**Course Syllabus:**
**Stem Cell Techniques: Differentiation into Neural Cell Types**

**Course Overview:** This four-module course explores the principles and techniques of neural progenitor cell (NPC) differentiation using stem cell technologies protocol and reagents. Students will gain a comprehensive understanding of the molecular and cellular processes involved in directing stem cells towards neural lineages. Through a combination of lectures, laboratory sessions, and interactive discussions, participants will learn the latest advancements in the field and gain hands-on experience in differentiating neural progenitor cells from various stem cell sources. Objectives include:

- To provide a foundational understanding of neural development and neural progenitor cell biology.
- To introduce students to the principles of stem cell technologies and their application in neural differentiation.
- To explore advanced techniques for generating and characterizing neural progenitor cells.
- To teach participants how to design and optimize differentiation protocols for specific neural lineages.
- To familiarize students with cutting-edge research in neural progenitor cell differentiation and its potential applications.

**Course Duration:**
Modules 1-3: Three days , Monday to Wednesday, two hours of class time and a one-hour discussion 1:00 pm to 4:00 pm,
Module 4: Four days of laboratory training, Monday through Thursday, 1:00 pm to 5:00 pm

**Note:** Enrollment may be limited to a max of 8 attendees per class to ensure individualized attention and optimal learning outcomes. The syllabus is designed to provide a comprehensive overview of the course content and may be adjusted based on participant needs and advancements in the field.

**Prerequisites:** Participants must complete the web-based Introduction to Stem Cell Laboratory Techniques. Participants should have a solid foundation in cell biology, molecular biology, and basic cell culture techniques. Familiarity with stem cell biology concepts is beneficial but not required.

**Module 1 Overview (Introduction to Neural Development and Progenitor Cells from Pluripotent Stem Cells):** This module will provide an overview of the neural development stage from embryogenesis to neural lineage commitment. Participants will learn how the neural development stages are recapitulated in vitro using iPSC.

**Day 1: Introduction to Neural Development and Progenitor Cells from Pluripotent Stem Cells** Overview of neural development stages

- Types and functions of neural progenitor cells
- Signaling pathways regulating neural differentiation
- Types of stem cells: embryonic, induced pluripotent, and adult stem cells
- Reprogramming techniques for neural lineage commitment
- Considerations for selecting stem cell sources

**Module 2 Overview (Neural Induction Commitment and Differentiation into Specific Neural Lineages):** This module will review mechanisms and pathways for how specific brain cells (different types of neurons, astrocyte, and oligodentrocytes) are formed in the in vitro differentiation.

**Day 1: Neural Induction Commitment and Differentiation Into Specific Neural Lineages**

- Factors influencing neural induction
- Generation of neural rosettes and neural tube structures
- Molecular markers for neural progenitor cell identification
- Differentiation into neurons, astrocytes, and oligodendrocytes
- Neurotransmitter phenotypes and electrophysiological properties
- Mimicking brain regions using tailored differentiation protocols

**Module 3 Overview (Characterization of Neural Progenitor Cells and Advances in Neural Progenitor Cell Differentiation):** This module will introduce methods of validation for neural cell differentiation and usage of the in vitro differentiation method in disease modeling.

**Day 1: Characterization of Neural Progenitor Cells and Advances in Neural Progenitor Cell Differentiation**

- Immunocytochemistry and molecular techniques for characterization
- Functional assays to assess neural differentiation success
- Quality control and validation of neural progenitor cells
- 3D culture systems and organoids
- CRISPR/Cas9-mediated genetic modifications
- Disease modeling and drug screening using NPC-derived cells

**Module 4 Overview (Practical Techniques for obtaining NPC and forebrain neurons from Pluripotent Stem Cells (iPSCs)):** This is a hands-on module that occurs in the laboratory and explores key stages needed for the generation of human Neural Progenitor Cells (NPC) using the embryoid bodies (EBs) intermediate. NPC will be further differentiated into Forebrain neurons. The protocol is based on the Stem Cell Technologies protocol and reagents.

**Day 1: EB Formation Laboratory Class**

- Evaluation of the hPSCs confluency and differentiation status
- Harvesting of the cells and cell count
- Transfer cells in aggrewells

**Day 2: Neural Rosette Evaluation and Selection**

- Identification of neural rosette, imaging, and scoring
- Neural rosette selection and replating

**Day 3: Maintenance, Characterization and Passaging of NPC**

- Evaluation of NPC shape and Imaging
- Passaging for expansion of NPC
- Preparing immunostaining with NPC markers for characterization
- Imaging of stained cells

**Day 4: Forebrain neuron differentiation**

- Harvesting and seeding NPC for Forebrain differentiation
- Characterization of differentiated forebrain neuron by using specific antibodies

- Imaging of stained cells

**Assessment Methods:**

- Attendance and participation in lectures and discussions
- Laboratory performance and reports