# M.Sc. (Nanobiotechnology) Syllabus (Credit System) for the PG Department of Biotechnology of the University of Pune

# COURSE STRUCTURE FORMAT Theory & Practical

Semester I	Course name	No. of
Theory		credits
Theory NBT 11*	Pielegiaal Chemistry	4
NBT 12*	Biological Chemistry	4
NBT 13	Cell Biology Quantitative Methods	4
Practical		4
NBT 14	Biological Chomistry	6
NBT 15	Biological Chemistry Cell Biology	3
NBT 16	Quantitative Methods	3
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Someotor II		24
Semester II Theory		
NBT 21*	Molecular Biology	4
NBT 22	Genetics	2
NBT 23a		2
NBT 23b	Microbial Technology	2
NBT 230	Immunology Fundamentals of Nanoscience &	2
INDI 24	Nanotechnology	Z
NBT 25	Bioinformatics	2
Practical	Bioinionnalics	Ζ
NBT 26	Molocular Biology	4
NBT 27	Molecular Biology Genetics	2
NBT 28		
	Microbiology & Immunology	3
NBT 29	Bioinformatics	
<u> </u>		25
Semester III		
Theory		
NBT 31*	Animal & Plant Biotechnoloy	4
NBT 32*	Genetic Engineering	2
NBT 33*	Nanobiotechnology	4
NBT 34*	Biochemical Engineering	2
NBT 35*	Pluripotent cell technology and reproduction	2
Practical		
NBT 36	Animal Tissue Culture	2
NBT 37	Genetic Engineering	4
NBT 38	Seminar	6
		26

# Semester IV

NBT 41	Research Project
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# TOTAL CREDITS = 100

\* All courses have been identified in terms of the prerequisites and lateral integration. All practical courses have lateral integration with theory.

# COURSE STRUCTURE FORMAT WITH MODULES, 2012-13

# THEORY

Course No.	Subjects	Theory 4C X 15 L= 60 L
Semester I		
NBT 11	Biological Chemistry	101
NBT 11.1	Biochemistry of macromolecules and building blocks	12 L
NBT 11.2 NBT 11.3	Organic reactions and stereochemistry	08 L 20 L
NBT 11.3 NBT 11.4	Introduction to enzymology, metabolism and bioenergetics Basic Biochemical techniques	20 L 20 L
NBT 12	Cell Biology	
NBT 12.1	Cell structure and methods in cell biology	15 L
NBT 12.2	Biomembranes and transmembrane signalling	15 L
NBT 12.3	Cell dynamics, cell differentiation, cell death and	15 L
	transformation	4 - 1
NBT 12.4	The plant cell	15 L
NBT 13	Quantitative Methods	
NBT 13.1	Biostatistics	15 L
NBT 13.2	Biomathematics	15 L
NBT 13.3	Basic Concepts in Computing & Networking	15 L
NBT 13.4	Introduction to Programming	15 L
Semester II		
NBT 21	Molecular Biology	
NBT 21.1	Genome Structure & Organization	15 L
NBT 21.2	DNA replication and DNA repair	15 L
NBT 21.3	Gene Expression in Prokaryotes & Eukaryotes	15 L
NBT 21.4	Protein Synthesis, modifications and transport	15 L
NBT 22	Genetics	
NBT 22.1	Basic Concepts in Genetics	15 L
NBT 22.1 NBT 22.2	Microbial Genetics	15 L 15 L
NBT 23a	Microbial Technology	
NBT 23a.1	Microbial Technology Microbial Characteristics	15 L
NBT 23a.2	Applied Microbiology	05 L
NBT 23a.3	Fungal Biotechnology	10 L
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BOS, Biotechno	ology, University of Pune, May, 2012 P	Page 3

<b>NBT 23b</b> NBT 23b.1 NBT 23b.2	<b>Immunology</b> Immunology I Immunology II		15L 15L
NBT 24	Fundamentals of Nanoscience & Nanotechnology		
NBT 24.1	Introduction, classifications and definition	15 L	
NBT 24.2	Physical and chemical fundamentals of nanomaterials	15 L	
NBT 25	Bioinformatics		
NBT 25.1 NBT 25.2	Biological Data Bases Applications of Bioinformatics	15 L 15 L	
Semester III			
NBT 31*	Animal and Plant Biotechnology		
NBT 31.1	Introduction to tissue culture techniques		15 L
NBT 31.2	Animal cell and organ culture		15 L
NBT 31.3	Plant cell, tissue and organ culture		15 L
NBT 31.4	Applications of tissue culture		15 L
NBT 32*	Genetic Engineering		
NBT 32.1	Basics of genetic Eng.& Cloning Strategies		10 L
NBT 32.2	Detection & Characterization of Transformants		05 L
NBT 32.3	Expression systems		10 L
NBT 32.4	Applications of Genetic Engineering		05 L
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NBT 33 NBT 33.1	Nanobiotechnology Properties & characterization of nanomaterials		15 L
NBT 33.2	Nanomaterials and biosystem interaction		15 L
NBT 33.3	Biomedical nanotechnology(Diagnostics, delival		15 L
	therapeutics)	<b>,</b>	
NBT33.4	Nanotoxicology		15 L
	Ricchomical Engineering		
<b>NBT 34</b> * NBT 34.1	Biochemical Engineering Theory and design of bioreactors		15 L
NBT 34.2	Transport and process control		15 L

<b>NBT 35</b> * NBT 35.1 NBT 35.2	Pluripotent cell technology and reproduction Cells of Reproduction and Early Development Stem Cell Concepts & Technologies	15 L 15 L
Semester IV		
No theory course, only Research project (Practical)		

# CREDIT COURSE STRUCTURE FORMAT, 2012-13 PRACTICAL

Course No.	Subjects	Practical
Semester I		
NBT 14	Biological Chemistry	6C = 6x15=90h
NBT 15	Cell Biology	3C = 3x15=45h
NBT 16	Quantitative Methods	3C = 3x15=45h
Semester II		
NBT 26	Molecular Biology	4C = 4x15=60h
NBT 27	Genetics	2C = 2x15=30h
NBT 28	Microbiology & Immunology	3C = 3x15=45h
NBT 29	Bioinformatics	2C = 2x15=30h
Semester III		
NBT 36	Animal Tissue culture	2C = 2x15=30h
NBT 37	Genetic Engineering	4C = 4x15=60h
NBT 38	SEMINAR	6C = 6x15=90h
Semester IV		
NBT 41	Research Project	25C = 25x15 =375h

# Detailed Syllabus of M.Tech. Biotechnology, 2012-13 THEORY

# SEMESTER I

# NBT 11 BIOLOGICAL CHEMISTRY

NBT 11.1: Biochemistry of Macromolecules and Building Blocks(12L)Macromolecules (Nucleic acids, proteins, carbohydrates and lipids) and their building<br/>blocks: amino acids, purine and pyrimidine bases, fatty acids and sugars.buildingSmall molecules of biological importance: vitamins and minerals.biological importancebiological importance

#### NBT 11.2: Organic Reactions and Stereochemistry

Basic reactions in organic chemistry: Oxidations, reductions, substitutions, molecular rearrangements

Stereochemistry: Stereoisomers, resolution, cyclohexanes, asymmetric synthesis.

# **NBT 11.3: Introduction to Enzymology, Metabolism and Bioenergetics** (20L)

Enzymes: classification, catalysis, kinetics, regulation (fine, coarse and metabolic control). Coenzymes and cofactors, and their relevant reactions. Allostery.

Metabolic pathways: glycolysis, Krebs cycle, pentose phosphate pathways, glycogen metabolism, oxidative phosphorylation, fatty acid biosynthesis and oxidation and photosynthesis.

Bioenergetics Thermodynamics in biological systems.

# NBT 11.4: Basic Biochemical Techniques

Spectroscopy, UV – VS, Fluorescence, pH and Conductivity. TLC, Concepts in a) Chromatography b) Radioactivity. Native and SDS Polyacrylamide gel electrophoresis, 2 D electrophoresis.

# NBT 12 CELL BIOLOGY

NBT 12.1: Cell Structure and Methods in Cell Biology	(15L)
Cell: structural and functional organization.	
Cell motility.	
Ultrastructure and Electron microscopy.	
Fractionation of subcellular organelles.	
Microscopy, Morphometry, Cell counting.	
NBT 12.2: Biomembranes and Trans-Membrane Signaling	(15L)

Biomembranes: structure-function relationship.

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Cell signaling: Cell surface, Hormone, receptors and signal transduction and second messengers.

# NBT 12.3: Cell Dynamics, Cell Differentiation, Cell death, and Transformation (15L)

Cell dynamics, cytoskeleton and cell surface.

Extracellular matrix.

Cell-cell interactions and cell matrix interaction. Cell lineages and the context of Developmental biology Cell differentiation, hormones and growth factors. Apoptosis. The transformed cell Oncogenes, protooncogenes and etiology of cancer

# NBT 12.4: The Plant Cell

Structure of Plant Cell, Plant cell wall - primary and secondary, role in growth and development, Plamodesmata, their role in virus transfer etc.

Plastids - biogenesis, structure and types, chloroplast-Nucleus interaction, Rubisco, photosynthesis.

Growth and development of plants Programmed Cell Death

# NBT 13 QUANTITATIVE METHODS

# **NBT 13.1: Biostatistics**

Statistical population, sample from population, random sample. Tabular and graphical presentation.

Mean and standard deviation of group and ungrouped data.

Probability, relative frequency, probability distribution.

Binomial, poisson and normal distribution.

Test of significance, test for proportion, means and standard deviations, F and t test, chisquare test for goodness of fit.

Theory of errors, errors and residuals, precision, measure of precision, probable error of function, rejection of observation.

Methods of averages and least squares.

Correlation and linear regression, associated test of significance.

Analysis of variance for one and two way classification.

Design of experiments, randomization, replication, local control, completely randomized and randomized block design.

Nonparametric tests.

# **NBT 13.2: Biomathematics**

Differential and integral calculus.

Derivative and its physical significance, basic rules for differentiation (without derivation) maxim and minima, their applications in chemistry, exact and inexact differentiation with specific emphasis on thermodynamic properties, partial differentiation.

Page 8

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Curve sketching.

Basic rules for integration (without derivations), definite and indefinite integrals, geometric meaning of integration, applications in the biology and chemistry.

Solutions to quadratic and cubic equations.

Differential equations.

Separable variable, homogeneous, exact and linear equation, equations of second order Applications of differential equations in chemistry.

Determinants evaluations of 3 x 3 determinants, matrices manipulations, simultaneous equations and inversion.

Interpolation and polynomial fitting.

Trigonometry: Trigonometric functions, identities and inverse functions

Complex numbers: Representation, complex algebra, complex conjugate, roots

Matrices: Definitions and types of matrices, Matrix algebra and matrix multiplication, Special square matrices, Determinant of a square matrix, Inverse of a matrix, rank of a matrix, Eigen vectors and eigen values, diagonalization.

Vectors: Vector algebra, coordinate systems, Basic vectors and components, Scalar and vector multiplications, Reciprocal vectors, coordinate transformations.

Fourier transform and inverse Fourier transform.

Logarithmic and exponential functions

# NBT 13.3: Basic Concepts in Computing and Networking (15L)

- a) Overview and functions of a computer system, devices, memory -Types of processing: Batch, Real-Time, Online, Offline Types of modern computers: The work station, the minicoputer, mainframe computers, parallel processing computer, the super computer etc
- a) Introduction to operating systems: Windows / Unix / Linux
- b) The internet and its resources, World Wide Web (www): associated tools, services, resources and various terminologies
- c) Computer Networking, Network and Data security:
- i) OSI reference model, TCP/IP, topologies and protocols, designing networks
- ii) Networking gadgets(router, switch, etc); Data Communication (ISDN, VPN, DSL, cable modem, cellular modem, etc); Communication links (wire pairs, coaxial cables, fibre optics, microwave, satellite, etc)
- iii) Network security fundamentals: types of attacks, firewall, packet filtering, classification of data security threats, protection mechanism (authentication, access control, access rules)
- iv) Encryption / Decryption techniques
- v) An overview of computer viruses: How do they get transmitted? What are the dangers? General precautions to be taken.
- vi) Current & future technologies (grid computing, VPN, wireless, mobile computing, biometrics etc)

# NBT 13.4: Introduction to programming

C programming C++

#### SEMESTER II

#### NBT 21 MOLECULAR BIOLOGY

#### NBT 21.1: Genome Structure & Organization

Organization of viral, prokaryotic and eukaryotic genomes: DNA reassociation kinetics (Cot curve analysis), repetitive and unique sequences, kinetics and sequence complexities, satellite DNA, DNA melting and buoyant density. Gene families, clusters, packaging of chromatins and evolutionary advantage Organelle genomes. Rearrangement and amplification of DNA in the genome. Genomics and proteomics.

#### NBT 21.2: DNA Replication and DNA Repair

DNA polymerases, mechanisms of DNA replication in prokaryotes and eukaryotes DNA replication models DNA damage, DNA repair and recombination.

#### NBT21.3: Gene Expression in Prokaryotes & Eukaryotes (15L)

Chromatin structure and remodeling in relation to gene expression, DNase hypersensitivity, DNA methylation.

Transcription: Basic mechanism in prokaryotes and eukaryotes. RNA polymerase, Reverse transcriptase, and regulation of transcription including transcription factors. Post-transcriptional processing and transport of RNA. Non coding RNAs Organization and structure-function of ribonucleoproteins.

#### NBT 21.4 Protein Synthesis, Modifications and Transport (15 L)

Components of protein synthesis Mechanism of protein synthesis Genetic code Regulation of protein synthesis Post translational modifications Transport of proteins Protein turnover and degradation

#### **NBT 22: GENETICS**

#### **NBT 22.1: Basic Concepts in Genetics**

a) Science of genetics
b) Animal models (*Drosophila, Caenorhabditis*) in the study of Genetics
c) Mendelian principles, Concept of Dominance, multiple allelic systems, sex-linked inheritance, Epistasis, Pleiotropy, Penetrance,
d) Mutation, Chromosomal aberrations. Linkage studies, genetic maps.

BOS, Biotechnology,	University of Pune,	May, 2012	Page 10
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- e) Sex determination and dosage compensation
- f) Genotoxicity: detection and assays.
- g) Population genetics: Hardy-Weinberg law.
- h) Quantitative genetics and applications
- i) Plant genetics: Inbreeding and heterosis, and plant improvement.

# **NBT 22.2: Microbial Genetics**

- a) Mutagenesis: mutagenic agents, mechanisms of mutagenesis
- b) Expression of mutations- gene mutation.
- c) Point mutations, isolation of auxotrophs, conditional lethals and suppressor mutations
- d) Gene mapping in phages, bacteria.
- e) Control of gene expression in bacteria. Operon concept- lactose, arabinose and tryptophane operons.
- f) Transposons in prokaryotes and eukaryotes.
- g) Lambda genetic control

# NBT 23a: MICROBIAL TECHNOLOGY

# NBT 23a.1: Microbial Characteristics

Microbial life: Prokaryotes, Eukaryotes, Archeas & Protozoa Structure of microbial cell : Spore, cell wall, flagella, cell membrane, capsule, pilli. Characteristics of aerobes, anaerobes, cyanobacteria, actinomycestes. Nutrition, Metabolism, Growth media, propagation, Cell counting, Growth kinetics, Yield constants, Growth Synchronous growth. Handling pathogens, Sterilization, Safety in microbiology laboratory. Microscopic identifications immuno probe tests PCP application in diagnostic

Microscopic identifications, immuno probe tests, PCR application in diagnostic microbiology.

Action of antibiotics and multi drug resistance

# NBT 23a.2: Applied Microbiology

Microbiology for public health: Mycobacteria, Enterobacteria, and Protozoa. Microbiology for agriculture : Agrobacterium, Nitrogen fixation. Extremophiles, Industrially important microbes, secondary metabolites

# NBT 23a.3 : Fungal Biotechnology

Fungi, a unique kingdom – Conventional and Molecular taxonomy, Evolution Fungal interactions – with fungus, plant, animal and insects Architecture of fungal cell : cell wall, membranes and cytoskeleton Growth and differentiation of fungi – dimorphism, sexual and asexual sporulation Metabolism, Biochemical and molecular basis of development, apoptosis and autolysis Mycotoxins, strain improvement Uses of fungi in industry including food industry, biosensors, fuel cells, coal sulubilization, cancer therapy etc

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Page 11

Uses of fungi in agriculture and environment: Biofertilizer and Bioremediation, Biological control Medical Mycology

# NBT 23b: IMMUNOLOGY

#### NBT 23b.1: Immunology - I

Introduction, History, Phylogeny. Immune system overview, innate and acquired immune system. Components of immune system. Structure and function of antibody. Inflammation, opsonization. Primary and secondary lymphoid organs. Complement. B cell, T cell ontogeny. Characteristics of antigen, T cell dependent and independent antigens. Hypersensitivity. Primary and Secondary immune responses. Techniques in humoral immunology.

# NBT 23b.2: Immunology - II

BCR and TCR structure, γδTCR.
Generation of diversity.
MHC I and II gene, polymorphism.
Generation of immune response.
T helper, T cytotoxic cells.
MHC peptide interaction.
Antigen presentation, secondary signaling.
Immunological disorders and autoimmune diseases.
Lymphocyte traffic.
Techniques in cellular immunology.
Immune response to viral and bacterial lymphatic infection.

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#### NBT 24: FUNDAMENTALS OF NANOSCIENCE & NANOTECHNOLOGY (30 L)

Introduction The nanoscale dimension and paradigm (4L) Definitions and historical evolution (colloids etc.) and current practice (2 L) Types of nanomaterials and their classifications (1D, 2D and 3D etc. Nanocrystal, Nanoparticle, Quantum dot, Quantum Wire and Quantum Well etc) (2 L) Polymer, Carbon, Inorganic, Organic and Biomaterials –Structures and characteristics (3 L) Physical and Chemical Fundamentals of Nanomaterials Overview of synthetic methods (1 L) Surfactants, polymers, emulsions. Micelles/reverse micelles and colloids (4 L) Top-down and bottom up approaches (4 L) Biological Methods (4 L) Growth and stabilization (4 L) Self-assembly (2 L)

# **NBT 25: BIOINFORMATICS**

#### NBT 25.1 Biological Data Bases

The need for computation in Biology: An introduction to Bioinformatics, Historical overview, the principles involved, development of tools, internet based access Introduction to Biological Databases, Database Browsing and Data Retrieval

- Sequence databases
- Genome Databases

#### **NBT 25.2 Applications of Bioinformatics**

Application of Bioinformatics Approaches for analysis and interpretation of Sequence Data and using : Homology Searches, Sequence Alignments, Pattern Searching Application of Bioinformatics Approaches for analysis and interpretation of Genome data

such as - Gene prediction, Full Genome comparison etc. Introduction to computational structural biology: Protein structure prediction using computational methods, Structure analysis, Classification of Proteins etc.

#### SEMESTER III

# NBT 31 : ANIMAL & PLANT BIOTECHNOLOGY

#### NBT 31.1: Introduction to Tissue Culture Techniques

Introduction to tissue culture: Definition, principle and significance of tissue culture. Animal tissue culture.

Maintenance of sterility and use of antibiotics, Mycoplasma and viral contaminants.

Various systems of tissue culture - their distinguishing features advantages and limitations.

Culture medium: Logic of formulation (natural media, synthetic media, and sera). Methodology: i. Primary culture: Behaviour of cells, properties, utility.

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ii Explant culture. iii. Suspension culture. History & Development of plant tissue culture. Nutrient media: obligatory and optional constituents. Plant Growth Regulators: mode and mechanism of action. Incubation systems: static & agitated culture systems. Maintenance of *in vitro* cultures.

# NBT 31.2: Animal Cell Organ Culture

Cell lines: Definition, development, maintenance and management and Cell adaptation. Established cell lines: Their characteristic features and utility, Cross contamination hazards.

Characteristics of cells in culture.

Contact inhibition, anchorage (in) dependence, cell-cell communication etc, Cell senescence.

Cell and tissue response to tropic factors, Culturing of different cells.

Designing of an experiment in tissue culture and response assessment. Significance of various controls.

Growth studies: Cell proliferation, cell cycle, mitosis in growing cells.

Organ culture: Methods, behaviour of organ explant, and utility of organ culture.

Organ transplants. Freeze storing of cells and transport of cultures.

Mass production of biologically important compound.

Harvesting of products, purification and assays.

Propagation of viruses (viral sensitivity of cell lines).

Cell cloning and cell synchronization.

Separation of cell types: Various methods: advantages and limitations; Flow cytometry. Nuclear transplantation, Cell hybridization, Transfection studies.

# NBT 31.3: Plant Cell, Tissue and Organ Culture

Growth and development of plant cells and tissues in vitro.

- Callus culture
- Cell suspension culture
- Organ culture
- Protoplast culture
- Organogenesis
- Embryogenesis

In vitro culture: physical, genetic, chemical and genotypic factors.

Assessment of growth and development *in vitro*.

Problems in plant tissue culture (Recalcitrance, Contamination, Phenolic Browning, Seasonal Variation)

# NBT 31.4: Applications of Tissue Culture

Commercial applications of animal tissue culture: Tissue culture as a screening system. Cytotoxicity and diagnostic tests.

Development and preparation of vaccines against infecting organisms, mammalian cloning.

Establishment of cell lines from tissues of genetic diseases. Applications of Genetic manipulations.

Page 14

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Commercial applications of plant tissue culture for clonally identical plants: Mass propagation by organogenesis and embryogenesis, Synthetic Seeds, Use in multiplication of specific genotypes, rare and/or improved varieties, endangered species, disease elimination.

Workings of a commercial laboratory (Design, aseptic techniques and control of contamination, quarantine, pathological indexing, packaging, cost analysis, marketing).

#### **NBT 32: GENETIC ENGINEERING**

NBT 32.1: Basics of Genetic Engineering & Cloning Strategies	(15L)
General introduction and concept.	
Biosafety guidelines and containment strategies	
DNA modifying enzymes and restriction enzymes	
Cloning strategies: Genomic libraries, cDNA libraries, single gene cloning.	
Vectors in gene cloning: Types of vectors and choice of vectors- Plasmids, c	cosmids,
lamda phage vectors, shuttle vectors, BACs and YACs	
Choice of hosts, Methods for transferring recombinant DNA to host cells (Transfer	ormation
and Transfection)	
NBT 32.2: Detection and Characterization of Transformants	(05L)
Screening and selection for transformants: Hybridizations- colony, Southern, N	orthern,
Western Detection (radioactive and non-radioactive procedures)	

Western, Detection (radioactive and non-radioactive procedures). DNA sequencing techniques including automated DNA sequencing. Site-directed mutagenesis.

NBT 32.3: Expression Systems	(05L)
Various expression vectors in bacteria and eukaryotes.	
Choice of appropriate hosts, Induced expression.	
Chimeric constructs, Expression of industrially important products.	

# NBT 32.4: Applications of Genetic Engineering techniques(05L)

# **NBT 33: NANOBIOTECHNOLOGY**

#### **NBT33.1: Properties and Characterizations:**

Optical (UV-Vis/Fluorescence) – 3 Lectures X-ray diffraction – 3 Lectures Imaging and size (Electron microscopy, light scattering, Zetapotential)-4 Lectures Surface and composition (ECSA, EDAX, AFM/STM etc) – 3 Lectures Vibrational (FT-IR and RAMAN), SERS -3 Lectures Magnetic, Electrical and Electrochemical – 3 Lectures

# NBT33.2: Applications of Nano-Materials in Biosystems:

Proteins - Lipids - RNA and DNA Protein Targeting - Small Molecule/Nanomaterial - Protein Interactions Nanomaterial-Cell interactions-Manifestations of Surface Modification (Polyvalency)

#### **NBT33.3: Nanomaterials and Diagnostics/Drug Delivery and Therapeutics** MRI, Imaging

Surface Modified Nanoparticles MEMS/NEMS based on Nanomaterials Peptide/DNA Coupled Nanoparticles Lipid Nanoparticles For Drug Delivery Inorganic Nanoparticles For Drug Delivery Metal/Metal Oxide Nanoparticles (antibacterial/anti fungal/anti viral) Anisotropic and Magnetic Particles (Hyperthermia)

# NBT33.4: Nanomaterials and Toxicity Evaluation

Cyto-toxicity, Geno-toxicity In vivo tests/assays etc.

# NBT 34: BIOCHEMICAL ENGINEERING

# NBT 34.1: Theory and Design of Bioreactiors

Concepts of basic modes of fermentation – Batch, Fed batch and Continuous Types of Bioreactors Mathematical aspects of enzyme reactions and bio-reactors. Simulation of reaction kinetics and reactors. Construction and design of bioreactors. Scaling up of processes.

# **NBT 34.2: Transport and Process Control**

Transport phenomena in biochemical engineering: mass transfer, heat transfer, mixing, rheology

# NBT 35 PLEURIPOTENT CELL TECHNOLOGIES AND REPRODUCTION

# NBT 35.1: Cells of reproduction and early development(15L)Gamets and fertilizationEarly development: Metabolic activation cytoplasmic rearrangement embryonic

Early development: Metabolic activation, cytoplasmic rearrangement, embryonic induction, cell lineages, pattern formation.

Molecular basis of development in animal and plants : a) homeobox gene expression and pattern formation b) DNA methylation and epigenetic gene regulation

NBT 35.2: Stem cell concept and technologies	(15L)
Committed cells and late development	
Stem cells, Embryonic stem cells, differentiation.	
ES cell technologies, Transgenics and knock outs.	
Concept of Cell replacement therapy and regenerative medicine	
Human cloning and Bioethics.	
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Page 16

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# SEMESTER IV

# **NBT 41: RESEARCH PROJECT (NANOBIOTECHNOLOGY)**

# DETAILED SYLLABUS OF M.Sc. BIOTECHNOLOGY, APRIL 2004

# PRACTICALS

# SEMESTER I:

# **NBT 14 BIOLOGICAL CHEMISTRY**

- 1. Introduction to measurements: balances and pipetting. Preparation of solutions of given normality and its standardisation.
- 2. pH meter: buffering capacity of a buffer, Indicators. To determine the pKa value and hence the dissociation constant of a given acid by using pH meter. Determination of the amount of a  $\alpha$ -amino nitrogen by formal titration method
- 3. Colorimetry: To determine the dissociation constant of a given indicator colorimetrically and to prepare the buffer solutions in the pH range of 2.2 to 8.0
- 4. Potentiometry: Redox potential of Fe  $^{+2}$  and Fe $^{+3}$
- 5. Conductivity meter: To determine the cell constant of 0.1 M KCl and perform the titration of strong acid vs strong base and to find out equivalent conductance of the salt formed.
- 6. Quality of water.
- 7. Viscometry: a) Radius determination: Glycerol molecule b) Molecular weight determination Proteins and DNA.
- 8. Radioactivity: To determine the half life, average life and decay constant of a given radioactive isotope.
- 9. Thin layer chromatography : lipids, mixture of dyes.
- 10. Spectrophotometry: Double beam and recording spectrophotometry, Derivatives and difference spectra: Indicators, cytochromes, haemoglobin.
- 11. ELISA Reader and spectrophotometer: Estimation of protein by Lowry, Biuret and Bradford methods, Analysis of Standard curves, linear regression and assessment of ranges and reliability.
- 12. Spectrophotometry: To find out absorption spectrum of a given chromophore and/or oxidised and reduced forms (sodium nitrite and borohydrate). a) Haemoglobin and Methaemoglobin b) NAD and NADH.
- 13. Enzyme assays  $\beta$ -galactosidase, time, temperature, protein concentration and cofactors.

- 14. LDH : Km and Vmax, Various kinetic plots.
- 15. Use of computer packages for parametric and non-parametric methods and non-linear regression.
- 16. Liposome : Preparation of Uni and multilamellar vesicles, use of sonication. Light scatter and Turbidity correction of multi and unilamellar vesicles.
- 17. Introduction to centrifugation.
- 18. Use of computer for data analysis and spectral changes, difference and derivative spectra.
- 19. Polyacrylamide gel electrophoresis : Native gel.
- 20. Isozymes and activity staining.
- 21. SDS-PAGE of proteins.

# NBT 15 CELL BIOLOGY

- 1. Microscopy: a) simple, b) compound c) phase contrast microscopes.
- 2. Cell Division: Mitosis and Meiosis.
- **3.** Permanent Slides: Polytene chromosomes, grass hopper spermatids and chromosomes.
- 4. Cell motility and flagellar staining, Photography and videotaping (motility, morphometry).
- 5. Micrometry: Calibration of stage and ocular micrometer and measurement of the given biological sample Haemocytometer: calibration and measurement of biological samples.
- 6. Electron microscopy : Demonstration and good photographs for interpretation.
- 7. Blood cells: WBC: types of polymorphs.
- 8. Demonstration of animal handling for experimental purposes: cervical dislocation, dissection of rat: cardiac puncture, blood sample preparation and its handling, Osmotic fragility of RBC's.
- 9. Density gradient: sucrose/percoll.

10. Embryo development: permanent mounts.

- 11. Developmental studies: chick: developmental stages and Gastrulation.
- 12. Programmed cell death during embryonic development.
- 13. Cell types of plants maceration of various tissue explants and identification of xylem vessels, trachieds, stomata, root hair etc.
- 14. Isolation of chloroplast.
- 15. Chlorophyll estimation: spectrum and light scatter turbidity correction in chloroplasts.

# **NBT 16 QUANTITATIVE METHODS**

#### **NBT 16.1 BIOSTATISTICS**

- 1. Descriptive statistics: systematic tabular summarisation of data (before analysis), measures of central tendency, measures of dispersion, measures of skewness (using calculators).
- 2. Correlations (product-moment coefficient, Spearman's rank coefficient) and regression (linear regression, curve fitting).
- 3. Data presentation (tables/figures): 1-D and 2-D bar charts, pie diagrams, graphs (using computer software packages).
- 4. Statistical distributions: fitting discrete uniform, binomial, Poisson and normal probability distributions to given data.
- 5. Testing of hypotheses: Tests of significance (mean, Standard Deviation, Correlation coefficient).
- 6. Chi-squared test for goodness-of-fit, test for independence of attributes, non parametric tests (run test) using calculators and printed tables and computers.
- 7. Sampling (drawing random samples using random number, tables, chits).
- 8. Computer programs for random number generation), Design of experiments, ANOVA (one-way and two-way).

# **NBT 16.2 BIOMATHEMATICS**

- 1. Ratio and proportion.
- 2. Factorial notation, permutation and combination.
- 3. System of algebric equations (verification of consistency and finding solutions).

- 4. Round-off error and how to minimise it while evaluating algebraic expression (can be included in computer course).
- 5. Functions and their graphs, Concept of limit of function.
- 6. Computing area under a curve of regular/arbitrary shape.
- 7. Trigonometry.
- 8. Calculus: i) Differentiation and higher order differentials ii) Application of derivatives iii) Integration: definite and indefinite iv) Differential equations v) Application of integration.

# NBT 16.3 COMPUTER

- 1. Introduction computers and its peripherals.
- 2. Maintenance of computers using antivirus programs, formatting computers.
- 3. Handling of computers files and folders, Use of Dos commands.
- 4. Introduction to various packages and softwares.
- 5. Use of MS Dos commands, Windows: Word and Excel.
- 6. Introduction to programming languages.
- 7. Basic C programming.
- 8. Advanced C programming.

# **SEMESTER II:**

# NBT 26 MOLECULAR BIOLOGY

- 1. Isolation of chromatin: Determination of mononucleosomal size.
- 2. Chromatin gel electrophoresis.
- 3. Endonuclease digestion of nuclei.
- 4. Analysis of DNA fragments by agarose electrophoresis.
- 5. Thermal melting of DNA.
- 6. Isolation and analysis of nuclear DNA.

- 7. Restriction endonuclease digestions of nuclear DNA.
- 8. Isolation of mitochondrial DNA, agarose gel electrophoresis and detection of modifications.
- 9. Restriction endonuclease digestion and separation of fragment by gel chromatography, density gradient centrifugation and base composition analysis of DNA.
- 10. Isolation plant genomic DNA, agarose gel electrophoresis and detection of plant DNA.
- 11. Restriction endonuclease digestions of plant genomic DNA.
- 12. Isolation of bacterial DNA.
- 13. Agarose gel electrophoresis and detection of bacterial DNA.
- 14. Restriction endonuclease digestions of bacterial DNA.
- 15. Transformation.
- 16. Preparation of beads and column packing for cationic exchange chromatography.
- 17. To find out the capacity and nature of the cationic exchange resin.
- 18. Preparation of beads and column packing for anionic exchange chromatography.
- 19. To find out the capacity and nature of the anionic exchange resin.
- 20. Gel filtration chromatography: Sephadex, Sepharose.

# **NBT 27 GENETICS**

- 1. Mutants of Drosophila, mono and dihybrid crosses in Drosophila Mendel's laws of genetics Sex linkage in Drosophila.
- 2. Sex linked lethals in Drosophila.
- 3. Dominant lethals in Drosophila.
- 4. Sister chromatid exchange in CHO cell line : Control and EMS treated.
- 5. Use of Drosophila as a model system in genetics: morphology, life history, mutants, culture, sexing pupae for setting up crosses etc.
- 6. Study of autosomal gene heritance, Study of sex linked gene inheritance.

- 7. Estimating gene frequencies in human population, estimation of heterozygotes frequencies, Pedegree analysis, analysis of human karyotes, chromosomal aberrations.
- 8. Isolation/identification of auxotroph mutants in bacteria, Recombination in Bacteria.
- 9. Micronucleus test for detecting genotoxins, study of sister chromatid exchange for genotoxicity study.
- 10. AME's test for screening genotoxins.

# NBT 28 MICROBIOLOGY & IMMUNOLOGY

# NBT 28.1 MICROBIOLOGY

- 1. Cleanliness, media preparation, sterilisation, culturing methods, dilution technique, and isolation of pure culture- techniques.
- 2. Staining techniques in microbiology i) simple staining ii) negative staining iii) positive staining iv) spore staining v) capsule staining and identification.
- 3. Culture characteristics of microbes, identification of unknown bacteria by biochemical tests.
- 4. Bacterial growth curve serial dilution plating and turbidity measurement.
- 5. Competent cell preparation, replica plating.
- 6. Extracellular enzymatic activities of microbes, immobilization of *Saccharomyces cerevisiae* and alcohol.
- 7. Standard qualitative analysis of water.
- 8. Antibiotic sensitivity test, LD50, Potency of drug/antibiotics and biotransformations.

# NBT 28.2 IMMUNOLOGY

- 1. Immunodiffusion.
- 2. Rocket immunoelectrophoresis.
- 3. Immunoelectrophoresis.
- 4. Western blotting.
- 5. Interferon induction in cells isolation and assay.

- 6. Development of monoclonal antibodies by hybridoma technology.
- 7. Production of polyclonal antibodies and testing-immunodiffusion, immunoelectrophoresis.
- 8. Crossed antigen-antibody electrophoresis.
- 9. Radioimmunoassay.
- 10. Immunofluorescence.
- 11. Agglutination, rosette-formation, complement fixation.
- 12. Antigen-induced T cell proliferation, Generation of cytotoxic T lymphocytes.

# **NBT 29 BIOINFORMATICS**

1. A guided tour of NCBI/EBI : Data acess – standard search engines : data retrievals tools – Entrez, DBGET and SRS (sequence retrieval systems); software for data building; submission of new revised data.

2. Sequence homology as product of molecular evolution, sequence similarity searches, sequence alignment-global, local, end free-space; measurement of sequence similarity, similarity and homology.

- 3. Multiple sequence alignment
- 4. Phylogeny reconstruction, PHYLIP package

# **SEMESTER III:**

# NBT 36 ANIMAL TISSUE CULTURE

# NBT 36.1: Tissue Culture Techniques

- 1. Acquaintance with tissue culture laboratory, Culture place: culture cubicals P1 to P4; Laminar flowsystem.
- 2. Preparatory techniques: Washing of glassware, dry and steam sterilisation. Maintenance of aseptic conditions, Sterilisation techniques, Preparation of culture media, Media preparation: Filter sterilisation, Sterility tests, and media storage. Serum inactivation.
- 3. Short term cultures. a. Primary culture of cells b. Organ culture.
- 4. Staining of cell cultures and observations under microscope.

# NBT 36.2: Animal Tissue Culture Techniques

- 1. Growth studies. Cell count, protein estimation, mitotic index.
- 2. Development and maintenance of a cell line.
- 3. Karyotyping.
- 4. Virus propogation in cells, cytopathogenic response of cells to viruses.
- 5. In vitro assay of drugs, predictive test for anticancer drugs.
- 6. Staining and screening of cells /sera for mycoplasma, viruses.
- 7. Cell cloning by single cell dilution method, Freeze storing and revival of cultured cells.
- 8. Clonogenic assay, Cell-cell interaction: Co-culture of normal and mutant cells, cell cloning by single cell dilution method.
- 9. Cell synchronization (determination of mitotic index and cell cycle time), LDH isozyme analysis of the given cell lines.
- 10. Purification of a product secreted by a functional cell line, Estimation of hormones secreted by a hormone secreting cell line.
- 11. Cell hybridization.
- 12. Immunohistochemical staining (oncogene expression).
- 13. Transplantations: tumors, organs, cells.

# **NBT 37 GENETIC ENGINEERING**

- 1. Isolation of plasmid DNA- i) minipreparation ii) large scale isolation.
- 2. In vitro DNA ligation, transformation of E.coli.
- 3. Characterisation of transformants: DNA gel electrophoresis, Restriction map analysis.
- 4. Southern blot analysis.
- 5. Isolation of cytoplasmic RNA.
- 6. Separation of poly A<sup>+</sup> RNA on oligo-dT column.
- 7. Electrophoresis of RNA on denaturing gels.

BOS, Biotechnology, University of Pune, May, 2012

- 8. Northern and dot blotting technique.
- 9. cDNA synthesis and cloning.
- 10. In situ detection of RNA in embryos/tissue.
- 11. PCR/ RT-PCR technique.
- 12. Sequencing and computer analysis.
- 13. In vitro translation.

# **NBT 38 : SEMINAR**

Each student will have to give 3-4 seminars on important recent scientific articles published in prestigious scientific journals.

# **SEMESTER IV:**

# NBT 41: RESEARCH PROJECT

Each student will have to do a research project on a topic in Nanobiotechnology.