# Experimental Biotechnology - Web course

### **COURSE OUTLINE**

Good lab practices and precautions, Spectroscopy: Recording UV/Visible absorption spectra for biomacromolecules and quantification; Fluorescence Spectroscopy: monitor equilibrium unfolding of a protein using tryptophan fluorescence; CD spectroscopy, IR spectroscopy. Chromatography: size exclusion chromatography of a crude mixture of proteins using standard matrix and dyes; HPLC, Fluorescence Microscopy: localization of antigen in cells; scanning and transmission electron microscopy. Atomic force microscope. Generation of polyclonal and monoclonal antibodies, Immuno-assays to detect and quantify antigens. Electrophoresis: electrophoresis of protein and nucleic acid. Characterization and quantification of proteins and nuceic acids. Molecular cloning techniques: Preparation and Transformation of competent cells, Small scale Biotechnology isolation of recombinant plasmid; Analysis of the recombinant plasmid using Restriction Endonucleases.

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## **COURSE DETAIL**

Module*	Topics and Contents	No of Lectures
1.Introduction	Introduction, Concept of buffers.	3
2. Spectroscopic experiments	Spectroscopic techniques: UV-Visible spectroscopy, Fluorescence spectroscopy, CD spectroscopy, IR spectroscopy.  Protein estimation 1, 2, DNA estimation, unfolding, Protein unfolding, CD, IR	10
3. electrophoretic experiments	Electrophoresis; Principle, performing electrophoresis techniques, application of electrophoresis in analyzing macromolecules.	7
4. Chromatographic experiments	Chromatographic techniques; Principles, Column chromatography, HPLC	5
5. Immunological experiments	Antibody generation and purification, Immuno-assays to detect and quantify antigens.	4
6. microscopic experiments	Light Microscopy; Fluorescence microscopy, Atomic force microscope, Electron microscope, Scanning electron microscopy, Transmission Electron microsope.	7
7. Molecular cloning	Preparation and transformation of competent cells; Small scale isolation of recombinant plasmid, Analysis of the recombinant plasmid using Restriction Endonucleases.	4

## \*Mid course examination after module 3 and finals after the completion of module 6.

## References:

1. A. J. Ninfa and D. P. Ballou, Fundamental Laboratory Approaches for Biochemistry and Biotechnology, Wiley; 2nd Edition,1998.

### **Coordinators:**

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<sup>\*\*</sup>Numbers of lectures are tentatively fixed.

- 2. J. Sambrook and D. W. Russell, *Molecular Cloning: A Laboratory Manual*, Cold Spring Harbor Laboratory Press, 3rd Edition, 2001.
- 3. Live Cell Imaging: A Laboratory Manual R. D. Goldman, J. R. Swedlow and D. L. Spector Cold Spring. Harbor Laboratory Press; 2nd edition, 2009
- 4. Basic Methods in Microscopy, Protocols and concepts from cells: A Laboratory Manual, D. L. Spector & R. D. Goldman (Editors.), Cold Spring Harbor Laboratory Press, 2006
- 5. A. Manz, N. Pamme and D. lossifidis, Bioanalytical Chemistry, World Scientific Publishing Company, 2004
- 6. R.J. Simpson, Proteins and Proteomics: A Laboratory Manual, CSHL press, 2003

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