

## Ph. D ENVIRONMENTAL BIOTECHNOLOGY(BY COURSE WORK)

### SYLLABUS

#### COURSE I -RESEARCH METHODOLOGY

(from 2014-2015 to 2018-2019)

**Unit I Introduction to research methodology:** Basic and applied research, Essential steps in research, Defining there search problem, Research/Experimental design, Literature collection, Literature citation, Research report: components, Format of thesis and dissertation, Manuscript/research article, Review monographs, Bibliography and Reference, Significance of research.

**Unit II Biophysical methods:** Analysis of biomolecules using UV/Visible, fluorescence, circulardichroism, NMR and ESR spectroscopy, structure determination using x-ray diffraction and NMR; Analysis using light scattering, different type of mass spectrometry and surface plasma resonance methods.

**Radio labeling techniques:** Properties of different types of radioisotopes normally used in biology, their detection and measurement; incorporation of radioisotopes in biological tissues and cells, molecular imaging of radioactive material safety guidelines.

**UNIT III Tools and techniques:** Techniques used for purification and characterization of biomolecules: Centrifugation, Ultrafiltration, Chromatography, electrophoresis, spectrophotometry, GC-MS, LCMS, NMR,MALDITOF, X-ray Crystallography, Microscopic techniques including Fluorescence microscopy, Confocal microscopy, Atomic force microscopy and live cell imaging FACS analysis. Real time PCR, DNA microarray, new generation DNA sequencing, Protein Microarray, protein sequencing, FRET analysis

**Unit IV Computational methods:** Nucleic acid and protein sequence database; data mining methods for sequence analysis, web-based tools for sequence searches motif analysis and presentation.

**Unit V** Measures of central tendency and dispersal; probability distributions (Binomial, Poisson and normal) Parametric and non-parametric; confidence interval; errors; levels of significance; regression and correlation; t-test; analysis of variance;  $\alpha$ 2 test Multivariable analysis etc..

#### REFERENCES

1. C.R. Kothari, 2<sup>nd</sup> Edition (2004) Research methodology, Methods and techniques, New Age International (P) Ltd, Publishers, New Delhi.
2. Jenod H. Zar (1999) Biostatistical analysis by, Prentice Hall International, Inc. Press, London.
3. Attwood. T.K and Parry-Smith D.J. (2002) Introduction to Bioinformatics, Pearson education Singapore.
4. Sharma.BK. instrumental methods of chemical analysis.
5. Upadhyay, Upadhyay and Nath, Biophysical chemistry.
6. Khandpur R.S. Handbook of biomedical instrumentation Tata Mc GrawHill.
7. Brigal.L.Williams,A biologist guide to principle and techniques of practical Biochemistry

## COURSE II-ADVANCES IN ENVIRONMENTAL BIOTECHNOLOGY

**UNIT I Basic concepts of molecular biology** - DNA structure and organization into chromosomes - DNA replication - Repetitive DNA - Coding and non-coding sequences in genomes - Gene structure and expression - Mechanics of transcription, translation and their regulation in both prokaryotes and eukaryotes

**UNIT II Basic techniques in genetic engineering** - Genetic manipulation - Restriction Endonucleases - Introduction of cloned genes into new hosts using plasmid and phage vector systems – Expression cloning - Affinity purification of expressed proteins - Nucleic acid hybridization and polymerase chain reaction - DNA sequencing

**UNIT III Biotechniques** Buffers - Methods of cell disintegration - Enzyme assays and controls - Detergents and membrane proteins – Dialysis - Ultrafiltration and other membrane techniques - *Spectroscopy Techniques*: Raman Spectroscopy, Fluorescence, MS, NMR, PMR, ESR, API-electrospray, MALDI-TOF - Mass spectrometry and Plasma Emission spectroscopy - Chromatography Techniques- Chromatographic methods for macromolecule separation - Gel permeation, FPLC - *Electrophoretic techniques*: Theory and application of Polyacrylamide and Agarose Gel Electrophoresis, Capillary electrophoresis, 2D Electrophoresis, Gradient electrophoresis, Pulsed field gel electrophoresis

**UNIT IV Applied Biotechnology** Practical aspects of genetic engineering with microorganisms from extreme environment: Use of extremophilic microorganisms in waste treatment and methane production from agro industrial wastes - Production of enzymes like cellulase, proteases, amylases - Alcohol and acetic acid production – Biocomposting and Biomining - *Alternate fuels*: Source and mechanism of various biofuel production

**UNIT V Metagenomics** Metagenomics: Environmental Genomics, ecogenomics or community genomics, the study of genetic material recovered directly from environmental samples and future applications in bioremediation - Genetically modified organisms and Biosafety- a general account

### REFERENCES

1. Agarwal SK. Environmental Biotechnology, APH Publishing Corp., New Delhi.
2. Brown TA (2000) Essential Molecular Biology. Practical Approach. Vol 1, 2nd edition, Oxford University Press, UK.
3. Brown TA (2006) Gene Cloning and DNA Analysis: An Introduction. John Wiley & Sons.
4. Chatterji AK. Introduction to Environmental Biotechnology, Prentice Hall of India Pvt. Ltd, New Delhi.
5. Dubey RC. A Text Book of Biotechnology, S. Chand & Company Ltd., New Delhi.
6. Glick and Pasternak. Molecular Biotechnology - Principles and Applications of Recombinant DNA, Panima Publishing Corporation, New Delhi.
7. Gupta PK. Elements of Biotechnology, Rastogi Publishing House, New Delhi.
8. InduShekhar Thakur I.K. Environmental Biotechnology-Basic Concepts and Applications International Pvt. Ltd. New Delhi.
9. Jogdand SN. Environmental Biotechnology, Himalaya Publishing House, New Delhi.
10. Singh BD. Biotechnology, KalyaniPublishers , New Delhi.

## **COURSE III -METAGENOMICS**

### **UNIT I**

History of the Culture divide - Early Microbiology and Microscope- Pure culture – rRNA analysis and culturing – Metagenomics – Culture independent insight – Microbial diversity – Uncultivable – Achaea.

### **UNIT II**

Bio prospecting- Population genetics and micro heterogeneity- Symbiosis – Competition – Communication – role of small molecules – Sequence based screening for small molecules – Antibiotics as signal molecules – Chemical ecology – Sargasso sea explorations.

### **UNIT III**

Community Genome Analysis - Methods – Microarray – Functional gene arrays – Community genome arrays - Phylogenetic oligonucleotide arrays – Whole genome ORF arrays- Environmental Gene Tags – Environmental genomics.

### **UNIT IV**

Metagenomic approaches - Marine drug discovery platform – Sequence based analysis – Functional metagenomics – Heterologous expression – Identifying active clones – Screens, Selections, Functional anchors – Search for potential producers – Polyketide synthases.

### **UNIT V**

Industrial applications - White biotechnology – Novelty – Diversity – Elusive metabolites - High throughput - screening - Multiparameter footprint analysis – screening for industrial enzymes – Bioactive molecules – synthons – Putative gene products.

### **REFERENCES**

1. Board on Life Sciences, The New Science of Metagenomics:Revealing the Secrets of Our Microbial Planet, The National Academies Press, Washington, DC
2. Alan T. Bull. Microbial Diversity and Bioprospecting. ASM press. Washington, D.C
3. Brenden Wren and Nick Dorrell, Functional Microbial Genomics (Volume 33) (Methods in Microbiology), Academic Press
4. Alexander Hillisch and Rolf Hilgenfeld. Modern Methods of Drug Discovery, Birkhauser, Switzerland
5. James N. Kyranos. High throughput Analysis for Early Drug Discovery. Esevier Academic Press

## COURSE IV - AREA OF SPECILIZATION

### 1-ENVIRONMENTAL GENOMICS

#### UNIT I

Environmental genomics- overview, goals and objectives. Genomic approaches for studying molecular events perturbed by environmental agents in microbial, invertebrate, aquatic, plant, and mammalian model systems.

#### UNIT II

Basic Structure of gene and its regulation. DNA Sequencing: Principles of DNA sequencing, Pyrosequencing, Automated DNA sequencing, Shotgun sequencing – contig assembly, High through put sequencing

#### UNIT III

Exploration of genome databases- Nucleic acid isolation from environmental samples- Real-time PCR, multiplexing, gene signature arrays- Cloning analysis- Denaturing gradient gel electrophoresis (DGGE), Microarrays, NNGS

#### UNIT IV

Large scale genome sequencing strategies, Genome assembly and annotation, Genome databases of Plants, animals and pathogens, Basic concepts on identification of disease genes, role of bioinformatics-OMIM database, reference, genome sequence, integrated genomic maps. Comparative Genomics: Basic concepts and applications, BLAST2, Mega Blast algorithms, PipMaker, AVID, Vista, MUMmer, applications of suffix tree in comparative genomics, synteny and gene order comparisons Comparative genomics databases: Clusters of Orthologous Groups (COGs) Systems biology – Use of computers in simulation of cellular subsystems. Metabolic networks, or network of metabolites and enzymes, Signal transduction networks, Gene regulatory networks, Metabolic pathways: databases such as KEGG, EMP, MetaCyc, AraCyc

#### UNIT V:

Functional Genomics; Gene expression profiling; identification of SNPs, SNP, database (dbSNP). Microarrays: DNA microarray: database and basic tools, Gene Expression Omnibus(GEO), Array Express, SAGE databases, SMD. DNA microarray: understanding of microarray data, normalizing microarray data, detecting differential gene expression, correlation of gene expression data to biological process and computational analysis tools

Sequence comparison, structure analysis (especially active sites, binding sites) and comparison, pattern identification, etc. Gene/Protein function prediction using Machine learning tools: supervised/unsupervised learning, Neural network, SVM etc.

#### REFERENCES:

1. Karp, G. "Cell and Molecular Biology: Concepts and Experiments, fourth edition", Eds. Fitzgerald, P., Osnato, G., Rumiello, B. Wiley, John & Sons, Incorporated, 2005.
2. NCBI outreach and education tutorial - <http://www.ncbi.nlm.nih.gov/About/tools/index.html>.
3. Real-time PCR vs. traditional PCR, Applied Biosystems.com.
4. The Gene Chip System: An integrated solution for expression and DNA analysis, www.Affymetrix.com.
5. Mardis, E.R., 2008. Next-Generation DNA sequencing methods. Annu. Rev. Genomics Hum. Genet. 9:387-402.
6. Felske, A., and A.M. Osborn. "DNA fingerprinting of microbial communities." Molecular Microbial Ecology.
7. Eds. A.M. Osborn and C.J. Smith. Taylor & Francis Group, 2005. 65-96.
8. Waters MD, Olden K, Tennant RW. (2003). Toxicogenomic approach for assessing toxicant-related disease. Mutat Res. Nov;544(2-3):415-24.
9. van Herwaarden AE, Wagenaar E, Karnekamp B, Merino G, Jonker JW, Schinkel AH. (2006) Breast cancer resistance protein (Bcrp1/Abcg2) reduces systemic exposure of the dietary carcinogens aflatoxin B1, IQ and Trp-P-1 but also mediates their secretion into breast milk. Carcinogenesis. Jan;27(1):123-30.

## COURSE IV– AREA OF SPECIALIZATION

### 2-ENVIRONMENTAL MICROBIOLOGY

**UNIT I General Microbiology:** History and scope of microbiology, Ultra structure of Prokaryotic and Eukaryotic cell, Sterilization techniques used in microbiology, Preparation of media for isolation and culture of microorganisms - Microbial growth and multiplication of bacteria, virus and fungi - MPN, total cell count - Nature of virulence, toxins, extra cellular enzymes of pathogenic bacteria

**UNIT II Microbes and Environment:** Classification and characteristics of Microorganisms, Microbial interactions-Entophytes-. *Microbial diversity of environment:* Microbes in air, water, waste water and soil; Introduction, distribution, sampling techniques and identification. Microbes of extreme environment. Microorganisms as bio-indicators in the environment, Role of microorganisms in element cycles – different cycles. Ecological impacts of microbes - Symbiosis (Nitrogen fixation and ruminant symbiosis) - Microbes and Nutrient cycles - Microbial communication system - Quorum sensing – Microbial fuel cells - Prebiotics and Probiotics – Vaccines

**UNIT III Microbial Interactions:** Microbes in the degradation of wastes, Bioremediation-Its role in Environmental management, advantages and disadvantages. Control of pests and diseases by microorganisms, Treatment of solid and liquid industrial wastes, Microbial degradation of pesticides. Microbes in metal extraction, mineral leaching and mining, copper extraction by leaching and microbes in petroleum product formation.

**UNIT IV Aquatic Microbiology:** Methods of water sampling for pollution analysis - *Biofilms in treatment of waste water:* Biofilm development and biofilm kinetics, aerobic biofilms. *Bioreactors for waste water treatments:* Reactor types and design, Reactors in series - Different types of water sampling tools and it uses – Geomicrobial transformations – Biomobilization of silicon, phosphate, nitrogen – Geomicrobiology of fossil fuel, methane, peat, coal and petroleum

**UNIT V Microbial techniques:** Molecular methods such as Denaturing Gradient Gel Electrophoresis (DGGE) - Temperature Gradient Gel Electrophoresis (TGGE) - Amplified rDNA Restriction Analysis and Terminal Restriction Fragment Length Polymorphism (T-RFLP) in assessing microbial diversity - 16S rDNA sequencing and Ribosomal Database Project.

#### REFERENCES :

1. Patrick K. Jemba. Environmental Microbiology Principles and Applications.
2. P D Sharma- Environmental Microbiology.
3. Brock TD, Madigan MT, Martinko JM and Parker J (1994) Biology of Microorganisms, VII Ed., Prentice Hall, New Jercey, USA.
4. Shree N. Singh, Rudra D. Tripathi Environmental bioremediation technologies. Introduction to Microbiology. A. S. Rao
5. Maria Csuros, Csaba Csuros. Microbiological examination of water and wastewater,
6. Raina M. Maier, Ian L. Pepper, Charles P. Gerba - Science. Environmental Microbiology
7. Ehrlich HL (1996) Geomicrobiology, Marcel Dekker Inc., New York.
8. Eweis JB, Ergas SJ, Change DPY and Schroeder ED (1998) Bioremediation – Principles, McGraw-Hill Inc., New York. 9. Joseph C Daniel (1999) Environment Aspects of Microbiology. 1st Edition, Bright Sun Publications, Chennai.
9. Pelczar MJ Jr., Chan ECS and Kreig NR (1993) Microbiology, Tata McGraw Hill, Delhi.
10. Reed G, Prescott and Dunn's (1987) Industrial Microbiology, 4th Edition, CBS Publishers.
11. Subba Rao NS (2004) Soil Microbiology. 4th Edition, Oxford & IBH Publishing Co. Pvt. Ltd., New Delhi.

## COURSE IV– AREA OF SPECIALIZATION

### 3-BIODIVERSITY AND CONSERVATION

**UNIT I Concept and Principles:** Origin of biodiversity/evolution, Definition of biodiversity, Types of biodiversity, Levels of biodiversity, Genetic resources, Conservation of biodiversity, endangered species, impact of pollution on biodiversity, loss of biodiversity. **Conservation of Biodiversity:** Need for conservation of biodiversity, types of conservation, role of biotechnology in biodiversity conservation, in vitro conservation, application of in vitro conservation, limitation of in vitro conservation.

**UNIT II Bio-diversity and Germplasm:** Germplasm conservation, classical and new approaches to conservation, collection and exchange of germ plasm, cryo preservation, stability of conserved germplasm. Endangered species/exotic species, effect of pollutants on species loss, loss of genetic diversity. Preventing biodiversity loss.

**Management of Biodiversity:** Identifying land for Natural resources, managing wild life resources, Study of rare and endangered animals, Project tiger, Project crocodile, Green Sea and olive Reedley turtles, Biodiversity in a changing world, Wealth of Nature.

**UNIT III Tools to study bio-diversity: DNA extraction** from difficult species and preserved specimens; **Screening methods:** Isozymes. RFLP analysis, DNA finger printing, PCR, RAPDs, AFLP, **DATA analysis:** measure of polymorphisms within and among populations, distance data, discrete data and Case studies.

**UNIT IV: Cellular and molecular aspects of Biodiversity:** Diversity of photosynthetic prokaryotes. Diversity in rRNA sequence. Diversity of microbial heterotrophic metabolism. Genetic diversity: Polymorphism, repetitive DNA, Tandem repeats, Interspersed repeats, SNPs etc. Characterization of DNA polymorphism – Single locus approach, Multi locus approach.

PCR base and Non PCR based methods of monitoring Diversity.

**UNIT V Applications of biotechnologies for conservation:** Transgenic farm animals, cryopreservation Seed banks exsitu conservation for the majority of endangered species. Genetic fingerprints origin of a seed or the relatedness of one plant variety to another. Tissue culture techniques, Micropropagation, DNA barcoding.

**E-Waste:** Sources of generation, effects and control measures, global strategies Management: waste characteristics, generation, collection, transport and disposal

#### REFERENCE

1. Plant biotechnology and Biodiversity conservation by U. Kumar and A.K. Sharma published by *Agrobios*(India)
2. Conservation and biodiversity by Andrew Dobson, *palgravemacmillan*
3. Essentials of conservation Biology 3rd Edition by Riicherd B. Primack; Sinauer Associates, Inc. Publishers.W.K.
4. Life by Purves.w.k; Sadava D., Orians, G.H.and Heller, H.C. published by Sinauer Associates Inc; &W.K. Freeman and Company.
5. Molecular tools for Screening Biodiversity Ed. Angela Karp, Peter G. Isaac and David S. Ingram published by *Chapman & Hall*.
6. Hazardous waste management by Prof. Anjaneyulu
7. Standard hand book of hazardous waste treatment and disposal by Harry M.Freeman Mc Graw Hill-1997
8. Environmental Science-towards sustainable future by Richard t wright.PHI learning New Delhi, 2008.