<table>
<tr><td>1</td><td>Name of Course</td><td>C. C. IN STEM CELL AND TISSUE ENGINEERING
(W.E.F. 2015-16)</td></tr>
<tr><td>2</td><td>Course Code</td><td>201122</td></tr>
<tr><td>3</td><td>Max.No.of Students Per Batch</td><td>25 Students</td></tr>
<tr><td>4</td><td>Duration</td><td>6 Month</td></tr>
<tr><td>5</td><td>Type</td><td>Part Time</td></tr>
<tr><td>6</td><td>No.Of Days / Week</td><td>6 Days</td></tr>
<tr><td>7</td><td>No.Of Hours /Days</td><td>4 Hrs</td></tr>
<tr><td>8</td><td>Space Required</td><td>Workshop = 400 Sq feet
Class Room = 200 Sq feet
TOTAL = 600 Sq feet</td></tr>
<tr><td>9</td><td>Minimum Entry Qualification</td><td>BSC (Biology)</td></tr>
<tr><td>10</td><td>Objective Of Course</td><td>To provide systematic training about stem cells and tissue engineering</td></tr>
<tr><td>11</td><td>Employment Opportunity</td><td>Job opportunities in hospitals, research institutes etc</td></tr>
<tr><td>12</td><td>Teacher’s Qualification</td><td>Diploma in Concern Field.</td></tr>
<tr><td>13</td><td>Training System</td><td>Training System Per Week
<table>
<tr><td>Theory</td><td>Practical</td><td>Total</td></tr>
<tr><td>6 Hours</td><td>18 Hours</td><td>24 Hours</td></tr>
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<tr><td>14</td><td>Exam. System</td><td>
<table>
<tr><th>Sr. No.</th><th>Paper Code</th><th>Name of Subject</th><th>TH/PR</th><th>Hours</th><th>Max. Marks</th><th>Min. Marks</th></tr>
<tr><td>1</td><td>20112211</td><td>Cell Culture & Cell Technologies</td><td>TH-I</td><td>3 hrs</td><td>100</td><td>35</td></tr>
<tr><td>2</td><td>20112212</td><td>Tissue Engineering</td><td>TH-II</td><td>3 hrs</td><td>100</td><td>35</td></tr>
<tr><td>3</td><td>20112221</td><td>Cell Culture & Cell Technologies</td><td>PR-I</td><td>3 hrs</td><td>100</td><td>50</td></tr>
<tr><td>4</td><td>20112222</td><td>Tissue Engineering</td><td>PR-II</td><td>3 hrs</td><td>100</td><td>50</td></tr>
<tr><td></td><td></td><td>Total</td><td></td><td></td><td>400</td><td>170</td></tr>
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Theory - I - Cell Culture & Cell Technologies

1. Introduction to human body & tissues, structure and functions of skin, (Layers and accessory organs].
2. Skeletal, muscular, cardiovascular, lymphatic and immune system.
1. Digestive, excretory & respiratory system
2. Nervous, endocrine & reproductive system.

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Histology

1. History and scope of Animal Cell Culture
2. Cell culture media and Growth factors**:** Metabolic requirements, equipments, aseptic technique, cryopreservation, cytotoxic assays, viability assays physicochemical properties, balanced salt solution, complete media, serum, selection of medium & serum, condition media and other supplements, determination of cell growth and kinetics,
3. Biology of Cultured Cell**:** The cell culture laboratory, GLP in cell culture, cell culture environment, adhesions, proliferation differentiation & dedifferentiation, cell lines evolution & origin.
4. Primary Cell cultures and established cell lines**:** Types, isolation procedures, tissue disaggregation, establishment, characteristics, and enrichment of the primary cells, secondary cell culture, commonly used cell lines, cell fusion, production of monoclonal antibodies, scale up methods for propagation of anchorage dependent and suspension cultures.
5. Genetic modifications & applications in animal cells**;** *in vitro* & *in vivo* transformation along with their applications, *in vitro* fertilization and embryo transfer technology, introduction to Stem Cell Biology,
6. Quality control of cell culture**:** Transportation of cell line, quarantine and initial cell handling, microbial quality control, authentication, regulatory aspects

Genomics & Proteomics

1. Chromosome**:** Chromatin structure, nucleosome, karyotype and chromosome nomenclature, structure, chromosomal aberrations, disorders, chromosomal mapping, gene mapping, construction of Genetic map
2. Clinical cytogenetics**:** Chromosomal abnormalities, autosomal and sex chromosomal abnormalities, chromosomal markers.
3**.** Inheritance of traits**:** Mendelian laws of Inheritance, allelic variance cytoplasmic inheritance, autosomal recessive inheritance, autosomal domoninant inheritance, X - linked dominant inheritance, X-linked recessive inheritance
4. Molecular defects affecting metabolic pathways**:** Overview of structure, function,
5 Metabolism and related diseases of amino acid carbohydrates, fats & lipids, Nucleic acids with special emphasis on inborn errors of metabolism & degenerative diseases.

1. Cell Biology**:** Prokaryotic cell, eukaryotic cell and their organelles, cytoskeleton and cell motility, cell interactions; extracellular matrix, adhesive proteins, cell-cell, cell matrix interactions, and transport across membranes
2. Eukaryotic Cell cycle & check points, Cell growth regulation & cell cycle progression, Programmed cell death
3. Molecular Biology: Central Dogma, functional definition of gene, physicochemical Properties of Nucleic Acids, Structure and types of nucleic acids (DNA &RNA),
4. Molecular processes and their mechanisms (Replication, Transcription, Translation), Protein sorting and transport, Gene regulatory processes, Mobile genetic elements, transposons & retrotransposons.

Cell and Molecular Techniques

1. **Analytical techniques:** Spectrophotometery, spectrofluorometry, chemiluminescence, chromatography and electrophoresis techniques radio-isotopic techniques, cell fractionation methods and centrifugation, density gradient centrifugation, ultracentrifugation.
2. **Techniques in cell biology :** Microscopy: Principle and applications of light, phase contrast, fluorescence, confocal, electron microscopy and atomic force microscopy.
3. **Techniques in molecular biology** : DNA Amplification- PCR, Real Time PCR, qPCR, RT-PCR Restriction digestion, Cloning, DNA Sequencing, Northern, Southern & Western blotting, dot blot, autoradiography, Genomic and cDNA library construction,, RFLP, RAPD, AFLP, mutagenesis, whole genome sequencing,
4. Electrophoretic mobility shift assay, CHIP assay, RNAi technique, phage display. Flow-cytometry, immunohistochemistry and immunocytometry

Stem Cell Biology

1. Basic biology of stem cells; Types & sources of stem cell with characteristics: embryonic, adult, haematopoetic, fetal, cord blood, placenta, bone marrow, primordial germ cells, cancer stem cells, induced pleuripotent stem cells.
2. Stem cell Characterizations: isolation & characterizations, markers & their identification, growth factor requirements and their maintenance in culture. feeder and feeder free cultures.
3. Cell cycle regulators in stem cells
4. Molecular basis of stem cell renewal and differentiation, Metaplasia and transdifferentiation.
5. Molecular basis of pleuripotency and stem cell niche
6. Nanoscale approaches to stem cell culture and applications of stem cells: Neurodegenrative diseases, spinal cord injury, heart disease, diabetes, burns and skin ulcers, muscular dystrophy, orthopedic applications, Stem cells and Gene therapy.

Practical - I - Cell Culture & Cell Technologies

Hemocytometer exercise, Practice handling new equipment and supplies. Inverted phase microscopes, trypan blue dye exclusion, incubators and ancillary equipment.
Aseptically prepare media in biosafety hood.
Cryopreservation of cells and retrieval of cells.
Cell passaging
Handling of secondary animal cell culture.
Identification of contaminants in animal cell culture
Subculturing of continuous cell line growing in monolayer and suspension
Construction and analysis of Growth curve
Isolation of primary cell culture.

Preparation of metaphase Chromosome.
Karyotyping
Banding Pattern (G-banding).
Quantitation of cellular macromolecules (DNA, RNA and Protein)
Preparation of blood smear for study of sex chromatin.
Study of human sex chromatin

To perform the absorption spectra
To study living matter under Phase contrast microscope.
To study the parts of compound microscope and phase contrast microscope and its maintenance.
Chromatographic techniques.
Radio isotopic counting techniques
Density gradient separation of human Blood Cells.
To perform Native PAGE for proteins.
To perform SDS-PAGE for separation of protein.
DNA electrophoresis

Restriction digestion
PCR analysis.
Gene cloning & subcloning

Western blotting
GST Pull down assays
Mass spectroscopic data analysis
FACS Analysis

Tissue processing
Fixation
Parafffin Embedding
Section cutting
Mounting techniques
Staining of tissue sections
Microscopical examination

Immunology & Immunogenetics

1. Overview and Generation of immune responses**:** History, innate and adaptive immunity,
Antigens: Immunogens, epitopes, haptens, adjuvants, antibodies: basic and fine structure, antibody classes and effector functions, antigen antibody interactions,. major histocompatiblity complex: MHC molecules and genes, cellular distribution, genetic organization and inheritance, MHC and disease susceptibility.
2. Cells and organs of immune system: Haematopoiesis, Haematopoietic stem cells, lymphoid cells: B and T cells, NK cells, Mononuclear phagocytes, granulocytic cells, dendritic cells, Primary lymphoid organs: Thymus, bone marrow, lymphatic system and Secondary lymphoid organs: Lymph nodes, spleen, MALT, CALT.
3. Immune effector mechanisms: Cytokines: properties, receptors, antagonists, diseases, therapies, Hypersensitivity: TypeI, II, III, and IV, Cell mediated cytotoxic responses: Effector T cells, Cytotoxic T cells, NK cells, ADCC, Complement system: Functions, components, different pathways, regulation and biological consequences.
4. Immune response in health and disease: Tolerance, Autoimmune diseases: organ specific and systemic, Animal models, Transplantation: Graft rejection, Immuno suppressive therapy,
Cancer: Tumor antigens, Cancer immunotherapy.

1. Histology of Lymphoid Organs.
2. Blood film preparation and identification of WBC from normal; and infected animals.
3. Blood group typing.
4. Demonstration of antigen- antibody interaction by oucterlony method
5. Seperation of WBC from blood by Density gradient centrifugation.
6. Detection of live/dead WBC using Acridine Orange/Ethidium Bromide staining.
7. To perform indirect fluorescent antibody test.

Stem Cell Biology

1. Preparation of feeder cell culture
2. Culturing of Stem cell line
3. Isolation of cells from blastocyst
4. Isolation of placenta
5. Isolation of cells from placenta
6. Growing mesenchymal stem cell
7. Differentiation of mesenchymal stem cells

Theory - II - Tissue Engineering-Biomaterials/Biopolymers

1. Introduction to biomaterials, Characterization of materials; mechanical properties; thermal properties, surface properties and adhesion, Biological tolerence.
2. Metals, ceramics and polymers, properties and Uses
3. Biologically Active scaffold based on tissue engineering, polysaccharide scaffold in tissue engineering, materials, modification and properties of scaffolds. tissue engineering applications
4. Applications of nanotechnology in tissue engineering. tissue engineering approaches to stem cell-based therapies

Translational & Ethics

1. Tissue engineering using embryonic, mesenchymal and adult stem cells, Soft tissue grafts, orthopedic tissue engineering engineered skin, skeletal tissue, neuronal tissue, intestine engineering etc.
2. Engineering embryonic stem cells with recombinase system, engineered tissues & regenerative medicine, Functional tissue engineering, Monitoring of engineered tissues.
3. Regulations & Ethics: Ethics in use of stem cell, regulatory bodies for use of material for human need, commercial developments and stem cell based products, bio- vigilance.
4. Patents and law: Introduction to the basic principles of the law of patents origin and function of the patent system, the nature of patents as property and legal instruments, comparisons with other forms of intellectual property, subject matter eligible for patenting, conditions for patent-ability of an invention and the disclosure requirements for a patent application, regulatory challenges in engineered tissue and regenerative medicine.

Stem Cell Signal Transduction and Epigenetic Mechanisms

1. Introduction to cell signaling: Crosstalk of extracellular signal to the physiological response, cell surface receptors, conserved components of signaling,
2. G-Protein signaling; Heterotrimeric and monomeric G-proteins, their types and G-protein coupled receptors (GPCR) regulating activities of adenylate cyclase, phosphodiesterase, ion channels, phopspholipase and calcium homeostasis. receptor switching
3. Genomic response through Signaling; GPCR, TGFβ, Cytokine signaling, Receptor tyrosine kinase, RAS/RAF and MAP kinase pathway, Wnt signaling classical and non-classical. Phosphoinositide signaling.
4. Signaling based on protein cleavage pathways (Hedge hog, Notch signaling),
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5. Role of signal transduction pathways in stem cell renewal and differentiation.
6. Epigenetic mechanisms: Role in development and cell remembrance, histone modifications, acetylation, methylation *etc.*, factors effecting these modifications, reprogramming, cellular response, DNA methylations and development, functional role of epigenetic in development.

Practical - II - Tissue Engineering

1) Isolation of collagen
2) Use of collagen scaffold for stem cell growth
3) Preparation of mixed scaffolds
4) SEM of scaffold
1. Preparation of synthetic scaffolds
2. Use of scaffolds for stem cell differentiation.
3. Scaffolds for bone
4. Tissue engineering using commercially available stem cell lines

Stem Cell Signal Transduction and Epigenetic Mechanisms

1. Demonstration of Calcium mediated signaling
2. Signal mediated activation of transcription factors.
3. Demonstration of wnt canonical pathway
4. GSK 3beta kinase pathway and stem cell differentiation

Tool and Equipment should be available in Institute

This section lists the minimal equipment required for basic culture and characterization of hPSCs. A given program may require specialized culture equipment, such as incubators that allow culture in low oxygen tension, or characterization and analysis equipment. This section lists the equipment required to establish a standard hPSC culture laboratory.

Name of Tool & Equipments
1.1. Tissue Culture Laboratory 1. Class II Biosafety Cabinet (BSC) 2. CO_2 incubator 3. Pipettors. 4. Vacuum flask/aspiration device. 5. Water bath (37°C). 6. Low-speed centrifuge (clinical grade, for spinning cells).
1.2. Microscopy 1. Phase–contrast microscope. 2. Dissecting microscope.

1.3. Storage

1. Cabinets and shelves for the storage of tissue culture supplies
2. Refrigerator (4°C).
3. Freezer (−20°C, nondefrosting).
4. Low-temperature freezer (−70 to −85°C).
5. Cryogenic freezer (storage below −140°C, usually liquid nitrogen).

1.4. Molecular Biology Laboratory/Quality Control Laboratory

1. RT-PCR.
2. Flow cytometer (might be in a Core facility).
3. Fluorescence microscope (might be located in a Microscopy Core).
4. Confocal microscope (might be located in a Microscopy Core).

1.5. Quarantine Laboratory

1. Class II Biosafety cabinet..
2. CO_2 incubator
3. Phase–contrast microscope.
4. Water bath (37°C).
5. Low-speed centrifuge (clinical grade, for spinning cells).
6. Pipettors.
7. Aspiration/vacuum flask.
8. Sink.

1.6. Additional Access to Common Equipment or Core Facilities

1. Microscopy.
2. Flow cytometry.
3. Microarray gene expression.
4. Genomics.
5. Proteomics.
6. Virus production.
7. Vivarium.

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