

Annexure I

DEPARTMENT OF BIOTECHNOLOGY
(UGC-SAP and DST-FIST & PURSE Sponsored Department)
ALAGAPPA UNIVERSITY
(A State University Accredited with “A” Grade by NAAC)
Karaikudi - 630 003

Syllabus for Ph.D. Course Work
For those who joined on or after 03.11.2011

Course Structure

S. No	Code	Name of the Course	Credits	Marks		
				Int.	Ext.	Total
Paper – I						
1	12311	Paper I- Research Methodology	4	25	75	100
Paper – II (Area of Specialization)						
2	12312	Medical Microbiology	4	25	75	100
3	12313	Metagenomics	4	25	75	100
4	12314	Microbial Quorum Sensing and Biofilms	4	25	75	100
5	12315	Bioactive Agents from Natural Sources	4	25	75	100
6	12316	Biology of <i>Caenorhabditis elegans</i> as a model system	4	25	75	100
7	12317	Gene Regulation using <i>Caenorhabditis elegans</i>	4	25	75	100
8	12318	Host-Pathogen Interactions	4	25	75	100
9	12319	Plant Biodiversity Conservation				
10	12320	Plant Genetic Engineering	4	25	75	100
11	12321	Plant Tissue Culture	4	25	75	100
12	12322	Seed Technology	4	25	75	100
13	12323	Evaluation of the Efficacy of Natural Products Against Dioxin Toxicity	4	25	75	100
14	12324	Neurodegenerative Disorders and Natural Products	4	25	75	100
15	12325	Anti-Cancer Potential of Natural Products	4	25	75	100
Additional Courses Introduced for those joined on or after 01.07.2015						
16	12326	Antibiotics	4	25	75	100
17	12327	Biomedical Implants and Microbial Infections	4	25	75	100
18	12328	Proteomics	4	25	75	100
19	12329	Recombinant DNA Technology	4	25	75	100
20	12330	Enzyme Technology	4	25	75	100
21	12331	Microbial Biotechnology	4	25	75	100
22	12332	Natural Products Chemistry	4	25	75	100

12311 - Research Methodology (4 credits)

Unit I

Microscopy Techniques

Light Microscopy: Microscopic optics, components of microscope, Basic principles and methods of Bright-field, Dark-field, Phase contrast, interference contrast, Fluorescence, Confocal Microscopy, Transmission Electron Microscopy, Scanning Electron Microscope, Atomic Force Microscopy.

Spectroscopy Techniques

Introduction to Spectroscopic Methods- Infrared Spectrometry, Nuclear Magnetic Resonance Spectroscopy, Molecular Mass Spectroscopy.

Unit II

Chromatography

Principles and applications of gel filtration, ion exchange and affinity chromatography, thin layer and gas chromatography, high-pressure liquid chromatography (HPLC) - Nano LC.

Electrophoresis

PAGE, SDS – PAGE and Agarose gel electrophoresis. Isoelectric focusing (IEF), 2D Electrophoresis.

Unit III

Molecular Biology Techniques

Isolation and amplification of nucleic acid – Plasmid isolation, chromosomal DNA isolation, Quality and quantity checking of DNA by AGE, UV Spectrometry and Real Time PCR, Polymerase Chain Reaction (PCR)-Principles, Types and applications. Gene cloning techniques – Physical and chemical methods – Lipofection – Electroporation – Gene Bombardment. Construction of cDNA and genomic DNA library – Screening of libraries. Blotting Techniques (Southern, Northern and Western blot), Hybridization –plaque, colony hybridization

Molecular Tools for Analysis of Genome

Principles and applications of RFLP, RAPD, AFLP and DNA fingerprinting. Principle and applications of DNA sequencing- Automated DNA sequencing – Next Generation Sequencing.

Unit-IV

Quantitative Methods (Biostatistics)

Principles and practice of statistical methods in biological research, basic statistics, data collection, significance tests, Students t-test, Analysis of variance-ANNOVA, correlation regression, chi – square test, and Duncan’s multiple test. Identifying Groups- Factor analysis and cluster analysis (eg., SPSS)

Unit-V

Computer Application and Bioinformatics

Windows – features -MS Word - Word Basic. Introduction to Excel - Data sort, data filters, valid criteria, invalid criteria, redundant criteria, functions (MAX, MIN, SUM, AVG etc.). Working with Power Point - Parts of power point, various toolbars, slide manipulation, fonts, graphs, tables and template. Adobe - Photoshop and Reader.

Computer program: Quantitative and Qualitative analysis - Softmax pro, Biospec nano. DNA and RNA Gel analysis tools- Peak Scanner, GelcomparII and Quantity-I, Image analysis: COMSTAT, ImageJ and NIS-elements F. HPLC- Realtime analysis, FPLC-UNICORN 5.11, Real time PCR- AB7500, Metagenome Analyser: MEGAN.

Biological Databases, Sequence retrieval, Sequence analysis- pairwise sequence alignment- Global vs local alignment, BLAST, EXPASY Translate tool, Checking vector contamination- VecScreen, CAP3, Multiple sequence alignment: T-COFFEE, MUSCLE, Clustal W and Clustal X, Significance of multiple sequence alignment. EMBOSS, Phylogenetic analysis. Protein structure prediction-Analysis and Structure visualization software. Proteomics-MASCOT, SEQUEST, Tandem X!, MASS-LYNX and SWISS 2D PAGE.

References

1. Microbiology (2005), Sixth edition by L.M. Prescott, J.P. Harley and D.A. Klein, McGraw Hill, Boston.
2. Molecular Biology of the Gene, Fifth Edition (2004) by James D Watson, Tania A. Baker, Stephen P. Bell, Alexander Gann, Michael Levine and Richard Losick, Benjamin Cummings.
3. Genomes 3 (2007) by T. A. Brown, Garland Science Publishing.
4. Microbial Genetics (2006) by S.R. Maloy, J. E. Cronan Jr., and D. Freifelder, Jones and Bartlett Publishers, Sudbury, Massachusetts.
5. Genes V111 (2004), B. Lewin, Pearson Prentice Hall.
6. Bioanalytical Chemistry (2004) by Susan R. Mikkelsen and Eduardo Corton, Wiley-Interscience.
7. Laboratory Manual for Analytical Biochemistry and Separation techniques (2000) by P. Palanivelu, Madurai Kamaraj University.
8. Sambrook, J. and Russell, D.W. (2003). Molecular Cloning-A laboratory Manual (3 rd Edition, Vol.1, 2 and 3), Cold Spring Harbor Laboratory Press, Cold Spring Harbor, New York.
9. David Mount (2001) Bioinformatics. Sequence and Genome Analysis, Cold Spring Harbor Laboratory Press.
10. Rashidi, H.H. and Buehler, L.K. (2002). Bioinformatics Basis: Applications in Biological Science and Medicine. CRC Press, London.
11. Primrose et al. (2005) Principles of gene manipulation, Black Well Science, London.

12312 - Medical Microbiology (4 credits)

Unit - I

Microorganisms - Introduction; Morphology and Physiology of Bacteria and Fungi. Sterilisation - Dry heat sterilisation; Moist heat sterilisation. Culture media - Basal media; Enriched media; Differential media; Selective media; Enrichment media; Transport media. Culture methods - Aerobic and anaerobic culture methods.

Unit - II

Bacteria: Classification based on morphology - Cocci and Rods; Classification based on physiological characters - Motility, Flagellar pattern, Spore formation, Nutritional requirements, Respiration; Gram's classification - Gram positive bacteria, Gram negative bacteria. Identification of Bacteria - Staining, motility, Culture methods, Biochemical methods - Oxidase, Catalase, IMViC tests, Carbohydrate fermentation tests, TSI test.

Unit - III

Virulence traits, Pathogenesis and clinical manifestations of Staphylococcus, Streptococcus, Corynebacterium, Bacillus, Propionibacterium, Escherichia, Pseudomonas, Proteus, Vibrio, Serratia.

Unit - IV

Medical Mycology: Classification of Fungi - Yeasts, Yeast like fungi, Moulds, Mushrooms. Morphology and Identification of Fungi. Pathogenesis and clinical manifestations of *Candida albicans*, *Cryptococcus neoformans*, Trichophyton, Microsporum, Epidermophyton, Malassezia.

Unit - V

Medically important Parasites: Classification - Protozoans, Helminths. Clinical manifestations of Amoebiasis, Giardiasis, Leishmaniasis, Malaria, Filariasis, Chagas disease, Sleeping sickness, Parasitic pneumonia (Round worm infection), Taeniasis (Tapeworm infection), Trypanosomiasis. Viruses: Classification - DNA viruses, RNA viruses, Reverse transcribing viruses. Structure of viruses. Pathogenesis and clinical manifestations of human viral infections - Common cold, Influenza, Rabies, Hepatitis, Dengue fever, Yellow fever, Polio, Small pox, AIDS.

References

1. Prescott, Harley and Klein's Microbiology (2008) 7th Ed, Willey JM, Sherwood L, Woolverton CJ. McGraw-Hill Higher Education.
2. Textbook of Microbiology (2006) 7th Ed, Ananthanarayan and paniker. Orient Blackswan.
3. Microbiology. Pelczar MJ, Chan ECS, Krieg NR. Tata McGraw-Hill Edition.

4. Medical Microbiology (1997) by D. Greenwood, R. Slack and J. Peutherer, ELST with Churchill Livingstone, Hong Kong.

12313 - Metagenomics (4 Credits)

Unit I

Introduction to Metagenomics, Applications of Metagenomics in Red Biotechnology and White Biotechnology

Unit II

Function driven analysis of metagenomics: Metagenomic library construction, High throughput Intracellular Screening, Identification of active clones against Quorum sensing signals and Heterologous Expression of active clones.

Unit III

Sequence Driven Analysis- Identification of novel and metabolic pathway specific genes, Whole genome metagenomic Sequencing using next generation sequencing techniques- 454 Pyrosequencing, Illumina Sequencing SOLiD sequencing, Primer Walking in Fosmid and Cosmid clones

Unit IV

Culture independent molecular analysis of environmental samples : T-RFLP, DGGE, TGGE, SSCP, 16S rRNA gene sequencing using Next Generation Sequencing.

Unit V

Identification and screening of Industrial important enzymes from environmental samples using metagenomic approaches- Amylase, Lipase, Protease, Esterase and Cellulase.

References

1. Muyzer, G. DGGE/TGGE A Method For Identifying Genes From Natural Ecosystems. *Current Opinion in Microbiology* **2**, 317-322 (1999).
2. LIU, W. *et al.* Characterization of Microbial Diversity by Determining Terminal Restriction Fragment Length Polymorphisms of Genes Encoding 16S rRNA. *Appl. Environ. Microbiol.* **63**, 4516–4522 (1997).
3. Williamson, L.L. *etal.* Intracellular Screen To Identify Metagenomic Clones That Induce or Inhibit a Quorum-Sensing Biosensor. *Appl. Environ. Microbiol.* **71**(10): 6335 (2005).
4. Jo Handelsman Metagenomics: Application of Genomics to Uncultured Microorganisms *Microbiology And Molecular Biology Review* **68** (4) 685-669 2004
5. Lorenz P, Eck J. Metagenomics and industrial applications. *Nat Rev Microbiol.* 2005 Jun;**3** (6):510-516.

12314 - Microbial Quorum Sensing and Biofilms (4 Credits)

Unit I

Bacterial communication - Quorum sensing (QS); signalling molecules - HSLs, AI-2, Pheromones, AIP. Role of QS in promoting microbial virulence and resistance to drugs.

Unit II

QS in Gram positive bacteria - Staphylococcus, Streptococcus, Bacillus; QS in Gram negative bacteria - Chromobacterium, Agrobacterium, Pseudomonas, Serratia, Vibrio; Virulence traits regulated by QS in bacteria.

Unit III

Biofilm: Definition, Composition, Structural organization and mechanism of biofilm formation. Importance of biofilm on environmental, industrial and medical perspectives. Resistance development in biofilm inhabitants. Role of quorum sensing in biofilm formation. Characteristics of biofilm in clinical devices. The role of biofilm in the dissemination of bacterial virulence. Impact of biofilm in aquaculture. Industrial importance of biofilm prevention. Consequences of biofilm in environment – biofouling and biocorrosion.

Unit IV

Biosensor strains used for QS based study - *Chromobacterium violaceum*, *Agrobacterium tumefaciens*, *Pseudomonas aeruginosa* and its mutants. Assays to measure the QS regulated virulence traits - Violacein production, LasA protease, LasB elastase, Pyocyanin, Pyoverdine, Prodigiosin, Serratia total protease, Lipase, Bioluminescence, Swarming and Swimming motility.

Unit V

In vitro biofilm formation; Quantification of *in vitro* and *in vivo* formed biofilms; Microscopical analysis of biofilm architecture - EPS, proteins, e-DNA; Fluorophores used for staining biofilms; Live-dead staining; Confocal microscopy for biofilm studies - Z-stack analysis, Optical sectioning.

References

1. Kievit, TR & Iglewski BH. Bacterial Quorum Sensing in Pathogenic Relationships. *Infect. Immun.* 68(9): 4839 (2000).
2. Stoodley, LH et al. Bacterial Biofilms: From the Natural Environment to Infectious Diseases. *Microbiology*.2 (2004).
3. Choi et al., Implication of Quorum Sensing in *Salmonella enteric* Serovar Typhimurium Virulence: the *luxS* Gene Is Necessary for Expression of Genes in Pathogenicity Island. 75, p. 4885–4890 (2007).
4. Rasmussen TB, Givskov M, Quorum-sensing inhibitors as anti-pathogenic drugs, *International Journal of Medical Microbiology*, 296, Pages 149-161, 2006.
5. Di Cagno R, De Angelis M, Calasso M, Gobbetti M. Proteomics of the bacterial cross-talk by quorum sensing. *J Proteomics*. 74(1):19-34. 2011.

12315 - Bioactive Agents from Natural Sources (4 Credits)

Unit I

Bioactive agents - Anti-bacterial, Anti-fungal, Antibiofilm, Anti-Quorum Sensing agents. Need for novel bioactive agents - Resistance development, Emergence of new pathogens. Bioactive agents from Natural resources - Bacteria and fungi associated with Soil, Marine sediment, Sea water, Sea weeds, Sea grass, Sponges, mangrove sediments, Plant natural products, Mushrooms. Merits of natural products.

Unit II

Antibacterial agents - Source, Types, Mode of action. Anti-fungal agents - Source, Types, Mode of action. Advantages and disadvantages.

Unit III

Strategies for quorum sensing inhibition - Inhibition of signal production, Degrading the signals, Blocking signal-receptor interactions. Quorum sensing inhibitors - Source, types - Enzyme inhibitors, Structural analogues, enzymes - lactonase, acylase. Assays to study Quorum sensing inhibition. Merits and demerits.

Unit IV

Antibiofilm agents - Source, mode of action - inhibition of biofilm formation, disruption of biofilms. Quorum sensing inhibitors with antibiofilm potential. Assays to study antibiofilm activity. Merits and demerits.

Unit V

Plant natural products - Medicinal plants, herbs, spices, fruits, vegetables. Phenolic compounds - types, identification. Ethnobotanical importance of plant natural products. Mushrooms - Nutritional value and industrial perspectives, medical importance.

References

1. Lindequist et al., The Pharmacological Potential of Mushrooms. eCAM; 2: 285–299 (2005).
2. Rasmussen TB, Givskov M, Quorum-sensing inhibitors as anti-pathogenic drugs, International Journal of Medical Microbiology, 296, Pages 149-161, 2006.
3. Hentzer M, Riedel K, Rasmussen TB, Heydorn A, Andersen JB, Parsek MR, Rice SA, Eberl L, Molin S, Hoiby N, Kjelleberg S, Givskov M. (2002) Inhibition of quorum sensing in *Pseudomonas aeruginosa* biofilm bacteria by a halogenated furanone compound. Microbiology 148: 87–102.
4. Kumar RJ, Zi-rong X (2004) Biomedical compounds from marine organisms. Marine Drugs 2:123-146.
5. Adonizio A, Kong KF, Mathee K 2008 Inhibition of quorum sensing-controlled virulence factor production in *Pseudomonas aeruginosa* by South Florida plant extracts. Antimicrob. Agents. Chemother. 52 198-203

12316 - Biology of *Caenorhabditis elegans* as a Model System (4 Credits)

Unit I

Introduction to *C. elegans* Biology. Evolution and ecology of *Caenorhabditis* species, Taxonomic overview and developmental biology of *C.elegans*, Metabolic pathways involved in fat storage.

Unit II

General biology and Maintenance of *Caenorhabditis elegans*. *C. elegans* as a model organism - Natural History, Life Cycle, Maintenance and Synchronization of *Caenorhabditis elegans*. Dauer development, Preservation of *C. elegans* Tissue Via High-Pressure Freezing and Freeze-Substitution for Ultrastructural Analysis and Immunocytochemistry - Techniques.

Unit III

Functional Genomic Approaches in *C. elegans*. Creation of Transgenic and Mutant Lines. Single-worm PCR. Assays for Toxicity. Drug discovery studies: Screening and targeting in *C. elegans*. Whole genome microarray analysis and genome-wide RNA interference (RNAi)-mediated phenotypic screening in *C. elegans*.

Unit IV

RNAi in *Caenorhabditis elegans*, Concepts and Strategies, silencing of gene expression. Construction of Plasmids for RNA Interference and *In vitro* transcription of Double-Stranded RNA. Methods for delivery and detecting gene silencing.

Unit V

WormBase: Methods for Data Mining and Comparative Genomics. *Caenorhabditis elegans* Gene Knockout Consortium

References

1. The Nematode *Caenorhabditis elegans* (1988) by Willam B.Wood Cold spring harbor monograph series, Cold spring Harbor Press.
2. *C. elegans*: Methods and Applications-Functional Genomic Approaches in *C. elegans*(2006) by Todd Lamitina , Humana Press.
3. Developmental Biology (9th edition) by Scott F. Gilbert, Sinauer Associates.
4. Obesity and the regulation of fat metabolism (2007), by Ashrafi, K, WormBook, ed.
5. RNAi A Guide to Gene Silencing (2003), by Gregory J. Hannon, Cold Spring Harbor Laboratory Press.
6. *C.elegans* : Methods and Applications- Preservation of *C. elegans* Tissue Via High-Pressure Freezing and Freeze-Substitution for Ultrastructural Analysis and Immunocytochemistry(2006) by Robby M.Weimer, Humana Press.

12317 - Gene Regulation using *Caenorhabditis elegans* (4 Credits)

Unit I

Genetic organization and regulation in eukaryotic system: Cell cycle: Mitosis and Meiosis. Genetics, physical mapping, Genome sequencing, Transposons; Aging: basic concepts and genes and pathway involved in *C. elegans*'s aging processes.

Unit II

Gene expression and regulation studies using semi-quantitative and qPCR analysis. Role of Innate immune signaling pathways in *C. elegans*.

Unit III

Translational regulations in *C. elegans* during adverse conditions (biotic and abiotic stresses). Methods and approaches in proteomic studies: SDS-PAGE, 1D & 2D electrophoresis, DIGE, MALDI and Mascot analysis.

Unit IV

Basic Cell Culture Techniques-Culture of Primary adherent and non adherent cells, Types of cell culture media, detection and eradication of Mycoplasma contamination. Gene regulation studies in cell lines.

Unit V

Cloning and characterization of regulatory genes in model cellular systems, specific vectors for cloning- bacterial and mammalian vectors, detection of recombinant molecules and over expression studies.

References

1. WormBook: The Online Review of *C. elegans* Biology (wormbase.org)
2. Microbiology (2005), Sixth edition by L.M. Prescott, J.P. Harley and D.A. Klein, McGrawHill, Boston.
3. Gene Cloning and DNA Analysis. An Introduction (2006) by T. A. Brown, Blackwell Scientific Publications.
4. Basic Cell Culture (2005). Edited by J.M.Davis. Oxford University Press
5. Molecular cloning: A Laboratory Manual (2001). Sambrook, J., Russell, D.W., Cold Spring Harbor Laboratory Press, Cold Spring Harbor, New York.
6. Gene expression changes associated with aging in *C. elegans* (2007) by Golden, T.R. and Melov, S. Wormbook.

12318 - Host-Pathogen Interactions (4 Credits)

Unit I

Normal microbiota, General introduction about human pathogens- bacterial, viral and fungal pathogens and its virulence factors, pathogenicity islands

Unit II

Recognition and entry processes of different pathogens, Evasion of normal host defenses against the infection, colonization, multiplication, toxin production, biofilm formation and antibiotic resistance.

Unit III

Host-pathogen interaction using *C. elegans* as a host – pathogen and its virulent factors, pathogenicity islands, Types of Secretion system, Antibiotic resistant mechanism, Biofilm formation and Molecular pathogenesis. Host response during the infection. Activation of immune system against the pathogen, primary and secondary line of defenses- antibody, neutrophils and AMPs productions.

Unit IV

Regulation of immune effectors and genes during the infection. Phenotypic alteration of host by pathogens.

Unit V

Types of infectious disease- bacterial, viral, fungal and protozoal diseases

References

1. Microbiology (2005), Sixth edition by L.M. Prescott, J.P. Harley and D.A. Klein, McGrawHill, Boston.
2. Medical Microbiology (1997) by D. Greenwood, R. Slack and J. Peutherer, ELST with Churchill Livingstone, Hong Kong.
3. Kuby Immunology (2007) by Kindt, Goldsby and Osborne. W.H.Freeman and Company
4. Immunology of Infectious Diseases (2002) Edited by Kaufmann, Sher and Ahmed. ASM Press.

12319 - Plant Biodiversity Conservation (4 Credits)

Unit I

Introduction to biodiversity, Types of biodiversity, Biodiversity concepts, Centres of diversity, Agro biodiversity, Loss of biodiversity, Biodiversity act of India 2002 and 2004.

Unit II

Phytogeographic zones, Vegetation types of India and Tamilnadu, Endemism, Wildlife Sanctuaries, National parks and Biosphere Reserves, Hotspot biodiversity areas in India, Red listed plants, Red Data Book.

Unit III

Threatened plants of India, Patenting life forms and their impact on biodiversity. Values and uses of biodiversity. The importance of long-distance dispersal in biodiversity conservation.

Unit IV

General overview of plant conservation, Sustainable use of plant genetic resources, Biotechnology assisted plant conservation (*in situ* and *ex situ* conservation).

Unit V

Conservation of biodiversity – National and international status. Emerging issues for biodiversity conservation. Conservation of biodiversity in a changing climate. Cryopreservation. Static conservation of seeds. Seed certification; seed banks. Synthetic seed preparation and plant conversion.

References

1. Biodiversity and Conservation (2004). Joshi P.C. and Namita Joshi, APH publishing company, New Delhi.
2. An advanced text book of Biodiversity (2004). K.V.Krishnamoorthy, Oxford &IBH, New Delhi.
3. Plant Conservation Biotechnology (2003). Edited by Erica E.Bensen, Taylor &Francis Ltd, London.
4. Recombinant DNA safety guidelines (January1990), Department of Biotechnology, Ministry of Science & Technology, Government of India, New Delhi.
5. Patents (2003), N.Subbaram, Pharma Book Syndicate, Hyderabad.
6. Molecular Biotechnology (1998), Second Edition, Glick, B.R., and Pasternack, J.J., ASM Press, Washington, DC.

12320 - Plant Genetic Engineering (4 Credits)

Unit I

Protoplast culture: Isolation of protoplasts – mechanical and enzymatic sources, Culture of protoplasts and protoplast fusion. Synthetic seed preparation, culture initiation, plant conversion and hardening.

Unit II

Agrobacterium and crown gall tumors. Mechanism of T-DNA transfer to plants. Co-integrate, binary and super binary Ti-plasmid based vectors for plant transformation. Introduction of binary plasmids into Agrobacterium cells by Triparental mating.

Unit III

Plant transformation technology- Agrobacterium and Biolistic mediated transformation. Transient β -glucuronidase (GUS) gene expression assays in transformed intact explants. Genetic engineering of plants for insect resistance, virus resistance, fungal resistance and herbicide tolerance.

Unit IV

Regulation of proline biosynthesis, degradation, transport and its implications in abiotic stress tolerance. Genetic engineering for abiotic stress tolerance in plants. Reporter genes used in plant gene expression vectors.

Unit V

Gene silencing in transgenic plants, Commercially important transgenic plants (Flavr Savr, Bt cotton, Golden rice), rDNA regulations in transgenic plants.

References

1. Introduction to Plant Biotechnology (2001), H.S.Chawla, Oxford & IBH Publishing Co. Pvt. Ltd.
2. Plant Tissue Culture: Theory and Practice, Revised Edition - 2004 (Studies in Plant Science), S.S.Bhojwani and M.K.Razdan, Elsevier Publications, Netherlands.
3. Plant Biotechnology: The Genetic Manipulation of Plants (2003) Adrian Slater, Nigel W.Scott and Mark R.Fowler, Oxford University Press.
4. Methods in Plant Molecular Biology (2005), Mary A.Schuler, Raymond E.Zielinski, Academic Press, An Imprint of Elsevier.
5. Plant Genetic Transformation Technology (1997). Wagdy A.Sawahel, Daya Publishing House, New Delhi.

12321 - Plant Tissue Culture (4 Credits)

Unit I

Preparation of stock solutions and nutrient media for callus culture initiation and plant regeneration. Processing of various explants (mature seed, leaf base, node) for culture initiation. Aseptic techniques-Sterilization of nutrient media and surface sterilization of explants collected from field for aseptic culture initiation.

Unit II

Callus initiation and maintenance from various explants of food crops and medicinal plants. Regeneration of shoots and roots from callus cultures. RAPD analysis of medicinal plants.

Unit III

Plant transformation technology - Direct and Indirect methods. Direct transformation of plants by physical methods (Biolistic, Microlaser, Ultrasonication and Silicon carbide WHISKER™ method). Transposon Tagging.

Unit IV

Genomic DNA extraction and purification – Principle and methods. Isolation and purification of Ti-plasmid DNA. Introduction of binary plasmids into Agrobacterium cells by Triparental mating. Restriction digestion and elution of genes from agarose gel for cloning.

Unit V

Cryopreservation and its applications in germplasm maintenance. Molecular Farming and case studies – Polyhydroxy butyrate (PHB), Polyfructons and Cyclodextrans.

References:

1. Plant Tissue Culture: Theory and Practice, Revised Edition - 2004 (Studies in Plant Science), S.S. Bhojwani and M.K. Razdan, Elsevier Publications, Netherlands.
2. Methods in Plant Molecular Biology (2005), Mary A.Schuler, Raymond E.Zielinski, Academic Press, An Imprint of Elsevier.
3. Introduction to Plant Biotechnology (2001), H.S.Chawla, Oxford & IBH Publishing Co. Pvt. Ltd.
4. Plant Biotechnology-New Products & Applications (2000). J. Hammond, P.McGarvey &V.Yusibov (Eds), Springer-Verlog.
5. Plant Biotechnology: The Genetic Manipulation of Plants (2003) Adrian Slater, Nigel W.Scott and Mark R.Fowler, Oxford University Press.

12322 - Seed Technology (4 Credits)

Unit I

Dicot and Monocot Seeds. Morphology and Types. Seed Reserves. Internal and External structures:- their functional significance. Albuminous and ex-albuminous seeds. Large and small seeds.

Unit II

Seed Dormancy: Types of dormancy - physical, physiological, morphological, chemical and mechanical. Primary and secondary dormancy. Skoto and photo dormancy. Methods to overcome dormancy. Ecological significance of seed dormancy.

Unit III

Seed Germination: Epigeal and hypogeal germination. Germination mechanism. Brief account of Germination value (Czabator) , Germination rate (Bartellete). Total germination percent and plant percent. Germination ecology: Environmental factors and germination behaviour. Seed storage proteins.

Unit IV

Seed Viability: Viability tests-their significance and importance. Orthodox and recalcitrant seeds. Critical role of seed moisture content and environmental factors on viability. Viability periods of selected Indian Forestry species.

Unit V

Seed Storage and Longevity: Seed germplasm and storage in different conditions. Cryopreservation. Static conservation of seeds. Clonal seed orchards. Seed certification; seed banks. Synthetic seed preparation and plant conversion.

REFERENCES

1. Introduction to Plant Biotechnology (2001), H.S.Chawla, Oxford & IBH Publishing Co. Pvt. Ltd.
2. Plant Biotechnology: The Genetic Manipulation of Plants (2003) Adrian Slater, Nigel W.Scott and Mark R.Fowler, Oxford University Press.
3. Biotechnology in Agriculture and Forestry - Somatic Embryogenesis and Synthetic Seed II (1993), edited by YPS Bajaj, Springer, Germany
4. Synthetic Seed: A Challenging Technology in Plant Propagation, Transportation and Conservation (2010), Bidhan Roy, Lambert Academic Publishing.
5. Tissue Culture Studies On Mulberry: Micropropagation, Gynogenic haploids, Synthetic seeds (2010), Kavyashree Rangappa, Lambert Academic Publishing.
6. Synthetic Seed For Commercial Crop Production (2011), U. Kumar, 2nd ED. Agrobios publishing.

12323 - Evaluation of the Efficacy of Natural Products against Dioxin Toxicity (4 Credits)

Unit-1

Environmental toxin- sources, classification and toxicological effects in human beings. Mechanism of toxicity induced by dioxin like compounds. Oxidative stress induced by environmental toxins and their pathological implications. Role of antioxidant enzymes and antioxidants from natural sources like polyphenols, carotenoids, flavonoids and vitamins in preventing oxidative stress induced by dioxin.

Unit-2

Natural products - Collection, processing and solvent extraction – Evaluation of phytoconstituents responsible for pharmacological properties (Terpenoids, alkaloids, flavonoids, tannins, cardiac glycosides) - Nutritional evaluation- Safety evaluation (Comet assay, Ames test, Cytotoxicity).

Unit-3

Protective effect of natural products against dioxin mediated toxicity in animal cells. Maintenance, preservation and storage of cell lines- Assessment of cytotoxicity (MTT assay)- Evaluation of antioxidant status (SOD, CAT, GPx, GR, GSSG, GST, intracellular ROS)- Assessment of macromolecular damage (LPO, DNA damage, Protein carbonyl content)- Evaluation of intracellular calcium level (AAS), cell membrane integrity (Light microscopy, Histopathology)- Expression studies of CYP1A1 and CYP1B1 and apoptotic genes (RT-PCR , Western Blot).

Unit-4

Immunoprotective effect of natural products against dioxin induced toxicity in PBMC:-Isolation of PBMC-cytotoxicity studies (MTT assay, LDH assay)-Cell cycle analysis (Flow cytometry). Apoptosis detection (DAPI, EtBr/AO) - Assessment of macromolecular damage (FTIR) - cell morphology changes (SEM, TEM). Protective potential of natural products against dioxin mediated toxicity in animals:-Determination of blood parameters and serum marker enzymes-Assessment of Phase I & II detoxification enzyme system (CYP 1A1, GST, NQO, SULT, EH, HO) - Estimation of membrane bound ATPase - Immunohistochemistry (CYP1A1, CYP1B1) – Assessment of Histopathological changes- Tissue storage, section processing, staining and interpretation (for liver, kidney, spleen and lungs)

Unit-5

Purification of plant compounds by chromatographic techniques (TLC, column chromatography, HPLC and GC-MS) and Spectroscopy (UV/Visible, NMR and IR)

References

1. Phytochemical methods – A guide to modern techniques of plant analysis, Third edition – L.B. Harborne. Springer Publishers (1998).
2. Plant drug Analysis, Second Edition, H.Wagner and S.Baldt. Springer Publishers (1996)
3. Basic Cell Culture- J.M.Davis. Oxford University Press (2005).

12324 - Neurodegenerative Disorders and Natural Products (4 Credits)

Unit I

Neurodegenerative disorders – Types – Causes – Pathways involved – Therapeutic strategies employed - Antioxidants, Cholinesterase inhibition, β -secretase inhibition, Monoamino oxidase inhibition, Inhibition of apoptosis, Metal chelation, anti-aggregation of toxic peptides.

Unit II

Marine natural products - Collection, processing and solvent extraction - Phytoconstituents responsible for pharmacological properties (Terpenoids, alkaloids, flavonoids, tannins, cardiac glycosides) – Nutritional evaluation (Proximate composition, Physico-chemical characteristics, mineral content, amino acids composition, fatty acid profile, vitamin analysis) – Safety evaluation (Ames test, Comet assay, MTT assay) - Therapeutic potentials of Mangroves and seaweeds (anti-cancer, neuroprotective, anti-inflammatory).

Unit III

Screening of marine natural products for neuroprotective agents in *in vitro* cell free systems - antioxidant activity (Free radical scavenging assay, Hydrogen peroxide scavenging assay, Hydroxyl radical scavenging assay, Nitric oxide scavenging assay, Reducing power, FRAP assays), Cholinesterase inhibitory activity (Acetylcholinesterase and Butyrylcholinesterase inhibitory assay), Metal chelating activity, β -secretase inhibitory activity, Aggregation and de-aggregation of abnormal proteins (Thioflavin T assay, TEM).

Unit IV

Purification and characterization of compounds from marine natural products by chromatographic techniques (TLC, column chromatography and HPLC) and Spectroscopy techniques (UV/Visible, NMR, GC-MS and IR).

Unit V

Neuroprotective effect of the purified compound using neuronal cells – Assessment of cell viability (MTT assay, trypan blue exclusion assay) — Evaluation of antioxidant status (SOD, Catalase, GPx, GR, GSH, GST, GSSG, Protein carbonyl content, NO production, LPO, Intracellular ROS determination) – Cholinesterase inhibitory activity - Caspase-3 activity - Assessment of intracellular calcium levels (AAS) and cell membrane integrity (Light microscopy and Histopathology) – Apoptotic morphological observation (DAPI, EtBr/AO) - Expression of apoptotic proteins (Real-Time PCR, Western blotting, Confocal microscopy)

References:

1. Phytochemical methods – A guide to modern techniques of plant analysis, Third edition – L.B. Harborne. Springer Publishers (1998).

2. Practical Biochemistry – Principles and techniques, Fifth edition – Keith Wilson and John Walker. Cambridge University Press (2000).
3. Molecular Cloning: A Laboratory Manual, Third Edition – Joseph Sambrook, David W. Russell. Cold Spring Harbor Laboratory Press (2001).
4. Kuby Immunology, Fourth edition - Kindt, Goldsby and Osborne. W.H.Freeman and Company (2007).
5. Basic Cell Culture- J.M.Davis. Oxford University Press (2005).
6. Principles of Gene Manipulation, Fifth edition - R.W. Old and S.B. Primrose. Blackwell Science Ltd (1994).

12325 - Anti-cancer Potential of Natural Products (4 Credits)

Unit I

Cancer – Classification – Symptoms - Causes – Angiogenesis – Metastasis – Involvement of signaling pathways (MAPK, ERK, Ras, Raf, JNK, AKT, PI3) – Involvement of apoptotic pathways (Intrinsic and extrinsic pathways) – Chemoprevention

Unit II

Chemical Carcinogens – Alkylating agents, Polycyclic Aromatic Hydrocarbons, Aromatic amines and Azo Dyes, Natural carcinogens, Nitrosamines - Stages of chemical carcinogenesis – Genotoxic and Non-genotoxic effects of carcinogens - Molecular targets of chemical carcinogens.

Unit III

Natural products - Collection, processing and solvent extraction- Phytoconstituents responsible for pharmacological properties (Terpenoids, alkaloids, flavonoids, tannins, cardiac glycosides) – Nutritional evaluation (Proximate composition, Physico-chemical characteristics, mineral content, amino acids composition, fatty acid profile, vitamin analysis) – Safety evaluation (Ames test, Comet assay, MTT assay) - Pharmacological properties of natural products – Antioxidants, Anti-inflammatory agents, Modulators of altered signaling pathways and apoptotic pathways.

Unit IV

Screening of natural products for antioxidant potentials - *In vitro* antioxidant activity (Free radical scavenging assay, Hydrogen peroxide scavenging assay, Hydroxyl radical scavenging assay, Nitric oxide scavenging assay, Reducing power, FRAP assays). Anticancer effect of the compounds using cancer cell lines – Assessment of cytotoxicity (MTT assay, trypan blue exclusion assay). Alteration in the expression of apoptotic proteins (Real-Time PCR, Western blotting, Confocal microscopy).

Unit V

Purification and characterization of compounds from natural products by chromatographic techniques (TLC, column chromatography and HPLC) and Spectroscopy techniques (UV/Visible, NMR, GC-MS and IR)

References:

1. Molecular Cell Biology, Fifth edition – Harvey Lodish, Arnold Berk, Paul Matsudaira, Chris A. Kaiser, Monty Krieger, Matthew P. Scott, S. Lawrence Zipursky, James Darnell. W.H. Freeman and Company (2004).
2. Animal Cell culture methods – Jennie P. Mather, David Barner. Academic Press (2006).
3. A handbook of Medicinal plants – A complete source book – Narayan Das Prajapati, S.S.Purohit, Arun K. Sharma, Tarun Kumar. Agrobios (2006).
4. Phytochemical methods – A guide to modern techniques of plant analysis, Third edition – L.B. Harborne. Springer Publishers (1998).

5. Practical Biochemistry – Principles and techniques, Fifth edition – Keith Wilson and John Walker. Cambridge University Press (2000).

12326 - Antibiotics (4 Credits)

Unit I

Discovery of antibiotics and its importance in therapeutical applications Types of antibiotics- antibacterial, antifungal, antiviral and antiparasitic agents - Evolution of antibiotics: first, second, third, fourth and fifth generation antibiotics – its drawbacks and consequences.

Unit II

Natural sources of antibiotics: microorganisms (bacterial, fungal, actinomycetes) and plants – Primary and secondary metabolism in relation to antibiotic production. Methods to screen antibiotic producing organisms – Isolation of antibiotics from microorganisms – Extraction of antibiotics from plants – Large scale production - fermentation, design of fermenter, control of different parameters, downstream processing and recovery of antibiotics - Semi-synthetic antibiotics - Production of antibiotics using enzymatic methods - Synthetic antibiotics – Chemical synthesis.

Unit III

Classification of antibiotics based on Chemical Structure: BetaLactams, Macrolides, Quinolones, Tetracyclines, Aminoglycosides - Bacterial Spectrum: broad and narrow - Type of Activity: bacteriocidal and bacteriostatic - Mechanism of action: Antibiotics that interfere with the biosynthesis of bacterial cell wall, protein biosynthesis in microorganisms, DNA and RNA biosynthesis, translational fidelity- Ribosome binding antibiotics.

Unit IV

Antibiotic resistance – Case studies: 1. New Delhi metallo-beta-lactamase (NDM-1) 2. MDR (Multi Drug Resistane) Tuberculosis. Molecular mechanisms underlying resistance: modification of the targets, membrane impermeability, inactivating enzymes, horizontal gene transfer, point mutations and efflux pumps - Superbugs: Innate and Acquired Resistance – Prevalence of antibiotic resistance in the environment.

Unit V

New methods for antibiotic discovery and delivery: Antisense antibiotics, Antisense oligonucleotides – Non-traditional methods to treat bacterial infection: Microbiome replacement therapy, Fecal bacteriotherapy, Phage therapy- Alternative to antibiotics: Antibiofilm and Anti-Quorum Sensing agents.

References

1. Betina, V. (1983). chemistry and biology of antibiotics. *Elsevier Scientific*.
2. Gottlieb, D., & Shaw, P. D. (2013). Antibiotics: Volume I Mechanism of Action. *Springer*.
3. Korzybski, T., Kowszyk-Gindifer, Z., & Kurylowicz, W. (2013). Antibiotics: origin, nature and properties (Vol. 1). *Elsevier*.

4. Bruggink, A. (Ed.). (2011). Synthesis of β -lactam antibiotics: chemistry, biocatalysis & process integration. *Springer Science & Business Media*.
5. Boman, H. G. (1995). Peptide antibiotics and their role in innate immunity. *Annual review of immunology*, 13(1), 61-92.
6. Chamany, K. (2006). Science and Social Justice. *Journal of College Science Teaching*, 36(2), 54.
7. Czekalski, N., T. Berthold, et al. "Increased Levels of Multi-resistant Bacteria and Resistance Genes after Wastewater Treatment and their Dissemination into Lake Geneva, Switzerland." *Frontiers in Microbiology* 3, no. 106 (2012).
8. Johnson, A. P., & Woodford, N. (2013). Global spread of antibiotic resistance: the example of New Delhi metallo- β -lactamase (NDM)-mediated carbapenem resistance. *Journal of medical microbiology*, 62(Pt 4), 499-513.
9. Ling, L. L., T. Schneider, et al. "A New Antibiotic Kills Pathogens without Detectable Resistance." *Nature* 517 (2015): 455–9.

12327 - Biomedical Implants and Microbial Infections (4 Credits)

Unit - I

Biomedical implants - Importance and its types - Cardiac implants - Ophthalmic implants - Orthopedic implants - Dental implants – Cochlear implants - Indwelling catheters - Foley catheter - Peripheral venous catheter (PVC).

Unit - II

Biomaterials - Surface properties of biomaterials - Synthetic, Metals and non-metallic alloys, Ceramics, Inorganics and polymers – Biopolymers - PEG, PVA, PMMA, PAH, Hydrogels and Methacrylates - Drug incorporation into polymer gels, Biocompatibility of polymers.

Unit - III

Methods for testing and evaluating biocompatibility: In Vitro Testing, In Vivo Testing; Hemocompatibility, Osteocompatibility, Odontocompatibility, Cytotoxicity Testing, Hypersensitivity/Allergic Responses, Genotoxicity, Tissue reaction to external materials, Blood/biomaterial interaction, Corrosion and wear of biomaterials, Treatment of materials for biocompatibility, Biodegradable materials and their applications, Rheological properties of biological solids- bone, tendons, blood vessels, biological liquids, mucus.

Unit - IV

Infections associated with Biomedical Implants – Septic arthritis, Prosthetic joint infections Peri-implant diseases/Peri-implantitis - Catheter-related urinary tract infection (UTI) - Formation of polymicrobial Biofilm on medical implants - Impact of biofilm on implants and Consequences – Common pathogens found in biofilm of implants.

Unit - V

Protective Surface coatings with anti-adhesive, anti-microbial, anti-biofilm, quorum quenching agents & Methods of coating - Characterization of coatings and surfaces - Measurement of coatings thickness, stability & adhesion of surface coatings - Surface microscopy & topography by scanning probe microscopy

References:

1. Biomaterials, Sujata V. Bhatt, Narosa Publishing House, 2002.
2. Biomaterials science and engineering, J. B. Park, Plenum press, 1984.
3. Biomaterials Science- An introduction to materials in medicine, Buddy D. Ratner, Academic Press, 1996.
4. Intelligent Polymer Thin Films and Coatings for Drug Delivery, H. Michelle Grandin and Marcus Textor, John Wiley & Sons, Inc., 2012
5. Arciola, Carla Renata, et al. "Biofilm-based implant infections in orthopaedics." Biofilm-based Healthcare-associated Infections. Springer International Publishing, 2015. 29-46.
6. Donlan, Rodney M. "Biofilms and device-associated infections." Emerging infectious diseases 7.2 (2001): 277.
7. Chen, Meng, Qingsong Yu, and Hongmin Sun. "Novel strategies for the prevention and treatment of biofilm related infections." International journal of molecular sciences 14.9 (2013): 18488-18501.

12328 - Proteomics (4 Credits)

Unit I:

Introduction to protein structure and function in a cell - Amino acids types, properties, and polypeptide synthesis – Four levels of protein structural organization - Enzymes -Types, kinetics, mode of action and Cofactors- Cellular functions of enzymatic and non-enzymatic proteins.

Unit II:

Introduction to System Biology approach- Proteome and Proteomics- Need for proteomic approach- promises of proteomics- Techniques commonly used in proteomic approach. Sample preparation for proteomics- Extraction and solubilization of proteins from cytoplasm, membrane, extracellular, Sub cellular organelles and biological fluids. Challenges associated with low- and high-abundant proteins- Sample pre-fractionation techniques- Liquid phase IEF, Molecular Weight Cut Off and IgG depletion.

Unit III:

Abundance based Proteomics- Gel based Proteomics - Two dimensional gel electrophoresis (2-DE) - Staining Methods and tools for gel analysis- Blue Native PAGE- Differential GE (DIGE) - Gel Scanner - Merits and demerits of Gel based Proteomics. Gel free Proteomics- Liquid Chromatography based separation - Two dimensional and Multidimensional based separation- MudPIT- Merits and demerits of gel-free proteomic technique. Quantitative Proteomics- gel-based quantitative proteomics DIGE- gel free mass spectrometry based quantitative proteomics- label-free, SILAC and iTRAQ. Merits and demerits of quantitative proteomic techniques. Application of gel based and gel-free quantitative proteomic techniques.

Unit IV:

Mass Spectrometry (MS) based Proteomics- Sample preparation for MS based proteomics- Enzymatic digestion of proteins- Criteria for enzyme selection- Basics of mass spectra for peptides. Types of Mass spectrometers- Ionization source- MALDI and ESI- Mass analyzers- Post source fragmentation and Ion detectors. Merits and demerits of different types of mass spectrometers- Applications of Mass spectrometry for proteomics-Tandem MS analysis. MS data analysis- computational tools- Search engines for MS protein identification- MASCOT, SEQUEST, X! TANDEM and Protein Prospector

Unit V:

Interactomics- Yeast Two-Hybrid - Immunoprecipitation- Protein microarrays-Computational tools for Protein-protein interactions- Pros and cons of using various interactomics techniques. Post-translational modifications (PTM) in proteins- Types- Reversible and Irreversible PTM- Techniques for characterization of PTM- Gel electrophoresis and staining procedures for PTM identification- Identification and quantitation of PTM by MS. Public protein databases and interfaces for PTM- Challenges in PTM for proteomics and bioinformatics.

References:

1. Kool, Jeroen, and Wilfried MA Niessen, eds. *Analyzing Biomolecular Interactions by Mass Spectrometry*. John Wiley & Sons, 2015
2. Dekker, Job, Marc Vidal, and AJ Marian Walhout, eds. *Handbook of Systems Biology: Concepts and Insights*, 2014
3. Twyman, Richard M. *Principles of proteomics*. Garland Science, 2013
4. Darvas, Ferenc, András Guttman, and György Dormán, eds. *Chemical Genomics and Proteomics*. CRC Press, 2013
5. Gross, Michael L., Guodong Chen, and Birendra Pramanik, eds. *Protein and peptide mass spectrometry in drug discovery*. John Wiley & Sons, 2011
6. Kannicht, Christoph. *Post-Translational Modifications of Proteins: Tools for functional proteomic*. Humana Press, 2010
7. Zaikin, Vladimir, and John M. Halket. *A handbook of derivatives for mass spectrometry*. IM Publications, 2009
8. Doyle, Sharon A. *High throughput protein expression and purification*. Humana Press; Springer, distributor, 2009
9. Totowa NJ. *Introduction to Proteomics: Tools for new biology*. Humana Press, 2002
10. Timothy P, Kluwer. *Proteomics*. Academic Publishers, 2002
11. Pennington A and Dunn MJ. *Proteomics: From protein sequences to function*. Springer Publications, 2001

Additional Readings:

1. Link, Andrew J., and Joshua LaBaer. *Proteomics: a cold spring harbor laboratory course manual*. Cold Spring Harbor Laboratory Press, 2009
2. Mayne, Janice, et al. "Bottom-Up Proteomics (2013–2015): Keeping up in the Era of Systems Biology." *Analytical chemistry* (2015)
3. Valledor, Luis, and Jesús Jorrín. "Back to the basics: maximizing the information obtained by quantitative two dimensional gel electrophoresis analyses by an appropriate experimental design and statistical analyses." *Journal of proteomics* (2011)
4. Ramu Muthu Selvam, et al. "Exoproteome of *Aspergillus flavus* corneal isolates and saprophytes: Identification of proteoforms of an oversecreted alkaline protease." *Journal of Proteomics* (2015)

12329 - Recombinant DNA Technology (4 Credits)

Unit - I

DNA modifying enzymes and their uses in Molecular Biology a) Restriction enzymes b) DNA Polymerase i) Klenow ii) DNA polymerase I iii) T4/T7 DNA Polymerase c) Reverse Transcriptase d) Terminal Transferases e) T4 Polynucleotide kinases & Alkaline phosphatase f) DNA dependent RNA polymerases. g) DNA ligases h) Nucleases: - Bal 31, S1 nucleases, DNase I, Mungbean nucleases, Ribonucleases, EXO III. Thermostable DNA polymerases used in PCR.

Unit - II

Host cells and Vectors- Host Cell Types (Prokaryotic and eukaryotic). Plasmid vectors for use in E. coli and Gram positive bacteria. Bacteriophage - Lambda and M13 vectors c) Cosmids d) Phagemids. Artificial chromosomes (YACs, PACs, BACs, MACs and HACs). Specialized vectors & their uses a) Expression vectors for Prokaryotes & Eukaryotes - Inducible vectors; vectors with tags (Histidine tags, signalling peptides for exportation), b) Gene fusion vectors.

Unit - III

Cloning strategies: DNA cloning a) Sticky ends b) Blunt ends c) Homopolymeric tailing d) Use of adapters & linkers. Construction of genomic DNA libraries (shotgun cloning) and cDNA libraries. Screening of recombinants - Antibiotic resistance, lacZ complementation (Blue-white selection), fluorescent markers (e.g. GFP). Preparation of radiolabelled/non-radiolabelled DNA & RNA probes. Southern/Northern/Western blot, dot blot and Zoo blot. Screening of genomic libraries with oligo-probe. Immunological screening for expressed genes.

Unit - IV

PCR – basic process, types and applications. DNA sequencing- Principle of chemical and enzymatic methods. Automated DNA sequencing, high throughput Pyrosequencing, next generation sequencing - Lynx Therapeutics' Massively Parallel Signature Sequencing (MPSS), Polony sequencing, Ligation based sequencing (SOLiD sequencing), Ion semiconductor sequencing, DNA nanoball sequencing, sequencing based on reversible dye-terminators (Illumina or Solexa sequencing), Real-Time DNA sequencing, Optical sequencing, Microchip based Sanger Sequencing of DNA, Deep sequencing. Site-directed mutagenesis and protein engineering. DNA footprinting, chromosome jumping, chromosome walking.

Unit - V

Biotechnological applications of rDNA technology: Synthesis and purification of proteins from cloned genes- Native and fusion proteins. Yeast expression system. Production of enzymes. Therapeutic products for use in human health care- insulin, growth hormones, TPA, alpha interferon, Hepatitis B vaccine and Factor VIII. Medical and forensic applications of rDNA technology- DNA Profiling, Multiplex PCR, Diagnosis of inherited disorders and infectious diseases, diagnosis and management of cancer. Treatment using rDNA technology- gene therapy. Gene therapy for ADA and cystic fibrosis.

References:

1. Principles of Gene Manipulation and Genomics (2006) by S. B. Primrose and R. M. Twyman, Blackwell Scientific Publications.
2. Gene Cloning (2007) by Julia Lodge, Pete Lund and Steve Minchin, Taylor and Francis.
3. An introduction to Genetic Engineering (2004) by Desmond S.T. Nicholl, Cambridge University Press.
4. Gene Cloning and DNA Analysis. An Introduction (2006) by T. A. Brown, Blackwell Scientific Publications.
5. Next-Generation Genome Sequencing (2008) by Michal Janitz, Wiley-Blackwell Publications.
6. Recombinant DNA (1992) by J.D. Watson, M. Gilman, J. Witowski and Mark Zoller, Scientific American Books.
7. From Genes to Clones: Introduction to gene technology (1987). Winnacker, E.L.
8. Molecular cloning: A Laboratory Manual (2001). Sambrook, J., Russell, D.W., Cold Spring Harbor Laboratory Press, Cold Spring Harbor, New York.
9. Comprehensive Biotechnology (Vol.1-4) (2004) by Moo-Young, Robinson Howell.
10. Next-Generation Genome Sequencing (2008) by Michal Janitz, Wiley Blackwell.

12330 - Enzyme technology (4 Credits)

Unit I

Enzyme nomenclature, classification, catalytic power, specificity of enzymes, mechanism of enzyme action, extraction and purification of enzymes. Structure of enzymes: determination of M_r (Molecular mass), amino acid composition, primary, secondary, tertiary & quaternary structure. Unfolding and folding of enzymes. Determination of protein structure by x-ray crystallography, NMR and CD.

Unit II

Enzyme production by fermentation method - submerged and solid state fermentations - Recovering enzymes from the fermentation broth - Disruption of cells and tissues - Clarification of culture broth – Filtration, Centrifugation, floatation, concentration, evaporation, Ultra filtration, precipitation, adsorption. Integrated systems for downstream processing. High resolution purification by chromatography - Adsorption chromatography, size exclusion chromatography - Crystallization for enzyme purification. Product formulation.

Unit III

Enzyme catalysis – mechanisms of catalysis, coenzyme - Classification of coenzymes: Isozymes, Apozymes and Synzyme, cofactor, metal activated enzymes and metalloenzymes. Control of enzyme activity. Enzyme inhibition – reversible and irreversible inhibition. Enzyme immobilization – properties and applications of immobilized enzymes.

Unit IV

Finding the gene that makes the enzyme – Transferring the gene to production organism(s). Protein engineering - Rational design, Directed evolution. Enzyme engineering - synthesis of artificial enzymes. Production of enzymes on industrial scale, large-scale purification of enzymes.

Unit V

Biotechnological applications of enzyme: Enzymes and recombinant DNA technology: restriction enzymes and ligases. Clinical aspects of enzymes. Role of enzymes in drug design, enzyme therapy. Industrial Enzymes - Enzymes in food, leather, textile and detergent industries

References

1. Enzymes – Biochemistry, Biotechnology and Clinical chemistry (2004) by Trevor Palmer, Affiliated East- West Press Pvt Ltd, India.
2. Fundamentals of Enzymology 3rd Ed. (2003) by Nicholas C. Price, Lewis Stevens, Oxford University Press, Oxford.
3. Handbook of Enzyme Biotechnology 3rd Edition (1999) by Alan Wiseman, Ellis Harwood Publications.
4. Enzymes: A practical introduction to structure, mechanism and data analysis 2nd edition (2000) by Robert A. Copeland, John Wiley and Sons, New York.
5. Lehninger Principles of Biochemistry (5th Edition) by Nelson DL and Cox MM, Macmillan worth publishers

6. Enzyme technology (2005) by Ashok Pandey, Colins Webb, Carlos Ricardo Soccol, Christian Larroche, Asiatech Publishers, Inc. New Delhi.
7. Molecular cloning: A Laboratory Manual 4th Edition (2012) by Sambrook, J., Russell, D.W., Cold Spring Harbor Laboratory Press, Cold Spring Harbor, New York.
8. Principles of Gene Manipulation and Genomics (2006) by S. B. Primrose and R. M. Twyman, Blackwell Scientific Publicaitions.
9. Isolation and purification of proteins (2003) by Rajni Hatti - Kaul Bo Mattiasson. Marcel Dekker. Inc. New York.
10. Proteolytic enzymes: A practical approach 2nd Edition (2001) by Rob Beynon and Judith S. Bond. Oxford University Press. New York.
11. Enzyme production. Rajni Hatti-Kaul, Lund Univ. Sweden. Biotechnology Vol.V – Enzyme production. Encyclopedia of Life Support System (EOLSS).
12. Tailoring new enzyme function by rational design current opinion in structural biology 10:405-410, 2000 Cedrone et al.

12331 - Microbial Biotechnology (4 Credits)

Unit-I : Fermentation Technology

Brief history of fermentation; Fermentation- general concepts, Applications of fermentation; Range of fermentation process- Isolation, preservation and maintenance of industrial microorganisms, kinetics of microbial growth and death, media formulation for industrial fermentation, Air and media sterilization.

Unit-II : Types of Fermentation

Types of fermentations- Aerobic and anaerobic fermentation, Submerged and solid state fermentation; Factors affecting submerged and solid state fermentation; Substrates used in SSF and its advantages; Culture media- types, components and formulations. Sterilization: Batch and continuous sterilization.

Unit-III: Bioprocess Development

Process development, Optimization of a process, Classical and statistical methods of optimization, Immobilization: different matrices, whole cell and enzyme immobilization; Scale up of bioprocess, Strain improvement, culture preservation and inoculum development, Microbial culture selection, fermented foods, probiotics.

Unit-IV : Concepts of Fermenter

General concept of a fermenter- Batch, fed-batch and continuous fermentation. Aeration and agitation- Effect of aeration and agitation on fermentation, Oxygen requirement and oxygen supply, Foam and antifoams, their effect on oxygen transfer; Fermentation economics.

Unit- V : Microbial Synthesis

Production of proteins and enzymes in bacteria yeast and fungus, recombinant and synthetic vaccines. Microbial polysaccharides and polyesters, Microbial biomass production, utilization of plant biomass by microorganisms (lignocellulose biodegradation), ethanol production, amino acids, antibiotics; Microbial electrosynthesis.

References

1. Prescott & Dunn (2002) Industrial Microbiology, Agrobios (India) Publishers.
2. S. N. Jogdan (2006) Industrial Biotechnology, Himalaya Publishing House
3. R.C. Dubey (2006) A text book of Biotechnology, S. Chand & Company Ltd.
4. Alexander N. Glazer, Hiroshi Nikaido (2007) Microbial Biotechnology – Fundamentals of applied microbiology (2nd Edition), Cambridge University Press.
5. Rabaey, Korneel, and René A. Rozendal. (2010) Microbial electrosynthesis—revisiting the electrical route for microbial production. *Nature Reviews Microbiology* 8, no. 10: 706-716.
6. Gopalarao vuppala, Rama Krishna, Krishna Murthy (March, 2015) Industrial Fermentation, *Research & Reviews: Journal of Microbiology and Biotechnology*, Vol 4 (1).

12332 - Natural Products Chemistry (4 Credits)

Unit I

Natural products: Collection, processing and solvent extraction. Secondary metabolites and their pharmacological properties: Terpenoids, alkaloids, flavonoids, tannins, cardiac glycosides.

Unit II

Drug development from natural products: Role of natural product in primary health care-anti-cancer, neuroprotective, anti-inflammatory, anti-diabetic, hepatoprotective, antioxidant. Protocols and screening methods for: antidiabetic, antiinflammatory, antihepatotoxic, antifertility, diuretic activity and CNS drugs.

Unit III

Nutritional evaluation of natural products: Proximate composition, Physico-chemical characteristics, mineral content, amino acids composition, fatty acid profile, vitamin analysis. WHO guidelines for safety evaluation of natural products. Safety evaluation tests: Ames test, Comet assay, MTT assay

Unit IV

Qualitative analysis for Terpenoids, alkaloids, flavonoids, tannins, cardiac glycosides, anthraquinones. Purification and characterization of compounds: (i) Chromatographic techniques (TLC, column chromatography and HPLC-analytical and preparatory) and (ii) Spectroscopy techniques (UV/Visible, ^{13}C -NMR, ^1H -NMR, GC-MS and IR).

Unit V

New Drug Development process: Pharmacokinetics: Absorption, bioavailability, distribution, metabolism, protein binding and excretion. Mechanism of drug action: receptors, agonists, antagonists, side effects and adverse events. IND Study Protocol. Clinical trial development: Preclinical Studies and Clinical (Phase I, II, III and IV)

References

1. Phytochemical methods – A guide to modern techniques of plant analysis, Third edition – L.B. Harborne. Springer Publishers (1998).
2. Practical Biochemistry – Principles and techniques, Fifth edition – Keith Wilson and John Walker. Cambridge University Press (2000).
3. Drug discovery and Clinical research. First edition - SK Gupta. Jaypee Brothers Medical publishers (2011).