SHIVAJI UNIVERSITY, KOLHAPUR
Biotechnology (Entire)
B. Sc III. Biotechnology (Entire)

Course Code	Title of the Course	Theory / Practical	Marks	Lecture /Practical Hours
BTE – 301	Genetic Engineering	Theory	100	90 L
BTE - 302	Industrial, Food and Microbial Biotechnology	Theory	100	90 L
BTE - 303	Applications of Biotechnology in Agriculture and Health	Theory	100	90 L
BTE - 304	Developmental Biology (Plant and Animal) and Bioinformatics.	Theory	100	90 L
BTE - 311	Techniques in Genetic engineering and Bioinformatics.	Practical	100	30 P
BTE - 312	Techniques in Industrial, Food, Agricultural, Health and Microbial Biotechnology	Practical	100	30 P
BTE - 313	Project	Practical	100	30 P

BTE 301 Genetic Engineering

Topic No.	Section I	Lectures 45
	Unit I	12
1.	**Molecular Tools in r-DNA technology** 1. Introduction and Scope 2. Molecular tools and its application 2.1 Restriction enzymes- types (I, II, III), nomenclature, recognition sequences, cleavage patterns, modification of cut ends (linkers and adaptors), application –RFLP, Restriction mapping. 2.2 Alkaline phosphatases 2.3 DNA ligases T4 and *E. coli* Ligases 2.4 Methylases 2.5 Reverse Transcriptases 2.6 Polymerases- Klenow enzymes, T4 DNA polymerases, Taq DNA polymerases 2.7 Polynucleotide kinase	
	Unit II	12
2.	**Cloning Vectors:** 1. Introduction 2. Properties of good vectors 3. Cloning & expression vectors 4. Types- 4.1 *E.coli* vector 4.1.1 plasmid – pBR 322 and pUC18 4.1.2 Bacteriophage vectors – λ phage vector, M 13 Vectors (λ replacement e. g. EMBL 3, EM BL 4and λ insertional e.g λ gt 10 and λgt 11) 4.2 Cosmid vector 4.3 Phagemid vector e.g pBlue script II KS/SK 4.4 Yeast vector- YAC and BAC 4.5 Animal vectors – Retroviral 4.6 Plant vector – Ti plasmid,Ri plasmid 4.7 shuttle vector- e.g pJBD 219 5. Selection of recombinant vector	

	Unit III	12
3.	**Nucleic Acid Hybridisation :** 3.1 Nucleic Acid and plasmid purification. 3.2 Probe Preparation 3.3 Methods of labelling probes. 3.3.1 Radio labelling – Nick translation, End labeling, Primer extension 3.3.2 Non Radiolabelling – Biotin, dioxygenin , fluorescent dyes, 3.3.3 Applications of probes.	
	Unit IV	
4.	**DNA Sequencing and blotting technique** 4.1 Maxam Gilbert method 4.2 Sanger Coulson method 4.3 Automated DNA sequencing 4.4 Southern Blotting 4.5 Northern Blotting. 4.6 Western blotting	09
	Section: II	Lectures 45
	Unit V	12
5.	**Isolation of Gene** 5.1 Chemical synthesis 5.2 Isolation desired gene from DNA 5.3 Isolation of specific gene with PCR 5.4 cDNA and genomic library . 5.5 Screening of libraries- immunological screening and colony or plaque hybridization.	
	Unit VI	12
6.	**PCR and its application** 6.1 Primer designing 6.2 Fidelity of thermostable enzymes. 6.3 Steps in PCR reaction 6.4 Types of PCR – RT-PCR,real time PCR, touch down PCR,hot start PCR,colony PCR 6.5 Applications- site directed mutagenesis, Molecular diagnostics ,viral and bacterial detection, RAPD, AFLP	

	Unit VII	12
7.	**Cloning methodologies** 7.1 Construction of plasmid – e. g. Somatostatin 7.2 Insertion of foreign DNA into host cells 7.2.1 Transformation 7.2.2 Transfection 7.2.3 Chemical methods- $CaCl_2$ coprecipitation, polycation mediated gene transfer. 7.2.4 Physical methods- Liposomes, microingection, electroporation, biolistics. 7.3 screening of recombinants	
	Unit VIII	09
8.	**Application of r-DNA technology** 8.1 Production of transgenics-knock out mice 8.2 In medicines –Insulin and Somatostatin 8.3 Gene Silencing- Introduction, Principle of Si-RNA and Si- RNA technology	

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References:

1. Molecular Biotechnology – Principles & applications of Recombinent DNA : Glick B. R. & Padtranak
2. Gene cloning & manipulating – Christopher
3. An introduction to genetic engineering – Nicholl D.S. T.
4. Principle of gene manipulation : An introduction to genetic engineering – Old R.W. & Primrose S. B.
5. Gene VIII – Lewin
6. Fundamentals of Biotechnology – S. S. Purohit
7. Fundamentals of Biotechnology – H. S. Chawala
8. Genetic engineering – P. K. Gupta
9. Priciple of Biochemistry – Wilson & Walker
10. Plant genetic engineering – P. K. Gupta
11. Molecular Biotechnology of gene – S. N. Jogdan
12. Protein Biotechnology – M. Philopse
13. Molecular Biotechnology – Principle & practices by Channarayappa
14. Biotechnology – R. C. Dubey
15. Molecular cloning (Vol I, II, III) – Sambrook and Russel

BTE 302 Industrial, Food and Microbial Biotechnology

Topic No.	Section –I	Lectures 45
	Unit I	12
1.	**Introduction to Industrial Biotechnology** 1.1 Concept and range of fermentation technology 1.2 Types of commercially important fermentations 1.3 Chronological developments of industrial fermentation technology.	
2.	**Basic Design of Typical Fermenter** 2.1 Components of fermenter and their functions 2.2 Comparison between fermenter and Bioreactor.	
	Unit II	10
3.	**Microbial Screening** 3.1 Selection strategy and techniques 3.2 Primary and secondary screening 3.3 Primary screening of antibiotics, organic acids, amines, amylases, proteases, vitamins and amino acid producers 3.4 Secondary screening of antibiotic producers 3.5 Scale up 3.6 Strain improvement--Concept - Isolation of microbial mutants 3.7 Stock culture maintenance and preservation 3.8 Contaminants and their role with respect to - -*Staphylococci* in penicillin fermentation -Phage in acetone-butanol fermentation	
	Unit III	10
4.	**Fermentation Media and Growth Kinetics** 4.1 Composition of typical fermentation media . 4.2 Criteria for typical fermentation medium 4.3 General role of media components- water, carbon source, nitrogen source, minerals, precursors, growth factors, buffers, antifoams, oxidation-reduction potentials, inducers,	

	inhibitors. 4.4 Optimization of media (Plakett and Burman design) 4.5 Types of fermentation media 4.6 Factors affecting fermentation process 4.7 Sterilization of air and media 4.8 Material and Energy Balance- Concept, Calculations.	
	UNIT IV	13
5.	**Downstream Process and Product Recovery** 5.1 Downstream Processes in - Fermentation and bioprocess technology - Solid and liquid separation - Flocculation and crystallization - Drying, filtration and centrifugation - Cell disruption by solid and liquid shear, ultrasonication, enzyme action and mechanical disruption 5.2 Product recovery and purification -Principle - Extraction and precipitation - Distillation (Fractional and Steam)- Principle, Process, Calculations - Separation of completely miscible (zeotropic and azeotropic) and partially miscible binary liquid-liquid mixtures -Evaporation - Chromatographic separation (Principles of all methods) - Adsorption and concentration - Lyophilization, spraying, drying and packing	
	Section- II	Lectures 45
	Unit-V	10
6.	**Microbial Cultures and Production** 6.1 Concept of pure and mixed culture	

	6.2 Microbial growth kinetics (Batch , Continuous and Fed Batch) 6.3 Microbial Production of - Enzymes (amylase) - Antibiotics (Penicillin ,Streptomycin) -Vitamins (B_{12}) - Single Cell Protein- (Yeast) - Amino acids (Lysine).	
	Unit- VI	12
7.	**Fermented Foods and Beverages** 7.1 Dairy Products – Cheese , Yoghurt, Indian Dairy Products 7.2 Indian Foods – Idli, Dahi , Gilebi 7.3 Bakery Products – Bread 7.4 Basics of Extruded Foods 7.5 Beverages – Beer, Wine 7.6 Fermented Pickles – Sauerkraut and Cucumber pickles .	
	Unit- VII	10
8	**Food Spoilage and Preservation** 8.1 Types of spoilage- Physical, Chemical and Biological (auto and microbial) 8.2 Preserving methods - High and Low temperatures - Controlled atmosphere and Aerobiosis - Radiations and Asepsis - Chemical preservatives (Salt, sugar, organic acids,SO_2)	
	Unit- VIII	13
9	**Integration of Genetic Engineering and Microbial Biotechnology** Uses of genetically engineered microbes in agriculture and medicines.	
10	**Introduction to Food Biotechnology** 10.1 Principle, Risk analysis and Regulations 10.2 Multidisciplinary perspectives of GM crops	

11	**Impact of food on human health** 11.1 Public health principles 11.2 Characteristics of food supply for public health 11.3 Food Toxicity – Mycotoxin (Aflatoxin), Exotoxin (*Staphylococcal*) . 11.4 Food borne illness- Shigellosis, Amoebiosis , Aspergillosis. 11.5 Nutrition 11.6 Threats to organic farming	

References:

1. Text Book of Biotechnology – Dr. H. K. Das
2. Industrial Microbiology & Biotechnology – Arnold L.
3. Fermentation Technology – Jayanto Acharekar
4. Basic Biotechnology – Colin and Bjrorn
5. Frontiers in Microbial Biotechnology – Bisel P.S.
6. Industrial Microbiology – Prescot and Dunn
7. Principle of Fermentation Technology – Stanbury P.F., Whitekar H., Hall S. J.
8. Principle of Management – Koontz & O. Donehl
9. Bioprocess Engineering : Principles – Nielson T. and Villadeson J.
10. Biochemical Engineering – Aiba.S, Humphray A.E., Millis F.
11. Industrial Microbology- L.E. Casida
12. Fermentation Biotechnology- H.A. Modi
13. Principles of Fermentation Technology- A.H.Patel
14. Food Biotechnology- Marwa, Arora

BTE 303 Application of Biotechnology in Agriculure and Health care

Topic No.	Section I	Lectures 45
	UNIT - I	12
1	**Methods for crop Improvement** 1.1 Introduction 1.2 Acclimatization 1.3 Breeding for self and cross pollinated plants and vegetatively reproducing plants , selection (pure line and mass) hybridization and mutation. 1.4 Somaclonal variations 1.5 Haploids 1.6 Micropropagation . 1.7 Somatic embryogenesis 1.8 Synseeds	
	Unit II	10
2	**Markers in plant breeding** 2.1 Introduction – Morphological , Biochemical, Molecular Markers 2.2 Molecular markers RFLP,RAPD,AFLP,STRS,QTL,SSR	
	Unit III	11
3	**GM Crops** 3.1 Herbicide resistance, bacterial, fungal, virus, insect resistance 3.2 GM Foods , ethical & social aspects 3.3 Concept of IPR and IPP, forms of protection 3.4 Molecular farming.	
	Unit IV	12
4	**A) Biofertilizers and Biopesticide** **Biofertilizers –** 4.1 Defination , Principal (Biological nitrogen fixation) 4.2 Mass production and field application –	

	Rhizobium, Azotobacter, Azospirillum, Acetobacter, Azolla, Cyanobacteria, PSB, VAM 4.3 Green manure and compost **Biopesticide –** Priciples and applications 4.4 Bacterial , fungal, viral , Plant origin Biopesticides **B) Biotechnology of secondary metabolites**	
	Section II	Lectures 45
	Unit V	12
5	**Applications of animal cell culture.** 5.1 Characteristics of stem cells , 5.2 Concept of stem cell progenitors. 5.3 Concept of stem cell technology and its application. 5.4 Transgenic technology & cloning in mammals	
	Unit VI	10
6	**Vaccines- Principle & practices** 6.1 Subunit vaccines- Hepatitis B vaccine, Foot and Mouth disease Vaccine, AIDS Vaccine 6.2 DNA Vaccines 6.4 Edible Vaccines 6.3 Recombinant vaccines- Cholera Vaccine, Vaccinia Virus Vaccine **Interferon Production**	
	Unit VII	12
7	**Monoclonal antibodies & hybridoma technology** 7.1 Production 7.2 Formulation 7.3 Applications- Diagnostics & Therapeutics **Biosensors-**	

	7.4 Principle & applications. 7.5 Biochips & microarray technology **Gene therapy –** 7.6 Types – Somatic, Germline, Argumentation 7.7 Gene therapy strategies for cancer	
	Unit VIII	11
8	**Forensic medicine** 8.1 Prepration of DNA sample 8.2 Approches of DNA analysis **Public health** 8.3 Epidemiology 8.4 Diagnosis of infectious diseases 8.5 Detection of genetic diseases 8.6 Diagnosis of cancers	

References:

1) Biotechnology – U. Satyanarayana
2) A textbook of plant breeding – B.D. Singh
3) Medical biotechnology – S. N. Jogdand
4) Advances in Biotechnology- S.N.Jogadand
5) Introduction to plant breeding – R. C. Chaudhary
6) A textbook of Biotechnology - R. C. Dubey
7) Pharmaceutical Biotechnology – S. P. Vyas ,V. K. Dixit
8) Biotchnology – B. D. Singh
9) Fundamentals of agriculture biotechnology – S. S. Purohit
10)Animal & cell biotechnology – Ian, Freshney
11) Animal cell biotechnology – Buttler
12) Methods in cell biology – Volume 57
13) Cell and Developmental Biotechnology.-Raj narian Desikar
14) Text Book of Bryophytes, Pteridophytes, Gymnosperms, and Paleobotany- Subramurti.
15) Agricultutre application of Microbiology- Neeelima Rajvaidya .

BTE 304 Developmental Biology (Plant and Animal) and Bioinformatics.

Topic No.	**SECTION I** **Developmental Biology (Plant and Animal)**	Lectures 45
	Unit I	10
1.	**Gametogenesis and Fertilization in plants** 1.1 Gametogensis in Plants 1.2 Development of male and female gametophyte 1.3 Process of fertilization in Angiosperm	6
2.	**Development of Embryo and Endosperm** 2.1 Development of embryo and endosperm 2.2 Types of endosperm in Angiosperm	4
	Unit II	10
3.	**Plant meristem** 3.1 Plant meristem , organization and differentiation 3.2 Organization of shoot apical meristem 3.3 Organization of root apical meristem.	8
4.	**Pollen germination** 4.1 Pollen germination. 4.2 Self incompatibility and its genetic control.	2
	Unit III	12
5.	**Gametogensis , gametes and fertilization in animals** 5.1 Gametogensis in animals. 5.2 Types of eggs and sperms in animals. 5.3 Fertilization in animals.	5
6.	**Early development in animals**	

	6.1 Types and patterns of cleavages in animals. 6.2 Blastulation ,gastrulation in frog and chick up-to the formation of three germ layers. 6.3 Embryonic induction .	7
	Unit IV	13
7	**Differentiation** 7.1 Differentiation 7.2 Didifferentiation 7.3 Rediffrentitation 7.4 Commitment 7.5 Transdifferentiation 7.6 Developmental plasticity	8
8	**Cell fusion , somatic cell genetics and immunoglobulin genes** 8.1 cell fusion and somatic cell genetics 8.2 Immunoglobulin genes and antibody Diversity	5
	Section- II **Bioinformatics**	Lectures 45
	Unit- V	10
9	9.1 Introduction to Bioinformatics:- History, Computers in Biology and Medicines, Internet, and related programs; Networking, WWW, Netscape, Internet Browsers, Telnet, FTP (File transfer protocol), HTTP, HTML, WAN, LAN, MAN, applications in communication 9.2 Information Resources:- Introduction, aim and objectives, National Centre for Biotechnology Information(NCBI), National Library of Medicine (NLM), and National Institute of Health (NIH), EBI, Sequence retrieval system (SRS):- Entrez, DBGet, EBI.	
	Unit- VI	11

10	10.1 Genomics:- Introduction, Genes, Genomes, Human Genome Project (HGP), Goals of HGP, Rough and final draft of HGP, Application of HGP in modern day discovery process. 10.2 Other Genome projects:- Mouse genome, *Arabidopsis thaliana* 10.3 Genome databases:- Introduction, Databases, Data, Nucleic acid sequence database, Gene Bank, EMBL, DDBJ, Expressed sequence tags (EST), STS, C-DNA libraries	
	Unit- VII	12
11	11.1 Proteomics:- Introduction to amino acids and protein, Proteome, Protein structure, Primary protein sequence database, SWISS-PROT, PIR, MIPS, NRL-3D, TrEMBL, Annotation and applications. 11.2 Secondary protein sequence databases:- PROSITE, PROFILE, PRINT, pfam, BLOCK, IDENTIFY; applications. 11.3 Other databases:- Literature database, PubMed, PubMed central, Journal database, Book, CATH, SCOP, PdbSum	
	Unit- VIII	12
12	12.1 Structural databases:- Introduction, Difference between Primary structure and 3D structure, Protein databank(PDB), Nucleic acid databank, Molecular modeling databank (MMDB), 12.2 Sequence Alignment:- Introduction, Protein sequence, Nucleic acid sequence, Pair wise sequence alignment, Multiple sequence alignment, Local and Global sequence alignment, Algorithm used in sequence alignment, Matrices- Dot matrix, PAM, BLOSSOM, 12.3 Bioinformatics in pharma industries :- Drug designing, Pharma informatics resources.	

References:-

1. Developmental Biology-Gilbert
2. Foundations of Embryology – Patten
3. Cell and Developmental Biotechnology – Raj Narian Desikar
4. Text book of Bryophytes, Pteridophytes , Gymnosperms and Paleobotany - Subramurti
5. Plant Anatomy and Embryology- S.N. Pandey, A. Chadha
6. Teresa K Attwood and David J. Parry-Smith, Introduction to Bioinformatics, Pearson Education Asia, 2001
7. Bexavanis & Francis, Bioinformatics-A practical guide to the analysis of genes and proteins, John Wiley and Sons, 2001
8. Rushidi, Basics of Bioinformatics, CRC Publications, 2001
9. Irfan Khan and Atiya Khanum, Emerging trends in Bioinformatics, Ukaaz Publishers, 2002
10. David M. Hill, Craig Martiz and Barke Mable, Molecular systematics
11. Khan Imtiyaz alam ,Rai University, Hydrabad:- Elementry Bioinformatics
12. N. Gautam Bioinformatics- Databases and algorithm
13. Plant Anatomy – E.Cutter.
14. The Embryology of Angiosperm – Bhojawani .S.S and Bhatnagar.S.P
15. An Introduction to the Embryology of Angiosperm. – P.Maheswari.

BTE– 311 Techniques in Genetic engineering And Bioinformatics

Sr. No.	Name of the Practicals	30 P
1.	Calculation of molecular size of digested DNA	01
2.	Construction of restriction map of plasmid DNA	02
3.	Western blotting technique	03
4.	Southern blotting technique	03
5.	DNA Amplification by PCR	01
6.	cDNA cloning by Reverse Transcription PCR	02
7.	Purification of DNA fragments from agarose gel	02
8.	Ligation of DNA	02
9.	Transformation of *E. Coli* and Selection of recombinants (β-galactosidase)	03
10.	*Agrobacterium* transformation in plants	02
11.	Expression of gene in *E. Coli* (GST)	02
	Bioinformatics	
12.	Introduction to NCBI	01
13.	Extraction and Analysis of Protein Sequence by using Protein Database	01
14.	Extraction and Analysis of Gene Sequencing by using genome database	01
15.	Introduction to Protein Data Bank (PDB) and Ras Mol- To visualize 3D structure of protein	02
16.	Compulsory visit to –Molecular Labs. (Introduction to DNA sequencing, PCR , DNA finger printing), Bioinformatics Laboratory	02

BTE – 312 Techniques in Industrial, Food, Agricultural, Health and Microbial Biotechnology

Sr. No.	**Name of the Practicals**	30P
01.	Primary screening of amylase producers by Replica Plate technique	02
02.	Primary screening of antibiotic producers by Agar Overlay method	02
03.	Production of primary metabolite (Amylase) - Purification, Immobilization and Activity	02
04.	Production of alcohol from sugarcane molasses or sugarcane juice	02
05.	Isolation of *Azotobacter, Rhizobium*, PSB and preparation of biofertilizer and to check its viability on 3^{rd} and 7^{th} day	02
06.	RAPD Analysis	01
07.	Determination of antibacterial activity of crude plant extract.	01
08.	Production of sauerkraut.	01
09	Mushroom Biotechnology: Demonstration, Cultivation, Practices.	01
11.	Isolation of vitamin B_{12} requiring mutants.	01
12.	Precipitation and recrystallization of citric acid.	01
13.	Bioassay- a) Vitamin B_{12} b) Penicillin	02
14.	Isolation of bacteria from fermented broth/ spoiled food.	02
15	Preparation of Biopesticide- *Trichoderma spp.*	01
16	Detection and isolation of pathogens (*E.Coli, Salmonella, Staphlococci*) from fermented milk.	02
17	Milk Analysis- a) Estimation of lactic acid. b) Estimation of total fat. c) MBRT	02
18	Alcoholometry- a) Estimation of alcohol in beverage by colorimetric method. b) To study kinetics of oxidation of	02

	alcohol by dichromate method. c) To distinguish ethanol from methanol using iodoform test.	
19.	Separation of crude caffeine from tea leaves/ tea powder.	01
20.	Industrial Visit- Vaccine Production , Tissue Culture Industries, Biofertilizers Industries. Wine Industry, Food Processing Industry.	

BTE 313: Project

The projects are to be prepared by the students on the subjects in consultation with the Project Guide in the academic year. The Guide will guide the students in selecting the topic of the project, working of the experiments, results of the same & writing the report.

The student has to submit this project report, duly signed by the project guide and Principal, before practical examination. The report will be assessed by both Internal examiner (The project guide), who will assign the marks out 50 and the external examiner (appointed by university), who will assign marks out of 50, Thus the total will be out of 100 marks.

Theory, practical and project report shall form separate heads of passing.

Practical Examination:-

A) The practical examination will be conducted on three (3) consecutive days for each practical not less than 5 hours on each day of the practical exanimation.

B) Each candidate must produce a certificate from the Head of the Department in his/her college stating that he/she has completed in a satisfactory manner the practical course on the guidelines laid down from time to time by Academic Council on the recommendation of Board of studies and has been recorded his/her observations in the laboratory journal and written a report on each exercise performed. Every journal is to be checked and signed periodically by a member teaching staff and certified by the Head of the Department at the end of staff and certified by the Head of the Department at the end of the year. Candidates are to produce their journal at the time of practical examination. Candidates have to visit the Biotechnological institutes as per the syllabus and satisfactorily complete project work. The visit and project report should be duly certified by the Head of the Department and submit the reports at the time of examination.

BTE– 311 :- Techniques in Genetic engineering and Bioinformatics

Q.1 A) Major Experiment	**20 Marks**
B) Major Experiment	**20 Marks**
Q.2 A) Minor Experiment	**10 Marks**
B) Minor Experiment	**10 Marks**
Q.3 Spotting	**10 Marks**
Q.4 Tour Report	**10 Marks**
Q.5 Journal	**10 Marks**
Q.6 Viva-voce	**10 Marks**

BTE – 312 Techniques in Industrial, Food, Agricultural, Health and Microbial Biotechnology

Q.1 A) Major Experiment	**20 Marks**
B) Major Experiment	**20 Marks**
Q.2 A) Minor Experiment	**10 Marks**
B) Minor Experiment	**10 Marks**
Q.3 Spotting	**10 Marks**
Q.4 Tour Report	**10 Marks**
Q.5 Journal	**10 Marks**
Q.6 Viva-voce	**10 Marks**

BTE 313: <u>Project</u>

A) Internal Examination	**50 Marks**
B) External Examination	**50 Marks**