

OBJECTIVES: To enable the students to

1. Understand the basic principles of plant tissue culture
2. Understand the methods in biotechnology
3. Get an insight into Recombinant DNA technology and Methods of gene transfer.
4. Appreciate the applications of Biotechnology

COURSE:

Unit I: PLANT TISSUE CULTURE – 1

1. History of plant tissue culture research - basic principles of plant tissue callus culture, meristem culture, organ culture, Totipotency of cells, differentiation and dedifferentiation.
2. Methodology - sterilization (physical and chemical methods), culture media, Murashige and Skoog's (MS medium), phytohormones, medium for micro-propagation/clonal propagation of ornamental and horticulturally important plants.
3. Callus subculture maintenance, growth measurements, morphogenesis in callus culture – organogenesis, somatic embryogenesis.

UNIT-II: Plant Tissue culture -2

1. Endosperm culture – Embryo culture -culture requirements – applications, embryo rescue technique.
2. Production of secondary metabolites.
3. Cryopreservation; Germ plasm conservation.

Unit III: Recombinant DNA technology

1. Restriction Endonucleases (history, types I-IV, biological role and application); concepts of restriction mapping.
2. Cloning Vectors: Prokaryotic(pUC 18, pBR322,Ti plasmid and Lambda phage, Eukaryotic Vectors (YAC and briefly PAC)
3. Gene cloning (Bacterial Transformation and selection of recombinant clones, PCR mediated gene cloning)
4. Construction of genomic and cDNA libraries, screening DNA libraries to obtain gene of interest by complementation technique, colony hybridization.

Unit IV: Methods of gene transfer

1. Methods of gene transfer- Agrobacterium-mediated, direct gene transfer by Electroporation, Microinjection, Micro projectile bombardment.
2. Selection of transgenics– selectable marker and reporter genes (Luciferase, GUS, GFP).

Unit V: Applications of Biotechnology

1. Applications of Plant Genetic Engineering – crop improvement, herbicide resistance, insect resistance, virus resistance.
2. Genetic modification – transgenic plants for pest resistant (Bt-cotton); herbicide resistance (Round Up Ready soybean); improved agronomic traits - flavrSavr tomato, Golden rice); Improved horticultural varieties (Moon dust carnations)

Text Book:

Botany-Plant tissue culture and its biotechnological applications, by B. R. C. Murthy & V. S. T. Sai, Venkateswara Publications, Guntur, 2017

Books for Reference:

1. Pullaiah. T. and M.V.Subba Rao. 2009. Plant Tissue culture. Scientific Publishers, New Delhi.
2. Bhojwani, S.S. and Razdan, M.K., (1996). Plant Tissue Culture: Theory and Practice. Elsevier Science Amsterdam. The Netherlands.
3. Glick, B.R., Pasternak, J.J. (2003). Molecular Biotechnology- Principles and Applications of recombinant DNA. ASM Press, Washington.
4. Bhojwani, S.S. and Bhatnagar, S.P. (2011). The Embryology of Angiosperms. VikasPublicationHouse Pvt. Ltd., New Delhi. 5th edition.
5. Snustad, D.P. and Simmons, M.J. (2010). Principles of Genetics. John Wiley and Sons, U.K. 5th edition.
6. Stewart, C.N. Jr. (2008). Plant Biotechnology & Genetics: Principles, Techniques and Applications. John Wiley & Sons Inc. U.S.A.

Suggested Activities: In vitro initiation of callus on artificial medium, seminars on utilization of rDNA technology, debates on applications of Biotechnology (whether it is a boon or bane to the society) studying growth patterns, vegetative characteristics of Bt.cotton and identifying the features of its pest resistance

ST. JOSEPH'S COLLEGE FOR WOMEN (AUTONOMOUS) VISAKHAPATNA
 VI SEMESTER **BOTANY** TIME: 2Hrs/week
 B () **Cluster Paper VIII:A2** Max. Marks: 50
Plant Tissue Culture & Plant Biotechnology
 W.e.f. 'U' (2017 – 2018) Batch **PRACTICAL VIII**

COURSE:

1. (a) Preparation of MS medium.
 (b) Demonstration of in vitro sterilization methods and inoculation methods using leaf and nodal explants of Tobacco/ Datura/ Brassica etc.
2. Study of embryo and culture, micro propagation of Banana, somatic embryogenesis, artificial seeds through photographs.
3. Construction of restriction map of circular and linear DNA from the data provided.
4. Study of methods of gene transfer through photographs: Agrobacterium-mediated, direct gene transfer by electroporation, microinjection, and micro projectile bombardment.
5. Different steps involved in genetic engineering for production of Bt. cotton, Golden rice, Flavr Savr tomato through photographs.
7. Isolation of plasmid DNA.
8. Restriction digestion and gel electrophoresis of plasmid DNA (optional)
9. Field visit to a lab involved in tissue culture
10. Study project under supervision of lecturer – tissue culture/ genetic engineering

Expected domain skills to be achieved: Ability to prepare artificial nutrient media, preparing independently, applying various sterilization procedures for media, glassware and biological materials, in vitro propagation of Banana callus, morphogenesis--s, clonal propagation methods, isolation of plasmid DNA individually and as a group.