

Tumkur University, Tumakuru
Department of Studies and Research in Microbiology

PART-A

Research Methodology (Microbiology)

Unit 1: Research Writing and Biostatistics

Empirical science; scientific method; manipulative experiments and controls; deductive and inductive reasoning; descriptive science; reductionist vs. holistic biology; research methods vs. methodology; types of research, data collection and analysis; research ethics and IPR; Concept of plagiarism, preparation of research; choosing a mentor, lab, and research question; research problem; literature review; hypothesis; gaps in research; research reports; publication of research. Types of reports, layout of a formal report, and scientific writing skills.

Introduction, scope, and application of biostatistics. Concepts of primary and secondary data. Methods of collection and editing of primary and secondary data. Designing a hypothesis and schedule. Measures of central tendency mean, median, mode, geometric mean, and harmonic mean. Measures of dispersion: range, quartile deviation, mean deviation, and standard deviation. Data graphics construction and labeling of graphs, histograms, pie charts, scatter plots, semilogarithmic plots. Basics of testing hypothesis related to statistics: null hypothesis, level of significance, power of test, p value, statistical estimation of confidence intervals. Level of significance: parametric data Student's t test (paired and unpaired), chi square test, analysis of variance (one way and two way); software: SPSS, Epi Info, SAS.

Unit 2: Microbial Research Instrumentation

Principles and applications of chromatography (TLC, column chromatography, ion exchange chromatography, affinity chromatography, gas chromatography, HPLC). Electrophoresis: agarose gel electrophoresis and SDS PAGE. Spectroscopy: UV visible spectroscopy, fluorescence spectroscopy, infrared spectroscopy, NMR, ESR. Radiography: autoradiography. Protein purification methods, mass spectrometry, X ray diffraction.

Analytical bacteriology: microscopy components of microscopes, basic principles, types of microscopes, and their applications. Working and applications of transmission electron microscope (TEM), scanning electron microscope (SEM), STEM, atomic force microscope (AFM), and electron microscopy.

Analytical virology: isolation and cultivation of viruses in embryonated eggs, experimental animals, and cell cultures primary, secondary, and continuous cell cultures. Assay of viruses principle, procedure, merits, and demerits. Physical assay electron microscopy; biological assay plaque assay; serological assay ELISA, RIA, Western blot.

Unit 3: *In silico* methods for Microbial Research

- Introduction, scope, and application of bioinformatics. Biological databases: types of databases nucleic acid sequence databases, protein sequence databases; database searching using BLAST and FASTA; structure databases, viral databases, immunodatabases, genome databases, and gene expression databases. Sequence alignment introduction, principle, and types of alignment; matrices of alignment DNA and protein matrices; multiple sequence alignment; structural alignment (DALI and SSAP). Genome annotation identification of genes (promoter, ribosome binding sites, initiation codons, intron exon boundaries in a gene, splice sites, termination codons), CpG islands, repetitive elements, DNA barcoding, phylogenetic analysis.

PART-B

Cognate Subject: Microbiology

Unit 4: Cell Biology

Prokaryotic cell structure and functions of cell wall, flagella, cilia, pili, fimbriae, periplasmic space, gas vesicles, chlorosomes, carboxysomes, magnetosomes, and phycobilisomes. Capsule structure, composition, and properties. Cell wall chemical composition, characteristics, and function (Gram positive and Gram negative bacteria: lipoproteins, lipopolysaccharides, matrix proteins); plasma membrane (fluid mosaic model), function of cell membrane; mesosomes, cytoplasm, ribosomes, subunits, and chemical composition; molecular chaperones, nucleoids, plasmids, cytoplasmic inclusions, endospores.

Eukaryotic cell: structural organization of endoplasmic reticulum, Golgi apparatus, lysosomes, peroxisomes, ribosomes, cellular cytoskeleton, mitochondria, chloroplasts and their genetic organization; nuclear compartment: nucleus, nucleolus, and chromosomes. Protein trafficking; cell cycle and its regulation; cell division: mitosis, meiosis, and cytokinesis. Study of cell cycle using FACS.

Unit 5: Biomolecules and Microbial Systematics

Biomolecules: carbohydrates general properties, structure, classification, functions, and examples of metabolism. Lipids classification, structure, properties, functions, and examples of metabolism. Proteins structure (primary, secondary, tertiary, and quaternary structure); peptide synthesis: chemical and Merrifield synthesis. Nucleic acids structure, a historical perspective leading up to the proposition of DNA double helical structure; difference between RNA and DNA structure and their importance in the evolution of DNA as the genetic material.

Definition of systematics, nomenclature rules, hierarchical organization. Classification systems artificial and phylogenetic dendrogram. Haeckel's three kingdom classification, Whittaker's five kingdom approach, three domain classification of Carl Woese. Major characteristics used in taxonomy morphological, physiological, metabolic, genetic, and molecular characteristics; comparison of proteins, nucleic acid hybridization, nucleic acid sequence comparison, DNA and RNA homology, G+C ratio, significance of rRNA in microbial taxonomy, and chemotaxonomy.

Unit 6: Microbial Diversity

Bacteriology: general characteristics, occurrence, shape, and arrangement of bacterial cells. Major types of bacterial classification morphological, phenotypic, genotypic, numerical, and analogical based classification. Taxonomic groups of bacteria based on Bergey's Manual; general characteristics, classification, ultrastructure, reproduction, and economic importance of eubacteria, actinomycetes, cyanobacteria, mycoplasma, rickettsia, chlamydia, photosynthetic bacteria, and bioluminescent bacteria.

Virology: brief outline on discovery of viruses, distinctive properties of viruses; biological properties of viruses host range, transmission (vector and non vector); physical properties of viruses morphology, structure. Biochemical characteristics chemical composition of viruses (proteins, nucleic acids, envelope, enzymes, lipids, carbohydrates), multiplication of viruses. Morphology and ultrastructure capsids and their arrangements; types of envelopes and their composition; ICTV nomenclature and classification of viruses. Major characteristics of different virus families viroids and prions, animal and plant viruses, bacteriophages.

Mycology: history and development of mycology, general characteristics, distribution, and ultrastructure of fungal cells, nutrition in fungi, reproduction of fungi vegetative, asexual, and sexual; fungal spores and fruiting bodies. Interaction between fungi and other organisms. Major fungal classification methods: general characteristics, structure, and life cycle of Oomycetes, Zygomycetes, Ascomycetes, Basidiomycetes, and Deuteromycetes.

Unit 7: Environmental Microbiology

Air microbiology: factors affecting air spora; important airborne pathogens and toxins. Techniques for trapping airborne microorganisms: vertical cylinder, Rotorod sampler, Burkard sampler, Andersen sampler, Hirst's trap, filtration, electrostatic precipitation, thermal precipitation, gravitational settling, impingement. Air sanitation: methods and applications.

Soil microbiology: characteristics and classification of soil. Interactions between microorganisms: mutualism, commensalism, amensalism, synergism, parasitism, predation, competition. Rhizosphere microflora and its beneficial activity. Biogeochemical cycles: carbon, hydrogen, nitrogen, phosphorus, sulfur, iron, and manganese cycles. Detrimental effects of diverted biogeochemical cycles. Biological nitrogen fixation in detail: symbiotic, asymbiotic, and associative nitrogen fixation. Structure, function, and genetic regulation of nitrogenase. Viable but non culturable microorganisms; soil microbiome, geomicrobiology, and its environmental applications.

Aquatic microbiology: distribution of microorganisms in freshwater and marine ecosystems (estuaries, mangroves, deep sea, hydrothermal vents, salt pans, coral reefs). Zonation of water ecosystem. Effect of aquatic microorganisms on ecosystem. Water pollution sources, source tracking of aquatic pollutants, biological indicators of water pollution, determination of sanitary quality of water. Wastewater microbiology primary, secondary, tertiary treatment, and reclamation of wastewater. Microbial assessment of potable water. Advances in potable water purification. Biofilms: types, mechanism, and significance.

- Extremophiles: introduction to extremophiles, isolation, classification, and properties of extremophiles (thermophiles, hyperthermophiles, psychrophiles, halophiles, acidophiles, methanogens, etc.). Adaptation mechanisms of extremophiles, biological applications of extremophiles, genome analysis from extremophiles protein stability in hyper extremophiles. Hyper extremophiles and their novel metabolic machinery and biomolecules unique applications.

Unit 8: Molecular Biology and Genetic Engineering

Genome structure and organization: definition and organization of viral, prokaryotic, and eukaryotic genomes; genome organization in mitochondria and chloroplasts; gene concept; unit of function; structure of chromatin, nucleosome, chromatin organization and remodeling; higher order organization chromosome, centromere, telomere. Histones and their effect on structure and function of chromatin. Recombination: homologous and site specific recombination; models for homologous recombination Holliday junction, NHEJ; proteins involved in recombination RecA, RuvA, RuvB, RuvC; gene conversion. rII locus and complementation analysis. Gene function: one gene one enzyme hypothesis.

Genetic engineering: impact of genetic engineering in modern society; general requirements for performing a genetic engineering experiment; restriction endonucleases and methylases; DNA ligase, Klenow enzyme, T4 DNA polymerase, polynucleotide kinase, alkaline phosphatase; cohesive and blunt end ligation; linkers, adaptors, homopolymeric tailing; labeling of DNA: nick translation, random priming, radioactive and non radioactive probes; hybridization techniques: Northern, Southern, Southwestern, far Western, and colony hybridization; fluorescence in situ hybridization. Determination of stringency conditions. Applications of nucleic acid hybridization.

Unit 9: Immunology and Medical Microbiology

Immune system and immune response: history and scope of immunology; innate and acquired immunity; structure and functions of immune cells T cells, B cells, macrophages, NK cells, dendritic cells, eosinophils, neutrophils, mast cells. Organs of immune system primary and secondary lymphoid organs. Immunoglobulins basic structure, classes, and subclasses. Antigen: types, properties, and functions. Primary and secondary immune response. Clonal selection theory.

Scope and importance of medical microbiology. Modern diagnostic microbiology techniques (overview). Normal human microbiota and its importance. Host microbe interactions. Opportunistic pathogens and infections. Sources of microbial infections. Routes of transmission of infectious agents. Microbial pathogenicity and virulence factors. Overview of communicable and non communicable diseases. Bacterial infectious diseases tuberculosis, typhoid, cholera, pneumonia, and urinary tract infections with causative organisms. Viral infectious diseases hepatitis, influenza, HIV/AIDS, rabies, and respiratory viral infections. Fungal infectious diseases candidiasis, aspergillosis, and dermatophytic infections affecting skin, nails, and mucous membranes. Hospital acquired (nosocomial) infections causes, common pathogens, transmission in hospital settings, and prevention measures.

Unit 10: Applications of Microbiology

a) Food microbiology: introduction to food processing of various foods, including dairy, bakery, brewing, fruit and vegetable products, plantation products; gut microbiome, probiotics, prebiotics, and nutraceuticals. Membrane technology: introduction to pressure activated membrane processes microfiltration, UF, NF, and RO and their industrial applications. High pressure processing: concept and its application in food processing. Principles of food preservation, hurdle technology.

b) Agricultural microbiology: historical development, scope, and concept of agricultural microbiology. Soil: composition and profile; physical, chemical, and biological properties. Distribution of microorganisms in soil and their importance in maintaining soil fertility; factors affecting soil microflora. Plant microbe interactions: rhizosphere, rhizoplane, PGPR, PGPF, PGPA, endophytes, phyllosphere, spermosphere, and mycorrhizal association. Phosphate solubilizing microbes and microbiome for sustainable agriculture.

c) Pharmaceutical microbiology: government regulatory practices and policies; regulatory aspects of quality control. Sterilization control and sterility testing chemical and biological indicators. Regulatory authorities for introduction of medicines in the market role of Food and Drug Administration; FDA guidelines for drugs/biologicals; validation (GMP, GLP, GCP, etc.). Clinical studies: Phase I, Phase II, Phase III, and Phase IV of clinical trials objectives, conduct of trials, outcome of trials.



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