**Class Hours:** Tuesday & Thursday 1 – 5 pm  
**Laboratory:** 284 SEB and other rooms as needed;  
**Instructor:** Professor Denis Callewaert; 203 SEB; 370-2349; callewae@oakland.edu  
**Office Hours:** Tuesday & Thursday 11 – 1 or by appointment  
**Text:** *Fundamental Laboratory Approaches for Biochemistry and Biotechnology* by Ninfa and Ballou plus handouts for additional experiments

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<thead>
<tr>
<th>Date</th>
<th>Experiments and Reading Assignments</th>
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| 8/31 (Th) | Introduction to the Biochemistry Laboratory  
*Reading Assignment: Chapter 1 and Handouts*  
- Safety in the laboratory  
- The laboratory notebook  
- Ethics in the recording, reporting and citing of scientific data  
- Operation of pipettors and spectrophotometers |
| 9/5 (Tu)  | Column Chromatography (Gel Filtration)  
*Reading Assignment: Chapter 4 (pp 89-100, 108-113)*  
Experiment 4-1: Sephadex chromatography/packing/calibration |
| 9/7 (Th)  | Analytical Methods for Amino Acid Separation and Identification  
*Reading Assignment: Handouts*  
- thin layer chromatography (TLC)  
- capillary zone electrophoresis (CZE) |
| 9/12 (Tu) | Measurement of Protein Concentration  
*Reading Assignment: Chapter 3 and Handouts*  
Experiment 3-1 and Handouts  
- Bradford method  
- BCA method  
- spectrophotometric assay |
| 9/14 (Th) | Measurement of Protein Concentration *  
- completion of measurements and calculations  
- introduction to protein purification |
9/19 (Tu)  **Isolation and Purification of Alkaline Phosphatase**  
*Reading Assignment: Chapter 6 and 7*  
Experiment 7-1 (first day)  
• preparation of Fraction 1

9/21 (Th)  **Purification of Alkaline Phosphatase**  
Experiment 7-1 (second day)  
• heat denaturation - Fraction 2  
• ammonium sulfate precipitation - Fraction 3  
• dialysis

9/26 (Tu)  **Purification of Alkaline Phosphatase**  
Experiment 7-1 (third day)  
• DEAE cellulose chromatography  
• enzyme assay on all fractions

9/28 (Th)  **Characterization of Alkaline Phosphatase**  
*Reading Assignment: Chapter 5*  
Experiment 5-2 (activity gel) [reference to Experiment 5-1]  
• native gel and activity assay  
• protein determination  
• enzyme assay (if necessary)

10/3 (Tu)  **Enzyme Kinetics - Alkaline Phosphatase**  
*Reading Assignment: Chapter 8*  
Experiments 8-1 and 8-2

10/5 (Th)  **Data Analysis and Discussion; Catch up and clean up**

10/10 (Tu)  **Exam I (written and practical)**  
*Practical based on Experiments 9-1 and 9-3*  
**Notebooks Due**

10/12 (Th)  **Plasmid Isolation**  
*Reading Assignment: Chapter 11, pp 277-299, and Handout*  
• microscale isolation of plasmid DNA

10/13 (F)  **Approval Required for Paper Topic**

10/17 (Tu)  **Restriction Digestion and Analysis of Plasmid DNA**  
• Agarose Gel Analysis  
*Reading Assignment: Handout*

10/19 (Th)  **Computer-Based PCR Primer Lab**  
*Reading Assignment: Chapter 12, Handout*
10/24 (Tu)  PCR Technology *
Reading Assignment:  Experiment 12-1 and Handout

10/26 (Th)  Characterization of PCR Product
  •  Laboratory clean up

10/31 (Tu)  Subcellular Fractionation
Reading Assignment: Handout
  •  preparation of homogenate
  •  differential centrifugation

11/2 (Th)  Subcellular Fractionation
  •  biuret assay
  •  marker enzyme assay

11/7 (Tu)  Microsomal Membrane Characterization
Reading Assignment: Handout and Experiment 5-1
  •  protein and lipid extraction
  •  SDS-PAGE gel casting

11/9 (Th)  Microsomal Membrane Characterization
  •  SDS-PAGE of membrane proteins
  •  TLC of membrane lipids

11/14 (Tu)  Catch up day, Rough Draft of Paper Due

11/16 (Tu)  Radioisotopic Assay of Fatty Acid Oxidation
  •  Radiation Safety Training (D. Luongo)
  •  NOTE: Must pass training exam prior to next lab

11/21 (Th)  Radioisotopic Assay of Fatty Acid Oxidation
  •  Experimental analysis of fatty acid oxidation
Reading Assignment: Handout Reading Assignment: Handout

11/28 (Tu)  Radioisotopic Assay of Fatty Acid Oxidation
  •  Laboratory Decontamination
  •  Data analysis & discussion

11/30 (Th)  Exam II (written)
Laboratory Notebooks Due

12/4 (M)  Paper Due

12/4 (Tu)  Oral presentations of Research Papers
LABORATORY CLEAN UP and check out
Grading

<table>
<thead>
<tr>
<th>Grading</th>
<th>Exams</th>
<th>100 points (2 @ 50 points)</th>
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<tbody>
<tr>
<td>Notebooks</td>
<td>150 points (2 @ 75 points)</td>
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<td>Technique</td>
<td>100 points</td>
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<td>Paper</td>
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<tr>
<td>Oral Presentation</td>
<td>50 points</td>
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<tr>
<td>Clean up and safety</td>
<td>1 point per lab period</td>
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Notes:

1. For obvious logistical and pedagogical reasons, prompt, regular attendance is very important and will factor into the technique portion of your grade.

2. For most experiments you will work in pairs. At the end of the semester your partner will be asked to comment on your contribution to group work and these comments will figure into the technique portion of your grade. Therefore, do not assume the group exercises are an opportunity for your partner to cover any deficiencies in your preparation.

3. Revised tuition refund policy: Students now have 14 calendar days to receive a 100% refund should they decide to drop classes. Periods for partial refunds are now eliminated. See www.oakland.edu/registrat for details.

Policy on Plagiarism

Often during the course of this semester we will be working in groups. In addition, there may be times during the semester when, for one reason or another, and with the instructors permission, you will be using data obtained by someone else for the write-up of your experiments. The sharing of information, data, or ideas in these circumstances is allowable, however, appropriate acknowledgment of the source of such contributions is required. Furthermore, the use of another’s data does not obviate the need to perform your own calculations, do your own data analysis, and draw and present your own conclusions. There is no reason for both members of a group to arrive at exactly the same conclusions, even if they are using the same data. There is plenty of room for interpretation based upon the results of these experiments. In fact, in the final discussion of an experiment the justification of your conclusions is a major portion of the grade for a particular experiment. Therefore, do not present the conclusions and ideas of another student as your own, you must think about and justify, your own conclusions. If you arrive at your interpretation as a result of discussions with other students, it is still necessary to provide your own analysis and summary of the experiment. If insufficient evidence of independent work is observed, the write-ups will be submitted to the academic conduct committee for adjudication.

Laboratory Notebooks and Reports:

All laboratory reports should be contained within a bound (non-perforated) composition notebook. Students will be required to purchase two of these books (one
for the 1st half of the semester and one for the 2nd half). All books should contain a table of contents (leave 2 pages in front for this) listing the experiment number, name, and the pages for which the documentation of data resides. All pages should be numbered before the start of the 2nd experiment. No pages should be removed and white out should not be used. Data pages can be pasted in (e.g. spectrophotometer printouts) but it is suggested that a small three-ring binder be purchased for high-volume printouts (e.g. Kinetics data). If a three ring binder is used, please refer to it where necessary within the bound composition book and have some kind of reference page within the binder. A detailed description of the way each lab report should be documented is on pages 47 and 48 of FLABB (your text book) but I will give a short description here.

1. The experiment title, number, and date for which it is to be started should be listed on the top of the first page of each experiment.

2. An abstract (~200 words) should be written about each experiment before it is started. This must be initialed before you will be able to start the experiment.

3. A brief introduction of the experiment and it purposes.

4. A materials and methods section: states the materials that were used and their purposes (e.g. a buffer, a solvent) and give references to methods used (e.g. Pierce, Lowry).

5. A results section: data, graphs, calculations (note: all raw data is required), and conclusions.

6. A discussion section where you talk about the results you obtained. This is the section where you can get retribution if your experiment failed. Discuss what went wrong and where (if you know) or discuss possible errors that may have caused poor results. Understand that poor pipetting skills does not qualify as an argument and any argument given must be supported by what you know of the experiment and what you will learn researching the subject matter on your own time.

7. A literature cited section: all references used should be cited (plagiarism is not tolerated). Note: if something is considered common knowledge it does not need to be cited (appears in most literature about the subject).

Most experiments will require the student to do outside research in order to develop a good discussion of the experiment and interpretation of the results obtained. It is recommended that you do so if you want to receive the maximum points for your reports. If you start one experiment prior to completing discussion of another, please skip some pages to allow room for calculations and discussion.
General Education Mandated Information

Catalog Course Description: Techniques of extraction, separation, identification and quantification of biomolecules, including electrophoresis, chromatography, and radioisotope techniques, with emphasis on mathematical treatment of experimental data. Identical with BCM 457

Prerequisites: CHM/BCM 453, which may be taken concurrently.

Course Objectives: This course satisfies the General Education writing intensive in the major and Capstone requirements.

As the Capstone Experience, for satisfactory completion the student will demonstrate:

- Appropriate uses of a variety of methods of inquiry and a recognition of ethical considerations that arise.

- Practical application of laboratory techniques involves a variety of methods of inquiry, in particular, different instruments and techniques are used to achieve the same sort of information. The library portion of the course will involve the use of published literature to examine problems similar to those encountered in the laboratory exercises.

- Ethical considerations with respect to acceptance and rejection of data, presentation of data, attribution of sources of information, and the sharing of information will be continuously encountered over the course of the semester. These topics will specifically addressed during the formal session on ethics.

- The ability to integrate the knowledge learned in general education and its relevance to the student’s life and career

The writing portion of the course will require a literature search, utilization of databases of scientific information, and the use of available internet-based research tools to answer specific scientific questions. The construction of a paper and presentation will require the distillation of knowledge gained into a coherent presentation capable of being understood by others.

Cross Cutting Capacities:

Effective Communication: The preparation of a manuscript, and a presentation based upon the manuscript, will require the development of effective communication skills including inter alia, the preparation of visual aids, the distillation of knowledge into a coherent position, attention to grammatical and organizational issues, and the ability to answer questions from the audience based upon the presentation.

Critical Thinking: The capacity for critical thinking is essential to successfully master and apply concepts in a scientific laboratory. Relevant and irrelevant conceptual information in the normal course of trying to understand the ideas underlying a particular laboratory exercise will be encountered. In addition, valid and invalid data points will be routinely obtained and the decision about whether or not to accept or reject these data points will be an almost daily occurrence. Both ethical and statistical concepts will be necessary to make these decisions. Once data have been accepted it will be necessary to interpret these data within the context of the conceptual basis of the
experiment. The need to explain and understand unexpected (albeit valid) outcomes will be frequent. During construction of the manuscript a large amount of information will be obtained that will require serious examination involving the critical thinking process.