Biology 3451: Cell and Molecular Biology Laboratory  
Fall 2012 Lab Syllabus and Schedule  
Room 330 Cherry Emerson  
Section A – Wednesday 12-3 pm; Section B – Wednesday 3-6 pm  

Instructor  
Dr. Shana Kerr  
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E-mail: shana.kerr@biology.gatech.edu  
Phone: 404-385-0065  
Office Hours: M 10:30-11:30 am or by appointment  

Teaching assistants – office hours by appointment  
Jennifer Goff  
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Matt Johnson  
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Dr. Merrill will also be visiting frequently from the Cell Biology lecture course, and may participate in the discussions and experiments.  

Suggested Texts*  
A Short Guide to Writing about Biology, 8th edition by Jan A. Pechenik  
A Student Handbook for Writing in Biology, 3rd edition by Karin Knisely  

*Note: these texts are not required; however, they are extremely useful resources and will significantly improve your writing if you utilize them. I recommend Pechenik over Knisely, but it is more expensive.  

Website  
This course makes extensive use of T-square (tsquare.gatech.edu). You are responsible for checking the website frequently for announcements. Quizzes, experimental procedures, and other important information will be posted on the site. Lab reports should be submitted via the assignments tab on T-square.  

Overview  
You will explore fundamental aspects of cell biology and current methods used in the cell biology laboratory by developing a group research project modeled on the experience of an individual student working under a research faculty member. Our model system is the mouse macrophage cell line, RAW264.7 and we will verify findings described in the paper “Regulation of the lipopolysaccharide signal transduction pathway by 17beta-estradiol in macrophage cells.” by E. Vegeto et. Al. (2004) J Steroid Biochem Mol Biol. 2004 Jun; 91(1-2):59-66. You will need to read this article and bring it to the lab, and you can find it using the Georgia Tech library website. We will be studying the response of RAW264.7 cells to LPS, a bacterial endotoxin. Five experiments will be performed to examine changes in cell morphology, proliferation and cell death, and gene expression that accompany LPS-induced activation.  

Course format  
Each of the five experiments will be performed over a three week period. During the first week, the relevant concepts will be discussed and the class will develop a protocol to be used in the experiment by adapting standard methods posted on the course T-square site. Students are expected to keep an accurate laboratory notebook and have the necessary procedures written out in full when they arrive in class the following week. During week two, you will work in groups to carry out the experiment, analyze the data, and organize your results. During week three, students will review their findings in preparation for writing their final reports. Discussion of the next experiment will also take place at this time.
Lab Safety and Personal Protection Equipment

Safety policies are mandated by federal, state, and institutional rules to keep everyone safe. Violation of safety policy will result in loss of all participation points for the lab in question. **Report all injuries or accidents to the TA or instructor immediately.** Safety glasses and lab coats must be worn in the lab during weeks we are conducting experiments. Lab coats must be purchased by students, and safety goggles will be provided. Shoes that completely enclose the foot must be worn at all times. During some laboratories, you will need to wear gloves (these will be provided). The following policies are non-negotiable, and failure to abide by them will result in removal and an unexcused absence from that day’s lab:

- You must wear shoes that cover your feet entirely (no flip flops, ballet slippers, sandals).
- No food or drinks, including water bottles.
- No cell phone use, including texting (phones must be silenced and off the lab bench).
- Clean up your lab station at the end of lab and report any mess left behind from previous lab sections to your TA.
- Properly dispose of trash, glassware, and biohazard waste. Other people’s safety may be compromised by your negligence.
- During “wet labs” you must wear long pants to the ankle, your lab coat and goggles, and long hair should be tied back. Long pants are recommended for every lab.
- Follow additional safety procedures for specific lab activities as indicated by your TA.

**Academic integrity**

All students should be familiar with their rights and responsibilities under the Georgia Tech Academic Honor Code and are expected to abide by its provisions. Academic dishonesty isn’t a “victimless” crime; it interferes with instruction, damages the reputation of the Institute, and ultimately harms the perpetrator who fails to learn course material or appreciate the value of individual effort. Plagiarism and other violations of the Honor Code ([http://www.honor.gatech.edu](http://www.honor.gatech.edu)) will not be tolerated and can result in severe disciplinary action, up to and including expulsion. Prohibited conduct includes, but is not limited to: copying from another student or allowing someone to copy your work (sharing group data when completing laboratory reports is permitted, but submission of identical written work is not), using notes in any form on a quiz without the express permission of the instructor, requesting a re-grade of an assignment after altering it, submitting someone else’s work as your own, or allowing your work to be submitted under another person’s name. When in doubt, ask your TAs or instructor whether an activity constitutes an Honor Code violation.

**Attendance and Participation**

All students are expected to be present each week in lab. (This includes being on time.) If you do not provide the instructor with a valid Georgia Tech excused absence (see the bylaws) within 24 hours of missing a lab, your grade will drop by 5%. Excused absences will require appropriate documentation (ie, physician’s note, letter from the Dean of Students, police report). Makeup work for excused absences is due within one week. If you miss a lab, you are still responsible for completing assignments and getting data from your group members.

Participation will be based mainly on contribution to class discussions. During the discussion sessions, participation will be scored based on the quality (not correctness) of answering questions and if you ask questions that allow for forward movement of the discussion.
Lab Notebooks
You will need a bound composition-style notebook for this course. Good record-keeping is an essential aspect of any research lab. All content should be written in ink. Each experiment should begin with a hypothesis and a brief statement explaining the approach to test that hypothesis. Detailed protocols should be written in your notebook by the day for that experiment. You should be using your handwritten protocols during the experiment. During the lab, you should note any changes to the protocol, clarification, etc. Also note any observations made during the experiment, as well as final observations and conclusions/interpretations. If the lab results were not as expected, include a statement about troubleshooting ideas and alternative interpretations. Graphs and other data should be assembled and taped into your notebook by the beginning of the discussion day for that experiment. Your lab notebook will be spot-checked regularly and graded at least twice during the semester. There will also be a final lab notebook grading at the end of the semester.

Online Quizzes
Quizzes will be posted on T-square throughout the semester and will be due half an hour before lab starts. A quiz submitted at 2:36 for 3:05 lab will be considered late. These quizzes will cover the necessary background for you to perform and understand the experiment that will follow the next week. Quizzes will be based on both material discussed in class as well as material posted on T-square.

Written reports
After the completion of an experiment, each student should prepare a journal style article for the lab report. This should include:

- Abstract: concise summary of what happened during the experiment (2-3 sentences for each experiment)
- Introduction: to provide adequate background pertaining to the cell line and experiments to give any reader knowledge of why you did the experiment. This should include your hypothesis.
- Materials and Methods: concise summary of what you did including how the cells were prepared and maintained.
- Results: This section will include both figures and written results. Here you state simply what you saw.
- Discussion: This section you will analyze your results and state why you observed what you did during the experiment. (Conclusions section)
- References: you will need to be looking for references to support your introduction and discussion. Don’t forget to cite the paper that the lab is based on!

Lab reports will be submitted via T-square half an hour before class begins. Lab reports will be reduced by 10% for each day they are late. A lab report submitted at 2:36 pm for 3:05 pm lab will be considered late.

Group Presentations
On the final lab meeting day, each group will lead the class through a discussion of Experiment 5. This will include background, rationale, methods, results, and an analysis of what conclusions could be made from those results. The presentations should be 10-15 minutes in length and may use Powerpoint, Prezzi, or other presentation formats. The electronic portion of the presentation must be submitted half an hour before the final lab session begins.

Grading
Quizzes and lab notebooks 20%
Lab reports, Experiments 1-3 30% (10% each)
Attendance and Participation 10%
Group presentation 10%
Final cumulative report 30%

Final scores will be rounded to the nearest whole number, and grades will be assigned according to the following scale: 90-100% A; 80-89% B; 70-79% C; 60-69% D; <60% F
<table>
<thead>
<tr>
<th>Date</th>
<th>Week #</th>
<th>Discussion Topics and Activities</th>
<th>Post-class Reading and Assignments (due via T-square no later than half an hour before the next lab period begins*)</th>
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</thead>
<tbody>
<tr>
<td>22-Aug</td>
<td>1</td>
<td>Introduction to the cell biology laboratory&lt;br&gt;Objectives and format&lt;br&gt;Introduction to the paper&lt;br&gt;Lab safety discussion</td>
<td>Reading: Vegeto et al.&lt;br&gt;Quiz 1: Introduction to the Paper</td>
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<td>29-Aug</td>
<td>2</td>
<td>In-depth discussion of paper&lt;br&gt;Lab skills and statistics exercise</td>
<td>Reading: Pechenik Chapter 4, pp. 49-68&lt;br&gt;Quiz 2: Statistics and Laboratory Skills</td>
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<td>5-Sep</td>
<td>3</td>
<td>Procedures used in cell culture&lt;br&gt;Cell culture exercise&lt;br&gt;Preparation for Experiment 1</td>
<td>Quiz 3: Experiment 1</td>
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<td>12-Sep</td>
<td>4</td>
<td><strong>Experiment 1: Viability of LPS-treated RAW264.7 cells</strong>&lt;br&gt;You will examine the effects of increasing LPS dosages on cell viability using two different assays: trypan blue exclusion and marker enzyme activity (WST-1)</td>
<td>Reading: Pechenik Chapter 9, pp. 149-220 or Knisely Chapter 4, pp.47-89</td>
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<td>19-Sep</td>
<td>5</td>
<td>Discussion of Experiment 1&lt;br&gt;Discussion on writing lab reports&lt;br&gt;Preparation for Experiment 2</td>
<td>Quiz 4: Experiment 2, part 1&lt;br&gt;Lab report: Experiment 1</td>
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<td>26-Sept</td>
<td>6</td>
<td><strong>Experiment 2, Part 1: Effects of LPS on RAW264.7 cells</strong>&lt;br&gt;You will use light and fluorescence microscopy to examine effects of LPS on RAW264.7 cell morphology, culture density, and phagocytosis relative to a negative control</td>
<td>Quiz 5: Experiment 2, part 2</td>
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<td>3-Oct</td>
<td>7</td>
<td><strong>Experiment 2, Part 2: Effects of LPS on RAW264.7 cells</strong>&lt;br&gt;You will use flow cytometry to examine effects of LPS RAW264.7 cells</td>
<td>Reading: Pechenik Chapter 2, pp. 21-33 or Knisely Chapter 2, pp. 9-33</td>
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<td>10-Oct</td>
<td>8</td>
<td>Discussion of Experiment 2&lt;br&gt;Preparation for Experiment 3</td>
<td>Quiz 6: Experiment 3&lt;br&gt;Lab report: Experiment 2</td>
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<td>17-Oct</td>
<td>9</td>
<td><strong>Experiment 3: Expression of MMP-9 during LPS-induced activation</strong>&lt;br&gt;You will use a quantitative reverse-transcription PCR (QPCR) assay to measure relative amounts of MMP-9 mRNA in treated and untreated cells</td>
<td>Reading: Pechenik Chapter 10, pp. 221-231 or Knisely Chapter 2, pp. 9-33&lt;br&gt;Literature review</td>
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<tr>
<td>24-Oct</td>
<td>10</td>
<td><strong>Experiment 3 gel analysis</strong>&lt;br&gt;Discussion of Experiment 3&lt;br&gt;Preparation for Experiment 4</td>
<td>Quiz 7: Experiment 4&lt;br&gt;Experiment 5 grant proposals</td>
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<td>31-Oct</td>
<td>11</td>
<td><strong>Experiment 4: Changes in MMP-9 enzymatic activity during cellular activation</strong>&lt;br&gt;You will determine the relative activity of MMP-9 protein in stimulated and unstimulated RAW256.7 cells using an <em>in situ</em> gel electrophoresis (zymography) assay&lt;br&gt;<strong>Experiment 5 Study Section</strong></td>
<td>Lab report: Experiment 3</td>
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<td>7-Nov</td>
<td>12</td>
<td>Discussion of Experiment 4&lt;br&gt;Preparation for Experiment 5&lt;br&gt;Experiment 5 protocol design sheet</td>
<td>Quiz 8: Experiment 5</td>
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<td>14-Nov</td>
<td>13</td>
<td><strong>Experiment 5: Original Experiments</strong></td>
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<td>21-Nov</td>
<td>14</td>
<td>Week of Thanksgiving – no lab</td>
<td>Group presentation (submit presentation slides to T-square by Wednesday, Nov 28 half an hour before lab begins)</td>
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<td>28-Nov</td>
<td>15</td>
<td><strong>Group Presentations and Discussion</strong>&lt;br&gt;Course wrap-up</td>
<td>Final lab report (cumulative) due Wednesday Dec 5 @ noon</td>
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<td>5-Dec</td>
<td>16</td>
<td>Final lab report due – no lab</td>
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*Note: An assignment turned in at 2:36 pm for the 3:05 pm lab section will be considered late.*