# CHEMISTRY 567

## OVERALL BIOCHEMISTRY LABORATORY COURSE SCHEDULE – FALL ‘18

<table>
<thead>
<tr>
<th>Week</th>
<th>Monday 2:00 pm lecture</th>
<th>Tuesday, 2:00 pm Lab</th>
<th>Thursday, 2:00 pm Lab</th>
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<tbody>
<tr>
<td>Aug 27</td>
<td>Course Overview Biochemistry Boot Camp</td>
<td>Boot camp continued…</td>
<td>1. 2:00 pm: Lecture on lab techniques, part 2 2. 3:30 pm: Interactive Biochemistry Computer Techniques (Chem computer room, GMCS245). All students must go through the computer-based interactive section of the Photosynthetic Reaction Center experiment prior to the lab phase beginning on Sept 8</td>
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<td>Sept 3</td>
<td>Labor day</td>
<td>1. Introductory Lecture on Photosynthetic Reaction Center Experiment 2. Locker Check-in</td>
<td>1. Begin Photosystems Experiment - isolation of spinach thylakoids (Introduction and Flow Chart due at beginning of class.)</td>
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<td>Sept 10</td>
<td>1. Introductory Lecture on Lactate Dehydrogenase Kinetics Experiment</td>
<td>1. Finish Photosystems Experiment- separation and characterization of the photosystems</td>
<td>1. Begin Lactate Dehydrogenase Kinetics Experiment (Introduction and Flow Chart due at beginning of class.) a) Progress Curves; initial velocities. b) Effects of enzyme concentration on initial velocity.</td>
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<td>Sept 17</td>
<td>EXAM on Photosynthetic Reaction Center Experiment (including lab techniques lecture)</td>
<td>1. Lab Report on Photosystems Experiment due at beginning of class. 2. Continue Lactate Dehydrogenase Kinetics pH effects on velocity.</td>
<td>1. Continue Lactate Dehydrogenase Kinetics Velocity vs. substrate concentration plot. Inhibition kinetics with cibacron blue.</td>
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<td>Sept 24</td>
<td>First Introductory Lecture on Aldolase Experiment</td>
<td>Kathy guest lectures in Forensic Anthropology, no class today</td>
<td>1. Begin Purification and Characterization of Aldolase experiment (Introduction and Flow Chart for first four days of Aldolase Experiment due at beginning of class, i.e., purification, dialysis, phosphocellulose column chromatography, gel filtration chromatography, etc.) a) Isolation of chicken breast muscle cytosol b) Prepare gel filtration column. c) Salt in excess sample</td>
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<td>Oct 1</td>
<td>EXAM on Lactate Dehydrogenase Kinetics Experiment.</td>
<td>1. LAB REPORT DUE at beginning of class on LDH Kinetics. 2. Continue Aldolase experiment</td>
<td>1. Continue Aldolase experiment a) Run phosphocellulose column. b) Determine elution volume of first set of gel</td>
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<td>Date</td>
<td>Event</td>
<td>Instructions</td>
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| Oct 7 | Second Introductory Lecture on Aldolase Experiment. | 1. Continue Aldolase experiment  
a) Run aldolase on gel filtration column.  
b) Continue protein concentration and specific activity assays.  
c) Prepare SDS polyacrylamide gel.  
1. Introduction and Flow Chart for second part of Aldolase Experiment (SDS gel and immunoblot) due.  
2. Continue Aldolase experiment  
a) Run SDS PAGE gel and blot to membrane (western transfer)  
b) Determine elution volume of second set of gel filtration column standards. |
| Oct 8 | Introductory Lecture on Plasmid Prep | 1. Continue Aldolase experiment  
a) Develop Immunoblot.  
b) Finish elution volume determination of final gel filtration column standards.  
Finish Aldolase (if needed, otherwise, day off) |

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<tr>
<th>Date</th>
<th>Monday 1:00 pm lecture</th>
<th>Tuesday, 2:00 pm Lab</th>
<th>Thursday, 2:00 pm Lab</th>
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| 9 Oct 22 | **Continue** introductory lecture on plasmid prep | 1. Begin Plasmid Preparation Experiment  
a) Prepare and autoclave media for 1 liter cultures  
b) Start overnight 25 ml culture from glycerol stock. | 1. **Introduction and Flow Chart for Plasmid Prep due at beginning of class**  
a) At **1:00 pm**, inoculate 1 liter cultures  
b) 2:00 pm: Begin monitoring cell growth by periodic readings of $A_{600}$  
c) Add chloramphenicol.  
d) Incubate overnight. TA will spin down the cells the following morning. You may observe or help if you would like to. |
| 10 Oct 29| **EXAM on Aldolase Module** | 1. **LAB REPORT on Aldolase Experiment DUE at beginning of Class (2:00 PM).**  
2. Continue Plasmid Prep  
a) Prepare clear lysate  
b) Set up first CsCl gradient  
c) Start first equilibrium gradient centrifugation run | 1. **Introductory Lecture on the Polymerase Chain Reaction (PCR) experiment, synthetic DNA oligonucleotide synthesis, and gel electrophoresis of nucleic acids**  
2. Continue Plasmid Prep  
a) Take down first CsCl gradient  
b) Start second CsCl gradient centrifugation |
| 11 Nov 5 | **Continue Introductory Lecture on PCR/STR’s.** | 1. Start PCR/STR experiment (**Introduction and Flow Chart Due at beginning of class**)  
2. Isolate cheek cell DNA  
3. Continue Plasmid Prep- Take down second CsCl gradient, extract ethidium bromide, and precipitate the DNA | 1. Continue PCR/STR experiment- Set up PCR  
a) Pour agarose / acrylamide gels  
2. Continue Plasmid Prep  
a) Spin down precipitated DNA, dissolve in water, and reprecipitate with ethanol.  
b) Spin down reprecipitated DNA, air dry pellet, resuspend in 1 ml TE buffer |
| 12 Nov 12| | 1. Run gels with DNA samples  
2. Finish Plasmid Prep  
a) Take absorbance readings of plasmid preps; calculate yield  
3. Introductory Lecture on DNA Sequencing | 1. Read STR gel  
2. Video on DNA sequencing  
3. Practice reading sequencing gel autoradiograms |
| 13 Nov 19| Lecture- Real time PCR | 1. **LAB REPORT on Plasmid Prep DUE at beginning of class.**  
2. Tissue culture and stem cells lecture | HW: Large scale carbohydrate digestion experiment |
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<td>14 Nov 26</td>
<td>Introductory Lecture on GST-SH2 Expression and Purification.</td>
<td>Getting a job/resumes</td>
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| 15 Dec 3  | Next generation sequencing lecture                                     | 1. Begin GST-SH2 Expression and Purification Experiment. (Introduction and Flow Chart for GST-SH2 Expression and Purification Due at beginning of class)  
              2. Lyse bacterial cells and bind supematant to resin.  
              3. Elute protein from resin.                                |
| 16 Dec 10 | Exam on Recombinant DNA module, including GST-SH2.                     | 1. Finish GST-SH2 Expression and Purification, destain gel, and photograph.  
              2. Check out.                                                  |
| Dec 17    | Lab Report on GST-SH2 due - 11:00 pm. (email)                          | No final exam                                                           |
|           |                                                                       |                                                                         |