Syllabus for B.Sc. (Biotechnology) CBCS- 2018 Group I Core Courses SEMESTER - I

BSCBTV 131: BIOCHEMISTRY AND BIOPHYSICS

48 hours

Unit I (12 hours)

Aims and scope of biochemistry and biophysics

General classification, structure and functions of biomolecules: carbohydrates, proteins, lipids and nucleic acids. Detailed structure and general properties of monosaccharides, glycosidic bond; structural polysaccharides - cellulose, chitin, peptidoglycans; storage polysaccharides - starch, glycogen. Classification of standard amino acids, peptide bonds, general properties of amino acids, titration curve. Protein structure - primary, secondary, tertiary and quaternary with examples.

Unit II (12 hours)

Enzymes: History, general properties, active site, Michelis Menton equation, allosteric enzymes; nomenclature and classification. Enzyme inhibition types- reversible, non-competitive and uncompetitive with examples. Multienzyme and isoenzyme with examples. Brief account of applications enzymes: enzymes in genetic engineering - restriction enzymes and polymerases; enzymes in clinical significance - LDH, SGOT, SGPT and diagnostic kits.

Unit III (12 hours)

Structure and function of water, pH impact on biomolecular reactions, Handerson and Hasselbach's equation with applications. Buffers- types and applications. Laws of thermodynamics, free energy, ATP as biological energy currency.

Lambert - Beer's law, absorption spectrum, absorption maxima. Chromophores; UV, Visible and Infrared spectrophotometry with applications. Fluorescence, phosphorescence and spectroflurometry with applications. Brief account of principles and applications to understand the structure of molecules: X-ray crystallography and NMR.

Unit IV (12 hours)

Microscopy: Magnification, Resolution power, Optical - Bright field, dark field, phase contrast and fluorescence; Electron microscopy - TEM and SEM. Partition coefficient, adsorption chromatography, paper and thin layer chromatography - principle, methodology, applications and significance of $R_{\rm f}$ value. Gel filtration chromatography, affinity chromatography, agarose and polyacrylamide electrophoresis - principle and applications. Centrifugation: differential, density gradient and ultra - principle, instrumentation and applications

References

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BSCBTP 132: BIOCHEMISTRY AND BIOPHYSICS PRACTICAL (12 × 3 hours)

Qualitative tests for carbohydrates - monosaccharides, disaccharides and polysaccharides.

Qualitative tests for proteins.

Qualitative tests for lipids.

Assay of enzymes – salivary amylase and urease.

Estimation of reducing sugar by DNS and Nelson Somogyi method

Estimation of proteins by Lowry's and Biuret method

Lambert – Beer's law

Absorption maxima of a solution

Paper chromatography

Thin layer chromatography

Electrophoresis

Differential centrifugation to separate cell organelles

Microscopy

48 hours

Unit I (12 hours)

Cell theory, classification of cell types. Levels of organisation in cell biology: cell, tissue, organs and organism. Cell locomotion - amoeboid, flagellar and ciliary. Prokaryotic and eukaryotic cells: ultrastructure, cell membrane and cell wall. Golgi complex, endoplasmic reticulum, mitochondria, chloroplasts, lysosomes, peroxysomes, interphase nucleus (nuclear membrane, nucleoplasm and nucleolus) and ribosomes. Cytosol and cytoskeletal structures - microfilaments, intermediate filaments and microtubules.

Unit II (12 hours)

Cell division: mitosis and meiosis, cell cycle, cell synchrony and its importance. Cell to cell interactions and signal molecules. Cell senescence and programmed cell death (apoptosis). Cell differentiation in plants - *Arabidopsis thaliana* and animals - *Drosophila melanogaster*.

Unit III (12 hours)

Chromosomes: chemical composition, structural organisation of chromatids, centromeres, telomeres, chromatin, nucleosome organisation. Euchromatin and heterochromatin. Special chromosomes: polytene and lampbrush chromosomes. Banding patterns in human chromosomes-G, C, R and T banding.

Mendelian laws of inheritance, gene interactions- complementary, supplementary, epistasis and codominence with suitable examples. Sex determination in plants and animals, sex linkage, non-disjunction as a proof of chromosomal theory of inheritance and gene mapping. Extrachromosomal inheritance, mitochondria and chloroplast genetic systems.

Unit IV (12 hours)

Fine structure of gene - recon, muton and cistron.

Spontaneous and induced mutations, chemical and physical mutagens, induced mutations in plants and microbes and its applications.

Structural and numerical aberrations of chromosomes; evolutions of wheat, cotton and rice. Hereditary defects: Kleinefelter, Turner, Cri-du- Chat and Down syndromes. Analysis of mutations in Biochemical pathways, one gene - one enzyme hypothesis. Population genetics: Hardy- Weinberg equilibrium, gene, genotypes and gene frequencies.

References

Dale JW. 1990. Molecular genetics of Bacteria. John Wiley and Sons.

De Robertis EDP and De Robertis EMF. 1995. *Cell and Molecular Biology*. 8th edition, BI Waverly Pvt. Ltd., New Delhi.

Gardner et al. 2003. Principle of Genetics - 8th edition. John Wiley and Sons, New York.

Gupta ML. and ML. Jangir. 2002. *Cell Biology- Fundamentals and Applications*. Argosies, Jodhpur, India.

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Taylor DJ. Green NPO and Stout GW. 1998. *Biological Science* 3rd Edition, Cambridge edition, Cambridge University Press, UK.

BSCBTP 182: CELL BIOLOGY AND GENETICS PRACTICAL (12×3 hours)

Study of stages of mitosis: staining of onion root tips.

Study of meiosis: staining of grass hopper testis or onion/*Rhoea/Tradescantia* flower buds. Microtomy: preparation of blocks for sectioning. Preparation of permanent slide and study of section.

Study of cell organelles: isolation and staining of mitochondria and chloroplast.

Separation of photosynthetic pigments by paper chromatography.

Separation of *Drosophila* eye pigments by circular paper chromatography.

Counting of RBC/WBC by Haemocytometer.

Micrometry.

Study of *Drosophila* (wild type and mutants), sex comb mounting.

Salivary gland chromosome isolation and staining.

Karyotyping.

Genetic problems.

BSCBTV 231: MICROBIOLOGY AND IMMUNOLOGY

48 hours

Unit I (12 hours)

Aim, scope and historical perspectives of microbiology. Contributions of early microbiologists: Leeuvenhoek, Louis Pasteur, Robert Koch and Edward Jenner. Concepts of sterilization and disinfection: dry heat, moist heat, radiation, chemical and filtration. Antibiotics: classification and mode of action. General classification of microbial kingdom - classical, nutritional and molecular approaches. Introductive concepts in virology-classification, structure and life cycle - lysogenic and lytic cycle.

Unit II (12 hours)

Selected representatives of archaebacteria (methanogens), eubacteria (*Escherichia coli*) and eukaryotic (*Saccharomyces*) microbes and their characteristics. Structure and functions of microbial cell wall, lipopolysaccharides, flagella, capsules, endospores, pili (fimbriae), cell membranes and cell inclusions.

Isolation techniques and media: Selective isolation of microorganisms (physical and chemical). Nutrition and growth kinetics. Microbes in extreme environments- thermophiles, psychrophiles, acidophiles, alkaliphiles, halophiles and barophiles. Microbial interactions – positive and negative interactions between microbes-microbes, interactions between microbes and plants, microbes and animals.

Unit III (12 hours)

Historical perspectives in immunology, origin and diversity of immune systems. Classification of immunity: innate and adaptive immunity. Immunity systems: organs, cells of immune system, major histocompatibility complexes (MHCs) - types, structure and their functions.

Unit IV (12 hours)

Bacterial conjugation, transduction and transformation. Structure, types and functions of antigens. Structure, types and functions of antibodies. Antigen-antibody reactions - precipitin test, agglutination test, complement fixation (or complement cascade) reaction and ELISA. Immunoblot - types, principle and applications. Immunological disorders -autoimmune diseases - Brief descriptions of two autoimmune diseases- Rheumatoid arthritis and myasthenia gravis. Acquired immunodeficiency syndrome: description of causative agent, and briefly description of mechanisms. Hypersensitivity and allergy. Vaccines-classical and modern, vaccination and immunization.

References

Abbas A, Lichtman AH and Pillai S. 2015. *Cellular and Molecular Immunology*. Elsevier Saunders Co.

Brock TB and Madigon. 1988. *Biology of Microorganisms*. Prentice Hall, New Jersey. Cassida, L.E. 1968. *Industrial Microbiology*. John Wiley & Sons.

Ivan Riott, Jonathan Brostoff and David Male. *Immunology* 3rd edition. Mosby Publishers. Janeway and Travers. *Immunobiology* 3rd edition. Churchill Livingstone Publications.

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Prescott LM, Harley JK and Oxford DA. 1993. *Microbiology*. WMC Brown Publishers, USA.

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Sharma PD. 1991. Microbiology, Rastogi Publications, Meerut.

Subba Rao N.S. 1974. *Soil Microbiology*, 4th edition, Oxford & IBH Publishers, New Delhi Torture GJ, Frank BR, and Case CL. 1992. *Microbiology- An Introduction*. Communing Publishing Company Inc, California.

BSCBTP 232: MICROBIOLOGY AND IMMUNOLOGY PRACTICAL (12X3 hours)

Instrumentation and microscopy with special reference to microbiology and immunology. Staining of microorganisms - Gram staining, capsule staining, spore staining, negative staining.

Preparation of media. Aseptic techniques.

Study of bacterial colony characteristics.

Biochemical activities of microorganisms - indole, methyl red, Voges Proskauer and catalase tests.

Antibiotic sensitivity of microorganisms. Isolation of microorganisms - air, water, human body and soil.

Isolation of bacteriophages from sewage.

Effect of pH and temperature on bacterial growth.

Blood grouping and Rh factor determination.

Study of different types of leucocytes.

Separation of macrophages.

Immunodiffusion studies.

SEMESTER - IV

BSCBTV 281: MOLECULAR BIOLOGY AND RECOMBINANT TECHNOLOGY 48 hours

Unit I (12 hours)

Discovery, structure and types of DNA. Experiments on DNA as genetic material. Replication of DNA in prokaryotes and eukaryotes. Structure of prokaryotic and eukaryotic genes. Mechanisms of DNA recombination in prokaryotes and eukaryotes. Discovery and types of transposons in prokaryotes and eukaryotes.

Unit II (12 hours)

Prokaryotic and eukaryotic transcription and translation. Prokaryotic gene expression - *lac* and *trip*. Eukaryotic gene expression - transcription factors, e.g. yeast.

Unit III (12 hours)

Aims, objectives and scope of gene cloning and recombinant DNA technology. Isolation and purification of DNA from bacterial, plant and animal cells. Tools of DNA modification: restriction enzymes- properties, classification, types with examples; ligation, DNA modifying enzymes. DNA vectors: plasmids, bacteriophages, phagemids, cosmids, plant and animal viruses.

Unit IV (12 hours)

Genomic and cDNA libraries: features, construction and application. Screening and selection of recombinants by selection media, insertional inactivation. PCR- principle, protocol and applications. Blotting techniques: Southern, Northern and Western- principle and applications. Probes - types, preparation and application. DNA finger printing- principle and applications. Hazards and biosafety measures for recombinant DNA technology and GMOs.

References

Alberts B, Bray D, Lewis J, Raff M, Roberts K and Watson JD. 2002. *Molecular Biology of the Cell* 4th edition. Garland Publishing, Inc., New York.

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Wu, W. et al., 2004. Gene Biotechnology. CRC Press.

BSCBTP 282: MOLECULAR BIOLOGY AND RECOMBINANT TECHNOLOGY PRACTICAL

 $(12 \times 3 \text{ hours})$

Separation and study of cell organelles.

Isolation of DNA from bacteria, plant and animal tissues.

Isolation of RNA

Tests for DNA/RNA/proteins isolated from tissues

Estimation of total DNA/RNA/protein from animal cells and plant cells

Agarose gel electrophoresis to separate DNA

Agarose gel electrophoresis to separate proteins – SDS PAGE

DNA ligation

Restriction digestion

Preparation of competent cells

Bacterial transformation

Western blotting technique

Plasmid isolation

BSCBTV 331: PLANT BIOTECHNOLOGY Unit I 40hours (10 hours)

History of plant tissue culture, technical terms and definitions in tissue culture. Establishing sterile cultures - plant tissue culture lab set up, sterilization methods for instruments and explants. Tissue culture media, plant growth regulators. Principles, methodology and applications of clonal or micropropagation – axillary bud culture, shoot tip culture, mesistem and mericlone culture. Haploid culture- principle, protocol and applications. Embryo culture – types, principle, protocol and applications. Embryo rescue.

Unit II (10 hours)

Callus induction, introduction to the process of embryogenesis- types and organogenesis. Synthetic seeds- principle, protocol and applications. Somaclonal variations- introduction, types, process, factors affecting process and applications. Single cell suspension cultures - types, methods, viability tests and applications. Secondary metabolites - introduction, classification, production *in vitro* methods and applications. Cryopreservation of plant tissues - introduction, principle, types, protocol and applications.

Unit III (10 hours)

Protoplast- introduction, principle; isolation methods- mechanical, enzymatic. CPW medium, source of enzymes. Isolation of protoplasts from intact tissue, callus, suspension cultures and haploid cells - protocols. Testing the viability of isolated protoplasts. Various steps and methods involved in the regeneration of protoplast. Markers used in the selection of hybrid cells. Somatic hybridization - introduction, principle, protocol; hybrids and cybrids. Protoplast fusion methods: chemical and electrical. Applications of protoplasts, hybrids and cybrids.

Unit IV (10 hours)

Genetic manipulations of plant cells- single cells, protoplasts protocols and applications. Structure of *Agrobacterium tumefaciens*, tumour formation in monocots and dicots, reporter genes used in genetic transformations. Root formation using *Agrobacterium rhizogenes* and applications. Genetic transformation – transgenic cotton, edible vaccines and transgenic brinjal: protocol and applications.

Diseases development in plants caused by bacteria (bacterial blight or rice), fungi (late blight of potato), virus (tobacco mosaic disease) and viroid (potato spindle tuber) – classification, symptoms, disease cycle and control measures. Systemic acquired resistance (SAR) and development of disease resistant plants.

References

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Srivatsava P.S. *Plant tissue culture and Molecular Biology*: Applications and prospects. Narosa Publishing House, New Delhi.

BSCBTP 333: PLANT BIOTECHNOLOGY PRACTICAL

(12x2hours)

Plant tissue culture laboratory set up

Different plant tissue culture media

Methods of sterilization of glassware, media and explants

Establishment of callus culture, seed, embryo culture

Anther and pollen culture

Clonal propagation - shoot tip and axillary bud culture

Establishment of suspension culture

Protoplast isolation and culture

Somatic embryogenesis and artificial seeds

Organogenesis from callus culture, and hardening of plantlets

Isolation of Agrobacterium from plants or soil and in vitro culture

Cocultivation of Agrobacterium with plant of interest

Cryopreservation methods

Unit I (10 hours)

History of the development of cell culture. Contributions of R.G. Harrison, Alexis Carrel. Hanging drop technique, watch glass technique.

Equipments and materials for animal cell culture. Essential, beneficial and useful equipments. Substrates (glass, plastic, treated surface, feeder layer).

Animal cell culture media. Media for immediate survival of cells (BSS). Media for prolonged survival of cells (natural and artificial). Natural media - embryo extract, lymph serum; artificial (defined) media- media with serum (DMEM), RPMI 1640 and media without serum (HITES). Importance of serum in culture media.

Basic techniques of mammalian cell culture *in vitro*: primary explants culture - technique, advantages and disadvantages; primary cell culture - technique. Disaggregation of tissue: trypsinization - cold and warm, collagenase treatment, mechanical methods. Measurement of cells - cell count and cell viability. Cell counting - hemocytometer, electronic cell counter; cell viability - trypan blue, MTT assay.

Cell separation techniques - density gradient centrifugation, immunopanning, MACS, centrifugal elutriation, FACS.

Maintenance of cell culture: medium change-need, method; subculturing- factors affecting, methods - monolayer and suspension culture.

Unit II (10 hours)

Cell lines: types (finite and continuous), characteristics, examples for commonly used cell lines - BHK 21 - C13, HeLa, CHO-K1, WI-38, Vero, 3T3, mouse L. Routine maintenance – medium change, subculturing.

Growth kinetics of cells in culture: growth curve – lag, log, stationery and plateu phase; PDT, multiplication rate, generation number.

Measurement of cell proliferation- MTT assay, ³[H]: thymidine incorporation. Cell

synchronization: methods - chemical blockade (³H: thymidine: double thymidine block, colcemid, vinblastin sulfate), low temperature procedure, starvation, centrifugation. Somatic cell fusion techniques: chemical/virus mediated, elctrofusion, LASER induced. Production of MAbs by hybridoma technology: technique, applications of MAbs. Selection of hybrids: HAT selection; cell cloning- types: dilution and suspension (agar gel and methocis). Stem cell cultures: types- totipotent, pluripotent, multipotent, unipotent; embryonic and adult. Methods of culturing applications. Cryopreservation - technique (freezing and thawing) and applications.

Unit III (10 hours)

Animal cloning - reproductive cloning (Dolly- nuclear transplantation), therapeutic cloning (Xenotransplantation). Gene manipulation in animals- cloning vectors and expression vectors. Gene transfer methods (transfection): chemical methods-CaPO₄ coprecipitation, DEAE dextran mediated, lipofaction; physical - microinjection, electroporation; biological method — retroviral infection. Reporter genes - GFP, antibiotic resistance markers (neomycin phosphotransferase). DNA microarray - method, applications. Gene therapy - somatic and germline. Somatic (SCID), gene therapy in cancer treatment (TNF gene, p53 gene replacement).

Study of animal diseases: Symptoms, disease diagnosis and treatment of AIDS, salmonellosis and Candidiasis and malaria.

Unit IV (10 hours)

Genetic engineering - Factor VIII, tissue plasmogen activator (tPA), hormones (growth hormone, insulin), hepatitis B vaccine. Silkworm as bioreactors: heterologus proteins (OFP, human growth hormone). Organ culture (3 dimensional cultures): methods- watch glass technique, raft method, agar gel method, grid method cyclic exposure to light and gas phase. Tissue engineering (artificial skin): methods-collagen method and mesh scaffolding method. Transgenic animals: transgenic cattle - tPA, AAT production; Animals as bioreactors (biopharming): mammary glands as bioreactors- production of regulatory proteins (alpha -1-antitrypsin[AAT], tPA), transgenic fish – GH induced fish, AFP (fish antifreeze protein) - method and production.

References

Butler M. 2004. *Animal Cell Culture and Technology* 2nd edition. BIOS Scientific Publishers

Cibelli JB, Lanza RP, Campbell K and West MD. 2002. *Principles of Cloning*. Academic Press

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BSCBTP 334: ANIMAL BIOTECHNOLOGY PRACTICAL (12 × 2 hours)

Lab set up and fumigation of the lab

Preparation and filtration of animal tissue culture media

Primary explants culture

Chick embryo culture (Spratt culture)

Isolation of bone marrow cells by flushing and primary culture

Culture of lymphocytes

Determination of viability of cells

Buffy coat preparation of WBC

Mammalian cell counting by Hemocytometer

Estimation of viability of cells by trypan blue dye exclusion

Staining for monolayer culture and suspension culture

Cryopreservation

BSCBTV 381: ENVIRONMENTAL BIOTECHNOLOGY Unit I 40 hours (10 hours)

Basic principles of environmental biotechnology and its relevance to environmental protection: definition, its role in waste management, manufacturing process and pollution control

Environmental pollution: definition, types- air, water and soil pollution – definition, causes, effects and control measures. Acid rain, photochemical smog, ozone depletion, greenhouse effect. BOD, eutrophification, Minamata disease, biomagnification. Biochemical cycles: definition, types – gaseous and sedimentary. Gaseous cycles- carbon and nitrogen. Sedimentary - phosphorous. Sulphur. Toxic element cycles- mercury, lead.

Unit II (10 hours)

Microbiological treatment solid wastes: composting, vermicomposting, land-farming. Biological treatment of liquid wastes (sewage): primary treatment, secondary treatment (activated sludge system, trickling filters), sludge digestion, septic tanks, oxidation ponds. Tertiary treatments.

Bioremediation: *In situ* and *ex situ* bioremediation. Phytoremediation, microbial bioremediation. Pollution control measures- control of air and water pollution. Indicator organisms. Permissible limits and indices for pollutants. Hazardous wastes and management: dyes and paints, distillery industry effluents, leather industry, radioactive wastes. Microbial mining, corrosion and remedies. Biomining (e.g. copper and gold). Microbiologically influenced corrosion (MIC) and remedies.

Unit III (10 hours)

Renewable and non-renewable resources. Conventional and non-conventional sources of energy. Biomass energy - firewood, plant and animal wastes, coal, gas and animal oils. Methanogenic bacteria and biogas, microbial H₂ production, bioethanol production, solar energy and solar energy converters, wind and tidal energy and its utilization. Energy gardens - *Pongamia* and *Jatropha*.

Unit IV (10 hours)

Microbial degradation of xenobiotics: pesticides, detergents, plastics. Degradation of organic compounds: cellulose, lignin, hydrocarbon. Degradation of economically valuable products: textiles, paper, leather, wood. Biocontrol agents: Bacterial, viral, fungal of plants as Biopesticides. Biofertilizers: utilization of Rhizobia, cyanobacteria, arbuscular mycorrhizae and ectomycorrhizae.

Coastal regulatory zone (CRZ), marine resources, environmental issues of freshwater and marine aquaculture. Genetically manipulated organisms (GMOs) - biopesticides e.g. *Bacillus thuringiensis*. GM foods and ethical issues.

References

Cassida, L.E. 1968. *Industrial Microbiology*. John Wiley & Sons.

Jogdand SN. 2010. *Environmental Biotechnology*. Himalaya Publishing House, Bangalore, New Delhi.

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Subba Rao N.S. 1974. Soil Microbiology, 4th edition, Oxford & IBH Publishers, New Delhi

Wang LK, Ivanov V, Tay JH and Hung YT. 2010. *Environmental Biotechnology*. Sringer publishers.

40 hours

Unit I (10 hours)

The set theory: basic concepts of sets, methods of describing a set. Tabular form, rule form renndiagram. Types of sets, subsets, power set, universal set. The binomial theorem: statements, based on theorem problems

Logarithm: basic concepts

Differentiation and integration: basic

Unit II (10 hours)

Measures of central tendencies: definitions with examples- mean, median, Geometric mean, Harmonic mean

Measures of dispersion: definitions with examples - range, quartile deviation, mean deviation, standard deviation

Probability: definition, sample space, event, complement of an event, subevent, union of events. Introduction of events, equally likely events, mutually exclusive and exhaustive events with illustrations.

Correlation: definition, types of correlation, Karl Pearson's coefficient of correlation, Spearman's rank correlation.

Regression: definition, two regression equations, properties of regression equations with problems

Unit III (10 hours)

Introduction to computer and organisation of computers

Digital and analogue computers, computer algorithm, computers in monitoring and automation.

Application of computers in coordination of solute concentration, pH and temperature of fermenters.

Computers as computational tools for bioinformatics.

Unit IV (10 hours)

Introduction to bioinformatics. Biology and bioinformatics: relevance of bioinformatics to study biomolecules, structure of RNA and DNA, genetic code, genes, protein structure, folding and functions.

Biological data bases and data tools: types of databases, database softwares, tools for genomics and proteomics, genome data visualisation tools, annotation, genome comparison and analysis and data submission.

Application of bioinformatics in agriculture, pharmacogenomics and aquaculture.

References

Arnold E. 1979. *Introductory statistics for Biology* 2nd edition, London.

Attwood T and Parry-Smith D. 1999. *Introduction to Bioinformatics*. Prentice Hall Publications.

Lewis AE. 2010. *Biostatistics*. Prentice Hall. New Jersey.

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Zar J. H. 1974. *Biostatistical analysis*. Prentice Hall, New Jersey

BSCBTP 383: ENVIRONMENTAL BIOTECHNOLOGY, BIOSTATISTICS AND BIOINFORMATICS PRACTICAL $(12 \times 2 \text{ hours})$

Estimation of alkalinity and salinity from water, soil or sewage Water microbiology and sewage analysis Determination of BOD and COD

Estimation of hardness of water

Estimation of total solids, dissolved and suspended solids

Estimation of inorganic phosphate and nitrogen in soil, sewage and water

Soil analysis- classification, water holding capacity and bulk density

Estimation of dissolves oxygen and carbon dioxide

Estimation of organic carbon.

Biogas production

Isolation of xenobiotic degrading microorganisms

Compost, vermicompost and biofertilizers

Studies on animals, plants and microbes in extreme habitats

Analysis of polluted water

Field trips to different biomes

Problems in biostatistics

Problems in bioinformatics

BSCBTP 384: Project

 $(12 \times 2 \text{ hours})$

Project Work