Biochemistry & Molecular Biology (BMB) 829
Special Problems in Macromolecular Analysis and Synthesis
Fall Semester 2022

Module 2: Recombinant DNA and Genome Editing
Course Syllabus, Policies, and Schedule

Credit Hours: 1
Course meeting days and time: Monday & Wednesday 2:40 – 4 pm
Course location: BCH 111

Instructors

Charles G. Hoogstraten; BMB 829 Course Coordinator
hoogstr3@msu.edu
302D Biochemistry Building
(517) 353-3978 (only checked occasionally)
Office hours: By appointment

Lee Kroos; BMB 829-302 Module 2 Lead Instructor
kroos@msu.edu
422 Biochemistry Building
Office hours: By appointment

Shireesh Srivastava; Module 2 Instructor; Recombinant DNA
srivas14@msu.edu
313 Biochemistry Building
Office hours: By appointment

Elena Demireva; Module 2 Instructor; Genome Editing
demireva@msu.edu
1111 IQ / Bio Engineering Facility
Office hours: By appointment
Course Description and Objectives

BMB 829 is a modular course that seeks to introduce students to modern molecular and structural/analytical techniques of interest in the biochemistry and molecular biology laboratories. Students may enroll in each of sections 301, 302, 303, 304, and 305 (corresponding to modules 1 through 5) once for a total of up to five credits; however, *Module 1 must be completed before any of the remaining modules can be taken, unless an exception has been granted due to a class scheduling conflict.*

**Required Textbook & Course Materials:**
No required textbooks; required reading material will be provided by instructors on D2L.

**Format:**
This course will be taught in-person, with any exceptions announced by the instructor both in class and on D2L. Because of effects of the ongoing COVID pandemic, lectures will be recorded (audio and video feeds) and posted to D2L whenever practicable. These recordings are not a replacement for class attendance, but students are encouraged to make use of them in cases of known or suspected COVID exposure or the observation of symptoms in order to avoid possible viral spread. Note that lecture recordings will not be possible in cases of hands-on workshops, facility visits, or technical difficulties.

**Required Technologies:**
This course will make extensive use of the D2L platform at Michigan State (d2l.msu.edu) to communicate course materials of various sorts. An internet connection and device capable of downloading documents, displaying Microsoft Word and PowerPoint and Adobe PDF documents and displaying video are required.

**Recommended Texts & Other Materials:**
Additional recommended reading and viewing materials will be provided on D2L.

**COVID-19 Statement:**
BMB 829 fully supports and expects compliance with all stated University policies relating to infectious disease safety. Students declining to follow guidelines relating to appropriate viral safety will be asked to leave the classroom.

The BMB 829 instructors are fully cognizant of the difficulties the pandemic has posed for many students. Our goal is to see all of our students succeed despite the ongoing challenges of the pandemic. Should you find yourself in scheduling, academic, or mental or emotional health difficulties, you are urged to contact the current course instructor and Prof. Hoogstraten as soon as possible so that appropriate accommodations may be made. In addition, MSU has made extensive resources available through the Keep Learning section of the University website, including academics ([https://remote.msu.edu/learning/additional-resources.html](https://remote.msu.edu/learning/additional-resources.html)) and for students facing challenges related to mental health ([https://remote.msu.edu/learning/mental-health.html](https://remote.msu.edu/learning/mental-health.html)). Students are encouraged to make full use of any or all of these resources as the need arises.
Learning Continuity Statement:
Should students be unable to attend class for an extended period of time, they should communicate this to the Course Coordinator (hoogstr3@msu.edu) and/or lead instructor for the module (kroos@msu.edu) as soon possible once the situation becomes evident. Students should work with course instructors to develop a schedule for regular communication and reasonable timelines for completing assignments including exams.

Course Continuity Statement:
Should an instructor be required to be absent for an extended period of time, scheduling of different course modules may be adjusted accordingly. Students may communicate with either the Course Coordinator or any of the other course instructors regarding grading and assessment modifications.

Prerequisites:
Recommended background equivalent to BMB 462. For all modules except Module 1, prior completion of Module 1 is expected, unless an exception has been granted due to a class scheduling conflict.

Late Work Policy:
Full credit for late submissions will only be considered if arranged with the instructor in advance of the due date. Otherwise, point penalties may be assigned or late work may not be accepted at the discretion of the individual instructor. Communication with the instructor about situations leading to late work as soon as possible is strongly advised.

Student Expectations:
All participants in this class are held to the standard set by MSU’s Policy on Integrity of Scholarship and Grades. The policy can be read in full at the MSU Ombudsperson’s website.

On March 22, 2016, The Associated Students of Michigan State University (ASMSU) adopted the following Spartan Code of Honor:

“As a Spartan, I will strive to uphold values of the highest ethical standard. I will practice honesty in my work, foster honesty in my peers, and take pride in knowing that honor is worth more than grades. I will carry these values beyond my time as a student at Michigan State University, continuing the endeavor to build personal integrity in all that I do.”

Disability Access:
Students must inform the instructor and course coordinator at the beginning of the semester, or as soon as reasonably possible after a situation arises during the semester, of any accommodations needed. Information related to disability access is available on the Resource Center for Persons with Disabilities (RCPD) website. Students: to make an appointment with a specialist, call: (517) 353-9642 Or TTY: (517) 355-1293 or visit the RCPD website.
Module Outline and Schedule

Assessments:
Grading for the module will be based on homework assignments (50%), in-class exams (40%), and in-class participation (10%). Dr. Srivastava’s part will count for 55% of the final grade and Dr. Demireva’s part for 45%. Each part will have two homework assignments and one in-class exam. These will be aimed at assessing basic concepts and understanding through problem-solving questions and/or reading of assigned original research papers or review articles. Letter grades will be assigned at the end of the semester and will be curved based on the final distribution of student scores.

Course Outline:
The planned schedule of lecture presentations is presented in the table below, with class meeting number, dates, instructors, and lecture topics.

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<tr>
<th>#</th>
<th>Date</th>
<th>Instructor</th>
<th>Topic</th>
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<tbody>
<tr>
<td>1</td>
<td>Wednesday, October 5</td>
<td>Srivastava</td>
<td><strong>Gene cloning: tools and analysis</strong> - Overview of steps in a typical cloning experiment, Restriction enzymes and their classes, Plasmid vectors – origin of replication/copy number, selectable marker(s), cloning sites, Polymerase chain reaction (PCR) – basics, design of primers, applications and pitfalls, common problems and solutions, Gel electrophoresis of DNA fragments – sample preparation, size determination and quantification, fragment purification</td>
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<td>2</td>
<td>Monday, October 10</td>
<td>Srivastava</td>
<td><strong>Gene cloning: traditional methods</strong> - Ligation cloning – compatible DNA ends, end modifications, T4 DNA ligase, ligation reaction conditions, Transformation – competent cells, electroporation, Analysis of transformants (clones) – colony PCR, DNA mini-preps, restriction digestion, DNA sequencing and analysis, choice of DNA sequencing methods</td>
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<td>3</td>
<td>Wednesday, October 12</td>
<td>Srivastava</td>
<td><strong>Gene cloning and alteration: advanced methods</strong> - Gibson assembly cloning – primer design, PCR conditions, isothermal assembly reaction conditions, Gateway cloning and Golden Gate Assembly, Gene Synthesis – its advantages and challenges, Overview of site-directed mutagenesis, Plasmid PCR – primer design, PCR conditions, gel analysis, DpnI digestion, Sequence overlap extension – primer design, PCR conditions, gel analysis</td>
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<td>4</td>
<td>Monday, October 17</td>
<td>Srivastava</td>
<td><strong>Gene expression</strong> - Overview of protein production, Plasmid vectors – inducible promoters, tags for protein purification, <em>E. coli</em> host strains, Codon Optimization to Express heterologous proteins, Culture/induction conditions, Gel electrophoresis of proteins – sample preparation, size determination and quantification, protein staining, immunoblot, Protein purification – metal-affinity chromatography, other affinity methods</td>
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<td>5</td>
<td>Wednesday, October 19</td>
<td>Srivastava</td>
<td><strong>Ethics, regulation and applications of recombinant DNA research</strong> – Ethical use of rDNA technology, regulations governing rDNA research; Applications of recombinant DNA technologies in various domains of biology</td>
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<td>6</td>
<td>Monday, October 24</td>
<td>Demireva</td>
<td><strong>Introduction to Genome Editing</strong> - review of canonical and recent transgenic and genome editing methods and approaches – retroviruses, transgenes, homologous recombination, site-specific nucleases. Editing in</td>
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different model organisms. Temporal and spatial control of gene and transgene expression.

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<tr>
<th>Date</th>
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<th>Demireva</th>
<th>Topic</th>
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<tr>
<td>7</td>
<td>Wednesday, October 26</td>
<td>CRISPR-Cas Genome Editing technologies – background on CRISPR systems in bacteria and adaptation for gene editing in eukaryotes. CRISPR experimental methods and approaches - using CRISPR platforms, design, cloning and delivery of genome editing reagents, screening and validation of CRISPR edits, indel analysis, off-target analysis.</td>
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<td>9</td>
<td>Wednesday, November 2</td>
<td>Demireva</td>
<td>Applications of CRISPR technologies and beyond – diagnostic and sensing applications, biomedicine and biotechnology, plants and agriculture, CRISPR screens. Future genome editing tools beyond CRISPR – exploiting other mobile genetic elements systems for larger scale genome editing.</td>
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