MICROBIAL CELL STRUCTURE & FUNCTION 45 LECTURES  S.MIC.1.01

LEARNING OBJECTIVES:

- Learn the fundamental aspects of Prokaryotic and Eukaryotic Cell structure and function, and the differences between these cells
- Learn and understand the principles of working of the light microscope and other modified microscopes and to know the differences between them. To be able to apply this knowledge in the laboratory.
- Develop analytical skills
- Think in a critical & creative manner

UNIT 1: PROCARYOTIC CELL STRUCTURE, FUNCTION AND STAINING 15 LECTURES

1. Members of the Microbial World 3L
   - The universal Phylogenetic tree
   - Discovery of Micro-organisms
   - Overview of Prokaryotic Cell Structure: Size, Shape, Arrangement, Micrometry
   - Diagram of Prokaryotic cell organization

2. Cell Wall Structure and Gram Stain  3L

3. Cell Membrane: Bacterial and Archaeal  2L

4. Cytoplasmic Matrix 3L
   - Cytoskeleton, Nucleoid, Plasmids, Ribosome
   - Inclusion granules: Composition, Function and Staining

5. Components External to Cell Wall 3L
   - Capsule, Slime, S-layer, Demonstration
   - Pili, Fimbriae
   - Flagella: Structure, Motility, Chemotaxis, Staining

6. Bacterial Endospires  1L
   - Examples of spore forming organisms, habitats, function, staining
   - Formation and Germination
ACTIVITY: Draw Table to include: names, morphology, arrangement, Gram nature with diagrams and kind of motility for each of 15 common microbes

UNIT 2: EUCARYOTIC CELL STRUCTURE AND FUNCTION  15 LECTURES

1. Overview of eukaryotic cell structure: General structure and types of cells  1L
2. External Cell coverings and Cell Membrane: Structure and Function  2L
3. Cytoplasmic Matrix  9L
   • Cytoskeleton: Structure and Function
   • Single Membrane Organelles - Endoplasmic reticulum, Golgi complex, Lysosomes, Vesicles, and Ribosomes: Structure and Function and the Endocytic, Biosynthetic and Secretory pathways involved
   • Double Membrane Organelles – Nucleus, Mitochondrion and Chloroplast: Structure and Function
   • Peroxisomes : Structure and Function
4. Organelles of motility – Structure and movement of flagella and cilia  2L
5. Comparison of Prokaryotic and Eukaryotic cells - Structure & Function  1L

UNIT 3: MICROSCOPY  15 LECTURES

1. History of the Microscope  1L
2. Lenses and bending of Light  1L
3. Light Microscopy  5L
   • Bright field Microscopy: Objectives, Eyepiece, Condenser
   • Characteristics of lenses: Resolution, Magnification, Numerical Aperture, Focal Length, Working distance, Depth of Focus.
   • Specimen Preparation and Principles of Bacterial cell staining.
4. Dark Field Microscopy  1L
5. Phase Contrast and Differential Interference Contrast Microscopy  1L
6. Fluorescence Microscopy  1L
7. Electron Microscopy: TEM & SEM and Specimen preparation  3L
8. Newer Techniques in Microscopy 2L

- Confocal Microscope.
- Scanning Probe Microscope

Student activity: History of the microscope, different types of light microscopes other than those mentioned above – draw or stick images of microbes using all of the above microscopes

CIA: Quiz

References

ELEMENTS OF MICROBIAL NUTRITION, GROWTH & CONTROL
45 LECTURES

LEARNING OBJECTIVES:

- Understand the basic concepts of microbial nutrition, growth and control
- Gain knowledge of the principles and basic methods involved in the study and control of microbes
- Develop analytical and problem solving skills
- Think in a critical & analytical manner

UNIT 1: MICROBIAL NUTRITION, CULTIVATION, ISOLATION AND PRESERVATION
15 LECTURES

1. Scope and Relevance of Microbiology 2L
2. Nutritional requirements- Carbon, Oxygen, Hydrogen, Nitrogen, Phosphorus, Sulphur and Growth factors 1L
3. Nutritional types of microorganisms 2L
4. Nutrient uptake mechanisms 2L
5. Types of culture media with examples 3L
6. Isolation of microorganisms and pure culture techniques 3L
7. Preservation of microorganisms 2L

UNIT 2: MICROBIAL GROWTH
15 LECTURES
1. Definition of growth, Mathematical expression, Growth curve 3L
2. Measurement of Growth 7L
   - Direct Microscopic count- Breeds, Petroff-Hausser counting chamber, Haemocytometer
   - Viable count- Spread plate and Pour plate technique, Membrane filtration
   - Electronic Counting
   - Measurement of cell mass
   - Turbidity measurements- Nephelometer and spectrophotometer techniques
   - Measurements of cell constituents.
3. Synchronous growth, Continuous growth (chemostat and turbidostat), Diauxic growth, Growth Yield (definition of terms) 1L

4. Influence of environmental factors on growth 3 L

5. Microbial growth in natural environments, viable non-culturable organisms, Quorum sensing 1L

UNIT 3: CONTROL OF MICROORGANISMS 15 LECTURES

1. Definitions of frequently used terms 1L

2. Pattern/Rate of Microbial Death 1L

3. Conditions influencing the effectiveness of Antimicrobial agents 1L

4. Physical Methods of Microbial Control 5L

- Heat: Moist and Dry
- Low temperature
- Filtration
- High pressure
- Desiccation
- Osmotic pressure
- Radiations

5. Chemical methods of Microbial Control 5L

- Phenolics
- Biguanides - chlorhexidine
- Alcohols
- Halogens
- Heavy Metals
- Quaternary ammonium compounds
- Surface active agents
- Aldehydes
- Sterilizing gases
- Peroxygens
- Chemotherapeutic agents

6. Evaluation of effectiveness of Antimicrobial agent 2L

C.I.A – Quiz /Problem solving
References
PRACTICALS

SEMESTER I

COURSE: S.MIC.1.PR

LEARNING OBJECTIVES:

- To learn, understand and practice Safety rules when in the Microbiology Laboratory and become proficient in Aseptic techniques
- To gain proficiency in the use of Micropipettes
- To learn principles of Microscopy, to gain proficiency in the use and care of the Compound Microscope and to successfully stain bacteria
- To gain proficiency in the techniques of cultivation, isolation and preservation of bacteria
- To use physical and chemical methods to control the growth of micro-organisms.
- To learn the techniques of enumeration of micro-organisms.
- To learn to critically observe and record the observation of all experimentation.

PRACTICAL 1

1. Biosafety in the Microbiology Laboratory- practices and rules involved with a short experimental study

2. Assignments –
   a) Contributions of one Scientist of the Golden Era
   b) Experiments that refuted the belief in Spontaneous Generation

3. The Light Microscope –
   a) Diagram of Path of Light through Compound Microscope
   b) Working Rules
4. Monochrome staining of bacteria
5. Negative staining and Micrometry
6. Gram staining of bacteria
7. Staining of Cell components – Cell wall, Capsule, Metachromatic & Lipid granules, Endospores
8. Staining of Flagella and Spirochaetes
9. Motility by Hanging drop technique
10. Staining of Yeasts
11. Wet mount of Hay Infusion and Pond water for observing bacterial, algal and protozoan forms.

C.I.A – Quiz/Staining technique
PRACTICAL 2

1. Preparation of culture medium:
   a) Liquid medium (Nutrient broth)
   b) Solid media (Nutrient agar, Sabouraud agar)
   c) Preparation of slants, butts and plates

2. Inoculation Techniques:
   a) Aseptic Transfer techniques using glass and micro pipettes
   b) Liquid medium
   c) Solid media (slants, butts and plates)

3. Cultivation of bacteria:
   a) Study of Colony Characteristics on Nutrient agar
   b) Study of Motility using Motility agar
   c) Use of differential, selective and enriched media
      (i) MacConkey’s agar
      (ii) Superimposed blood agar

4. Determination of optimum growth conditions (Temperature, pH, aeration)

5. Measurement of Microbial growth
   a) Microscopic cell count (Haemocytometer, Breed’s Count)
   b) Brown’s opacity tubes
   c) Viable count (Pour plate and surface spread)
   d) Growth curve of *E.coli* and determination of generation time (Group Experiment)

6. Physical methods of control of microorganisms:
   a) Heat: Autoclaving, Fractional sterilization, Dry heat.
   b) Bacteria proof filtration (Demonstration of Membrane filtration)
   c) Effect of U-V rays
   d) Effect of desiccation.
   e) Effect of high osmotic pressure

7. Chemical Methods of Control of microorganisms:
   a) Effect of phenolics (Disc Method) and other disinfectants used at home
   b) Oligodynamic action of Copper foil and Mercurochrome
   c) Effect of Cetrimide
   d) Effect of Dyes (Disc Method)
   e) Effect of Chemotherapeutic agents

CIA- Isolation and Motility Techniques