Mike Dorrell: BIOLOGY 350 LAB

The laboratory component of Advanced Cell comprises approximately 15% of your overall course grade. The lab grade will be based upon an electronic lab book, group participation evaluations, and lab quizzes.

Lab Schedule

Date	Lab activity	Text	Due
9-4 (Tues)	Cryosectioning mouse tissue	Read: Cryosectioning handout 1; Quiz on handout	
9-11 (Tues)	Staining cryosectioned tissue	Quiz on handout and previous cryosectioning handout / previous work	
9-18 (Tues)	Fluorescence imaging and photomicroscopy Group 5 primary literature presentation (membranes)	Class notes on Chapters 8-9. No quiz	*HeLa (Immortal Life of Henrietta Lacks) discussion – Part 1, 'Life'*
9-25 (Tues)		Exam 1; Chapter 1,3, 8-11	
10-2 (Tues)	Artificial culturing of L-cells;	Read: Cell-culture handout1; Quiz on handout	Lab report - mouse brain sectioning (Mon night)
10-9 (Tues)	Introduction to chamber slides Group 4 primary literature presentation (cellular transport)	Read: chamber slides protocol Quiz on handout Continue growing L-cells	*HeLa discussion – Part 2, 'Death'*
10-16 (Tues)		Exam II ; Chapters 12-13 *Reminder; early next 2 weeks, you must set up the chamber slides*	
10-23 (Tues)	Fluorescent microscopy #1 Demonstrate cell counting discuss cell viability exp.	Read: Staining L-cells handout 1; Quiz on handout / previous work	
10-30 (Tues)	Cell culture viability assay / Group 3 primary literature presentation (cell signaling)	Read: cell culture viability handout Quiz on handout / previous work	
11-6 (Tues)	Fluorescent microscopy #2 stem cell discussion	Read: Staining L-cells handout 2; Quiz on handout / previous work	Stem cell homework
11-13 (Tues)	Bone marrow cell culturing, day 1 Group 2 primary literature presentation (cytoskeleton)	Read: Mouse BMCs handout-1 Quiz on handout / previous work	Lab report and E-notebook for cell culture (Fri night;11-20)
11-20 (Tues)		Exam III ; Chapters 15, 16, 19	
11-27 (Tues)	Bone marrow cell culturing; day 2	Read: Mouse BMCs handout-2 Quiz on handout / previous work	Lab report for BMCs due Fri Dec. 9
12-4 (Tues)	HeLa discussion – part 3 Group 1 primary literature presentation (cancer)	Stem cells handout / discussion assignment	*HeLa discussion – Part 3, 'Immortality'*

Attendance:

Attendance at lab sessions is required and role will be taken. Absence from a lab, results in a zero for any work done on that day. Extenuating circumstances will be considered as long as the instructor is notified up front.

Laboratory Work – Brief Introduction

There will be three main pieces of laboratory work for this course. For each of them, you will be working as part of a group of four people. Your group may split the efforts equally, but you are always expected to perform the work at least in pairs to minimize errors. **Handouts with pertinent background information and procedural details for each lab can be found on canvas.** There will be a quiz at the beginning of the lab period, which will cover the material in this introductory handout. The goal is to insure that you have put some effort into reading and digesting the handout material, so that a high quality lab experience results. At the conclusion of a body of laboratory work, you will be responsible for turning in a "summary product" for me to evaluate. ***There will be an opportunity for each student to evaluate the quality of the group experience (by evaluating yourself and other group members) as we reach major assignment "milestones" for the semester. The intent is to ensure that there is accountability within each group.***

(1) Cryosectioning and staining of mouse heart tissue (20% of lab grade)

We will be practicing an important technique used by cell biologists to study cells within a tissue system. I will provide some mouse hearts in a cryomold and groups will prepare sections of this tissue so that we can stain them for various markers of cardiac muscle and myocardial precursor cells. Your grade will be based on the success of the cryosectioning, the success of the staining, and your individual lab report.

(2) Artificial Cell Culture/Fluorescence Microscopy (40% of lab grade)

The laboratory student teams as constituted above will conduct the work involving the artificial culture of cells, and their use as subjects for microscopy. This work will stretch over several weeks. You will also be responsible for preparation of cells for use in the microscopy labs listed in this syllabus. As microscopic data accumulate (e.g. digital images) you will also be responsible for selecting a set of these and including them, with your analytical comments. I will give you a handout later on with more details on this. Grades will be based on successful maintenance of cells over the course of several weeks, success during fluorescent staining of the cultured cells, and your electronic lab notebook that you keep throughout the project.

(3) Mouse Bone Marrow Clonal Progenitor Cell assay (15% of lab grade)

You and your lab group will prepare a set of special culture dishes which will support the growth of primitive bone marrow "progenitor" cells, which are responsible for populating the marrow and peripheral blood. This is the same culture technology which is used in research to study the control of progenitor cell physiology, and to assess the growth of marrow cells from patients with hemopoietic abnormalities. As a class we will collect data from the growth of bone marrow cells under control, and an experimental treatment condition. Your grade will be slightly based on successful growth of the BMCs, quantification of the bone marrow colonies, and your electronic lab notebook (details further below).

Lab Quizes: (15% of lab grade)

There will be several short quizzes that will be given at the beginning of lab. These quizzes will cover material from the lab handout for that day to ensure that you have read the handout thoroughly and come prepared to perform the lab activities. You should come to lab prepared and knowledgeable of the experiment you are about to begin. Some of the questions will also cover activities and material from previous laboratories to ensure that you are understanding and maintaining the desired material.

The Immortal Life of Henrietta Lacks (10% of lab grade)

As part of the lab for this course, we will be reading a book that covers some of the history of cell culture, one of the most important aspects of cell biology research for the past 50 years and a technique that we will be learning in lab. This book will also give us an opportunity to discuss some of the bioethical questions that have been a historical part of biomedical research, many of which continue to be an issue today. Understanding these questions and both sides of the argument, along with the implications on people and important biomedical research and advances is critically important for you who will be the future doctors, scientists, and teachers. We will be discussing parts of this book in lab throughout the course, culminating in a big discussing during the last lab period. At this point in your career, it is important to have some level of accountability on your own.

Thus, a relatively small percentage of your grade is based on this book. However, I truly believe that your understanding (and hopefully interest) of this class will be greatly enhanced by reading of this book. It is a pretty quick read altogether. Your points will be mainly based on participation in the discussion and evidence of having read and understood the book. There will be some questions on the lab quizzes, and perhaps a couple of questions on the main exam, based on this book

Electronic Lab Notebook (Cell culture project)

Keeping a detailed notebook procedures performed, data accumulated, and analyses of the data and subsequent next steps is critical for any researcher. Be sure to keep this updated as we go, and **make sure that you write exactly what was done and how (don't just copy and paste the protocol from the lab handout**). Things do not always go perfectly according to the protocol so you need to keep good detailed notes on exactly what was done (concentrations, timing of incubations, etc.) for both the parts that corresponded perfectly with the protocol as well as mistakes or necessary alterations. Sometimes, changes to the protocol are the reason that the lab does not work, and sometimes they turn out to be what helps a particular procedure work. You need to note exactly what was done so that you can either replicate it at some later date, or determine areas that need improvement for a protocol to work. I suggest keeping a written lab book to take notes on, followed by completing an electronic lab book. Your lab notebook should contain:

1) A section describing the purpose (what are we trying to do, and why). Be sure to relate the lab procedure to its relevance and importance in broader cell biology experimentation and/or clinical techniques.

2) A detailed log of the procedures performed including the date, exact methods used, and any pertinent notes or results of that work. For procedures that were repeated multiple times (e.g. cell culture procedures), you should include the detailed protocol once, and then any problems and/or deviations from the standard procedure (and the reason for changes) should be noted each time.

3) Results of the lab including any images obtained pertinent to the lab (such as images of cells, images obtained from fluorescent staining, etc.). Be sure to directly reference the images along with a thorough description of what is being observed and the relevance of this. Is the staining real? Is it what you would expect? Why or why not? Etc. Finally, discuss conclusions and next steps based on that data.

For the 5 – 6 week cell culture lab, your grade will be based on the lab notebook (rather than a standard lab report). I would strongly suggest that your group set up a Googledocs or OfficeLive file online so that each member can access and add to the same file as appropriate. Further information on the electronic lab notebook will be given throughout the course. **If you have any questions at all, please ask.**

Lab Report for cryosectioning, and for hematopoietic stem cell labs.

- The **introduction** should start broad and become more specific as you go along. The analogy I like to use is that of an hourglass. Start with general information about the techniques or lab and its purpose and get more specific as you go. For example, in the cryosectioning lab report, you should start by introducing cryosectioning and its purpose / advantage, followed by immunofluorescence, and then get more specific by discussing the tissue we are using and the markers we are staining for, and what you expect to observe.
- The **methods** section describes the procedures by which this was accomplished (***in past tense; this should always be formatted in a "blank was done" manner***). This should not be in the form of a bulleted protocol, but rather a paragraph. Avoid the first or third person (don't say "we did this", say "this was done)
- The brief **results** section should include the **labeled** images from your slides, complete with figure legends. **This section should also have a short 1-2 paragraph description of what the results show** within the body. Describe your observations thoroughly in the main paragraph and reference the figures appropriately. **The separate figure legends should briefly describe the image being shown**.
- The **discussion** should relate what you see in your images to what was expected, along with some modifications that could be made to improve the results, and future directions for similar experiments (be creative and show some understanding of the utility of this method). This is the bottom of the hourglass. You start specific describing and analyzing the meaning of the observations / results. Then put them into broader context and discuss issues / future directions, etc.

Points Breakdown for lab work

Quizzes (9 x 5 pts each; drop lowest score)		**40 points**
Cryosectioning and staining		**30 points**
Quality of work	10 pts	
Lab report (not a lab notebook)	20 pts	
Artificial cell culture (groups of 3)		**70 points**
Quality of work	20 pts	
Ability to keep cells alive		
Requirement for cell culture replacement = -5 points per replacement		
Quality of fluorescence staining		
Health of cells		
Electronic lab book	30 pts	
Group participation / effort	20 pts	
Bone marrow cells in culture (groups of 3)		**25 points**
Quality of work	10 pts	
Data analysis	10 pts	
Group participation / effort	5 pts	
Henrietta Lacks discussions		**20 points**
Total		**185 pts**