloning, stem cell differentiation, stem cell plasticity, preservation of stem cells. Organogenesis through stem cells for transplantation. Applications of stem cell therapy-Parkinson's disease and Alzheimer's disease
- Vaccines: Principles of vaccination, Design of vaccines. Conventional vaccines – Whole organism, live and attenuated, purified macromolecules. New generation vaccines- Recombinant antigen vaccines, recombinant vector antigens, DNA vaccines, synthetic vaccines, edible vaccines. Vaccine delivery systems – Liposomes, micelles, ISCOMS. Strategies for developing vaccines for malaria, HIV and

Salmonellosis.

- Gene therapy –Types and use of rDNA constructs for gene therapy. Microarrays and biochips. Principle and applications of Metabolic engineering. Principle and applications of Nanotechnology.
- Genome Projects**:** General introduction to genome projects. Human Genome Project (HGP) - Science behind HGP, facts at genome, chromosome and gene level. Benefits of HGP in medicine, agriculture, evolution, forensic science. Ethical Legal Social Implications (ELSI) of HGP- fairness in the use of genetic information, privacy and confidentiality, psychological impact and stigmatization. Genetic testing - standards, quality control and commercialization.
- Introduction to Bioinformatics**:** Various disciplines and applications of Bioinformatics- Databases, Genome analysis, functional genomics, proteomics, metabolomics, phylogenetics, structure prediction, drug design, pharmacogenomics.

- Biological Databases**:** General introduction, need of DB, database searching options-sequence alignment, An overview of Public molecular DATABASES (EMBL, GenBank, DDBJ, GSDB) - SWISS-PROT database, PDB, Database querying NCBI
- Proteomics: Introduction, principle, technique, 2-D database. Gel analysis software, post gel analysis, MALDI-TOF, MS analysis. Significance and applications of proteomics in biology.
- Molecular Modeling & Drug Design: Homology modeling, Model building tools-Preparation of alignment and template files. Building protein 3-D model with SwissPDB viewer, energy minimization. Molecular Visualization Packages: Rasmol - basic utilities and commands for protein visualization in 3-D. study of protein-DNA interactions, stereo mode, visualization of hetero atoms, HOH, metals, electron density maps, N and C terminals, disulfide bonds. Phases of drug development and drug design in genomicera.