University of California
Department of Bioengineering & Molecular & Cell Biology

Course Number: BioE C218 & MCB C237
Course Title: Stem Cells and Directed Organogenesis
Offered: Spring
Instructor: Irina and Michael Conboy, conboymj@berkeley.edu, 408-621-2063
Units: 3 units.
Course Format: six lab hours plus 1 hour of discussion per week
Prerequisites: B.A./B.S. in the sciences or Consent of Instructor
Grading: Letter & S/U
May be repeated for credit: No
Course that may restrict credit and how much: n/a
Estimated Additional Number of Required Hours of Student work per week: 3
Time and Location: Mondays and Wednesdays **2-5 (may change to 1-4!)** in Stanley B144 Teaching Lab; Office hours Fridays 11 am., Stanley B108 or by appointment.

Description:

This course will teach the main concepts and key methods of human embryonic stem cell (hESC) and adult stem cell derivation, propagation and characterization, focusing on currently developing stem cell technologies.

The course will teach the in vitro techniques used in stem cell research and stem cell engineering. Specific examples include a choice of substrates and biomaterials for in vitro expansion and sustained pluripotency of ESC; controlled and normalized ecto-, meso- and endoderm formation in 3D embryoid bodies, directed tissue specific differentiation of stem cells and conventional as well as single-cell microfluidics-based analysis of hESC gene expression. Comparison between hESCs and adult organ stem cells, e.g. muscle or neuronal stem cells, in self-renewal, pluripotency and in tissue-specific differentiation will be discussed based on the experimental evidence obtained in class.

Objectives:

The purpose of this course is to introduce the student to the technologies associated with the molecular regulation of embryonic and adult stem cells, to the methods currently suited for the derivation, propagation and characterization of stem cells and provide knowledge of current stem cell engineering. The level of course-work presupposes knowledge of fundamentals of cellular and molecular biology and of biomaterials at the senior undergraduate level, and will require competence in basic sterile cell culture technique and of handling basic micro/ molecular biology/bioengineering laboratory equipment. Through class lectures

and experimental methods of stem cell science, material science and bioengineering the student will gain a fundamental understanding of the principles and techniques guiding the current stem cell-based research and will be able to culture self-renewing and differentiating hESCs and two lineages of adult stem cells, and to conduct basic characterization of their gene expression profile. In addition, this course will aid the student in cultivating broad knowledge of the stem cell field and in learning about the interface with biomedical and translational sciences.

Overview:

This course will meet twice per week for ~180 minute experimental laboratory
and once per week for <sixty minute office hours and discussion. Students are required to attend class, participate in the experimental laboratory and discuss their results in the context of the knowledge in the field of advanced stem cell research. Students will make a paper presentation from the reading list in the clinical, life and materials science literature. Additionally, students are encouraged to seek out reference material to complement the readings assigned. Students will present as mid-terms their findings from embryonic and adults stem cell laboratory work, much like a short "lab meeting". Presentations will be of the common research lab Journal Club/Lab Meeting format and level of presentation. To encourage preparation, homework will be assigned most weeks of lab to answer a short list of relevant questions for the planned procedures. There is a Berkeley Stem Cell Center Retreat planned for a day in April at Asilomar, CA, that students are strongly encouraged to attend. The final exam is a short research proposal using stem cells to answer a question of interest to the student or of relevance to the field.

Grading:

10 points, one per homework: Homework: Before each week of lab each student will answer a short series of questions to confirm the student's understanding of the lab work ahead.

10 points, Paper Presentation: In a Journal Club format, each student will present a relevant paper, chosen from a list or by consent of the Instructor.

20 points: Midterm 1: Presentation of Findings for hESC work: Student lab partners will make a short oral presentation of the findings from the recent labs, including problems and pitfalls.

20 points: Midterm 2: Presentation of Findings for adult stem cell work: Student lab partners will make a short oral presentation of the findings from the recent labs, including problems and pitfalls.

10 points: Laboratory Performance: preparation/understanding of protocol,

notebook keeping, development of laboratory skills, clean up. The student's performance in these areas will be reviewed at or before the midterm with suggestions for improvement, but graded only on the performance after the midterm, after improvements may be implemented.

30 points: Final Research Proposal: Each student will submit a short research proposal using stem cells to answer a research question.

Total: 100 points

Daily Plans:

Week 1
Lab "0" (23 Jan. Wed.)
Introduction and overview
course requirements
grading: homework, labwork, paper presentation, result presentation, final
introductions and student projects
skill set survey
lab safety
Literature list and presentation schedule
Homework assignments
Culture feeding schedule
Discussion of growth requirements
Plan outgrowth of hESCs
Practice identifying good/bad hESC colonies
Tissue culture sterile technique skills test.

Office hours (25 Jan. Fri.)
Refresher on tissue culture sterile technique, as needed.

Week 2
Lab 1 (28 Jan. Mon.)
Homework 1 due!
Growth and culture of hESCs.
Papers: derivation of mESCs: Martin 1981; Evans 1981.

Lab 2 (30 Jan. Wed.)
Observation, scoring and weeding of regular hESC colonies.
Passage of hESC.
Paper discussion.
Paper: hESC: Thompson 1998.

Office hours (1 Feb. Fri.)

Week 3
Lab 3 (4 Feb. Mon.)
Homework 2 due!
Observation, scoring and weeding of regular hESC colonies.
Embryoid body formation.
Paper: iPS: Takahashi 2006.

Lab 4 (6 Feb. Wed.)
Re-feeding hESCs and EBs.
Paper: core pluripotency regulation: Kalmar 2009.

Office hours (8 Feb. Fri.)

Week 4
Lab 5 (11 Feb. Mon.)
Homework 3 due!
Plating of EBs for outgrowth.
Observation, scoring and weeding of regular hESC colonies.
Paper: iPS by RNA: Yakubov 2010, Warren 2011.

Lab 6 (13 Feb. Wed.)
EB scoring, picking and plating to slides.
New EB cell plating.
Paper: iPS by protein: Zhou 2009.

Office hours (15 Feb. Fri.)

Week 5
(18 Feb. Mon.)
Monday is a holiday - no lab

Lab 7 (20 Feb. Wed.)
Homework 4 due!
Immunostaining of hESC on slides.

Office hours (22 Feb. Fri.)

Week 6
Lab 8 (25 Feb. Mon.)
Homework 5 due.
Analysis of immunostained slides.

Lab 9 (27 Feb. Wed.)
Analysis of immunostained slides.
Paper: Engler 2006.

Office hours (1 Mar. Fri.)

Week 7
Lab 10 (4 Mar. Mon.)
Homework 6 due.
Harvest EB derived cells.

Lab 11 (6 Mar. Wed.)
Overview of flow cytometry.
Paper: stem cell renewal, He 2009

Office hours (8 Mar. Fri.)

Week 8
Lab 12 (11 Mar. Mon.)
Homework 7 due.
Cell cycle analysis of hESC on the flow cytometer.

Lab 13 (13 Mar. Wed.)
Cell cycle analysis of hESC on the flow cytometer.

Office hours (15 Mar. Fri.)
Help with FACS analysis.

Week 9
Lab 14 (18 Mar. Mon.)
Midterm: presentation of ESC work.

Lab 15 (20 Mar. Wed.)
Midterm: presentation of ESC work.

Office hours (22 Mar. Fri.)

Week 10
25-29 March is Spring Break! Woohoo!

Week 11
Lab 16 (1 Apr. Mon)
Discussion of Adult stem cells

Preparation of reagents
Papers: muscle stem cells: Collins 2005

Lab 17 (3 Apr. Wed.)
Mouse adult stem cells:
Dissect mouse muscle, digest and plate muscle fibers
Dissect mouse brain, digest, plate for neural stem cells

Office hours (5 Apr. Fri.)

Week 12
Lab 18 (8 Apr. Mon.)
Homework 8 due.
Plan differentiation of Adult stem cells
Collect myogenic cells and selective adherence
Paper: neuronal stem cells: Karpowics 2005.

Lab 19 (10 Apr. Wed.)
Collect neurospheres and replate
Differentiate myogenic cells.

Office hours (12 Apr. Fri.)

Week 13
Lab 20 (15 Apr. Wed.)
Homework 9 due.
Discussion of Western blotting
Feed/pass myogenic cultures
Check neurospheres

Lab 21 (17 Apr. Mon.)
Harvest differentiated and undifferentiated muscle stem cells.
Process frozen cell pellets for protein and quantify.
Papers: hematopoietic stem cells, Osawa 1986, Maximow 1909

Office hours (19 Apr. Fri.)

Week 14
(21-22 April, Sun.-Mon.)
Berkeley Stem Cell Center Retreat at Asilomar!
Attendance is encouraged but not required.

Lab 22 (24 Apr. Wed.)

Discuss Retreat talks.
Set up any biochemically defined niche experiments

Office hours (27 Apr. Fri.)

Week 15
Lab 23 (29 Apr. Mon.)
Homework 10 due.
PAGE separation of proteins and Western Blotting

Lab 23 (1 May. Wed.)
Immunodetection of Western.
Paper: hair: Hsu 2011

Office hours (4 May Fri.)

Week 16
6 May, Mon.
Midterm 2: Presentation of mouse adult stem cell findings

8 May, Wed.
Midterm 2: Presentation of mouse adult stem cell findings

Office hours (10 May Fri.)

Week 17
17 May, Friday
Final Exam Due: Research Proposal using stem cells.