Course title: Plant Biotechnology Laboratory – Part 3			
Course code: BBP 103	**No. of credits:** 7	**L-T-P:** 0-0-210	**Learning hours:** 210
Pre-requisite course code and title (if any): Science graduate			
Department: Department of Biotechnology			
Course coordinator: Dr Chaithanya Madhurantakam		**Course instructor:** Dr Chaithanya Madhurantakam	
Contact details: chaithanya.madhurantakam@terisas.ac.in			
Course type: Core		**Course offered in:** Semester 3	

Course description:

Through this lab course students are provided with an expertise in computational biology and proteomics along with experimental exposure to protein biochemistry and stress physiology. The four modules covered under this laboratory course are bioinformatics, stress physiology, proteomics and biochemical assays. As bioinformatics and proteomics have been major contributors to the surge in biological research in the last decade, students will have an opportunity to have hands-on experience with a series of *in-silico* and *in-vitro* assays to generate, analyse and store the data using advanced bioinformatics and proteomics approaches. Further, the in-vivo and in-vitro assays from stress physiology and biochemistry will allow students to apply the principles in future research projects. Ultimately, the lab course aims at creating a professional pool of students to engage with academia and industry on a larger scale.

Course objectives:

1. To teach a range of techniques, to empirically compute the physiological responses in terms of biochemical and proteomic indicators.
2. To train an impart knowledge of theories and techniques relevant to computational biology
3. To enable students to assess the stress perception in plant systems
4. To provide students with principles of protein biochemical techniques and assays.

Course contents

Module	Topic	L	T	P
1	**Bioinformatics** • To introduce various biological databases and sequence formats in which biological data is stored. • To analyse DNA, RNA and protein sequences in biological databases. • To submit sequence in biological databases. • To analyse biochemical pathway using KEGG. • To predict RNA secondary structures using different software. • To study and use various programs for gene prediction. • To perform sequence similarity tools viz. BLAST. • To identify primers and open reading frames (ORF) using different software. • To perform multiple sequence alignment for the given set of sequences. • To perform and analyse phylogenetic analysis. • To study various protein sequence analysis tools on ExPASY web site .			

	• To build a model of the protein by homology modelling • To dock a ligand into the active site of the protein • To dock two proteins and study their interacting surface. • To study and learn to use the suite of programs EXOME.			
2	**Physiological Studies for abiotic/biotic stressed plants** • Relative water content measurement • Proline measurement • Cell membrane stability: Lipid Peroxidation measurement • Pigment estimation • Total peroxide measurement			
3	**Proteomics** • Total plant protein isolation • Soluble protein isolation • Fractionation and purification of subcellular organelles • Subcellular protein isolation • Quantification of proteins: Lowry and Bradford's method • Non-denaturing polyacrylamide gel electrophoresis (Native PAGE) • Denaturing polyacrylamide gel electrophoresis (SDS-PAGE) • Visualization of proteins on polyacrylamide gels: Linear vs Non Linear stains (Coomassie blue, silver stain, Sypro ruby staining techniques) • Western Blotting			
	Biochemical assays • Alkaline phosphatase enzyme assay • Superoxide dismutase/ ascorbate peroxidase enzyme assay • Marker enzyme assays for subcellular fractions • Inorganic Pi estimation			
	Total	**0**	**0**	**210**

Evaluation criteria:

1. Preparation of report(s)/lab record(s): 20
2. Lab work: 25
3. Major Exam: 40
4. Viva voice: 15

Learning outcomes:

1. Experimental evaluation of dynamic plant physiological responses in terms of quantifiable biochemical and proteomic parameters.
2. Ability to employ *in- silico* tools to retrieve, assimilate and analyze secondary molecular information.
3. Development of an integrative approach towards designing an experiment, execution to generate primary data, analysis and interpretation of the results.
4. Ability to analyse and document the protein characterization methods either biochemically or by *in-silico* methods.
5. Development of understanding a biological problem using proteomic and physiological tools for deciphering the molecular mechanisms in stress.

Pedagogical Approach:

1. Online/classroom lectures and discussions
2. Case studies and examples from original research articles

Skill Set:

1. Formulation of media preparation for plant and animal cell cultures
2. Initiation and maintenance of plant and animal cell cultures
3. Genetic transformation of plants

Employability:

1. Academic organisations
2. Tissue culture facilities and horticulture companies
3. Agri-biotechnology and seed companies
4. Pharmaceutical and drug research companies
5. IPR consultancy firms

Materials:

Suggested Readings

1. Molecular Cloning, a Laboratory Manual Set of Volumes 1, 2, and 3, Third Edition, Joseph Sambrook.
2. Current Protocols in Molecular Biology (2007), John Wiley and Sons, Inc.
3. Methods in Plant Molecular Biology: A Laboratory Course Manual, Pal Maliga, Daniel F. lessig, Anthony
4. R. Cashmore, Wilhelm Gruissem, Joseph E. Varner (1995), Cold Spring Harbor Laboratory Press, US

Additional information (if any): Coordinator may choose experiments from this list, which should be considered merely representative, not exhaustive. The objective is to give students sufficient exposure to several aspects of experimental modern biology.

Student responsibilities:

1. Good Lab Practices
2. Ensuring safety and security of fellow students within the lab premises
3. Requisite attendance and lab record maintenance.

Course reviewers:

The course outline was reviewed and commented by the following experts.

1. Dr. Gitanjali Yadav, Staff Scientist IV, NIPGR
2. Dr. Niranjan Chakraborty, Staff Scientist VII, NIPGR