# 2015-2016

# B.Sc., I year –Semester-I MICROBIOLOGY AND CELL BIOLOGY (w.e.f 2015-2016)

#### UNIT I

#### HISTORY, DEVELOPMENT AND MICROSCOPY

History and development of microbiology: contributions of Louis Pasteur, Robert Koch and Edward Jenner. Microscopy: Compound microscopy: Numerical aperture and its importance, resolving power, oil immersion objectives and their significance, principles and applications of dark field, phase contrast, fluorescent microscopy. Electron microscopy: Principle, ray diagram and applications, TEM and SEM, comparison between optical and electron microscopy.

Stains and staining procedures: Acidic, basic and neutral stains, Gram staining, Acid fast staining, Flagella staining, Endospore staining.

#### UNIT-II BACTERIA & VIRUSES A.BACTERIA

Bacterial morphology and subcellular structures, general morphology of bacteria, shapes and sizes, generalized diagram of typical bacterial cell.Slime layer and capsule, difference between the structure, function and the position of the two structures. Cell wall of gram +ve and Gram -ve cells, Prokrayotic classification.

General account of flagella and fimbriae.Chromatin material, plasmids; definition and kind of plasmids (conjugative and non-conjugative) F, R, and Col plasmids.

A brief idea Bergey's manual. Morphology of archaea, archaeal cell membrane (differences between bacterial and archaeal cell membrane), other cell structures, concept of the three distinct archaea groups.

#### B.VIRUSES:

General Characters of viruses, difference between virus and typical microbial cell structure ,diferent shapes and symmetries with one example of each type, classification of viruses on the basis of nucleic acids, phage and animal cell viruses, example of each and their importance. Brief idea of lytic cycle and lysogeny.

#### UNIT III

#### **MICROBIAL NUTRITION:**

Types of Microbial nutrition.

Basic nutritional requirements: Basic idea of such nutrients as water, carbon, nitrogen, sulfur and vitamins etc., natural and synthetic media, nutritional classification of bacteria. Selective and Differential media, Enriched media, Enrichment media.

#### UNIT IV: MICROBIAL GROWTH AND CONTROL:

#### **GROWTH:**

Growth rate and generation time, details of growth curve and its various phases. Concepts of synchronous cultures, continuous and batch cultures (chemostat and turbidostat). **Measurement of growth.** 

Physical conditions required for growth : Temperature (Classification of microorganisms on the basis of temperature requirements), pH etc. Pure cultures and cultural characteristics.

Maintenance of pure culture.

**Microbial Control:** Terminologies - Sterilization, disinfection, antiseptic, sanitization, germicide, microbistasis, preservative and antimicrobial agents.

Mechanism of cell injury: Damage to cell wall, cell membrane, denaturation of proteins, inhibition of protein synthesis, transcription, replication, other metabolic reactions and change in supercoiling of DNA.

**Physical control:** Temperature (moist heat, autoclave, dry heat, hot air oven and incinerators), dessication, surface tension, osmotic pressure, radiation, UV light, electricity, ultrasonic sound waves, filtration.Chemical control: Antiseptics and disinfectants (halogens, alcohol, gaseous sterilization. Concept of biological control. Air filtration.

# UNIT V

# **CELL BIOLOGY**

**Eukaryotic Cell** – Structure and function of the following nucleus , nuclear membrane , nucleoplasm, nucleolus, golgi complex, Mitochandria, Chloroplast, endoplasmic reticulum, lysosomes, peroxisomes, glyoxisomes and vacuoles.

# Plant Cell wall :

Cytoskeleton (Micro and Macro filaments, microtubules) and cell locomotion. Mitosis and meiosis. Brief idea of cell cycle.Muscle and nerve cell structure, synaptic transmission and neuromuscular junctions.

# B.Sc., - I year SEMESTER-I Practicals BIOTECHNOLOGY MICROBIOLOGY & CELL BIOLOGY

- 1. Demonstration, Use and care of microbiological equipments
- 2. Preparation of media, sterilization and isolation of bacteria
- 3. Isolation of bacteria from water, soil and vegetables
- 4. Demonstration of motility of bacteria
- 5. Simple staining of Bacteria
- 6. Gram's Staining of bacteria
- 7. Acid fast Staining of bacteria- Demonstration
- 8. Endospore staining
- 9. Demonstration of Starch hydrolysis by bacterial cultures
- 10. Growth of fecal coliforms on selective media
- 11. Isolation of pure culture by Pour plate method
- 12. Isolation of pure culture by Streak plate method
- 13. Anaerobic cultivation of microorganisms
- 14. Cultivation of yeast/Mucor/Rhizopus
- 15. Antibiotic Sensitivity test

- 16. Oligodynamic action of Metals-Demonstration
- 17. To study Germicidal effect of UV-Light on bacterial Growth
- 18. Stages of Mitosis-Demonstration
- 19. Stages of Meiosis (Permanent slides)

Note: - Mandatory to perform at least ten practicals.

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### B. Sc., I – Semester II

## MACROMOLEULES, ENZYMOLOGY AND BIOENERGETICS (w.e.f 2015-16)

# UNIT I

#### NUCLEIC ACIDS AND CHROMOSOMES

Chemical structure and base composition of nucleic acids, Chargaff's rules, Watson Crick Model (B-DNA), deviations from Watson-Crick model, other forms of DNA (Aand Z-DNA), forces stabilizing nucleic acid structures, (hydrogen bonds and hydrophobic associations, base stacking). Maxam and Gilbert DNA sequencing and Sanger's method.

Concept of prokaryotic genes and eukaryotic genes: Definition of a gene, concept of split genes, introns, exons, spacers, C-value and C-value paradox, basic idea of Cot curves.

Chromatin structure: Nucleosome structure (10 nm fibre, experiments leading to discovery of nucleosomal structure, types of histones, arrangement of histones in the octamer, H1 histone and its role, role and length of linker DNA), 300 nm fibers (arrangement of nucleosome in a helical structure), domain and loop structure (further compacting of 300 nm fibre, role of scaffolding proteins). Role of telomere and centromere, telomeric and centromeric repeat sequences.

#### UNIT II

#### **Amino acids and Proteins**

Amino acids: Structure of amino acids occurring in proteins, classification of amino acids (pH based, polarity based and nutrition based), Physico-chemical properties of amino acids (solubility, boiling and melting points, reactions like Edman's, Sanger's, Dansyl chloride, ninhydrin). Titration curves of neutral, basic and acidic amino acids.

Primary structure of proteins: Determination of primary structure (end group analysis, cleavage of disulfide bonds, amino acid composition, use of endopepticlase specificity, sequence determination, assignment of disulfide position).

Secondary structure of proteins: The -helix, -structures (parallel, antiparallel, mixed, -turn).

Tertiary structure of proteins: Forces that stabilize the structure (electrostatic forces, hydrogen and disulfide bonds, hydrophobic associations), myoglobin as an example of tertiary structure, concept of domains, protein denaturation.

Quaternary structure of proteins: Forces stabilizing quaternary structure, advantages of oligomeric proteins.

#### UNIT-III

# CARBOHYDRATES

Definition, classification, nomenclature of carbohydrates, structures of monosaccharides, disaccharides and polysaccharides (structures of starch and glycogen as examples of homopolysaccharides). Concept and examples of heteropolysaccharides.

**Lipids** - Types of lipids, structures of saturated and unsaturated fatty acids, triglycerides, phospholipids, plasmalogens, gangliosides and sphingolipids. Terpenoids and isoprenoids - definition and representative structures, steroids. Concept of acid value, saponification value and iodine value. Chemistry of Porphyrines, Heme, Cytochromes, and Chlorophylls

# UNIT -V ENZYMES :

Terminology: Active site , allosteric site, Holo enzyme, apoenzyme, co-enzyme

substrate, inhibitor, activator, modulator etc.Classification and nomenclature. Concept of isoenzymes (example Lactate Dehydrogenase) and multienzymes (example pyruvate dehydrogenase) Substrate Specificity (bond specificity, group specificity, absolute specificity, stereo-specificity, proof-reading mechanism), lock and key and induced fit models.

Concept of Allosteric enzymes( Brief idea of ATPase as an example, mechanisms of calalysis

: Acid base, covalent and metal ion catalysis.

Assay of Enzymes: Concept of activity, specific activity, turnover number, units of enzyme activity (katal, international unit), spectrophotometric methods of assay of enzymes (simple and coupled assay), very brief idea of other methods.

Enzyme kinetics: Michaelis-Menten equation, effect of substrate concentration, effect of enzyme concentration, effect of p H and temperature, temperature quotient, single reciprocal( Eadie-Hoffstee equation) and double reciprocal plots( Lineweaver-Burke plots), enzyme inhibition kinetics (reversible inhibition types – competitive, uncompetitive and non-competitive), brief idea of irreversible inhibition.

### UNIT V

**Bioenergetics**: Concept of free energy, Entropy, Enthalpy & Redox Potential. Concept of high energy bonds as related to the structure of ATP, Phosphoenolpyruvate, Creatine phosphate etc.

Glycolosis ( Pathway , entry of other monosaccharides and di saccharides , regulations,

inhibitors) Gluconeogenesis : Bypass reactions.

## B.Sc., - I year SEMESTER-II Practicals BIOTECHNOLOGY MACRO MOLECULES & ENZYMOLOGY

- 1. Formal titration of Glycine- Demonstration
- 2. Quantitative estimation of proteins by Biuret method
- 3. Demonstration of Albumin and Ag ratio in Serum
- 4. Estimation of DNA by DPA method
- 5. Estimation of RNA by Orcinol method
- 6. Quantitative estimation of Amino acids using Ninhydrin reaction
- 7. Quantitative analysis of Sugars and proteins
- 8. Quantitative estimation of Sugars ( Dinitro salisilic acid method ) Demonstration
- 9. Estimation of Glucose by Benedicts Quantitative method
- 10. Quantitative estimation of proteins by Lowry's method
- 11. Extraction and quantification of total Lipids
- 12. Determination of Saponification value of fats
- 13. Determination of Acid value of fats
- 14. Isolation of Urease and demonstration of its activity
- 15. Assay of protease activity
- 16. Preparation of starch from potato and its hydrolysis by Salivary amylase
- 17. Assay of Alkaline phosphatase
- 18. Immobilization of enzymes / cells by Entrapment in Alginate gel
- 19. Effect of temperature / pH on enzyme activity

Note: - Mandatory to perform at least ten practicals.

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### II YEAR SEMESTER –III PAPER III: BIO MOLECULES AND CHEMISTRY (w.e.f 2013-14)

# UNIT I

**Carbohydrates**: Classification, structure and properties of monosaccharides (glucose, fructose, mannose) and disaccharides (maltose, lactose, sucrose) Polysaccharides (starch, glycogen. cellulose).

# UNIT II

Amino acids: Classification and properties of amino acids.

**Proteins**: Classification of proteins based on structure and functions, structural organization of proteins (primary, secondary, tertiary and quaternary structures) e.g. Keratin and Hemoglobin.

# UNIT III

**Lipids**: Classification, structures, properties and function of Cholesterol, Phospholipids and Triglyceride.

# UNIT IV

**Metabolisms**: Glycolysis, TCA cycle, energy yield, HMP pathway, Electron transport and oxidative phosphorylation.

# PRACTICAL BIOCHEMISTRY

- 1. Estimation of reducing sugars- Glucose, Maltose and lactose.
- 2. Estimation of protein by Biuret method and Lowry's method.
- 3. Separation of Sugars by TLC
- 4. Estimation of Amino acids.
- 5. Estimation of inorganic phosphate
- 6. Estimation of lipids by soxhlet method

# **Reference:**

1. Principles of Biochemistry- AlbertL. Lehninger CBS Publishers & Distributors

- 2. Biochemistry Lubert stryer Freeman International Edition.
- 3. Biochemistry Keshav Trehan Wiley Eastern Publications
- 4. Fundamentals of Bochemistry-J.L.Jain S.Chand and Company
- 5. Biochemistry- Prasaranga, Bangalore University
- 6. Fundamental of Biochemistry Dr.A.C.Deb
- 7. Textbook of Organic Chemistry (A Modern Approach)

8. The Biochemistry of Nucleic acid – Tenth Edition-Roger L.P.Adams, John T. Knowler and David

P.Leader, Chapman and Hall Publications

#### II YEAR SEMESTER –IV <u>PAPER IV: MICROBIOLOGY</u> (w.e.f 2013-14)

# UNIT I

#### Introduction and Scope of Microbiology

Definition and history of microbiology, contributions of Antony van Leeuwenhoek, Louis Pasteur, Robert Importance and scope of Microbiology as a modern Science **Microbial Techniques** 

Sterilization: Principles and Applications of

- a. Physical Methods. Autoclave, Hotairoven, Laminar airflow, Seitz filter, Sintered glass filter, and membrane filter.
- b. Chemical Methods: Alcohol, Aldehydes, Phenols, Halogens and Gaseous agents.
- c. Radiation Methods: UV rays and Gamma stains.

**STAINS AND STAINING TECHNIQUES**: Principles of staining, Types of stains – simple stains, structural stains and Differential stains.

### UNIT II

#### **Microbial Taxonomy**

Classification of bacteria based on – morphology (shape and flagella), staining reaction, nutrition and extreme environment.

**General Account of Viruses and Bacteria** 

A. Structure and classification of Plant Viruses – CaMV Animal viruses – Hepatitis B Bacterial Virus – Lamba Phage **UNIT III** 

Beneficial Microorganisms: General characteristics of Penicillium, Rhizobus and Mucor.

#### **Pathogenic Microorganisms**

A. Bacterial diseases of man – Tuberculosis, Pneumonia and Cholera.B. Viral diseases: AIDS (HIV)Plant Viral diseases: TMV.

#### UNIT IV

Growth and growth measurement: Definition of growth. Growth curve.

**Factors affecting growth**: nutrients, temperature, oxygen, pH, osmotic pressure. Measurement of growth by measuring cell number, cell mass and cell activity Cell count, direct and indirect method, turbidometric method. Plate count method, membrane filter count method, dry weight and wet weight method by measurement of cellular activity.Synchronous culture, continuous culture and batch culture.

# PRACTICAL - MICROBIOLOGY

- 1. Safety measures in microbiology laboratory
- 2. Cleaning and sterilization of glass ware
- 3. Study of instruments: Compound microscope, Autoclave, Hot air oven, pH meter, Laminar
- 4. airflow and centrifuge
- 5. Staining Techniques: Simple, Negative staining, Gram staining, Endospore staining and fungal staining.
- 6. Media preparation: Nutrients agar, MRBA and Nutrient broth.
- 7. Isolation of bacteria and fungi from soil, air, and water dilution and pourplate methods
- 8. Estimation of microorganisms Total Count (Haemocytometer)
- 9. Antibiotic sensitivity test starch hydrolysis, catalase &
- 10. Biochemical tests starch hydrolysis, catalase & gelatin liquefaction.
- 11. Study of Rhizobium from root nodules of legumes.
- 12. Determination of growth curve and generation time of E. coli

# **REFERENCE:**

- 1. Microbiology pelezar, chan, krieg Tata McGraw Hill Publications.
- 2. Microbiology concepts and application by Paul A.Ketchum, Wiley Publications
- 3. Fundaments of Microbiology- Frobisher, Sauders & toppan publications.
- 4. Microbiology Ronald M.Atlas
- 5. Introductory Biotechnology R.B. Singh C.B.D. India (1990)
- 6. Industrial Microbiology casidal.E.Wiley Eastern Ltd.
- 7. Fundamentals of Bacteriology Salley
- 8. Fontiers in Microbial technology P.S.Bisen, CBS Publishers
- Biotechnology: International Trends of perspectives A.T.Bull, G.Holl M.D.Lilly Oxford &
- 10. TBH publishers.
- 11. General Microbiology-C.B.Powar, H.F. Daginawala, Himalayan Publishing House

# <u>3rd YEAR SYLLABUS</u> <u>SEMESTER - V</u> <u>PAPERL - V: IMMUNOLOGY</u> (w.e.f 2014-15)

# UNIT-I

Historical perspectives - overview of immune system, innate and acquired immunity, primary and secondary lymphoid organs, cell involved in immune system.

# UNIT-II

Antigen and antigenicity, Immunoglobulins – structure and types complement system, antigen - antibody interaction, precipitation, agglutination, RIA, ELISA and western blotting – polyclonal and monoclonal antibodies production and applications.

Organisation and expression of immunoglobulin genes.

# UNIT-III

Cytokines: Structure and function-cytokine receptors-biological functions of cytokines, histocompatability complex - cell mediated immunity; T cell activation - Humoral response; B cell activation and proliferation - hypersensitive reactions.

# UNIT-IV

Immunodeficiency diseases (AIDS) - immune suppression & transplantation. Vaccines – Conventional and modern approaches.

# Practicals

- 1. Identification of Immune organs in Rat.
- 2. Antigen antibody reaction and blood grouping.
- 3. Aseptic technique, sterilization, cleaning of glassware.
- 4. Preparation of media, (microbial & tissue culture)
- 5. ELISA

#### Reference

- 1. Immunology (V Edition),- Richard A.Goldsby, Thomas. J. Kindt, A. Osborne, Janis Kuby, 2003. W.H. Freeman and company
- 2. Immunology, Ivan Roitt, 2001. Harcourt publishers, ltd.
- 3. Essential immunology, Ivan Riott, 2000. Blackwell Science, 9th Edition.
- 4. Immunology An Introduction, Tizard.

# $\underline{SEMESTER - V}$

# <u>PAPER – VI: MOLECULAR BIOLOGY & GENETIC ENGINEERING</u> (w.e.f 2014-15)

### **Molecular Biology**

### **UNIT – 1**

An introduction. Experimental Proof of DNA and RNA as genetic material. Structure and functions of DNA and RNA. Transformation, conjugation and transduction. DNA Replication, types – rolling circle,  $\sigma$ , single strand DNA. Prokaryotic and Eukaryotic – Enzymes and proteins involved in replication of prokaryotic and eukaryotic DNA. DNA damage and repair.

### UNIT - II

**Transcription:** in prokaryotes and Eukaryotes, Promoters and RNA polymerase, transcription factors.

**Translation**: Mechanism of translation in Prokaryotes and Eukaryotes, Post translational modifications of proteins.

**Regulation of Gene expression:** Regulation of Gene expression in Prokaryotes – Operon concept (Lac and Trp), Regulation of Gene expression in Eukaryotes - transcriptional ctivation, galactose metabolism in yeast.

# **Genetic Engineering**

#### UNIT-III

**Vectors:** Plasmids – types and salient features (pBR322, pUC19), Expression vectors, shuttle vectors.

Phages: Insertional and replacement vectors

Cosmids: YAC and BAC, Ti plasmids

**Enzymes:** Restriction enzyme, types, nomenclature and applications; other nucleases, ligase, kimase, methylase, gyrase, phosphatase. DNA & RNA polymerase – T4, T7, SP6, Polymerase, Tag polymerase, Reverse transcriptase.

#### UNIT- IV

rDNA Technology – generation of rDNA molecules, introduction of rDNA into cells – microinjection, transformation, electroporation.

Screening: DNA and RNA probes, Hybridization. Transgenic plants (Bt cotton) and transgenic animals (fish)

# Practicals

# **Molecular Biology**

- 1. Estimation of DNA by DPA/ RNA by Orcinal method
- 2. Isolation of Plasmid by alkaline lysis method
- 3. Transformation of pB322 into *E. coli*.
- 4. DNA ligase
- 5. Restriction digestion

# **Reference:**

# MOLECULAR BIOLOGY

- 1. Glick, B.T and Pastermak J.J (1998) Moleclar biotechnology, Principles and application of recombinant DNA, Washington D.C. ASM press.
- 2. Howe.C. (1995) Gene Cloning and manioulation, Cambridge University Press, USA
- 3. Lewin, B., Gene VI New York, Oxford University Press.
- 4. Rigby, P.W.J. (1987) Genetic Engineering Academic Press Inc. Florida, USA.
- 5. Sambrook et al (2000) Molecular cloning Volumes I,II, & III Cold spring Harbor Laboratory
- 6. Press, New York, USA
- 7. Walker J.M. and Gingold, E.B. (1983)Molecular Biology & Biotechnogy (Indian Edition)
- 8. Royal Society of Cemistry U.K
- 9. Karp.G (2002) Cell & Molecular Biology, 3rd Edition, John Wiley & Sons; INC

# GENETIC ENGINEERING

- 1. Gene Cloning An introduction, T.A. Brown. Van Reinhold, 1988.
- 2. Recombinant DNA Watson JD, Gilman M, Witkowski J and Zoller M, 1992. Second Ed. Scientific American Books.
- 3. DNA Cloning I and II, D.M. Glover and B.D. Hames, 1995. IRL press.
- 4. Genetic Engineering An introduction, D.S.T. Nicholl.
- 5. Principles of Gene Manipulation, R.N.Old and S.B. Primrose, 1994. Blackwell Publishers, New York.

# <u>SEMESTER – VI</u>

# <u>PAPERL – VII: PLANT BIOTECHNOLOGY AND ANIMAL BIOTECHNOLOGY</u> (w.e.f 2014-15)

#### **Plant Biotechnology**

## UNIT – I

In-vitro Methods in plant tissue culture, Aseptic Techniques, Nutrient media, and use of growth regulators (Auxins, Cytokininis and Gibberellins). Clonal Propogation of elite species (Micropropagation).

**Organ Culture** – Anther, Embryo and Endosperm culture and their applications. Organogenesis and Somatic Embryogenesis – Techniques and applications.

**Protoplast Culture** –methods of protoplast fusion –chemical and electro fusion. Somaclonal Variation and their significance. Hybrids and cybrids.

# UNIT – II

*In-Vitro* production of secondary metabolites – Techniques and significance. Role of tissue culture in agriculture, horticulture and forestry.

**Transgenic plants:** Technique of transformation – Agrobacterium mediated and physical methods (Microprojectile and electroporation) Applications of transgenic plants. Edible Vaccines from plants – Banana, Watermelon.

#### **Animal Biotechnology**

# UNIT – III

Culture Media: Composition and importance of Serum.

a) Primary Culture – disaggregation of tissue, isolation of tissue, enzyme disaggregation, and mechanical disaggregation.

b) Secondary Culture – transformed animal cells and continious cell lines.

# UNIT – IV

Artificial insemination (AI) and separation of semen by flow cytometry.

*In vitro* fertilization (IVF), embryo transfer, super ovulation, synchronization of Estrus, collection and transfer of embryos, cryopreservation.

**Transgenic Animals:** Techniques and Applications and Transgenic mice and sheep. Intellectual property rights (IPR), Patents, trade secrests, copyright and trademark.

#### Practicals

- 1. Preparation of media, and initiation of callus from any one selected plant species
- 2. Micropropogation of plants (any one)
- 3. Anther culture for production of haploid plants
- 4. Preparation of synthetic seeds
- 5. Isolation and fusion of protoplast
- 6. Preparation of media and culture of animal cells/tissues
- 7. Cell disaggregation and cell counting
- 8. Cytotoxicity of the cells using the dye MTT method
- 9. Sperm analysis sperm count, mobility and viability.

### **Reference:**

#### **Plant Biotechnology**

- 1. Ravishankar G.A. and Venkataraman L.V. (197) Biotechnolgoy Applications of plant Tissue & culture. Oxford & IBH Publishing Co, Pvt. Ltd.
- 2. Bhan (1998) Tissue Culture, Mittal Publications, New Delhi.
- 3. Islan A.C (1996) Plant Tissue Culture, Oxford & IBH Publishing Co. Pvt. Ltd.
- 4. Lydiane Kyte & John Kelyn (1996) Plants from test tubes. An introduction to Micropropogation Edition) Timber Press, Partland.
- 5. Kumar H.D. (1991) A text book on Biotechnology (2nd Edition) Affiliated East West Press Priva Ltd. New Delhi.
- 6. Chrispeel M.J. and Sdava D.E. (1994) Plants, Genes and Agriculture. Jones and Barlett Publisher Boton.
- 7. Reinert J. and Bajaj Y.P.S. (197) Applied and Fundamental Aspects of Plant Cell, Tissue, and Orga Culture, Narosa Publishing House.

# **Animal Biotechnology**

- 1. Ian Freshney (4th Edition)
- 2. Buttler.
- 3. Elements of Biotechnology P.k. Gupta (1st Edition -2000) Rastogi Publications.

# <u>SEMESTER – VI</u>

#### <u>PAPERL – VIII: MICROBIAL BIOTECHNOLOGY AND ENVIRONMENTAL</u> <u>BIOTECHNOLOGY</u> (w.e.f 2014-15)

### UNIT I

Microbial Biotechnology: Scope, Techniques, Chronological Development, Microbial diversity and its use.

Industrial Production: amylase, protease, xanthan, ethanol, citric acid and HBsAg by microbes.

### UNIT II

Production of microbial metabolites: Amino acids (glutamine), Antibiotics (Ampicillin) and other secondary metabolites (Streptomycin, Penicillin).

### UNIT – III

Environmental components, Environmental pollution and its types, Non-renewable and renewable energy resources.

Conventional fuels and their major impacts: Global warming and greenhouse effect, Global Ozone Problem, Acid rain, Eutrophication, Biomagnification, Concept of clean fuel technology: Biomass energy and biofuels

#### UNIT – IV

Biodegradation and bioremediation of major pollutants, Superbug biomineralisation: Use of microbial technology for mining.

Treatment of municipal solid and liquid wastes, Environmental impact assessment and Environmental audit.

Bioassessment of Environmental Quality, Biofertilizers and Biopesticides.

#### Practicals

- 1. Estimation of citric acid from Aspergillums Culture.
- 2. Estimation of lactic acid and lactose.
- 3. Immobilization of Yeast cells.
- 4. Preparation of wine.
- 5. Estimation of Alcohol by Specific gravity method.
- 6. Estimation of BOD (2 Samples)
- 7. VAM staining

# References

- 1. Microbial Biotechnology (Fundamental of applied Microbiology) Alexandar N. Glazer & Hiroshi Nikaido
- 2. Fermentation Microbiology and Biotechnology. El mans, E.M.T., and Bryce, C.F.A. 2002. Taylor & Francis group.
- 3. Fundamentals of Biotechnology, Prave, P. Faust, V. Sitting, W. and Sukatseh, D.A. (eds). 1987. WCH Weinhein.
- 4. Principles of Fermentation Technology. Stanbury P.F. and Whitaker, A. 1984. Pergamon Press.
- 5. Waste water engineering treatment, disposal and reuse, Metcalf and Eddy Inc., Tata McGraw Hill, New Delhi.
- 6. Environmental Chemistry, AK. De, Wiley Eastern Ltd, New Delhi.
- 7. Introduction to Biodeterioration, D.Allsopp and K.J. Seal, ELBS / Edward Arnold.
- 8. Bioremidation, Baaker, KH and Herson D.S., 1994. Mc.GrawHill Inc, NewYork.
- 9. Industrial and Environmental Biotechnology Nuzhat Ahmed, Fouad M. Qureshi and Obaid Y. Khan, 2006. Horizon Press.
- 10. Environmental Molecular Biology, Paul. A, Rochelle, 2001. Horizon Press.

# **MODEL QUESTION PAPER**

#### **B.Sc.**, Three year Degree Examinations, November December

Model Question Paper I Semester Biotechnology 1119-Paper –I Microbiology and Cell Biology (W.e.f. 2015-16)

#### **Time -3hrs**

,

Max Marks: 70

Section – A Answer any FIVE Questions (Each question carries 4 Marks )

5x4=20M

- 1. Endospore staining
- 2. Dark field Microscope
- 3. Flagella
- 4. Autotrophs
- 5. Artificial media
- 6. Conditions required for growth
- 7. Disinfection
- 8. Turbidostat
- 9. Neuromuscular junction
- **10.** Ribosomes

#### Section – B

#### Answer ALL of the following questions

5x10=50M

#### (Each question carries 8 Marks)

11.(A) Describe the contributions of Louis Pasteur and Edward Jenner to microbiology

(Or)

- (B) Explain about Phase-Contrast Microscope with neat labeled Diagram
- 12.(A)Give an account on Ultra structure of Bacterial cell wall with a neat labeled diagram

(Or)

(B)Describe the structure and lifecycle of Bacteriophase- phase

13. (A) Write about nutritional groups of Bacteria

#### (Or)

- (B) Write a note on nutritional requirements of bacteria
- 14. (A).Describe different phases of Growth Curve (Or)
- (B) Explain the different methods of physical sterilization
- 15. (A) Describe in detail Mitosis

(Or)

(B)Explain about structure and functions of Golgi complex

# B.Sc., I year –Semester-I MICROBIOLOGY AND CELL BIOLOGY (w.e.f 2015-2016)

#### UNIT I

# HISTORY, DEVELOPMENT AND MICROSCOPY

History and development of microbiology: contributions of Louis Pasteur, Robert Koch and Edward Jenner. Microscopy: Compound microscopy: Numerical aperture and its importance, resolving power, oil immersion objectives and their significance, principles and applications of dark field, phase contrast, fluorescent microscopy. Electron microscopy: Principle, ray diagram and applications, TEM and SEM, comparison between optical and electron microscope, limitations of electron microscopy.

Stains and staining procedures: Acidic, basic and neutral stains, Gram staining, Acid fast staining, Flagella staining, Endospore staining.

## UNIT-II BACTERIA & VIRUSES A.BACTERIA

Bacterial morphology and subcellular structures, general morphology of bacteria, shapes and sizes, generalized diagram of typical bacterial cell.Slime layer and capsule, difference between the structure, function and the position of the two structures. Cell wall of gram +ve and Gram -ve cells, Prokrayotic classification.

General account of flagella and fimbriae.Chromatin material, plasmids; definition and kind of plasmids (conjugative and non-conjugative) F, R, and Col plasmids.

A brief idea Bergey's manual. Morphology of archaea, archaeal cell membrane (differences between bacterial and archaeal cell membrane), other cell structures, concept of the three distinct archaea groups.

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General Characters of viruses, difference between virus and typical microbial cell structure ,diferent shapes and symmetries with one example of each type, classification of viruses on the basis of nucleic acids, phage and animal cell viruses, example of each and their importance. Brief idea of lytic cycle and lysogeny.

# UNIT III MICROBIAL NUTRITION:

Types of Microbial nutrition.

Basic nutritional requirements: Basic idea of such nutrients as water, carbon, nitrogen, sulfur and vitamins etc., natural and synthetic media, nutritional classification of bacteria. Selective and Differential media, Enriched media, Enrichment media.

# UNIT IV: MICROBIAL GROWTH AND CONTROL:

# **GROWTH:**

Growth rate and generation time, details of growth curve and its various phases. Concepts of

synchronous cultures, continuous and batch cultures (chemostat and turbidostat). **Measurement of growth.** 

Physical conditions required for growth : Temperature (Classification of microorganisms on the basis of temperature requirements), pH etc. Pure cultures and cultural characteristics. Maintenance of pure culture.

**Microbial Control:** Terminologies - Sterilization, disinfection, antiseptic, sanitization, germicide, microbistasis, preservative and antimicrobial agents.

Mechanism of cell injury: Damage to cell wall, cell membrane, denaturation of proteins, inhibition of protein synthesis, transcription, replication, other metabolic reactions and change in supercoiling of DNA.

**Physical control:** Temperature (moist heat, autoclave, dry heat, hot air oven and incinerators), dessication, surface tension, osmotic pressure, radiation, UV light, electricity, ultrasonic sound waves, filtration.Chemical control: Antiseptics and disinfectants (halogens, alcohol, gaseous sterilization. Concept of biological control. Air filtration.

# UNIT V CELL BIOLOGY

**Eukaryotic Cell** – Structure and function of the following nucleus , nuclear membrane , nucleoplasm, nucleolus, golgi complex, Mitochandria, Chloroplast, endoplasmic reticulum, lysosomes, peroxisomes, glyoxisomes and vacuoles.

## Plant Cell wall :

Cytoskeleton (Micro and Macro filaments, microtubules) and cell locomotion. Mitosis and meiosis. Brief idea of cell cycle.Muscle and nerve cell structure, synaptic transmission and neuromuscular junctions.

# B.Sc., - I year SEMESTER-I Practicals BIOTECHNOLOGY MICROBIOLOGY & CELL BIOLOGY

- 20. Demonstration, Use and care of microbiological equipments
- 21. Preparation of media, sterilization and isolation of bacteria
- 22. Isolation of bacteria from water, soil and vegetables
- 23. Demonstration of motility of bacteria
- 24. Simple staining of Bacteria
- 25. Gram's Staining of bacteria
- 26. Acid fast Staining of bacteria- Demonstration

- 27. Endospore staining
- 28. Demonstration of Starch hydrolysis by bacterial cultures
- 29. Growth of fecal coliforms on selective media
- 30. Isolation of pure culture by Pour plate method
- 31. Isolation of pure culture by Streak plate method
- 32. Anaerobic cultivation of microorganisms
- 33. Cultivation of yeast/Mucor/Rhizopus
- 34. Antibiotic Sensitivity test
- 35. Oligodynamic action of Metals-Demonstration
- 36. To study Germicidal effect of UV-Light on bacterial Growth
- 37. Stages of Mitosis-Demonstration
- 38. Stages of Meiosis (Permanent slides)

#### Note: - Mandatory to perform at least ten practicals.

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# B. Sc., I – Semester II

### MACROMOLEULES, ENZYMOLOGY AND BIOENERGETICS (w.e.f 2015-16)

## UNIT I

#### NUCLEIC ACIDS AND CHROMOSOMES

Chemical structure and base composition of nucleic acids, Chargaff's rules, Watson Crick Model (B-DNA), deviations from Watson-Crick model, other forms of DNA (A- and Z-DNA), forces stabilizing nucleic acid structures, (hydrogen bonds and hydrophobic associations, base stacking). Maxam and Gilbert DNA sequencing and Sanger's method.

Concept of prokaryotic genes and eukaryotic genes: Definition of a gene, concept of split genes, introns, exons, spacers, C-value and C-value paradox, basic idea of Cot curves.

Chromatin structure: Nucleosome structure (10 nm fibre, experiments leading to discovery of nucleosomal structure, types of histones, arrangement of histones in the octamer, H1 histone and its role, role and length of linker DNA), 300 nm fibers (arrangement of nucleosome in a helical structure), domain and loop structure (further compacting of 300 nm fibre, role of scaffolding proteins). Role of telomere and centromere, telomeric and centromeric repeat sequences.

#### UNIT II

#### Amino acids and Proteins

Amino acids: Structure of amino acids occurring in proteins, classification of amino acids (pH based, polarity based and nutrition based), Physico-chemical properties of amino acids (solubility, boiling and melting points, reactions like Edman's, Sanger's, Dansyl chloride, ninhydrin). Titration curves of neutral, basic and acidic amino acids.

Primary structure of proteins: Determination of primary structure (end group analysis, cleavage of disulfide bonds, antino acid<sup>2</sup> composition, use of endopeptidase <sup>8</sup>pecificity, sequence determination, assignment of disulfide position).

Secondary structure of proteins: The -helix, -structures (parallel, antiparallel, mixed, -turn).

Tertiary structure of proteins: Forces that stabilize the structure (electrostatic forces, hydrogen and disulfide bonds, hydrophobic associations), myoglobin as an example of tertiary structure, concept of domains, protein denaturation.

Quaternary structure of proteins: Forces stabilizing quaternary structure, advantages of oligomeric proteins.

#### UNIT-III

# CARBOHYDRATES

Definition, classification, nomenclature of carbohydrates, structures of monosaccharides, disaccharides and polysaccharides (structures of starch and glycogen as examples of homopolysaccharides). Concept and examples of heteropolysaccharides.

Lipids - Types of lipids, structures of saturated and unsaturated fatty acids, triglycerides, phospholipids, plasmalogens, gangliosides and sphingolipids. Terpenoids and isoprenoids -

definition and representative structures, steroids. Concept of acid value, saponification value and iodine value. Chemistry of Porphyrines, Heme, Cytochromes, and Chlorophylls

# UNIT -V ENZYMES :

Terminology: Active site , allosteric site, Holo enzyme, apoenzyme, co-enzyme

substrate, inhibitor, activator, modulator etc.Classification and nomenclature. Concept of isoenzymes (example Lactate Dehydrogenase) and multienzymes (example pyruvate dehydrogenase) Substrate Specificity (bond specificity, group specificity, absolute specificity, stereo-specificity, proof-reading mechanism), lock and key and induced fit models.

Concept of Allosteric enzymes( Brief idea of ATPase as an example, mechanisms of calalysis :

Acid base, covalent and metal ion catalysis.

Assay of Enzymes: Concept of activity, specific activity, turnover number, units of enzyme activity (katal, international unit), spectrophotometric methods of assay of enzymes (simple and coupled assay), very brief idea of other methods.

Enzyme kinetics: Michaelis-Menten equation, effect of substrate concentration, effect of enzyme concentration, effect of p H and temperature, temperature quotient, single reciprocal( Eadie-Hoffstee equation) and double reciprocal plots( Lineweaver-Burke plots), enzyme inhibition kinetics (reversible inhibition types – competitive, uncompetitive and non-competitive), brief idea of irreversible inhibition.

#### UNIT V

**Bioenergetics**: Concept of free energy, Entropy, Enthalpy & Redox Potential. Concept of high energy bonds as related to the structure of ATP, Phosphoenolpyruvate, Creatine phosphate etc. Glycolosis (Pathway, entry of other monosaccharides and di saccharides, regulations,

inhibitors) Gluconeogenesis : Bypass reactions.

#### B.Sc., - I year SEMESTER-II Practicals BIOTECHNOLOGY MACRO MOLECULES & ENZYMOLOGY

- 20. Formal titration of Glycine- Demonstration
- 21. Quantitative estimation of proteins by Biuret method
- 22. Demonstration of Albumin and Ag ratio in Serum
- 23. Estimation of DNA by DPA method
- 24. Estimation of RNA by Orcinol method
- 25. Quantitative estimation of Amino acids using Ninhydrin reaction
- 26. Quantitative analysis of Sugars and proteins
- 27. Quantitative estimation of Sugars ( Dinitro salisilic acid method ) Demonstration
- 28. Estimation of Glucose by Benedicts Quantitative method
- 29. Quantitative estimation of proteins by Lowry's method
- 30. Extraction and quantification of total Lipids
- 31. Determination of Saponification value of fats
- 32. Determination of Acid value of fats
- 33. Isolation of Urease and demonstration of its activity
- 34. Assay of protease activity
- 35. Preparation of starch from potato and its hydrolysis by Salivary amylase
- 36. Assay of Alkaline phosphatase
- 37. Immobilization of enzymes / cells by Entrapment in Alginate gel
- 38. Effect of temperature / pH on enzyme activity

#### Note: - Mandatory to perform at least ten practicals.

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#### **MODIFIED SYLLABUS**

# B.Sc., SEMESTER III BIOPHYSICAL TECHNIQUES (w.e.f 2016-17)

#### UNIT -I:

**Spectrophotometry :** Spectrum of light, absorption of electromagnetic radiations, Beer's law – derivation and deviations, extinction coefficient. Instrumentation of UV and visible spectrophotometry, Double deam spectrometer ; dual – wavelength spectrometer, Applications of UV and visible spectrotometry. Spectrofluorometry : principle , instrumentation and application.

#### **UNIT II :**

**Chromatography:** Partition principle, coefficient, nature of partition forces, brief account of paper chromatography. Thin layer chromatography and column chromatography. Gel filtration: Concept of distribution coefficient, types of gels and glass beads, applications. Ion-exchange chromatography: Principle, types of resins, choice of buffers, applications including amino acid analyzer. Affinity chromatography: Principle, selection of ligand, brief idea of ligand attachment, specific and non-specific elution, applications. HPLC.

#### **UNIT III**

**Electrophoresis :** Migration of ions in electric fields, Factors affecting electrophoretic mobility. Paper electrophoresis, Gel electrophoresis :- Types of gels, Solubilizers, Procedure, Column & slad gels Detection, Recovery & Estimation of macromolecules. SDS – PAGE Electrophoresis and applications. Isoelectric focusing, Puised-field gel electrophoresis.

#### UNIT IV

**Isotopic tracer technique :** Radioactive & stable isotopes, rate of radioactive decay. Units of radioactivity. Measurement of radioactivity :- Ionization chambers, proportional counters, Geiger – Muller counter, Solid and liquid scintillation counters (basic principle, instrumentation and rechnique), Cerenkov radiation. Measurement of Stable isotopes : Falling drop method for deuterium measurement, Mass spectrometry. Principles of tracer technique, advantages and

limitations, applications of isotopes in biotechnology (distribution studies, metabolic studies, isotope dilution technique, metabolic studies, clinical applications, autoradiography).

# $\mathbf{UNIT} - \mathbf{V}$

**Centrifugation :** Basic principles, concept of RCF, types of centrifuges (clinical high speed and ultracentrifuges). Preparative centrifugation: Differential and density gradient centrifugation, applications (Isolation of cell components). Analytical centrifugation: Sedimentation coefficient, determination of molecular weight by sedimentation velocity and sedimentation equilibrium methods.

Biostatistics Basic concepts of mean, median, mode Standard deviation and Standard error. Introduction to ANOVA Applications of basic statistics

Bio Informatics: Introduction, Databases, Search tolls, Applications in Bo Technology.

# **PRACTICALS : BIOPHYSICAL TECHNIQUES**

- 1. Spectrophotometric analysis of DNA denaturation.
- 2. Protein estimation by Biuret/Lowry method
- 3. Paper chromatography of amino acids/sugars.
- 4. TLC of sugars/amino acids- A demonstration by using of Photograph / reference diagram
- 5. Cellular fractionation and separation of cell organelles using centrifuge
- 6. Isolation of mitochondria and assay of marker enzyme
- 7. Estimation of Urea by diacetyle monoxide method
- 8. Estimation of Sugars by Folin Phenol method.
- 9. Preparation of standard buffers and determination of pH of a solution
- 10. Titration of a mixture of strong & weak acid
- 11. Paper electrophoresis of proteins- A demonstration by using of reference diagram
- 12. Gel electrophoresis of protein
- 13. SDS-PAGE of an oligomeric protein.
- 14. Calculation of mean, median, and mode (manual / computer aided)
- 15. Calculation of standard deviation and standard error (manual/ computer aided).
- 16. Biostatistical problem based on standard deviation.

# Note : - Mandatory to perform at least 8 practical's

# **B.Sc., SEMESTER IV**

# **IMMUNOLOGY**

# UNIT I(w.e.f 2016-17)

Immune system : Organs and cells of immune system immunity, innate immune mechanism Acquired immune mechanism, Antigen,

Humoral immunity, main pathways of complement system.

UNIT II

Antibody and Antigen : Antibody structure and classes, Antibody diversity, types of Antigens Antigenecity (factors affecting antigenecity). Complement system.

UNIT III

**Immunity :** Cell mediated immunity : TC mediated immunity, NK cell mediated immunity, ADCC brief description of cytokines and MHC (MHC types and diversity) Autoimmunity.

UNIT IV

**Hypersensitivity and vaccination :** General features of hypersensitivity various types of hypersensitivity, Vaccination : Discovery, principles, significance, Types of Vaccines

UNIT V

**Immunological Techniques :** Antigen – antibody reactions : Precipition, agglutination, complement fixation, immunodiffusion, ELISA. Hybridoma technology : Monoclonal antibodies and their applications in immunodiagnosis.

# **PRACTICALS : IMMUNOLOGY**

- 1. Antigen antibody reaction
- 2. Determination of blood group
- 3. Widal test
- 4. Ouchterloney immunodiffusion-A demonstration
- 5. Radial immunodiffusion-A Demonstration
- 6. ELISA A demonstration
- 7. Coombs Test
- 8. Isolation of casein by isoelectric precipitation
- 9. Production of antibodies and their titration

# Note : - Mandatory to perform at least 5 Practical's

## <u>3rd YEAR SYLLABUS</u> <u>SEMESTER - V</u> <u>PAPER - V: IMMUNOLOGY</u> (w.e.f 2014-15)

# UNIT-I

Historical perspectives - overview of immune system, innate and acquired immunity, primary and secondary lymphoid organs, cell involved in immune system.

# UNIT-II

Antigen and antigenicity, Immunoglobulins – structure and types complement system, antigen - antibody interaction, precipitation, agglutination, RIA, ELISA and western blotting – polyclonal and monoclonal antibodies production and applications.

Organisation and expression of immunoglobulin genes.

# UNIT-III

Cytokines: Structure and function-cytokine receptors-biological functions of cytokines, histocompatability complex - cell mediated immunity; T cell activation - Humoral response; B cell activation and proliferation - hypersensitive reactions.

#### UNIT-IV

Immunodeficiency diseases (AIDS) - immune suppression & transplantation. Vaccines – Conventional and modern approaches.

#### Practicals

- 6. Identification of Immune organs in Rat.
- 7. Antigen antibody reaction and blood grouping.
- 8. Aseptic technique, sterilization, cleaning of glassware.
- 9. Preparation of media, (microbial & tissue culture)
- 10. ELISA

#### Reference

- 5. Immunology (V Edition),- Richard A.Goldsby, Thomas. J. Kindt, A. Osborne, Janis Kuby, 2003. W.H. Freeman and company
- 6. Immunology, Ivan Roitt, 2001. Harcourt publishers, ltd.
- 7. Essential immunology, Ivan Riott, 2000. Blackwell Science, 9th Edition.
- 8. Immunology An Introduction, Tizard.

# $\underline{SEMESTER - V}$

## PAPER – VI: MOLECULAR BIOLOGY & GENETIC ENGINEERING (w.e.f 2014-15)

#### **Molecular Biology**

## UNIT – 1

An introduction. Experimental Proof of DNA and RNA as genetic material. Structure and functions of DNA and RNA. Transformation, conjugation and transduction. DNA Replication, types – rolling circle,  $\sigma$ , single strand DNA. Prokaryotic and Eukaryotic – Enzymes and proteins involved in replication of prokaryotic and eukaryotic DNA. DNA damage and repair.

#### UNIT - II

**Transcription:** in prokaryotes and Eukaryotes, Promoters and RNA polymerase, transcription factors.

**Translation**: Mechanism of translation in Prokaryotes and Eukaryotes, Post translational modifications of proteins.

**Regulation of Gene expression:** Regulation of Gene expression in Prokaryotes – Operon concept (Lac and Trp), Regulation of Gene expression in Eukaryotes - transcriptional ctivation, galactose metabolism in yeast.

# Genetic Engineering

# UNIT-III

**Vectors:** Plasmids – types and salient features (pBR322, pUC19), Expression vectors, shuttle vectors.

**Phages:** Insertional and replacement vectors

**Cosmids:** YAC and BAC, Ti plasmids

**Enzymes:** Restriction enzyme, types, nomenclature and applications; other nucleases, ligase, kimase, methylase, gyrase, phosphatase. DNA & RNA polymerase – T4, T7, SP6, Polymerase, Tag polymerase, Reverse transcriptase.

#### UNIT-IV

rDNA Technology – generation of rDNA molecules, introduction of rDNA into cells – microinjection, transformation, electroporation.

Screening: DNA and RNA probes, Hybridization. Transgenic plants (Bt cotton) and transgenic animals (fish)

# Practicals

# **Molecular Biology**

- 6. Estimation of DNA by DPA/ RNA by Orcinal method
- 7. Isolation of Plasmid by alkaline lysis method
- 8. Transformation of pB322 into E. coli.
- 9. DNA ligase
- 10. Restriction digestion

# **Reference:**

# **MOLECULAR BIOLOGY**

- 10. Glick, B.T and Pastermak J.J (1998) Moleclar biotechnology, Principles and application of recombinant DNA, Washington D.C. ASM press.
- 11. Howe.C. (1995) Gene Cloning and manioulation, Cambridge University Press, USA
- 12. Lewin, B., Gene VI New York, Oxford University Press.
- 13. Rigby, P.W.J. (1987) Genetic Engineering Academic Press Inc. Florida, USA.
- 14. Sambrook et al (2000) Molecular cloning Volumes I,II, & III Cold spring Harbor Laboratory
- 15. Press, New York, USA
- 16. Walker J.M. and Gingold, E.B. (1983)Molecular Biology & Biotechnogy (Indian Edition)
- 17. Royal Society of Cemistry U.K
- 18. Karp.G (2002) Cell & Molecular Biology, 3rd Edition, John Wiley & Sons; INC

# **GENETIC ENGINEERING**

- 6. Gene Cloning An introduction, T.A. Brown. Van Reinhold, 1988.
- 7. Recombinant DNA Watson JD, Gilman M, Witkowski J and Zoller M, 1992. Second Ed. Scientific American Books.
- 8. DNA Cloning I and II, D.M. Glover and B.D. Hames, 1995. IRL press.
- 9. Genetic Engineering An introduction, D.S.T. Nicholl.
- 10. Principles of Gene Manipulation, R.N.Old and S.B. Primrose, 1994. Blackwell Publishers, New York.

## <u>SEMESTER – VI</u>

# <u>PAPERL – VII: PLANT BIOTECHNOLOGY AND ANIMAL BIOTECHNOLOGY</u> (w.e.f 2014-15)

#### **Plant Biotechnology**

UNIT – I

In-vitro Methods in plant tissue culture, Aseptic Techniques, Nutrient media, and use of growth regulators (Auxins, Cytokininis and Gibberellins). Clonal Propogation of elite species (Micropropagation).

**Organ Culture** – Anther, Embryo and Endosperm culture and their applications. Organogenesis and Somatic Embryogenesis – Techniques and applications.

**Protoplast Culture** –methods of protoplast fusion –chemical and electro fusion. Somaclonal Variation and their significance. Hybrids and cybrids.

#### UNIT – II

*In-Vitro* production of secondary metabolites – Techniques and significance. Role of tissue culture in agriculture, horticulture and forestry.

**Transgenic plants:** Technique of transformation – Agrobacterium mediated and physical methods (Microprojectile and electroporation) Applications of transgenic plants. Edible Vaccines from plants – Banana, Watermelon.

#### **Animal Biotechnology**

#### $\mathbf{UNIT} - \mathbf{III}$

Culture Media: Composition and importance of Serum.

a) Primary Culture – disaggregation of tissue, isolation of tissue, enzyme disaggregation, and mechanical disaggregation.

b) Secondary Culture – transformed animal cells and continious cell lines.

#### $\mathbf{UNIT} - \mathbf{IV}$

Artificial insemination (AI) and separation of semen by flow cytometry.

*In vitro* fertilization (IVF), embryo transfer, super ovulation, synchronization of Estrus, collection and transfer of embryos, cryopreservation.

**Transgenic Animals:** Techniques and Applications and Transgenic mice and sheep. Intellectual property rights (IPR), Patents, trade secrests, copyright and trademark.

# Practicals

- 10. Preparation of media, and initiation of callus from any one selected plant species
- 11. Micropropogation of plants (any one)
- 12. Anther culture for production of haploid plants
- 13. Preparation of synthetic seeds
- 14. Isolation and fusion of protoplast
- 15. Preparation of media and culture of animal cells/tissues
- 16. Cell disaggregation and cell counting
- 17. Cytotoxicity of the cells using the dye MTT method
- 18. Sperm analysis sperm count, mobility and viability.

# **Reference:**

# **Plant Biotechnology**

- 8. Ravishankar G.A. and Venkataraman L.V. (197) Biotechnolgoy Applications of plant Tissue & culture. Oxford & IBH Publishing Co, Pvt. Ltd.
- 9. Bhan (1998) Tissue Culture, Mittal Publications, New Delhi.
- 10. Islan A.C (1996) Plant Tissue Culture, Oxford & IBH Publishing Co. Pvt. Ltd.
- 11. Lydiane Kyte & John Kelyn (1996) Plants from test tubes. An introduction to Micropropogation Edition) Timber Press, Partland.
- 12. Kumar H.D. (1991) A text book on Biotechnology (2nd Edition) Affiliated East West Press Priva Ltd. New Delhi.
- 13. Chrispcel M.J. and Sdava D.E. (1994) Plants, Genes and Agriculture. Jones and Barlett Publisher Boton.
- 14. Reinert J. and Bajaj Y.P.S. (197) Applied and Fundamental Aspects of Plant Cell, Tissue, and Orga Culture, Narosa Publishing House.

# Animal Biotechnology

- 4. Ian Freshney (4th Edition)
- 5. Buttler.
- 6. Elements of Biotechnology P.k. Gupta (1st Edition -2000) Rastogi Publications.

#### <u>SEMESTER – VI</u> <u>PAPERL – VIII: MICROBIAL BIOTECHNOLOGY AND ENVIRONMENTAL</u> <u>BIOTECHNOLOGY</u> (w.e.f 2014-15)

## UNIT I

Microbial Biotechnology: Scope, Techniques, Chronological Development, Microbial diversity and its use.

Industrial Production: amylase, protease, xanthan, ethanol, citric acid and HBsAg by microbes.

## UNIT II

Production of microbial metabolites: Amino acids (glutamine), Antibiotics (Ampicillin) and other secondary metabolites (Streptomycin, Penicillin).

#### UNIT – III

Environmental components, Environmental pollution and its types, Non-renewable and renewable energy resources.

Conventional fuels and their major impacts: Global warming and greenhouse effect, Global Ozone Problem, Acid rain, Eutrophication, Biomagnification, Concept of clean fuel technology: Biomass energy and biofuels

# $\mathbf{UNIT} - \mathbf{IV}$

Biodegradation and bioremediation of major pollutants, Superbug biomineralisation: Use of microbial technology for mining.

Treatment of municipal solid and liquid wastes, Environmental impact assessment and Environmental audit.

Bioassessment of Environmental Quality, Biofertilizers and Biopesticides.

#### Practicals

- 8. Estimation of citric acid from Aspergillums Culture.
- 9. Estimation of lactic acid and lactose.
- 10. Immobilization of Yeast cells.
- 11. Preparation of wine.
- 12. Estimation of Alcohol by Specific gravity method.
- 13. Estimation of BOD (2 Samples)
- 14. VAM staining

#### References

- 11. Microbial Biotechnology (Fundamental of applied Microbiology) Alexandar N. Glazer & Hiroshi Nikaido
- 12. Fermentation Microbiology and Biotechnology. El mans, E.M.T., and Bryce, C.F.A. 2002. Taylor & Francis group.
- 13. Fundamentals of Biotechnology, Prave, P. Faust, V. Sitting, W. and Sukatseh, D.A. (eds). 1987. WCH Weinhein.
- 14. Principles of Fermentation Technology. Stanbury P.F. and Whitaker, A. 1984. Pergamon Press.
- 15. Waste water engineering treatment, disposal and reuse, Metcalf and Eddy Inc., Tata McGraw Hill, New Delhi.
- 16. Environmental Chemistry, AK. De, Wiley Eastern Ltd, New Delhi.
- 17. Introduction to Biodeterioration, D.Allsopp and K.J. Seal, ELBS / Edward Arnold.
- 18. Bioremidation, Baaker, KH and Herson D.S., 1994. Mc.GrawHill Inc, NewYork.
- 19. Industrial and Environmental Biotechnology Nuzhat Ahmed, Fouad M. Qureshi and Obaid Y. Khan, 2006. Horizon Press.
- 20. Environmental Molecular Biology, Paul. A, Rochelle, 2001. Horizon Press.

# B.Sc., II year Degree Examinations, November/ December III Semester Biotechnology 3319Biophysical Techniques (W.E.F 2016-17)

**Time -3hrs** 

Max Marks: 70

Section – A

# Answer any FIVE Questions (Each question carries 4 Marks )

- 1. Spectroflorometry applications
- 2. Beer's law derivation
- 3. Partiotion principle
- 4. TLC
- 5. Migration of Ions in electric field
- 6. Types of gels
- 7. Ionization chambers
- 8. Autoradiography
- 9. Basic principles of RCF
- 10. ANOVA

## Section – B Answer ALL of the following questions (Each question carries 10 Marks )

11.(A)Describe about the Derivations and deviations of Beer's law

(Or)

(B) Explain the types, instrumentation and Applications of Spectroscopy

12.(A)Discuss the theory of TLC, Ion -exchange and Affinity Chromatography

(Or)

(B)Describe the principle, instrumentation and applications of HPLC

13.(A) Explain about Principle and applications of Paper electrophoresis.

(Or)

(B) Write complete details of SDS-PAGE

14.(A).Explain about radio activity- in detail

(Or)

(B) Write an essay about Isotopes

15.(A) Explain the types of Centrifugation

(Or)

(B)Describe Databases, Search tools and applications in Biotechnology

# 2017-2018

# B.Sc., I year –Semester-I MICROBIOLOGY AND CELL BIOLOGY (w.e.f 2017-2018)

#### UNIT I

## HISTORY, DEVELOPMENT AND MICROSCOPY

History and development of microbiology: contributions of Louis Pasteur, Robert Koch and Edward Jenner. Microscopy: Compound microscopy: Numerical aperture and its importance, resolving power, oil immersion objectives and their significance, principles and applications of dark field, phase contrast, fluorescent microscopy. Electron microscopy: Principle, ray diagram and applications, TEM and SEM, comparison between optical and electron microscopy.

Stains and staining procedures: Acidic, basic and neutral stains, Gram staining, Acid fast staining, Flagella staining, Endospore staining.

#### UNIT-II BACTERIA & VIRUSES A.BACTERIA

Bacterial morphology and subcellular structures, general morphology of bacteria, shapes and sizes, generalized diagram of typical bacterial cell.Slime layer and capsule, difference between the structure, function and the position of the two structures. Cell wall of gram +ve and Gram -ve cells, Prokrayotic classification.

General account of flagella and fimbriae.Chromatin material, plasmids; definition and kind of plasmids (conjugative and non-conjugative) F, R, and Col plasmids.

A brief idea Bergey's manual. Morphology of archaea, archaeal cell membrane (differences between bacterial and archaeal cell membrane), other cell structures, concept of the three distinct archaea groups.

#### B.VIRUSES:

General Characters of viruses, difference between virus and typical microbial cell structure ,diferent shapes and symmetries with one example of each type, classification of viruses on the basis of nucleic acids, phage and animal cell viruses, example of each and their importance. Brief idea of lytic cycle and lysogeny.

#### UNIT III

#### **MICROBIAL NUTRITION:**

Types of Microbial nutrition.

Basic nutritional requirements: Basic idea of such nutrients as water, carbon, nitrogen, sulfur and vitamins etc., natural and synthetic media, nutritional classification of bacteria. Selective and Differential media, Enriched media, Enrichment media.

# UNIT IV: MICROBIAL GROWTH AND CONTROL:

#### **GROWTH:**

Growth rate and generation time, details of growth curve and its various phases. Concepts of synchronous cultures, continuous and batch cultures (chemostat and turbidostat). **Measurement of growth.** 

Physical conditions required for growth : Temperature (Classification of microorganisms on the basis of temperature requirements), pH etc. Pure cultures and cultural characteristics. Maintenance of pure culture.

**Microbial Control:** Terminologies - Sterilization, disinfection, antiseptic, sanitization, germicide, microbistasis, preservative and antimicrobial agents.

Mechanism of cell injury: Damage to cell wall, cell membrane, denaturation of proteins, inhibition of protein synthesis, transcription, replication, other metabolic reactions and change in supercoiling of DNA.

**Physical control:** Temperature (moist heat, autoclave, dry heat, hot air oven and incinerators), dessication, surface tension, osmotic pressure, radiation, UV light, electricity, ultrasonic sound waves, filtration.Chemical control: Antiseptics and disinfectants (halogens, alcohol, gaseous sterilization. Concept of biological control. Air filtration.

### UNIT V

#### CELL BIOLOGY

**Eukaryotic Cell** – Structure and function of the following nucleus , nuclear membrane , nucleoplasm, nucleolus, golgi complex, Mitochandria, Chloroplast, endoplasmic reticulum, lysosomes, peroxisomes, glyoxisomes and vacuoles.

#### Plant Cell wall :

Cytoskeleton (Micro and Macro filaments, microtubules) and cell locomotion. Mitosis and meiosis. Brief idea of cell cycle.Muscle and nerve cell structure, synaptic transmission and neuromuscular junctions.

### B.Sc., - I year SEMESTER-I Practicals BIOTECHNOLOGY MICROBIOLOGY & CELL BIOLOGY

- 39. Demonstration, Use and care of microbiological equipments
- 40. Preparation of media, sterilization and isolation of bacteria
- 41. Isolation of bacteria from water, soil and vegetables
- 42. Demonstration of motility of bacteria
- 43. Simple staining of Bacteria
- 44. Gram's Staining of bacteria
- 45. Acid fast Staining of bacteria- Demonstration
- 46. Endospore staining
- 47. Demonstration of Starch hydrolysis by bacterial cultures
- 48. Growth of fecal coliforms on selective media
- 49. Isolation of pure culture by Pour plate method
- 50. Isolation of pure culture by Streak plate method
- 51. Anaerobic cultivation of microorganisms
- 52. Cultivation of yeast/Mucor/Rhizopus
- 53. Antibiotic Sensitivity test
- 54. Oligodynamic action of Metals-Demonstration
- 55. To study Germicidal effect of UV-Light on bacterial Growth
- 56. Stages of Mitosis-Demonstration

57. Stages of Meiosis (Permanent slides)

#### Note: - Mandatory to perform at least ten practicals.

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### B. Sc., I – Semester II

# MACROMOLEULES, ENZYMOLOGY AND BIOENERGETICS (w.e.f 2017-18)

#### UNIT I

#### NUCLEIC ACIDS AND CHROMOSOMES

Chemical structure and base composition of nucleic acids, Chargaff's rules, Watson Crick Model (B-DNA), deviations from Watson-Crick model, other forms of DNA (Aand Z-DNA), forces stabilizing nucleic acid structures, (hydrogen bonds and hydrophobic associations, base stacking). Maxam and Gilbert DNA sequencing and Sanger's method.

Concept of prokaryotic genes and eukaryotic genes: Definition of a gene, concept of split genes, introns, exons, spacers, C-value and C-value paradox, basic idea of Cot curves. Chromatin structure: Nucleosome structure (10 nm fibre, experiments leading to discovery of nucleosomal structure, types of histones, arrangement of histones in the octamer, H1 histone and its role, role and length of linker DNA), 300 nm fibers (arrangement of nucleosome in a helical structure), domain and loop structure (further compacting of 300 nm fibre, role of scaffolding proteins). Role of telomere and centromere, telomeric and centromeric repeat sequences.

### UNIT II

### Amino acids and Proteins

Amino acids: Structure of amino acids occurring in proteins, classification of amino acids (pH based, polarity based and nutrition based), Physico-chemical properties of amino acids (solubility, boiling and melting points, reactions like Edman's, Sanger's, Dansyl chloride, ninhydrin). Titration curves of neutral, basic and acidic amino acids.

Primary structure of proteins: Determination of primary structure (end group analysis, cleavage of disulfide bonds, amino acid composition, use of endopeptidase specificity, sequence determination, assignment of disulfide position).

Secondary structure of proteins: The -helix, -structures (parallel, antiparallel, mixed, -turn).

Tertiary structure of proteins: Forces that stabilize the structure (electrostatic forces, hydrogen and disulfide bonds, hydrophobic associations), myoglobin as an example of tertiary structure, concept of domains, protein denaturation.

Quaternary structure of proteins: Forces stabilizing quaternary structure, advantages of oligomeric proteins.

### UNIT-III

### CARBOHYDRATES

Definition, classification, nomenclature of carbohydrates, structures of monosaccharides, disaccharides and polysaccharides (structures of starch and glycogen as examples of homopolysaccharides). Concept and examples of heteropolysaccharides.

**Lipids** - Types of lipids, structures of saturated and unsaturated fatty acids, triglycerides, phospholipids, plasmalogens, gangliosides and sphingolipids. Terpenoids and isoprenoids - definition and representative structures, steroids. Concept of acid value, saponification value and iodine value. Chemistry of Porphyrines, Heme, Cytochromes, and Chlorophylls

### UNIT -V ENZYMES :

Terminology: Active site , allosteric site, Holo enzyme, apoenzyme, co-enzyme

substrate, inhibitor, activator, modulator etc.Classification and nomenclature. Concept of isoenzymes (example Lactate Dehydrogenase) and multienzymes (example pyruvate dehydrogenase) Substrate Specificity (bond specificity, group specificity, absolute specificity, stereo-specificity, proof-reading mechanism), lock and key and induced fit models.

Concept of Allosteric enzymes( Brief idea of ATPase as an example, mechanisms of calalysis

: Acid base, covalent and metal ion catalysis.

Assay of Enzymes: Concept of activity, specific activity, turnover number, units of enzyme activity (katal, international unit), spectrophotometric methods of assay of enzymes (simple and coupled assay), very brief idea of other methods.

Enzyme kinetics: Michaelis-Menten equation, effect of substrate concentration, effect of enzyme concentration, effect of p H and temperature, temperature quotient, single reciprocal( Eadie-Hoffstee equation) and double reciprocal plots( Lineweaver-Burke plots), enzyme inhibition kinetics (reversible inhibition types – competitive, uncompetitive and non-competitive), brief idea of irreversible inhibition.

#### UNIT V

**Bioenergetics**: Concept of free energy, Entropy, Enthalpy & Redox Potential. Concept of high energy bonds as related to the structure of ATP, Phosphoenolpyruvate, Creatine phosphate etc.

Glycolosis (Pathway, entry of other monosaccharides and di saccharides, regulations, inhibitors) Gluconeogenesis : Bypass reactions.

39. Formal titration of Glycine- Demonstration

40. Quantitative estimation of proteins by Biuret method

41. Demonstration of Albumin and Ag ratio in Serum

42. Estimation of DNA by DPA method

43. Estimation of RNA by Orcinol method

44. Quantitative estimation of Amino acids using Ninhydrin reaction

B.Sc., - I year SEMESTER-II Practicals BIOTECHNOLOGY MACRO MOLECULES & ENZYMOLOGY

45. Quantitative analysis of Sugars and proteins

46. Quantitative estimation of Sugars ( Dinitro salisilic acid method ) – Demonstration

47. Estimation of Glucose by Benedicts Quantitative method

48. Quantitative estimation of proteins by Lowry's method

49. Extraction and quantification of total Lipids

50. Determination of Saponification value of fats

51. Determination of Acid value of fats

52. Isolation of Urease and demonstration of its activity

- 53. Assay of protease activity
- 54. Preparation of starch from potato and its hydrolysis by Salivary amylase
- 55. Assay of Alkaline phosphatase
- 56. Immobilization of enzymes / cells by Entrapment in Alginate gel
- 57. Effect of temperature / pH on enzyme activity

Note: - Mandatory to perform at least ten practicals.

### MODIFIED SYLLABUS

### B.Sc., SEMESTER III BIOPHYSICAL TECHNIQUES (w.e.f 2016-17)

#### UNIT – I :

**Spectrophotometry :** Spectrum of light, absorption of electromagnetic radiations, Beer's law – derivation and deviations, extinction coefficient. Instrumentation of UV and visible spectrophotometry, Double deam spectrometer ; dual – wavelength spectrometer, Applications of UV and visible spectrotometry. Spectrofluorometry : principle , instrumentation and application.

#### UNIT II:

**Chromatography:** Partition principle, coefficient, nature of partition forces, brief account of paper chromatography. Thin layer chromatography and column chromatography. Gel filtration: Concept of distribution coefficient, types of gels and glass beads, applications. Ion-exchange chromatography: Principle, types of resins, choice of buffers, applications including amino acid analyzer. Affinity chromatography: Principle, selection of ligand, brief idea of ligand attachment, specific and non-specific elution, applications. HPLC.

### UNIT III

**Electrophoresis :** Migration of ions in electric fields, Factors affecting electrophoretic mobility. Paper electrophoresis, Gel electrophoresis :- Types of gels, Solubilizers, Procedure, Column & slad gels Detection, Recovery & Estimation of macromolecules. SDS – PAGE Electrophoresis and applications. Isoelectric focusing, Puised-field gel electrophoresis.

#### UNIT IV

**Isotopic tracer technique :** Radioactive & stable isotopes, rate of radioactive decay. Units of radioactivity. Measurement of radioactivity :- Ionization chambers, proportional counters, Geiger – Muller counter, Solid and liquid scintillation counters (basic principle, instrumentation and rechnique), Cerenkov radiation. Measurement of Stable isotopes : Falling drop method for deuterium measurement, Mass spectrometry. Principles of tracer technique, advantages and limitations, applications of isotopes in biotechnology (distribution studies, metabolic studies, isotope dilution technique, metabolic studies, clinical applications, autoradiography).

### UNIT – V

**Centrifugation :** Basic principles, concept of RCF, types of centrifuges (clinical high speed and ultracentrifuges). Preparative centrifugation: Differential and density gradient centrifugation, applications (Isolation of cell components). Analytical centrifugation: Sedimentation coefficient, determination of molecular weight by sedimentation velocity and sedimentation equilibrium methods.

Biostatistics Basic concepts of mean, median, mode Standard deviation and Standard error. Introduction to ANOVA Applications of basic statistics

Bio Informatics: Introduction, Databases, Search tolls, Applications in Bo Technology.

### B.Sc., SEMESTER IV IMMUNOLOGY (w.e.f 2016-17)

### UNIT I

Immune system : Organs and cells of immune system immunity, innate immune mechanism Acquired immune mechanism, Antigen,

Humoral immunity, main pathways of complement system.

UNIT II

**Antibody and Antigen :** Antibody structure and classes, Antibody diversity, types of Antigens Antigenecity (factors affecting antigenecity). Complement system.

### UNIT III

**Immunity :** Cell mediated immunity : TC mediated immunity, NK cell mediated immunity, ADCC brief description of cytokines and MHC (MHC types and diversity) Autoimmunity.

UNIT IV

**Hypersensitivity and vaccination :** General features of hypersensitivity various types of hypersensitivity, Vaccination : Discovery, principles, significance, Types of Vaccines **UNIT V** 

**Immunological Techniques :** Antigen – antibody reactions : Precipition, agglutination, complement fixation, immunodiffusion, ELISA. Hybridoma technology : Monoclonal antibodies and their applications in immunodiagnosis.

### **PRACTICALS : IMMUNOLOGY**

- 10. Antigen antibody reaction
- 11. Determination of blood group
- 12. Widal test
- 13. Ouchterloney immunodiffusion-A demonstration
- 14. Radial immunodiffusion-A Demonstration
- 15. ELISA -A demonstration

16. Coombs Test

17. Isolation of casein by isoelectric precipitation

18. Production of antibodies and their titration

### Note : - Mandatory to perform at least 5 Practical's

### B. Sc. III –Semester V MOLECULAR BIOLOGY (w.e.f 2017-18)

### Unit I:

**Genome Structure:**Watson and Crick model of DNA; Genome organization with specific reference to prokaryotic and eukaryotic genomes; Genome size. Concepts of Genetic Material, Gene, Chromosome and Genome. Histone Proteins. Experiments to prove DNA as genetic material (Griffith experiment, Hershey- Chase experiment)

### Unit II

**DNA Replication**:Enzymology of replication (DNA polymerase I, pol II and III, helicases, topoisomerases, single strand binding proteins, DNA melting proteins, primase. Proof of semiconservative replication, Replication origins, initiation, elongation, and termination. Rolling circle replication of DNA. Difference between Prokaryotic and Eukaryotic DNA Replication.

### Unit III

**Transcription : Central dogma of molecular biology.** Transcription in Prokaryotes: Enzymatic synthesis of RNA: Basi& features of transcription, classes of RNA molecules. E.coli RNA polymerases, types and structure (core enzyme and holo enzyme, sigma factor), transcription mechanism in prokaryotes-Promoter, initiation, elongation, proof reading and termination (Rho dependent and Rho independent). Trancription in Eukaryotes: Promoter, RNA polymerases of eukaryotes, Reverse transcription.

### Unit IV:

### **Genetic Code and Protein Synthesis**

Genetic code: Features of genetic code, Structure of mRNA Post-transcriptional modifications of eukaryotic hnRNA – Capping, methylation, polyadenylation and RNA splicing and splicing mechanisms. brief structure of tRNA, the adaptor hypothesis, attachment of amino acids to tRNA. Codon-anticodon interaction - the wobble hypothesis. Initiation, elongation, termination of protein. post translational modifications.(Post translational modifications).

### Unit V

### Gene Expression and regulation

Regulation of gene expression; Regulation of transcription (regulatory elements and transcription factors).Clustered genes and the operon concepts - Negative and positive control of the Lac Operon, trp operon, Control of gene expression. Poly and Mono cistronic m-RNA. Translational regulation of gene expression in prokaryotic and eukaryotic organisms.

### PRACTICALS 55191P MOLECULAR BIOLOGY

- 1. Isolation of plasmid DNA from bacteria
- 2. Isolation of genomic DNA from *E.coli*
- 3. Isolation of DNA from sheep liver
- 4. Isolation of DNA from plant material (Onion bulb)
- 5. Quantitative estimation and purity analysis of nucleic acids
- 6. Separation of DNA by Agarose gel Electrophoresis
- 7. Demonstrate Spliceosome
- 8. Demonstrate cloverleaf model of tRNA
- 9. Demonstrate different forms of DNA
- 10. Demonstrate Lac Operon.
- 11. Demonstrate blue white colonies

#### **Recommended Books:**

- 1. Molecular Biology of the Cell B. Alberts, D. Bray, J. Lewis, M. Raff, K. Roberts and J.D.Watson (Garland Publishing, New York and London)
- 2. Molecular Biology A Comprehensive Introduction to Prokaryotes and Eukaryotes D. Freifelder (Jones and Bartlett, USA)
- 3. Cell and Molecular Biology by Robertis and Roberties (Publ: Waverly)
- 4. Recombinant DNA : A Short Course J.D. Watson, J. Tooze and D.T.Kurtz (Scientific American Book, W.A.Preemon).
- 5. Modern Genetics (2<sup>nd</sup> Edition, 1984) A.J. Ayala and W. Castra (Goom Helns, London).
- 6. Text book of Biotechnology by HK Das (Wiley Publication)
- 7. Genes VIII. (1997) Benjamin Lewin (Oxford University Press).
- 8. Molecular Cloning: A Laboratory manual, J. Sambrook, E.F frisch and T. Maniatis, Old Spring Harbor Laboratory Press New York, 2000.

#### B. Sc. III – Semester V RECOMBINANT DNA TECHNOLOGY (Elective Theory)

#### (w.e.f 2017-18)

#### Unit I:

**Tools of rDNA Technology-** Restriction and Modification enzymes. Classification of restriction endo nucleases (Type-I, Type-II and Type-III). Enzymes used in molecular cloning; Polymerases, ligases, phosphatases, kinases, nucleases and terminal transferase. Cloning vehicles - Plasmid, Bacteriophage, cosmids, shuttle vectors, expression vectors, Ti derived Vectors, artificial chromosomes (BAC, YAC).

#### Unit II

**Cutting and joining DNA** (cohesive end ligation, methods of blunt end ligation). Use of linkers, adaptors and homopolymer tails. Selection of transformed cells. Screening methods (Genetic marker and blue white screening). Construction of genomic and cDNA libraries. Advantages of cDNA libraries.

#### Unit III:

**Techniques of rDNA Technology:** Polymerase chain reaction technique (Components in PCR and PCR conditions) and Reverse transcription-PCR. Principles involved in blotting techniques-Southern, Northern, and Western blotting Methods of gene sequencing – Maxam - Gilberts and Sanger's dideoxy chain termination methods.

#### Unit IV:

**Methods of gene transfer**-Transformation, Electroporation, microinjection, microprojectile bombardment (gene gun method) and Plant transformation techniques *–Agrobacterium tumefaciens* mediated, Particle bombardment, electroporation and viral mediated. Transfection.

#### Unit V:

**Applications** of recombinant DNA technology in Agriculture (Transgenic Plants) Medicine (production of Insulin, Growth harmone, Tissue plasmogen activator and HBsAg vaccine. Principal and applications of DNA fingerprinting technique.

Application of plant transformation (Herbicide resistance-Phosphoinothricin, glyphosate, Insect resistance – transgenic Bt crops).

#### PRACTICALS 55192P rDNA TECHNOLOGY (Elective Lab)

- 1. Isolation of plasmid DNA.
- 2. Restriction digestion of DNA and its electrophoretic separation.
- 3. Ligation of Restriction digested DNA
- 4. Transformation in Bacteria using plasmid DNA
- 5. Demonstration of PCR
- 6. primer designing
- 7. Activity of DNase and RNase on DNA and RNA (Demo)

8. Study of vectors PBR, PUC

### **Recommended Books:**

- 1. Molecular Cloning: A Laboratory manual, J. Sambrook, E.Ffrisch and T. Maniatis, Old Spring Harbor Laboratory Press New York, 2000
- 2. DNA Cloning : a Practical Approach, DM Glover and BD Hames, IRL Press
- 3. Methods in Enzymology Vol. 152, Guide to Molecular Cloning Techniques, SL Berger and AP Kimmel, Academic Press, Inc San Diego, 1998
- 4. DNA Science. A first Course in Recombinant Technology, DA Mickloss and GA Freyer, Cold Spring Harbor Laboratory Press, New York 1990
- 5. Milestones in Biotechnology. Classic papers on Genetic Engineering. JA Davies and WS Reznikoff, Butterworth-Heinemann, Boston, 1992
- 6. Concepts of Biotechnology D. Balasubramanian.
- 7. Principles of Gene Manipulation by Old and Primrose, Blackwell
- 8. Gene cloning by T.A. Brown

### **GENETICS** (Elective Theory)

#### (w.e.f 2016-17)

### UNIT I

**Mendels Laws and Inheritance:** Mendel experiments, Mendel Laws and deviations: incomplete dominance and Co dominance, Back cross, Test cross Penetration and pleiotropism, Recessive and Dominant epistatic gene interactions. Concept of multiple alleles

### UNIT II

**Genes and their variations:** Structure of gene, gene and environment, gene copies of prokaryotic and Eukaryotic chromosomes. Eukaryotic chromosome organization, histone proteins. Karyotype, special chromosomes – Lampbrush and polytene

### Unit III:

**Gene mutations:** Mutagenesis – Spontaneous and induced (Chemical and physical) mutations; Natural and induction of mutations, point mutations, frameshift mutations, auxotrophic conditional and suppressor mutations. Chromosomal aberrations.

### **UNIT IV:**

**DNA Damage and DNA Repair**: Light induced repair, Excision repair and mismatch repair, Post replication repair, Rec gene and its role in DNA repair, SOS repair and SOS response

### Unit V:

**Transposable elements:** Structure and Molecular basis of AC-DS transposition in maize, "P" element of Drosophila and hybrid dysgenesis, Yeast "T<sub>7</sub>" elements, Retroposans

### **PRACTICAL GENETICS (Elective Lab)**

- 1. Study of different phases of mitosis in onion root tips and meiosis in *Allium cepa* flower buds.
- 2. Karyotyping in Allium or Drosophila.
- 3. Determination of multiple allele frequencies of leaf scars in Trifolium.
- 4. Problems and assignments in Mendilian genetics Monohybrid, dyhybrid, Testcross and backcross
- 5. Induction of chromosomal aberrations by chemical mutagenesis in Allium (or any plant).
- 6. Isolation of auxotrophic mutants (plants or insects).
- 7. Repair of DNA by Photo activation of Photolyase in bacteria.
- 8. Mutation of bacteria by UV.
- 9. Demo on Chemical induced mutation in bacteria

### **Recommended Books**

- 1. Molecular Biology. Freifelder, D. 1990. Narosa Publication House, New Delhi
- 2. Principles of Genetics. Gardner, E.J and D.P. Snustad. 1996. John Willey, New York.
- 3. Genetics. Sambamurthy, A.V.S.S. 1999. Narosa Publishing House, New Delhi.
- 4. Principles of Genetics. Sinnot, E.W., L.C Dunn and T. Dobzhansky 1958. McGraw Hill, New York.
- 5. Theory and Problems in Genetics. Stansfield, W.D. 1991. McGraw Hill, New York.
- 6. Genetics. Strickberger, M.W. 1996. 3<sup>rd</sup> Edn. McMillan, New York.
- 7. Genetics A Conceptual Approach. Pierce, B. A.2006. 2<sup>nd</sup> Edn. W.H.Freeman and Company, New York.

#### B. Sc. III – Semester VI PLANT AND ANIMAL BIOTECHNOLOGY (w.e.f 2017-18)

#### UNIT I:

**Cell and tissue culture:** Histological perspective of Plant tissue culture. Basic requirement for tissue culture laboratory. Totipotency, Tissue culture media (composition and MS media preparation). Aseptic techniques/sterilization methods used in plant tissue culture. Initiation and maintenance of callus and suspension cultures.

#### **UNIT II:**

**Applications of Plant Tissue culture**, meristem culture, clonal propogation, production of haploids (Anther culture), somatic embryogenesis, protoplast culture and somatic hybridization-Cybrids, Hybrids. Somoclonal variation-sources and its applications. Cryopreservation.

Production of secondary metabolites –Classication isolation, characharization and biosynthetic pathway of Secondary metabolites.

### UNIT III:

**Tools and techniques of animal cell and tissue culture**: Culture media, growth factors, laboratory facilities. CO2 incubator, Cell count-haemocytometer, coulter method, MTT based Assay. Characteristics of cells in culture: Contact inhibition, anchorage dependence, cell-cell communication etc.; Cell senescence; cell and tissue response to trophic factors. Primary culture, immortal cells, cell lines. Maintenance of cell lines in the laboratory-subculture, trypsinisation and cryopreservation.

### UNIT IV:

**rDNA products:** Brief idea about recombinant DNA products in medicine (insulin, somatostatin, vaccines), Concept of Gene therapy, Production of recombinant vaccines – hepatitis. Concept of transgenic animals. In vitro fertilization and embryo transfer in humans and farm animals. Cloning-Concepts of nuclear transfer and creation of Dolly. Stem cells-types and applications.

#### UNIT V:

**IPR:** Intellectual property rights. Types and importance of IPRs (Copy rights. Patents, GIs...) International and national organizations, agencies and treaties. Regulation of GMO/transgenics. ethical aspects of Biotechnology.

### PRACTICALS: 66191-P PLANT AND ANIMALBIOTECHNOLOGY

- **1.** Preparation of MS medium
- 2. Sterilization methods
- **3.** Callus culture
- **4.** Demonstration of Suspension Culture
- **5.** Determination of seed viability by TTC
- **6.** Establishing primary cell culture of chicken embryo fibroblasts.
- 7. Animal tissue culture maintenance of established cell lines.
- **8.** Cell count by Haemocytometer.
- **9.** Microphotography Cell line photographs.

### **Recommended Books**

- 1. Culture of Animal Cells A Manual of Basic Technique. Freshney, R .I. 2006. 5<sup>th</sup> Edn. John Wiley and Sons, USA.
- 2. Cell Culture. Yadav, P.R and R. Tyagi. 2005. Discoery Publishing House, New Delhi.
- 3. Principles of Cloning. Jose Cibelli, Robert P. Lanza. Keith H.S. Campbell. 2002. Academic Press, London.
- 4. Animal Cell Technology Principles and practices. Butter, M. 1987. Oxford Press, USA.
- 5. Elements of Biotechnology. Gupta, P.K. 1996. Rastogi and Company, Meerut.
- 6. Plant Tissue Culture: Theory and Practice, a Revised Edition. Bhojwani, S.S and M.K.Razdan.2004.Panima Publishing Corporation, New Delhi.
- 7. Biotechnology-Fundamentals and applications. Purohit, S. 2000. 3<sup>rd</sup> Edn. Agrobios, New Delhi.
- 8. An Introduction to Plant Tissue Culture. Razdan, M.K. 1995. Oxford and IBH Publishing Company, New Delhi.
- 9. Plant Tissue Culture and Molecular Biology, Applications and Prospects. Srivastava, P.S. 1998. Narosa Publishing House, New Delhi.
- 10. Intellectural Property rights-Deborah, E. Bouchoux, Cengage Learning

### 66192- ENVIRONMENTAL BIOTECHNOLOGY (Elective theory)

### (w.e.f 2016-17)

### Unit I

**Principles of Ecology,** Water and terrestrial ecosystems, Bio-geo chemical cycles - Carbon, Nitrogen cycles. Role of microbes in bio-geochemical cycles.

### Unit II

**Inorganic and Organic pollutants** of air, land and water; maintenance of standards, Environmental monitoring. Detection, treatment and prevention of pollution. Biological indicators

### Unit III

**Biocides,** Four stage alternatives, Refuse disposal - Treatment methods, effluent from pharmaceuticals, fertilizers, pulp and paper industry.

### Unit IV

Waste water management - Aerobic and anaerobic treatment, primary, secondary and tertiary treatment of municipal wastes, Solid waste management.

### Unit V

**Bioremediation**, Biodegradation of recalcitrant compounds and the role of genetically engineered microbes and genetically modified organisms in the environmental management.

### PRACTICALS-66192- ENVIRONMENTAL BIOTECHNOLOGY (Elective Lab)

- 1. Detection of coliforms for determination of the purity of potable water.
- 2. Determination of total dissolved solids of water
- 3. Determination of Hardness and alkalinity of water sample.
- 4. Determination of dissolved oxygen concentration of water sample
- 5. Determination of biological oxygen demand of sewage sample
- 6. Determination of chemical oxygen demand (COD) of sewage sample.
- 7. Isolation of xenobiotic degrading bacteria by selective enrichment technique
- 8. Estimation of heavy metals in water/soil
- 9. Estimation of nitrate in drinking water.
- 10. Preparation and formulation of microbial biopesticide (bacteria, fungi and viruses)
- 11. In vitro evaluation of medicinal plants against pathogenic microbes.
- 12. Effect of mycorrhizal fungi on growth promotion of plants.
- 13. Production of microbial fertilizers (Rhizobium, Azotobacter and AMF).

### **Recommended Books**

1. Ecology and Environment P.D.Sharma

2. Brock Biology of Microorganisms (9th edition) by Madigan, Martinko & Parker.2000. Prentice Hall.

- 3. Environmental Microbiology by Mitchell. 2009. John Wiley and Sons.
- 4. Environmental Microbiology by Grant & Long. 1981. Wiley.

5. Environmental Microbiology: A laboratory manual by Peppler, Gerba & Brendecke.1995. Academic Press.

### Industrial Biotechnology (Elective theory) (w.e.f 2016-17)

### Unit I:

**Isolation, Screening**, Preservation and Improvement of Industrially Important Microorganisms. Synthetic and Natural Medium, Precursors, Antifoams, Sterilization Methods and Inoculum Preparation.

### Unit II:

**Definition of bioreactor**, Basic principles of bioreactor. Classification of bioreactors. Analysis of batch, continuous, fed batch and semi-continuous bioreactors.

### Unit III:

**Ethanol Production** by Fermentation using Molasses, Starchy Substances. Production of Alcoholic Beverages like Beer and Wine. Production of Citric Acid by Submerged and Solid State Fermentations.

### Unit IV:

**Sources of Industrial Enzymes**, Production of Microbial Enzymes like Amylase and protease. Backer's Yeast and SCP Production. Production of Antibiotics : Penicillin .

### Unit V:

**Biotechnology Products**- Production of recombinant proteins having therapeutic and diagnostic applications( Insulin, Growth Harmone, Recombinant vaccines, Monoclonal Antibody).

### PRACTICALS Industrial Biotechnology (Elective Lab)

- 1. Isolation of bacteria / fungi from soil by serial dilution technique
- 2. Estimation of amylase from seeds.
- 3. Production of  $\alpha$  amylase from *Bacillus Spp.* by shake flask culture.
- 4. Production of alcohol or wine using different substrates.
- 5. Estimation of alcohol by titrimetry.
- 6. Estimation of alcohol by colorimetric method .
- 7. Production of citric acid.
- 8. Citric acid production by submerged fermentation.
- 9. Estimation of citric acid by titrimetry.
- 10. Demonstration of Fermentor and bioreactor

### **Recommended Books**

- 1. Biogas technology by b.T.Nijaguna
- 2. Food microbiology by m.R.Adams and M.O.Mass

- 3. Industrial microbiology by A.H.Patel
- 4. Industrial microbiology, A text book W.Clark
- 5. Industrial Microbiology by L.E.Casida
- 6. Introduction to Biotechnology by P.K.Gupta
- 7. Text book of Biotechnology by H.K.Das (Wily Publications)
- 8. Bioprocess engineering by Shuler (Pearson Education)
  - 9. Biotechnology: A Text Book of Industrial microbiology 2nd edition by Crueger. & Crueger. 1990. Sinauer Associates.
- 10.Microbial Biotechnology: Fundamentals of Applied Microbiology (2nd edition) by Glazer & Nikaido. 2007. Cambridge University Press.
- 11. Prescott & Dunn's Industrial Microbiology 4th edition Editor Reed. 1982. AVI Pub. Co.,

### MODEL QUESTION PAPER B.Sc., I year Degree Examinations, November December Model Question Paper I Semester Biotechnology 1119-Paper –I Microbiology and Cell Biology (W.e.f. 2017-18)

**Time -3hrs** 

#### Max Marks : 60

Section – A Answer any FIVE Questions (Each question carries 4 Marks )

5x4=20M

16. Conditions required for growth

19. Neuromuscular junction

12. Fluorsect microscope

11. Endospore staining

13. Pili

14. Heterotrophs

15. Artificial media

**20.** Ribosomes

17. Disinfection

18. Chemostat

#### Section – B

### Answer ALL of the following questions

5x8=40M

#### (Each question carries 8 Marks)

- 11.(A) Describe the contributions of Louis Pasteur and Edward Jenner to microbiology (Or)
  - (B) Explain about TEM with neat labeled Diagram
- 12.(A)Give an account on Ultra structure of Bacterial cell wall with a neat labeled diagram

(Or) (B)Describe the Lytic cycle with labeled diagrams

- 13. (A) Write about nutritional groups of Bacteria
  - 1. (Or) (B) Write a note on types of nutrient media
- 14. (A).Describe different phases of Growth Curve

2. (Or) (B) Explain the different methods of chemical sterilization

15. (A) Describe in detail about meosis

### 3. (Or)

(B)Explain about structure and functions of ER and Golgi complex

#### 2018-2019

### B.Sc., I year –Semester-I MICROBIOLOGY AND CELL BIOLOGY (w.e.f 2017-2018)

#### UNIT I

#### HISTORY, DEVELOPMENT AND MICROSCOPY

History and development of microbiology: contributions of Louis Pasteur, Robert Koch and Edward Jenner. Microscopy: Compound microscopy: Numerical aperture and its importance, resolving power, oil immersion objectives and their significance, principles and applications of dark field, phase contrast, fluorescent microscopy. Electron microscopy: Principle, ray diagram and applications, TEM and SEM, comparison between optical and electron microscopy.

Stains and staining procedures: Acidic, basic and neutral stains, Gram staining, Acid fast staining, Flagella staining, Endospore staining.

#### UNIT-II BACTERIA & VIRUSES A.BACTERIA

Bacterial morphology and subcellular structures, general morphology of bacteria, shapes and sizes, generalized diagram of typical bacterial cell.Slime layer and capsule, difference between the structure, function and the position of the two structures. Cell wall of gram +ve and Gram -ve cells, Prokrayotic classification.

General account of flagella and fimbriae.Chromatin material, plasmids; definition and kind of plasmids (conjugative and non-conjugative) F, R, and Col plasmids.

A brief idea Bergey's manual. Morphology of archaea, archaeal cell membrane (differences between bacterial and archaeal cell membrane), other cell structures, concept of the three distinct archaea groups.

#### B.VIRUSES:

General Characters of viruses, difference between virus and typical microbial cell structure ,diferent shapes and symmetries with one example of each type, classification of viruses on the basis of nucleic acids, phage and animal cell viruses, example of each and their importance. Brief idea of lytic cycle and lysogeny.

#### UNIT III MICROBIAL NUTRITION:

Types of Microbial nutrition.

Basic nutritional requirements: Basic idea of such nutrients as water, carbon, nitrogen, sulfur and vitamins etc., natural and synthetic media, nutritional classification of bacteria. Selective and Differential media, Enriched media, Enrichment media.

#### UNIT IV: MICROBIAL GROWTH AND CONTROL:

#### **GROWTH:**

Growth rate and generation time, details of growth curve and its various phases .Concepts of synchronous cultures, continuous and batch cultures (chemostat and turbidostat). **Measurement of growth.** 

Physical conditions required for growth : Temperature (Classification of microorganisms on the basis of temperature requirements), pH etc. Pure cultures and cultural characteristics.

Maintenance of pure culture.

**Microbial Control:** Terminologies - Sterilization, disinfection, antiseptic, sanitization, germicide, microbistasis, preservative and antimicrobial agents.

Mechanism of cell injury: Damage to cell wall, cell membrane, denaturation of proteins, inhibition of protein synthesis, transcription, replication, other metabolic reactions and change in supercoiling of DNA.

**Physical control:** Temperature (moist heat, autoclave, dry heat, hot air oven and incinerators), dessication, surface tension, osmotic pressure, radiation, UV light, electricity, ultrasonic sound waves, filtration.Chemical control: Antiseptics and disinfectants (halogens, alcohol, gaseous sterilization. Concept of biological control. Air filtration.

### UNIT V

### CELL BIOLOGY

**Eukaryotic Cell** – Structure and function of the following nucleus , nuclear membrane , nucleoplasm, nucleolus, golgi complex, Mitochandria, Chloroplast, endoplasmic reticulum, lysosomes, peroxisomes, glyoxisomes and vacuoles.

### Plant Cell wall :

Cytoskeleton (Micro and Macro filaments, microtubules) and cell locomotion. Mitosis and meiosis. Brief idea of cell cycle.Muscle and nerve cell structure, synaptic transmission and neuromuscular junctions.

### B.Sc., - I year SEMESTER-I Practicals BIOTECHNOLOGY MICROBIOLOGY & CELL BIOLOGY

- 58. Demonstration, Use and care of microbiological equipments
- 59. Preparation of media, sterilization and isolation of bacteria
- 60. Isolation of bacteria from water, soil and vegetables
- 61. Demonstration of motility of bacteria
- 62. Simple staining of Bacteria
- 63. Gram's Staining of bacteria
- 64. Acid fast Staining of bacteria- Demonstration
- 65. Endospore staining
- 66. Demonstration of Starch hydrolysis by bacterial cultures
- 67. Growth of fecal coliforms on selective media
- 68. Isolation of pure culture by Pour plate method
- 69. Isolation of pure culture by Streak plate method
- 70. Anaerobic cultivation of microorganisms
- 71. Cultivation of yeast/Mucor/Rhizopus
- 72. Antibiotic Sensitivity test
- 73. Oligodynamic action of Metals-Demonstration
- 74. To study Germicidal effect of UV-Light on bacterial Growth
- 75. Stages of Mitosis-Demonstration
- 76. Stages of Meiosis (Permanent slides)

#### Note: - Mandatory to perform at least ten practicals.

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### B. Sc., I – Semester II

### MACROMOLEULES, ENZYMOLOGY AND BIOENERGETICS (w.e.f 2017-18)

### UNIT I

#### NUCLEIC ACIDS AND CHROMOSOMES

Chemical structure and base composition of nucleic acids, Chargaff's rules, Watson Crick Model (B-DNA), deviations from Watson-Crick model, other forms of DNA (Aand Z-DNA), forces stabilizing nucleic acid structures, (hydrogen bonds and hydrophobic associations, base stacking). Maxam and Gilbert DNA sequencing and Sanger's method.

Concept of prokaryotic genes and eukaryotic genes: Definition of a gene, concept of split genes, introns, exons, spacers, C-value and C-value paradox, basic idea of Cot curves.

Chromatin structure: Nucleosome structure (10 nm fibre, experiments leading to discovery of nucleosomal structure, types of histones, arrangement of histones in the octamer, H1 histone and its role, role and length of linker DNA), 300 nm fibers (arrangement of nucleosome in a helical structure), domain and loop structure (further compacting of 300 nm fibre, role of scaffolding proteins). Role of telomere and centromere, telomeric and centromeric repeat sequences.

#### UNIT II

#### **Amino acids and Proteins**

Amino acids: Structure of amino acids occurring in proteins, classification of amino acids (pH based, polarity based and nutrition based), Physico-chemical properties of amino acids (solubility, boiling and melting points, reactions like Edman's, Sanger's, Dansyl chloride, ninhydrin). Titration curves of neutral, basic and acidic amino acids.

Primary structure of proteins: Determination of primary structure (end group analysis, cleavage of disulfide bonds, amino acid composition, use of endopepticlase specificity, sequence determination, assignment of disulfide position).

Secondary structure of proteins: The -helix, -structures (parallel, antiparallel, mixed, -turn).

Tertiary structure of proteins: Forces that stabilize the structure (electrostatic forces, hydrogen and disulfide bonds, hydrophobic associations), myoglobin as an example of tertiary structure, concept of domains, protein denaturation.

Quaternary structure of proteins: Forces stabilizing quaternary structure, advantages of oligomeric proteins.

### UNIT-III

#### CARBOHYDRATES

Definition, classification, nomenclature of carbohydrates, structures of monosaccharides, disaccharides and polysaccharides (structures of starch and glycogen as examples of homopolysaccharides). Concept and examples of heteropolysaccharides.

**Lipids** - Types of lipids, structures of saturated and unsaturated fatty acids, triglycerides, phospholipids, plasmalogens, gangliosides and sphingolipids. Terpenoids and isoprenoids - definition and representative structures, steroids. Concept of acid value, saponification value and iodine value. Chemistry of Porphyrines, Heme, Cytochromes, and Chlorophylls

UNIT -V ENZYMES : Terminology: Active site , allosteric site, Holo enzyme, apoenzyme, co-enzyme

substrate, inhibitor, activator, modulator etc.Classification and nomenclature. Concept of isoenzymes (example Lactate Dehydrogenase) and multienzymes (example pyruvate dehydrogenase) Substrate Specificity (bond specificity, group specificity, absolute specificity, stereo-specificity, proof-reading mechanism), lock and key and induced fit models.

Concept of Allosteric enzymes( Brief idea of ATPase as an example, mechanisms of calalysis

: Acid base, covalent and metal ion catalysis.

Assay of Enzymes: Concept of activity, specific activity, turnover number, units of enzyme activity (katal, international unit), spectrophotometric methods of assay of enzymes (simple and coupled assay), very brief idea of other methods.

Enzyme kinetics: Michaelis-Menten equation, effect of substrate concentration, effect of enzyme concentration, effect of p H and temperature, temperature quotient, single reciprocal( Eadie-Hoffstee equation) and double reciprocal plots( Lineweaver-Burke plots), enzyme inhibition kinetics (reversible inhibition types – competitive, uncompetitive and non-competitive), brief idea of irreversible inhibition.

### UNIT V

**Bioenergetics**: Concept of free energy, Entropy, Enthalpy & Redox Potential. Concept of high energy bonds as related to the structure of ATP, Phosphoenolpyruvate, Creatine phosphate etc.

Glycolosis (Pathway, entry of other monosaccharides and di saccharides, regulations,

inhibitors) Gluconeogenesis : Bypass reactions.

### B.Sc., - I year SEMESTER-II Practicals BIOTECHNOLOGY MACRO MOLECULES & ENZYMOLOGY

- 58. Formal titration of Glycine- Demonstration
- 59. Quantitative estimation of proteins by Biuret method
- 60. Demonstration of Albumin and Ag ratio in Serum
- 61. Estimation of DNA by DPA method
- 62. Estimation of RNA by Orcinol method
- 63. Quantitative estimation of Amino acids using Ninhydrin reaction
- 64. Quantitative analysis of Sugars and proteins
- 65. Quantitative estimation of Sugars ( Dinitro salisilic acid method ) Demonstration
- 66. Estimation of Glucose by Benedicts Quantitative method
- 67. Quantitative estimation of proteins by Lowry's method
- 68. Extraction and quantification of total Lipids
- 69. Determination of Saponification value of fats
- 70. Determination of Acid value of fats
- 71. Isolation of Urease and demonstration of its activity
- 72. Assay of protease activity
- 73. Preparation of starch from potato and its hydrolysis by Salivary amylase
- 74. Assay of Alkaline phosphatase
- 75. Immobilization of enzymes / cells by Entrapment in Alginate gel
- 76. Effect of temperature / pH on enzyme activity

## Note: - Mandatory to perform at least ten practicals.

#### **B.Sc., SEMESTER III**

#### BIOPHYSICAL TECHNIQUES (w.e.f 2018-19)

#### UNIT – I

**Spectrophotometry :** Spectrum of light, absorption of electromagnetic radiations, Beer's law – derivation and deviations, extinction coefficient. Instrumentation of UV and visible spectrophotometry, Double deam spectrometer ; dual – wavelength spectrometer, Applications of UV and visible spectrotometry. Spectrofluorometry : principle , instrumentation and application. Mass spectrometry

#### UNIT II :

**Chromatography :** Partition principle, coefficient, nature of partition forces, brief account of paper chromatography. Thin layer chromatography and column chromatography. Gel filtration : Concept of distribution coefficient, types of gels and glass beads, applications. Ion-exchange chromatography : Principle, types of resins, choice of buffers, applications including amino acid analyzer. Affinity chromatography : Principle, selection of ligand, brief idea of ligand attachment, specific and non-specific elution, applications. HPLC

### UNIT III

**Electrophoresis :** Migration of ions in electric fields, Factors affecting electrophoretic mobility. Paper electrophoresis, Gel electrophoresis :- Types of gels, Solubilizers, Procedure, Column & slad gels Detection, Recovery & Estimation of macromolecules. SDS – PAGE Electrophoresis and applications. Isoelectric focusing, Puised-field gel electrophoresis. **UNIT IV** 

**Isotopic tracer technique :** Radioactive & stable isotopes, rate of radioactive decay. Units of radioactivity. Measurement of radioactivity :- Ionization chambers, proportional counters, Geiger – Muller counter, Solid and liquid scintillation counters (basic principle, instrumentation and rechnique), Cerenkov radiation. Measurement of Stable isotopes : Falling drop method for deuterium measurement, Principles of tracer technique, advantages and limitations, applications of isotopes in biotechnology (distribution studies, metabolic studies, isotope dilution technique, metabolic studies, clinical applications, autoradiography).

#### $\mathbf{UNIT} - \mathbf{V}$

**Centrifugation :** Basic principles, concept of RCF, types of centrifuges (clinicale high speed and ultracentrifuges). Preparative centrifugation : Differential and density gradient centrifugation, applications (Isolation of cell components). Analytical centrifugation : Sedimentation coefficient, determination of molecular weight by sedimentation velocity and sedimentation equilibrium methods.

### **PRACTICALS : BIOPHYSICAL TECHNIQUES**

- 17. Spectrophotometric analysis of DNA denaturation.
- 18. Protein estimation by Biuret/Lowry method
- 19. Paper chromatography of amino acids/sugars.
- 20. TLC of sugars/amino acids- A demonstration by using of Photograph / reference diagram
- 21. Cellular fractionation and separation of cell organelles using centrifuge
- 22. Isolation of mitochondria and assay of marker enzyme
- 23. Estimation of Urea by diacetyle monoxide method
- 24. Estimation of Sugars by Folin Phenol method.
- 25. Preparation of standard buffers and determination of pH of a solution
- 26. Titration of a mixture of strong & weak acid
- 27. Paper electrophoresis of proteins- A demonstration by using of reference diagram
- 28. Gel electrophoresis of protein
- 29. SDS-PAGE of an oligomeric protein.
- 30. Calculation of mean, median, and mode (manual / computer aided)
- 31. Calculation of standard deviation and standard error (manual/ computer aided).
- 32. Biostatistical problem based on standard deviation.

### Note : - Mandatory to perform at least 8 practical's

### B.Sc., SEMESTER IV IMMUNOLOGY (w.e.f 2018-19)

### UNIT I

Immune system : Organs and cells of immune system immunity, innate immune mechanism Acquired immune mechanism, Antigen,

Humoral immunity, main pathways of complement system.

### UNIT II

**Antibody and Antigen :** Antibody structure and classes, Antibody diversity, types of Antigens Antigenecity (factors affecting antigenecity). Complement system.

### UNIT III

**Immunity :** Cell mediated immunity : TC mediated immunity, NK cell mediated immunity, ADCC brief description of cytokines and MHC (MHC types and diversity) Autoimmunity.

### UNIT IV

**Hypersensitivity and vaccination :** General features of hypersensitivity various types of hypersensitivity, Vaccination : Discovery, principles, significance, Types of Vaccines

#### UNIT V

**Immunological Techniques :** Antigen – antibody reactions : Precipition, agglutination, complement fixation, immunodiffusion, RIA,ELISA. Hybridoma technology : Monoclonal antibodies and their applications in immunodiagnosis.

### **PRACTICALS : IMMUNOLOGY**

- 19. Antigen antibody reaction
- 20. Determination of blood group
- 21. Widal test
- 22. Ouchterloney immunodiffusion-A demonstration
- 23. Radial immunodiffusion-A Demonstration

#### 24. ELISA – A demonstration

- 25. Coombs Test
- 26. Isolation of casein by isoelectric precipitation
- 27. Production of antibodies and their titration

Note : - Mandatory to perform at least 5 Practical's

B. Sc. III –Semester V

### 5191: MOLECULAR BIOLOGY (w.e.f 2017-18)

### Unit I:

**Genome Structure:** Watson and Crick model of DNA; Genome organization with specific reference to prokaryotic and eukaryotic genomes; Genome size. Concepts of Genetic Material, Gene, Chromosome and Genome. Histone Proteins. Experiments to prove DNA as genetic material (Griffith experiment, Hershey- Chase experiment)

### Unit II

**DNA Replication**:Enzymology of replication (DNA polymerase I, pol II and III, helicases, topoisomerases, single strand binding proteins, DNA melting proteins, primase. Proof of semi conservative replication, Replication origins, initiation, elongation, and termination. Rolling circle replication of DNA. Difference between Prokaryotic and Eukaryotic DNA Replication.

### Unit III

**Transcription: Central dogma of molecular biology.** Transcription in Prokaryotes: Enzymatic synthesis of RNA: Basi& features of transcription, classes of RNA molecules. E.coli RNA polymerases, types and structure (core enzyme and holo enzyme, sigma factor), transcription mechanism in prokaryotes-Promoter, initiation, elongation, proof reading and termination (Rho dependent and Rho independent). Transcriptions in Eukaryotes: Promoter, RNA polymerases of eukaryotes, Reverse transcription.

### Unit IV:

### **Genetic Code and Protein Synthesis**

Genetic code: Features of genetic code, Structure of mRNA Post-transcriptional modifications of eukaryotic hnRNA – Capping, methylation, polyadenylation and RNA splicing and splicing mechanisms. brief structure of tRNA, the adaptor hypothesis, attachment of amino acids to tRNA. Codon-anticodon interaction - the wobble hypothesis. Initiation, elongation, termination of protein. post translational modifications.(Post translational modifications).

#### Unit V

### Gene Expression and regulation

Regulation of gene expression; Regulation of transcription (regulatory elements and transcription factors).Clustered genes and the operon concepts - Negative and positive control of the Lac Operon, trp operon, Control of gene expression. Poly and Mono cistronic m-RNA. Translational regulation of gene expression in prokaryotic and eukaryotic organisms.

### PRACTICALS 5191P-MOLECULAR BIOLOGY

- 12. Isolation of plasmid DNA from bacteria
- 13. Isolation of genomic DNA from *E.coli*
- 14. Isolation of DNA from sheep liver
- 15. Isolation of DNA from plant material (Onion bulb)
- 16. Quantitative estimation and purity analysis of nucleic acids
- 17. Separation of DNA by Agarose gel Electrophoresis
- 18. Demonstrate Spliceosome
- 19. Demonstrate cloverleaf model of tRNA
- 20. Demonstrate different forms of DNA
- 21. Demonstrate Lac Operon.
- 22. Demonstrate blue white colonies

#### **Recommended Books:**

- 9. Molecular Biology of the Cell B. Alberts, D. Bray, J. Lewis, M. Raff, K. Roberts and J.D.Watson (Garland Publishing, New York and London)
- 10. Molecular Biology A Comprehensive Introduction to Prokaryotes and Eukaryotes D. Freifelder (Jones and Bartlett, USA)
- 11. Cell and Molecular Biology by Robertis and Roberties (Publ: Waverly)
- 12. Recombinant DNA : A Short Course J.D. Watson, J. Tooze and D.T.Kurtz (Scientific American Book, W.A.Preemon).
- 13. Modern Genetics (2<sup>nd</sup> Edition, 1984) A.J. Ayala and W. Castra (Goom Helns, London).
- 14. Text book of Biotechnology by HK Das (Wiley Publication)
- 15. Genes VIII. (1997) Benjamin Lewin (Oxford University Press).
- 16. Molecular Cloning: A Laboratory manual, J. Sambrook, E.F frisch and T. Maniatis, Old Spring Harbor Laboratory Press New York, 2000.

#### B. Sc. III – Semester V 5192: RECOMBINANT DNA TECHNOLOGY (Elective Theory) (w.e.f 2018-19)

#### Unit I:

**Tools of rDNA Technology-** Restriction and Modification enzymes. Classification of restriction endo nucleases (Type-I, Type-II and Type-III). Enzymes used in molecular cloning; Polymerases, ligases, phosphatases, kinases, nucleases and terminal transferase. Cloning vehicles - Plasmid, Bacteriophage, cosmids, shuttle vectors, expression vectors, Ti derived Vectors, artificial chromosomes (BAC, YAC).

#### Unit II

**Cutting and joining DNA** (cohesive end ligation, methods of blunt end ligation). Use of linkers, adaptors and homopolymer tails. Selection of transformed cells. Screening methods (Genetic marker and blue white screening). Construction of genomic and cDNA libraries. Advantages of cDNA libraries.

#### Unit III:

**Techniques of rDNA Technology:** Polymerase chain reaction technique (Components in PCR and PCR conditions) and Reverse transcription-PCR. Principles involved in blotting techniques-Southern, Northern, and Western blotting Methods of gene sequencing – Maxam - Gilberts and Sanger's dideoxy chain termination methods.

#### Unit IV:

**Methods of gene transfer**-Transformation, Electroporation, microinjection, microprojectile bombardment (gene gun method) and Plant transformation techniques *–Agrobacterium tumefaciens* mediated, Particle bombardment, electroporation and viral mediated. Transfection.

#### Unit V:

**Applications** of recombinant DNA technology in Agriculture (Transgenic Plants) Medicine (production of Insulin, Growth harmone, Tissue plasmogen activator and HBsAg vaccine. Principal and applications of DNA fingerprinting technique.

Application of plant transformation (Herbicide resistance-Phosphoinothricin, glyphosate, Insect resistance – transgenic Bt crops).

### PRACTICALS 5192P: rDNA TECHNOLOGY (Elective Lab)

- 1. Isolation of plasmid DNA.
- 2. Restriction digestion of DNA and its electrophoretic separation.
- 3. Ligation of Restriction digested DNA
- 4. Transformation in Bacteria using plasmid DNA
- 5. Demonstration of PCR
- 6. primer designing
- 7. Activity of DNase and RNase on DNA and RNA (Demo)

8. Study of vectors PBR, PUC

#### **Recommended Books:**

- 9. Molecular Cloning: A Laboratory manual, J. Sambrook, E.Ffrisch and T. Maniatis, Old Spring Harbor Laboratory Press New York, 2000
- 10. DNA Cloning : a Practical Approach, DM Glover and BD Hames, IRL Press
- 11. Methods in Enzymology Vol. 152, Guide to Molecular Cloning Techniques, SL Berger and AP Kimmel, Academic Press, Inc San Diego, 1998
- 12. DNA Science. A first Course in Recombinant Technology, DA Mickloss and GA Freyer, Cold Spring Harbor Laboratory Press, New York 1990
- 13. Milestones in Biotechnology. Classic papers on Genetic Engineering. JA Davies and WS Reznikoff, Butterworth-Heinemann, Boston, 1992
- 14. Concepts of Biotechnology D. Balasubramanian.
- 15. Principles of Gene Manipulation by Old and Primrose, Blackwell
- 16. Gene cloning by T.A. Brown

### 5193: GENETICS (Elective Theory) (w.e.f 2017-18)

### UNIT I

**Mendels Laws and Inheritance:** Mendel experiments, Mendel Laws and deviations: incomplete dominance and Co dominance, Back cross, Test cross Penetration and pleiotropism, Recessive and Dominant epistatic gene interactions. Concept of multiple alleles

#### UNIT II

**Genes and their variations:** Structure of gene, gene and environment, gene copies of prokaryotic and Eukaryotic chromosomes. Eukaryotic chromosome organization, histone proteins. Karyotype, special chromosomes – Lampbrush and polytene

#### Unit III:

**Gene mutations:** Mutagenesis – Spontaneous and induced (Chemical and physical) mutations; Natural and induction of mutations, point mutations, frameshift mutations, auxotrophic conditional and suppressor mutations. Chromosomal aberrations.

#### **UNIT IV:**

**DNA Damage and DNA Repair**: Light induced repair, Excision repair and mismatch repair, Post replication repair, Rec gene and its role in DNA repair, SOS repair and SOS response

#### Unit V:

**Transposable elements:** Structure and Molecular basis of AC-DS transposition in maize, "P" element of Drosophila and hybrid dysgenesis, Yeast "T<sub>7</sub>" elements, Retroposans

#### **PRACTICALS 5193P: GENETICS (Elective Lab)**

- 10. Study of different phases of mitosis in onion root tips and meiosis in *Allium cepa* flower buds.
- 11. Karyotyping in Allium or Drosophila.
- 12. Determination of multiple allele frequencies of leaf scars in Trifolium.
- 13. Problems and assignments in Mendilian genetics Monohybrid, dyhybrid, Testcross and backcross
- 14. Induction of chromosomal aberrations by chemical mutagenesis in Allium (or any plant).
- 15. Isolation of auxotrophic mutants (plants or insects).
- 16. Repair of DNA by Photo activation of Photolyase in bacteria.
- 17. Mutation of bacteria by UV.
- 18. Demo on Chemical induced mutation in bacteria

#### **Recommended Books**

- 8. Molecular Biology. Freifelder, D. 1990. Narosa Publication House, New Delhi
- 9. Principles of Genetics. Gardner, E.J and D.P. Snustad. 1996. John Willey, New York.
- 10. Genetics. Sambamurthy, A.V.S.S. 1999. Narosa Publishing House, New Delhi.
- 11. Principles of Genetics. Sinnot, E.W., L.C Dunn and T. Dobzhansky 1958. McGraw Hill, New York.
- 12. Theory and Problems in Genetics. Stansfield, W.D. 1991. McGraw Hill, New York.
- 13. Genetics. Strickberger, M.W. 1996. 3<sup>rd</sup> Edn. McMillan, New York.
- 14. Genetics A Conceptual Approach. Pierce, B. A.2006. 2<sup>nd</sup> Edn. W.H.Freeman and Company, New York.

#### B. Sc. III – Semester VI 619EL01: PLANT AND ANIMAL BIOTECHNOLOGY (w.e.f 2017-18)

### UNIT I:

**Cell and tissue culture:** Histological perspective of Plant tissue culture. Basic requirement for tissue culture laboratory. Totipotency, Tissue culture media (composition and MS media preparation). Aseptic techniques/sterilization methods used in plant tissue culture. Initiation and maintenance of callus and suspension cultures.

#### UNIT II:

**Applications of Plant Tissue culture**, meristem culture, clonal propogation, production of haploids (Anther culture), somatic embryogenesis, protoplast culture and somatic hybridization-Cybrids, Hybrids. Somoclonal variation-sources and its applications. Cryopreservation.

Production of secondary metabolites –Classication isolation, characharization and biosynthetic pathway of Secondary metabolites.

#### **UNIT III:**

**Tools and techniques of animal cell and tissue culture**: Culture media, growth factors, laboratory facilities. CO2 incubator, Cell count-haemocytometer, coulter method, MTT based Assay. Characteristics of cells in culture: Contact inhibition, anchorage dependence, cell-cell communication etc.; Cell senescence; cell and tissue response to trophic factors. Primary culture, immortal cells, cell lines. Maintenance of cell lines in the laboratory-subculture, trypsinisation and cryopreservation.

### **UNIT IV:**

**rDNA products:** Brief idea about recombinant DNA products in medicine (insulin, somatostatin, vaccines), Concept of Gene therapy, Production of recombinant vaccines – hepatitis. Concept of transgenic animals. In vitro fertilization and embryo transfer in humans and farm animals. Cloning-Concepts of nuclear transfer and creation of Dolly. Stem cells-types and applications.

#### UNIT V:

**IPR:** Intellectual property rights. Types and importance of IPRs (Copy rights. Patents, GIs...) International and national organizations, agencies and treaties. Regulation of GMO/transgenics. ethical aspects of Biotechnology.

#### PRACTICALS: 619EL01PLANT AND ANIMALBIOTECHNOLOGY

- **11.** Preparation of MS medium
- **12.** Sterilization methods
- **13.** Callus culture
- **14.** Demonstration of Suspension Culture
- **15.** Determination of seed viability by TTC
- **16.** Establishing primary cell culture of chicken embryo fibroblasts.
- **17.** Animal tissue culture maintenance of established cell lines.
- **18.** Cell count by Haemocytometer.
- **19.** Microphotography Cell line photographs.
- **20.** Synthetic seeds

#### **Recommended Books**

- 11. Culture of Animal Cells A Manual of Basic Technique. Freshney, R .I. 2006. 5<sup>th</sup> Edn. John Wiley and Sons, USA.
- 12. Cell Culture. Yadav, P.R and R. Tyagi. 2005. Discoery Publishing House, New Delhi.
- 13. Principles of Cloning. Jose Cibelli, Robert P. Lanza. Keith H.S. Campbell. 2002. Academic Press, London.
- 14. Animal Cell Technology Principles and practices. Butter, M. 1987. Oxford Press, USA.
- 15. Elements of Biotechnology. Gupta, P.K. 1996. Rastogi and Company, Meerut.
- 16. Plant Tissue Culture: Theory and Practice, a Revised Edition. Bhojwani, S.S and M.K.Razdan.2004.Panima Publishing Corporation, New Delhi.
- 17. Biotechnology-Fundamentals and applications. Purohit, S. 2000. 3<sup>rd</sup> Edn. Agrobios, New Delhi.
- 18. An Introduction to Plant Tissue Culture. Razdan, M.K. 1995. Oxford and IBH Publishing Company, New Delhi.
- 19. Plant Tissue Culture and Molecular Biology, Applications and Prospects. Srivastava, P.S. 1998. Narosa Publishing House, New Delhi.
- 20. Intellectural Property rights-Deborah, E. Bouchoux, Cengage Learning

#### 619EL02- ENVIRONMENTAL BIOTECHNOLOGY (Elective theory)

## (w.e.f 2017-18)

#### Unit I

**Principles of Ecology,** Water and terrestrial ecosystems, Bio-geo chemical cycles - Carbon, Nitrogen cycles. Role of microbes in bio-geochemical cycles.

#### Unit II

**Inorganic and Organic pollutants** of air, land and water; maintenance of standards, Environmental monitoring. Detection, treatment and prevention of pollution. Biological indicators

#### Unit III

**Biocides,** Four stage alternatives, Refuse disposal - Treatment methods, effluent from pharmaceuticals, fertilizers, pulp and paper industry.

#### Unit IV

Waste water management - Aerobic and anaerobic treatment, primary, secondary and tertiary treatment of municipal wastes, Solid waste management.

#### Unit V

**Bioremediation**, Biodegradation of recalcitrant compounds and the role of genetically engineered microbes and genetically modified organisms in the environmental management.

# **PRACTICALS : ENVIRONMENTAL BIOTECHNOLOGY (Elective Lab)**

- 1. Detection of coliforms for determination of the purity of potable water.
- 2. Determination of total dissolved solids of water
- 3. Determination of Hardness and alkalinity of water sample.
- 4. Determination of dissolved oxygen concentration of water sample
- 5. Determination of biological oxygen demand of sewage sample
- 6. Determination of chemical oxygen demand (COD) of sewage sample.
- 7. Isolation of xenobiotic degrading bacteria by selective enrichment technique
- 8. Estimation of heavy metals in water/soil
- 9. Estimation of nitrate in drinking water.
- 10. Preparation and formulation of microbial biopesticide (bacteria, fungi and viruses)
- 11. In vitro evaluation of medicinal plants against pathogenic microbes.
- 12. Effect of mycorrhizal fungi on growth promotion of plants.
- 13. Production of microbial fertilizers (Rhizobium, Azotobacter and AMF).

#### **Recommended Books**

1. Ecology and Environment P.D.Sharma

2. Brock Biology of Microorganisms (9th edition) by Madigan, Martinko & Parker.2000. Prentice Hall.

3. Environmental Microbiology by Mitchell. 2009. John Wiley and Sons.

4. Environmental Microbiology by Grant & Long. 1981. Wiley.

5. Environmental Microbiology: A laboratory manual by Peppler, Gerba & Brendecke.1995. Academic Press.

## 619EL03-Industrial Biotechnology (Elective theory) (w.e.f 2017-18)

# Unit I:

**Isolation, Screening**, Preservation and Improvement of Industrially Important Microorganisms. Synthetic and Natural Medium, Precursors, Antifoams, Sterilization Methods and Inoculum Preparation.

# Unit II:

**Definition of bioreactor**, Basic principles of bioreactor. Classification of bioreactors. Analysis of batch, continuous, fed batch and semi-continuous bioreactors.

# Unit III:

**Ethanol Production** by Fermentation using Molasses, Starchy Substances. Production of Alcoholic Beverages like Beer and Wine. Production of Citric Acid by Submerged and Solid State Fermentations.

# Unit IV:

**Sources of Industrial Enzymes**, Production of Microbial Enzymes like Amylase and protease. Backer's Yeast and SCP Production. Production of Antibiotics : Penicillin .

# Unit V:

**Biotechnology Products-** Production of recombinant proteins having therapeutic and diagnostic applications(Insulin, Growth Harmone, Recombinant vaccines, Monoclonal Antibody).

# PRACTICALS Industrial Biotechnology (Elective Lab)

- 11. Isolation of bacteria / fungi from soil by serial dilution technique
- 12. Estimation of amylase from seeds.
- 13. Production of  $\alpha$  amylase from *Bacillus Spp*. by shake flask culture.
- 14. Production of alcohol or wine using different substrates.
- 15. Estimation of alcohol by titrimetry.
- 16. Estimation of alcohol by colorimetric method .
- 17. Production of citric acid.
- 18. Citric acid production by submerged fermentation.
- 19. Estimation of citric acid by titrimetry.
- 20. Demonstration of Fermentor and bioreactor

# **Recommended Books**

- 1. Biogas technology by b.T.Nijaguna
- 2. Food microbiology by m.R.Adams and M.O.Mass
- 3. Industrial microbiology by A.H.Patel
- 4. Industrial microbiology, A text book W.Clark
- 5. Industrial Microbiology by L.E.Casida
- 6. Introduction to Biotechnology by P.K.Gupta
- 7. Text book of Biotechnology by H.K.Das (Wily Publications)

- 8. Bioprocess engineering by Shuler (Pearson Education
  - 9. Biotechnology: A Text Book of Industrial microbiology

2nd edition by Crueger. & Crueger. 1990. Sinauer Associates.

- 10.Microbial Biotechnology: Fundamentals of Applied Microbiology (2nd edition) by Glazer & Nikaido. 2007. Cambridge University Press.
- 11. Prescott & Dunn's Industrial Microbiology 4th edition Editor Reed. 1982. AVI Pub. Co.,

# **CLUSTERS**

#### 619CLA1 - TISSUE CULTURE (w.e.f 2018-19)

# <u>UNIT - I</u>

Animal tissue culture – Introduction to culture medium– natural and artificial medium preparation and sterilization of medium, , Disagrigation of and subculturing.

# <u>UNIT - II</u>

Cell cultures, suspension cultures, maintenance of cell lines, organ culture – techniques, advantages, applications,

# <u>UNIT - III</u>

Plant tissue culture, Introduction to culture room, Culture vessels, their working. Sterilization of nutrient medium, callus cultures, subculture.

# UNIT - IV

Somatic embryogenesis, meristem culture, somatic hybridization, micro propagation, somatic hybrids & cybrids.

# UNIT - V

Invitro pollination, parthenogenisis, applications of tissue culture both in agriculture and animal husbandry. Embryo culture technique.

# 619CLAP -Practical -TISSUE CULTURE

- 1. Preparation of plant tissue culture medium
- 2. Initiation of callus from any one selected plant species
- 3. Micro propagation of plants
- 4. Cell disaggregation and cell counting
- 5. Study of gene transfer techniques (Transformation)
- 6. Preparation of animal tissue culture medium

# III B. Sc. (SEMESTER - VI) 619CLA2 - INDUSTRIAL BIOTECHNOLOGY

#### (w.e.f 2018-19)

# <u>UNIT - I</u>

Introduction to industrial biotechnology. Primary and secondary metabolic products of microorganisms. Screening, isolation and preservation of industrially important microorganisms, <u>UNIT - II</u>

Fermentation technology: principles of fermentation, fermentation medium, types of fermenters, fermentation conditions and sterilization methods. Factors involved in fermenter design. Fermenter configurations. Principle operating characteristics of fermenters.

# <u>UNIT - III</u>

Factors effecting fermentation. Fermented products like – fermentation of molasses, production of alcoholic beverages like beer, wine. Commercial production of fuels and chemicals by microbial fermentations,

#### <u>Unit - IV</u>

penicillin and streptomycin. Downstream process techniques. Production and application of recombinant proteins – Insulin, Growth harmone, Recombinant vaccines Monoclonal Antibodies and interferons.

# <u>UNIT - V</u>

Fermentative production of foods and dairy products. Production of vitamins – B12, B2 & C. Good manufacturing practices. Bio safety issues. Intellectual property rights and patenting issues.

# Practical 619CLA 2P - : INDUSTRIAL BIOTECHNOLOGY

- 1. Isolation of Amylase producing organisms from soil.
- 2. Production of Amylase from bacteria / fungi
- 3. Assay of Amylase activity.
- 4. Production of alcohol (or) wine from grapes
- 5. Estimation of alcohol by colorimetric method
- 6. Production of citric acid from bacteria / fungi
- 7. Estimation of citric acid by titrimetric method.

#### 619CLA 3 - ENVIRONMENTAL BIOTECHNOLOGY (w.e.f 2018-19)

## **Unit - I : Environmental Concepts**

Introduction to environmental biotechnology and ecology. Environmental Concepts – Atmosphere, Ozone layer, Hydrosphere, Lithosphere

#### **Unit - II : Ecosystem and Ecology**

Ecosystem and community. Factors effecting Ecosystem. Ecological pyramids. Endangered and Extinct species. Renewable and non renewable energy resources.

#### **Unit - III : Pollution**

Air, Water, Soil, types of pollutants i.e pesticides, insecticides, heavy metals, toxins, radiation, hazardous waste and biodegradation with microbes. Biopesticides and biofertlizers.

#### **Unit – IV : Protection and Restoration of Ecosystem**

Major protection acts in India (Forest conservation act 1980, wild life protection act 1972), Bioremediation, Phytoremediation, waste water treatment, solid waste management. Microbial ore leaching.

#### **Unit - V : Environmental Protection**

Bio gas production, Global warming, Green house effect, Rain water harvesting, Energy conservation, acid rains, ozone depletion, deforestation and its effects, reforestation.

# 619CLA3P -Practical ENVIRONMENTAL BIOTECHNOLOGY

- 1. Estimation of BOD of water
- 2. Estimation of COD of polluted water
- 3. Collection of endangered and extinct species data
- 4. Study production of hydrogen (or) biogas using Cow / cattle dung
- 5. Most Probable Number for water

# MODEL QUESTION PAPER B.Sc., Three year Degree Examinations-2018 III Semester Biotechnology 3191Biophysical Techniques (W.E.F 2018-19)

### Time -3hrs

Max Marks : 60

#### Section – A

# Answer any FIVE Questions

5X4 =20M

# (Each question carries 4 Marks )

- 1. Spectroflorometry applications
- 2. Beer Lambert's Law
- 3. Gel filtration Chromatography
- 4. TLC
- 5. Factors effecting Electrophoretic mobility
- 6. Types of gels
- 7. Liquid Scintillation counter
- 8. Autoradiography
- 9. Ultra centrifuges
- 10. Density gradient centrifugation

# Section – B Answer ALL of the following questions (Each question carries 10 Marks )

11. (A)Write about Principle, Instrumentation and applications of UV – Visible spectrophotometry

# (Or)

(B) Write about Principle, Instrumentation and applications of Spectro Flurometry

12.(A) Write about column chromatography

# (Or)

(B)Describe the principle, instrumentation and applications of HPLC

13.(A) Explain about Principle ,procedure and applications of Isoelectric focussing

(Or)

(B) Write complete details of SDS-PAGE

14.(A).Describe the importance of GM Counter for measurement of radioactivity

# (Or)

(B) Define about radio isotopes and write different applications of radioisotopes

15.(A) Write about different types of centrifuges

#### (Or)

(B) How you isolate the cell organelles by using centrifugation technique

### 2019-2020

# B.Sc., I year –Semester-I MICROBIOLOGY AND CELL BIOLOGY (w.e.f 2017-2018)

# UNIT I

# HISTORY, DEVELOPMENT AND MICROSCOPY

History and development of microbiology: contributions of Louis Pasteur, Robert Koch and Edward Jenner. Microscopy: Compound microscopy: Numerical aperture and its importance, resolving power, oil immersion objectives and their significance, principles and applications of dark field, phase contrast, fluorescent microscopy. Electron microscopy: Principle, ray diagram and applications, TEM and SEM, comparison between optical and electron microscopy.

Stains and staining procedures: Acidic, basic and neutral stains, Gram staining, Acid fast staining, Flagella staining, Endospore staining.

# UNIT-II BACTERIA & VIRUSES A.BACTERIA

Bacterial morphology and subcellular structures, general morphology of bacteria, shapes and sizes, generalized diagram of typical bacterial cell.Slime layer and capsule, difference between the structure, function and the position of the two structures. Cell wall of gram +ve and Gram -ve cells, Prokrayotic classification.

General account of flagella and fimbriae.Chromatin material, plasmids; definition and kind of plasmids (conjugative and non-conjugative) F, R, and Col plasmids.

A brief idea Bergey's manual. Morphology of archaea, archaeal cell membrane (differences between bacterial and archaeal cell membrane), other cell structures, concept of the three distinct archaea groups.

#### B.VIRUSES:

General Characters of viruses, difference between virus and typical microbial cell structure ,diferent shapes and symmetries with one example of each type, classification of viruses on the basis of nucleic acids, phage and animal cell viruses, example of each and their importance. Brief idea of lytic cycle and lysogeny.

# UNIT III

# **MICROBIAL NUTRITION:**

Types of Microbial nutrition.

Basic nutritional requirements: Basic idea of such nutrients as water, carbon, nitrogen, sulfur and vitamins etc., natural and synthetic media, nutritional classification of bacteria. Selective and Differential media, Enriched media, Enrichment media.

#### UNIT IV: MICROBIAL GROWTH AND CONTROL:

#### **GROWTH:**

Growth rate and generation time, details of growth curve and its various phases. Concepts of synchronous cultures, continuous and batch cultures (chemostat and turbidostat). **Measurement of growth.** 

Physical conditions required for growth : Temperature (Classification of microorganisms on the basis of temperature requirements), pH etc. Pure cultures and cultural characteristics. Maintenance of pure culture.

**Microbial Control:** Terminologies - Sterilization, disinfection, antiseptic, sanitization, germicide, microbistasis, preservative and antimicrobial agents.

Mechanism of cell injury: Damage to cell wall, cell membrane, denaturation of proteins, inhibition of protein synthesis, transcription, replication, other metabolic reactions and change in supercoiling of DNA.

**Physical control:** Temperature (moist heat, autoclave, dry heat, hot air oven and incinerators), dessication, surface tension, osmotic pressure, radiation, UV light, electricity, ultrasonic sound waves, filtration.Chemical control: Antiseptics and disinfectants (halogens, alcohol, gaseous sterilization. Concept of biological control. Air filtration.

#### UNIT V

#### **CELL BIOLOGY**

**Eukaryotic Cell** – Structure and function of the following nucleus , nuclear membrane , nucleoplasm, nucleolus, golgi complex, Mitochandria, Chloroplast, endoplasmic reticulum, lysosomes, peroxisomes, glyoxisomes and vacuoles.

#### Plant Cell wall :

Cytoskeleton (Micro and Macro filaments, microtubules) and cell locomotion. Mitosis and meiosis. Brief idea of cell cycle.Muscle and nerve cell structure, synaptic transmission and neuromuscular junctions.

# B.Sc., - I year SEMESTER-I Practicals BIOTECHNOLOGY MICROBIOLOGY & CELL BIOLOGY

- 77. Demonstration, Use and care of microbiological equipments
- 78. Preparation of media, sterilization and isolation of bacteria
- 79. Isolation of bacteria from water, soil and vegetables
- 80. Demonstration of motility of bacteria
- 81. Simple staining of Bacteria
- 82. Gram's Staining of bacteria
- 83. Acid fast Staining of bacteria- Demonstration
- 84. Endospore staining
- 85. Demonstration of Starch hydrolysis by bacterial cultures
- 86. Growth of fecal coliforms on selective media
- 87. Isolation of pure culture by Pour plate method
- 88. Isolation of pure culture by Streak plate method
- 89. Anaerobic cultivation of microorganisms
- 90. Cultivation of yeast/Mucor/Rhizopus
- 91. Antibiotic Sensitivity test

- 92. Oligodynamic action of Metals-Demonstration
- 93. To study Germicidal effect of UV-Light on bacterial Growth
- 94. Stages of Mitosis-Demonstration
- 95. Stages of Meiosis (Permanent slides)

# Note: - Mandatory to perform at least ten practicals.

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#### B. Sc., I – Semester II

### MACROMOLEULES, ENZYMOLOGY AND BIOENERGETICS (w.e.f 2017-18)

## UNIT I

#### NUCLEIC ACIDS AND CHROMOSOMES

Chemical structure and base composition of nucleic acids, Chargaff's rules, Watson Crick Model (B-DNA), deviations from Watson-Crick model, other forms of DNA (Aand Z-DNA), forces stabilizing nucleic acid structures, (hydrogen bonds and hydrophobic associations, base stacking). Maxam and Gilbert DNA sequencing and Sanger's method.

Concept of prokaryotic genes and eukaryotic genes: Definition of a gene, concept of split genes, introns, exons, spacers, C-value and C-value paradox, basic idea of Cot curves.

Chromatin structure: Nucleosome structure (10 nm fibre, experiments leading to discovery of nucleosomal structure, types of histones, arrangement of histones in the octamer, H1 histone and its role, role and length of linker DNA), 300 nm fibers (arrangement of nucleosome in a helical structure), domain and loop structure (further compacting of 300 nm fibre, role of scaffolding proteins). Role of telomere and centromere, telomeric and centromeric repeat sequences.

#### UNIT II

#### **Amino acids and Proteins**

Amino acids: Structure of amino acids occurring in proteins, classification of amino acids (pH based, polarity based and nutrition based), Physico-chemical properties of amino acids (solubility, boiling and melting points, reactions like Edman's, Sanger's, Dansyl chloride, ninhydrin). Titration curves of neutral, basic and acidic amino acids.

Primary structure of proteins: Determination of primary structure (end group analysis, cleavage of disulfide bonds, amino acid composition, use of endopepticlase specificity, sequence determination, assignment of disulfide position).

Secondary structure of proteins: The -helix, -structures (parallel, antiparallel, mixed, -turn).

Tertiary structure of proteins: Forces that stabilize the structure (electrostatic forces, hydrogen and disulfide bonds, hydrophobic associations), myoglobin as an example of tertiary structure, concept of domains, protein denaturation.

Quaternary structure of proteins: Forces stabilizing quaternary structure, advantages of oligomeric proteins.

## UNIT-III

#### CARBOHYDRATES

Definition, classification, nomenclature of carbohydrates, structures of monosaccharides, disaccharides and polysaccharides (structures of starch and glycogen as examples of homopolysaccharides). Concept and examples of heteropolysaccharides.

**Lipids** - Types of lipids, structures of saturated and unsaturated fatty acids, triglycerides, phospholipids, plasmalogens, gangliosides and sphingolipids. Terpenoids and isoprenoids - definition and representative structures, steroids. Concept of acid value, saponification value and iodine value. Chemistry of Porphyrines, Heme, Cytochromes, and Chlorophylls

UNIT -V ENZYMES : Terminology: Active site , allosteric site, Holo enzyme, apoenzyme, co-enzyme

substrate, inhibitor, activator, modulator etc.Classification and nomenclature. Concept of isoenzymes (example Lactate Dehydrogenase) and multienzymes (example pyruvate dehydrogenase) Substrate Specificity (bond specificity, group specificity, absolute specificity, stereo-specificity, proof-reading mechanism), lock and key and induced fit models.

Concept of Allosteric enzymes( Brief idea of ATPase as an example, mechanisms of calalysis

: Acid base, covalent and metal ion catalysis.

Assay of Enzymes: Concept of activity, specific activity, turnover number, units of enzyme activity (katal, international unit), spectrophotometric methods of assay of enzymes (simple and coupled assay), very brief idea of other methods.

Enzyme kinetics: Michaelis-Menten equation, effect of substrate concentration, effect of enzyme concentration, effect of p H and temperature, temperature quotient, single reciprocal( Eadie-Hoffstee equation) and double reciprocal plots( Lineweaver-Burke plots), enzyme inhibition kinetics (reversible inhibition types – competitive, uncompetitive and non-competitive), brief idea of irreversible inhibition.

#### UNIT V

**Bioenergetics**: Concept of free energy, Entropy, Enthalpy & Redox Potential. Concept of high energy bonds as related to the structure of ATP, Phosphoenolpyruvate, Creatine phosphate etc.

Glycolosis (Pathway, entry of other monosaccharides and di saccharides, regulations,

inhibitors) Gluconeogenesis : Bypass reactions.

#### B.Sc., - I year SEMESTER-II Practicals BIOTECHNOLOGY MACRO MOLECULES & ENZYMOLOGY

- 77. Formal titration of Glycine- Demonstration
- 78. Quantitative estimation of proteins by Biuret method
- 79. Demonstration of Albumin and Ag ratio in Serum
- 80. Estimation of DNA by DPA method
- 81. Estimation of RNA by Orcinol method
- 82. Quantitative estimation of Amino acids using Ninhydrin reaction
- 83. Quantitative analysis of Sugars and proteins
- 84. Quantitative estimation of Sugars ( Dinitro salisilic acid method ) Demonstration
- 85. Estimation of Glucose by Benedicts Quantitative method
- 86. Quantitative estimation of proteins by Lowry's method
- 87. Extraction and quantification of total Lipids
- 88. Determination of Saponification value of fats
- 89. Determination of Acid value of fats
- 90. Isolation of Urease and demonstration of its activity
- 91. Assay of protease activity
- 92. Preparation of starch from potato and its hydrolysis by Salivary amylase
- 93. Assay of Alkaline phosphatase
- 94. Immobilization of enzymes / cells by Entrapment in Alginate gel
- 95. Effect of temperature / pH on enzyme activity

Note: - Mandatory to perform at least ten practicals.

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#### **B.Sc., SEMESTER III**

#### BIOPHYSICAL TECHNIQUES (w.e.f 2018-19)

# UNIT – I

**Spectrophotometry :** Spectrum of light, absorption of electromagnetic radiations, Beer's law – derivation and deviations, extinction coefficient. Instrumentation of UV and visible spectrophotometry, Double deam spectrometer ; dual – wavelength spectrometer, Applications of UV and visible spectrotometry. Spectrofluorometry : principle , instrumentation and application. Mass spectrometry

#### UNIT II :

**Chromatography :** Partition principle, coefficient, nature of partition forces, brief account of paper chromatography. Thin layer chromatography and column chromatography. Gel filtration : Concept of distribution coefficient, types of gels and glass beads, applications. Ion-exchange chromatography : Principle, types of resins, choice of buffers, applications including amino acid analyzer. Affinity chromatography : Principle, selection of ligand, brief idea of ligand attachment, specific and non-specific elution, applications. HPLC

#### UNIT III

**Electrophoresis :** Migration of ions in electric fields, Factors affecting electrophoretic mobility. Paper electrophoresis, Gel electrophoresis :- Types of gels, Solubilizers, Procedure, Column & slad gels Detection, Recovery & Estimation of macromolecules. SDS – PAGE Electrophoresis and applications. Isoelectric focusing, Puised-field gel electrophoresis. **UNIT IV** 

**Isotopic tracer technique :** Radioactive & stable isotopes, rate of radioactive decay. Units of radioactivity. Measurement of radioactivity :- Ionization chambers, proportional counters, Geiger – Muller counter, Solid and liquid scintillation counters (basic principle, instrumentation and rechnique), Cerenkov radiation. Measurement of Stable isotopes : Falling drop method for deuterium measurement, Principles of tracer technique, advantages and limitations, applications of isotopes in biotechnology (distribution studies, metabolic studies, isotope dilution technique, metabolic studies, clinical applications, autoradiography).

### $\mathbf{UNIT} - \mathbf{V}$

**Centrifugation :** Basic principles, concept of RCF, types of centrifuges (clinicale high speed and ultracentrifuges). Preparative centrifugation : Differential and density gradient centrifugation, applications (Isolation of cell components). Analytical centrifugation : Sedimentation coefficient, determination of molecular weight by sedimentation velocity and sedimentation equilibrium methods.

# **PRACTICALS : BIOPHYSICAL TECHNIQUES**

- 33. Spectrophotometric analysis of DNA denaturation.
- 34. Protein estimation by Biuret/Lowry method
- 35. Paper chromatography of amino acids/sugars.
- 36. TLC of sugars/amino acids- A demonstration by using of Photograph / reference diagram
- 37. Cellular fractionation and separation of cell organelles using centrifuge
- 38. Isolation of mitochondria and assay of marker enzyme
- 39. Estimation of Urea by diacetyle monoxide method
- 40. Estimation of Sugars by Folin Phenol method.
- 41. Preparation of standard buffers and determination of pH of a solution
- 42. Titration of a mixture of strong & weak acid
- 43. Paper electrophoresis of proteins- A demonstration by using of reference diagram
- 44. Gel electrophoresis of protein
- 45. SDS-PAGE of an oligomeric protein.
- 46. Calculation of mean, median, and mode (manual / computer aided)
- 47. Calculation of standard deviation and standard error (manual/ computer aided).
- 48. Biostatistical problem based on standard deviation.

#### Note : - Mandatory to perform at least 8 practical's

# B.Sc., SEMESTER IV IMMUNOLOGY (w.e.f 2018-19)

# UNIT I

Immune system : Organs and cells of immune system immunity, innate immune mechanism Acquired immune mechanism, Antigen,

Humoral immunity, main pathways of complement system.

# UNIT II

Antibody and Antigen : Antibody structure and classes, Antibody diversity, types of Antigens Antigenecity (factors affecting antigenecity). Complement system.

# UNIT III

**Immunity :** Cell mediated immunity : TC mediated immunity, NK cell mediated immunity, ADCC brief description of cytokines and MHC (MHC types and diversity) Autoimmunity.

# UNIT IV

**Hypersensitivity and vaccination :** General features of hypersensitivity various types of hypersensitivity, Vaccination : Discovery, principles, significance, Types of Vaccines **UNIT V** 

**Immunological Techniques :** Antigen – antibody reactions : Precipition, agglutination, complement fixation, immunodiffusion, RIA,ELISA. Hybridoma technology : Monoclonal antibodies and their applications in immunodiagnosis.

# **PRACTICALS : IMMUNOLOGY**

- 28. Antigen antibody reaction
- 29. Determination of blood group
- 30. Widal test
- 31. Ouchterloney immunodiffusion-A demonstration
- 32. Radial immunodiffusion-A Demonstration
- 33. ELISA A demonstration
- 34. Coombs Test
- 35. Isolation of casein by isoelectric precipitation
- 36. Production of antibodies and their titration

# Note : - Mandatory to perform at least 5 Practical'

#### B. Sc. III –Semester V 5191: MOLECULAR BIOLOGY (w.e.f 2019-20)

#### Unit I:

**Genome Structure:** Watson and Crick model of DNA; Genome organization with specific reference to prokaryotic and eukaryotic genomes; Genome size. Concepts of Genetic Material, Gene, Chromosome and Genome. Histone Proteins. Experiments to prove DNA as genetic material (Griffith experiment, Hershey- Chase experiment)

#### Unit II

**DNA Replication**:Enzymology of replication (DNA polymerase I, pol II and III, helicases, topoisomerases, single strand binding proteins, DNA melting proteins, primase. Proof of semi conservative replication, Replication origins, initiation, elongation, and termination. Rolling circle replication of DNA. Difference between Prokaryotic and Eukaryotic DNA Replication.

#### Unit III

**Transcription: Central dogma of molecular biology.** Transcription in Prokaryotes: Enzymatic synthesis of RNA: Basi& features of transcription, classes of RNA molecules. E.coli RNA polymerases, types and structure (core enzyme and holo enzyme, sigma factor), transcription mechanism in prokaryotes-Promoter, initiation, elongation, proof reading and termination (Rho dependent and Rho independent). Transcriptions in Eukaryotes: Promoter, RNA polymerases of eukaryotes, Reverse transcription.

#### Unit IV:

#### **Genetic Code and Protein Synthesis**

Genetic code: Features of genetic code, Structure of mRNA Post-transcriptional modifications of eukaryotic hnRNA – Capping, methylation, polyadenylation and RNA splicing and splicing mechanisms. brief structure of tRNA, the adaptor hypothesis, attachment of amino acids to tRNA. Codon-anticodon interaction - the wobble hypothesis. Initiation, elongation, termination of protein. post translational modifications.(Post translational modifications).

#### Unit V

#### Gene Expression and regulation

Regulation of gene expression; Regulation of transcription (regulatory elements and transcription factors).Clustered genes and the operon concepts - Negative and positive control of the Lac Operon, trp operon, Control of gene expression. Poly and Mono cistronic m-RNA. Translational regulation of gene expression in prokaryotic and eukaryotic organisms. Bioinformatics: Introduction ,Data Bases, Search Tools and Applications in Biotechnology

#### PRACTICALS 5191P-MOLECULAR BIOLOGY

- 23. Isolation of plasmid DNA from bacteria
- 24. Isolation of genomic DNA from E.coli
- 25. Isolation of DNA from sheep liver
- 26. Isolation of DNA from plant material (Onion bulb)
- 27. Quantitative estimation and purity analysis of nucleic acids
- 28. Separation of DNA by Agarose gel Electrophoresis
- 29. Demonstrate Spliceosome
- 30. Demonstrate cloverleaf model of tRNA
- 31. Demonstrate different forms of DNA
- 32. Demonstrate Lac Operon.
- 33. Demonstrate blue white colonies

#### **Recommended Books:**

- 17. Molecular Biology of the Cell B. Alberts, D. Bray, J. Lewis, M. Raff, K. Roberts and J.D.Watson (Garland Publishing, New York and London)
- Molecular Biology A Comprehensive Introduction to Prokaryotes and Eukaryotes D. Freifelder (Jones and Bartlett, USA)
- 19. Cell and Molecular Biology by Robertis and Roberties (Publ: Waverly)
- 20. Recombinant DNA : A Short Course J.D. Watson, J. Tooze and D.T.Kurtz (Scientific American Book, W.A.Preemon).
- 21. Modern Genetics (2<sup>nd</sup> Edition, 1984) A.J. Ayala and W. Castra (Goom Helns, London).
- 22. Text book of Biotechnology by HK Das (Wiley Publication)
- 23. Genes VIII. (1997) Benjamin Lewin (Oxford University Press).
- 24. Molecular Cloning: A Laboratory manual, J. Sambrook, E.F frisch and T. Maniatis, Old Spring Harbor Laboratory Press New York, 2000.

#### B. Sc. III – Semester V 5192: RECOMBINANT DNA TECHNOLOGY (Elective Theory) (w.e.f 2019-20)

#### Unit I:

**Tools of rDNA Technology-** Restriction and Modification enzymes. Classification of restriction endo nucleases (Type-I, Type-II and Type-III). Enzymes used in molecular cloning; Polymerases, ligases, phosphatases, kinases, nucleases and terminal transferase. Cloning vehicles - Plasmid, Bacteriophage, cosmids, shuttle vectors, expression vectors, Ti derived Vectors, artificial chromosomes (BAC, YAC).

#### Unit II

**Cutting and joining DNA** (cohesive end ligation, methods of blunt end ligation). Use of linkers, adaptors and homopolymer tails. Selection of transformed cells. Screening methods (Genetic marker and blue white screening). Construction of genomic and cDNA libraries. Advantages of cDNA libraries.

#### Unit III:

**Techniques of rDNA Technology:** Polymerase chain reaction technique (Components in PCR and PCR conditions) and Reverse transcription-PCR. Principles involved in blotting techniques-Southern, Northern, and Western blotting Methods of gene sequencing – Maxam - Gilberts and Sanger's dideoxy chain termination methods.

#### Unit IV:

**Methods of gene transfer**-Transformation, Electroporation, microinjection, microprojectile bombardment (gene gun method) and Plant transformation techniques *–Agrobacterium tumefaciens* mediated, Particle bombardment, electroporation and viral mediated. Transfection.

#### Unit V:

**Applications** of recombinant DNA technology in Agriculture (Transgenic Plants) Medicine (production of Insulin, Growth harmone, Tissue plasmogen activator and HBsAg vaccine. Principal and applications of DNA fingerprinting technique.

Application of plant transformation (Herbicide resistance-Phosphoinothricin, glyphosate, Insect resistance – transgenic Bt crops).

# PRACTICALS 5192P: rDNA TECHNOLOGY (Elective Lab)

- 1. Isolation of plasmid DNA.
- 2. Restriction digestion of DNA and its electrophoretic separation.
- 3. Ligation of Restriction digested DNA
- 4. Transformation in Bacteria using plasmid DNA
- 5. Demonstration of PCR

- 6. primer designing
- 7. Activity of DNase and RNase on DNA and RNA (Demo)
- 8. Study of vectors PBR, PUC

#### **Recommended Books:**

- 17. Molecular Cloning: A Laboratory manual, J. Sambrook, E.Ffrisch and T. Maniatis, Old Spring Harbor Laboratory Press New York, 2000
- 18. DNA Cloning : a Practical Approach, DM Glover and BD Hames, IRL Press
- 19. Methods in Enzymology Vol. 152, Guide to Molecular Cloning Techniques, SL Berger and AP Kimmel, Academic Press, Inc San Diego, 1998
- 20. DNA Science. A first Course in Recombinant Technology, DA Mickloss and GA Freyer, Cold Spring Harbor Laboratory Press, New York 1990
- 21. Milestones in Biotechnology. Classic papers on Genetic Engineering. JA Davies and WS Reznikoff, Butterworth-Heinemann, Boston, 1992
- 22. Concepts of Biotechnology D. Balasubramanian.
- 23. Principles of Gene Manipulation by Old and Primrose, Blackwell
- 24. Gene cloning by T.A. Brown

#### 5193: GENETICS (Elective Theory) (w.e.f 2019-20)

## UNIT I

**Mendels Laws and Inheritance:** Mendel experiments, Mendel Laws and deviations: incomplete dominance and Co dominance, Back cross, Test cross Penetration and pleiotropism, Recessive and Dominant epistatic gene interactions. Concept of multiple alleles

#### UNIT II

**Genes and their variations:** Structure of gene, gene and environment, gene copies of prokaryotic and Eukaryotic chromosomes. Eukaryotic chromosome organization, histone proteins. Karyotype, special chromosomes – Lampbrush and polytene

#### Unit III:

**Gene mutations:** Mutagenesis – Spontaneous and induced (Chemical and physical) mutations; Natural and induction of mutations, point mutations, frameshift mutations, auxotrophic conditional and suppressor mutations. Chromosomal aberrations.

#### **UNIT IV:**

**DNA Damage and DNA Repair**: Light induced repair, Excision repair and mismatch repair, Post replication repair, Rec gene and its role in DNA repair, SOS repair and SOS response

#### Unit V:

**Transposable elements:** Structure and Molecular basis of AC-DS transposition in maize, "P" element of Drosophila and hybrid dysgenesis, Yeast "T<sub>7</sub>" elements, Retroposans

# PRACTICALS 5193P: GENETICS (Elective Lab)

- 19. Study of different phases of mitosis in onion root tips and meiosis in *Allium cepa* flower buds.
- 20. Karyotyping in Allium or Drosophila.
- 21. Determination of multiple allele frequencies of leaf scars in Trifolium.
- 22. Problems and assignments in Mendilian genetics Monohybrid, dyhybrid, Testcross and backcross
- 23. Induction of chromosomal aberrations by chemical mutagenesis in Allium (or any plant).
- 24. Isolation of auxotrophic mutants (plants or insects).
- 25. Repair of DNA by Photo activation of Photolyase in bacteria.
- 26. Mutation of bacteria by UV.
- 27. Demo on Chemical induced mutation in bacteria

#### **Recommended Books**

- 15. Molecular Biology. Freifelder, D. 1990. Narosa Publication House, New Delhi
- 16. Principles of Genetics. Gardner, E.J and D.P. Snustad. 1996. John Willey, New York.
- 17. Genetics. Sambamurthy, A.V.S.S. 1999. Narosa Publishing House, New Delhi.
- 18. Principles of Genetics. Sinnot, E.W., L.C Dunn and T. Dobzhansky 1958. McGraw Hill, New York.
- 19. Theory and Problems in Genetics. Stansfield, W.D. 1991. McGraw Hill, New York.
- 20. Genetics. Strickberger, M.W. 1996. 3<sup>rd</sup> Edn. McMillan, New York.
- 21. Genetics A Conceptual Approach. Pierce, B. A.2006. 2<sup>nd</sup> Edn. W.H.Freeman and Company, New York.

#### B. Sc. III – Semester VI 619EL01: PLANT AND ANIMAL BIOTECHNOLOGY (w.e.f 2019-20)

# UNIT I:

**Cell and tissue culture:** Histological perspective of Plant tissue culture. Basic requirement for tissue culture laboratory. Totipotency, Tissue culture media (composition and MS media preparation). Aseptic techniques/sterilization methods used in plant tissue culture. Initiation and maintenance of callus and suspension cultures.

#### UNIT II:

**Applications of Plant Tissue culture**, meristem culture, clonal propogation, production of haploids (Anther culture), somatic embryogenesis, protoplast culture and somatic hybridization-Cybrids, Hybrids. Somoclonal variation-sources and its applications. Cryopreservation.

Production of secondary metabolites –Classication isolation, characharization and biosynthetic pathway of Secondary metabolites.

#### UNIT III:

**Tools and techniques of animal cell and tissue culture**: Culture media, growth factors, laboratory facilities. CO2 incubator, Cell count-haemocytometer, coulter method, MTT based Assay. Characteristics of cells in culture: Contact inhibition, anchorage dependence, cell-cell communication etc.; Cell senescence; cell and tissue response to trophic factors. Primary culture, immortal cells, cell lines. Maintenance of cell lines in the laboratory-subculture, trypsinisation and cryopreservation.

#### UNIT IV:

**rDNA products:** Brief idea about recombinant DNA products in medicine (insulin, somatostatin, vaccines), Concept of Gene therapy, Production of recombinant vaccines – hepatitis. Concept of transgenic animals. In vitro fertilization and embryo transfer in humans and farm animals. Cloning-Concepts of nuclear transfer and creation of Dolly. Stem cells-types and applications.

#### UNIT V:

**IPR:** Intellectual property rights. Types and importance of IPRs (Copy rights. Patents, GIs...) International and national organizations, agencies and treaties. Regulation of GMO/transgenics. ethical aspects of Biotechnology.

Biostatistics: Basic concepts of Mean, Median, Mode ,Standard Deviation and Standard Error. Introduction to ANOVA

## PRACTICALS: 619EL01PLANT AND ANIMALBIOTECHNOLOGY

- **21.** Preparation of MS medium
- **22.** Sterilization methods
- 23. Callus culture
- 24. Demonstration of Suspension Culture
- 25. Determination of seed viability by TTC
- **26.** Establishing primary cell culture of chicken embryo fibroblasts.
- 27. Animal tissue culture maintenance of established cell lines.

- **28.** Cell count by Haemocytometer.
- **29.** Microphotography Cell line photographs.
- **30.** Synthetic seeds
- **31.** Calculation of Mean ,Median and Mode(Manual/Computer aided)
- **32.** Calculation of Standard deviation and Standard Error (Manual/Computer aided)
- **33.** Biostatical problems based on Standard Deviation

#### **Recommended Books**

- 21. Culture of Animal Cells A Manual of Basic Technique. Freshney, R .I. 2006. 5<sup>th</sup> Edn. John Wiley and Sons, USA.
- 22. Cell Culture. Yadav, P.R and R. Tyagi. 2005. Discoery Publishing House, New Delhi.
- 23. Principles of Cloning. Jose Cibelli, Robert P. Lanza. Keith H.S. Campbell. 2002. Academic Press, London.
- 24. Animal Cell Technology Principles and practices. Butter, M. 1987. Oxford Press, USA.
- 25. Elements of Biotechnology. Gupta, P.K. 1996. Rastogi and Company, Meerut.
- 26. Plant Tissue Culture: Theory and Practice, a Revised Edition. Bhojwani, S.S and M.K.Razdan.2004.Panima Publishing Corporation, New Delhi.
- 27. Biotechnology-Fundamentals and applications. Purohit, S. 2000. 3<sup>rd</sup> Edn. Agrobios, New Delhi.
- 28. An Introduction to Plant Tissue Culture. Razdan, M.K. 1995. Oxford and IBH Publishing Company, New Delhi.
- 29. Plant Tissue Culture and Molecular Biology, Applications and Prospects. Srivastava, P.S. 1998. Narosa Publishing House, New Delhi.
- 30. Intellectural Property rights-Deborah, E. Bouchoux, Cengage Learning

#### 619EL02- ENVIRONMENTAL BIOTECHNOLOGY (Elective theory)

#### (w.e.f 2019-20)

# Unit I

**Principles of Ecology,** Water and terrestrial ecosystems, Bio-geo chemical cycles - Carbon, Nitrogen cycles. Role of microbes in bio-geochemical cycles.

# Unit II

**Inorganic and Organic pollutants** of air, land and water; maintenance of standards, Environmental monitoring. Detection, treatment and prevention of pollution. Biological indicators

# Unit III

**Biocides,** Four stage alternatives, Refuse disposal - Treatment methods, effluent from pharmaceuticals, fertilizers, pulp and paper industry.

# Unit IV

Waste water management - Aerobic and anaerobic treatment, primary, secondary and tertiary treatment of municipal wastes, Solid waste management.

# Unit V

**Bioremediation**, Biodegradation of recalcitrant compounds and the role of genetically engineered microbes and genetically modified organisms in the environmental management.

# PRACTICALS BTP: 604- ENVIRONMENTAL BIOTECHNOLOGY (Elective Lab)

- 1. Detection of coliforms for determination of the purity of potable water.
- 2. Determination of total dissolved solids of water
- 3. Determination of Hardness and alkalinity of water sample.
- 4. Determination of dissolved oxygen concentration of water sample
- 5. Determination of biological oxygen demand of sewage sample
- 6. Determination of chemical oxygen demand (COD) of sewage sample.
- 7. Isolation of xenobiotic degrading bacteria by selective enrichment technique
- 8. Estimation of heavy metals in water/soil
- 9. Estimation of nitrate in drinking water.
- 10. Preparation and formulation of microbial biopesticide (bacteria, fungi and viruses)
- 11. In vitro evaluation of medicinal plants against pathogenic microbes.
- 12. Effect of mycorrhizal fungi on growth promotion of plants.
- 13. Production of microbial fertilizers (Rhizobium, Azotobacter and AMF).

# **Recommended Books**

1. Ecology and Environment P.D.Sharma

2. Brock Biology of Microorganisms (9th edition) by Madigan, Martinko & Parker.2000. Prentice Hall.

- 3. Environmental Microbiology by Mitchell. 2009. John Wiley and Sons.
- 4. Environmental Microbiology by Grant & Long. 1981. Wiley.

5. Environmental Microbiology: A laboratory manual by Peppler, Gerba & Brendecke.1995. Academic Press.

#### 619EL03-Industrial Biotechnology (Elective theory) (w.e.f 2019-20)

# Unit I:

**Isolation, Screening**, Preservation and Improvement of Industrially Important Microorganisms. Synthetic and Natural Medium, Precursors, Antifoams, Sterilization Methods and Inoculum Preparation.

# Unit II:

**Definition of bioreactor**, Basic principles of bioreactor. Classification of bioreactors. Analysis of batch, continuous, fed batch and semi-continuous bioreactors.

# Unit III:

**Ethanol Production** by Fermentation using Molasses, Starchy Substances. Production of Alcoholic Beverages like Beer and Wine. Production of Citric Acid by Submerged and Solid State Fermentations.

#### Unit IV:

**Sources of Industrial Enzymes**, Production of Microbial Enzymes like Amylase and protease. Backer's Yeast and SCP Production. Production of Antibiotics : Penicillin .

#### Unit V:

**Biotechnology Products-** Production of recombinant proteins having therapeutic and diagnostic applications( Insulin, Growth Harmone, Recombinant vaccines, Monoclonal Antibody).

#### PRACTICALS BTP: 606 Industrial Biotechnology (Elective Lab)

- 21. Isolation of bacteria / fungi from soil by serial dilution technique
- 22. Estimation of amylase from seeds.
- 23. Production of  $\alpha$  amylase from *Bacillus Spp.* by shake flask culture.
- 24. Production of alcohol or wine using different substrates.
- 25. Estimation of alcohol by titrimetry.
- 26. Estimation of alcohol by colorimetric method .
- 27. Production of citric acid.
- 28. Citric acid production by submerged fermentation.
- 29. Estimation of citric acid by titrimetry.
- 30. Demonstration of Fermentor and bioreactor

#### **Recommended Books**

- 1. Biogas technology by b.T.Nijaguna
- 2. Food microbiology by m.R.Adams and M.O.Mass
- 3. Industrial microbiology by A.H.Patel
- 4. Industrial microbiology ,A text book W.Clark
- 5. Industrial Microbiology by L.E.Casida

- 6. Introduction to Biotechnology by P.K.Gupta
- 7. Text book of Biotechnology by H.K.Das (Wily Publications)
- 8. Bioprocess engineering by Shuler (Pearson Education
- 9. Biotechnology: A Text Book of Industrial microbiology

2nd edition by Crueger. & Crueger. 1990. Sinauer Associates.

- 10.Microbial Biotechnology: Fundamentals of Applied Microbiology (2nd edition) by Glazer & Nikaido. 2007. Cambridge University Press.
- 11. Prescott & Dunn's Industrial Microbiology 4th edition Editor Reed. 1982. AVI Pub. Co.,

# CLUSTERS

# 619CLA1 - TISSUE CULTURE(w.e.f 2019-20)

## <u>UNIT - I</u>

Animal tissue culture – Introduction to culture medium Culture Vessels – natural and artificial medium preparation and sterilization of medium, Isolation of Tissue, Disagrigation of Tissue and subculturing.

## <u>UNIT - II</u>

Cell cultures, suspension cultures, maintenance of cell lines, organ culture – techniques, advantages, applications, Hybridoma technology – Applications and Limitations

#### UNIT - III

Plant tissue culture, Introduction to culture room, Culture vessels, their working. Sterilization of nutrient medium, callus cultures, subculture.

#### UNIT - IV

Somatic embryogenesis, meristem culture, somatic hybridization, micro propagation, somatic hybrids & cybrids.

#### <u>UNIT - V</u>

Invitro pollination, parthenogenisis, applications of tissue culture both in agriculture and animal husbandry. Embryo culture technique.

#### 619CLAP -Practical -TISSUE CULTURE

- 1. Preparation of plant tissue culture medium
- 2. Initiation of callus from any one selected plant species
- 3. Micro propagation of plants
- 4. Cell disaggregation and cell counting
- 5. Study of gene transfer techniques (Transformation)
- 6. Preparation of animal tissue culture medium

# III B. Sc. (SEMESTER - VI) 619CLA2 - INDUSTRIAL BIOTECHNOLOGY(w.e.f 2019-20)

# <u>UNIT - I</u>

Introduction to industrial biotechnology. Primary and secondary metabolic products of microorganisms. Screening, isolation and preservation of industrially important microorganisms, Synthetic and Natural medium, Precursors and antifoams

# <u>UNIT - II</u>

Fermentation technology: principles of fermentation, fermentation medium, types of fermenters, fermentation conditions and sterilization methods. Factors involved in fermenter design. Fermenter configurations. Principle operating characteristics of fermenters.

# <u>UNIT - III</u>

Factors effecting fermentation. Fermented products like – fermentation of molasses, production of alcoholic beverages like beer, wine. Commercial production of fuels and chemicals by microbial fermentations, Production of Citric acid

# <u>Unit - IV</u>

Basics in the production of antibiotics – penicillin and streptomycin. Downstream process techniques. Production and application of recombinant proteins – Insulin, Growth harmone, Recombinant vaccines(Rabies Vaccine) Monoclonal Antibodies and interferons.

# <u>UNIT - V</u>

Fermentative production of foods and dairy products. Production of vitamins – B12, B2 & C. Good manufacturing practices. Bio safety issues. Intellectual property rights and patenting issues.

# Practical 619CLA 2P - : INDUSTRIAL BIOTECHNOLOGY

- 1. Isolation of Amylase producing organisms from soil.
- 2. Production of Amylase from bacteria / fungi
- 3. Assay of Amylase activity.
- 4. Production of alcohol (or) wine from grapes
- 5. Estimation of alcohol by colorimetric method
- 6. Production of citric acid from bacteria / fungi
- 7. Estimation of citric acid by titrimetric method.
- 8.Immobilization of cells or Enzymes
- 9.Starch Hydrolysis
- 10.Patent Fileing
- 11.Growth curve(Bacterial)

# 619CLA 3 - ENVIRONMENTAL BIOTECHNOLOGY (w.e.f 2019-20)

#### **Unit - I : Environmental Concepts**

Introduction to environmental biotechnology and ecology. Environmental Concepts – Atmosphere, Ozone layer, Hydrosphere, Lithosphere and Biosphere.Biogeochemical Cycles(Nitrogen, Carbon and Sulphar)

## **Unit - II : Ecosystem and Ecology**

Ecosystem and community. Factors effecting Ecosystem. Ecological pyramids. Endangered and Extinct species. Renewable and non renewable energy resources.

#### **Unit - III : Pollution**

Air, Water, Soil, types of pollutants i.e pesticides, insecticides, heavy metals, toxins, radiation, hazardous waste and biodegradation with microbes. Biopesticides and biofertlizers.

#### **Unit – IV : Protection and Restoration of Ecosystem**

Major protection acts in India (Forest conservation act 1980, wild life protection act 1972), Bioremediation, Phytoremediation, waste water treatment, solid waste management. Microbial ore leaching.

# **Unit - V : Environmental Protection**

Bio gas production, Global warming, Green house effect, Rain water harvesting, Energy conservation, acid rains, ozone depletion, deforestation and its effects, reforestation.

# 619CLA3P -Practical ENVIRONMENTAL BIOTECHNOLOGY

- 1. Estimation of BOD of water
- 2. Estimation of COD of polluted water
- 3. Collection of endangered and extinct species data
- 4. Study production of hydrogen (or) biogas using Cow / cattle dung
- 5. Most Probable Number for water
- 6. Estimation of Organic matter in water samples
- 7. Isolation of Nitrifying Bacteria
- 8.Production of Biofertilizers (Azolla)
- 9. Isolation of Rhizobium

10.In vitro toxicity of mercuric chloride on Succinic dehydrogenase activity

# **MODEL QUESTION PAPER**

B.Sc., Three year degree examinations-2019 V Semester Biotechnology 5191A-Molecular Biology (W.E.F 2019-20)

Time -3hrs

Max Marks : 60M

Section – A Answer any FIVE Questions

5x4=20M

(Each question carries 4 Marks )

- 1. Eukaryotic genome
- 2. Structure of DNA
- 3. Polymerases
- 4. Rolling circle replication of DNA
- 5. Core enzymes
- 6. hnRNA
- 7. Operons
- 8. BLAST

#### Section – B

#### Answer ALL of the following questions 5x8=40M

#### (Each question carries 8 Marks )

1. (A) Genomic organization of eukaryotic genomes Or

(B)Write the experiments to prove DNA as genetic material

10(A) Describe about the enzymes that are involved in the DNA replication Or

(B)Write about the DNA Replication in Eukaryotes

11(A) Describe about the transcription in prokaryotes

(B) Describe about the transcription in Eukaryotes

12(A)Write the regulation of gene expression in Lac Operon

Or

- (B)Write about poly and Mono cistronic m-RNA
- 13(A)Draw the diagram and describe about the structure of t RNA

#### Or

B) write about bioinformatics and the applications of search tools