Brooklyn College – CUNY Department of Chemistry Fall 2017 Syllabus – Professor Davenport Chemistry 4572 (Section RQ9; 17192) – Biochemistry Laboratory I (2 credits; 4-hours)

Course Goals and Learning Objectives. The goal of this course is to provide the student with both a theoretical and practical understanding of basic biochemical methodologies. Specifically students will perform methods for the isolation, purification and characterization of proteins, lipids and DNA. Students are also introduced to the application of database management for biotechnological applications.

Required Text

Laboratory procedures are posted on Blackboard and can be downloaded ahead of the laboratory meeting.

Instructor Contact Information

NAME	Extension	Room
Prof. Davenport	2825	344NE
LDvnport@brooklyn.	cuny.edu	

Office hours:

Monday:	2:00pm - 4:00pm	
Wednesday:	11:00am - 12:00noon	
or by appointment (please email first)		

Class Meeting Schedule

The class meets weekly. Thursday (9:00am - 12:50pm) Room 451NE Ingersoll Extension

Please bring a written protocol (see Pre-Lab Outline below) to each lab session (in your lab notebook) for the day's experiment based on the instructions in the lab manual in order that you have a personalized guide for performing the experiment. NOTE: You will <u>not</u> be given any extra time for lab sessions.

Experimental Schedule

<u>Day One</u>: Check-In; Protein Determination Using the BCA Assay; Construction of a Protein Standard Plot.

Day Two: Spectrophotometric Estimation of pK_a Values.

<u>Day Three</u>: Quantitative Spectrophotometric Determination of Total DNA and RNA in Animal Cells.

<u>Day Four</u>: Thermal Melting Profile of DNA Helix Unfolding (Demonstration).

<u>Day Five</u>: Isolation and Identification of Lipids from Egg Yolk and Animal Cells Using TLC Analysis.

<u>Day Six</u>: Ferrimyoglobin Fluoride Titration and Binding Kinetics.

<u>Day Seven</u>: Protein Chemistry and Enzymology (Isolation of Crude Alkaline Phosphatase from E.coli; Alkaline Phosphatase Activity Assay; Purification Using Binding to DEAE Sephadex A-50).

<u>Day Eight</u>: Ion Exchange Chromatography-Ammonium Sulfate Precipitation.

<u>Day Nine</u>: Electrophoresis and Activity Assay of Purified Alkaline Phosphatase.

<u>Day Ten</u>: Determination of Molecular Weight of Alkaline Phosphatase by Analytical Gel Filtration.

<u>Day Eleven</u>: Enzyme Kinetics (K_M and V_{MAX} Determinations).

<u>Day Twelve</u>: Competitive and Noncompetitive Inhibition.

<u>Day Thirteen</u>: Alkaline Phosphatase: Internet Exercise in Protein/Gene Database Management (NCBI and ExPASy).

Day Fourteen: Check Out.

Laboratory Requirements

Please bring detergent, paper towels and a laboratory notebook (this will NOT be handed in or graded and is solely for documenting your observations and acquired/measured data during the lab sessions). Safety goggles <u>must</u> be worn at all times (see below). Some procedures may soil clothing and some may involve hazardous chemicals. You are expected to understand the preparation of laboratory reagents and solutions, and should review UNITS such as percent (w/w, w/v, v/v) and molarity.

Grade Breakdown

The final grade for the course is based on the sum of the scores received for the following five lab reports, one quiz (held at the end of the semester) and laboratory performance:

- 10% Spectrophotometric Methods
- 15% DNA/RNA Lab
- 10% Lipid Lab
- 10% Ferrimyoglobin Binding Kinetics Lab
- 35% Alkaline Phosphatase Lab
- 15% Lab Quiz
- 5% Lab Performance

Given that most labs are conducted with a lab partner, the lab performance component is designed to encourage students to engage in the practical aspects of the experiment rather than serve as a spectator.

Laboratory Reports

Submit an electronic version of your lab reports using Blackboard. In the "Assignment" folder on Blackboard you will find an appropriate link to upload your report prepared in either pdf or Word format.

Please submit/upload lab reports for grading <u>within 2</u> <u>weeks</u> after completion of the experiment. If you need access to word processing and other software, you may use the Chemistry Department's personal computers in the lab on the fourth floor or other campus computers. Please answer any questions that may be posed in the lab handouts in the Results or Discussion sections of your report.

Note: you will be sharing data with your partner but NOT preparing a joint report. Your report will be returned to you if the writing is not grammatically correct and the format does not follow the guidelines below; your grade will be penalized for subsequent submissions that do not follow the expected guidelines. For cases of plagiarism, students will be assigned a zero for their report and the instructor may take further action. Plagiarism is easy to detect and is considered Academic Dishonesty (see below). If you are unsure of what plagiarism means, please consult the CUNY quidelines about plagiarism at: http://www.brooklyn.cuny.edu/web/about/initiatives/pol icies/integrity.php. In addition, the following link has many examples of plagiarism and is very instructive: http://www.gc.cuny.edu/About-the-GC/Governance,-

Policies,-Procedures/Detail?id=4827. This document is also available on the Blackboard course site.

Please prepare lab reports in the format of a research article as published in the American Chemical Society's journal Biochemistry. If possible the lab report should be <u>typed</u>, but must be organized into the following sections:

<u>Abstract</u>: The abstract <u>concisely</u> states the problem, the experimental approach used, the most important results, and conclusion(s). Only a few sentences are needed.

<u>Introduction</u>: This section provides <u>brief</u> background information about the biochemistry addressed by the experiment. This information will justify why you carried out the experiment. The introduction usually ends with a statement summarizing what you will show the reader as your report progresses.

<u>Materials and Methods</u>: This section describes the materials used (any dangerous chemicals involved) and the experimental procedures followed. Refer to the handouts instead of rewriting all the details of the procedures. For example, "A series of buffer solutions was prepared from pH 4 to 8 according to the hand out"; or, "Absorbance measurements were made at the desired wavelengths using the Spectronic20 spectrophotometer". Avoid details such as: "I pipetted 20 mL of solution A into a test tube and then added 20 mL of solution B into the test tube".

<u>Results and Calculations</u>: The results section includes your experimental results presented in <u>tabulated</u> and/or figure format as appropriate, accompanied by a <u>concise</u> description. You need to <u>show calculations</u> (and <u>errors</u> where possible) with <u>appropriate equations</u>. All graphs can be computer generated (using SigmaPlot or Excel) with the axes accurately labeled and legends (or titles) clearly describing the data presented. Calculations (only) can be hand-written (<u>no page limit</u>).

<u>Discussion</u> (approx. two paragraphs): The discussion section provides a <u>short and comprehensive</u> description of your findings or conclusions. This should <u>NOT</u> take the form that includes comments such as "this was a fun experiment..." or "I learned how to...". Rather, this section provides a discussion of the principles involved in the experiment and any conclusions that you can make based on your data. Discuss experimental errors or problems, which occurred and attempt to make a simple conclusion. When you evaluate your data or a concept that is already known for a chemical species and is available in the chemical literature, <u>you must compare your results</u> to literature values, report a <u>% error</u> and <u>cite the</u> source of the known value(s). The handouts may not

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always specify what and when to do this, so use your judgment.

Reports are evaluated based on: 1) the quality of the results but only to a degree that coincides with how well things go in the lab, which varies from group to group and year to year. For example, if all your enzyme sample is accidentally spilled during the fourth week of a 6week experiment, you will not be penalized greatly for this kind of mishap and will be given enzyme from one of the other student groups or from the stock room; 2) the quality of the written text (in terms of explanations and interpretation of results); 3) presentation of data. If you are not yet familiar with SigmaPlot or other data handling software (Excel), this is a great opportunity to learn; 4) explanation of why the experiment did not go according to plan or the results do not match literature values, sources of errors, etc.

Lab Quiz

A short written lab quiz, based on the experiments/concepts covered throughout the semester, will take place at the end of the semester. The quiz is worth 15% of the course grade.

Pre-lab Outline

Please prepare a brief pre-lab summary (<u>one-page or</u> <u>less</u>) for each of the labs to be performed. This can be uploaded before the appropriate lab session using the "Assignments" folder located on Blackboard. The prelab is particularly important for the alkaline phosphatase/protein purification and characterization experiments, since the overall report will cover seven weeks of work and the pre-lab will provide you with guidance for preparing the full written (graded) lab report. You will NOT be allowed to start the experiment without a pre-lab. Your pre-lab outlines (not copied or pasted from the lab procedure hand-outs) need to briefly address the following key points:

- 1. Purpose of Experiment.
- 2. Techniques and Apparatus to be used.
- 3. Type of Data Collected (what will be measured and calculated).
- 4. List of Main Reagents and Dangerous Chemicals (if any).

Safety

It is a New York State law that safety goggles be worn at all times by all students in the laboratory. You will lose marks if you are observed not wearing safety goggles. Goggles are provided as part of the lab equipment rental fee. Students who consistently refuse to properly wear safety goggles during the lab period

Eating and drinking are NOT permitted during lab sessions.

Pregnant students are encouraged to defer taking Chemistry 4572 laboratory.

Accommodations for Students with Disabilities In order to receive disability-related academic accommodations, students must first be registered with the Center for Student Disability Services. Students who have a documented disability or suspect they may have a disability are invited to set up an appointment with the Director of the Center for Student Disability Services, Ms. Valerie Stewart-Lovell, at 718-951-5538 (vstewart@brooklyn.cuny.edu). If you have already registered with the Center for Student Disability Services, please provide your professor with the course accommodation form and discuss your specific accommodation with him/her.

Academic Integrity

The faculty and administration of Brooklyn College support an environment free from cheating and plagiarism. Each student is responsible for being aware of what constitutes cheating and plagiarism and for avoiding both.

The complete text of the CUNY Academic Integrity Policy and the Brooklyn College procedure for implementing that policy can be found at this site: http://www.brooklyn.cuny.edu/bc/policies. If a faculty member suspects a violation of academic integrity and, upon investigation, confirms that violation, or if the student admits the violation, the faculty member MUST report the violation.

All students should read carefully and thoroughly the 2017-2018 Brooklyn College Bulletin (http://www.brooklyn.cuny.edu/web/about/administrat ion/enrollment/registrar/bulletins.php) for a complete listing of academic regulations of the College.

The State law regarding non-attendance because of religious beliefs shall be followed as given in the Brooklyn College Bulletin, Undergraduate Programs 2017-2018, page 66 (http://www.brooklyn.cuny.edu/web/about/administrat ion/enrollment/registrar/bulletins.php). If you miss a

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lab session for religious observances, please make arrangements ASAP with your instructor and stockroom for a make-up time.

Important Dates (Fall 2017)

<u>Last Day to Add a Course</u> Thursday, August 31st

<u>Makeup Class/Conversion Date</u> TUESDAY, September 19th

<u>No Classes</u> Thursday, September 21st and November 23rd.

Last Day to Resolve INC Grades from the Spring/Summer Semesters Thursday, November 9th

Last Day to Withdraw From a Course with a W (Non-Penalty) Grade Friday, November 10th