M.Sc. Microbiology Kumaun University, Nainital *New* Curricula/Syllabi (<u>Note:</u> New Students taking admission (in <u>first semester)</u> after July 2017 will follow this syllabus)

SEME	STER – I			Credits
1S1	Biochemistry			3 Credits
1S2	Cell & Developmental Biology			3 Credits
1S3	Molecular Biology			3 Credits
1S4	Microbiology and Industrial Ap	plications		3 Credits
1S5	Biostatistics and Computer Ap	plications		3 Credits
1S6	Seminar/Journal Club/Assignm	ient		1 Credit
	Lab I- Biochemistry and Microb	piology		4 Credits
	Lab II- Molecular Biology	57		4 Credits
		Total		24 Credits
SEME	STER- II			
2S1	Immunology and Immunotechr	ology		3 Credits
2S2	Analytical Techniques	0,		3 Credits
2S 3	Genetic Engineering			3 Credits
2S4	Molecular Genetics			3 Credits
2SM1	Bacterial Metabolism			3 Credits
2S6	Seminar/Journal Club/Assignm	nent		1 Credit
	Lab III- Immunology			3 Credits
	Lab IV- Analytical Techniques			2 Credits
	Lab V- Genetic Engineering			3 Credits
		Total		24 Credits
SEMESTER-III				
3SM1	Applied Microbiology			3 Credits
3S2	Environmental Biotechnology			3 Credits
3S3	Animal Biotechnology			3 Credits
3SM2	Microbiological Techniques			3 Credits
3SM3	Molecular Virology			3 Credits
3S6	Seminar/Journal Club/Assignm	nent		1 Credit
	Lab VI: Microbiological Technic	ques		4 Credits
	Project Proposal Presentation	-		2 Credits
		Total		- 22 Credits
SEMESTER-IV				
	Project/Thesis Work			20 Credits
		Total		20 Credits

Marks- (75+25) =100

1S1 Biochemistry

- 3 Credits

Unit-I

Chemical basis of life: Composition of living matter; Water- properties, pH, pKa,_Titration curves of weak acids, Buffers, Handerson-Hasselbach equations, ionization and hydrophobicity; Emergent properties of biomolecules in water; Water as a reactant.

Unit-II

Proteins: Amino acids as building blocks of proteins and their chemical properties, pl and pKa values, Primary, Secondary, Tertiary and Higher order structure of Proteins, Protein Sequencing, Ramchandran Plot, Conjugated proteins- Glycoproteins, Lipoproteins, Heamproteins.

Unit-III

Enzymes: General principles of catalysis, Quantitation of enzyme activity and efficiency, Enzyme characterization and Michaelis-Menten kinetics, Relevance of enzymes in metabolic regulation, activation, inhibition and covalent modification; Single substrate enzymes

Unit-IV

Carbohydrates: Mono- Di- and Polysaccharides, Optical isomerism, Structure of Carbohydrates, Glycolysis, Gluconeogenesis, Pentose phosphate pathways, Citric acid cycle.

Unit V

Lipids: Classification and structural analysis of fatty acids, Glycerols, Waxes, Phospholipids, Sphingolipids, Sterols, Lipoproteins, β -oxidation, Glycolipids. Biosynthesis of Cholesterol and Fatty acids.

Unit- VI

Nucleic acids: Biosynthetic pathways of purines and pyrimidines, degradation pathways

Unit-VII

Bioenergetics- Basic principles; Equilibria and concept of free energy; Group transfer, concept of Entropy, Enthalpy and free energy, Oxidation and Reduction reactions, Electron Transport Chain, Oxidative phosphorylation; photosynthesis. Metabolic regulations including the role of hormones.

Marks- (75+25) =100

-3 Credits

1S2 Cell and Developmental Biology Unit- I

Cell Theory and Methods of Study

Microscope and its modifications- Light, phase contrast and interference, Fluorescence, Confocal, Electron (TEM and SEM), Electron tunneling and Atomic Force Microscopy, etc.

Membrane Structure and Function

Structural models; Composition and dynamics; Transport of ions and macromolecules; Pumps, carriers and channels; Endo- and Exocytosis; Membrane carbohydrates and their significance in cellular recognition; Cellular junctions and adhesions; Structure and functional significance of plasmodesmata.

Unit- II Organelles

Cellular compartments and intracellular sorting of proteins, ER & Lysosomes, peroxisomes, synthesis and sorting of proteins (lysosomal proteins, membrane proteins, secretory proteins). Nuclear transport.

Unit-III

Endo-membrane System and Cellular Motility

Organization of nucleus and nuclear membrane, structure and organization of chromatin. Cytoskeleton: Actin filaments and cell cortex, cilliary movements and cytoplasmic microtubules and intermediate filaments.

Unit IV

Cell Communication

Endocrine, Exocrine and synaptic signaling, surface and intracellular receptors, G proteins and generation of secondary messengers, mode of action of cAMP, Ca²⁺, calmodulin.

Unit-V

Differentiation of specialized cells

Stem cell differentiation. Differentiation of cancerous cells and role of proto-oncogenes *Plant Meristem Organization and Differentiation*

Organization of shoot Apical Meristem (SAM); Organization of Root Apical Meristem (RAM); Pollen germination and pollen tube guidance; Phloem differentiation; Self-incompatibility and its genetic control; Embryo and endosperm development; Heterosis and apomixes.

1S3 Molecular Biology

Unit-I Genome Organization

Organization of bacterial genome; Structure of eukaryotic chromosomes; Role of nuclear matrix in chromosome organization and function; Matrix binding proteins; Heterochromatin and Euchromatin; DNA reassociation kinetics (Cot curve analysis); Repetitive and unique sequences; Satellite DNA; DNA melting and buoyant density; Nucleosome phasing; DNase I hypersensitive region; DNA methylation & Imprinting.

Unit-II DNA Structure; Replication; Repair & Recombination

Structure of DNA-A-,B-, Z- and triplex DNA; Measurement of properties-Spectrophotometric, CD, AFM and Electron microscope analysis of DNA structure; Replication initiation, elongation and termination in prokaryotes and eukaryotes; Enzymes and accessory proteins; Fidelity; Replication of single stranded circular DNA; Gene stability and DNA repair- enzymes; Photoreactivation; Nucleotide excision repair; Mismatch correction; SOS repair; Recombination: Homologous and non-homologous; Site specific recombination; Chi sequences in prokaryotes; Gene disruption; FLP/FRT and Cre/Lox recombination.

Unit III Prokaryotic & Eukaryotic Transcription

Prokaryotic Transcription; Transcription unit; Promoters- Constitutive and Inducible; Operators; Regulatory elements; Initiation; Attenuation; Termination-Rho-dependent and independent; Anti-termination; Transcriptional regulation-Positive and negative; Operon concept- lac, trp, ara, his, and gal operons; Transcriptional control in lambda phage; Transcript processing; Processing of tRNA and rRNA

Eukaryotic transcription and regulation; RNA polymerase structure and assembly; RNA polymerase I, II, III; Eukaryotic promoters and enhancers; General Transcription factors; TATA binding proteins (TBP) and TBP associated factors (TAF); Activators and repressors; Transcriptional and post-transcriptional gene silencing

Unit-IV Post Transcriptional Modification

Processing of hnRNA, tRNA, rRNA; 5'-Cap formation; 3'-end processing and polyadenylation; Splicing; RNA editing; Nuclear export of mRNA; mRNA stability; Catalytic RNA.

Translation & Transport

Translation machinery; Ribosomes; Composition and assembly; Universal genetic code; Degeneracy of codons; Termination codons; Isoaccepting tRNA; Wobble hypothesis; Mechanism of initiation, elongation and termination; Co-and post-translational modifications; Genetic code in mitochondria; Transport of proteins and molecular chaperones; Protein stability; Protein turnover and degradation

Unit-V Mutation; Oncogenes and Tumor suppressor gene

Nonsense, missense and point mutations; Intragenic and Intergenic suppression; Frameshift mutations; Physical, chemical and biological mutagens; Transposition- Transposable genetic elements in prokaryotes and eukaryotes; Mechanisms of transposition; Role of transposons in mutation; Viral and cellular oncogenes; Tumor suppressor genes from humans; Structure, function and mechanism of action of pRB and p53 tumor suppressor proteins; Activation of oncogenes and dominant negative effect; Suppression of tumor suppressor genes; Oncogenes as transcriptional activators.

Marks- (75+25) =100 -3 Credits

SEMESTER-I (M.Sc. Microbiology) Marks- (75+25) =100

1S4 Microbiology & Industrial Applications -3 Credits Unit I

Microbial Diversity & Systematics.

The Milestones in Microbiology: The discovery of microbial world by Antony van Leeuwenhocek, The controversy over spontaneous generation, Golden age of Microbiology. Criteria for classification of microorganism; Classification of Bacteria according to Bergey's manual; Molecular methods such as Denaturing Gradient Gel Electrophoresis (DGGE), Temperature Gradient Gel Electrophoresis (TGGE), Amplified rDNA Restriction Analysis and Terminal Restriction Fragment Length Polymorphism (T-RFLP) in assessing microbial diversity; 16S rDNA sequencing and Ribosomal Database Project.

Unit II

Microbial Growth & Physiology

Cell Structure and Functions: Prokaryote cell, size, shape and arrangement of bacterial cells, Cell wall, External and Internal structures to the cell wall of Eubacteria. Ultrastructure of Archaea (Methanococcus); Unicellular Eukaryotes (Yeast). Microbial growth: Batch, fed-batch, continuous kinetics, synchronous growth, methods of growth estimation, stringent response, Thermal death of a bacterial cell. Methods in Microbiology: Pure culture techniques, The theory and practice of sterilization, Principles of microbial nutrition, Construction of culture media, Enrichment of culture techniques, Pure culture and its maintenance

Unit III

Microbial Interactions and Infection

Host-pathogen interactions; Microbes infecting animals and plants; Disease reservoirs, Epidemiological terminologies, Infectious diseases transmission, Pathogenicity islands and their role in bacterial virulence.

Unit IV

Microbes and Environment

Salient features of extremophiles (halophiles, thermophiles, psychrophiles) archaeabacteria. Aerobic and Anaerobic bacteria, Phototrophic and Gliding bacteria, Prosthecate and budding bacteria. Ecological impacts of microbes; Symbiosis (Nitrogen fixation and ruminant symbiosis); Microbes and Nutrient cycles; Microbial communication system; Quorum sensing;.

Unit V

Industrial Applications

Role of microorganisms in natural system and artificial system. Scope and importance of Microbiology in Biotechnology. Microbial fuel cells; Prebiotics and Probiotics; Vaccines. Microbial processes-production, optimization, screening, strain improvement, for the production of ethanol, organic acids, antibiotics etc. Basic principles in bioprocess technology; Media Formulation; Sterilization; Batch and continuous sterilization systems; Bioprocess control and monitoring variables such as temperature, agitation, pressure, pH.

SEMESTER-I (M.Sc. Microbiology) Marks- (75+25) =100

1S5 Biostatistics and Computer Applications -3 Credits

- 1. Brief description and Tabulation of data and its graphical representation.
- 2. Measure of central tendency and description: Mean, Mode, Median, Range, Standard deviation, Variance, Idea of two types of errors and level of significance, Tests of significance (F and T test), Chi-Square tests.
- 3. Simple linear regression and Correlation.
- 4. Introduction of digital computers: Organizations, Low-level and High-level languages, Binary systems.
- 5. Flow charts and Programming techniques.
- 6. Introduction to data structures and data base concepts, Introduction to internet and its applications.
- 7. Introduction to MS-office software covering word processing, spread sheets and presentation software.
- 8. Introduction to Harvard graphics/Sigma plotter.
- 9. Computer oriented statistical techniques: Frequency table of single discrete variable. Bubble sort, Computation of mean, Variance and standard deviations, T-test, Correlation coefficient.
- 10. Bio-informatics- Internet access and using web search engines to access biological databases, sequence, structure and strain database, Secondary and sequence analysis of DNA, RNA and proteins.

Lab on Biochemistry and Microbiology

General Biochemistry (Practical)

- 1. Titration of Amino Acids.
- 2. Colorimetric determination of pKa.
- 3. Quantitative estimation of Proteins and Sugars.
- 4. Separation techniques- Centrifugation, Chromatography (Gel Permeation, Ion exchange, TLC, etc.)

Lab on Microbiology

- 1. Sterilization, disinfection, safety in microbiological laboratory.
- 2. Preparation of media for growth of various microorganisms.
- 3. Isolation and maintenance of organisms by plating, Streaking and Serial dilution methods- slants and stab cultures, Storage of microorganisms.
- 4. Gram Staining and enumeration of microorganisms.
- 5. Growth curve, measure of bacterial population by turbidometry and studying the effect of temperature, pH, carbon and nitrogen.
- 6. Assay of antibiotics production and demonstration of antibiotic resistance.
- Isolation and screening of industrially important microorganisms.
 Determination of thermal death point and thermal death time of microorganisms.

– 2 Credits

-4 Credits

Lab on Molecular Biology

-4 Credits

- 1. Plasmid DNA isolation and DNA quantitation
- 2. Restriction digestion
- 3. Preparation of competent cells
- 4. Agarose gel electrophoresis
- 5. Restriction Enzyme digestion of DNA
- 6. Purification of DNA from an agarose gel
- 7. DNA Ligation
- 8. Transformation of *E.coli* with standard plasmids, Calculation of transformation efficiency
- 9. Restriction mapping of recombinant plasmid.
- 10. Polymerase Chain reaction
- 11. RFLP analysis of the PCR product

Marks- (75+25) =100

– 3 Credits

2S1 Immunology and Immunotechnology

Unit I- Immunology- fundamental concepts and anatomy of the immune system

Components of innate and acquired immunity; Phagocytosis; Complement and Inflammatory responses; haematopoesis; Organs and cells of the immune system- primary and secondary lymphoid organs; Lymphatic system; Lymphocyte circulation; Lymphocyte homing; Mucosal and Cutaneous associated Lymphoid tissue. (MALT & CALT); Mucosal Immunity; Antigens and antigenicity – immunogens and immunogenicity, Immune modulators: Adjuvants, hapten-carrier system; Toxins and Toxoids.

Major Histocompatibility Complex – MHC genes, MHC and immune responsiveness and disease susceptibility.

Unit II- Immune responses generated by B and T lymphocytes

Immunoglobulins- basic structure, classes & subclasses of immunoglobulins, antigenic determinants (Epitopes); Antigen-Antibody interaction, affinity, cross reactivity, specificity, Multigene organization of immunoglobulin genes; B-cell receptor; Immunoglobulin superfamily; Basis of self –non-self discrimination; Generation of antibody diversity; T-cell receptors; Functional T Cell Subsets; Cell-mediated immune responses, ADCC Antigen processing and presentation- endogenous antigens, exogenous antigens, non-peptide bacterial antigens and super-antigens; Cytokines-properties, receptors and therapeutic uses.

Unit III- Antigen-antibody interactions

Precipitation, agglutination and complement mediated immune reactions; Antibodies as in-vitro and in-vivo probes; Advanced immunological techniques – RIA, ELISA, Western blotting, ELISPOT assay, Flow cytometry: Instrumentation and Applications; Identification of Immune Cells; Surface Plasmon resonance, Biosenor assays for assessing ligand–receptor interaction, CMI techniques- lymphoproliferation assay, Mixed lymphocyte reaction, Cell Cytotoxicity assays, Apoptosis.

Unit IV- Vaccine Technology

Principles of Immunization, Techniques for analysis of immune response. General Idea of Active and passive immunization; Live, killed, attenuated, sub unit vaccines; recombinant DNA and protein based vaccines, plant-based vaccines, reverse vaccinology; Peptide vaccines, conjugate vaccines; Hybridoma, antibody engineering - chimeric and hybrid monoclonal antibodies; Transfusion of Immuno-competent cells; stem cell therapy; Cell based vaccines.

Unit V-Clinical Immunology

Immunity to Infection : Bacteria, viral, fungal and parasitic infections (with examples from each group); Hypersensitivity – Type I-IV; Autoimmunity; Types of autoimmune diseases; Treatment of autoimmune diseases; Transplantation – Immunological basis of graft rejection; General Idea of Tumor immunology, Cancer immunotherapy; Immunodeficiency-Primary immunodeficiencies, Acquired or secondary immunodeficiencies.

2S2 Analytical Techniques

Marks- (75+25) =100 -3 Credits

Unit-I Basic Techniques

Buffers; Methods of cell disintegration; Enzyme assays and controls; Detergents and membrane proteins; Dialysis, Ultrafiltration and other membrane techniques.

Spectroscopy Techniques

Basic Principle, Instrumentation and Biological applications of: UV and Visible light absorption spectroscopy, Spectrofluorometry, CD and ORD, Atomic spectroscopy (Absorption and emission). Infrared spectroscopy, Raman Scattering, Application of FT-IR in the study of biomolecules, Nuclear Magnetic Resonance (NMR) spectroscopy, and EPR; Mass spectroscopy and mass analyzers like ion trap, quadrupole, magnetic sector, time of flight (ToF).

Unit-II

Chromatography Techniques

TLC and Paper Chromatography; Column chromatography Chromatographic methods for macromolecule separation-Gel permeation, Ion exchange, Hydrophobic, Reverse-phase and Affinity chromatography; HPLC and FPLC.

Electrophoretic Techniques

Theory and application of Polyacrylamide and Agarose gel electrophoresis; Native and SDS-PAGE electrophoresis; Capillary electrophoresis; 2D Electrophoresis; Disc gel electrophoresis; Gradient electrophoresis; Pulsed field gel electrophoresis

Unit III

Centrifugation

Basic principles; Mathematics & theory (RCF, Sedimentation coefficient etc); Types of centrifuge- Microcentrifuge, High speed & Ultracentrifuges; Preparative centrifugation; Differential & density gradient centrifugation; Application (Isolation of cell components); Analytical centrifugation.

Unit-IV

Radioactivity

Radioactive & stable isotopes; Radioactive decay; Units of radioactivity; Measurement of radioactivity; Geiger-Muller counter; Solid & Liquid scintillation counters (Basic principle, instrumentation & technique); Autoradiography; Applications of isotopes in biochemistry, Clinical application; Radioimmunoassay

Unit-V

Advanced Techniques

Protein crystallization; Enzyme and cell immobilization techniques;

Marks- (75+25) =100

2S3 Genetic Engineering

-3 Credits

Unit I Basics Concepts

DNA structure and properties; Restriction enzymes; DNA ligase, Klenow enzyme, T4 DNA polymerase, Polynucleotide kinase, Alkaline phosphate, cohesive and blunt end ligation; Linkers; Adaptors; Homopolymer tailing, Labeling of DNA: Nick translation, Random priming, Radioactive and Non radioactive probes, Hybridization technique: Northern, southern and colony hybridization, fluorescence in situ hybridization; Chromatin Immunoprecipitation; DNA Protein Interactions; electrophoretic shift assay, DNase I footprinting.

Unit II

Cloning Vectors

Plasmids; M13 mp vector; PUC19 and Bluescript vectors, Phagemids, Lambda vectors; Cosmids; Artificial chromosome vectors (YACs; BACs); Mammalian expression vectors & retroviral vectors; Prokaryotic Expression vectors with GST-, His- and MBP- tags; Affinity purification of recombinant fusion proteins; Inclusion bodies; Methodologies to reduce formation of inclusion bodies; Baculovirus vectors system, Plant based vectors, Yeast vectors.

Unit III

Cloning Methodologies

Insertion of Foreign DNA into Host Cells; Transformation; Construction of libraries; Isolation of mRNA and total RNA; cDNA and genomic libraries; cDNA and genomic cloning; Expression cloning; Phage display

Unit-IV

PCR and Its Applications

Primer design; Fidelity of thermostable enzymes; DNA polymerases; Types of PCR- multiplex, nested, reverse transcriptase, real time PCR, hot start PCR, colony PCR, cloning of PCR products; T-vectors; Proof reading enzymes; PCR in site specific mutagenesis; PCR in molecular diagnostics; Viral and bacterial detection.

Unit-V

Sequencing methods; Enzymatic DNA sequencing; Automated DNA sequencing; RNA sequencing; Chemical Synthesis of oligonucleotides; Introduction of DNA into mammalian cells; Transfection techniques; Gene silencing techniques; RNA interference and siRNA Gene knockouts and Gene Therapy; Somatic and germ-line therapy: in-vivo and ex-vivo.

Marks- (75+25) =100 -3 credits

2S4 Molecular Genetics

Unit I

Bacterial Mutants and mutations

Isolation; Useful phenotypes (auxotrophic, conditional, lethal, resistant); Mutation rate; Types of mutations (base pair changes; frameshift; insertions; deletion; tandem duplication); Reversion vs. suppression; Mutagenic agents; Molecular Mechanisms of mutagenesis; Assay of mutagenic agents (Ames test)

Gene transfer in bacteria

History; Transduction- generalized and specialized; Conjugation- F, F', HFr; F transfer; Hfrmediated chromosome transfer; Transformation- natural and artificial transformation; Merodiploid generation; Gene mapping; Transposable genetic elements; Insertion sequences; Composite and Complex transposons; Replicative and non-replicative transposition; Genetic analysis using transposons.

Unit II

Bacteriophages and Plasmids

Bacteriophage-structure; Assay; Lambda phage – genetic map, lysogenic and lytic cycles; Gene regulation; Filamentous phages such as M13; Plasmids – natural plasmids; their properties and phenotypes; Plasmid biology – copy number and its control; Incompatibility; Plasmid survival strategies; Antibiotic resistance markers on plasmids (mechanism of action and resistance); Genetic analysis using phage and plasmid

Unit III

Mendelian Genetics

Introduction to human genetics; Background and history; Types of genetic diseases; Role of genetics in medicine; Human pedigrees; Patterns of single gene inheritance-autosomal recessive; Autosomal dominant; X linked inheritance; Complicating factors – incomplete penetrance; variable expression; Multiple alleles; Co dominance; Sex influenced expression; Hemoglobinopathies – Genetic disorders of hemoglobin and their diseases.

Non Mendelian inheritance patterns

Mitochondrial inheritance; Genomic imprinthing; Lyon hypothesis; isodisomy; Complex inheritance-genetic and environmental variation; Heritability; Twin studies; Behavioral traits; Analysis of quantitative and qualitative traits.

Unit IV-

Molecular Genetics of Lambda

The genome packaging, replication and recombination, Regulation of Lytic and Lysogenic Cycles

Unit V

Gene mapping and human genome project

Physical mapping; linkage and association

Population genetics and evolution

Phenotype; Genotype; Gene frequency; Hardy Weinberg law; Factors distinguishing; Hardy Weinberg equilibrium; Mutation selection; Migration; Gene flow; Genetic drift;

SEMESTER-II (M.Sc. Microbiology) Marks- (75+25) =100

2SM1 Bacterial Metabolism

3 credits

- **Unit 1.**Detailed study of metabolic pathways involved in the release and dissimilation of substrates by heterotrophs and autotrophs with emphasis on the reactions of industrial and environmental concern.
- **Unit 2**.Thermodynamic considerations of biological reactions, mechanisms of ATP synthesis, Photosynthesis and photometabolism in eubacteria, Respiration: aerobic and anaerobic, electron transport chain and Metabolism of secondary metabolites
- **Unit 3**.Fermentation of lactic acid bacteria, ethanol fermenting organisms, propionic acid bacteria, butane-diol, butyric acid bacteria, enterobacteriaceae and clostridia
- **Unit 4**.Biochemistry of xenobiotics degradation; aliphatic, aromatic and polycyclic compounds and Heavy metal toxicity: biochemical and genetic basis, resistance
- **Unit 5.**Fixation of molecular nitrogen and regulation, Biochemistry of methanogenesis and Regulation: enzyme synthesis and enzyme activity.

SEMESTER-II (M.Sc. Microbiology) Lab on Immunology – 3 Credits

1. Preparation of human blood smear and identification of cells.

- 2. Determination of blood groups.
- 3. Determination of Rh antigen.
- 4. Estimation of antiserum by Mancini method.
- 5. Estimation of antiserum by Ouchterlony method.
- 6. Antiserum titer determination by ELISA.
- 7. DOT ELISA for the presence of specific antigen.
- 8. Immunization, Collection of Serum.
- 9. Immunoelectrophoresis.
- 10. Immunodiagnostics (Demonstration using commercial kits).

Analytical Techniques (Practical)

2 Credits

- 1. Paper Chromatography of amino acids.
- 2. T.L.C of lipids.
- 3. Isolation of plasmid DNA from *E.coli*.
- 4. Agarose gel electrophoresis of isolated plasmid DNA.
- 5. Extraction and purification of protein from plant and animals.
- 6. SDS PAGE of BSA and extracted proteins.

SEMESTER-II (M.Sc. Microbiology) Lab on Genetic Engineering - 3 Credits

- 1. Isolation of genomic DNA from E. coli
- 2. PCR amplification of bacterial/plant/animal-cell genomic region and analysis by agarose gel electrophoresis.
- 3. Preparation of plasmid DNA from *E.coli* DH5 α and gel analysis.
- 4. Restriction digestion of vector (gel analysis) with Restriction endonucleases
- 5. a. Vector and Insert ligation
 - b. Transformation in *E.coli* DH5α.
- 6. Plasmid isolation and confirming recombinant by PCR and RE digestion.
- 7. Transformation of recombinant plasmid in *E.coli* Laboratory strain.
- 8. Induction of recombinant protein with IPTG and analysis on SDS-PAGE.

9. Purification of protein on Ni-NTA/Glutathione/Mannose column and analysis of purified protein by SDS- PAGE.

SEMESTER-III (M.Sc. Microbiology) Marks- (75+25) =100

3SM1 Applied Microbiology

-3 Credits

Unit I

Study of microflora with special reference to silage, agroindustrial waste, environment, soil fertility and management.

Unit II

Scope and importance of microbiology as applied to environment and industry, Peteroleum and mining microbiology, Biopesticides and Microbiology of paints, films, pharmaceuticals and other stored products, fermented food products and Biotransformation of steroids

Unit III

Environmental quality; Biodegradation of waste and pollutants; (i) solid waste disposal, sanitary, landfills and composting (ii) Treatment of liquid waste, sewage treatment, (iii) treatment and safety of water supply, Role of microbes in bioremediation, genetic engineering and biotechnology, Microbial degradation of pesticides and hydrocarbons

Unit IV

Microbial deterioration of cotton, jute, coir, wool, leather and wood and methods of preservation, Microbiology of biogas generation, utilization of alternate sources of energy, Utilization of agroindustrial waste for microbial biomass and protein

Unit V

Soil fertility and management of agricultural soil: soil microflora and organic matter decomposition, rhizosphere, Soil-plant-microbe interactions and Biofertilizers.

Marks- (75+25) =100

3S2 Environmental Biotechnology Unit I

- 3 Credits

Introduction

Environment; Basic concepts; Resources; Eco system: plants, animals, microbes; Ecosystem management; Renewable resources; Sustainability; Microbiology of degradation and decay; Role of Biotech in environmental protection; Control and management of biological processes.

Unit II

Pollution

Environmental pollution; Source of pollution; Air, water as a source of natural resource; Hydrocarbons, substituted hydro carbons; Oil pollution; Surfactants; Pesticides; Measurement of pollution; Water pollution; Biofilm; Soil pollution; Radioactive pollution; Radiation; Ozone depletion; Green house effect; Impact of pollutants; Measurement techniques; Pollution of milk and aquatic animals.

Unit III

Control, remediation and management

Waste water collection; control and management; Waste water treatment; Sewage treatment through chemical, microbial and biotech techniques; Anaerobic processes; Anaerobic filters; Anaerobic sludge blanket reactors; Bioremediation of organic pollutants and odorous compounds; Use of bacteria, fungi, plants, enzymes, and GE organisms; Plasmid borne metabolic treatment; Bioaugmentation; Bioremediation of contaminated soils and waste land; Bioremediation of contaminated ground water; Macrophytes in water treatment; Phytoremediation of soil metals; Treatment for waste water from dairy, distillery, tannery, sugar and antibiotic industries.

Unit IV

Alternate source of energy

Biomass as source of energy; Bioreactors; Rural biotechnology; Biocomposting; Biofertilizers; Vermiculture; Organic farming; Bio-mineralization; Biofuels; Bioethanol and biohydrogen; Solid waste management.

Unit V

Environment and health in respect to genetics

Gene and environment; Effect of carbon and other nanoparticles upon health; Gene mutation; Genetic testing; Genetic sensors; Environmental pollution and children; Human biomonitoring.

SEMESTER-III (M.Sc. Microbiology) ELECTIVE PAPER –I Marks- (75+25) =100

3S3 Animal Biotechnology

- 3 Credits

Unit I- Animal cell culture

History of animal cell culture; Basic requirements for animal cell culture; Cell culture media and reagents; Animal cell, tissue and organ cultures; Primary culture, secondary culture; Continuous cell lines; Suspension cultures; Transfection and transformation of cells; Stem cells and their application; Induced Pluripotency, Application of animal cell culture for in vitro testing of drugs; Application of cell culture technology in production of pharmaceutical proteins.

Unit II- Animal health Biotechnology

Recombinant approaches to vaccine production; Hybridoma technology; Phage display technology for production of antibodies; Antigen-antibody based diagnostic assays including radioimmunoassay and ELISA; Immunoblotting; Nucleic acid based diagnostic methods including nucleic acid probe hybridization; PCR, Real time PCR; Branched DNA technology, Nucleic acid sequencing; Animal disease diagnostic kits; Probiotics.

Unit III-Animal Reproductive Biotechnology

Cryopreservation of sperms and ova of livestock; Artificial insemination; Super ovulation; in vitro fertilization; Culture of embryos; Cryopreservation of embryos; Embryo transfer; Micromanipulation of animal embryos; Transgenic animal technology and its different applications; Different methods of Transgenic animal production; Targeted gene transfer, Detection of transgene and transgene function; Animal cloning- basic concepts; Cloning from embryonic cells and adult cells; Ethical, social and moral issues related to cloning; in situ and ex situ preservation of germplasm, Pregnancy diagnostic kits.

Unit IV-Animal genomics

Introduction to animal genomics; Different methods for characterization of animal genomes, SNP, STR, QTLS, RFLP, RAPD, proteomics, metobolomics; Genetic basis for disease resistance; Gene knock out technology and animal models for human genetic disorders.

Unit V-DNA Forensics

Immunological and nucleic acid based methods for identification of animal species; DNA Barcoding; Detection of adulteration in meat using DNA based methods; Detection of food/feed adulteration with animal protein; Identification of wild animal species using DNA based methods; Microbial forensics; Bioterror agents; Biocrimes and Bioterrorism.

SEMESTER-III (M.Sc. Microbiology) Marks- (75+25) =100

3SM2 Microbiological Techniques -3 Credits

Unit I

Experiments designed to familiarize students with the handling, identification and characterization of microorganisms and their activities particularly those of interest to man from different habitats.

Unit II

Introduction to terms and equipments used in Microbiological laboratory, safety precautions, Microscopy: Microscope and its operations, components, types. Staining: types of dyes, preparation, staining techniques, their applications, Motility (Hanging drop method), Cell cytometry

Unit III

Preparation of culture media-nutritional needs of microbes-dehydrated-selective-differentialautotrophic-heterotrophic, Culture techniques-adjustment of pH, buffers, pure culture techniques, prepartion of slants, sub-culturing, Preservation of microorganisms-slants, mineral oil, paraffin, Norris-bead method, lyophilization etc.

Unit IV

Isolation of pure microbial flora from natural and extreme environments-air, soil, water, food, Halophilic, thermophilic, psychrophilic and Acidophilic, Microbial growth measurements-Direct and indirect methods-cell count, turbidity measurement, percentage transmission, optical density, serial dilution, standard plate count, haemocytometry

Unit V

Biochemical characterization of bacteria-BIOLOG plant method, carbohydrate fermentation, catalase, peroxidase, indole, methyl red, vogus-prausker, citrate utilization test (IMViC) etc. Assignment: Identification of unknown isolated pure culture upto genus level.

Marks- (75+25) =100

3SM3 Molecular Virology Unit I

– 3 Credits

Structure of animal viruses and plant viruses; Classification of animal and plant viruses; Satellite viruses; Viroids; Virusoids, Prions etc.; Transmission of Viruses; Vectors for Virus transmission, Cell to cell and systemic movement of viruses. Impact of Viruses on Health and Economy: (Diseases causes by animal viruses and plant viruses; Economic loss due to important viruses); Bacterial Viruses: Lysogenic and Lytic Phages, Bacteriophage Typing.

Unit II

General Genomic organization of animal viruses; Replication and Life cycle of: Poliovirus, Human Immunodeficiency virus (HIV), Influenza Virus, Rabies Virus, Poxvirus, Herpesvirus and Hepatitis viruses; Introduction to Cancer causing viruses and their mechanism of host-cell transformation.

Unit III

General Genomic organization of plant viruses; Replication and Life cycle of plant viruses: Cauliflower Mosaic Virus (CMV), Tobacco Mosaic Virus (TMV), Rice Dwarf Virus, Citrus triesteza Virus.

Unit IV

Methods to diagnose animal virus infections: Electron microscopy, Tissue culture growth of viruses and Cytopathic effects, Virus quantitation assays, Viral serology: ELISA, neutralization assays; Molecular methods: hybridization, Real-time PCR, gene silencing and antiviral assays.

Unit V

Methods to study plant viruses; Infectivity assays – Sap transmission, insect vector transmission, agroinfection (using Agrobacterium); serological methods, immunelectrophoresis in gels, direct double-antibody sandwich method, Dot ELISA, Immunosorbent electron microscopy (ISEM), Polymerase chain reaction; Gene silencing, and viral suppressors of gene silencing.

Lab on Microbiological Techniques

- 4 Credits

- 1. Preparation of culture media-nutritional needs of microbes-dehydrated-selectivedifferential-autotrophic-heterotrophic, adjustment of pH, buffers, pure culture techniques, preparation of slants, sub-culturing.
- 2. Isolation of pure microbial flora from natural and extreme environments, serial dilution, Microbial growth measurement, standard plate count, haemocytometry.
- 3. Staining: Dye preparation, staining techniques, their applications, Motility (Hanging drop method). Microscopy: Microscope and its operations, components, type. Preservation of microorganisms.
- 4. Biochemical characterization of bacteria-BIOLOG plate method, carbohydrate fermentation, catalase, peroxidase, indole, methyl red, vogus-prausker, citrate utilization test (IMViC), Nitrate Reduction Test etc.