

Microbiology

Agriculture and Food Microbiology

Description of the Course

Course Title: Agriculture and Food Microbiology

Course No: MB 401 (Major)

Nature of the Course: Theory

Full Marks: 100

Pass Marks: 35

Year: IV

Total Lecture Hours: 150

Course Objectives

After completion of the course, the students will be able to:

- a) understand microbial ecology and their role in environment
- b) understand the role of micro-organisms in soil fertility, mineral cycles of nature, and plant disease management
- c) understand various microbes found in different kinds of food, food borne infections and food preservation methods.

Course Contents

Soil and its constituents

3 hrs

Introduction and formation of soil, physical, chemical and biological factors, mineral and organic matters, soil solution, gases

Microorganisms in soil and their roles

3 hrs

Bacteria, fungi, actinomycetes, protozoa, blue-green algae (cyanobacteria)

Microbial ecology

5 hrs

Microbial association in soil, water and air, components of microbial ecology, ecosystem and energy, tools and techniques of experimental microbial ecology

Rhizospheric and phyllospheric microorganisms

3 hrs

Introduction, functions, factors influencing their growth and activities

Microbial interactions

6 hrs

Microbial interactions in ecosystem, types of interactions- synergism, commensalism, mutualism, competition, amensalism, predation, and parasitism

Microbiology of extreme environments	6 hrs
Growth and survival of microorganisms in extreme temperature, pH, humidity, salinity, and applications of extremophiles	
Biogeochemical cycles and role of microorganisms	12 hrs
Introduction, nitrogen cycle, carbon cycle, phosphorous cycle, sulphur cycle	
Microbial degradation	10 hrs
Difference between degradation and deterioration, mechanisms of microbial degradation of cellulose, hemi-cellulose, lignin, chitin, pectin, recycling of organic waste	
Bioremediation	5 hrs
Principles of bioremediation, <i>in situ</i> and <i>ex situ</i> bioremediation of soil, water and air pollution, steps and approaches in bioremediation, removal of xenobiotics, bioleaching, petroleum degradation	
Aerobic and anaerobic decomposition of organic compounds and application	12 hrs
Anaerobic decomposition of typical organic compounds, mechanism of aerobic and anaerobic composting, mechanism of methane gas production from agricultural waste including animal waste, biochemistry of methane formation and application	
Plant pathogens	10 hrs
Agents of plant diseases: a) Bacterial diseases- Citrus canker causing microorganism, Rickettsia, Mycoplasma, (MLD), Spiroplasma b) Fungal diseases c) Diseases caused by Nematodes, d) Mesobiotic-viral disease	
Microorganisms in various foods	10 hrs
Bacteria, molds, yeasts, primary sources of microorganisms in food contamination	
Factors affecting microbial growth in food	5 hrs
Intrinsic parameters, extrinsic parameters	
Techniques for determination of microorganisms in food	10 hrs
Techniques of detection and enumeration of microorganisms in food, sampling methods (various food industries, dairy, meat market)	
Food handling and spoilage	15 hrs

Different types of food handling in industries and market, spoilage of fruits and vegetables, fresh and processed meat and poultry product, egg and egg products, milk and milk products, canned foods, flour cereals and bakery products, fermented foods and beverages, soft drinks, seafoods

Food preservations **10 hrs**

Chemical, irradiation, low temperature, high temperature, drying

Food quality evaluation **15 hrs**

Quality standard of milk, quality standard of bakeries, quality standard of meat and eggs, quality control of food, Nepal standard, national food regulatory system

Role of microorganisms in food poisoning **10 hrs**

Gram positive cocci (*Staphylococcus* spp.), gram positive spore formers (*Bacillus cereus*, *Clostridium* spp.), gram negative bacteria (*Salmonella* spp.), mycotoxin (aflatoxin), seafood toxicants

Recommended Readings

Text books

1. Alexander M (1961). Introduction to Soil Microbiology, Academic Press.
2. Atlas RM and Bartha R (1998). Microbial Ecology: Fundamentals and Applications. The Benjamin Cummins Publication Co. Inc.
3. Banwart GJ (1989) Basic Food Microbiology. CBS Publication and Distributions, Delhi.
4. Jay JM (1987). Modern Food Microbiology (3rd Edition). CBS Publications and Distributions, Delhi.

Reference books

1. Frazier WC and Westhoff DC (1986). Food Microbiology (3rd Edition). Tata McGraw-Hill Publishing Company Limited, New Delhi.
2. Rangaswami G and Bagyaraja PT (1993). Agricultural Microbiology (2nd Edition). Prentice Hall of India.
3. Stolp H (1988). Microbial Ecology: Organisms, Habitats, Activities. Cambridge.

Agriculture and Food Microbiology Practical

Description of the Course

Course Title: Agriculture and Food Microbiology Practical

Course No: MB 402 (Major)

Nature of the Course: Practical

Full Marks: 50

Pass Marks: 20

Year: IV

Total Lecture Hours: 180

Course Objectives

After completion of the course, the students will be able to:

- a) develop practical skills on handling, isolation and identification of soil, water, and food microorganisms.

Course Contents

Microorganisms present in soil: Total viable number of bacteria, fungi, actinomycetes in different kinds of soil

Buried slide technique for microbial flora of soil: Different types of microorganisms in soil, identification of microbes by using various techniques

Azotobacter: Introduction, morphological structure, culture of *Azotobacter*

Rhizobium: Morphological structure, cultural properties, methods of isolation and identification

Cellulase produced by organisms: Demonstration of cellulolysis, use of cellulase

Micro organisms from biofertilizers: Isolation and identification of organisms responsible for formation of biofertilizers

Observation of various organisms present in soil: Demonstration of Winogradsky's column, uses of column in differentiation of microbial ecology

Differentiation of various organisms from soil samples: To show the distribution of organisms, their methods of isolation and antibiotic activity

Cyanobacteria: Isolation, growth characteristics and identification

Phosphorus solubilisers: Methods of culture, identification of solubilisers

Count of micro-organisms in different foods: Methods of isolation and counting techniques of bacteria from various foods

Dairy products; milk, cheese and ice-cream: Enumeration, culture of organisms, isolation, detection and demonstration of microorganisms

Tests for milk quality: Methylene Blue Reduction Time (MBRT) test, Resazurin test, phosphatase test, adulteration tests in milk (starch, soda, sodium hydroxide, table sugar), alcohol test, acidity test, SNF and fat tests

Presence of microorganisms in meat and meat products: Isolation and identification of types of contaminating microorganisms in meat and meat products

Microorganisms present in fruits: Culture of microorganisms from fruits (bacteria, yeast and molds), methods of isolation and identification

Medical and Public Health Microbiology

Description of the Course

Course Title: Medical and Public Health Microbiology
Course No: MB 403 (Major)
Nature of the Course: Theory

Full Marks: 100
Pass Marks: 35
Year: IV
Total Lecture Hours: 150

Course Objectives

After completion of the course, the students will be able to:

- a) understand basics of epidemiology and health and disease measurements
- b) understand public health of infectious diseases
- c) understand the immunity process in human body
- d) describe biology, pathogenesis and diagnostic methods of bacteria, virus, fungi and parasites

Course Contents

Health and disease and epidemiological measurements	5 hrs
Definitions of epidemiology, applications of epidemiology, definitions of health and disease, indicators of health and disease, disease frequency measures (mortality, morbidity, incidence, prevalence, incidence density), measures of effect	
Methods of transmission of diseases	6 hrs
Epidemic, endemic, pandemic, sporadic, outbreak, investigation of disease outbreaks, mode of transmission of diseases, chain of infection, cases, carriers, hosts	
Management of diseases	4 hrs
Disease prevention, control, elimination and eradication	
Drinking water microbiology	10 hrs
Types of water, safe drinking water, physico-chemical and microbiological parameters of water quality, biological indicators of water pollution, national and WHO guidelines for drinking water quality standards, principle and procedures of drinking water treatment for large water supply system, methods for monitoring water quality	
Waste management	10 hrs
Introduction, solid waste and its types, solid waste management, sewage and industrial effluents, composition and microbiology of sewage, methods for the treatment of waste water	
Microbial air pollution	6 hrs
Introduction, methods of enumeration and identification of microorganisms in air (indoor and out door), indicator microorganisms of air pollution, air-borne diseases, air-pollution control	
Historical background of medical microbiology	2 hrs
Historical aspects of medical microbiology, major contributors in medical microbiology	
Normal flora of the human body	5 hrs
Normal flora of human body (skin, gastrointestinal tract, respiratory tract, genito-urinary tract), opportunistic pathogens	

Immunity process	30 hrs
Types of immunity, cells and tissues of immune system, antigens, immunoglobulins and their types, antigen antibody reactions, hypersensitivity	
Safety measures in clinical laboratory	5 hrs
Principles of laboratories safety, decontamination and safe disposal of contaminated materials, bio-safety level laboratories, risk and hazard group of microorganisms	
Methods of specimen collection, transportation, processing and culture of clinical samples for detection of pathogens	8 hrs
Cerebrospinal fluid, blood, sputum, urine, stool, other body fluids, pus and wound exudates, bacteria culture procedures, possible pathogens in different clinical specimens, maintenance of temperature and transportation of samples, culture of virus in chick embryo and cell lines, cytopathic effects, detection of virus from culture, serological tests for the diagnosis of viruses, samples for fungal infections, types of samples for parasite detection, sample processing and detection methods for blood, intestinal and tissue parasites	
Common pathogenic bacteria	18 hrs
Biology, epidemiology of diseases/infections, diagnostic methods, prevention and control of: <i>Mycobacterium</i> , <i>Staphylococcus</i> , <i>Streptococcus</i> , <i>Escherichia coli</i> , <i>Salmonella</i> , <i>Shigella</i> , <i>Vibrio</i> , <i>Rickettsia</i> , <i>Treponema</i> , <i>Clostridium</i>	
Common pathogenic viruses	18 hrs
Biology, epidemiology of diseases/infections, diagnostic methods and prevention and control of: Herpes viruses, hepatitis viruses, measles virus, influenza virus, HIV, rotavirus, polio virus, rabies virus, Japanese encephalitis virus, dengue virus	
Medically important fungi	8 hrs
Introduction, classification, and characteristics of medically important fungi and yeasts Biology, infections, diagnostic methods, prevention and control of: Dermatophytes, <i>Aspergillus</i> , <i>Histoplasma</i> , <i>Candida</i> , <i>Cryptococcus</i>	
Common pathogenic parasites	15 hrs
Biology, epidemiology of diseases/infections, diagnostic methods and prevention and control of: <i>Entamoeba</i> , <i>Giardia</i> , <i>Plasmodium</i> spp., <i>Wuchereria bancrofti</i> , <i>Leishmania</i> spp., <i>Taenia</i> , <i>Ascaris lumbricoides</i> , <i>Ancylostoma duodenale</i>	

Recommended Readings

1. Park K (2008). Park's Textbook of Social and Preventive Medicine. 18th Edition.
2. Gordis L (2004). Epidemiology, 3rd Edition, Elsevier Saunders.
3. Maier RM, Pepper IL and Gerba CP (2006). Environmental Microbiology. Academic Press, Elsevier Publication.
4. Cheesbrough M (2007). Medical Laboratory Manual for Tropical Countries Vol. 2 ELBS London.
5. Tille P (2014). Bailey & Scott's Diagnostic Microbiology (13th edition). Elsevier.
6. Collee JG, Fraser AG, Marmion BP and Simmons A (1996). Mackie & McCartney Practical Medical Microbiology (14th edition). Churchill Livingstone.
7. Greenwood D, Slack RCB and Peutherer J (2001). Medical Microbiology ELBS, Dunclue Livingstone.
8. Pelczar MJ, Chan ECS and Krieg NR (1993). Microbiology 5th edition, Tata McGraw Hill, New Delhi.

Medical and Public Health Microbiology Practical

Description of the Course

Course Title: Medical and Public Health Microbiology Practical

Course No: MB 404 (Major)

Marks: 20

Nature of the Course: Practical

Full Marks: 50

Pass

Year: IV

Total Lecture Hours: 180

Course Objectives

After completion of the course, the students will be able to:

- a) conduct analysis of environmental samples.
- b) perform field level tests for the diagnosis of diseases.
- c) collect, transport, and process the clinical samples for the diagnosis of microbial diseases.

Course Contents

To perform bacteriological examination of drinking water: Most Probable Number (MPN), membrane filter (MF) methods, physico-chemical parameters of water tests, DO, BOD, COD, residual chlorine, ammonia, nitrate/nitrite, sulphate, chloride, iron

To demonstrate water treatment station: Field visit to water treatment station and report submission

To assess air pollution: Air microbes in indoor and outdoor environments

To perform rapid diagnosis of viral diseases using test kits: HIV, hepatitis B, hepatitis C, rotavirus

To understand the disease reporting system of Nepal: Visit to District Public/Health Office and report submission

To demonstrate safety precautions in microbiology laboratories: Demonstrate various safety measures and precautions to be taken in the laboratories

To collect and transport various clinical specimens: Blood, urine, stool, sputum, swabs

To perform different staining techniques: Gram's staining, capsule staining, spore staining, Ziehl Neelson, Albert stain, Giemsa stain

To prepare different culture media and monitoring their quality: Nutrient agar, blood agar, MacConkey agar, chocolate agar, SS agar, XLD agar, MSA, anaerobic culture medium

To prepare biochemical media and reagents for identification of bacteria: MR test, VP test, citrate test, urease test, SIM test, indole test, O/F test, TSI/KIA.

To differentiate different types of bacteria from biochemical tests: Carbohydrate utilization test, Nitrate reduction test, interpretation of the result

To perform enzymatic test of the bacteria: Perform important enzymatic tests, coagulase test, catalase test, oxidase test, DNase test, Gelatin, Casein and lipid hydrolysis.

To perform antibiotic susceptibility test of the bacterial isolates

Serotyping and molecular typing of medically important bacteria

To demonstrate serological tests: Rapid diagnostic test kits, ELISA, hemagglutination test

To learn various sample collection techniques for diagnosis of mycotic infections: Skin scrapping, nails clipping, sputum collection, hair plucking

To prepare fungal culture media: Preparation of media; Sabouraud dextrose agar, potato dextrose agar, malt extract agar

To detect the fungi by direct microscopic methods: Detection of fungal elements: KOH preparation, iodine preparation, India ink preparation, lacto-phenol cotton blue staining

To examine the samples for intestinal and tissue parasites: *Ascaris*, *Entamoeba histolytica*, *Giardia lamblia*, *Plasmodium* spp., *Leishmania* spp.

Methods in Microbiology

Description of the Course

Course Title: Methods in Microbiology
Course No: MB 405 (Applied Microbiology)
Nature of the Course: Theory

Full Marks: 100
Pass Marks: 35
Year: IV
Total Lecture Hours: 150

Course Objectives

After completion of the course, the students will be able to:

- a) understand the principles, procedures and applications of methods used in the fields of microbiology

Course Contents

Safety measures in microbiology laboratory

10 hrs

Principles of laboratories safety, biosafety level of laboratories and bio-hazards, risk group of microorganisms, decontamination and safe disposal of contaminated materials, sterilization and sterility techniques

Methods of specimen collection, transportation and processing of clinical samples for bacteria detection

20 hrs

Cerebrospinal fluid, blood and other body fluids, sputum, urine, discharges and pus, stool, culture procedures, test algorithms for diagnosis of bacteria, antibiotic susceptibility tests (Kirby Bauer disc diffusion method, minimum inhibitory concentration determination)

Method of collection, transportation and processing of clinical samples for virus detection

20 hrs

Introduction, types of samples, maintenance of temperature and transportation, identification and interpretation, culture of virus in chick embryo and cell lines, cytopathic effects, detection of virus from culture, serological tests for the diagnosis of viruses

Sample collection and laboratory diagnosis of mycotic infections

10 hrs

Samples for fungal infections, sputum, nasal swab, skin scraping, hair and nails, CSF for fungal meningitis, microscopy, staining, culture

Method of collections of samples and processing for detection of parasites

15 hrs

Introduction, types of samples for parasite detection, sample processing and detection methods for blood, stool and tissue parasites

Immunological and serological tests

15 hrs

Principles, procedures, advantages and applications of precipitation, agglutination, complement fixation, ELISA, radio-immunoassay

Method of collections of water samples and microbiological analysis **10 hrs**

Introduction, types of water samples, water sample processing and detection methods, MPN, MF, BOD

Field level tests for disease diagnosis **10 hrs**

Principles, procedures and applications of rapid tests for malaria, kala-azar, lymphatic filariasis, dengue, HIV, HBV, HCV, rotavirus, JE, leptospirosis, typhoid

Molecular tests in microbiology laboratories **15 hrs**

Samples for molecular diagnostic tests, DNA/RNA extraction, PCR, Real Time PCR, PCR-RFLP, sequencing, western blotting

Microbiological quality tests of foods **15 hrs**

Quality test of milk and milk products, egg and egg products, meat and meat products, cereal and cereal products, HACCP, detection methods of carcinogens and toxins in food

Microbiology laboratory in agriculture **10 hrs**

Methods for preparation of bio-fertilizers, detection methods of pesticide, herbicide, insecticide, fungicide in soil, isolation and detection of pectinolytic, lignolytic, lipolytic, cellulolytic microorganisms from soil

Recommended Readings

Text books

1. Cheesbrough M (2007). Medical Laboratory Manual for Tropical Countries Vol. 2 ELBS London.
2. Brown AE (2012). Benson's Microbiological Applications. Laboratory Manual in General Microbiology. (12th edition). McGraw-Hill Publisher.
3. Collee JG, Fraser AG, Marmion BP and Simmons A (1996). Mackie & McCartney Practical Medical Microbiology (14th edition). Churchill Livingstone.

Project Writing and Presentation

Description of the Course

Course Title: Project Writing and Presentation

Course No: MB 406 (Major)

Nature of the Course: Project work

Full Marks: 100

Pass Marks: 40

Year: IV

Total Lecture Hours: 150

Course Objectives

After completion of the course, the students will be able to:

- a) carry out laboratory based mini research
- b) develop knowledge and skills in writing scientific research report

Course Contents

Students in group will be assigned relevant research topics related to their study by concerned department/campus. Students will perform laboratory experiments within fourth academic year. The research will be supervised by faculty member(s) of microbiology of concerned department/campus. After completion of laboratory work, the student should write the research report in standard format on the basis of data/findings generated during the laboratory works. The student will submit required number of copies of their research report to concerned department or campus for evaluation. The final evaluation of the project work will be made by a panel of external and internal examiners, head of the department and supervisor(s).

Bioinstrumentation

Description of the Course

Course Title: Bioinstrumentation
Course No: MB 407 (Interdisciplinary subject)
Nature of the Course: Theory

Full Marks: 50
Pass Marks: 17.5
Year: IV
Total Lecture Hours: 75

Course Objectives

After completion of the course, the students will be able to:

- a) understand the working principles and procedures of instruments used in microbiology laboratory

Course Contents

Buffers	2hrs
Physiological solutions and buffers, importance of buffers in biological researches	
General principle and approaches of biochemical investigations	8hrs
Introduction, concept, significance of disruption of cells, extraction buffer, chemical methods, enzymatic digestion, mechanical methods, selection of protein source, methods of solubilization of protein, solubility of protein, chromatographic separations, electrophoresis, ultracentrifugation of protein samples	
Working principle, instrumentation and application of	10hrs
Phase contrast, electron microscopy, fluorescence microscopy	
Working principle, instrumentation and applications of	15 hrs
Centrifugation techniques, Electrophoretic techniques: Agarose gel, Polyacrylamide gel electrophoresis, Isoelectric focusing, Two- dimensional electrophoresis	
Principles, instrumentation and uses of chromatographic techniques	20 hrs
Ion exchange chromatography, Affinity Chromatography, Paper and Thin layer chromatography, Gel Permeation chromatography, Gas Chromatography, High performance Liquid Chromatography (HPLC)	
Principle, instrumentation and application of	10 hrs

Lambert Beer's law, X-ray and gamma ray spectroscopy, spectrofluorometry, optical rotation dispersion, electron spin resonance (ESR), Infrared spectroscopy, nuclear magnetic resonance spectroscopy, mass spectrometry, ultraviolet and visible spectroscopy

Principle, instrumentation and application of molecular techniques

10 hrs

Thermocycler, Probes, Sequencer, Gel documentation system, Microarray

Recommended Readings

Text books

1. Skoog DA, Holler FJ and Nieman TA (2005), Principles of Instrumental Analysis, 5th Edition, Thomson Books/Cole
2. Wilson K and Walker J (Eds) (2005), Principles and Techniques of Biochemistry and Molecular Biology, 6th Edition, Cambridge University Press
3. Mendham J, Denny RC, Barnes JD and Thomas M (2008), Vogel's Text Book of Quantitative Chemical Analysis, 6th Edition, Pearson Education .