INTRODUCTION TO ESSENTIAL BIOLOGICAL TECHNIQUES

NUTR 225

SYLLABUS

FALL 2012

Course Instructor
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Overall Goals

To become familiar with biochemical and molecular techniques commonly used in nutrition research: Five broad classes of techniques will be covered: The course will emphasize how these different classes of techniques are used in a complementary and mutually-enforcing manner to address a research question.
1. Partitioning techniques for the separation and analysis of complex biomaterials
   1. Physical separation of cell constituents and biomolecules
2. Analysis of these using antibody-based techniques in conjunction with…
3. Fluorescence and Bioluminescence
4. Gene Expression: Analysis and In Vitro Manipulation

General Approach

The intent of this intensive, short course is to familiarize you with fundamental techniques used to study biology at the molecular, cell, tissue and whole organism level. The techniques that are covered in this course will be referred to in classes and seminars throughout your training at the Friedman School of Nutrition.

The basic theory for individual laboratory techniques is standardized across many disciplines, and can be learned in the form of self-study. An additional goal of this course is to familiarize you with the “virtual” communities of scientists and the vast resources available to you in public domain websites managed by scientific consortia. For each technique or group of techniques we have provided initial web-based materials describing particular techniques. You are expected to complete these and the assignments designated by **** before class. Class time will be devoted to a 20 minute quiz, discussion of the answers to the quiz and expanded discussion of techniques covered in your reading. Yes, there will be a quiz at the first class.

Prerequisites

Undergraduate biochemistry course

Grading

This 0.5 credit course is graded S/U and is required of all Biochemical and Molecular Nutrition (BMN) students. Grading is based on the average of thee three quizzes (75%) and classroom participation (25%). A grade of “S” requires an overall average of 70% or above.

2012 Course Schedule

Class will meet Tuesday September 4th and Friday September 7th, 14th, and 21st from 1:00-2:30. All classes will meet in the mezzanine conference room at the HNRCA (next to the auditorium).
Class #1. Tuesday, September 4: Introduction; Lab Basics; Partitioning Techniques

A. Levels of Biological Organization; Lab Books; On-Line Tools; ISI

I. [http://staff.jccc.net/PDECELL/lifeis/biorgnew.html](http://staff.jccc.net/PDECELL/lifeis/biorgnew.html)
II. [http://techtransfer.tufts.edu/?pid=11&c=36](http://techtransfer.tufts.edu/?pid=11&c=36) (links describing lab books and e-lab books)
III. [http://www.currentprotocols.com/](http://www.currentprotocols.com/) (Invaluable resource of online methods and protocols; explore)
IV. **Assignment:** Using Current Protocols, find and bring to class (1) a glossary of bioinformatics terms, and (2) a downloaded figure from the Reactome Database showing pathways involved in integration of energy metabolism.
V. [http://www.scientistsolutions.com/](http://www.scientistsolutions.com/) that addresses how to determine energy expenditure in mouse models of metabolic diseases such as obesity.
VI. [http://physics.nist.gov/cuu/Units/index.html](http://physics.nist.gov/cuu/Units/index.html) - (ISI; understand difference between base units and derived units and be facile with ISI prefixes). Two guides for appropriate use of ISI are:

B. Separation / Partitioning Techniques: Gas and Liquid Chromatography:

II. [http://www.youtube.com/watch?v=ddfeyLgeKx8&NR=1](http://www.youtube.com/watch?v=ddfeyLgeKx8&NR=1)
IV. [http://www.youtube.com/watch?v=q0pM-kOsVQ&feature=related](http://www.youtube.com/watch?v=q0pM-kOsVQ&feature=related)
V. [http://www.youtube.com/watch?v=xXkOte0dPfg&feature=related](http://www.youtube.com/watch?v=xXkOte0dPfg&feature=related)

C. Mass Spectrometry

II. [http://www.chemguide.co.uk/analysis/masspec/howitworks.html](http://www.chemguide.co.uk/analysis/masspec/howitworks.html) (up to “understanding what’s going on”)
IV. [http://www.youtube.com/watch?v=rBymrFzcaPM&feature=fvwrel](http://www.youtube.com/watch?v=rBymrFzcaPM&feature=fvwrel) (lectures 1 and 2)

Class #2: Friday, September 7: Working With Mammalian Cells; Electrophoresis

A. Cell Culture, Cell Fractionation Using Centrifugation or Commercial Kits

X. [http://nobelprize.org/educational/medicine/dna/a/translation/svedberg_unit.html](http://nobelprize.org/educational/medicine/dna/a/translation/svedberg_unit.html)
XI. [http://homepages.gac.edu/~cellab/chpts/chpt3/figure3-5.html](http://homepages.gac.edu/~cellab/chpts/chpt3/figure3-5.html) (no need to memorize- only appreciate the principle)


**B. Separation and Initial Characterization of Proteins and Nucleic Acids**


II. [http://www.davidson.edu/academic/biology/courses/Molbio/SDSPAGE/SDSPAGE.html](http://www.davidson.edu/academic/biology/courses/Molbio/SDSPAGE/SDSPAGE.html)

III. [http://www.youtube.com/watch?v=UuuN05ZApb0&feature=related](http://www.youtube.com/watch?v=UuuN05ZApb0&feature=related)

IV. [http://www.dnalc.org/resources/animations/gelelectrophoresis.html](http://www.dnalc.org/resources/animations/gelelectrophoresis.html)

V. [http://www.youtube.com/watch?v=6_4AY3iYRgo&feature=related](http://www.youtube.com/watch?v=6_4AY3iYRgo&feature=related)

**** Assignment: Bring to class an on-line protocol for separating RNA complexes by gel electrophoresis.-Be prepared to discuss the concepts behind the protocol.

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**Class #3. Friday September 14. Antibody and Fluorescence-Based Techniques**

**A. Measuring Antigens in Biological Samples (blood, tissue homogenates, etc)**

I. [http://www.youtube.com/watch?v=Ys_V6FcYD5I&feature=related](http://www.youtube.com/watch?v=Ys_V6FcYD5I&feature=related)

II. [http://www.youtube.com/watch?v=hQmaPwP0KRI&feature=BFa&list=PL12D59D28513CA093&index=1](http://www.youtube.com/watch?v=hQmaPwP0KRI&feature=BFa&list=PL12D59D28513CA093&index=1)


V. [http://www.sumanasinc.com/webcontent/animations/content/ELISA.html](http://www.sumanasinc.com/webcontent/animations/content/ELISA.html)

**B. Fluorescence and Flow Cytometry**


**C. Imaging Techniques (Immunofluorescence, Confocal Microscopy and In Vivo Imaging)**


II. [http://www.youtube.com/watch?v=OH2GFeaGV6w](http://www.youtube.com/watch?v=OH2GFeaGV6w)


V. [http://www.youtube.com/watch?v=g5U-n4Toq60&feature=related](http://www.youtube.com/watch?v=g5U-n4Toq60&feature=related)

VI. [http://www2.healthsci.tufts.edu/saif/mods.htm](http://www2.healthsci.tufts.edu/saif/mods.htm)

VII. [http://circ.ahajournals.org/content/117/3/379.full.pdf+html](http://circ.ahajournals.org/content/117/3/379.full.pdf+html) (introduction and figures)

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**Class #4. September 21: Measuring and Manipulating Gene Expression**

**A. PCR-based approaches: PCR, RT-PCR and Real-Time PCR;**


B. Cell Transfection, Reporter (Promoter) Assays and RNA Interference

I. http://www.promega.com/multimedia/ (view animation on transfection and reporter assays)
II. http://www.jove.com/index/details.stp?ID=240
III. http://www.cellbiolabs.com/viral-expression
V. http://www.nature.com/focus/rnai/animations/animation/animation.html (modules 1 and 2)
VI. http://www.promega.com/resources/multimedia/rna-analysis/introduction-to-rna-interference/
VII. http://www.jove.com/details.php?id=1499
VIII. http://www.youtube.com/watch?v=QA67v4vSg00&feature=player_embedded

C. Bioinformatics