Annexure No.	**42 H**
SCAA Dated	**29.02.2008**

BHARATHIAR UNIVERSITY, COIMBATORE

Syllabus for PG Diploma in Microbial Biotechnology (Affiliated Colleges)

Duration of the course **One year (two semesters)**

Scheme of Examination

Semester	Papers	Title of paper	Max. Marks *	Inst. hrs	Exam hrs
I	Paper I	Fundamentals of microbiology	75	75	3
	Paper II	Microbial genetics	75	75	3
	Paper III	Gene cloning	75	75	3
	Paper IV	Industrial biotechnology	75	75	3
		Practical –I	-	60	-
II	Paper V	Clinical biotechnology	75	75	3
	Paper VI	Plant microbial interaction	75	75	3
	Paper VII	Environmental biotechnology	75	75	3
	Paper VIII	Introduction to bioinformatics	75	75	3
		Practical I	100		6
		Practical II	100	60	6
		Total marks	**800**		

∗ Includes 25% continuous internal assessment marks.

PAPER I: FUNDAMENTALS OF MICROBIOLOGY

UNIT I:

Microscopy - light and laser optic system, Fluorescence microscopy, Inverted Microscopy, Image Analyser and Flow Cytometry.
World of Microorganisms: Characteristics of microorganisms. Taxonomy – Classification of viruses, bacteria and fungi – Methods of studying microorganisms.
Microbial form and function: General structural organization of bacteria, viruses, Actinomycetes. Molecular architecture of nucleus, mitochondria, chloroplast, cell wall, ribosome, cilia, flagella, vacuole and other microbodies.

UNIT II:

Microbial nutrition: Media composition – Preparation of media. Sterilization methods - Thermal death kinetics. Factors influencing the choice of carbon, nitrogen sources, vitamins, minerals, precursors and antifoam agents.
Microbial growth: Growth of microorganisms in different media – growth curve of microbes and different methods of enumeration of multiplying microorganisms. Effect of pH, temperature, ionic concentration on growth.

UNIT III:

Microbial kinetics: Batch, fed-batch and continuous cultures – Phases of batch growth. Kinetics of cell growth – Kinetic models and methods of model parameter estimation – Yield concept and productivity.

UNIT IV:

Replication of microorganisms: Multiplication of bacteriophages, bacteria and differentiating organisms such as yeast, fungi and actinomycetes. Sexual and asexual reproduction in bacteria and fungi. Strain selection and improvement methods.

UNIT V:

Microbial metabolism: Metabolic pathways and bioenergetics. Aerobic and anaerobic growth – product formation and substrate utilization – endogenous and maintenance metabolism. Production of secondary metabolites and their application in industry.

REFERENCES:

1. Microbiology by Pelczar, Reid and Chan, McGraw Hill Book Company.
2. Microbiology, Fundamental and Applications by R.A. Atlas, McMillan Publishers.
3. General Microbiology by Powar and Daginawala, Himalaya Publishing House.
4. Microbial genetics by David friefelder.

PAPER II: MICROBIAL GENETICS

UNIT I:

Genetics – Historical introduction – Mendelian principles – microbial genetics versus Mendelian Genetics. DNA as the genetic material – The duplex DNA – chemical composition – physical structures of DNA – circular and triple helical DNA – Transposons – Repetitive DNA – Satellite DNA – Cytoplasmic DNA - cytoplasmic and nuclear gene interaction
Genetic exchange in bacteria (transformation, transduction and conjugation) – Linkage and genetic maps – phage genetics – phage T mutants – genetic recombination – genetic mapping of phage T.

UNIT II:

DNA replication – Enzymes of replication – DNA editing - rolling circle replication – DNA damage and repair – mutation – mutation versus adaptation – Luria Delbruck experiments and significance – Mutagenesis – Spontaneous and induced mutations – deletions, insertion and point mutations – Physicochemical agents of mutation - mutant selection.

UNIT III:

Protein synthesis: Genetic code – relation between genes and proteins – DNA transcription – Promoters and enhancers - RNA polymerases – post-transcriptional modification - RNA editing – Ribosome structure and function - tRNA structure and amino acid activation – RNA translation – polypeptide synthesis – post-translational modifications.

UNIT IV:

Regulation of gene activity – operon model – autoregulation – transcriptional and translational regulations. Antibiotics on DNA/protein synthesis and mode of action.

UNIT V:

Restriction enzymes: Types, nomenclature and restriction site. Isolation and purification of restriction enzymes.

REFERENCES:

1. Friefelder D. 1995 Molecular biology Narosa Publishing house, New Delhi
2. Maloy S.R. Cronan J.E.Jr and David Friefelder Microbial Genetics, 2nd edition Jones and Bartlett publishers.
3. Yamerin R.H. 1996 Principles of Genetics, 5th edition Wm.C. Brown publishers.
4. Benjamin Lewin 1996 Genes VI Oxford University press. Inc. New York.
5. Klug W.S.and Cummings M.R. 1996. Essentials of Genetics, Prentice Hall New Jersey.

PAPER III GENE CLONING

UNIT I:

Cloning vectors: Types of cloning vectors viz. Plasmids, cosmids, ssDNA phages, Yeast cloning vectors, animal viruses, Ti-plasmid and Cauliflower Mosaic Virus.

UNIT II:

Plasmid biology: Structural and functional organization of plasmids, plasmid replication, Stringent and relaxed plasmids, Incompatibility of plasmid maintenance. Nucleotide sequence and restriction map of p322 plasmid.
Biology of λ-phage: λ-phage as a natural in vivo vector - in vitro construction of lambda vector – Structural organization of λ-phage. Restriction map of λ-phage.

UNIT III:

Enzymes of genetic engineering: DNA polymerase/Klenow's fragment, DNA ligase, Polynucleotide kinase, Nick translation system, Terminal deoxynucleotidyl transferase, Reverse transcriptase and Taq polymerase.

UNIT IV:

Cloning and subcloning strategies: Making genomic and cDNA libraries in plasmids and phages. PCR product cloning (TA cloning). Cloning strategies in yeast, *E. coli* and *B subtilis*. Direct gene transfer methods – Biolistic gun and fusion techniques.
Selection of rDNA clones and their expression products: Direct and indirect methods of gene transfer – Particle bombardment and fusion. Drug / antibiotic resistance and reporter genes. Gene inactivation. DNA hybridization - colony hybridization and in situ hybridization (Southern, Northern and Dot Blots and immunological techniques, Western Blotting).

UNIT V:

Molecular probes - Production, labeling and uses - PCR and its application. Exon cloning, chromosome walking, RFLP, gene transfer, gene therapy and transgenic animals
Protein synthesis in mini and maxi cells, genetic changes for overproduction of biomolecules such as insulin, interferon and growth hormones
Bioethics - Safety guidelines of rDNA research, containment facilities and disposal. IPR

REFERENCES:

1. "Principles of Gene manipulation" by R.W. Old and S.B. Primrose Third Edition. Blackwell Scientific Publication 1985.
2. "Genes VII" by Lewin
3. "Genes to clones" by L. Winnecker.

PAPER IV: INDUSTRIAL BIOTECHNOLOGY

UNIT I:

Bioreactors: Types and modes of operation (Stirred tank, air-lift, bubble column, fluidized bed, packed-bed etc) – Design and construction of bioreactors for different products. Fundamentals of process control and monitoring – control and monitoring theory. On-line and off-line analysis – Sensors for physical, chemical and biological environments. Regulatory and feedback control. – PID controller – regulators and actuators. Computer aided control.

UNIT II:

Transport Phenomena: Introduction to mass, energy and momentum transfer – dimensionless numbers. Rheological properties of fermentation broths (Newtonian and non-Newtonian behaviours). Oxygen mass transfer; Determination of oxygen mass transfer rate – role of aeration and agitation – factors affecting oxygen transfer. Theory of mixing (mixing time and circulation time). Determination of aerated and un-aerated power consumption – Correlation for oxygen transfer coefficient. Heat transfer requirements of microbial cultivations including correlations for determining heat transfer coefficients in natural and forced convections – Various approaches to scale-up the bioprocess. Unit operations involved in upstream and downstream processing of antibiotics, organic acids, alcoholic beverages and industrial enzymes.

UNIT III:

Downstream processing: Characteristics of biotechnological products. Primary separation – removal of insoluble (centrifugation, filtration and sedimentation). Cell disruption (mechanical, enzymatic and chemical). Product isolation – Methods including solvent extraction, adsorption, aqueous two-phase system and precipitation. Purification techniques – Chromatography (ion exchange, gel permeation and affinity), membrane separation (micro-filtration, ultra-filtration and reverse phase electrophoresis). Product polishing (crystallization, drying and diafiltration)

UNIT IV:

Enzyme technology: Practical aspects of large-scale protein purification. Use of soluble enzymes, large scale application of microbial enzymes in food and allied industries, leather industry, textile, paper industries and antibiotics production. Medical application of enzymes in reverse glycosidase in synthetic reaction. Inter-esterification of lipids.

UNIT V:

Immobilization technology: Merits and demerits of cell and enzyme immobilization – methods of immobilization – properties and applications of immobilized enzymes and microorganisms. Characterization of immobilized biocatalysts.

REFERENCES

1.Microbial Biotechnology-Fundamentals of applied Microbiology by A.N.Glazer and H.Nikaido. W.H Freeman and company

2.Principles of Fermentation Technology, P.F.stanbury & A. Whitaker, Pergamon Press.

3.Microbial Process Development by H.W Woelle, World Scientific

4.Biotechnology Text book of Industrial Microbiology by W.Creuger and A Creuger

5.Industrial Microbiology by Casida

6.Industrial Microbiology by Prescott

7.Biochemical Engineering Fundamentals (2^{nd} Ed) by J.E Bailey and D.Ollis, Mc Graw-Hill Book Company.

PAPER V: PRACTICAL I

LAB ON PAPERS I, II, III AND IV

1. Isolation of microorganisms - Media preparation – sterilization techniques
2. Pure culture techniques
3. Staining – gram, simple
4. Estimation of reducing sugar and protein
5. Western hybridization
6. Southern hybridization
7. Genomic DNA isolation
8. Plasmid DNA isolation
9. RNA isolation
10. Library construction (Restriction, digestion and transformation)
11. PCR-RFLP
12. PAGE
13. TLC and Paper chromatography
14. Effect substrate concentration of bacterial growth and estimation of monod parameters
15. Effect of pH and temperature on bacterial growth kinetics
16. Effect of inoculum, age and size of bacterial growth kinetics
17. Effect of substrate concentration on enzyme activity (Vmax and Km)
18. Effect of pH and temperature on enzyme activity
19. Solvent extraction of product from fermentation broth
20. Purification of a fermentation product by ion exchange, gel exclusion and affinity chromatography

PAPER V: CLINICAL MICROBIOLOGY

UNIT I:

Pathogenic/parasitic organisms: Bacterial, viral and protozoal infections of the gastrointestinal system, nervous system, lung, liver and eye; Sexually transmitted diseases, skin infections, zoonoses, arthropod borne diseases. Transmission and spread of diseases – Disease epidemiology.

UNIT II:

Control and prevention of infections – drugs and antibiotics – drug resistance. Mycobacteria, leprosy and malarial parasite – importance, lifecycle, spread and control.

Biochemical changes due to infections – blood test and tissue analysis. Isolation and identification of organisms from tissue samples. Disease detection – conventional and molecular techniques.

UNIT III:

Antigens – types and hapten. Antibody production – Types and structure of antibodies. Antigen-antibody interaction. Role of complement. Cell mediated immunity – cell types in blood and tissues and mechanism of action. Antibody producing genes and antibody synthesis. Methods of antibody measurement. Immune deficiency and autoimmune diseases.
Production of antibody against specific proteins/molecules – applications. Monoclonal antibodies and hybridoma technology

UNIT IV:

Molecular architecture of AIDS virus; Viral hepatitis and jaundice. Septicaemia – causes, clinical features and prevention. Hospital infection and control.

UNIT V:

Vaccines – types and methods of action. Biotechnological approaches to disease control and vaccine production. Genetic disorders and Gene therapy. Control of vectors – Mosquito control - Biotechnological approaches.

REFERENCES

1. Immunology, Roitt, I.M., Brestoff and Male, D.K, 1996
2. Principles of Gene manipulation" by R.W. Old and S.B. Primrose Third Edition. Blackwell Scientific Publication 1985.
3. Molecular Biotechnology – Glick
4. Clininal microbiology - Ananthanarayanan

PAPER VI: PLANT AND MICROBIAL INTERACTION

UNIT I:

Biofertilisers: Symbiotic nitrogen fixation in legumes by Rhizobia – nodule formation and biochemistry and molecular biology of nitrogen fixation; Non-symbiotic nitrogen fixing bacteria; Nitrifying and denitrifying bacteria. Enzymes of nitrogen fixation and assimilation – Nitrogenase, Glutamate dehydrogenase, glutamine synthetase, glutamate synthase and allantoinase. Nitrogen fixation by blue green algae. Engineering *nif* genes and applications. Azospirillum, Vesicular arbuscular mycorrhiza and phosphobacteria as biofertilizers. Commercial production of biofertilizers and quality control.

UNIT II:

Plant tissue culture: Callus culture – sterilization methods – media types and composition - organogenesis - meristem culture - anther, pollen and embryo culture and their applications. Protoplast isolation and culture and its application. Cell culture and secondary metabolite production.

UNIT III:

Plant transformation: *Agrobacterium* - Crown gall tumours - Mechanism of T-DNA transfer to plants, Ti-plasmid vectors for plant transformation, agroinfection. *Bacillus thuringiensis* – Plant transformation with *Bt* genes and *cry* proteins. Success stories and achievements in plant transformation – disease resistance, pest resistance, herbicide tolerance, nutritional quality improvement, production of antibodies, viral antigens, vaccines and peptide hormones.

UNIT IV:

Plant-pathogen interaction: Classification of plant viruses. Molecular biology of tobacco mosaic virus. Plant pathogens – host recognition - plant-pathogen interaction – virulence and avirulence; Toxin production, deranged metabolism in plants and symptom development. Disease detection and control.

UNIT V:

Biopesticides – predators and parasites of plant pests and pathogens. Production of biocontrol agents and quality control. Botanical pesticides – Neem products and other naturally occurring components toxic to pests and pathogens.

REFERENCE:

1.Invitro Culture of Higher Plants, by R.L.M.Pierik

2.Plant Cell Culture, A Practical Approach, II Edition, R.A Dixon and R.A Gonzales.

3.Plant Molecular Biology by Grierson and S.V Convey

4.Genetic Engineering of Crop Plants. Edited by G.W Lycett and D.Grierson

5.Plants, Genes and Agriculture by M.J Chrispeels and D.F Sadava.

6. Plant Biotechnology by Slater et. al., 2003, Oxford University Press

PAPER VII: ENVIRONMENTAL BIOTECHNOLOGY

UNIT I:

Water purification: Microbiology and biochemistry of drinking water and water purification. Waste /sewage water treatment and recycling – Biological processes for industrial effluent treatment – aerobic, anaerobic and biological treatment, periodic biological reactors, membrane bioreactors, use of immobilized enzymes and microbial cells.
Effluent treatment: Biotechnological applications for pesticide industry, tannery industry, textile, food, distillery and paper industry – processing and effluent treatment – Common Effluent Treatment Plant – Recycling of waste and recovery of byproducts. Disposal of radioactive wastes.

UNIT II:

SCPs – large scale production - nutritional quality and disadvantages. Algae as food. Edible mushroom production. Biotechnology in food industry – genetic engineering for protein, amino acid, vitamin and food additive production.

UNIT III:

Bioremediation: Definition - Case histories, constraints and priorities of bioremediation. Bio-augmentation; bioreactors for remedial processes, types of bioremediation, application – examples, biotechnology and oil spills. Bioremediation of mine sweepings.
Bioremediation of heavy metals: Microorganisms for ore concentration and leaching. Reclamation of mine sweepings - Biosorption and bioleaching. Heavy metal elimination from sewage water and effluents – Biotechnological approaches.

UNIT IV:

Xenobiotic compounds: Recalcitrance – hazardous wastes – disposal of radioactive wastes. Biodegradation of xenobiotics - Biological detoxification; Biodegradation of DDT, BHC and malathion in soil, plants and insects. Biotechnological methods for hazardous waste management.

Biotechniques for **air pollution** abatement and odor control: Deodorization process, application.

UNIT V:

Solid waste management: Hospital wastes and their disposal. Agricultural wastes and Municipal waste – Incineration – Composting and vermicompost; Biogas production – methanogenic bacteria – design of biogas plants. Electricity production from waste.

Environmental applications of microbiology: Organic farming - Biofertilizers and biopesticides. Pollution control through use of consortium of microorganisms. Disposal of plastics - Bioplastics.

REFERENCES:

1.Waste Water Engineering Treatment and Disposal and Reuse by Metcalf and Eddy.

2.Water Pollution management hand book by Lepathak

3.Waste Water management by Arceivala

4.Environmental biotechnology by C.F forster and D.A. J. wase

5. New Processes of Waste Water Treatment and Recovery by G.Mallock (ED) Ellis Horwood.

6.Biochemical Engineering Fundamentals, 2nd ed by J.E Bailey and D.F Ollis Mc Graw-Hill (1986) Chapters 13 & 14.

7.Environmental Biotechnology by jogdand.

PAPER VIII : INTRODUCTION TO BIOINFORMATICS

UNIT I:

Elements of computer science – hardware, software – hierarchies in software, operating systems and application software algorithms and computational complexity examples – traveling salesman problem – the Internet and applications.

UNIT II:

Bioinformatics – definition, scope and uses. The digital nature of biological information – elements of molecular biology – the transfer of information in biological systems. Representation of biological molecules as strings of symbols – correspondences to other branches of computation, including computational linguistics, pattern recognition, image processing etc.

UNIT III:

Databases – computer databases – biomolecular databases – structural databases – details of organization access and deposition – derived and specialized databases – Mapping databases. Data mining

UNIT IV:

DNA sequencing by chemical, enzymic and big-bye terminator methods. Sequence projects – structure projects – definitions – structural and functional genomics.

UNIT V:

Molecular modeling softwares - structures of compounds - Three dimensional structure – Change of functional groups and change of sequences in proteins and nucleic acids.

Algorithms for analysis of sequence – homology versus similarity – dot matrices – sequence comparison using Needleman and Wunsch method – Hash coding – Sequence databases - BLAST and FASTA – structure analysis – distance matices – examples. Dendrogram and Phylogenetic analysis.

REFERENCES:

1.Sequence Analysis in Molecular Biology, Gyon Heijane Academic Press (1987)

2.Bioinformatics: a practical guide to the analysis of genes and protein A.D Baxevanis and B.F Francis (eds) John Wiley and sons(1998)

3.Bioinformatics methods and protocols S.Misener and S.A Frawetz, Humana Press (2000)

4.Introduction to Bioinformatics T.E Atwood and D.J. Parry-Smith Addision Wesley Longman Ltd (1990)

PAPER X: PRACTICAL II

LAB ON PAPERS V, VI, VII, AND VIII

1. Callus induction and differentiation
2. Isolation, fusion and regeneration of protoplasts
3. Agrobacterium mediated gene transfer
4. Batch cultivation of bacteria in fermentor; Different phases of bacterial growth (Estimation of μM, K_s and $Y_{X/S}$)
5. Medium optimization for bacterial growth by statistical method
6. Immobilization of bacteria and enzyme by calcium alginate method
7. Production of alcohol from molasses
8. Production of citric acid
9. Production of amylase
10. Estimation of oxygen transfer coefficient (KLa)
11. Analysis of pH, turbidity, color, total solids, suspended solids, dissolved solids
12. Estimation of COD, BOD
13. Estimation of Iron and chromium
14. MPN test
15. IMViC test
16. Calculation of bond length, bond angles and tension angles
17. Comparison of 3D structures of protein/nucleic acids
18. Searches of MEDLINE, CD_ROM and bibliographics data bases