Semester – I

BRANCH	Subject Title	Subject Code
B.Sc. (Microbiology – Minor/Generic Elective Course)	MICROBIAL TECHNIQUES	MBM2/MB

Course Learning Outcomes (CLO) -

After completing this course in Microbiology, a student shall have understanding of -
- Recall the basic lab glassware to be used in the laboratory.
- Summarize different methods of sterilization and isolation of pure cultures.
- Understand the working of different kinds of instruments and microscopes.
  Apply serial dilution technique to isolate the bacteria.
- Practice different methods to culture bacteria in the laboratory
- Illustrate a method to differentiate between Gram positive and Gram negative bacteria.

CONTENT

Unit - I

Microscopy and Staining

1.1 Microscopy - Principles and applications of simple and compound Bright-field microscopy, Dark-field microscopy, Fluorescence microscopy, Phase-contrast microscopy, Transmission electron microscopy and Scanning electron microscopy.

1.2 Preparation for Light Microscope Examination - Wet- mount and hanging-drop techniques.

(iii). Preparation for smear and fixation.

1.3 Staining - Principles of staining, negative staining, simple staining, differential staining (Gram and acid fast staining), flagella staining, capsule and endospore staining.

Unit - II

Instruments

Electronic Balance, Autoclave, Centrifuge, Colony counter, Deep freezer, Homogenizer, Hot air Oven, Incubator, Laminar air flow, Magnetic stirrer, pH Meter, Spectrophotometer, Vortex mixture, Water bath, Water distiller, Chromatography Chambers, Anaerobic chamber and Electrophoresis apparatus.

Unit - III

Sterilization and Culture Medium

3.1 Physical methods of sterilization - Dry heat, Moist heat, Filtration and Incineration.

3.2 Chemical methods of sterilization - Phenol and phenolic compounds, Alcohol, Halogens and Detergents.

3.3 Types of culture media -Natural, synthetic, complex, enriched and selective. Anaerobic (Thioglycolate broth, Robertson's media, Microaerophilic), broth culture of aerobic bacteria.

Unit - IV

Isolation, Cultivation and Preservation

4.1 Natural microbial population - Pure culture.

4.2 Isolation of microbial population - From air, water and soil.

4.3 Methods for isolation - Streak plate, Pour plate and Spread plate. Serial dilution and Micromanipulator methods. Cultivation on liquid and solid media. Isolation of microorganisms on potato slice and bread.

4.4 Maintenance and preservation for short term and long term.

4.5 Cultivation of anaerobic bacteria and accessing non- cultivable microorganisms.

Part B-Content of Practical Course

1. Demonstration and briefing about principles and working of basic instruments.

2. Basic media preparation technique, autoclaving, cleaning and sterilization of glassware.

3. Preparation of liquid culture media - Peptone water, nutrient broth

4. Preparation of solid culture media - Nutrient agar (agar slant/ agar plate)

5. Isolation of microbes from water, soil and air by serial dilution agar plating method.

6. Isolation of fungi from water, soil and air by serial dilution agar plating method.

7. Isolation of microorganisms by pour plate method.

8. Isolation of microorganisms by streak plate method.

9. Isolation of microorganisms by spread plate method.

10 Any other experiment may be designed on the basis of theoretical aspects.