Semester- III

Course MI-201

Microbial Physiology

Unit I. Microbial Nutrition and Factors Affecting

1. Culture media: Types of culture media: Routine and specialized media; selective media, differential media, enriched media, enrichment media, enumeration media, assay media and maintenance media (4 hr)
2. Modes of nutritional uptake (3 hr)
   A. Entry of nutrition in the cell, passive diffusion, facilitated diffusion and active transport,
   B. Utilization of nutrients that cannot enter the cell
3. Classification of bacteria on the basis of growth supporting environmental factors such as oxygen, temperature, pH, osmotic pressure, salt and hydrostatic pressure. (3 hr)

Unit II. Enzymes

1. General introduction (4 hr)
   A. Physical and chemical properties
   B. Structure of enzymes: Prosthetic group, apoenzyme, coenzymes, cofactors
   C. Localization of enzymes: Extra cellular and intra cellular
   D. Nomenclature and classification of enzymes, IUB system of enzyme classification
2. Enzyme action (6 hr)
   A. Active sites of enzymes
   B. Mechanism of enzyme action
   C. Factors affecting enzyme activity
   D. Inhibition of enzyme activity: Competitive and noncompetitive

Unit III. Microbial growth

1. Methods of reproduction in bacteria and new cell formation (2 hr)
2. Growth (5 hr)
   A. Introduction to growth rate, generation time
   B. Criteria for growth measurement: Cell mass and cell number, methods of their measurement
   C. Normal growth curve of bacteria
   D. Continuous growth and synchronous growth
3. Chemotherapeutic agents as growth inhibitors (3 hr)
   A. Principles of chemotherapy

B. **G**eneral mode of action of various chemotherapeutic agents: Sulfonamides, antibiotics (penicillin, streptomycin, Polymixin)

Unit IV. Biomolecules and metabolism

1. Biommolecules: Chemical structure, properties, classification and biological significance of carbohydrates, proteins, lipids and nucleic acids (7 hr)
2. Introduction to metabolism (3 hr)
   A. Anabolism, catabolism, primary and secondary metabolism
   B. Role of reducing power, precursor metabolites and energy rich compounds in cell metabolism

Text Books:

1. Pelczar Jr, M J, Chan E C S., Krieg N R, (1986) ***Microbiology,*** 5th edn, McGraw-Hill Book Company, NY
2. Ingraham J L, and Ingraham, C L, (2000) ***Introduction to Microbiology***, 2nd edn, Brooks/Cole, Singapore
3. Black J G, (2002) ***Microbiology: Principles and Explorations***, 5th edn, John Wiley and Sons, Inc. NY

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Semester- III

Course MI-202

Soil and Water Microbiology

Unit I. Microbiology of Soil

1. Physicochemical characteristics of soil, soil as a culture medium (1 hr)
2. Soil microflora: Diversity in soil microflora (1 hr)
3. Methods of studying soil micro flora: (3 hr)
   A. Direct microscopic method, agar plate technique, enrichment culture technique, and buried slide method
   B. Use of Winogradsky column in studying microbial diversity in soil
4. Microbial interactions in soil (5 hr)
   A. Neutral, positive and negative associations
   B. Interaction between plant roots and microorganisms:
      i. Rhizosphere and its significance
      ii. Mycorrhiza and root nodule formation

Unit II. Microorganisms as Biogeochemical Agents

1. Introduction to biogeochemical transformations in soil: Mineralization and immobilization of elements (1 hr)
2. Rotation of elements in nature (7 hr)
   A. Nitrogen cycle: Proteolysis, ammonification, nitrification, denitrification and nitrogen fixation
   B. Sulfur cycle: Sulfur oxidation and reduction
   C. Carbon cycle: Degradation of complex organic compounds, carbon dioxide fixation, humus and its significance
   D. Iron cycle: Iron oxidation and reduction
   E. Phosphorus cycle: Mineralization, immobilization and solubilization of phosphorus
3. Soil fertility: Role of microorganisms in soil fertility, biofertilizers (2 hr)

Unit III. Microbiology of Drinking Water

1. Natural waters: Sources of contamination (1 hr)
2. Microbial indicators of fecal pollution (3 hr)
   A. Coliforms as indicator, need for differentiation
   B. Methods of differentiation: IMViC test and Elevated temperature test
   C. Microbial indicators other than coliforms
3. Nuisance organisms in water: Slime forming bacteria, iron and sulfur bacteria and algae (1 hr)

4. Water-borne diseases (1 hr)
5. Bacteriological examination of drinking water (3 hr)
   A. Sampling
   B. Quantitative analysis: Standard plate count
   C. Qualitative analysis: Multiple tube fermentation method (presumptive, confirm and completed test), MPN, membrane filter technique, defined substrate test, P-A (Presence-Absence) test
6. Purification of drinking water: Sedimentation, filtration and disinfection (1 hr)

Unit IV. Microbiology of Wastewater

1. Types of wastewater, chemical and microbiological characteristics of waste water (1 hr)
2. BOD, COD and TOD as indicators of strength of wastewater, pollution problems due to disposal of untreated wastewater (3 hr)
3. Methods of wastewater treatment (6 hr)
   A. Primary treatment and secondary treatment: Principles and role of microorganisms in septic tank, Imhoff tank, trickling filters, activated sludge process, oxidation ponds
   B. Advanced treatment and final treatment
   C. Solid waste processing: Anaerobic sludge digestion and composting
   D. Efficiency of wastewater treatment procedures

Text Books:

1. Pelczar Jr. M J, Chan E C S, Krieg N R, (1986), ***Microbiology***, 5th edn, McGraw-Hill Book Company, NY
2. Alexander M, (1977), **Soil Microbiology,** 2nd edn. Krieger Publ. Co., Melbourne, FL
3. Atlas R M., (1997), ***Principles of Microbiology***. 2nd edn. Wm. C. Brown Pub., Iowa, USA.

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Semester- III

Course MI-203

Microbiology Practicals

1. Study of different types of media and their ingredients.
   A. Selective media: Rose Bengal agar medium
   B. Differential media: MacConkey's medium, EMB agar medium, triple sugar iron agar medium
   C. Enrichment media: Selenite broth
   D. Enriched media: Blood agar medium, glucose yeast extract agar medium
   E. Natural media: Soil extract agar, potato dextrose agar medium
2. Qualitative analysis of biomolecules:
   A. Carbohydrates: Iodine test, Molisch's test, Benedict's test, Barfoed test, Bial's test and Saliwanoff's test
   B. Proteins: Biurate test, Ehrlich's test, glyoxilic acid test, xanthoproteic test.
3. Determination of absorption maxima of a colored solution (use methylene blue 1:20,000 dilution)
4. Study of effect of antibiotics on bacteria
   A. Study of sensitivity spectrum of antibiotic against the test organism by use of paper disc method
   B. Determination of spectrum of activity of an antibiotic by use of agar ditch method
5. Study biochemical reaction of bacteria
   A. Based on carbon source
      i. Oxidative and fermentative breakdown of glucose
      ii. Fermentation of sugars and sugar alcohol: glucose, xylose, mannitol, lactose, maltose and sucrose
      iii. Glucose breakdown product: Methyl red test, Voges-Proskauer's test
      iv. Citrate utilization test
      v. Starch utilization test
      vi. Lipid utilization test
   B. Based on nitrogen source
      i. Indole production test
      ii. H_2S production test
      iii. Urea utilization test
      iv. Casein hydrolysis test
      v. Gelatin hydrolysis test
      vi. Deamination test

D. Other tests
   i. Catalase test
   ii. Dehydrogenase test
   iii. Oxidase test

6. Microbiological analysis of soil
   A. Enumeration of organisms from soil (standard plate count from soil)
   B. Isolation of symbiotic & non-symbiotic nitrogen fixing bacteria & actinomycetes from soil

7. Microbiological analysis of drinking water
   A. Standard plate count of drinking water
   B. Detection of fecal pollution of water by performing presumptive test, confirmed test and completed test.
   C. Determination of MPN of coliforms in water

Scheme for Examination

Ex		Marks
1.	Microbiological analysis of soil / water (any one)	15
	A. Standard plate count of water / soil sample	
	B. Determination of MPN for coliforms in water sample	
	C. Presumptive and confirmed test for water	
	D. Confirmed and completed test for water	
2.	Biochemical reactions of bacteria (any five)	15
3.	General Exercise: (any one)	15
	A. Study of effect of antibiotics on test organism by paper disc method	
	B. Determination of spectrum of activity of an antibiotic by use of agar ditch method	
	C. Determination of absorption maxima	
	D. Qualitative analysis of protein or carbohydrates	
	E. Study of cultural and morphological characteristic of actinomycetes	
	F Cultivation and study of nitrogen fixing bacteria from soil	
4	Spotting	10
5	Viva	10
6	Journal and slides	05
	Total	**70**

Semester- IV

Course MI-204

Microbial Biodiversity

Unit I. Introduction

1. What is biodiversity? (1 hr)
2. Origin of life, evolution and origin of biodiversity, species concept (3 hr)
3. Evolutionary tree of microorganisms (3 hr)
4. Value of biodiversity, microbial biodiversity as index of environmental change (3 hr)

Unit II. Methods of Assessing Biodiversity

1. Microscopic methods (3 hr)
2. Cultural methods (4 hr)
3. Molecular and genomic methods: Molecular context of microbial diversity, importance of DNA and rRNA sequence comparison, determination of GC content (3 hr)

Unit III. Biodiversity among Bacteria & Archaea

1. Morphological and cellular diversity (4 hr)
   A. Diversity in major cell shape and grouping
   B. Diversity in ultra structure of cell with reference to cell envelope, cell membrane, cell wall, surface appendages, other cell organelles and spore.
2. Physiological and metabolic diversity (4 hr)
   A. Diversity in photosynthetic, heterotrophic and autotrophic metabolism
3. Ecological diversity (2 hr)
   A. Diversity in major ecosystems
   B. Diversity in aquatic, marine and extreme environment

Unit IV. Biodiversity among Eukaryotic and Acellular Microorganisms

1. Eucarya: Morphological, cellular, physiological, metabolic and ecological characteristics of (8 hr)
   A. Protozoans
   B. Slime molds
   C. Fungi
   D. Algae
   E. Lichens as consortium of algae and fungi
2. Acellular organisms: Viruses and prions (2 hr)

Text Books:

1. Cambell R., (1983), ***Microbial Ecology,*** 2nd edn. Blackwell Scientific Publications, London
2. Ogunseitan O., (2005) ***Microbial Diversity: Form and Function in Prokaryotes,*** Blackwell Publishing, Malden, MA, Oxford, Victoria
3. Atlas R M, Bartha R(1998), ***Microbial Ecology: Fundamentals & Applications***. 4th edn. Pearson Education.

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Semester - IV

Course MI-205

Food and Dairy Microbiology

Unit I. Microbes in Food Infection and Poisoning

1. Food as a substrate for microorganisms (1 hr)
2. Microbial flora of foods: Milk, fruits, vegetables, meat, eggs (2 hr)
3. Factors affecting kinds and numbers of microorganisms, intrinsic and extrinsic factors (2 hr)
4. Food and milk borne infections (2 hr)
   - A. Sources of contamination
   - B. Major food and milk borne diseases
5. Food poisoning (3 hr)
   - A. Microorganisms involved, sources of contamination
   - B. Role of *Staphylococcus aureus, Clostridium botulinium* and *Salmonella* spp
   - C. Molds as poisoning agents

Unit II. Microbial Food Spoilage and Preservation

1. Microbial Spoilage of food (4 hr)
   - A. Causes of spoilage
   - B. Biochemical changes caused by microbes
   - C. Spoilage of milk and milk products, fruits, vegetables, eggs, meat
   - D. Spoilage of canned foods
2. Preservation of food and Milk (6 hr)
   - A. General principles
   - B. Methods of preservation
     - i. Use of aseptic handling
     - ii. High temperature: Pasteurization, sterilization, canning
     - iii. Low temperature: Refrigeration and freezing
     - iv. Dehydration
     - v. Osmotic pressure
     - vi. Preservatives
     - vii. Radiations: Ionizing and non ionizing radiation

Unit III. Microbes as Food and Food Products

1. Fermented dairy products (5 hr)
   A. Starter culture
   B. Cheese: Types, curdling, processing, ripening
   C. Other fermented dairy products- Yogurt, cultured buttermilk, acidophilus milk, Kefir and cultured sour milk
   D. Introduction to probiotics, prebiotics and synbiotics
2. Indian fermented food products: Pickles, idli, Khaman and bread (2 hr)
4. Microbes as food: Mushrooms, spirulina and yeasts (3 hr)

Unit IV. Methods in Food Microbiology

1. Biological methods: Generalized scheme for microbiological examination (5 hr)
   A. Direct microscopic examination, colony forming units (CFU),
   B. Most probable number (MPN),
   C. Identification of specific group or species of microorganisms
2. Bacteriological analysis of milk (3 hr)
   A. Grading of milk: Resazurin test
   B. Determination of efficiency of pasteurization: Phosphatase test
   C. Determination of MPN
   D. Acid-fast staining
3. Microbiological criteria of food safety (2 hr)

Text Books:

1. Pelczar Jr, M J, Chan E C S, Krieg N R, (1986), ***Microbiology: An Application Based Approach***, 5th edn. McGraw-Hill Book Company, NY
2. Frazier W C and Westhoff D C (1988), ***Food Microbiology***, 4th edn. McGraw-Hill Book Company, NY.
3. Prescott L, Harley J P, and Klein D A, (2008), ***Microbiology***, 7th edn. Wm C. Brown - McGraw Hill, Dubuque, IA.

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Semester- IV

Course MI-206

Microbiology Practicals

1. Study of ecological diversity amongst bacteria at extreme conditions: Cultivation of acidotolerant (pH-4), alkalitolerant (pH-8), halotolerant (NaCl 10%), thermotolerant (temp:50 ^{o}C) bacteria

   [Cultivation using nutrient broth (as basal medium) at different environmental variable(s), results to be observed in form of turbidity followed by Gram's staining. Use routine nutrient broth as control tube. Soil sample to be used for cultivation].

2. Study of microbial diversity in soil by using Winogradsky Column (Demonstration only)

3. Study of morphological and cultural diversity of *Escherichia coli, Enterobacter aerogenes, Staphylococcus aureus, Bacillus subtilis, Bacillus megaterium and Bacillus cereus*.

   A. Study of morphological diversity by performing Gram's staining, capsule staining and spore staining.

   B. Study of cultural / growth diversity using nutrient broth and nutrient agar media

4. Study of metabolic diversity amongst bacteria: *Escherichia coli, Enterobacter aerogenes, Proteus vulgaris, Staphylococcus aureus, and Bacillus subtilis* by performing various biochemical tests:

   A. Based on carbon metabolism

      i. Methyl Red Test
      ii. Voges-Proskauer (V-P) test
      iii. Fermentation of sugars and sugar alcohol: glucose, xylose, mannitol, lactose, maltose and sucrose
      iv. Citrate utilization test
      v. Starch utilization test
      vi. Lipid utilization test

   B. Based nitrogen metabolism

      i. Indole production test
      ii. H_2S production test
      iii. Urea utilization test
      iv. Casein hydrolysis test
      v. Gelatin hydrolysis test

   C. Presence of respiratory enzymes

      i. Catalase test
      ii. Dehydrogenase test
      iii. Oxidase test

5. Study of diverse groups of eukaryotic microorganisms
   A. Fungi: Cultural and microscopic characters of *Mucor, Rhizopus, Aspergillus, Penicillium* and yeast
   B. Algae: Study of algae present in pond water; study of permanent slides of spirogyra and diatoms
   C. Protozoa: Study of presence of protozoa in pond water; study of permanent slides of Amoeba, Euglena and Paramecium
6. Microbiological analysis of food
   A. Standard plate count of food sample
   B. Determination of MPN of coliforms
7. Microbiological analysis of milk
   A. Standard plate count of milk sample
   B. Determination of microbial load of milk by use of MBRT of raw milk, boiled milk and pasteurized milk
   C. Detection of acid-fast organisms in milk sample

Scheme for Examination

Ex		Marks
1.	Microbiological analysis of food / milk (any one) A. Standard plate count of food / milk sample B. Determination of MPN for coliforms in food sample C. Determine microbial load of milk sample by performing MBRT and check for presence of acid-fast bacteria.	15
2.	Diversity in bacteria (any one) A. Study cultural diversity and morphological diversity in given bacterial cultures (two bacterial cultures) B. Study metabolic diversity based on metabolism of nitrogen source / carbon source / presence of respiratory enzymes of the given bacterial cultures (two bacterial cultures, three tests)	15
3.	Identification of fungi A. Identify the given fungal culture based on its growth and morphological characters.	15
4	Spotting	10
5	Viva	10
6	Journal and slides	05
	Total	**70**