# APRCET 2023-24 62-BIOTECHNOLOGY

### 1. CELL BIOLOGY

Mechanism of cell division Cell cycle – Molecular events including cell cycle check points and Cdk – cyclin complexes and their role in cell cycle regulation. Ultra structure of plasmamembrane – Components and membrane asymmetry. Transport processes-active transport, ionophores and ionchannels. Exo and endocytosis. Phago and pinocytosis.

General morphology and functions of endoplasmic reticulum. Signal hypothesis. Ribosomes- eukaryotic and prokaryotic. Ribosomal proteins. Role of Golgi in protein secretion. Lysosomes and peroxisomes. Cytoskeletal elements. Cell-cell interaction.

Mitochondria – structure, biogenesis and enzymatic compartmentation. Organization of mitochondrial respiratory chain, mechanism of oxidative of phosphorylation. Ultra structure of the chloroplast. Photosynthesis- photophosphorylation. Carbon dioxide fixation in C-3, C-4 and CAM plants. Photorespiration.

Organic evolution: Origin of life. Species concept, population, dones, races, and subspecies. Mechanisms of speciation. Role of isolating mechanisms. Lamarckism, Darwinism, Neo-Darwinism, Synthetic theory of evolution. Micro, Macro and mega evolution, sequential and divergent evolution. Natural selection.

# 2. BIOMOLECULES

Chemical foundations of Biology – pH, pK, acids, bases, buffers, weak bonds and covalent bonds . Classification, Structure, properties and biological significance of carbohydrates. Biological role of peptidoglycans, glycosamino glycans and Lectins. Lipids – Classification, Structure and properties of fatty acids, triglycerides, phospholipids, sphingolipids and cholesterol.

Amino acids – Classification, structure and physico-chemical properties. Chemical synthesis of peptides –solid phase peptide synthesis. Proteins – classification, purification and criteria of homogeneity. Structural organization, sequence determination and characterization of proteins. Confirmation of proteins – Ramachandran plots. Denaturation of proteins. Hetero cyclic compounds – Heme and Chlorophylls.

Structure and properties of purines, pyrimidines, nucleosides, and nucleotides. Covalent structure of DNA and different forms of DNA – A,B and Z. DNA super coiling. Types of RNA and covalent structure of t-RNA. Classification, structure and physiological roles of Vitamins.

Hormones-classification and mechanism of action of steroid and protein hormones. Signal transduction cascade by cyclic AMP, Phospho inositate and calcium (Ca), G- proteins, growth factors and membrane receptor tyrosine kinases. Phytohormones and their physiological role

3. Microbial Physiology & Genetics

Bacterial reproduction and grown curve. Preparation of bacteriological media. Staining techniques. Differences between gram positive and gram negative bacteria. Methods of sterilization, pasteurization and disinfection. Microbes as pathological agents in plant and animals. Clinically important bacteria. Biohazards-safety precautions.

Chemical nature and classification of bacteriophages. Parasitic and temperate phages. Plant and animal viruses – multiplication of viruses. General characteristics of T Phase,  $\phi x 174$ , SV40, TMV. Clinically important viruses, retroviruses, HIV, Hepatitis B Virus and viral infections. General account of algae molds and yeasts. Clinically significant protozoans.

Microbial genetics: Recombination in prokaryotes, Transformation, conjugation, transduction and sexduction. Mapping of prokaryotic gene. Transposons, retrotransposons and mechanism of transposition. Viral genetics. Biology of plasmids. Extra chromosomal inheritance.

Genetics of Eukaryotes: Gene & Environment, Genotype and phenotype, Mendel's experiments, Dominance relationships. Multiple alleles, Gene Interaction, Gene mutations, Sex determination, Sex linkage, Linkage and recombination in diploids. Tetrad analysis. Elements of gene mapping, Pedigree analysis.

4. Analytical Tools and Techniques in Biotechnology

Principles and applications of light, phase contract, fluorescence, scanning and transmission electron microscopy. Properties of electromagnetic radiations. Principles, instrumentation and applications of UV, visible, NMR spectroscopy, mass spectrometry, X-ray diffraction. Flow cytometry.

Principles and applications of gel-filtration, ion-exchange and affinity chromatography. TLC, GLC and HPLC. Basic principles of sedimentation. Applications of preparative and analytical ultra centrifuges. Principles and applications of lyophilisation.

General principles of electrophoretic techniques. Poly AcryI amide GelElectrophoresis. Iso-electric focusing. Isotachophoresis. 2-D Electrophoresis. Capillaryelectrophoresis. Agarose gel electrophoresis of DNA and RNA. Blotting techniques. DNA fingerprinting.

Stable and radioactive isotopes. Detection and measurement of radioactivity. Applications of radioisotopes in biological sciences. Autoradiography. Non-isotopic tracer techniques. Principles and range of electrochemical techniques. Operation ofpH electrodes. Principles and applications oflon-selective and gas sensing electrodes. Oxygen electrodes.

### 5. ENZYMOLOGY & METABOLISM

Classification and Nomenclature of Enzymes, Enzyme kinetics, Factros affecting the rates of enzyme catalysed reactions. Assay of enzyme activity-units of enzyme activity. Multisubstrate reactions. Enzyme- substrate (protein ligand) binding. Methods for measurement of km. Coenzymes, metalloenzymes, and isoenzymes with examples.

Active site determination. Mechanism of enzyme action of Chymotrypsim & Trypsin, carboxy peptidase – A and ribonuclease A. Multienzyme systems. Covalent modification. Zymogen activation. Enzyme inhibition – Competitive, non-competitive and uncompetitive. Allosteric enzymes, Ribozymes and catalytic antibodies.

Glycolysis, Glycogenolysis, Glycogensis, Gluconeogenesis, HMP shunt path way and their regulation. Tricarboxylic acid (TCA) cycle, Glyoxylate cycle and its significance. Biosynthesis andoxidation of fatty acids. Metaboliam of cholesterol. Ketone bodies. Biosynthesis of Heme and chlorophylls

Protein turnover. General metabolic reactions of amino acids. Urea cycle. Nitrogen fixation. Essential and non-essential amino acids. Biosynthesis and degradation of aromatic and branches chain aminoacids. Inborn errors of amino acid metabolism. Biosynthesis of purine and pyrimidinenucleotides and their regulation. Catabolism of purines and pyrimidines.

### 6. MOLECULAR BIOLOGY

Organization of genetic material – Packing of DNA in to chromatin – protein components of chromatin, histones, nucleosome organization. Solenoids loops, domains & scaffolds. Gene amplification, polytene chromosomes. DNA replication – apparatus, enzymes involved and mechanism. Replication at telomeres. DNA damage and repair mechanism. Nuclear genome. C – value paradox. Mitochondrial & plastid genomes and genes. Fine structure of the eukaryotic gene. Split genes. Different kinds of genes: overlapping, assembled, polyprotein & nested genes.

Transcription in prokaryotes and eukaryotes. Mechanism of transcription, enzymes and transcription factors, zine finger, leucine zipper mechanism. Maturation and processing of m-RNA, splicing, 5' end capping & 3' end tailing, RNA editing and transport. RNAi and small RNAs.

Translation in prokaryotes and eukaryotes: Genetic code – properties of the genetic code, deciphering of the genetic code. Ribosome as a translation factory. t -RNA as an adaptor, its mode of function. Post translational modifications. Leader sequences & protein targeting.

Regulation of gene expression in prokaryotes –The operon concept, lac & tryp operons. Transcriptional control. Post translation control. Regulation in eukaryotes – Control by promoter, enhancer and silencers. Cis-trans elements. Environmental & developmental regulation. DNA methylation & gene expression. Chromatin structure & gene expression.

# 7: GENETIC ENGINEERING

DNA sequencing by chemical and enzymatic methods. Nucleic acid blotting – southern and northern blotting. DNA cloning. Enzymes used in genetic engineering : Restriction endonucleasestypes, nomenclature and properties. DNA polymerase-I polyncleotide kinase, DNA ligase, terminal nucleotide transferase, Reverse transcriptase, alkaline phosphatase,  $S_1$  nuclease.

Salient features of cloning vectors, types of cloning vectors – plasmids, cosmids, phages (lamda and M13 phages), animal (SV40, Baculo) and plant (CMV) viruses, Artificial chromosomes – YACs and MACs, Preparation of gene libraries and c-DNA libraries.

Techniques of gene transfer – transformation, transfection, micro injection, electroporation, lipofection and biolistics. Selection of r-DNA clones and their expression. Nucleic acid probes, colony and fluorescent in-situ hybridization.

Polymerase Chain Reaction and its applications. DNA micro array technology. Genomicsgenome sequencing by shot gun and hierarchical method. Genome annotation – identification of genes, promoters and exon – intron boundries.

# 8. BIOLOGY OF IMMUNE SYSTEM

Organisation and structure of lymphoid organs – bone marrow, thymus, spleen and lymphnodes. Cells of the immune system – B - Lymphocytes, T – Lymphocytes. Types of cell mediated immunity and lymphokine activated killer cells. Adjuvants and immunological tolerance.

Nature of antigens and antibodies. Structure and function of antibodies. Isotypes, Allotyes and Idiotypes. Antigen – antibody interactions. The generation of antibody diversity, antigen receptors on B & T ltymphocytes. Major Histocompatibility Complex (MHC). Human leukocyte antigens (HLA), MHC restriction and typing. Lymphokines. Complement system.

Immunological techniques – ELISA, RIA, Western Blot, Immunoblot and Immuno fluorescent techniques. FACS. Hybridoma technology – production and applications of monoclonal antibodies. Antibody engineering, chimeric antibodies.

Hypersensitivity –types of hypersensitivity – immediate and delayed hypersensitivity, autoimmunediseases, transplantation and immunity, immunity to infectious agents. Vaccines and Vaccination, types of vaccines including new generation vaccines. Tumor immunology.

# 9: CELL CULTURE TECHNOLOGY AND TISSUE ENGINEERING

Plant tissue culture technology: culture media – composition and preparation. Factors governing in vitro behavior, Somatic embryogenesis, organogenesis and plant regeneration. Culture types. Micro propagation, Haploids, somaclonal variations, metabolite production in cultures. Isolation of protoplasts, protoplast fusion and culture. Somatic hybridization.

Animal cell and tissue culture. Primary culture, balanced salt solutions and simple growth medium. Serum and protein free defined media. Cell lines, primary and established cell line cultures. Basic techniques of mammalian cell culture in vitro. Tissue and organ culture. Production and use of artificial tissues and organs –skin, liver and pancreas. Apoptosis – mechanism and significance.

The biology of stem cells – Different types of stem cells – embryonic stem cells, fetal tissue stem cells, adult stem cells; stem cell differentiation, stem cell plasticity – Differentiation versus stem cell renewal. Isolation and propagation of embryonic stem cells; chimeras; generation of knockout miceand knock-in technology.

Hematopoietic stem cells and bone marrow transplantation: Cells for hematopoietic reconstitution – Cord blood stem cells; cells for adoptive cellular immunotherapy; bone marrow transplantation – advantages and disadvantages. Allogenic, autologous, syngenic and congenic transplantation. Clinical applications of stem cell therapy; neurodegenerative diseases – Pakinson's disease, Alzhemers, spinal cord injury and other brain syndromes.

# 10: PLANT BIOTECHNOLOGY

Plant Genetic engineering: Gene cloning techniques, Techniques for gene transfer into plants. Mechanism of gene transfer by  $T_1$  and  $R_1$  plasmids as vectors. Reporter genes, transient gene assays and identification of transgenic plants. Molecular markers and their significance. RFLP, AFLP and QTL in plants. RAPD for molecular mapping and crop improvement.

Agricultural Biotechnology: Engineering of herbicide tolerance in plants, production of disease resistant plants by gene transfer; Development of insect resistant plants. Biotechnological strategies for engineering stress tolerance.

Altering protein and oil quality traits in seeds. Chloroplast transformation – advantages in tobacco and potato, plants for expression of bacterial, viral and eukaryotic genes. Edible vaccines and plantibodies. The genetic manipulation of crop yield by enhancement of photosynthesis.

Algal Biotechnology: Marine micro algae/sea weeds and their products. Edible sea weeds and their cultivation. Bio fertilizers – Blue green algal fertilizers – Azolla, Anabaena, symbiotic association.

Sea weed fertilizers. Mycorrhizal bio fertilizers, bacterial fertilizers. Bio pesticides in agricultural production.

#### 11: ANIMAL BIOTECHNOLOGY

Types and causes of male and female infertility, sperm collection, Cryopreservation, artificial insemination, Oocyte recovery, superovulation, oocyte maturation in vitro, In vitro fertilization in humans and cattle. Embryo culture, embryo transfer in farm animals. Immunocontraception – hormonal methods. Biotechnological approaches for the management of pests, mosquitoes and nematodes. Live stock improvement.

Production of transgenic animals – mice, sheep and fish. Molecular pharming and animal cloning. Somatic cell nuclear transfer in humans – Legal and ethical aspects. Potential applications of transgenic animals – Animal models for diseases and disorders.

The concept of aquatic biotechnology and blue revolution. Economically important aquatic resources form fresh water, brackish water and marine habitats – the finfish, shellfish, lime fish, algae, corals, and holothurians. Bioactive compounds from corals. Fish by products. Pearl culture technology – principles and applications.

Aquaculture – Fresh water fish culture practices and types. Freshwater prawn culture. Brackish water fish, shrimp and crab culture practices. Fresh water fish hatchery and seed production. Hypophysation and induced breeding techniques. Eyestalk oblation. Techniques involved in transgenic fish production. Postharvest technology. Diagnosis of shrimp & fish diseases caused by bacterial, fungal and viral pathogens using molecular methods.

# 12: MEDICAL AND ENVIRONMENTAL BIOTECHNOLOGY

Health care products. Products from recombinant DNA Technology – insulin, growth hormone, factor VIII, tissue plasminogen activator, interferons, lymphokines and Hepatitis-B vaccines.

Disease diagnosis: DNA probes, Enzyme probes – glucose oxidase, lactate oxidase, monoamine oxidase, PCR amplification and diagnosis – Applications in forensic medicine. Genetic diseases and gene therapy. Current strategies for development of vaccines against HIV, Malaria, Tuberculosis.

Environmental pollution – types, sources and control, Reduction of environmental impact of industrial effluents, chemical herbicides and fertilizers. Removal of oil spills. Environmental monitoring and bio monitoring. Bioremediation – solid and liquid waste treatment. Biomass and energy production from waste. Bioleaching. – Microbial recovery of metals and acid mine drainage. Water pollution and its control. Microbiology of waste water treatment.

Environment and energy: Renewable sources of energy – Biogas, waste materials, energy corps, cellulose. Production of energy and fuel using microorganism – Biofuels and Biodiesal. Global environmental problems: Ozone depletion, UV-B, Greenhouse effect, Biodiversity, benefits to mankind – Conservation. Ecology and sustainable development.

# 13: HETEROLOGOUS EXPRESSION AND DOWN STREMAM PROCESSING

Heterologous Expression: Expression vectors and hosts Generally Regarded As Safe (GRAS) organisms. Production of active recombinant proteins of mammalian/Eukaryotic origin in prokaryotes. Large scale production of proteins from recombinant micro orgnisms. Principles of microbial growth – Batch fermentation, feed –batch fermentation – continuous fermentation, high density cell cultures – Bioreactors – Large scale fermentation system- tandem Airlift reactors – Single Stirred tank reactors.

Downstream processing: Harvesting microbial cells – Membrane filtration system, high speed semi continuous centrifugation – disrupting microbial cells. Large scale purification of recombinant proteins – Chromatography systems and analytical methods for large scale purification. Stabilization of the proteins.

Processing technology: Microbial metabolites – Organic solvents (Alcohol, Acetone, Butanol), Organic acids (Citric acid, lactic, acid), Wines and beers, Antibiotics (penicillin, streptomycin, tetracycline, semi synthetic penicillins), Vitamins (Vitamin B<sub>12</sub> and Riboflavin), Amino acids (lysine, glutamic acid). Production of single cell proteins.

Enzyme technology: Sources production, isolation and purification of enzymes for the industrial use. Application of enzymes in pharmaceutical, food processing and other industries. Different techniques of immobilization of enzymes, application and kinetics of immobilized enzymes. Design and operation of immobilized enzyme systems and bioreactors. Whole cell immobilization. Biosensors – principle and types.

### 14: BIOINFORMATICS AND BIOSTATISTICS

Scope of computers in current biological research. Basic operations. Architecture of computer. Introduction of digital computers. Organization, low level and high level languages, binary number system. The soft side of the computer – Different operating systems – windows, Linux. Introduction of Programming in C. Introduction to Internet and its Application. Introduction to Bioinformatics – Genomics and Proteomics. Bioinformatics – Online tools and offline tools. Biological databases. Types of data bases – Gen bank, Swiss port, EMBL, NCBL, and PDB. Database searching using BLAST and FASTA.

Multiple sequence alignment and Dynamic programming. Gene and Genome annotation – Tools used. Physical map of genomes. Molecular phylogeny – Concept methods of tree construction. Protein secondary structure prediction. Protein 3D structure prediction. Protein docking. Introduction to homology modeling. Computer Aided Drug Design (CADD) in Drug discovery. Brief description and tabulation of data and its graphical representation. Measures of central tendency and dispersion – mean, median, mode, range, standard deviation, variance. Simple linear regression and correlation. Types of errors and level of significance. Tests of significance – F & t tests, chi – square tests, ANOVA.