Genomics Lab: Methods in Data Collection and Analysis

BSC 4445C-RI
Genomics Lab: Methods in Data Collection and Analysis
Spring 2022 - 4 credits

Dr. Anna Forsman (anna.forsman@ucf.edu)

Course Description
The field of genomics focuses on understanding the collective function of all components encoded in an organism’s genomic blueprint. Over the past decade, there has been an explosion of new and cost-effective methodologies to sequence the genetic material of life. Originally, high-throughput sequencing was slow, costly, and was used only to sequence the genomes of model organisms. Today genome-scale datasets are essential to most molecular biology applications in any taxonomic group, including phylogenetics and population genetics, metagenomic and targeted amplicon sequencing of entire biological communities, functional genomics of the expressed portion of the genome, and whole-genome assembly and characterization of an organism’s complete biological code. An integral, related, and emerging field of study is bioinformatics, which focuses on developing computational tools to analyze these massively large sequence data sets. In this class, we take a hands-on approach to understanding (1) how genomic datasets are generated in the lab, and (2) how they are analyzed computationally using bioinformatics pipelines. We begin, in both lecture and lab, with the fundamental biochemistry of DNA and the tools biologists have developed to isolate and manipulate genomic elements. We then scale up to address the “Next Generation” Sequencing (NGS) revolution, including how and why new methodologies have made genome-scale analyses achievable for nearly any organism, and the detailed methodologies and skills necessary to prepare samples for whole-genome, whole-transcriptome, and whole-community profiling (DNA metabarcoding). We spend the second half of the course learning about and utilizing bioinformatics pipelines to manipulate and analyze the sequencing datasets we have generated. Imparting a working knowledge of practical methods for generating and analyzing genomic datasets is the fundamental goal of this course. By generating, analyzing, and presenting (in both written and oral formats) novel genomic data, students will hone their writing, critical thinking and problem solving skills. The importance of genomics for all facets of life sciences will be emphasized, particularly the impacts of genomic datasets on recent advances in phylogenetics, evolutionary ecology, epigenetics, functional genomics, and health and medicine.

Research Intensive Course Designation
BSC 4445C is designated as a Research-Intensive (RI) course. This designation will be noted on your transcripts. Your active engagement in the research and/or creative scholarship process will be the core of your learning experience in this course. A significant portion of your grade for the Genomics Lab Course will be derived from both your active participation in the research process and the tangible course-related project(s) that comes out of said project. If you have any questions about this designation, please ask your course instructor.

Updated: 06 January 2022
Course Objectives

- To understand the molecular biology underlying existing methods for isolating, manipulating, and sequencing DNA and RNA
- To understand the NGS revolution and the computational demands of modern molecular analyses
- To understand the types of research and analyses that fall under the umbrella of genomics, including data generation, manipulation, and bioinformatics
- To gain hands-on skills in how to manipulate genome-scale datasets by conducting analyses of group project data using basic computer programming and established bioinformatics pipelines for metabarcoding taxonomic classification and analysis.
- To gain skills in how to present and communicate research by (1) creating figures, tables, and a written narrative in the style of a peer-reviewed publication and (2) presenting results in the format of a 15-minute oral powerpoint presentation using data and analyses from semester-long group research projects.
- To become a skilled molecular biologist actively engaging in scientific inquiry by conducting, in a group and with guidance, the (1) formation of a scientific hypothesis that utilizes genomic data, (2) maintenance of a laboratory notebook, and (3) successful performance of core molecular protocols, including:
  - purifying nucleic acids
  - quantifying nucleic acids
  - DNA amplification (endpoint PCR)
  - quantitative DNA amplification (qPCR)
  - gel electrophoresis
  - cDNA library construction
  - library preparation for NGS of targeted genomic regions (e.g., 16S r RNA gene) for compositional characterization of eukaryotic and microbial communities.

Prerequisites

A grade of B or better in undergraduate genetics or consent of the instructor. Completion of EHS Lab Safety Training online course and practical by end of add/drop period (mandatory in order to conduct research in the Genomics Core Facility where the course is held).

Class Meetings: M/W 1:30pm – 3:50pm

Lecture: Lectures will be posted to webcourses at the start of each week. Each lecture will be 60-90 minutes long and will cover the material that will appear on exams. Students will be required to complete a one-question assignment each week to indicated that they have watched the lecture.

Lab: Genomics is a research intensive course where you will have the opportunity to learn many new skills that will allow you to complete a group-based research project. Real lab work takes a lot of time and so we will be using the entirety of our class meetings for hands-on lab work.

Updated: 06 January 2022
Research activities often do not fit perfectly into scheduled time blocks due to the trial-and-error nature of conducting protocols on novel samples and hypotheses where outcomes are unknown. Thus, the Genomics Core Facility will also be open throughout the week for students to complete protocols or re-do failed samples; during this time, the lab manager, GTA, and/or instructor will be present to answer questions and provide guidance. Additional lab time should be scheduled with the instructor.

**Lecture Instructor:** Dr. Anna M. Forsman

Office: Biology 439, 407-823-0766  
E-mail: Anna.Forsman@ucf.edu  
Office/Lab Hours: M/W 11:30pm – 1:30pm and by appointment. During office hours, Anna will either be in her office (Bio 439) or prepping for class in Bio 414. Virtual meetings via Zoom can also be arranged by appointment.

**Teaching Assistants**

GTA: George Zaragoza  
Email: george.zaragoza@Knights.ucf.edu

UTA: Devin Burris  
E-mail: devinburris@Knights.ucf.edu

Teaching assistants will help maintain lab and reagents for the course and will help students with lab procedures during class. They will also be available for questions, troubleshooting and help with protocols outside of class hours by appointment.

If you would like to schedule a meeting or labtime outside of class hours, please do so in advance by email to ensure that someone is available to help you during that particular time. This will avoid you having to wait for assistance if Dr. Forsman or the TA’s are in meetings or working with other students.

**Webcourses Site**

There is a course web site available through Webcourses (https://webcourses.ucf.edu) that will be used to post materials for the course, including the syllabus, assignment due dates, lecture presentations, and grades.


This is a fantastic resource, and the best part is that you do not need to purchase this textbook. The e-book version of this text is available for free through the UCF Library. This textbook provides excellent conceptual background for the DNA metabarcoding protocols that we will be using throughout your research projects.

**Class Policies**

1. **Attendance** is not strictly required but will contribute to each student’s participation grade. A large portion of the grading for this hands-on methodology
course will be based on lab participation, and success in this class requires completion of lab procedures. It is the student’s responsibility to arrange to make up any missed work outside of class hours.

With that said, we are in the midst of a global pandemic and it is not unlikely that some of us will become sick or exposed to COVID during this semester. If you are sick, please do not come to class. If you test positive for COVID, please call the UCF COVID line at 407-823-2509. Students will be working in groups of 2-3 on research projects and so lab work will be completed even in your absence. Please contact the instructor and we will make arrangements to catch you up over Zoom.

The last few years have been difficult for many of us because of COVID, and this is likely to continue throughout Spring 2022. As your instructor and research mentor, I want to facilitate your success in this course. If you are struggling outside of school in a way that impacts your well-being or performance in this course, please let me know. We will work together to make sure that you can be successful and that you can get the most out of this course as possible.

2. **Exams** will be administered through webcourses. The format for these exams will be a) fill in the blank, b) short answer, and c) essay. You may use course materials and your notes while taking these exams, but you may not consult with each other or anyone else while taking an exam. Please notify me if you are experiencing circumstances that prevent you from completing these online assessments during the defined time-frame so that we can coordinate.

3. Assigned readings should be completed before attending class and will be provided via webcourses or handed out in class.

4. You are encouraged to discuss any and all portions of the class with me. Please feel free to come to my office hours or make an appointment to discuss the class, especially if you are having trouble.

5. Respect should be given to fellow students, the TA’s, and the instructor. Please do not arrive late to lab or leave early.

6. Hateful or offensive speech or writing will not be tolerated.

7. Cell phones, iPods, and other electronic devices should be silenced and put away before class starts.

8. Academic dishonesty (cheating and plagiarism) is strictly prohibited and will be taken very seriously and will result at least in an "F" for that assignment (and may, depending on the severity of the case, lead to an "F" for the entire course) and may be subject to appropriate referral to the Office of Student Conduct for further action. See the UCF Golden Rule for further information.

**Course Accessibility**

My goal for this class is to provide an accessible and welcoming experience for all students, including those with disabilities that may impact learning in this class. If anyone
believes the design of this course poses barriers to effectively participating and/or
demonstrating learning in this course, please meet with me to discuss reasonable
options or adjustments. You may also contact SAS (Ferrell Commons 185; 407-823-
2371; sas@ucf.edu) to talk about academic accommodations.

Grading

Grades will be assigned according to the following scale:
A: 90-100; B: 80-89; C: 70-79; D: 60-69; F: <60

The final grade for this course will be based on six components:

<table>
<thead>
<tr>
<th>Assignment</th>
<th>% of final grade</th>
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<tbody>
<tr>
<td>Week 1 attendance assignment</td>
<td>1%</td>
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<tr>
<td>Exams (4)</td>
<td>20%</td>
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<tr>
<td>Participation</td>
<td>30%</td>
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<tr>
<td>Lab notebook</td>
<td>20%</td>
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<tr>
<td>Final report</td>
<td>19%</td>
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<tr>
<td>Oral Presentation</td>
<td>10%</td>
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</tbody>
</table>

(1) Week one attendance assignment on Webcourses to determine work groups (1%)

(2) Four semester exams will be given in class on the dates indicated on the schedule
(5% each; **20% total**). They will consist of fill in the blank, short answer, and problem-
solving questions (essay) based on lecture material. The purpose of these exams is to
challenge students to synthesize and apply their knowledge at a level that may be
expected of independent student researchers and graduate students. Students will be
assessed on their ability to understand genomics lab methodologies, the molecular
biology behind how those methodologies work, and how genomic datasets are analyzed
and interpreted.

(3) Students will be graded on **lab participation (30% total)**, including asking questions,
performing protocols, and generating the necessary data for each week to move forward
to the next lab session. Additional lab time outside of the scheduled class hours will be
necessary to complete protocols for the majority of labs. Make-ups during office hours
will be offered for up to two missed lab sessions; additional missed lab sessions will
each result in a 5% drop in participation grade.

(4) The **lab notebook**, to be maintained by each group of three students working on a
unique project, will be evaluated throughout the semester and given a letter grade (**20%
total**). The lab notebook record keeper will rotate weekly among the three group
members during the nine weeks of molecular lab, and each student will be graded
individually on their own entries. Students will be given feedback on how to improve their
note-taking and record-keeping of all lab activities and results. At any time before lab
notebooks must be turned in, students can also check with the instructor to determine
whether lab notebook content is sufficient or lacking, and the instructor will provide
feedback. The final lab notebook grade will be based on both mid-semester notebook
grades as well as improvement over the course of the semester.
(5) Each group will submit a **final report (19%, group receives one grade)** detailing the results of their research project. The report must be written in the format of a scientific journal article (i.e., manuscript), including the following sections: Abstract (5 pts) Introduction (10 pts), Methods (25 pts), Results (with figures/tables; 35 pts) and Discussion (10 pts). Groups must turn in drafts of the introduction, methods and results sections on the dates listed in the schedule below. Drafts will be evaluated and the instructor will give feedback on how to make improvements before the final draft is due. Groups will receive 15 pts of their overall final report grade for turning in drafts of these sections (5 pts for each deadline and section).

(6) During the final exam period, each group will give a 15-minute **oral presentation (10% total)** on their project, including powerpoint slides illustrating their research. The presentation must include the following sections: overview (10 pts), introduction (20 pts), methods (30 pts), results (30 pts), and conclusions (10 pts). Each student will speak for 5 minutes and be graded individually. Students may opt to pre-record their presentation using Zoom or a similar platform.

**Schedule:**

*The following schedule is approximate and dates may be changed at any time.*

**Week 1:**
- **Lecture 1:** Intro and overview of course, overview of two focal project types (microbial and parasite metabarcoding) and methods to be used. **Week 1 webcourses assignment due.**
- **Molecular Lab 1:** Keeping a lab notebook, lab safety and etiquette, and pipetting.

**Week 2:**
- **Lecture 2:** Basics of molecular biology; isolating and characterizing nucleic acids
- **Molecular Lab 2:** DNA extractions. Quantitative and qualitative analysis of DNA using MicroSpot plate on the 96-well plate reader

**Week 3:**
- **Lecture 3:** Polymerase Chain Reaction (PCR) and electrophoresis.
- **Molecular Lab 3:** DNA extractions. Quantitative and qualitative analysis of DNA using MicroSpot plate on the 96-well plate reader

**Week 4:**
- **Lecture 4:** Genomic and cDNA libraries. **Exam 1 (Wed).**
- **Molecular Lab 4:** DNA extractions. Quantitative and qualitative analysis of DNA using MicroSpot plate on the 96-well plate reader

**Week 5:**
- **Lecture 5:** Overview of Sanger and next-generation sequencing
- **Molecular Lab 5:** PCR1 to amplify genomic targets based on project track, bead cleanups to remove primer dimers. PCR visualization with gel

**Week 6:**
- **Lecture 6:** Overview of Next Generation Sequencing technologies. **Project outline due**
Molecular Lab 6: PCR1 to amplify genomic targets based on project track, bead cleanups to remove primer dimers. PCR visualization with gel

Week 7:
Lecture 7: Genomic analysis 1: Quality control, NCBI, GenBank, and BLAST;
Molecular Lab 7: PCR2 to attach sequencing adapters and indexes, bead cleanups.

Week 8:
Lecture 8: Genomic analyses 2: Bioinformatics overview; Exam 2!
Molecular Lab 8: Library quantification by Quibit, completion of pooling calculations, preparation and QC (TapeStation) of group pools and QC by TapeStation.

Week 9: Spring Break!

Week 10: Lecture 9: Whole genome assembly, genomes and genome browsers
Molecular Lab 9: qPCR on serial dilutions of group library pools to precisely quantify the amount of available material for sequencing, equimolar pooling of group libraries, final prep of libraries for sequencing, MiSeq Run!

Week 11:
Lecture 10: Genome-scale analyses: RadSeq, SNPs and GWAS.
Computer Lab 1: Intro to Geneious and gene identification using GenBank

Week 12:
Lecture 11: Microarrays, transcriptomics, and RNAseq Exam 3 (Wed)
Computer Lab 2: basics of command line, bioinformatics pipeline tutorials for QIIME2

Week 13:
Lecture 12: Metagenomics and metabarcoding
Computer Lab 3: Manipulating Illumina data and Quality Control (QC) using FastQC; start running pipeline on lab-generated data (students work in groups).

Week 14:
Lecture 13: Presenting genomic data
Computer Lab 4: Continue running pipelines on lab-generated data; analyze results/generate summary statistics (students work in groups). FULL DRAFT DUE (Wed)

Week 15:
Lecture 14: Exam 4 (Wed)
Computer Lab 5: Finish/troubleshoot lab-generated data analysis; finish analyzing results and work on tables, figures and reports (students work in groups).

Week 16:
Monday, April 25: FINAL REPORT DUE & Final group presentations

Updated: 06 January 2022
The following schedule is approximate and dates may be changed at any time.

<table>
<thead>
<tr>
<th>Week</th>
<th>Date</th>
<th>Topic</th>
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<tbody>
<tr>
<td>1</td>
<td>10 Jan M</td>
<td>Lecture: Class intro ~ lab project intro ~ overview of lab notebooks</td>
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<tr>
<td></td>
<td>12 Jan W</td>
<td>Group project placements ~ Pipetting and dilutions ~ webcourses assignment</td>
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<tr>
<td>2</td>
<td>17 Jan M</td>
<td><strong>No Class – Martin Luther King Day</strong></td>
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<tr>
<td></td>
<td>19 Jan W</td>
<td>Lecture/Lab: Molecular biology ~ DNA and RNA extractions</td>
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<tr>
<td>3</td>
<td>24 Jan M</td>
<td>Lecture: Polymerase Chain Reaction (PCR) and electrophoresis</td>
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<tr>
<td></td>
<td>26 Jan W</td>
<td>Lab: DNA extractions ~ DNA quantification</td>
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<tr>
<td>4</td>
<td>31 Jan M</td>
<td>Lecture: Genomic and cDNA libraries</td>
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<tr>
<td></td>
<td>2 Feb W</td>
<td>Lab: DNA extractions ~ DNA quantification. <strong>Exam 1</strong></td>
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<tr>
<td>5</td>
<td>7 Feb M</td>
<td>Lecture: Sequencing technologies I</td>
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<tr>
<td></td>
<td>9 Feb W</td>
<td>Illumina library prep week 1 (PCR1, bead cleanups, quantification)</td>
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<tr>
<td>6</td>
<td>14 Feb M</td>
<td>Lecture: Sequencing technologies II</td>
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<tr>
<td></td>
<td>16 Feb W</td>
<td>Illumina library prep week 2. <strong>Project outline due.</strong></td>
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<tr>
<td>7</td>
<td>21 Feb M</td>
<td>Lecture: Genomic analysis I</td>
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<tr>
<td></td>
<td>23 Feb W</td>
<td>Illumina library prep week 3 (PCR2, bead cleanups, quantification)</td>
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<tr>
<td>8</td>
<td>28 Feb M</td>
<td>Lecture: Genomic analysis 2</td>
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<tr>
<td></td>
<td>2 Mar W</td>
<td>Continue Illumina library prep as needed. <strong>Exam 2.</strong></td>
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<tr>
<td>9</td>
<td>7-9 March</td>
<td><strong>SPRING BREAK!</strong></td>
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<tr>
<td>10</td>
<td>14 Mar M</td>
<td>Lecture: Whole genome assembly, genomes and genome browsers</td>
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<tr>
<td></td>
<td>16 Mar W</td>
<td>Library QC ~ library pooling ~ MiSeq sequencing</td>
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<tr>
<td>11</td>
<td>21 Mar M</td>
<td>Lecture: Genome-scale analyses: RadSeq, SNPs and GWAS</td>
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<tr>
<td></td>
<td>23 Mar W</td>
<td>Intro to Geneious &amp; gene identification using GenBank ~ Work on tutorials</td>
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<tr>
<td>12</td>
<td>28 Mar M</td>
<td>Lecture: Genome-scale analyses: Microarrays, transcriptomics, and RNAseq</td>
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<tr>
<td></td>
<td>30 Mar W</td>
<td>Finish pipeline tutorials ~ QC and manipulation of Illumina data. <strong>Exam 3.</strong></td>
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<tr>
<td>13</td>
<td>4 Apr M</td>
<td>Lecture: Metagenomics and metabarcoding</td>
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<td></td>
<td>6 Apr W</td>
<td>Lab: Data analysis</td>
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<tr>
<td>14</td>
<td>11 Apr M</td>
<td>Lecture: Presenting genomic data</td>
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<tr>
<td></td>
<td>13 Apr W</td>
<td>Continue analysis of project data ~ <strong>FULL DRAFT DUE</strong></td>
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<tr>
<td>15</td>
<td>18 Apr M</td>
<td>Presenting genomic data ~ Compete analysis of project data. <strong>Exam 4.</strong></td>
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<tr>
<td></td>
<td>20 Apr W</td>
<td>Complete analysis of project data</td>
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<tr>
<td>16</td>
<td>24 Apr</td>
<td>1:30pm-3:50pm <em>FINAL PRESENTATIONS</em>FINAL REPORT DUE*</td>
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