

**Name of the Programme:** M.Sc. Biotechnology

**Course Code:** GBT-601

**Title of the Course:** LAB VII: RECOMBINANT DNA TECHNOLOGY

**Number of Credits:** 2

**Effective from AY:** 2022-23

<b>Pre-requisites for the Course:</b>	A theory course in Recombinant DNA technology	
<b>Course Objectives:</b>	The students will learn 1) Understand cloning strategies and expression of foreign genes 2) setting up reactions for DNA manipulation. 3) to interpret the results of DNA manipulation studies and use 4) appropriate tools for the validation of recombinant DNA.	
<b>Content:</b>	<p style="text-align: center;"><b><u>MODULE I</u></b></p> <ul style="list-style-type: none"><li>● Plasmid DNA isolation (Alkaline lysis, Boiling method , column based method)</li><li>● Plasmid DNA quantification.</li><li>● Restriction Enzyme digestion of plasmid DNA.</li><li>● Polymerase Chain reaction (RAPD/RFLP).</li><li>● Real Time PCR.</li><li>● Reverse transcriptase PCR</li></ul>	<b>No. of hours</b>  30
	<p style="text-align: center;"><b><u>MODULE II</u></b></p> <ul style="list-style-type: none"><li>● Cloning of insert into a plasmid vector</li><li>● Transformation of <i>E.coli</i> with standard plasmids, Calculation of transformation efficiency.</li><li>● Confirmation of the insert by Colony PCR and Restriction mapping</li><li>● Expression of recombinant protein, the concept of soluble proteins and inclusion body formation in <i>E.coli</i>, SDS-PAGE analysis</li><li>● Purification of His-Tagged protein on Ni-NTA columns</li><li>● Southern blotting hybridization.</li></ul>	30
<b>Pedagogy:</b>	Hands-on experiments in the laboratory, online videos.	

<b>References/ Readings:</b>	<ol style="list-style-type: none"> <li>1. S. Carson, Manipulation and expression of recombinant, DNA a laboratory manual Elsevier Academic Press, 2006.</li> <li>2. M.R Green and J. Sambrook, Molecular Cloning: A Laboratory Manual Three-volume CSH Press, 2012.</li> <li>3. J.S. Vennison, Laboratory Manual for GENETIC ENGINEERING, PHI Learning, 2009.</li> </ol>
<b>Course Outcomes:</b>	<p>The student will be able to</p> <ol style="list-style-type: none"> <li>1. Create recombinant DNA molecules.</li> <li>2. Conceptualize the various steps in cloning DNA in an appropriate vector and evaluate gene expression.</li> <li>3. Apply and use the knowledge to create tools in diagnostics, medical and forensic science.</li> <li>4. Apply and use PCR for diagnostic applications</li> </ol>