



Department of Biotechnology	LP: BY22211 Rev. No: 02 Date: 21.01.2025
B.E/B.Tech/M.E/M.Tech : Biotechnology PG Specialisation : Biotechnology Sub. Code / Sub. Name : BY22211/ ADVANCED MICROBIAL AND IMMUNOTECHNOLOGY LABORATORY	Regulation: 2022

OBJECTIVE:

- Develop skills and competencies in standard microbiological laboratory techniques.
- Comprehend the ethical consideration and techniques for observing, collecting, and handling live animals and their tissues.
- Isolation of mononuclear cells and splenocytes for immunological studies.
- Identification of lymphocytes using immunological techniques.
- Develop an immunodiagnostic prototype.

Session No*	List of Experiments
1	Disinfection, safety instructions; Preparation of media and Sterilization.
2	Screening and isolation of antibiotic producing microbes from soil.
3	Enumeration of microorganisms by serial dilution.
4	Growth curve, measure of bacterial population by turbidometry.
5	Ethics, selection and handling of animals for immunological experiments (Eg. Mice, Rats, Rabbits).
6	Preparation of antigen and immunization through different routes (Eg. Intra-peritoneal, Subcutaneous, Intramuscular, Intra-nasal, Oral).
7	Methods of bleeding (Eg. Tail bleeding, Intravenous, Intraorbital).
8	Collection of serum, storage and purification of total IgG (salt precipitation).
9	Separation of splenocytes and proliferation against mitogens.
10	Separation of mononuclear cells by Ficoll-Hypaque.
11	Measurement of cytokines secreted from immune cells by ELISA.
12	Immunofluorescence staining of B-cell and T-cell populations of mouse spleen and thymus.
13	Analysis of CD4 expression using RT-PCR and Agarose Gel Analysis.
14	Flow cytometry and FACS analysis of mouse splenocytes and thymocytes.
15	Methods for prototype development of Immunodiagnostics (ICT card).
Content beyond syllabus (if any): Nil	

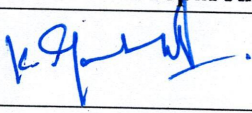

* Session Duration: 200 minutes



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LABORATORY

TEXTBOOKS:

1. Collee, J.G., Fraser, A.G., Marmion, B.P., Simmons, A., Mackie & McCartney. Practical Medical Microbiology, 14th Edition, Churchill Livingstone, 1996.
2. John, E. C. Current Protocols in Immunology, Wiley Interscience, 2003.
3. Freshney, R. Ian. Culture of animal cells: a manual of basic technique and specialized applications, John Wiley & Sons, 2015.
4. Lawson, P. Timothy, ed. Assistant Laboratory Animal Technician, American Association for Laboratory Animal Science, 1998.
5. Melamed, Myron R., Tore Lindmo, Mortimer L. Mendelsohn, and Robert D. Bigler. Flow cytometry and sorting, 2nd Edition, John Wiley & Sons, 1991.

	Prepared by	Approved by
Signature		
Name	Dr. K. Ganesh Prasath	Prof. E. Nakkeeran
Designation	Assistant Professor, Biotechnology	HOD
Date	21/01/2025	21/01/2025
Remarks*:	This lesson plan will be followed in the subsequent years	
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*If the same lab plan is followed in the subsequent semester/year it should be mentioned and signed by the Faculty and the HOD

