

**MB 301: BIOSTATISTICS, COMPUTER APPLICATIONS AND RESEARCH
METHODOLOGY**

----- Marks: 50-----

Unit I: Introduction to biostatistics. (08)

Basic definitions and applications, sampling representative sample size, sampling bias and sampling techniques. Data collection and presentation: types of data, methods of collection of primary and secondary data, methods of data collection, graphical representation by histogram, polygon, ogive curves and pie diagram.

Unit II: Measures of central tendency. (09)

Measures of central tendency: mean, median, mode.

Measures of variability of variation. Correlation and regression: positive and negative correlation and calculation of Karl Pearson's coefficient of correlation. Linear regression and regression equation and multiple linear regressions. ANOVA, one and two way classification. Calculation of an unknown variable using regression equation.

Unit III: Tests of significance. (10)

Tests of significance: small test (Chi-square t-test, F-test), large sample test (Z-test) and standard error. Introduction to probability theory and distribution (concept without deviation) binomial poisson and normal (only definitions and problems) computer oriented statistical techniques. Frequency table of single discrete variable, bubble plot. Computation of mean, variance and standard deviations, t test, correlation coefficient.

Unit IV: Computer: Introduction and application. (10)

Introduction to computers and computer applications: Introduction to computers, Computer applications in research, basics, organization, PC, mainframes and Super-computers, concept of hardware and software, concept of file, folders and directories, commonly used commands, flow charts and programming techniques. Introduction in MS Office software concerning Word processing, spreadsheets and presentation software.

Unit V: Scientific writing in research.

(08)

Research: Definition, importance and meaning of research, characteristics of research, types of research, steps in research, identification, selection and research problems, formulation of hypothesis.

Scientific writing- characteristics. Logical format for writing thesis and papers. Essentials features of abstract, introduction, review of literature, materials, methods, and discussion. Effective illustration- table and figures. Reference styles- Harvard and Vancouver systems.

PRACTICALS:

- 1) Representation of statistical data by
 - a) Histogram
 - b) Ogive curve
 - c) Pie diagram.
- 2) Determination of statistical averages / central tendencies.
 - a) Arithmetic mean
 - b) Median
 - c) Mode.
- 3) Determination of measure of dispersion.
 - a) Mean deviation.
 - b) Standard deviation and coefficient of variation.
 - c) Quartile deviation.
- 4) Tests of significance-Application of following.
 - a) Chi-square test.
 - b) t-test
 - c) standard error
- 5) Creating files, folders and directories.
- 6) Application of computers in biology using MS-office.
 - a) MS-word
 - b) Excel
 - c) Power point.

- 7) Creating and e-mail account, sending and receiving mails.
- 8) An introduction to Internet, search engines, websites, browsing and downloading.

REFERENCES:

- 1) Biostatistical methods by Lachin.
- 2) Biostatistics- 7th edition by Daniel.
- 3) Fundamentals of biostatistics by Khan.
- 4) How computer work (2000) by Ron White. Tech Media.
- 5) How the internet work 2000 by Preston Garlla Tech. Media.
- 6) Practical statistics for experimental biologist by Wardlaw A.C (1985).
- 7) Research methodology methods and statistical techniques- Santosh Gupta.
- 8) Research methodology methods and techniques- C.R. Kothari.
- 9) Research methods in Biological sciences. Palanisamy S. and M. Shanmugavelu. 1997. Palani Paramount publications, Tamildanu. India
- 10) Statistics for biologist by Campbell R.C (1974). Cambridge University Press, UK.
- 11) Statistics in biology Vol:1, by Bliss, C.I.K (1967) Mc Graw Hill, New York.

MB 302: MOLECULAR IMMUNOLOGY

----- Marks: 50-----

Unit I: Organs and cells of immune system. (10)

Primary lymphoid organs - thymus, bone marrow - structure and function. Lymphatic system - transporter of antigen - introduction. Secondary lymphoid organs - spleen and lymph nodes structure and functions. Mucosal associated lymphoid tissue, (MALT) - tonsils. Cutaneous associated lymphoid tissue - keratinocytes and langerhans cells - Location and immunological functions.

Lymphoid cells - B-lymphocytes and T-lymphocytes - maturation, activation and differentiation. Receptor on B and T cells. Null cells. γ δ T cells - Intraepithelial lymphocyte (IEL)- function, Mesangial cells, Microglial cells - Structures and secretions - interleukin I, hydrolytic enzymes, complement proteins, α -Interferon, Tumor necrosis factor α (TNF- α) (IL-6, GM-CSF, G-CSF, M-CSF). Growth factors associated in haematopoiesis, Granulocytes - Neutrophile, Basophile, Eosinophile - immune response generated against parasite by granulocytes. Mast cell - Structure, function in innate immunity and acquired immunity. Dendritic cell - structure and function.

Unit II: Immunogens and Immunoglobulins. (09)

Types of antigens - Exogenous, Endogenous, Autologous, Xenogenic and Allogenic. General properties of antigens - Molecular size, chemical composition, foreignness, specificity, Haptens, Superaantigens and Adjuvants: Freund, complete and incomplete adjuvants, Depot effect, Macrophage activation, Effect of lymphocyte, antitumor action,

Epitopes: A.A.sequence /structure. Immunoglobulins: Classes, Structure, distribution and function. Isotypic, Allotypic, Idiotypic determinants. Idiotypic network. Antibody production theories.

Unit III: Organization and Expression of Immunoglobulin genes. (10)

Genetic model for Ig structure, Germ line and somatic variation models, Dryer and Bennett two gene models, K chain genes, λ chain genes, Heavy chain genes, VH gene segments, Gene rearrangement in VH region - In light chain, In heavy chain, Mechanism of variables region DNA rearrangement, Generation of antibody diversity, Regulation of Ig gene transcription

Unit IV: Major and Minor Histocompatibility Complexes. (08)

MHC class-I, MHC class-II - Structure of molecules, gene organization. Genetic polymorphism of molecule, Peptide interaction with molecule, MHC and immune responsiveness, MHC and susceptibility to infectious diseases, Minor MHA - structure, role and genetics, HLA system, Antigen processing and presentation

Unit V: Clinical immunology. (08)

Hypersensitivity, Immunology of Tumors, Immunodeficiency diseases, autoimmune diseases, Immunomodulation / Immunological tolerance.

PRACTICALS:

1. Ag – Ab reaction

- Agglutination - Slide – widal test
 - Tube - Dreyer's technique
 - Bordet Durham's technique
 - Quantitative widal test.
- * Precipitation - Slide - VDRL, RPR, RA
- * Complement fixation test - Coomb's test (demonstration)

2. Radial Immunodiffusion.

3. Immunohaematology.

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- * DLC, TLC, RBC count
 - * Blood grouping - ABO system
 - Rh grouping
4. Separation of serum proteins by electrophoresis.
 5. Preparation of 'H' antigen of *S. typhi* by Craigies tube method.
 6. Preparation of 'O' antigen of *S. typhi* by phenol agar method.

REFERENCES -

1. Genes VII; Benjamin Lewin, Oxford University Press.
2. Immunology ; Weir,
3. Immunology; C. Vaman Rao, 2nd edition, Narosa publication.
4. Immunology; Janis Kuby, 2nd edition, W. H. Freeman and company.
5. Practical Immunology ; Talwar,
6. Roitt's Essential Immunology; Ivan Roitt, 9th edition, Blackwell Sciences.

MB 303 : MICROBIAL DIVERSITY AND EXTREMOPHILES

----- Marks: 50-----

Unit 1: Biodiversity (09)

Introduction to microbial diversity-Distribution, Abundance, Ecological Niches. Types-Bacterial, Archaeal, Eucaryal, Characteristics and Classification of Archae (Methanogens).

Unit 2: Thermophiles (09)

Classification, Hyper-thermophilic habitat and ecological aspects. Molecular basis of thermo-stability, Heat stable enzymes and metabolism, Genetics of thermophiles, Minimal complexity model systems.

Commercial aspects of thermophiles and application of thermoenzymes

Unit 3: Acidophiles and Alkalophiles (09)

Acidophiles- Classification, life at low pH, acido-tolerance, applications.

Alkalophiles- Isolation, Distribution and Taxonomy. Cell structures-Flagella, Cell wall, Cell membrane. Physiology- Growth conditions, Mutants, Antiporters and alkaliphily. Intracellular enzymes. Molecular biology- Alkalohiles as DNA sources, secretion vectors, promoters

Enzymes of alkaliphiles and their applications

Unit 4: Psychrophiles (09)

Conditions for microbial life at low temperature Climate of snow and ice, limits for life at subzero temperature.

Microbial diversity at cold ecosystem – snow and glaciers ice, subglacial environments, psychropiezophiles, permafrost, anaerobic and cyanobacteria in cold ecosystem, microalgae in Polar Regions.

Molecular adaptations to cold habitats – Membrane components and cold sensing, cold adapted enzymes, cryoprotectants and ice binding proteins , role of exopolymers in microbial adaptations to sea ice

Unit 5: Halophiles and Barophiles (09)

Halophiles- Classification, Halophilicity and Osmotic protection, Hypersaline Environments, Eukaryotic and prokaryotic halophiles Halobacteria – cell wall. Membranes, compatible solutes, osmo-adaptations or halotolerance, Applications of halophiles and their extremozymes.

Barophiles- Classification, high pressure habitat, life under pressure, barophily, death under pressure.

PRACTICALS:

1. Isolation of thermophiles from hot water spring (Study at least one thermostable enzyme).
2. Studies on halophiles isolated from high salt habitat. (Study its pigmentation and salt tolerance phenomenon).
3. Studies on alkalophiles and its enzymes (any one) isolated form extreme alkaline environment.
4. Biogenic methane production using different wastes.
5. Isolation of *Thiobacillus ferrooxidans* and *Thiobacillus thiooxidans* culture from metal sulfides, rock coal and acid mine water.

REFERENCES:

1. Advances in applied microbiology. Vol.X, edited by Wayne W. Umbreit and D. Pearlman Academic Press.
2. Brock biology of Microorganisms. XI edition. By Michael T. Madigan, John M. Martinko. Pearson Education International.
3. Extreme environment. Metabolism of microbial Adaptation. Milton R., Heinirich Academic Press.

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4. Extremophiles by Johri B.N. 2000. Springer Werlag, New York.
5. Microbial diversity by Colwd D., 1999, Academic Press.
6. Microbial ecology. Fundamental and applications by Ronald M. Atlas and Richard Bartha. II and IV edition.
7. Microbial Ecology. Ii edition by R. Campbell. Blackwell scientific publication.
8. Microbial life in extreme Environment. Edited by D.J. Kushner. Academic Press.
9. Microbiology : Dynamics and Diversity by Perry.
10. Microbiology of Extreme Environment . Edited by Clive Edward. Open University Press. Milton Keynes.
11. Microbiology of extreme Environment and its potentials for Biotechnology. Edited by N. S. Da Coasta, J. C. Duarata,, R.A.D. Williams. Elsisver applied science, London
12. Thermophiles. General, Molecular and applied Microbiology. Thomas D.Brock. Wiley Interscience publication.

MB 304: RECOMBINANT DNA TECHNOLOGY

----- Marks: 50-----

Unit I: Techniques and enzymes used in genetic engineering. (10)

Core techniques of gene cloning and essential enzyme used in genetic engineering: restriction endonucleases type I, II, III, restriction modification system: nomenclature and classification of type II endonucleases, their activity, DNA ligase: properties and specificities, S₁ nuclease, BAL 31 nuclease, DNA polymerase, polynucleotide kinase, phosphatase, reverse transcriptase and its activity and mode of action. Restriction digestion, ligation and transformation. Hybridization techniques: Northern, southern and colony hybridization, fluorescence *in situ* hybridization. Restriction map and mapping techniques, DNA fingerprinting, chromosome walking and jumping.

Unit II: Cloning vectors. (08)

Gene cloning vectors: plasmids and their properties, pBR 322 and pUC18 its derivatives and construction, single stranded plasmid, promoter probe vectors, runaway plasmid vectors. Bacteriophage as cloning vectors, EMBL, λgt 10/11, λZAp etc. cosmid vectors. Artificial chromosome vectors (YAC, BACs). Animal virus derived vectors, SV₄₀vaccina/bacculo and retroviral vectors. Expression vectors, pMal, GST, pET based vectors.

Unit III: Cloning methodologies (10)

Insertion of foreign DNA into the host cells: transformation, transfection: chemical and physical method, liposomes, microinjection, electroporation, biolistic, somatic cell fusion, gene transfer by pronuclear microinjection, plant transformation technology: Basic of tumor formation, hairy root, features of Ti and Ri plasmids, mechanism of DNA transfer, role of virulence gene, use of Ti and Ri as plasmids vectors. Cloning and expression in yeast (*Saccharomyces*, *pichia* etc), animal and plant cells, methods of selection and screening, cDNA and genomic cloning, expression cloning, jumping and hopping libraries, phage display, construction of cDNA libraries in

plasmids and screening methodology, construction of cDNA and genomic DNA libraries in plasmids in lambda vectors, principles in maximizing gene expression.

Unit IV: Polymerase Chain Reaction. (08)

Primer design, fidelity of thermal enzymes, DNA polymerase, multiplex, nested reverse transcriptase, realtime PCR touchdown PCR, hot start PCR, colony PCR, cloning of PCR products, T vectors, proof reading enzymes, PCR in gene recombination , deletion , addition , overlap extension and SOEing , site specific mutagenesis, PCR in molecular diagnostics, viral and bacterial detection, PCR based mutagenesis.

Unit V: PCR application. (09)

sequencing methods: enzymatic DNA sequencing, chemical DNA sequencing of DNA, principles of automated DNA sequencing, RNA sequencing, chemical synthesis of oligonucleotides, gene silencing techniques: introduction to si RNA and si RNA gene technology, micro RNA, construction of si RNA vectors, principle and application of gene silencing and germ line therapy in vivo and ex-vivo, suicide gene therapy, gene replacement, gene targeting, RFLP, RAPD, AFLP analysis. Application of recombinant DNA technology in medicine, agriculture and veterinary sciences.

PRACTICALS:

1. Demonstration of gene cloning,
2. DNA fingerprinting.
3. DNA ligation by T4 DNA ligase.
4. DNA molecular size determination.
5. Isolation of genomic DNA and it's confirmation by Southern blotting
6. Isolation of plasmid DNA and it's Restriction digestion.
7. PCR amplification from genomic DNA and analysis by agarose gel electrophoresis.
8. RAPD application.
9. Restriction mapping.

REFERENCES:

1. Biotechnology: A guide to genetic engineering by Peteres.
2. DNA cloning: A practical approach. D.M. Glover and D.D. Harmes, RL press, Oxford 1995.
3. Essentials of molecular biology vol. I (A Practical Approach). Brown T.A., IRL press Oxford. 1995.
4. From Gene to Clone. Winnaker.
5. Genetic engineering, principles and practice, Sandhya Mitra. Macmillan india Ltd.
6. Genome mapping and sequencing edited by Ian Dunham. Horizon Scientific press.
7. Manipulation and expression of Recombinant DNA. Robertson.
8. Methods in enzymology gene expression technology, vol. 185 D.A Godgel. Academic press Inc, San Diego, 1990.
9. Methods in enzymology guide to molecular cloning techniques, vol. 152 S. L. Berger. Academic press .Inc, san Diegn, 1996.
10. Molecular biotechnology, 2nd edition. S.B. Primrose, Blackwell Scientific publishers, Oxford. 1994.
11. Molecular biotechnology: principles and application of Recombinant DNA II edition. 1998. Bernard R. Glick and J. Pastemak, ASM publication.
12. Nicholl D.S.T., An introduction to genetic engineering 2nd edition, Cambridge University press, Cambridge, U.K.
13. PCR application. Protocol for functional genomics. Edited by Michael A. innis. David H., Gelfand John J. Sninsky, Academic Press.
14. PCR technology- principles and application for DNA amplification, Henry A Erilch (Ed) Stockton Press. 1989.
15. Principles of gene manipulation, Old and Primrose, Blackwell Scientific publication. 1994.

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16. Route maps in gene technology, M.R. Walker and R.Rapley, Blackwell science, Oxford, 1997.
17. Sambrook J, Fritsch E.F and Maniatis (1989) molecular cloning vol. I, II, III. IInd edition, cold spring harbor laboratory press, New York.