

Annexure No.	42 J
SCAA Dated	29.02.2008

BHARATHIAR UNIVERSITY, COIMBATORE – 641046

PG DIPLOMA IN GENETIC ENGINEERING (Colleges)

For the students who are admitted from the academic year 2007-08 batch and onwards.

DURATION: ONE YEAR (TWO SEMESTERS)

ELIGIBILITY FOR ADMISSION:

B.Sc. Degree in Biology, Biochemistry, Botany, Microbiology, Zoology, Agriculture or B.Pharm or M.B.B.S. or B.V.Sc. of a recognised Indian or Foreign University.

SCHEME OF EXAMINATIONS:

SEMESTER	SUBJECTS	MAX.MARKS *	
I	PAPER I	PRINCIPLES OF GENETIC ENGINEERING	75
	PAPER II	MOLECULAR GENETICS	75
	PAPER III	GENOMICS AND PROTEOMICS	75
	PAPER IV	INTRODUCTION TO BIOINFORMATICS	75
II	PAPER V	MICROBIAL STRAIN MANIPULATION	75
	PAPER VI	PLANT TRANSGENICS	75
	PAPER VII	ANIMAL TRANSGENICS	75
	PAPER VIII	BIOETHICS, BIOSAFETY AND IPR	75
	PRACTICAL I	BASICS OF GENETIC ENGINEERING	100
	PRACTICAL II	APPLIED GENETIC ENGINEERING	100

DURATION OF EACH PRACTICAL EXAMINATIONS 6 HRS.

* Includes 25% continuous internal assessment marks.

Paper I - PRINCIPLES OF GENETIC ENGINEERING

Preamble

Scope: This paper provides the student a thorough knowledge in principles and methods in genetic engineering, vectors in gene cloning, transformation in higher organisms and other research works of importance like The Human Genome Project and Gene therapy.

Objective: The students will have an exposure to the application of Recombinant DNA techniques in various aspects of biotechnological research areas.

Goal: This paper will help the student to understand and appreciate the latest advances in recombinant technology and will expose the students to the novel techniques in genetic engineering.

Unit- I

Principles and methods in genetic engineering: Isolation and purification of Nucleic Acids - Agarose Gel Electrophoresis - Southern, Northern and South-Western blotting techniques- Polymerase Chain Reaction: Methods and advancements - Enzymes in Molecular Biology: Nucleases, Restriction endonucleases, DNA Ligases, topoisomerases, gyrases, methylases, other modifying enzymes – Bacterial Transformation: Principles and methods.

Unit-II

Vectors in Gene Cloning: Plasmids, Bacteriophages, Phagemids, Cosmids - Artificial Chromosomes: PAC, BAC, YAC - Cloning in Prokaryotes *E.coli* - Cloning in Organisms other than *E.coli*: *Pseudomonas*, *Bacillus subtilis*, Yeast and Fungi – Two and three hybrid systems - Gene expression in Prokaryotes: merits and demerits - examples – Operon systems: lac, trp – Sigma factors – Transposons: Transposable elements.

Unit-III

Transformation in higher organisms: Plants - T-DNA and Transposon tagging – *Agrobacterium* mediated transformations – Alternate transformation strategies: chloroplast transformation etc. - Construction of Transgenic crop plants - Examples – Markers for Transgenics. Animal transgenics: DNA mediated transformation – Gene transfer by viral transduction – Viral vectors for Animal transgenics - Genetic manipulation of mammals – DNA transfer to other vertebrates – - Transgenic and gene knock out Technologies – Animal models for transgenic studies.

Unit-IV

Site-directed Mutagenesis - Fusion proteins: GFP and other examples. Genomic library - cDNA library: Screening Representative cDNA libraries – RT PCR – RACE PCR, DD-PCR. Site directed Mutagenesis: Screening recombinant clones. Functional genomics: Nucleic acid microarray and Real Time PCR. RNAi and Gene Silencing. – Genome analysis: DNA Sequencing and annotations: Methods and advancements - The Human Genome Project.

Unit-V

Gene Therapy: Different strategies for Gene Therapy, Gene therapy for inherited diseases, ADA, FH, Cystic Fibrosis, DMA, Neoplastic disorders, infectious diseases., DNA forensics, , Somatic Cell Gene therapy, Triple helix therapeutics, Antisense oligonucleotides, Ribozymes, Aptamers. Molecular Breeding of Plants – Production of interferons – DNA vaccines – Gene targeting: Applications in medicine, agriculture and industry.

Reference

1. Primrose. S.B., Twyman R.M., Old. R.W. (2001) Principles of Gene Manipulation. Blackwell Science Limited.
2. Molecular and cellular methods in Biology and Medicine, P.B. Kaufman, W.Wu, D.Kim and L.J. Cseke, CRC Press, Florida, 1995
3. Methods in Enzymology Vol.152, guide to molecular cloning Techniques, S.L. Berger and A.R.Kimmel, Academic Press, Inc. San Diego, 1998
4. Methods in Enzymology Vol 185, Gene Expression Technology, D.V. Goeddel, Academic Press, Inc., San Diego, 1990
5. DNA Science, A First Course in Recombinant Technology, D.A.Mickloss and G.A.Freyar, Cold Spring Harbor Laboratory Press, New York, 1990.
6. Molecular Biotechnology (2nd Edition), S.B.Primrose, Blackwell Scientific Publishers, Oxford, 1994.
7. Milestones in Biotechnology. Classic papers on Genetic Engineering, J.A. Davies and W.S. Reznikoff, Butterworth-Heinemann, Boston, 1992.
8. Route Maps in Gene Technology, M.R.Walker and R.Rapley, Blackwell Science Ltd., Oxford, 1997.
9. Genetic Engineering. An introduction to gene analysis and exploitation in Eukaryotes, S.M. Kingsman and A.J. Kingsman, Blackwell Scientific Publications, Oxford, 1998
10. Molecular Biotechnology – Glick
11. Human Molecular Genetics, Tom Strachan and Andrew P.Read, Bios Scientific Publishers, 1996.

Paper II - MOLECULAR GENETICS

Preamble

Scope: This paper in genetics has been structured to provide an in depth knowledge of the structural organization of the genome of various organisms, their principle of inheritance and their role in health and disease.

Objective: The major objective of this paper is to envisage a thorough knowledge regarding genetics and genome organizations in organisms.

Goal: This paper will help the student to get an insight on the genetic organization and an over all view of the genetic makeup; the significant differences between individuals, thereby its varied use.

UNIT I INTRODUCTION TO MOLECULAR GENETICS:

Principles of Mendelian inheritance, Chromosome structure and function: Gene structure. Genetic code, Genome organization and genetics of Bacteria, Actinomycetes, Viruses, Fungi, , Mycoplasma, protozoan and higher organisms. Genetic rearrangement: Organization of coding sequences and repetitive sequences .

UNIT II GENETICS IN INHERITANCE:

Human Chromosomes - Pedigree analysis, Mendalian traits and sex-linked traits in human - Extra chromosomal inheritance (Episomes, Mitochondria and Chloroplast), Genetic polymorphism, Mechanism of sex determination. Population genetics – Hardywein berg genetic equilibrium.

UNTI III CHROMOSOMAL DAMAGE:

The law of DNA constancy and C - value paradox; Numerical and structural changes in chromosomes: Molecular basis of spontaneous and induced mutations. Ames test for

mutagenesis. DNA damage and repair Holliday junction, gene targeting, gene disruption, FLP/FRT and Cre /Lox recombination, Rec A and other recombinases.

UNIT IV REGULATION OF GENE EXPRESSION IN PROKARYOTES AND EUKARYOTES:

Attenuation and anti termination, Operon concept: lac, trp, sigma factors. DNA methylation: Hetero chromatinization: Environmental regulation of genes - Transposons in Bacteria, Maize, Yeast. - Insertional elements.

UNIT V GENOME SEQUENCING

Genetic mapping - Molecular markers, RFLP, RAPD, SNP, DNA micro arrays - Human genome project - DNA finger printing: Applications of molecular markers in genetic polymorphism.

References

1. The science of Genetics by Alan G. Atherton, Jack. R, Girton, John. F, McDonald. Saunders college publishers.
2. Genes VII by Lewine
3. Molecular cell Biology, Darnell, Lodish, Baltimore, Scientific American Books, Inc., 1994.
4. Molecular and cellular Biology, Stephen L. Wolfe, Wadsworth Publishing company, 1993

Paper III GENOMICS AND PROTEOMICS

Preamble

Scope: Understanding the basics in the genome and will help to design and develop new drugs for the various human disorders and the treatment based on gene therapy

Objective: To expose the students to genome and protein structure and function at molecular level.

Goal: this paper provides the students an insight on various aspects of genome organization and protein structure and thereby its significance molecular mechanism behind it.

Unit – I

Overview : Genomes of bacteria . Archaea and Eucarya ; genome shape and topology ; chromatin , supercoiling and packaging ; study of genomes – mapping, genetic and physical mapping, single nucleotide polymorphisms and RFLPs.

Unit – II

Genome sequencing; chain termination and automated DNA sequencing; shotgun and gene cloning strategies; library construction; sequence assembly and gap closure; genome resources – NCBI Map viewer, ORF finder, Locuslink, etc.

Unit – III

Gene finding and functional annotation: sequence annotation and bioinformatics tools for genomics and genome comparison; Analyzing gene expression – DNA microarrays – design, analysis and visualization of data. RNA data handling/manipulation; using gene expression arrays for disease profiling.

Unit-IV

Protein structure and function – methods to quantitate proteins; densitometry and classical methods: proteomics methods; two dimensional gel electrophoresis. Mass spectrometry. Protein expression profiling, protein-protein interactions; RNA interference.

Unit – V

Genome mapping projects – human genome projects – methods and insights obtained; transcriptome analysis of *E. coli* and *S. cerevisiae*. Biochemical pathway databases – WIT and KEGG.

References

1. Genomes – T. A. Brown, Wiley – Liss Publications, 2002
2. Mount, David W. Bioinformatics sequence and genome analysis. Cold Spring Harbor Laboratories press, CSH New York, 2001
3. Liebler, Introduction to proteomics. Tools for the new biology. Humana Press 1st Ed. December, 2001
4. Proteomics: From protein sequence to function S. Pennington, M. J. Dunn Bios Scientific Publications Ltd. 2nd Ed. (Jan 15, 2001)

Paper IV - INTRODUCTION TO BIOINFORMATICS

Scope: This paper involves the study of computer assisted management of data that is generated for biotechnological application which helps to make use of data that are fast accumulating in massive databases.

Objective: To expose the students to study various databases and tools

Goal: It is prerequisite for the students to do research by using the various Data bases and bioinformatics tools for the analysis of biomolecules at the sequence level. Exposure to the tools and databases will help them to design research and their application

Unit – I

Principles of computing: Operating systems, application and advantages of Unix/Linux in bioinformatics. Basic word processing and database management soft wares.

Unit – II

Data acquisition and management: Types of data-DNA, RNA and protein sequences, protein structure data, gene and protein expression data. Databases: the public biological databases (Ex. Genbank, SWISSPROT, PDB, etc)- searching and retrieving data form databases- FASTA and BLAST and PHI-BLAST).

Unit – III

Sequence analysis with acquired data: Sequence comparison with pair wise and multiple sequence alignment. Deducing phylogenetic relationships from multiple sequence alignment. Building phylogenetic trees. Profiles and motifs.

Unit – IV

Bioinformatics in structure analysis: Format of a protein structure data. Retrieving protein structures from PDB. Using molecular visualization tools to study protein structure. Predicting protein structure from sequence by comparative modelling and other methods.

Unit – V

Genomics and proteomics: Use of micro arrays to study gene expression. Genome sequencing projects, 2D-PAGE as a tool in proteomics. Bioinformatics and drug discovery.

References

1. Bioinformatics-A beginner's guide by Jean – Michel Claverie and Cedric Notredame, Wiley- Dream Tech India Pvt. Ltd.
2. Developing bioinformatics computer skills by Cynthia Gibas and Per Jambeck, O' Reilly publications.
3. Introduction to bioinformatics by T.K. Attwood and D.J. Parry –smith, Pearson Education Asia.
4. Bioinformatics by David.W.Mount, CBS publishers and distributors.
5. Instant notes in bioinformatics by D.R. Westhead, J.H.Parish and R.M.Twyman.

Paper V. MICROBIAL STRAIN MANIPULATION

Preamble

Scope: This paper provides the thorough knowledge Isolation, Culturing and Maintenance of Microorganisms, their applications and products of industrial and commercial uses.

Objective: The major objective of this course is to provide basic knowledge on microbial strain manipulation for industrial and other uses.

Goal: This paper will help students to get an idea on the economic importance of microorganisms and new methods to product development by means of in vivo manipulation.

Unit I

Isolation and screening of microorganisms – strain selection and improvement methods; principles of microbial nutrition and media formulations for cell growth and product formation; factors influencing the choice of various carbon and nitrogen sources; vitamins, minerals, precursors and antifoam agents. Importance of medium pH and temperatures. Development of inocula for industrial fermentations

Unit II

Industrial and agricultural microbiology – microbial synthesis of commercial products, organic acids, alcohols, alcoholic beverages and industrial enzymes; Biofertilizers and bioinsecticides

Unit III

Food microbiology – incidence and types of microorganisms in food, meat, poultry, sea foods, vegetables, dairy products; production of baker's yeast, various types of cheese and other dairy products; SCP; indicators of food microbial quality and safety

Unit IV

Medical microbiology – vaccine production from microbes; antibiotics production; methods of isolation and identification of pathogenic organisms; antibiotic susceptibility testing

Unit V

Industrial production – problems in scaled up productions; optimisation of production using statistical packages; various operations involved in upstream and down stream processing

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 by P.F. Stanbury and A. Whitaker, Pergamon Press.
3. Microbial process development by H.W. Woelle, World Scientific.
4. Product recovery in bioprocess technology, by J. Krijgsman, BIOTOL, Butterworth-Heinemann.
5. Bioseparations: down stream processing for biotechnology by P.A. Belter, E.L.Cussler and W.S. Hu, Wiley Interscience.
6. Biotechnology Text book of Industrial Microbiology by W.Creuger and A.Creuger.

7. Industrial Microbiology by Casida.
8. Industrial Microbiology by Prescott
9. Comprehensive Biotechnology (Multivolumes) Editor M.Moo-Young, Pergamon Press.
10. Biotechnology Editor (Multivolumes) H.S. Rehm and G. Reed, Verlag Chomie Weinheim.

Paper VI - PLANT TRANSGENICS

Preamble

Scope: students will get detailed idea about genome organization in plants, basic techniques in genetic transformations in plants, metabolic engineering for products of industrial use.

Objective: To give an insight into the various important research areas, basic genetic tools regarding plant biotechnology .

Goal: This paper has been designed to give a comprehensive coaching in the research field for developing transgenic plants that benefit agricultural production, environment, nutrition and health.

UNIT – I

Genome organization in plants. Conventional plant breeding. Molecular Marker-aided Breeding: RFLP maps, linkage analysis, RAPD markers, STS, microsatellites, SCAR (sequence characterized amplified regions), SSCP(single strand conformational polymorphism), AFLP, QTL, map based cloning, molecular marker assisted selection. Arid and semi-arid technology. Green house and Green-home technology.

UNIT – II

Cell and tissue culture in plants; Tissue culture media(composition and preparation), Primary culture; cell line; Cell clones; Callus and suspension culture; Somoclonal variation; Micropropagation; Organogenesis; Somatic embryogenesis; transfer and establishment of whole plants in soil. Embryo culture and embryo rescue. Haploidy; Protoplast fusion and somatic hybridization; Cybrids; Anther, pollen and ovary culture for production of haploid plants and homozygous lines. Cryopreservation, slow growth and DNA banking for germ plasm conservation. Allopheny; Artificial seeds.

UNIT – III

Plant transformation technology: features of Ti and Ri plasmids, use of Ti and Ri as vectors, binary vectors, use of 35S and other promoters, genetic markers, use of reporter genes, reporter gene with introns, use of scaffold attachment regions, methods of nuclear transformation, viral vectors and their applications. Transgenic biology; role of virulence genes. Gene transfer methods in plants: multiple gene transfers, vector-less or direct DNA transfer. Transformation of monocots. Transgene stability and gene silencing. Chloroplast transformation: advantages, vectors, success with tobacco and potato.

UNIT - IV

Application of plant transformation for productivity and performance: herbicide resistance, phosphinothricin, glyphosate, sulfonamide, atrazine, insect resistance, Bt genes, Non-Bt like protease inhibitors, alpha amylase inhibitor, virus resistance, coat protein mediated, nucleocapsid gene, disease resistance, chitinase, 1-3 beta glucanase, RIP, antifungal proteins, thionins, PR proteins, nematode resistance, abiotic stress, post-harvest losses, long shelf life of fruits and flowers, use of ACC synthase, polygalacturonase, ACC oxidase, male sterile lines, bar and barnase systems, carbohydrate composition and storage, ADP glucose pyrophosphatase.

UNIT - V

Metabolic engineering and Industrial Products: plant secondary metabolites, control mechanisms and manipulation of phenylpropanoid pathway, shikimate pathway; alkaloids, industrial enzymes,

biodegradable plastics, polyhydroxybutyrate, therapeutic proteins, lysosomal enzymes, antibodies, edible vaccines, purification strategies, oleosin partitioning technology.

REFERENCES

1. An introduction to genetic engineering in plants, Mantel S.H, mathews J.A. Mickee R.A.1985. Blackwell Scientific Publishers. London.
2. In Vitro culture of higher plants by R.L.M. Pierik, 1987. Martinus Nijhoff Publisher, Dordrecht.
3. Plant cell culture. A practical approach. Second edition. Edited by R.A. Dixon and R.A. Gonzales.1994.Oxford University Press. Oxford.
4. Plant molecular biology by Donal Grierson and S.V. Convey.1984. Blackie & Son Limited. Newyork
5. Plant molecular genetics by Monica. A. Hughes.1999. Pearson Education limited, England.
- 6.Plant Biotechnology by mantell, S.H and Smith, H. 1983. Cambridge University press
- 7.Plants, genes and agriculture by M.J. Chrispeels and D.F. Sadava.2000.The American scientific Publishers
- 8.Practical Application of plant molecular Biology by R.J. Henry.1997. Chapamans and Hall.
9. Elements of Biotechnology, by P.K.Gupta.1996. Rastogi and Co. Meerut.
10. Plant Biotechnology by J.Hammond, P. Mc Garvey and V. Yusibov (Eds) 2000 Springer Verlag.
11. Plant cell and tissue culture in the production of Food ingredients by T.J. Fu, G-Singh, and W.R. Curtis Kluwer Academic/ Plenum Press
12. Biotechnology in crop improvement by H.S. Chawla.1998. International book distributor company.

Paper VII - ANIMAL TRANSGENICS

Preamble

Scope: The study of animal cells will help us gain an insight not only in the structure and function of cells and tissues but also in different physiological, biochemical and immunological processes. studies on gene regulation and novel techniques for gene manipulation helps novel product development in biomedical industry as well as other applications.

Objective: The major objective of this course is to provide basic techniques for gene manipulation and thereby help in understanding this interdisciplinary research area.

Goal: This paper will help students interested in careers in the field of veterinary or human health biotechnology.

Unit I

Animal cells: Culture media; Balanced salt solutions and simple growth medium, Physical, chemical and metabolic functions of different constituents of culture medium; Role of carbon dioxide, serum, growth factors, glutamine in cell culture; Serum and protein free defined media and their applications.

Unit II

Types of cell culture: primary and established culture; organ culture; tissue culture; three dimensional culture and tissue engineering; feeder layers; disaggregation of tissue and primary cell culture; cell separation; cell synchronization; cryopreservation.

Biology and characterization of cultured cells: tissue typing; cell-cell interaction; measuring parameters of growth; measurement of cell death; Apoptosis and its determination; cytotoxicity assays.

Unit III

Molecular techniques in cell culture: cell transformation; physical, chemical and biological methods; manipulation of genes; cell cloning and micro manipulation; hybridoma technology and its applications; cell fusion methods; gene mapping; vaccine production; gene therapy, targeting, silencing and knockout.

Unit IV

Embryology: Collection and preservation of embryos; culturing of embryos; Gametogenesis and fertilization in animals; types of cleavage pattern; role of maternal contributions in early embryonic development; genetic regulation of embryonic development in Drosophila; homeotic genes in development.

Unit V

Transgenics: Transgenic animal: production and application; transgenic animals as models for human diseases; transgenic animals in live-stock improvement; expression of the bovine growth hormone; transgenics in industry; chimera production; Ethical issues in animal biotechnology.

References

1. Animal cell culture; A practical approach, 4th Edition, by Freshney. R.I. John Wiley publication.
2. Methods in cell biology; Volume 57, Animal cell culture methods, Ed. Jennie P. Mather, David Barnes, Academic press.
3. Mammalian cell biotechnology; A practical approach, Ed. M. Butler, Oxford university press.
4. Exploring genetic mechanism; Ed. Maxine Singer and Paul Berg.
5. Principles of genetic manipulation; Ed. Old and Primrose, 6th Edition. Blackwell science publication.

Paper VIII - BIOETHICS, BIOSAFETY AND IPR

Preamble

Scope: This course has been designed to provide the students insights into the valuable areas of biotechnology, which plays a crucial role in determining its future use and applications.

Objective: Students get an idea about the advantages and disadvantages of biotechnological applications, ethical implications and intellectual property rights.

Goal: To study the diversity of plants and animal life in a particular habitat, ethical issues and potential of biotechnology for the benefit of man kind.

Unit I

Introduction to ethics/bioethics – framework for ethical decision making; biotechnology and ethics – benefits and risks of genetic engineering – ethical aspects of genetic testing – ethical aspects relating to use of genetic information – genetic engineering and biowarfare

Unit II

Ethical implications of cloning: Reproductive cloning , therapeutic cloning ; Ethical, legal and socio-economic aspects of gene therapy, germ line, somatic, embryonic and adult stem cell research- GM crops and GMO's – biotechnology and biopiracy – ELSI of human genome project

Unit III

Introduction to biosafety – biosafety issues in biotechnology – risk assessment and risk management – safety protocols: risk groups – biosafety levels – biosafety guidelines and regulations (National and International) – operation of biosafety guidelines and regulations – types of biosafety containment

Unit IV

Introduction to intellectual property and intellectual property rights – types: patents, copy rights, trade marks, design rights, geographical indications – importance of IPR - world intellectual property rights organization (WIPO)

Unit V

what can and what cannot be patented? – patenting life – legal protection of biotechnological inventions – Patenting in India: Indian patent act.

References:

1. Principles of cloning, Jose Cibelli, Robert P. Ianza, Keith H. S. Campbell, Michael D. West, Academic Press, 2002
2. <http://books.cambridge.org/0521384737.htm>
3. <http://online.sfsu.edu/%7Erone/GEessays/gedanger.htm>
4. http://www.actahort.org/members/showpdf?booknr=447_125
5. <http://www.cordis.lu/elsa/src/about.htm>
6. <http://www.csmt.ewu.edu/csmt/chem/jcorkill/bioch480/bioLN98.html>
7. <http://www.accessexcellence.org/AE/AEPC/BE02/ethics/ethintro.html>
8. <http://lawlib.samford.edu/cio/ciofeb03.pdf>
9. <http://www.biomedcentral.com/content/pdf/1472-6939-2-2.pdf>
10. http://lifesciences.cornell.edu/vision/accelerating_focus05.php
11. <http://thompson.com/libraries/fooddrug/>
12. <http://assets.cambridge.org/0521792495/sample/0521792495WS.PDF>
13. http://europa.eu.int/eurlex/pri/en/oj/dat/1998/l_213/l_21319980730en00130021.pdf
<http://www.clubofamsterdam.com/content.asp?contentid=281>
14. Biosafety issues related to transgenic crops, DBT guidelines, Biotech Consortium India Limited, New Delhi

SEMESTER I PRACTICAL I – BASICS OF GENETIC ENGINEERING

Scope: Getting trained in practical knowledge on these experiments would help the students in further research.

Objective: To expose the student in practical knowledge in advanced techniques in molecular biology and genetic engineering.

Goal: Upon completion of this course will emphasize the students to get the Practical knowledge this will help them doing the further research

S.No.	Title
1.	Preparation of Antibiotic selection media and competent
2	Isolation of plasmid DNA (pBS/pTaq) from <i>E.coli</i> .
3	Isolation of Genomic DNA from <i>Bacillus subtilis/thuringiensis</i>
4	Restriction analysis of plasmid/phage DNA
5	Elution of DNA from Agarose gel (Electro elution/Column/Alternate methods)
6	Cloning in <i>E.coli</i> : ligation and transformation

7	Gene expression and analysis of gene (cry/Taq polymerase) product
8	Western blotting
9	Production of His-tag protein and purification
10	Polymerase chain reaction and RAPD
11	Site directed mutagenesis
12	Sequence comparison through Bioinformatics (BLAST & FASTA)
13	Analysis of Protein structure in Silico (Swiss – PDB veiwser
14	Multiple sequence alignment
15	Phylogenetic tree construction

SEMESTER II
PRACTICAL 2 – APPLIED GENETIC ENGINEERING

1.	Isolation of DNA from blood
2	Preparation of primary cells from chick embryo
3	Cell counting and cell viability
4	Isolation of DNA from animal tissue by non enzymatic method
5	Liposome mediated transformation in animal cells
6	Isolation of RNA from blood
7	Superovulation in mice
8	Embryo (blastocyst) collection in mice / rat
9	Basics of plant tissue culture, preparation of media stocks and <i>in vitro</i> seed germination.
10	Induction of callus and suspension cultures
11	Protoplast isolation and viability test
12	Production of synthetic seeds.
13	Floral dip method of gene transfer in <i>Arabidopsis</i> .
14	<i>GUS</i> assay using histochemical method.
15	<i>Agrobacterium tumefaciens</i> mediated transformation
	Contd.
16	Southern blotting (Colony hybridization/Dot blotting)
17	rRNA library construction
18	Total genomic DNA isolation from plants by CTAB method
19	Isolation total RNA from plants by Trizol method
20	Analysis of transgenic plants by PCR.
21	cDNA synthesis by RT-PCR