

# UNIVERSITY OF RAJASTHAN JAIPUR

## **SYLLABUS**

# M.SC. Biotechnology

## Semester Scheme

III/IV Semester Examination

2017-2018

Dy. Registrar
(Academic)
University of Rajasthan

## Semester-III (2017-18)

### **BTH 901:- Genetic Engineering**

Genetic engineering tools and their applications: Restriction-modification system & enzymes, modification enzymes (methylases and other enzymes needed in genetic engineering), DNA and RNA markers. Gene Cloning Vectors- Plasmids, bacteriophages, phagemids, cosmids. Artificial chromosome vectors (YAC, BAC, MAC), CHEF analysis, virus derived vectors-SV40, M13, retroviral vectors, and general applications.

8

Nucleic Acid Sequencing and Amplification: Sequencing methods and their Applications-Maxim & Gilbert's and Sanger's methods; Pyrosequencing, Thermal PCR, Shot gun sequencing and Automated method. Nucleic Acid purification and Yield Analysis; PCR – Types and applications.

Gene manipulation: cDNA Synthesis and its Cloning; mRNA enrichment, DNA primers, linkers and adaptors, Library (cDNA and Genomic) construction and screening. Alternative Strategies of Gene Cloning- Two and three hybrid systems, cloning of genes in expression vectors and regulation, DNA microarrays.

10

Study Gene Regulation and analysis of gene Expression: DNA transfection methods, Northern blot, Primer extension, SI mapping, RNase protection assays, Reporter assays.

Southern and Western blotting, DNA fingerprinting, Chromosome walking, Southern and Fluorescence *in situ* hybridization;

3

Mutagenesis, Protein Engineering & Processing of Recombinant proteins - Directed Mutagenesis- Oligonucleotide with M13 DNA, PCR amplified oligonucleotide and Random mutagenesis. Protein Engineering: adding disulfide bonds, reducing number of free sulfhydril residues, changing aminoacids, increasing and modifying enzymatic activity. Processing of Recombinant proteins: Purification and refolding, characterization of recombinant proteins, stabilization of proteins.

T-DNA and Transposon Tagging: Role of gene tagging in gene analysis, T-DNA and Transposon tagging, Identification and isolation of genes through T-DNA or transposon. Transgenic and Gene Knockout Technologies Targeted gene replacement, Chromosome engineering.

Expression Strategies for Heterologous Proteins: Vector engineering, host engineering, in vitro transcription and translation, expression in hacteria, yeast, insects and insect cells, expression in mammalian cells and plants.

Gene Therapy-Vector engineering Strategies of gene delivery, gene replacement/

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Application of genetic engineering: UsesofTransgehic plants and animals; production of recombinant pharmaceuticals, disease diagnoses and nanotechnology.

#### **Suggested Laboratory Exercises**

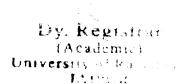
- 1. Growth characteristics of E. coli using plating and turbidometric methods. Growth
- 2. Bacterial culture and antibiotic selection on media.
- 3. Isolation of plasmid from E. coli by alkaline lysis method and its quantification by spectrophotometer.
- 4. Amplification of DNA by PCR.
- 5. Restriction enzyme digestion of genomic DNA and plasmid DNA from *E.coli* and estimation of size of DNA fragments after electrophoresis using DNA markers.
- 6. RFLP analysis
- 7. RAPD analysis
- 8. Demonstration of DNA fingerprinting. Humane Forensic
- 9. Restriction digestion of the plasmid and estimation of the size of various DNA fragments & Construction of Restriction digestion map.
- 10. Cloning of DNA fragment in a plasmid vector.
- 11. Transformation of the given bacterial population and selection of recombinants.
- 12. Co-cultivation of the plant material (e.g. leaf discs) with Agrobacterium and study GUS activity histochemically.
- 13. Any other practical based on theory syllabus.

#### Suggested Reading:

- 1. Sambrook, J., Fritsch, EF. and Maniatis, T. (2000). Molecular Cloning: A Laboratory Manual Cold Spring Harbor Laboratory Press, New York.
- Glover, DM. and Hames, BD. (1995). DNA Cloning: a practical Approach IRL Press Oxford.
- 3. Kaufman, PB., Wu, W., Kim, D. and Cseke, LJ. (1995). Molecular and cellular Methods in Biology and Medicine CRC Press.Florida.
- 4. Berger, SL. and Kimmel, AR. (1998). Guide to Molecular Cloning Techniques, Academic press Inc. San Dlogo.
- 5 (Gooddol DV (1990) Gene Uspression Technology Academic Press for San Diego 1990

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- 6. Mickloss, DA. and Greyer, GA. (1990). DNA Science A First Course in Recombinant Technology, Cold Spring Harbor Laboratory Press, New York.
- 7. Primorso, SB. (1994). Molecular Biotechnology (2nd Edn.), Blackwell Scientific Publishers, Oxford.
- 8. Davies, JA. andRoznikolf, WS. (1992). Milestones in Biotechnology. Classic papers on genetic Engineering Butterworth-Helnomann, Boston.
- 9. Walker, MR. and Repley, R. (1997). R<sub>Out</sub>e Maps in Gene Technology. Blackwell Science Ltd.Oxford.
- 10. Kingsman, SM. and Kingsman, AJ. (1998). Genetic Engineering: An Introduction to gene analysis and exploitation in eukaryotes. Blackwell Scientific blications.Oxford,1998.
- 11. Glick BR. and Thompson, JE. (1993). Methods in Plant Molecular Biology and Biotechnology. CRC Press, Boca Raton, Florida.
- 12. Glover, D.M. and Hames, B.D. (Eds.) (1995). DNA Cloning 1: A Practical Approach, Core Techniques, 2nd edition. pAS, IRL Press at Oxford University Press, Oxford.
- 13. Hackett, PB., Fuchs, JA. and Meesing, JW. (1988). An Introduction to Recombinant DNA Techniques: Basic Experiments in Gene Manipulation. Benjamin/Cummings Publishing Co., Inc. Menlo Park, California.
- 14. Glick, BR. And Pasternak, JJ. (1994). Molecular Biotechnology Principles and Applications of Recombinant DNA. Panima Publishing Corp, New Delhi.
- 15. Watson, JD., Gilman, M., Witkowski, J and Zollar, M. (1992). Recombinant DNA (Sec. Ed.). Scientific American Books, New York.



## BTH 902:- Animal Biotechnology

Tools and Culture Media: Equipments and materials for animal cell culture technology. Introduction to the balance salt solutions and simple growth medium. Brief account on the chemical, physical and metabolic functions of different constituents of culture medium. Role of carbon dioxide. Role of serum and supplements, Serum & protein free defined media and their application.

Basic understanding for cell culture: Structure and Organization of animal cell, Cell physiology. Primary and established cell line cultures. Biology and characterization of the cultured cells and measuring their growth.

Techniques of cell culture: Basic techniques of mammalian cell culture in vitro; disaggregation of tissue and primary culture; maintenance of cell culture; cell separation. Scaling-up of animal cell culture, measurement of viability and cytotoxicity. Cell synchronization. Cell cloning, micromanipulation and types of cloning. Stem cell culture, embryonic stem cells and their applications. Measurement of cell death. Apoptosis. Three dimensional culture and tissue engineering.

Mammalian Cell transformation: Establishment of Immortal cell lines, transfection, selection by selectable markers, gene amplification for high level protein expression. Specialized methods to transfer difficult cell types; Uses of viral vectors, Vaccinia and Baculovirus and Retrovirus in gene transfer; and use of antisense RNA and DNA in controlling gene function. Mice as the experimental material for gene introduction.

10

Impact of Recombinant DNA on human Genetics: Mapping and cloning human disease genes-Positional cloning, subchromosomal mapping and markers, in situ hybridization to chromosomes and RFLP.

Applications of Animal cell and Recombinant DNA technology: Cell culture based vaccines. Somatic cell genetics. Organ and histotypic cultures. Development of Transgenic animals (Mice, Cattle, Sheep, Goat, Pigs, Birds and Fish) and their uses. DNA- based diagnosis of genetic diseases: Human somatic cell gene therapy for single-gene disorders.

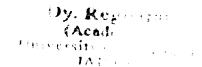
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Suggested Laboratory Exercises

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- 1. Preparation of tissue culture medium and membrane filtration.
- 2. Preparation of single cell suspension from spleen and thymus.
- 3. Cell counting and cell viability.
- 4. Macrophage monolayer from PEC, and measurement of pathogencity activity.
- 5. Trypsinization of monolayer and subculturing.
- 6. Cryopreservation and thawing.
- 7. Measurement of doubling time.
- 8. Role of serum in cell culture.
- 9. Preparation metaphage chromosome from cultured cells.
- 10. Isolation of and demonstration of apoptosis of DNA laddering.
- 11. MTT assay for cell viability and growth.
- 12. Cell fusion with PEG.
- 13. Any other practical based on theory syllabus

- 1. Watson, JD., Gilman, M., Witkowski, J and Zollar, M. (1992). Recombinant DNA (Sec. Ed.). Scientific American Books, New York.
- 2. Glick, BR. And Pasternak, JJ. (1994). Molecular Biotechnology Principles and Applications of Recombinant DNA. Panima Publishing Corp, New Delhi.
- 3. Froshney, RI. Culture of Animal Cells, (3rd Edition), Wiley-Liss.
- 4. Mesters, JRW. (Ed) Animal Cell Culture-Practical Approach,Oxford.
- 5. Basega, R. (Ed), Cell Growth and Division: A Practical Approach, IRL Press.
- Butler, M. & Dawson, M. (Eds) Cell Culture Lab Fax. Eds., Bios Scientific Publications Ltd. Oxford.
- 7. Martin Clynes. M. (Ed). Animal Cell Culture Techniques. Springer.
- 8. Jenni, Mathur P. and Barnes, D (Eds). Methods in Cell Biology, Vol. 57, Animal Cell Culture Methods. Academic Press.
- 9. Glick, BR. And Pasternak, JJ. (1994). Molecular Biotechnology Principles and Applications of Recombinant DNA. Panima Publishing Corp, New Delhi.
- Watson, JD., Gilman, M., Witkowski, J and Zollar, M. (1992). Recombinant DNA (Sec. Ed.). Scientific American Books, New York.
- 11 Kumar, IID (1998) Modern Concept of Biotechnology, VikasPublishing House, New Delin



## BTH 903:- Seminar, Scientific Writing& Presentations

## **Practicals through Assignments:**

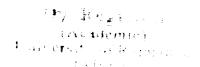
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- 1. Writing Review of Literature: Review of the Literature/Annotated Bibliography
  Locate, analyze, summarize, and evaluate at least 3 scholarly and 3 popular sources in
  an area of interest and produce an Annotated Bibliography/Review of the Literature
  using appropriate citation style, to support the Long Article (4-6 pages).
- 2. Data analysis and presentation: How to present data? Which diagrams to use? What programs and data formats to use for data visualization?
- 3. Types of graphs (Microsoft graphics 3D): Different graphs for different purposes (poster, talk, manuscript). Students will be able to correctly identify and describe four kinds of graphs commonly used in science: pie charts, line graphs, scatter plots, and bar graphs.
- 4. How to give a Seminar: introduction and background information on topic. What relevant research has been performed previously? State the problem(s) that remain unanswered. State objectives and specific hypotheses you wish to test. Describe the methodology used to test your hypotheses. Present Data ,Results, Discussion and Interpretations (fact vs. fiction) and explain the significance of your findings.
- 5. Scientific presentations/Poster/Power Point (short and long): Document Analysis Presentation (Information Literacy) Through multimedia presentation (PowerPoint or Poster Session) by analyzing selected articles and scholarly and popular science writing and journals. Use visuals. What does a good poster need? Structure of a good poster.

### Suggested Reading:

- 1. Scientific Writing and Communication: Papers, Proposals, and Presentations, 1st Edition, by Angelika Hoffman, ©2010 Oxford University Press, ISBN 978-0-19-539005-6
- 2. The Craft of Scientific Presentations: Critical Steps to Success and Critical Errors to Avoid by Michael Alley, ©2013 Springer, ISBN 978-1441982780
- 3. Bennett, B. 2001. The three P's of scientific talks: Preparation, practice, and presentation. Society for Economic Botany Newsletter. 15: 6-9.

can be found on line at .http://www.econbol.org/webmaster/oubs\_p&p/issues/2001\_spring.pdf



Examination		MM:-100
15	Presentation	50
	Poster Presentation (on Computer)	25
	Practical Record	15
10	Viva-Voce	10

## BTH A01:- Entrepreneurship & Ethics

Entrepreneurship: concept, definition, structure and theories of entrepreneurship. Types of start- ups. Types of entrepreneurship, environment, process of entrepreneurial development. Entrepreneurial culture, entrepreneurial leadership, product planning and development. Project management. Search for business idea. Concept of projects. Project identification, formulation, design and network analysis. Project report and project appraisal.

15

Ethical issues: introduction- causes of unethical acts, ignorance of laws, codes, policies and procedures, recognition, friendship, personal gains. Professional ethics- professional conduct. Ethical decision making, ethical dilemmas. Teaching ethical values to scientists, good laboratory practices, good manufacturing practices, laboratory accreditation.

A15

- Álvaro Cuervo, Domingo Ribeiro and Salvador Roig, 2007, Entrepreneurship Concepts, Theory and Perspective. Part II, 155-170
- 2. <u>Hannafey</u>, FT. (2004). Entrepreneurship and Ethics: A Literature Review. J. of . Bussiness Ethics, Volume 46, Number 2, 99-110
- 3. Hassan, E., Yaqub, O., Diepeveen, D. (2010). Intellectual Property and Developing Countries: A review of the literature, the RAND Corporation, 1776 Main Street, P.O. Box 2138, Santa Monica, CA 90407-2138
- 4. Krattiger et al (2007) "Intellectual Property Management in Health and Agricultural Innovation: A Handbook of Best Practices", Managing Innovation for a Better World
- 5. Hahn, RW. (2005). Intellectual Property Rights in Frontier Industries: Software and Biotechnology, AEI Press.
- 6. Miller, Raphael, Λ. and Michael HD. (2000) Intellectual Property: Patents, Trademarks, and Copyright. 3rd ed. New York: West/Wadsworth.
- 7. Creswell, J. (1998). Qualitative inquiry and research design: Choosing among five traditions. Thousand Oaks, California: Sage Publications.
- 8. Creswell, J. (2003). Research Design: Qualitative, Quantitative, and Mixed Methods Approaches. Thousand Oaks, California: Sage Publications.
- 9. John W. Creswell, 2009, Research Design: Qualitative, Quantitative, and Mixed Methods Approaches, Third Edition, www.sagepub.com. ISBN: 978-1-4129-6557-6

- Dahlia K. Remler, Gregg G., Van Ryzin, R. (2011). Research Methods in Practice, Strategies for Description and Causation. , <u>www.sagepub.com</u>, ISBN: 978-1-4129-6467-
- 11. Glenn, MacDonald, L. (2011). Ethical Issues in Genetic Engineering and Transgenics
- 12. McGee, G. "Primer on Ethics and Human Cloning" <a href="http://www.actionbioscience.org/biotech/mcgee.html">http://www.actionbioscience.org/biotech/mcgee.html</a>
- 13. "Primer on Ethics and Crossing Species Boundaries "http://www.actionbioscience.org/biotech/baylis\_robert.html
- 14. Grey, ST. "Genetic Engineering and Xenotransplantation" <a href="http://www.actionbioscience.org/biotech/grey.html">http://www.actionbioscience.org/biotech/grey.html</a>
- 15. Kolehmainen, S.M. "The Dangerous Promise of Gene Therapy" <a href="http://www.actionbioscience.org/biotech/kolehmainen.html">http://www.actionbioscience.org/biotech/kolehmainen.html</a>
- 16. Sherlock, R. and Morrey, JD. (2002). -Ethical issues in biotechnology, Rowman& Littlefield Publishers, Inc., Maryland.
- 17. <u>Paul B. Thompson</u> (2007). <u>Food biotechnology in ethical perspective</u>, The Springer, 2<sup>nd</sup> Ed., The Netherlands.
- 18. <u>Krishna R. Dronamraju</u>, (2008). Emerging consequences of biotechnology: biodiversity loss and IPR issues. World Sc. Publ. Co. Pvt. Ltd., Singapore.

## **BTH B02:- Protein Engineering**

Structure of Proteins and Prediction: Overview of protein structure, PDB, structure based classification, databases, visualization tools, structure alignment, domain architecture databases, protein-ligand interactions. Primary structure and its determination, secondary structure prediction and determination of motifs, profiles, patterns, fingerprints, super secondary structures, protein folding pathways, tertiary structure, quaternary structure, methods to determine tertiary and quaternary structure, post translational modification.

12

Introduction to Protein engineering: definition, basic principles; Features or characteristics of protein that can be engineered (definition and methods of study), affinity and specificity; Spectroscopic properties; Stability to changes in parameters as pH, temperature and amino acid sequence, aggregation propensities etc.

8

Method for Protein engineering: Rational design, Directed mutagenesis, Random mutagenesis, DNA shuffling, Evolutionary methods/directed evolution, Homology modelling, De novo enzyme engineering, strategies and case studies: Addition of disulfide bonds- T4 Lysozyme, Xylanase, Human pancreatic Ribonuclease; changing asparagine to other amino acids, reducing the number of free sulphydryl residues, increasing enzyme activity, modifying metal cofactor requirements, decreasing protease sensitivity, modifying protein specificity FokI endonuclease, Antibodies; increasing enzyme stability and specificity-altering multiple properties (Subtilisin, Peroxidase).

15

Computational approaches to protein engineering: sequence and 3D structure analysis, bioinformatics tools for protein study, Data mining, Ramachandran map, Mecahnism of stabilization of protein from psychrophiles and thermophiles vis-a-vis those from mesophiles; Protein design.

16

Application of Protein engineering: Food and detergent industry applications, Environmental challenges and protein engineering, therapeutic protein production, antibody modelling, biopolymer production, applications in Nanobiotechnology, Applications with redox proteins and enzymes, industrially important enzymes, role in gene regulation, Other new applications

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#### Reference book

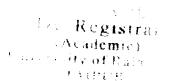
- 1. Carl Brandon & John Tooze, "Introduction to Protein Structure," "2nd Edition" Garland Publishing, 1999
- 2. Paul R. Carey, "Protein Engineering and Design," Academic Press, 1996.
- 3. Daniel C. Liebler, "Introduction to Proteomics Tools for the New Biology," Humana Press, 2001
- 4. C Branden, T. Tooze. 1999. Introduction to Protein Structure (2nd Ed.), Garland Science, Taylor and Francis Group, New York, USA.
- 5 T.F. Creighton 2002 Proteins Structures and Molecular Properties (3rd Ed.), W.H. Freeman and Company, New York, USA

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- 6. R. H. Pain. 2000. Mechanisms of Protein Folding, Oxford University Press, Oxford, England. 5. J. Cavanagh, W.J. Fairbrother, A.G. Palmer III, M. Rance, N. J. Skelton. 2007.
- 7. S. Lutz, U. T. Bornscheuer. 2008. Protein Engineering Handbook, Wiley-VCH, Weinheim, Germany.
- 8. D. W. Mount. 2004. Bioinformatics: Sequence and Genome Analysis, Cold Spring Harbor Laboratory, Plainview, New York, USA.
- 9. V. N. Uversky, A.L. Fink. 2006. Protein Misfolding, Aggregation and Conformational Diseases: Part A: Protein Aggregation and Conformational Diseases (Protein Reviews), Springer, New York, USA.
- 10. M. Zvelebil, J.O. Baum. 2007. Understanding Bioinformatics (1s Ed.), Garland Science, Taylor and Francis Group, New York, USA.

#### Lab Exercise for Protein Engineering:-

- 1. Isolation and purification of protein
- 2. SDS PAGE
- 3. Demonstration of Mass spectroscopy MOLDI TOF
- 4. Gel filtrations chromatography
- 5. Affinity chromatography
- 6. To find out capacity & nature of the given ion exchange resin
- 7. Effect of pH, temperature on activity or stability of protein
- 8. Protein structure prediction by bioinformatics
- 9. Protein structure prediction and classification
- 10. Application of Bioinformatics tools in support of protein research
- 11 Searching protein sequence databases
- 12. Protein structure visualization
- 13. Secondary structure prediction
- 14 Protein structure prediction
- 15. Structural prediction through homology modelling
- 16. Ant other exercise based on theory paper content



## **BTH C02:- Proteomics and Genomics**

Genomics: Genome sequencing strategies and programs, new technologies for highthroughput sequencing, methods for sequence alignment and gene annotation; Approaches to analyze differential expression of genes - ESTs, SAGE, microarrays and their applications; gene tagging; gene and promoter trapping; knockout and knock-down mutants; dynamic modulation of protein structure and function, chip in chip.

15

Genome and Gene databases, Brief Outlook of Various Plant Genome Projects and their Outcome (Arabidopsis, Tomato, Potato, Rice), Non-coding F.NAs, Transcriptomics, RNA interference and gene silencing, genome imprinting, small RNAs-biogenesis and functions, role of small RNAs in heterochromatin formation and gene silencing, tools to study methylome and histone modifications Humane and Mouse.

15

Proteomics: Analysis of proteins by different biochemical and biophysical procedures like CD (Circular Dichroism), NMR, UV/Visible and fluorescent spectroscopy, protein identification and analysis on ExPASy server, other protein related databases, 1-D and 2-D gel electrophoresis for proteome analysis, Sample preparation, gel resolution and staining.

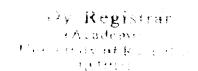
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Mass spectrometry based method for protein identification like PMF (protein mass fingerprinting) and LCMS, (Image analysis of 2D gels) Data acquisition, spot detection & quantitation, gel matching, data analysis, presentation, databases, conclusions; DIGE (Differential In Gel Electrophoresis), alternatives to 2-DE for protein expression analysis; Analysis of post-translational modifications and protein-protein interactions; protein chips and arrays, future directions in proteomics, scope of functional proteomics, and Protein databases, Protein-protein interaction, Interactome.

15

#### **Practical Exercises:**

- 1. Demonstration and listing of sequence retrieval online tools.
- 2. Demonstration and listing of sequence submission online tools.
- 3. Listing and demonstration of Protein and DNA Sequence Databases and their utilities.
- 4. Demonstration of DNA and Protein Array Technology and applications.
- 5. Reverse transcription-PCR to examine gene expression.
- 6. Real-time PCR to quantify gene expression.
- 7. Northern and Western Blotting analysis.
- 8. Demonstration of Instrumentation (MALDI/TOF, LC-MS-MS, 2DGE) by visit or audio-visual medium



- 9. Protein separation techniques (Chromatography-Ion-Exchange, Gel Filtration, Affinity; Ultrafiltration, Recombinant protein separation techniques).
- 10. Comparison of Next-generation sequencing methods (by Chart/ poster preparation).
- 11. Any other exercises designed by course teacher as per the syllabus.

#### **SUGGESTED READINGS:**

- 1. Buchanan B, Gruissem G, and Jones R (2000) Biochemistry and Molecular Biology of Plants, American Society of Plant Physiologists, USA.
- 2. Hammes GD (2005) Spectroscopy for the Biological Sciences; Wiley Interscience, USA.
- 3. Harlow and Lane D (Eds.) (1988) Antibodies A Laboratory Manual; Cold Spring Harbor Laboratory, USA.
- 4. Lieber DC (2006) Introduction to Proteomics: Tools for New Biology; Humana Press, NJ.
- 5. Pennington SR, Dunn MJ (Eds.) (2002) Proteomics: From Protein Sequence to Function, BIOS Scientific Publishers, United Kingdom.
- 6. Sambrook J and Russell DW (2001). Molecular Cloning A Laboratory Manual, Vols I III, Cold Spring Harbor Laboratory, USA.
- 7. Singer M and Berg P (1991). Genes and Genomes: A Changing Perspective; University Science Books, CA, USA.

## M.Sc. Biotechnology Semester-IV (2017-18)

## BTH X01:- Plant Biotechnology

Plant tissue culture: Principles, Concept, History of development of plant tissue culture, Concept of totipotency, PTC laboratory facilities, operation and management, General methodology. Different PTC media and their nutritional components, media preparation and sterilisation techniques. aseptic techniques and preparation of explants, histological techniques for plant tissue culture. Cryopreservation and slow growth for germplasm preservation.

Plant tissue culture technology: Shoot morphogenesis and organogenesis, rooting, hardening and field transfer; Micropropagation, production of virus free plants, callus and suspension cultures, single cell culture. Ovary, anther and microspore culture for production of haploid plants. Somatic embryogenesis, synthetic seeds and its cryopreservation. Plant tissue culture as a technique to produce novel plants, somaclonal variations. Overview of Plant Tissue Culture Applications.

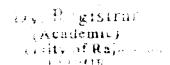
**Protoplast technology**: Protoplast isolation, purification, viability tests, plating efficiency, culture, Somatic cell hybridization, selection of hybrid, cybrids and their regeneration.

6

Plant transformation (Recombinant DNA) technology: Tools and techniques, Vectors for plant transformation (Viral and Bacterial), Basic molecular characteristics of Agrobacterium, Basis of tumor and hairy-root formation, Characteristic features of vectors (Co-integrative and binary vectors, Ti, Ri plasmids, 35S and other promoters and terminators, selectable markers, reporter genes, origin of replication etc.)

7

Agrobacterium-mediated plant transformation: Cloning of selected gene, its integration into Agrobacterium. Agrobacterium-mediated gene transfer - mechanism of T-DNA transfer and its integration into plant genome, role of virulence gene, selection of transformed cells/



tissues, expression of the integrated gene in plants. Multiple gene transfer. Practical applications of Agrobacterium-mediated gene transfer.

Direct gene transfer methods: Particle bombardment, electroporation and micro injection.

Transgenic gene incorporation, stability and expression; gene silencing. Cryopreservation and Genebanks.

Plant Breeding: Brief idea about conventional Plant Breeding Methods- Character identification, incorporation (hybridization), selection and release of variety; Role of Molecular markers: RFLP, RAPD, STS, SCAR, SSCP, AFLP in plant breeding applications. Green house and green-home technology.

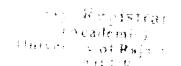
Transgenic approaches to crop improvement: Resistant against biotic (virus, fungi, bacteria, nematode, insect, weed) and abiotic stress (salinity, drought, herbicide, cold, metals), longer shelf life. Improvement of crop yield and quality - golden rice and other developments. Extension of flower life, pigmentation and fragrance.

8

Manufacture of valuable products: Industrial applications of plant cell culture; Plant cell culture and biosynthesis of secondary products; Manufacture of - antigens, antibodies, edible vaccines, enzymes, proteins.

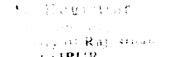
#### Suggested Laboratory Exercises:

- 1. Preparation of Stock solutions for MS medium.
- 2. Preparation of medium.
- 3. Micro propagation technique
- 4. Surface sterilization and Organ culture.
- 5. Callus induction, propagation, and differentiation
- 6. Organogenesis- Shoot and root formation and their organic connection.
- 7. Hardening and transfer of plants to soil.
- 8. Study of somatic embryogenesis.
- 9. Anther culture, production of Haploids.
- 10. Ovary culture
- 11 Somatic embryogenesis using appropriate explants and Preparation of synthetic seeds
- 1.7 Protoplast isolation and cutture



- 13. Demonstration of protoplast fusion employing PEG.
- 14. Cytological examination of regenerated plants.
- 15. Isolation & Identification of Sec. metabolite from Plant Cell Cultures.
- 16. Agrobacterium culture, selection of transformants, reporter gene (GUS) assays.
- 17. Any other practical based on theory syllabus

- 1. Bhojwani, S.S. and Razdan, M.K. (1996). Plant Tissue Culture: Theory and Practice (a revised edition). Elsevier Science Publishers, New York. USA.
- 2. Slater A, Scott N, Fowler M (2010). Plant biotechnology: the genetic manipulation of plants. Oxford: Oxford University Press.
- 3. Hammond, J. McGarvey P. and Yusibov V.(Eds.) (2000). Plant Biotechnology. Springer Verlag. Germany.
- 4. Fu,T –J., Singh, G. and Curtis, WR. (Eds) (1999). Plant Cell and Tissue Culture for the Production of Food ingredients. Kluwer Academic/Plenum Press.
- 5. Chawla, HS. (1998). Biotechnology in Crop improvement. International Book Distributing Company.
- 6. Henry, RJ. (1997). Practical Application of plant Molecular Biology. Chapman and hall.
- 7. Butenko, RG. (2000). Plant Cell Culture, University Press of Pacific.
- 8. Collin, H.A. and Edwards, S. (1998). Plant Cell Culture. Bios Scientific Publishers, Oxford, UK.
- 9. Dixon, RA. (Ed.) (1987). Plant Cell Culture: Practical Approach. IRL Press, Oxford.
- 10. George, EF. (1993). Plant Propagation by Tissue Culture. Part 1. The Technology, 2nd edition. Exegetics Ltd., Edington, UK.
- 11. Hall, RD. (Ed.) (1999). Plant Cell Culture Protocols. Humana Press, Inc., New Jersey, USA.
- 12. Shaw, CH. (Ed.) (1988). Plant Molecular Biology: A Practical Approach, IRL Press, Oxford.
- 13. Smith, RH. (2000). Plant Tissue Culture: Techniques and Experiments. academic press, New York.
- 14. Kumar, A. and Roy, S. (2006). Plant Biotechnology & is applications in Tissue Culture. I.K. International Pvt. Ltd.
- 15 Kumar, A. and Roy, S. (2011). Plant Tissue Culture and Applied Biotechnology, Aavishkar Publishers, Jaipan.
- 16 Mascarenhas, Ab. (1991). Handbook of Plant Tusius, Culting, ICAR, New Delhi.



- 17. Ramawat, KG. (2000). Plant Biotechnology, S. Chand & Co. Ltd. New Delhi.
- Rajdan, MK. (1993). An Introduction to Plant Cell Culture. Oxford & IBH Publishing
   Co. Pvt. Ltd. New Delhi.
- 19. Narayanaswamy, S. (1994). Plant Cell and Tissue Culture. Tata McGraw-Hill Pub. Com. Ltd. New Delhi.
- 20. Ammirato, PV, Evans, DA, Sharp, WR. And Yamada, Y. (1984). Hand Book of Plant Cell Culture, Vol. 1-3, Macmillan Pub. Co. NY & Collier Macmillan Pub. London.
- 21. Gupta, PK. (2010). Plant biotechnology, Rastogi Pub. Meerut.
- 22. Natesh,S, Chopra, VL. And Ramachandran, S. (1987). Biotechniques in Agriculture. Oxford & IBH Publishing Co. Pvt. Ltd. New Delhi.

## BTH X002:- IPR & Biosafety

Introduction to Intellectual Property: Types of IP: Patents, Trademarks, Copyright & Related Rights, Industrial Design, Traditional Knowledge, Geographical Indications, International framework for the protection of IPIP as a factor in R&D; IPs of relevance to Biotechnology and few Case Studies; Introduction to History of GATT, WTO, WIPO and TRIPS

#### Concept of 'prior art'

Invention in context of "prior art"; Patent databases; Searching International Databases; Country-wise patent searches (USPTO, EPO, India etc.); Analysis and report formation Basics of Patents:-

Types of patents; Indian Patent Act 1970; Recent Amendments; Filing of a patent application; Precautions before patenting-disclosure/non-disclosure; WIPO Treaties; Budapest Treaty; PCT and Implications; Role of a Country Patent Office; Procedure for filing a PCT application

#### Patent filing and Infringement

Patent application- forms and guidelines, fee structure, time frames; Types of patent applications: provisional and complete specifications; PCT and convention patent applications; International patenting-requirement, procedures and costs; Financial assistance for patenting-introduction to existing schemes; Publication of patents-gazette of India, status in Europe and US Patenting by research students, lecturers and scientists-University/organizational rules in India and abroad, credit sharing by workers, financial incentives Patent infringement- meaning, scope, hitigation, case studies and examples

#### **Biosafety**

Introduction; Historical Backround; Introduction to Biological Safety Cabinets; Primary Containment for Biohazards; Biosafety Levels; Biosafety Levels of Specific Microorganisms; Recommended Biosafety Levels for Infectious Agents and Infected Animals; Biosafety Protection of New GMOs; guidelines - Government of India; Definition of GMOs & LMOs; Roles of Institutional Biosafety Committee, RCGM, GEAC etc. for GMO applications in food and agriculture; Environmental release of GMOs; Risk Analysis; Risk Assessment; Risk management and communication; Overview of National Regulations and relevant International Agreements including Cartagena Protocol.

#### **Important Links**

http://www.w3.org/IPR/

http://www.wipo.int/portal/index.html.en

http://www.ipr.co.uk/IP\_conventions/patent\_cooperation\_treaty.html www.patentoffice.nic.in www.iprlawindia.org/ - 31k - Cached - Similar page

http://www.cbd.int/biosafety/background.shtml

http://www.cdc.gov/OD/ohs/symp5/jyrtext.htm

http://web.princeton.edu/sites/ehs/biosafety/biosafetypage/section3.html

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## BTH X003:-Dissertation & Industrial Training

The project work will involve in depth practical work on a problem suggested by the supervisor of the candidate. The evaluation of the dissertation will be done by the external examiner and carry 50 marks. The dissertation submitted by the candidate shall be evaluated by one external expert, Head of the department and supervisor of the candidate. The seminars, in-plant training and industrial visit reports will also be submitted by the candidate to the Head of the Department who will submit these to the external examiner. The examination shall be held in the department and the dissertation etc. will NOT be required to be mailed to the external examiner. The distribution of the marks will be as under:

$\mathcal{M} \cdot \mathcal{M}$ .	<i>G0</i> /
Disseration Report	20
Evaluation (PPT & VIVE Voce)	50

## **BTH A01:-VIROLOGY**

Vaccines - 4 Credits

#### Innate Immunity;

Activation of the Innate Immunity through TLR mediated signaling; Adaptive Immunity; T and B cells in adaptive immunity; Immune response in infection; Protective immune response in bacterial; Viral and parasitic infections; Correlates of protection

#### Vaccination and immune response;

Appropriate and inappropriate immune response during infection: CD4+ and CD8+ memory T cells; Memory B cells; Generation and Maintenance of memory T and B cells; Dendritic cells in immune response

#### Adjuvants in Vaccination;

Induction of Th1 and Th2 responses by using appropriate adjuvants; Microbial, Liposomal and Microparticles as adjuvant; Chemokines and cytokines; Role of soluble mediators in vaccination; Oral immunization and mucosal Immunity

#### Conventional vaccines;

Bacterial vaccines; Live attenuated and inactivated vaccine; Subunit Vaccines and Toxoids; Peptide Vaccine

#### New Vaccine Technologies;

Rationally designed Vaccines; DNA Vaccination; Mucosal vaccination; New approaches for vaccine delivery; Engineering virus vectors for vaccination; Vaccines for specific targets; Tuberculosis Vaccine; Malaria Vaccine; HIV vaccine

#### Texts/References

- 1. Edited by Stefan H.E. Kaufmann, Novel Vaccination Strategies, Wiley-VCH Verlag GmbH & Co. KgaA, 2004 or later edition.
- 2 Topley& Wilson's, Microbiology and Microbial Infections Immunology Edited by Stefan H.E. Kaufmann and Michael W Steward Holder Arnold, ASM Press, 2005 or later edition.
- 3. Edition Charles A Janeway. Jr, Paul Travers, Mark Walport and Mark J Shlomchik, Immuno Biology, The Immune system in health and Disease, 6th Edition, Garland Science, New York, 2005 or later edition.
- 4. Annual Review of Immunology: Relevant issues 5. Annual Review of Microbiology: Relevant issues

Suggested Brechal : Based on Many Syllabus

## BTH B02:- Applied Environmental Biotechnology

Credit: 4

Bioremediation & Phytoremediation: Biofeasibility, applications of bioremediation, Bioreduction, Phytoremediation.

Bioabsorption and Bioleaching of heavy metals: Cadmium, Lead, Mercury, Metal binding targets and organisms, Bioabsorption, Metal microbial interaction, Biomethylation of elements (Methylation of mercury and arsenic), Commercial biosorbants, bioleaching, metal precipitation, advantages and disadvantages of bioleaching.

Waste water Treatment: Biological treatment system (Oxidative ponds, aerobic and anaerobic ponds, facultative ponds, aerated ponds), Biological waster treatment, activated sludge treatment, microbial pollution in activated sludge, percolating filters, waster water treatment by biofilms. Treatment scheme of Dairy, Distillery, Tannery, Sugar, Fertilizers, Refinery, Chemical and Antibiotic waste.

Solid waste pollution and its management: Current practice of solid waste management, composting systems, vermicomposting, sewage treatment. Anaerobia digestion, Land Filling.

#### PRACTICALS—CREDIT 2

Visit to

Sewage/waste water treatment plants, Farms doing floriculture, vegetable farming, Dairy/sugar/dyc industry treatment plants Krishivaigyanikkendras for composting Report writing of various visits.

#### REFERENCES---

- 1. INDUSTRIAL WASTE WATER TREATMENT (2008) by A D PATWARDHAN, Prentice hall of India, New Delhi.
- 2. APPLIED BIOREMEDIATION AND PHYTOREMEDIATION(2004) Ed by AJAY SINGH and OWEN P WARD, SPRINGER
- 3. THE COMPLETE BOOK ON ORGANIC FARMING AND PRODUCTION OF ORGANIC COMPOST(2008) By NPCS BOARD OF CONSULTANTS AND ENGINEERS ASIA PACIFIC BUSINESS PRESS Inc.
- 4. The Complete Technology Book on Biofertilizer and Organic Farming (2nd Revised Edition) [NI 15] by NIIR Board.
- 5. The Complete Technology Book on Vermiculture and Vermicompost [NI116] by NPCS Board of Consultants and Engineers,
- 6 Biopesticides Handbook [NI210] by NPCS Board of Consultants & Engineers,
- 7 Manufacture of Biofertilizer and Organic Farming [N1237] by H. Panda,
- 8 Integrated Organic Farming Handbook [NI248] by Dr. Li, Panda.

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## BTH C03:- Nano Biotechnology

Introduction: History- The purple of Cassius, the Lycurgus cup, Michael Faraday and "divided metals". Richard Feynman, Itis theory and predictions, Moore's Law, Miniaturization of microprocessors., the story of the Damascus sword. What is "nano'? Comparison with familiar objects. From nanoscience to nanotechnology: Eric Drexler.

5

Nanoscience: The multidisciplinary science, Bottom-up and top-down approach of research with examples, Different types of Nanonaterials, one-dimensional: CNTs, its types and characteristics; Two-dimensional: Nanofilms, nanosheets, nanowalls.

Properties of Nanomaterials, Different types of Nanomaterials and their Size, Quantum effects. Seeing Nanomaterials: Microscopes (SEM, TEM,STM, AFM) How do the different types of microscope work with Nanomaterials. Preparation of Nanomaterials: Physical Method (hydrothermal and solvothermal) Chemical methods, Biological methods (Green synthesis using plants, microbes & other living organisms).

10

Nanobiotechnology: Nanomedicine, Nanocosmetics, Textiles, Nanosensors (Biological, chemical, Biosensors, Gas sensors, mechanical), Drug delivery, Cancertherapy, Tissue engineering, water purification, Lab-on-a-chip (LOC), nanocomputers, DNA computer, MRI with magnetic nanoparticles. Current trends of research in Nanobiotechnology, particularly health sciences.

Effect of Nanomaterials on growth and development of plants: Biouptake, Localization, and transformation of nanoparticles within plants. Nanoagriculture for sustainable agricultural crop producton, application and Perspectives, Nanoparticles for herbicide, pesticides and fertilizers delivery, Role of Nanoparticles in photosynthesis, Mode of action of nanomaterials at gene level, Nanotoxicity and its mechanism. Safety Issues. Current trends of research in Nanotechnology pertaining to plant sciences.

15

- 1. ManasiKarkare.Nanotechnology: Fundamentals and Applications.2008.

  1. K.International
- 2. K. Eric Drexler, Chris Peterson and Gayle Pergamit Unbounding the future: The Nanotechnology Revolution. 1991. William Morrow and Company, Inc., New York.
- 3 C N R Rao Nanoworld: An Introduction to nanoscience and Technology.2010.

  Jawaharlal Nehru Centre for Advanced Scientific Research, Bangalore.
- 4 Manzer H. Siddiqui, M.H. Al Whabhi, F. Mohammad (Editors). Nanotechnology and Plant Sciences. 2015. Springer.
- 5 C.M. Niemeyer and C.A. Mirkin, Nanobiotechnology, 2012. Wiley-VCH
- 6. CM Niemeyer and CA Makin Nanobiotechnology 41, 2012. Wiley VCII.



#### Suggested Exercises:

- 1. Synthesis of Nanoparticles by physical method & Chemical Method.
- 2. Green synthesis of Nanoparticles using Plant system from Leaves, fruit, callus etc.
- 3. Characterization of Nanoparticles using UV Visible Spectroscopy, XRD, FTIR.
- 4. Visualization of Nanomaterials using SEM, TEM, AFM.
- 5. To check the bioactivity of Nanomaterials on various pathological Fungi and Bacteria.
- 6. To check the effect of Nanomaterials on Plant germination parameters.

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