Biology425 Biochemistry, Molecular Biology and Genetics Superlab Fall 2022
Syllabus

THIS LITTLE PIGGY: BIOMARKERS OF ANIMAL WELFARE

Instructors
Dr. John Wagner/ L53 NBS/ jwagner@sas.upenn.edu/ 215 746-2159
Dr. Jenni Punt/Rosenthal 217A/ punt@vet.upenn.edu/ 610 389-1782
Developed in collaboration with Dr. Tom Parsons
(https://www.vet.upenn.edu/research/centers-laboratories/center/swine-teaching-research-center/swine-center-team)

TAs
TBA

Lab
Th 1:30-4:30 Levin L57 Labs
The lab is also available for use outside of designated lab times
(Please check with John to assure that the lab is unlocked).

Course description
We invite you to join us this fall to perform original experiments exploring the impact of chronic stress on the immune system of domesticated pigs and identifying immune parameters that correlate with pig well-being. Our immune system is responsible for protecting us against pathogen infection and for maintaining a healthy community of cells and microbes. The immune system is influenced not just by encounters with pathogens, but by stressors with environmental, genetic and behavioral origins. Aspects of the immune system, in fact, reflect the overall well-being of human and non-human animals, including those we keep as livestock. The quality of care for livestock animals is quite variable, and a topic of concern for consumers, farmers, and the public in general. The ability to assess these animals' well-being is in part hampered by a lack of understanding of the effects that farming practices have on stress responses, which, in turn, is reflected by differences in immune cell phenotype, genotype, function, and frequency.

In this lab, we will work together to identify differences in immune cell phenotype, genotype and frequency that may be useful as biomarkers of psychosocial stress, one variable that has an impact on well-being. We will focus on the welfare of individual sows in Penn Vet's research colony. Using flow cytometry, we will examine cellular and molecular profiles of white blood cells from sows that hold different positions in the social hierarchy of their community. Parameters that have been proposed to reflect psychosocial stress in humans include frequencies of inflammatory and exhausted cell types, as well as telomere length of chromosomes in specific subpopulations of blood cells. However, much is still not known and we will evaluate reports of biomarkers of human stress in current literature for other promising clues that can be gained from blood samples. We will ask you to generate and test original hypotheses about stress biomarkers, introduce you to the techniques required to evaluate these markers, then set you free to evaluate your own questions and hypotheses through experimentation, from design through implementation and evaluation.

Lab philosophy.
Over the past fifty years since the discovery of the structure of the DNA double helix, there has been an explosion in new discoveries about how living organisms function. These discoveries have fueled great changes in medicine and human health and in our understanding of the world around us.

Yet these far reaching advances are primarily the result of reiterative application of the scientific method to biology on a very practical level by, summarized as follows:
1) identifying a key unanswered question;
2) choosing a suitable biological system to address this question;
3) designing experiments likely to yield informative results;
4) conducting the experiments;
5) interpreting the outcomes; and
6) sharing the results with the community.

BIOL425 is a course designed to employ all these aspects of the scientific method and experimental approach to try to address an unanswered question.

In BIOL425, we invite you to join in the process of scientific discovery. The course may be different from courses that you have taken previously in several important ways, so we would urge you to read the following overview carefully and make sure to discuss any of your questions with us and other students. Experimental science is all about collaboration and discussion; your thoughts and ideas matter!

BIOL425 is project-based. All experiments are designed (or will be designed by you!) with the goal of better understanding a specific biological process through answering a defined open question in this process. Each semester focuses on a different topic developed in collaboration with researchers active in that field, enabling us to conduct "real" science. Consequently, the choice of experiments, and the week-to-week progression of experiments are determined by the underlying scientific question and the evolution of your own projects.

You will get the most out of this class therefore if you prepare prior to class time, with an eye to making sure you are clear about what you will be doing (the techniques and experimental protocols) and particularly, why you are doing each step. Staying on top of both "the big picture" and the details is well worth the effort, and will be rewarded in the evaluation of your work. This project-based approach is an exciting alternative to "demonstration" type laboratory classes; we urge you to join in to make this class your own.

BIOL425 is open-ended. What draws many of us to experimental science is the allure of discovery. It is a very special feeling indeed to see a brand new, novel result and realize its implications, knowing that you and your colleagues are the only people in the world that know this. Of course, discoveries with major implications are rare, and are not going to occur on a weekly basis in a laboratory class. Nevertheless, since this is such a key element in our motivation for doing experiments, the experiments that we will conduct in class are novel- we do not know what the outcomes will be. As a consequence, we often do not know what the next step will be until we have obtained the result, so we will need to maintain some flexibility in scheduling as to what we will be doing in class time each week. Students are particularly encouraged to read all e-mail communications about the course carefully, and to check the course Canvas site frequently to stay abreast of any modifications to our overall plan. We hope that you will enjoy and contribute to this style of course, which we feel reflects the modern research experience.

BIOL425 encourages independence. Designing and conducting your own research can feel very intimidating. But like most research, the class will be conducted as a collaboration. The instructors have thought about suitable approaches to address the question and have prepared protocols. We will also prepare the reagents such that nearly all of the procedures can be conducted during class time. However, your participation is critically important. We fully ascribe to the notion that people learn best by doing, and that "doing" includes not only obtaining a result, but also the interpretation and evaluation of the result and subsequent experimental planning. So do not be surprised if we ask you for your interpretation or opinion about data generated in class and future experiments class planning before we give you our own. We may go with your suggestion. We ask because your intellectual input will help you make this project your own. We ask because experimental research is most successful when all contribute; your ideas are valuable! We are convinced that the extra time that discussion and collaborative work entails is fully worth it.

On another note, we also emphasize that you are free to pursue your projects further outside of class time as you wish. We have a fully equipped lab (Levin L57) available for your use (simply contact John, jwagner@sas.upenn.edu, to coordinate). Of course, this is not required- it is up to you. But should you wish to pursue any aspect of our project that interests you, you are most welcome.

Course expectations.
Your grade for this quarter will be based on five components:

1. Lab performance		20%
2. Lab notebook:		
	-mid semester	10%
	-semester end	10%
3. Lab reports:		
In your three reports (at 10% each), you will:		30%
a) Annotate a bibliography		
b) Generate hypotheses		
c) Present an experimental result		
4. Final presentation (oral)		30%

1. Lab Performance

Lab performance is a measure of several factors. These include:

a. an understanding of the theory (*why* we are doing what we are doing) and practice (the design and protocols) underlying the methods that we will be using in the laboratory;
b. the ability to carry out an experiment properly and efficiently;
c. the ability to think independently while working together effectively as a team;
d. respect for the safety and well-being of the other students in the laboratory.

2. **Lab Notebook**

You will be working in pairs in the BIO425 lab. However, each student must keep his or her own individual laboratory notebook, since an important aim of this laboratory is to learn how to keep complete and clearly-written experimental records. This year, you will be keeping your lab notebook online.

The following information should be recorded in your notebook:

a. The date. This will be recorded automatically online
b. A short (1-2 sentences) summary of the aim or goal of each experiment. Be sure to include the relevant detail that would be important to an outsider in understanding what ***and why*** you are doing the procedure (e.g. the *nam*e of the plasmid you are isolating, *why* you are conducting gel electrohoresis).
c. Log of procedures used. These can be copied and pasted from the Canvas protocols, but be sure to find a format for reviewing them carefully before class. *It is my experience that this brief preparation can save you a substantial amount of time in class!* As you do this, imagine yourself doing each step, and think about what questions you need to ask before you do. I personally prefer to write them out by longhand (as do many professional scientists) as a strategy to work through them, but there are other ways of preparing, of course. If you repeat a procedure later, you may simply record it as, e.g. "RNA purification, as per xxx" and make a link. ***Be sure to note any alterations you made in that protocol during the actual experiment.***
e. All data, calculations and results obtained at the time you obtain them. If you realize at a later date that you need to correct an entry made previously, add the correction and date it appropriately.
f. Comments on significant results, explanations for unexpected findings, etc . written while data are still fresh in your mind (don't wait until after dinner!).
g. Appropriate attribution. It will sometimes happen that one lab group will have difficulty with a particular experiment and may then obtain a reagent or even a set of results from another group. On other occasions, different groups each contributing part of an experiment will need to share data. These situations reflect what happens during collaborative science in the real world. It is therefore terribly important that you learn now to credit the people involved directly in your book. Complete and accurate citation is an important manifestation of your scientific integrity.
h. A brief and concise discussion of your data. At the end of each set of experiments, you need to briefly note down the answers to such questions as:

What did your experiments show?
Was the result what you expected? If so, why; if not, why not?
What problems did you encounter in doing the experiments? How would you alter the protocol for next time?

Please note, you should **never** be embarrassed to show your own data. A careful, thoughtful discussion of ambiguous or negative results is worth more in our book than a sloppy treatment of nice data. Consider your notebook to be your scientific diary. **You must never record your data in a separate notebook or on scraps of paper or paper towels and rewrite later!!** This wastes your time, results in needless mistakes and, most importantly, is regarded as poor, and potentially dishonest, scientific practice.
i. Files of your data.
During your experiments, you will accrue data files, that will be initially posted on Canvas ("our data" tab). Copies of this information should be annotated and incorporated in your lab notebook **as the experiments proceed**.

3. Assignments
The lab reports are a series of written assignments that will be due over the course of the semester. The assignments are designed to give you the opportunity to interpret and write about research. These assignments will be graded on a scale from 1-20 points, but if you pass in anything on time, you will receive a minimum of 14 points.

The first assignment we will ask you to develop an underlying bibliography for the project. You will annotate relevant literature that you and your partner have identified with an eye towards developing and articulating ideas for experimental exploration. Don't worry! Immunology is complicated but we are excited to work with each of you to draw you into this fascinating and important field.

For the second assignment (*essay format*), you will be asked to *identify* a research question of interest then *articulate* a specific, experimentally testable hypothesis of your own. The assignment due date is placed such that- through class discussion- we can hone your ideas into experimentally accessible and novel questions that we can then approach in class experiments. Project and experimental design are some of the most important and fun aspects of doing research.

For the third assignment (*essay format with one Figure*), you will have the opportunity to present one of your results in a standard written format (as you might see results presented in a published paper). Presentation and interpretation of your own data is a creative, challenging and exciting aspect of research science that you will engage in this tightly focused assignment.

4. Oral presentation of Lab Results.
An important part of scientific research is presenting *and discussing* your results with your colleagues, for example at a scientific conference. During the last week of the quarter, we would like for you to take this opportunity. Plan to make a short presentation to your classmates about your results. You may use Powerpoint, overheads and/or the blackboard as you like. This presentation should be about 15 minutes in length and ***explicitly*** present:1) ideas or hypotheses that you have tested; 2) the experimental results; and 3) your interpretation of the results.

Note that our goal is to engage one another in scientific discussion, so please plan to contribute accordingly!