CHEM 567-1, General Biochemistry
Spring 2023

COURSE INFORMATION

Lectures: Mondays in GMCS 305 1:00-1:50 pm
Laboratories: Tuesdays and Thursdays in CSL 226 2:00-4:40 pm

Instructor: Peter van der Geer
Email: pvanderg@sdsu.edu
During the week, I usually respond to emails within 24 hours.
Office location: CSL 322
Office hours: Monday, 9:30–11:00; Wednesday, 9:30-11:00, Friday 9:30-11:00

Instructor: Richard Schroeder
Office: CSL 505
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Phone:
Office hours:

Instructor: Kathy McNamara Schroeder
Office: CSL 505
Email: kmcnamara@sdsu.edu
Phone: 619-582-5332
Office hours: By appointment

THIS COURSE IN YOUR DEGREE:

Prerequisites: Chemistry 560 or equivalent.
It is strongly recommended that you take one or more of the biochemistry electives (Chemistry 562, 563, 564) before or concurrently with Chemistry 567.
This course fulfills a requirement for majors in Chemistry or Biochemistry with an emphasis in Biochemistry

COURSE DESCRIPTION

This is a combined lecture and laboratory class. The lectures will cover what you need to know to successfully complete the labs, but additional material is often covered to expand your knowledge of modern biotechnology. We will cover isolation of membranes, separation of membrane components, enzyme kinetics, protein purification and analysis, DNA isolation, PCR, recombinant DNA, and associated topics such as tissue culture, sequencing, and use of radioactivity. You will work with a group to complete all tasks so communication and planning is essential.

COURSE MATERIALS

- Chem 567 "Biochemistry Laboratory" by Stumph, Metzger, and Adams, available in electronic version from the Aztec Shops bookstore. This is shelved under the name "Stumph". The 2008-2009 version includes material excerpted from Methods in Enzymology. Do not use older versions.
- An actual bound laboratory notebook.
- Please note the university charges a lab fee for this class.
- You may wish to wear a lab coat during the class, it will prevent stained clothing which is certainly possible during some of the experiments. It may be kept in the locker if desired. You will also need gloves for some of the experiments.

STUDENT LEARNING OUTCOMES

Students that have completed this course should be able to:

1. Explain the purpose of each chemical component used in an experiment, and how it aids in the ultimate goal of the experiment.
2. Safely set up and use standard laboratory equipment such as superspeed and ultracentrifuges, pipets, acrylamide and agarose gel electrophoresis modules, spectrophotometers, shakers, thermocycler, fraction collector, gravity chromatography columns.
3. Carry out measurements and calculations of results obtained using standard biochemistry laboratory equipment.
4. Perform techniques commonly used in biochemical research including PCR, electrophoresis, western blotting, enzyme kinetics, expression of recombinant proteins, isolation and purification of DNA, and growth of bacterial cells.
5. Observe safe practices in the laboratory, follow proper procedures and regulations for safe use and disposal of chemicals.
6. Interpret results of experiments in lab reports that include background information on the topic, a plan to accomplish the experiment, an outline of the theory behind relevant lab techniques, and results/discussion of the experimental outcome that allow students to draw reasonable, accurate conclusions from analysis of the results.
7. Conduct work in lab both independently and as part of a team.
8. Troubleshoot an experiment with the intention of improving the outcome.
9. Devise an experiment using previously practiced techniques

ASSIGNMENTS AND ASSESSMENTS

Lab notebook inspection: There will be one unannounced inspection of the lab notebook.

4 lab reports: Introduction and flow-chart are always due **before** the onset of the experiment (see schedule). Complete reports are usually due 1 week after completion of the experiment (see schedule).

There are 4 exams:

Exam 1, Photosystems experiment and lab techniques	Monday February 6
Exam 2, LDH Kinetics	Monday February 20
Exam 3, Aldolase purification	Monday March 20
Exam 4, DNA section	Monday May 1

Bring a **calculator** and **ruler** to the exam.

Homework: DNA sequencing

There is no final exam

KEEPING A LAB NOTEBOOK

1. Leave several pages blank at the beginning of the notebook to keep an up-to-date **Table of Contents**.
2. **Introduction and Flow-chart**: To be written **prior** to starting the experiment. Copies are to be submitted electronically on the date due, but copies must also be kept permanently in the notebook, placed there on the date each intro is due.
3. **Experimental Procedures and Results**: Record any and all deviations from the protocols provided in the lab manuals or handouts, whether these are intentional or done in error. Record primary data (obtained during the lab period) and derived data. That is, record your raw data as it is gathered, and then show how you get from your raw data to your final tabulated data. An example of every single calculation you do to get from collected data to any result needs to be shown.
4. **Conclusion and Discussion:** Same as for lab report, this is also placed in the notebook the day it is due in class.
5. Your entries into the lab notebook do not have to be pretty. However, one should be able to read and follow what was written. The goal of keeping a notebook is to be able to take a primary protocol along with your research notebook and be able to completely **understand and reproduce** the work that was accomplished.

LAB REPORTS

A. Introduction

1. Background: Give a brief background of the topic of research (at least half a page)
2. Purpose: Give the goal of the experiment (one sentence, **not more**)
3. General plan: Describe in general terms how you are going to carry out the experiment (three to four sentences, **not more**)
4. Theory: Describe for all **lab techniques** involved in the experiment, how they work and why they were used. Describe the function of **all chemicals** that are used in the various steps of the experiment.

- Sections should be separated and have clearly visible headings

B. Flow chart

One page, unless otherwise noted. This should be a general outline of the experimental procedures to be followed.

Important notes:
Sections A and B (i.e., Introduction and Flow Chart) should be written as if you were planning out this experiment in your own mind prior to initiating work on the experiment. Sections A and B must also be included in your lab notebook. (It is suggested to type A and B, copy them, and then tape these copies into your notebook)
Sections A and B must be submitted before the **beginning** of the lab period on the day they are due. This is usually, but not always, the day on which you begin the experiment. See schedule for due dates.

C. Experimental procedures

If you are provided with a detailed step-by-step protocol, you only need to mention any actual variations from the protocol provided. However, be sure to mention **any and all** deviations. You do not have to re-copy the protocols from the laboratory manuals. If there is not a detailed protocol provided for a particular experiment, you will need to include more details in this section. This section will be handwritten in your notebook but should be typed for your lab report.

D. Results

The length of this section will vary from experiment to experiment. Present the results in the most clear, logical, and understandable fashion possible. In the results section you describe, superficially, what you did (details are in the theory and experimental procedure sections) and what you found. The results section should be readable independently of the rest of the report, which means that someone who has not done the experiment understands what you did and what you found after reading this section. The results section should also be readable when charts, figures and tables have been removed. In your narrative you should refer to charts, figures, and tables after you describe what they show.

Data shown in a graph also must be presented in a table (preferable side-by-side). The results section should include equations and examples of all calculations.

E. Conclusion and discussion

Discuss the results of your experiment. In your introduction there was a purpose statement. Address this statement by explaining the results gathered, and if the experiment accomplished what was said in the purpose. Finally, do include error analysis. Describe what errors may have taken place **and the impact that these had on the results.**

GRADING POLICY

Photosystems report	100 points
LDH report	100 points
Aldolase report	150 points
Plasmid isolation report	75 points
PCR/STR report	75 points
GST-SH2 domain purification report	75 points
Photosystems exam	150 points
LDH exam	150 points
Aldolase exam	150 points
DNA exam	150 points
Notebook inspection	100 points
DNA sequencing homework	25 points
Total	1300 points (100%)

DUE DATE POLICY

Failure to hand in the **Introduction and Flow Chart** at the date due will result in a **10-point penalty per week (or part thereof).** This penalty will be subtracted from the total points. Moreover, if you do not hand in a copy when initially due, you are still expected to prepare and include these sections as part of your lab report, or you will **lose even more points** in the grading of your report.

Late lab reports will be accepted up to the last day of wet lab work (but not after). Failure to hand in the lab report at the due date result in a **10-point penalty per week (or part thereof).**

LETTER GRADES

Percentage	Points	Letter grade
93 and above	1209 and above	A
90	1170-1208	A-
87	1131-1169	B+
83	1079-1130	B
80	1040-1078	B-
77	1001-1039	C+
73	949-1000	C
70	910-948	C-
67	871-909	D+
63	819-870	D
60	780-818	D-
below 60	779 and below	F

FLOW-CHART **EXAMPLE**

Large Scale Plasmid Preparation - Flow Chart

Day 1

1. Prepare and autoclave media.

Day 2

1. Innoculate 1 liter cell cultures. Incubate with shaking at 37^{o}C.
2. Read absorbance of culture at 2, 2.5, 3 hrs etc.
3. When A_{600} reaches 0.55-0.6, add chloramphenicol. Incubate overnight and centrifuge cells on the following morning.

Day 3

1. Thaw cell pellet and lyse cells on ice.
2. Remove cell debris by centrifugation in Sorvall high speed centrifuge. Save supernatant containing the plasmid DNA.
3. Set up CsCl-ethidium bromide equilibrium gradient centrifugation. Spin in ultacentrifuge at 41,000 rpm in TV-850 rotor at least 18 hours.

Day 4

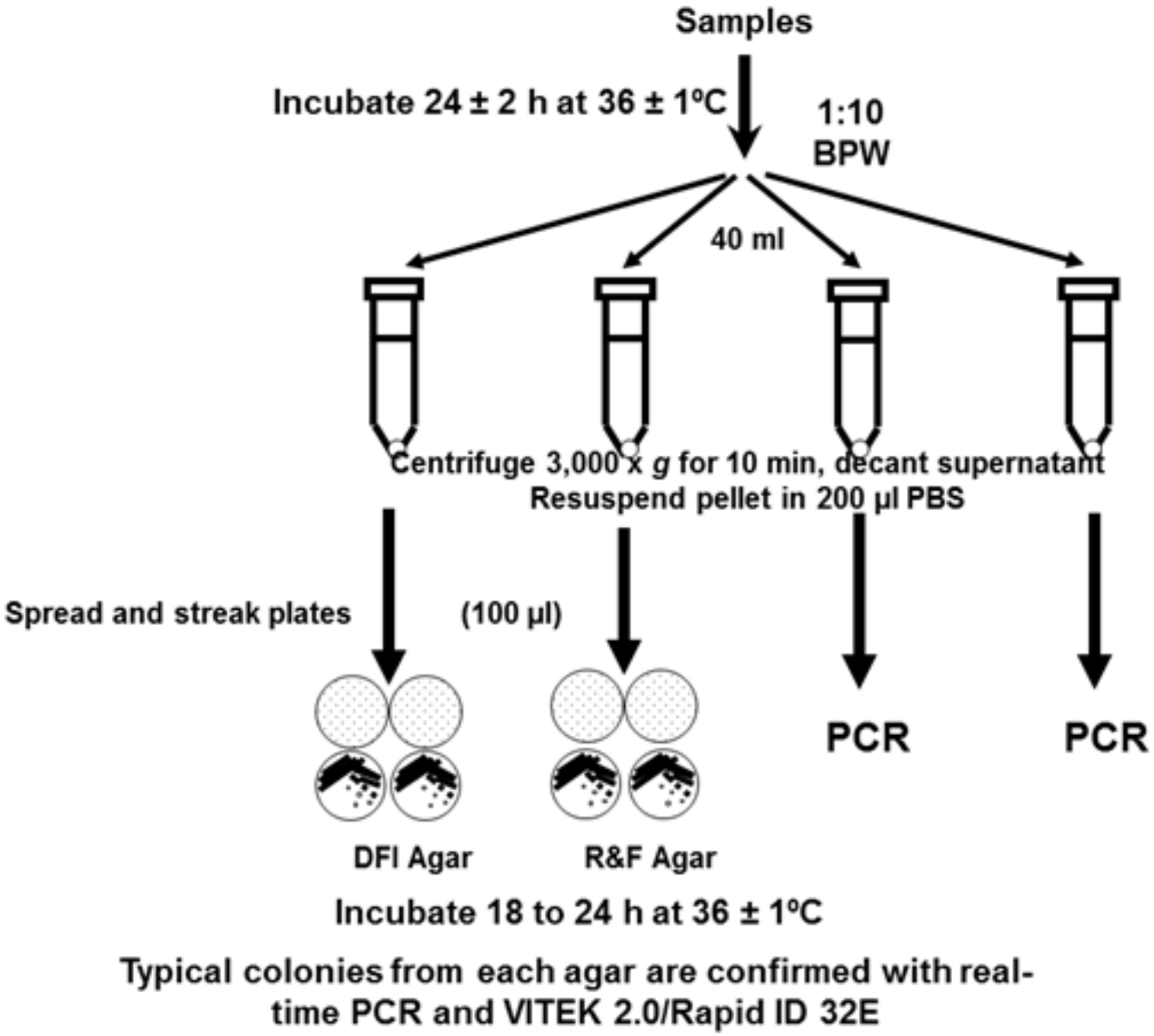
1. Bring down ultracentrifugation run. While illuminating with UV light, remove plasmid DNA band from centrifuge tube using needle and syringe.
2. Set up second CsCl-ethidum bromide centrifugation in TV650 rotor. Spin at 41,000 rpm at least 18 hours.

Day 5

1. etc.

Sometimes, depending upon the particular experiment, it may be preferable to **diagram** the flow chart as shown on the following page. Choose whichever method (or combination of the two) is best for getting across the points most clearly.

Alternative Flow Chart Format



Include or leave out as much detail as required to meet the 1 page limit. Aim for 2/3 to 1 page in length. Most importantly, demonstrate that you have planned out the experiment and that you know what you are going to be doing.

CHANGES TO THE SYLLABUS

Consistent with University policy, I retain the right to adjust course design, including assignments, assessments, and deadlines. Major departures from the syllabus shall be made only for compelling reasons. "Any major changes to the course syllabus will be announced in class, communicated to all students electronically, and incorporated into an updated and posted version of the syllabus."

UNIVERSITY POLICIES

Accommodations: If you are a student with a disability and are in need of accommodations for this class, please contact Student Ability Success Center at (619) 594-6473 as soon as possible. Please know accommodations are not retroactive, and I cannot provide accommodations based upon disability until I have received an accommodation letter from Student Ability Success Center.

Student Privacy and Intellectual Property: The Family Educational Rights and Privacy Act (FERPA) mandates the protection of student information, including contact information, grades, and graded assignments. I will use [Canvas / Blackboard] to communicate with you, and I will not post grades or leave graded assignments in public places. Students will be notified at the time of an assignment if copies of student work will be retained beyond the end of the semester or used as examples for future students or the wider public. Students maintain intellectual property rights to work products they create as part of this course unless they are formally notified otherwise.

Religious observances: According to the University Policy File, students should notify the instructors of affected courses of planned absences for religious observances by the end of the second week of classes.

Resources for students: A complete list of all academic support services--including the Writing Center and Math Learning Center--is available on the Student Affairs' Academic Success website. Counseling and Psychological Services (619-594-5220) offers confidential counseling services by licensed therapists; you can Live Chat with a counselor at http://go.sdsu.edu/student_affairs/cps/therapist-consultation.aspx between 4:00pm and 10:00pm, or call San Diego Access and Crisis 24-hour Hotline at (888) 724-7240.

SDSU Economic Crisis Response Team: If you or a friend are experiencing food or housing insecurity, or any unforeseen financial crisis, visit sdsu.edu/ecrt, email ecrt@sdsu.edu, or walk-in to Well-being & Health Promotion on the 3rd floor of Calpulli Center.

Academic Honesty: The University adheres to a strict policy prohibiting cheating and plagiarism. Examples of academic dishonesty include but are not limited to:

- copying, in part or in whole, from another's test or other examination;
- obtaining copies of a test, an examination, or other course material without the permission of the instructor;

- collaborating with another or others in work to be presented without the permission of the instructor;
- falsifying records, laboratory work, or other course data;
- submitting work previously presented in another course, if contrary to the rules of the course;
- altering or interfering with grading procedures;
- assisting another student in any of the above;
- using sources verbatim or paraphrasing without giving proper attribution (this can include phrases, sentences, paragraphs and/or pages of work);
- copying and pasting work from an online or offline source directly and calling it your own;
- using information you find from an online or offline source without giving the author credit;
- replacing words or phrases from another source and inserting your own words or phrases.

The California State University system requires instructors to report all instances of academic misconduct to the Center for Student Rights and Responsibilities. Academic dishonesty will result in disciplinary review by the University and may lead to probation, suspension, or expulsion. Instructors may also, at their discretion, penalize student grades on any assignment or assessment discovered to have been produced in an academically dishonest manner.

Classroom Conduct Standards: SDSU students are expected to abide by the terms of the Student Conduct Code in classrooms and other instructional settings. Violation of these standards will result in referral to appropriate campus authorities. Prohibited conduct includes:

- Willful, material and substantial disruption or obstruction of a University-related activity, or any on-campus activity.
- Participating in an activity that substantially and materially disrupts the normal operations of the University or infringes on the rights of members of the University community.
- Unauthorized recording, dissemination, or publication (including on websites or social media) of lectures or other course materials.
- Conduct that threatens or endangers the health or safety of any person within or related to the University community, including
    1. physical abuse, threats, intimidation, or harassment.
    2. sexual misconduct.

Medical-related absences: Students are instructed to contact their professor/instructor/coach in the event they need to miss class, etc. due to an illness, injury or emergency. All decisions about the impact of an absence, as well as any arrangements for making up work, rest with the instructors. Student Health Services (SHS) does not provide medical excuses for short-term absences due to illness or injury. When a medical-related absence persists beyond five days, SHS will work with students to provide appropriate documentation. When a student is hospitalized or has a

serious, ongoing illness or injury, SHS will, at the student's request and with the student's consent, communicate with the student's instructors via the Vice President for Student Affairs and may communicate with the student's Assistant Dean and/or the Student Ability Success Center.

Sexual violence / TItle IX mandated reporting: As an instructor, one of my responsibilities is to help create a safe learning environment on our campus. I am a mandated reporter in my role as an SDSU employee. It is my goal that you feel able to share information related to your life experiences in classroom discussions, in your written work, and in our one-on-one meetings. I will seek to keep the information you share private to the greatest extent possible. However, I am required to share information regarding sexual violence on SDSU's campus with the Title IX coordinator, Jessica Rentto 619-594-6017. She (or her designee) will contact you to let you know about accommodations and support services at SDSU and possibilities for holding accountable the person who harmed you. Know that you will not be forced to share information you do not wish to disclose and your level of involvement will be your choice. If you do not want the Title IX Officer notified, instead of disclosing this information to your instructor, you can speak confidentially with the following people on campus and in the community. They can connect you with support services and discuss options for pursuing a University or criminal investigation. Sexual Violence Victim Advocate 619-594-0210 or Counseling and Psychological Services 619-594-5220, psycserv@sdsu.edu. For more information regarding your university rights and options as a survivor of sexual misconduct or sexual violence, please visit titleix.sdsu.edu or sdsutalks.sdsu.edu.

COURSE SCHEDULE

WEEK	LECTURE Monday 1:00-1:50	LABORATORY Tuesday 2:00-4:40	LABORATORY Thursday 2:00-4:40
1 Jan 16			• **Course overview** • **Biochemistry bootcamp** All things you learned and may have forgotten
2 Jan 23	• **Lecture on photosystems experiment**	• **Lecture on lab techniques, part 1**	• **Lecture on lab techniques, part 2** • **Locker Check-in**

WEEK	LECTURE Monday 1:00-1:50	LABORATORY Tuesday 2:00-4:40	LABORATORY Thursday 2:00-4:40
3 Jan 30	• **Lecture on LDH kinetics**	• **Introduction and flow-chart photosystems due** • **Photosystems experiment, day** Isolation of thylakoids	• **Photosystems experiment, day 2** Separation of photosystems
4 Feb 6	• **Exam 1: Photosystems experiment and lab techniques**	• **Introduction and flow-chart LDH kinetics due** • **LDH kinetics, day 1** a. Progress curves; initial velocity b. Effects of enzyme concentration	• **Lab report photosystems due** • **LDH kinetics, day 2** a. Effect of the pH on initial velocity
5 Feb 13	• **Lecture on aldolase purification, part 1**	• **LDH kinetics, day 3** a. Velocity vs substrate concentration b. Inhibition with Cibacron blue	• **Introduction and flow-chart first 4 days of aldolase purification due** • **Aldolase purification, day 1** a. Isolation of chicken breast cytosol b. Pour gel filtration column c. 40% ammonium sulfate cut
6 Feb 20	• **Exam 2: LDH kinetics**	• **Lab report LDH kinetics due** • **Aldolase purification, day 2** a. Ammonium sulfate precipitation b. Determine void volume gel filtration column c. Prepare phosphocellulose column d. Start dialysis	• **Aldolase purification, day 3** a. Run phosphocellulose column b. Determine elution volume first set of gel filtration standards c. Start protein concentration and enzyme assays
7	• **Lecture on aldolase**	• **Aldolase purification, day 4**	• **Introduction and flow-chart last part of aldolase**

WEEK	LECTURE Monday 1:00-1:50	LABORATORY Tuesday 2:00-4:40	LABORATORY Thursday 2:00-4:40
Feb 27	**purification, part 2**	a. Run aldolase on gel filtration column b. Continue protein concentration and enzyme assays c. Pour SDS-polyacrylamide gel	**purification (SDS-PAGE and immunoblot) due** • **Aldolase purification, day 5** a. Run SDS-PAGE b. Transfer half the gel to immobilon c. Stain the second half of the gel d. Determine elution volume second set of gel filtration standards
8 Mar 6	• **Lecture on plasmid isolation, part 1**	• **Aldolase purification, day 6** a. Develop immunoblot b. Determine elution volume of final gel filtration standards	**Day off**
9 Mar 13	• **Lecture on plasmid isolation, part 2**	• **Plasmid isolation, day 1** a. Prepare and autoclave media b. Start overnight 25 mL culture	• **Introduction and flow-chart plasmid isolation due** • **Plasmid isolation, day 2** a. At **1:00 pm** inoculate 1 L culture b. At **2:00 pm** start monitoring A_{600} c. Add chloramphenicol d. Incubate overnight
10 Mar 20	• **Exam 3: Aldolase purification**	• **Lab report aldolase due** • **Plasmid isolation, day 3** a. Prepare clear lysate b. Set up first CsCl gradient c. Start equilibrium gradient centrifugation	• **Lecture on PCR and molecular biology, part 1** • **Plasmid isolation, day 4** a. Take down first CsCl gradient b. Set up and start second CsCl gradient
11	**Spring break**	**Spring break**	**Spring break**

WEEK	LECTURE Monday 1:00-1:50	LABORATORY Tuesday 2:00-4:40	LABORATORY Thursday 2:00-4:40
Mar 27			
12 Apr 3	• **Lecture on PCR and molecular biology, part 2**	• **Introduction and flow-chart PCR due** • **PCR/short tandem repeat experiment, day 1** a. Isolate cheek cell DNA • **Plasmid isolation, day 5** a. Take down second CsCl gradient b. Extract EtBr c. Precipitate DNA	• **PCR/short tandem repeat experiment, day 2** a. Set up PCR b. Pour agarose and polyacrylamide gel • **Plasmid isolation, day 6** a. Spin down precipitated DNA b. Dissolve c. Reprecipitate d. Spin down e. Dry and dissolve
13 Apr 10	**Getting a job** **Writing a resume**	• **PCR/short tandem repeat experiment, day 3** a. Run gels • **Plasmid isolation, day 7** a. Determine DNA concentration, determine yield • **Lecture on DNA sequencing**	• **PCR/short tandem repeat experiment, day 4** a. Read gel b. Practice reading sequencing gel
14 Apr 17	• **Lecture on real time PCR**	• **Lab report plasmid isolation due** • **Lecture on tissue culture and stem cells** • **Lecture on SH2 domain purification**	**Lecture on GST-SH2 purification** **Lecture on tissue culture and stem cells**
15 Apr 24	• **Lecture on next generation sequencing**	• **Introduction and flow-chart GST-SH2 purification due** • **GST-SH2 purification, day 1** a. Lyse bacteria b. Bind GST-SH2 to resin c. Elute GST-SH2	• **Homework DNA sequencing due** • **Lab report PCR due** • **GST-SH2 purification, day 2** a. Run samples on SDS-PAGE b. Stain gel

WEEK	LECTURE Monday 1:00-1:50	LABORATORY Tuesday 2:00-4:40	LABORATORY Thursday 2:00-4:40
		d. Pour gel	
16 May 1	• **Exam 4: Molecular biology and GST-SH2 purification**	• **GST-SH2 purification, day 3** a. Photograph gel • Check out	• **Lab report GST-SH2 purification due on May 9**