SEMESTER IV
COURSE 2 TECHNIQUES IN MICROBIOLOGY
Theory | Credits: 3 | 3 hours per week | Total teaching hours: 45

I Learning Objectives

1. Learn principles and use of microscopes and basic staining methods.
2. Understand preparation of media and isolation of pure cultures.
3. Learn methods to estimate microbial numbers and assess growth.
4. Understand physical and chemical sterilization methods and their applications.
5. Learn biosafety levels, PPE usage, and safe handling of biohazardous materials.

II Course Outcomes

On completion of this course, students will be able to:

CO1. Identify microbial morphology, arrangements, and structures using different stains and microscopes.

CO2. Demonstrate isolation of microbes and maintain pure cultures effectively.

CO3. Quantify microbial populations and monitor growth patterns.

CO4. Sterilize media, glassware, instruments, and workspaces effectively.

CO5. Apply good laboratory practices to prevent contamination and ensure safety.

Activity Implementation

The activities listed under each unit provide options for classroom and independent learning. The instructor shall select and schedule appropriate activities, communicate the expected submission and assessment criteria in advance, and provide feedback. Activity time shall be planned within the course delivery and independent study schedule; the theory allocation remains nine hours per unit.

III Syllabus of Theory

Unit 1 Microscopy and Staining Techniques

Teaching hours: 9 | Related course outcomes: CO1 and CO5

1.1 Principles and applications of light microscopy, phase-contrast, and fluorescence microscopy.

1.2 Electron microscopy: SEM and TEM, sample preparation, and applications.

1.3 Staining methods: Simple, differential, and special staining (Gram, Acid-fast, Capsule, Endospore).

Suggested Learning Activities

Activity	Task	Expected submission
Assignment	Compare light, phase-contrast, fluorescence, SEM and TEM microscopy by principle, image characteristics and applications.	A comparison chart with labelled illustrations and references.

Activity	Task	Expected submission
Seminar	Explain the principle and application of one staining method, using instructor-provided images to discuss interpretation.	A 5–7 minute presentation and responses to questions.
Quiz	Identify morphology, arrangements and stained structures from supplied images; select suitable microscopy methods for stated purposes.	A 20-mark individual image-based quiz.
Project	Create an illustrated microscopy and staining atlas using prepared-slide images or credited reference images.	An atlas with at least eight annotated images, method labels and interpretations.
Peer teaching	Explain magnification and resolution, or interpretation of Gram-stained images, using a labelled diagram.	A 5-minute lesson and three questions to check understanding.

Evaluation

The instructor shall prepare a dedicated rubric for each selected activity before implementation, adapting the activity-specific criteria in Section V to the task. Share the rubric and maximum marks with students, assess the submitted evidence and individual contribution, and record feedback and marks.

Unit 2 Culture and Isolation Techniques

Teaching hours: 9 | Related course outcomes: CO2 and CO5

2.1 Media preparation: Types of media: selective, differential, and enrichment media.

2.2 Isolation techniques: Streak plate, spread plate, pour plate.

2.3 Pure culture maintenance: Sub-culturing and preservation methods.

Suggested Learning Activities

Activity	Task	Expected submission
Assignment	Compare selective, differential and enrichment media. Match instructor-provided examples to their purpose and explain the choices.	A referenced comparison table and brief justifications.
Seminar	Discuss pure culture maintenance and preservation methods, comparing their purposes, advantages and limitations.	A 5–7 minute presentation and source list.
Quiz	Distinguish streak, spread and pour plate methods; select appropriate media and identify contamination risks in supplied scenarios.	A 20-mark individual quiz.
Project	Develop a visual guide to isolation and culture maintenance using diagrams or a paper simulation, highlighting checkpoints for purity.	A labelled flowchart, observation checklist and short explanation.

Activity	Task	Expected submission
Peer teaching	Use a paper-plate simulation to explain the purpose of streaking and how isolated colonies differ from mixed growth.	A 5-minute lesson, annotated plate diagram and three check questions.

Evaluation

The instructor shall prepare a dedicated rubric for each selected activity before implementation, adapting the activity-specific criteria in Section V to the task. Share the rubric and maximum marks with students, assess the submitted evidence and individual contribution, and record feedback and marks.

Unit 3 Quantitative and Qualitative Microbial Analysis

Teaching hours: 9 | Related course outcomes: CO3

3.1 Microbial enumeration: Serial dilution, standard plate count, most probable number (MPN).

3.2 Microscopic counts: Direct counts using counting chambers with vital staining.

3.3 Assessment of microbial growth: Turbidity measurement, spectrophotometry, and dry weight.

Suggested Learning Activities

Activity	Task	Expected submission
Assignment	Solve instructor-provided enumeration problems using dilution factors, plate counts, counting-chamber data and an MPN table. Show units and assumptions.	Worked calculations and a short comparison of the methods.
Seminar	Compare plate counts, direct counts, turbidity and dry weight as measures of growth; discuss what each method measures and its limitations.	A 5–7 minute presentation with an evidence-based comparison.
Quiz	Interpret supplied counting and growth data; calculate microbial numbers and select a suitable enumeration method for a stated purpose.	A 20-mark quiz with numerical and interpretation items.
Project	Analyse an instructor-provided dataset of optical density and viable counts. Plot results and discuss agreement, variation and limitations.	A graph or spreadsheet, calculations and a 500-word interpretation.
Peer teaching	Explain a dilution and plate-count calculation or a counting-chamber example with correct units.	A 5-minute lesson, worked example and short peer exercise.

Evaluation

The instructor shall prepare a dedicated rubric for each selected activity before implementation, adapting the activity-specific criteria in Section V to the task. Share the rubric and maximum marks with students, assess the submitted evidence and individual contribution, and record feedback and marks.

Unit 4 Sterilization Techniques

Teaching hours: 9 | Related course outcomes: CO4 and CO5

4.1 Physical methods: Autoclaving, dry heat, filtration, and radiation techniques.

4.2 Chemical methods: Alcohols, phenolics, aldehydes, quaternary ammonium compounds.

4.3 Sterilization: Sterilization of media, glassware, instruments, and workspace.

Suggested Learning Activities

Activity	Task	Expected submission
Assignment	Prepare a method-selection chart for media, glassware, instruments and workspaces. Distinguish sterilization from disinfection and justify each choice.	A referenced chart with material compatibility and method limitations.
Seminar	Explain one physical or chemical method of microbial control, its applications, limitations and appropriate verification measures.	A 5–7 minute presentation and responses to questions.
Quiz	Match methods to materials, distinguish sterilization from disinfection and interpret instructor-provided process-check scenarios.	A 20-mark individual quiz.
Project	Design a decision chart for selecting an appropriate microbial-control method using supplied material and equipment scenarios.	A decision chart, justification sheet and sample record form.
Peer teaching	Teach peers how to choose between heat, filtration and chemical methods for stated materials and purposes.	A 5-minute lesson and three scenario-based check questions.

Evaluation

The instructor shall prepare a dedicated rubric for each selected activity before implementation, adapting the activity-specific criteria in Section V to the task. Share the rubric and maximum marks with students, assess the submitted evidence and individual contribution, and record feedback and marks.

Unit 5 Biosafety and Handling Biohazardous Materials

Teaching hours: 9 | Related course outcomes: CO5

5.1 Laboratory biosafety levels (BSL-1 to BSL-3) and PPE.

5.2 Handling, transport, and disposal of biohazardous materials.

5.3 Good microbiological and laboratory practices.

Suggested Learning Activities

Activity	Task	Expected submission
Assignment	Compare BSL-1, BSL-2 and BSL-3 by purpose, containment features and the role of PPE. Explain why PPE selection depends on the task.	A comparison chart with references and a brief explanation.

Activity	Task	Expected submission
Seminar	Discuss good microbiological practices using a fictional laboratory incident to identify lapses, reporting needs and preventive measures.	A 5–7 minute presentation and a prevention checklist.
Quiz	Identify appropriate PPE, waste categories and contamination-prevention practices in instructor-provided scenarios.	A 20-mark individual scenario-based quiz.
Project	Prepare a laboratory safety orientation poster and a mock audit checklist using the institution's approved procedures.	A poster, checklist and short rationale for selected items.
Peer teaching	Demonstrate PPE use and waste segregation in a classroom simulation with clean props, explaining each choice.	A 5-minute demonstration, peer checklist and corrective feedback.

Evaluation

The instructor shall prepare a dedicated rubric for each selected activity before implementation, adapting the activity-specific criteria in Section V to the task. Share the rubric and maximum marks with students, assess the submitted evidence and individual contribution, and record feedback and marks.

IV Co-curricular Activities

1. Hands-on session for students to explore different microscopes (light, phase-contrast, fluorescence) and practise staining techniques.

2. Organise a mini-competition on serial dilution, colony counting and turbidity measurement to reinforce quantitative techniques.

3. Simulation activity in which students demonstrate proper use of personal protective equipment, handling of biohazardous materials and sterilization practices.

Practical activities shall be instructor-supervised and follow the institution's approved laboratory procedures. Use prepared materials, teaching strains or simulations suited to the available teaching facilities; the BSL-2 and BSL-3 comparison is a classroom learning activity.

V Evaluation of Learning Activities

The instructor is required to prepare a dedicated assessment rubric for each activity assigned. The templates below are proposed for use and adaptation; they do not prescribe institutional internal-assessment weightage. For each task, specify the unit, course outcome, expected evidence, deadline, criteria, performance descriptors and maximum marks before the activity begins.

VI Reference Books

1. Appl, M. (1999). Microbiology of Extreme Soils. Springer.

2. Atlas, R. M. (2010). Handbook of Microbiological Media (4th ed.). CRC Press.

3. Cappuccino, J. G., & Sherman, N. (2014). Microbiology: A Laboratory Manual (10th ed.). Pearson Education.

4. Frazier, W. C., & Westhoff, D. C. (2014). Food Microbiology (5th ed.). McGraw-Hill Education.

5. Pelczar, M. J., Chan, E. C. S., & Krieg, N. R. (2009). Microbiology: Concepts and Applications (6th ed.). McGraw-Hill Education.

6. Prescott, L. M., Harley, J. P., & Klein, D. A. (2005). Microbiology (6th ed.). McGraw-Hill Education.

7. Tortora, G. J., Funke, B. R., & Case, C. L. (2020). Microbiology: An Introduction (13th ed.). Pearson Education.

8. Willey, J. M., Sherwood, L. M., & Woolverton, C. J. (2017). Prescott's Microbiology (10th ed.). McGraw-Hill Education.

SEMESTER-IV

COURSE 2: TECHNIQUES IN MICROBIOLOGY

Practical **Credits: 1** **2 hrs/week**

I. Course Outcomes: On successful completion of this practical course, student shall be able to:

1. Perform basic staining and microscopic techniques to study microorganisms.

2. Handle culture media and isolate pure microbial cultures using aseptic methods.

3. Estimate bacterial population using quantitative methods and follow laboratory biosafety practices.

II. Laboratory/Field exercises:

1. Compound Light microscope: Parts, handling and observation of the cultures.

2. Simple Staining Technique.

3. Gram Staining Technique.

4. Observation of electron microphotographs of viruses (Lambda, T4, TMV, HIV, SARS CoV-2, Polio).

5. Demonstration of biosafety cabinet usage.

6. Sterilization of Glassware using Hot Air Oven.

7. Preparation and Sterilization of Media using Autoclave.

8. Aseptic transfer of microbial cultures using loop and pipette.

9. Isolation of pure culture of bacteria by streak plate,pour plate and spread plate method.

10. Determination of bacterial count using standard plate count method.

11. Determination of bacterial count using turbidity (spectrophotometric) method