

ANSI 5573 - TECHNIQUES IN ANIMAL MOLECULAR BIOLOGY SPRING 2009 SYLLABUS

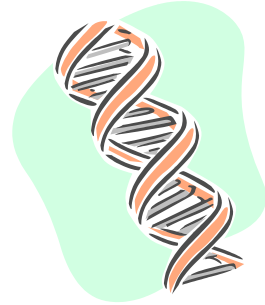
Objective: To understand the principles of major basic animal molecular biology techniques and gain hands-on experience with those techniques

Credit hrs: 3

Time: Lecture: 3:30 pm - 5:10 pm, Mondays (125 ANSI)
Lab: 3:30pm - 5:10 pm, Tuesdays (214 ANSI)

Instructor: Dr. Glenn Zhang, Associate Professor of Molecular Biology
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TA: Tulasi Sunkara, PhD Student
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Prerequisites:

A good understanding of the basic knowledge in biochemistry, molecular genetics, and molecular biology at the college level, equivalent to ANSI 3423 – Animal Genetics; BIOC 3653 – Survey of Biochemistry, BIOC 4113 – Biochemistry, or above.

References and Preclass Reading:

There will be no textbooks for this class, but students may check out one of the following books from the OSU library. In addition to textbooks, additional materials on each major topic will be handed out in advance. To enable productive classes, you need to read all relevant materials prior to the class. Special class sessions will be designed to discuss the handouts.

1. Mulhardt, C. Molecular Biology and Genomics (The Experimenter Series), 4th Ed, 2007, Academic Press.
2. Brown, T.A. 2000. Essential Molecular Biology: Practical Approach. 2nd Edition. Oxford University Press.
3. Sambrook, J. and Russell, D.W. 2001. Molecular Cloning: A Laboratory Manual. 3rd Edition. Cold Spring Harbor Laboratory Press, NY.

Class and Lab Attendance:

Students are expected to attend all lectures, labs, and computer exercises. The final grade will be deducted 5 points for each session missed for up to 50 points, unless the student has a legitimate reason. In addition, failure to attend classes will present a disadvantage to students as it is also likely to miss quizzes that are frequently but randomly conducted in lecture sessions throughout the semester. You are expected to talk with the instructor and/or TA to make up the missed session(s).

Lab Notebooks:

Although students will be working in groups on experiments, each is required to maintain a bound lab notebook with numbered pages. The lab notebook should contain a descriptive title, date, purpose, protocols, results, discussion, and other details necessary to repeat your work (see p. 5 for detailed instructions). Students are required to wear lab coats and complete the entries for “Purpose” and “Protocols” sections of each experiment prior to the lab session. Failure to do so will result in suspension of that lab session. Notebooks will be turned in three times during the semester and graded. No late assignments will be accepted.

Lab Reports:

Lab report is required for each student to summarize a series of experiments in a session. The format used to prepare manuscripts for publication in the *Journal of Biological Chemistry* will be followed. The report should include the following sections: (a) title, (b) summary, (c) introduction, (d) experimental procedures, (e) results, (f) discussion, (g) references, (h) footnotes (optional), (i) figure legends, (j) tables, and (k) figures. Details can be found at <http://www.jbc.org/misc/ifora.dtl>. Two lab reports will be turned in during the semester and graded. No late assignments will be accepted.

Quizzes and Exams:

A total of nine (9) unannounced quizzes will be conducted throughout the semester and seven (7) with the highest scores will be counted toward your final performance. A final comprehensive, closed book, written exam will be conducted in the finals week. Make-up exams and quizzes will be provided only for those missed for legitimate reasons. It is your responsibility to inform the instructor and/or TA as early as possible and, if deemed necessary, to provide official documentation (e.g., from your physician, advisor, or dean) stating that you could not take the exam due to this emergency or illness, in which case we will schedule a make-up exam. Missed exams and quizzes will otherwise be assigned a grade of 0.

Grading:

Your attendance and your performance on three lab notebooks, two lab reports, quizzes, and final exam will be counted. All assignments are expected to be finished on your own and to be well organized and neatly presented. No late assignments will be accepted. Each component is weighted as follows:

Class/Lab Attendance	50 pts
Lab notebooks (2 @ 50 pts)	100 pts
Lab Reports (1 @ 50 pts)	50 pts
Quiz (7 @ 10 pts)	70 pts
<u>Final Comprehensive Exam</u>	<u>180 pts</u>
Course Total:	450 pts

Final grades will be assigned based on point totals as follows:

100% to 90%:	A
89% to 80%:	B

79% to 70%:	C
69% to 60%:	D
59% to 0%:	F

Student Conduct:

Academic dishonesty or misconduct will not be tolerated. When the instructor feels beyond reasonable doubt that dishonesty has occurred, he will take disciplinary action in accordance with university policies and procedures.

Disabilities:

If any member of this class feels that he/she has a disability and needs special accommodations of any nature, we will work with you and the Office of Student Disability Services, 315 Student Union or <http://sds.okstate.edu> to provide reasonable accommodations to ensure that you have a fair opportunity to perform in this class. Please advise the instructor of such disability and the desired accommodations as early as possible to address your needs in a timely manner.

OSU Syllabus Attachment:

We are very concerned about your success as a student at OSU. The information at <http://osu.okstate.edu/acadaffr/aa/CurrentStudents.htm> is provided to answer questions most often asked by students.

Drop and Add Dates

Last day to add a class (without instructor permission)	1/20/09
Last day to drop a course with no grade and no fees charged for courses	1/20/09
Last day to withdraw completely from the University and receive a 100% refund	1/20/09
Last day to enroll (late fee charged beginning 1/17)	1/23/09
Last day to add a class (with instructor permission)	1/23/09
Last day to drop a course with an automatic “W” and receive a 50% refund	1/23/09
Last day to withdraw completely from the University and receive a 50% refund	1/23/09
Last day to drop a class with an automatic “W”	4/10/09

Pre-Finals week	4/27 – 5/1/09
Finals	5/04 – 5/8/09

Spring Semester Holidays

Martin Luther King Day*	1/19/09
Spring Break	3/14 – 3/22/09

HAVE A GREAT SEMESTER!!

LECTURE SESSIONS

SESSION I - DNA CLONING:

Central dogma of molecular biology
Preparation and quantification of DNA
PCR
Agarose gel electrophoresis
Digestion and ligation of DNA
Preparation and manipulation of plasmid
Common enzymes used in molecular biology



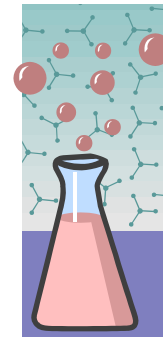
SESSION II - mRNA EXPRESSION:

Mechanisms of gene transcription
Preparation and analysis of RNA
RT-PCR
Primer design
Real-time RT-PCR
Northern blotting
Microarray
RNA interference



SESSION III - PROTEIN EXPRESSION:

Mechanisms of protein translation
Types of posttranslational modifications
Quantification and purification of proteins
Recombinant protein expression techniques
Protein electrophoresis
Western blotting
ELISA



LABORATORY SESSIONS

SESSION I – DNA CLONING:

January 13

Exp. 1. Pipetting

January 20

Exp. 2. Isolation of genomic DNA

Exp. 3. Determination of DNA concentration

January 27

Exp. 4. PCR

February 3

Exp. 5. Agarose gel electrophoresis of PCR products

February 10

Exp. 6. Recovery of DNA from agarose gel

Exp. 7. DNA ligation

February 17

Exp. 8. Transformation of *E. coli* with recombinant DNA

February 24

Exp. 9. Isolation of plasmid DNA

March 3

Exp. 10. Confirmation of the insert:

a) Restriction enzyme digestion

b) PCR

c) Checkup of the results by agarose gel electrophoresis

First lab report: *Cloning of the gene for mouse interleukin-1 β*
(Due 3:30 pm, Monday, March 23 prior to the class)

SESSION II - mRNA EXPRESSION:

March 10

Exp. 11. Isolation of RNA

March 24

Exp. 12. Determination of RNA concentration and quality

March 31

Exp. 13. Real-time RT-PCR

April 7

Exp. 14. Primer design (computer exercise)

Second lab report: *Tissue expression pattern of the mRNA for mouse interleukin-1 β*
(Due 3:30 pm, Monday, April 27 prior to the class)

SESSION III - PROTEIN EXPRESSION:

April 14

Exp. 15. Determination of protein concentration

April 21

Exp. 16. SDS-PAGE

Exp. 17. Coomassie Blue staining of SDS-PAGE gels

Additional Instructions on Laboratory Sessions

All labs will be in the Animal Molecular Biology Lab (214 ANSI) unless otherwise indicated.

Safety Precautions:

- ✓ Always wear lab coats in the lab
- ✓ Always wear disposable gloves during experiments
- ✓ Always wear protective eyewear under UV light
- ✓ Don't access to areas with "biohazard", "infectious agents" or "radioactive material" sign
- ✓ Carefully follow experimental protocols and instrument user's manuals
- ✓ Failure to adhere to these rules will result in a loss of your privilege to work in the lab.

Laboratory Notebook:

Each student is required to have a bound lab notebook with numbered pages before lab work can begin. Remember the purpose of keeping a notebook is to allow you to determine why you did an experiment and exactly how the experiment was performed, so that someone else of similar training can read, understand, and reproduce your work. No pages will be torn off. Any mistakes should be crossed out neatly. All entries should be ink. All data should be recorded promptly and directly in the notebook, not anywhere else.

The notebook should include a table of contents, for which the first page is reserved. The table of contents can be organized in columns as follows:

<u>Experiment No.</u>	<u>Title of Experiment</u>	<u>Page No.</u>	<u>Date</u>
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For each experiment, a descriptive title of the experiment and the date experiment is performing should be provided at the top of the first page. The following sections are to follow in your laboratory write-up:

Purpose: A short statement indicating the objective of the experiment

Protocol: Include a detailed description of the procedure, including volumes of solutions added, time and speed of centrifugation, the composition of the solutions, and all pertinent information that is essential for reproduction of the exp by someone else. If the same experiments are repeated later on, identical procedures can be written in a simplified manner, but detailed protocols should be referred to by giving the page number and changes in the procedures should be indicated clearly in detail.

Results: Include actual raw data collected and final data analyzed. Pictures, diagrams, graphic plots, and calculations involved should be included as well.

Discussion: Include a brief summary of the conclusions of your experiment. For example, did the experiment succeed? Did the controls work? What do the results mean? How do the results compare to your expected results? What are you going to do next?

Since the execution of an experiment is always more successful if you are prepared, you are required to complete the notebook entries for each experiment through the “Protocol” section before beginning the experiment. You are also expected to finish the remaining sections within hours (NOT DAYS) after you finish the experiment. You may discuss the purpose and procedures of the experiment with fellow students or instructor, but you must compose these parts and other parts of the notebook individually. Your notebook needs to be turned in at predetermined dates three times and graded. No late assignments will be accepted.

Laboratory Report:

In addition to the lab notebook, each student is to work independently to prepare written reports to summarize each session’s experiments. The title of each report has been provided at the end of each lab session. The format used to prepare manuscripts for publication in the *Journal of Biological Chemistry* will be followed. You may go to “JBC Instructions to Authors” at <http://www.jbc.org/misc/ifora.dtl> for detailed descriptions on the organization of papers. The written report should include the following in the order listed: (a) title, (b) summary, (c) introduction, (d) experimental procedures, (e) results, (f) discussion, (g) references, (h) footnotes (optional), (i) figure legends, (j) tables, and (k) figures. You may also consult recent JBC papers online at <http://www.jbc.org>. Reports must be typed and organized strictly in JBC format and need to be turned in at predetermined dates. No late assignments will be accepted.

