

## **MCB 5427-002: Real-Time PCR Techniques (Spring 2020)**

Instructor: Dr. Jeremiah Marden ([jeremiah.marden@uconn.edu](mailto:jeremiah.marden@uconn.edu))

Assistant: Todd Testerman ([todd.testerman@uconn.edu](mailto:todd.testerman@uconn.edu))

### **Course Overview**

This course offers an introduction to the key concepts of real-time PCR. The course includes a strong theoretical background of real-time PCR in a form of lectures. Students will have an opportunity to learn about practical applications of real-time PCR methods through demonstrations that will available online.

### **Objectives**

At the conclusion of this course, students should be able to perform the following tasks:

- Determine if real-time PCR is the best method to address a given problem
- Design and perform real-time PCR experiments
- Analyze real-time PCR data

### **Course Overview**

At 4:30 pm on April 24<sup>th</sup> a live question/answer period will be held using HuskyCT that will last ~30 min to describe the online format of the course and address questions. An additional question and answer period will be held at the conclusion of the course from 2 to 4 pm on Sunday, April 26<sup>th</sup>. Details regarding the set-up of these live sessions will be provided prior to the beginning of the course.

The outline below describes the general format of the course. The videos of lectures and lab techniques demonstrations should be watched in the indicated order, and the videos can be watched at any time on the indicated days (the videos for April 26<sup>th</sup> must be watched prior to the question and answer session). Details on how to access the videos will be provided prior to the beginning of the course.

### **Day 1 (April 24)**

1. Live question/answer session on HuskyCT (4:30 pm); brief overview of the course and its format
2. Lecture to introduce real-time PCR techniques and project overview

### **Day 2 (April 25)**

1. Lecture on RNA extraction and cDNA generation
2. Total RNA extraction
3. Generation of cDNA
4. Semi-quantitative PCR and analysis

5. Lecture on qPCR
6. Set up and qPCR assay
7. Lecture on droplet digital PCR (ddPCR) and analysis

### **Day 3 (April 26)**

1. Lecture on qPCR data analysis
2. Gel electrophoresis of qPCR samples
3. qPCR data analysis and interpretation of results
4. Live question/answer session on HuskyCT (2-4 pm)

### **Grading**

#### Quizzes (40%)

Short quizzes designed to assess knowledge of protocols

#### Exam (40%)

The exam will consist of multiple choice and short essay questions

#### Wet lab write-up (20%)

The data from the experiment will be provided to you, and you will be responsible for writing a short write-up of the study in the following format.

- **Purpose:** Provide a brief description of the goal of this module/experiment.
- **Procedure:** Describe the general workflow of the lab. Feel free to write this information in the format of a flowchart showing your experimental plan overview.
- **Results:** Provide a formal presentation of the analysis of your raw data that can include graphs, summary tables or other figures.
- **Discussion and Conclusions:** You should have some interpretation of your data and final conclusions from the experiment here. It is useful to comment on whether the goal/purpose of the lab was achieved and why or why not that was the case. In addition, note any sources of error and propose changes for future work.