

Chemistry 437/837: Chemical Biology Laboratory

Fluorescent biomolecule design and application

University of Nebraska – Lincoln

Spring 2021



Instructor: Dr. Catherine Eichhorn, Assistant Professor of Chemistry

Class meets: Tuesdays and Thursdays 9:30 am – 12:20 pm in Hamilton 305

How to reach me: email on Canvas or ceichhor@unl.edu *include CHEM837 in subject line*



Virtual student (office) hours: *By appointment*

Teaching Assistant: Gloricelly Roman, Contact on Canvas

COURSE DESCRIPTION

This course integrates chemical synthesis, biomolecule structure and ligand recognition, and biological applications to advance understanding of the rational design of fluorescent biomolecules.

In this course, you will be performing novel research and forming your own hypotheses. Like research in the real world, experiments may fail and/or you may have negative results – that is ok! Your work in this class will lead to generating new knowledge in the areas of fluorogen synthesis, RNA aptamer design, RNA-small molecule interactions, and more. Your contributions may result in authorship in peer-reviewed publications.

REQUIRED RESOURCES

1. Truong, L. and Ferre-D'Amare, A.R. (2019) From fluorescent proteins to fluorogenic RNAs: Tools for imaging cellular macromolecules. *Protein Sci*, **28**, 1374-1386.
2. Chen, X., Zhang, D., Su, N., Bao, B., Xie, X., Zuo, F., Yang, L., Wang, H., Jiang, L., Lin, Q., Fang, M., Li, N., Hua, X., Chen, Z., Bao, C., Xu, J., Du, W., Zhang, L., Zhao, Y., Zhu, L., Loscalzo, J. and Yang, Y. (2019) Visualizing RNA dynamics in live cells with bright and stable fluorescent RNAs. *Nat Biotechnol*, **37**, 1287-1293.
3. Lab manual (on Canvas)

EARNING YOUR GRADE

Your course grade is based on your performance on various course assessments, including formative and summative assessments as outlined below. Your grade is not dependent on experiment success.

Formative assessments

- Pre-lab and post-lab assignments
- Lab notebook

Summative assessments

- End-of-module lab reports detailing experiment procedures, measurements, observations, and results
- Final report/proposal (required for students enrolled in 837 only)

GRADING SCHEME

Assignment	Point Allotment	Points Earned	Percent	Letter Grade
Final report & proposal	250	970–1000	97.0+	A+
End-of-Module reports	250	930–969	93.0+	A
Lab notebook	200	900–929	90.0+	A–
Pre/post-lab assignments	200	870–899	87.0+	B+
Lab safety and participation	100	830–869	83.0+	B
		800–829	80.0+	B–
Total Points Available	1000	770–799	77.0+	C+
		730–769	73.0+	C
		700–729	70.0+	C–
		670–699	67.0+	D+
		630–669	63.0+	D
		600–629	60.0+	D–
		< 600	< 60.0	F

Because life happens:

- Notify the instructor if you are sick or are in quarantine to make arrangements for make-up lab
- Lowest 2 pre-lab assignments are dropped
- All late assignments will have maximum late penalty of 20% and can be turned in anytime before the last day of class
 - Assignment is turned in between one minute and one week late : 10% penalty
 - Assignment is turned in after one week late: 20% penalty

ASSIGNMENTS

Pre/post-lab assignment

Several experiments require a pre-lab assignment to be completed and submitted through Canvas. These assignments are designed to prompt you to read through the procedure, understand the purpose and expected outcomes of the experiment, and form hypotheses. Post-lab assignments will be turning in data (e.g. NMR spectrum, spreadsheet of data points).

Lab notebook

It is essential to document a complete recording of all experimental conditions and observations. At the end of each class period your lab notebook will be submitted for grading. Grading will be based on legibility, complete documentation of procedures (and deviations), documentation of observations, and inclusion of data and analysis (e.g. reporting yield, NMR spectrum, picture of gel)/

End-of-module reports

At the end of each module, a written report of results and conclusions will be submitted. These reports are structured for you to review the experiments performed and data collected, interpret and critically evaluate the data, and form conclusions in the context of your initial hypothesis. Each report is worth 50 points.

FINAL REPORT & PROPOSAL

The final project is a combination report and proposal that will combine your end-of-module reports and ask you to propose new experiments to move the project forward.

CLASSROOM POLICIES & RESOURCES

Class format:

The course format is in person. Make up labs are available – it is important to notify the instructor prior to absence to discuss make-up options.

Making up labs:

Contact the instructor if you will miss a lab to make arrangements to make up the lab.

Attendance policy:

If you will miss class during class time (in person or online) you must notify the instructor or TA.

If you do not feel well **DO NOT** come to class in person. Students who are sick or who are engaging in self-quarantine in accordance with guidance from the Lincoln-Lancaster County Health Department or their health care professional **should not physically attend in-person classes**. You can receive a free COVID19 test on-campus: <https://covid19.unl.edu/campus-covid-19-testing>

Health and safety:

Classrooms have health & wellness stations by the doors: apply hand sanitizer when entering the room, and use disinfectant to wipe down your desktop surface. Face coverings are required inside any University building, including Hamilton Hall. Students who do not comply with the University policy regarding wearing face coverings will be asked to leave the class. See University Policy below regarding face coverings.

Accessibility & Accommodations

There are several resources to ensure you can learn and perform coursework in a safe environment. Please consult <https://covid19.unl.edu/students-and-covid-19-risk> for guidance on how to receive accommodations you may need. Please also let the instructor know privately so reasonable accommodations can be made. See the University Policy below (American Disabilities Act) for more information. If you have trouble acquiring the resources needed for the class, especially textbook and computer, please let the instructor know immediately so accommodations can be made. The UNL Library has laptop to check out (<https://libraries.unl.edu/laptops>).

Academic Integrity

Your intellectual growth depends on responsibility, honesty, and doing your own work. Presenting the work of others as your own by taking ideas from others (plagiarism) or copying other's work is dishonest, hurts your reputation and credibility, and will result in a failing grade on the assignment and potentially disciplinary action. See the University Policy below on Academic honesty for more information.

Health and well-being

These are not normal times, and this is not a normal semester. Everyone has been under a great deal of stress the past several months and for the foreseeable future. Please be kind to yourself, and to others. If you are struggling please reach out to the instructor. Please see the University Policy on Counseling and psychological services for more information.

UNIVERSITY POLICIES & RESOURCES

Academic honesty

Academic honesty is essential to the existence and integrity of an academic institution. The responsibility for maintaining that integrity is shared by all members of the academic community. The University's Student Code of Conduct addresses academic dishonesty (<https://studentconduct.unl.edu/student-code-conduct>). Students who commit acts of academic dishonesty are subject to disciplinary action and are granted due process and the right to appeal any decision.

Incomplete policy

A grade of Incomplete means that some of the coursework was not completed. Therefore, a grade of "I" will be given only if the student has completed over 75% of the course material, earning a B- or better. To make up an incomplete means that you only re-take only the portion of the course that you missed. Incompletes will only be submitted for students who are unable to complete the final 25% of the course because of illness, military service, hardship, a death in the immediate family, or other serious life problem.

American Disabilities Act

The University strives to make all learning experiences as accessible as possible. If you anticipate or experience barriers based on your disability (including mental health, chronic or temporary medical conditions), please let me know immediately so that we can discuss options privately. To establish reasonable accommodations, I may request that you register with Services for Students with Disabilities (SSD). If you are eligible for services and register with their office, make arrangements with me as soon as possible to discuss your accommodations so they can be implemented in a timely manner. SSD contact information: 117 Louise Pound Hall; 402-472-3787.

Counseling and psychological services

UNL offers a variety of options to students to aid them in dealing with stress and adversity.

Counseling and Psychological & Services (CAPS) is a multidisciplinary team of psychologists and counselors that works collaboratively with Nebraska students to help them explore their feelings and thoughts and learn helpful ways to improve their mental, psychological and emotional well-being when issues arise. CAPS can be reached by calling 402-472-7450, or by scheduling an appointment online at caps.unl.edu.

Big Red Resilience & Well-Being (BRRWB) provides one-on-one well-being coaching to any student who wants to enhance their well-being. Trained well-being coaches help students create and be grateful for positive experiences, practice resilience and self-compassion, and find support as they need it. BRRWB can be reached by calling 402-472-8770, or by scheduling an appointment online at resilience.unl.edu.

Required use of face coverings for on-campus shared learning environments

As of July 17, 2020 and until further notice, all University of Nebraska–Lincoln (UNL) faculty, staff, students, and visitors (including contractors, service providers, and others) are required to use a facial covering at all times when indoors except under specific conditions outlined in the COVID 19 face covering policy found at: <https://covid19.unl.edu/face-covering-policy>. To protect the health and well-being of the University and wider community, UNL has implemented a policy requiring all people, including students, faculty, and staff, to wear a face covering that covers the mouth and nose while on campus. The classroom is a community, and as a community, we seek to maintain the health and safety of all members by wearing face coverings when in the classroom. Failure to comply with this policy is interpreted as a disruption of the classroom and may be a violation of UNL's Student Code of Conduct.

Individuals who have health or medical reasons for not wearing face coverings should work with the Office of [Services for Students with Disabilities](#) (for students) or the Office of [Faculty/Staff Disability Services](#) (for faculty and staff) to establish accommodations to address the health concern. Students who prefer not to wear a face covering should work with the course instructor to arrange a fully online course schedule that does not require their presence on campus.

Students in the classroom:

1. If a student is not properly wearing a face covering, the instructor will remind the student of the policy and ask them to comply with it.
2. If the student will not comply with the face covering policy, the instructor will ask the student to leave the classroom, and the student may only return when they are properly wearing a face covering.

3. If the student refuses to properly wear a face covering or leave the classroom, the instructor will dismiss the class and will report the student to Student Conduct & Community Standards for misconduct, where the student will be subject to disciplinary action.

Instructors in the classroom:

1. If an instructor is not properly wearing a face covering, students will remind the instructor of the policy and ask them to comply with it.
2. If an instructor will not properly wear a face covering, students may leave the classroom and should report the misconduct to the department chair or via the TIPS system for disciplinary action through faculty governance processes.

Tentative Schedule:

DATE	TOPIC	DUE BEFORE LAB	DUE AFTER LAB
JAN 26	Orientation: Syllabus, Policies, Resources Introduction to software		
JAN 28	RNA folding and structure	• Pre-lab assignment	• Post-lab assignment
FEB 2	PyMol tutorial and RNA-ligand interactions	• Pre-lab assignment	• Lab notebook • Post-lab assignment
FEB 4	RNA construct and DNA template design	• Pre-lab assignment	• Lab notebook • Post-lab assignment
FEB 9	HBC synthesis part 1: synthesis	• Pre-lab assignment • Module 1 Report	• Lab notebook • Post-lab assignment
FEB 11	HBC synthesis part 2: purification	• Pre-lab assignment	• Lab notebook • Post-lab assignment
FEB 16	HBC characterization: NMR	• Pre-lab assignment	• Lab notebook • Post-lab assignment
FEB 18	HBC characterization: MS	• Pre-lab assignment	• Lab notebook • Post-lab assignment
FEB 23	HBC characterization: HPLC	• Pre-lab assignment	• Lab notebook • Post-lab assignment
FEB 25	Pipetting calibration and molecular biology basic techniques	• Pre-lab assignment • Module 2 Report	• Lab notebook • Post-lab assignment
MAR 2	PCR	• Pre-lab assignment	• Lab notebook • Post-lab assignment
MAR 4	RNA transcription control RNA	• Pre-lab assignment	• Lab notebook • Post-lab assignment
MAR 9	RNA purification control RNA	• Pre-lab assignment	• Lab notebook • Post-lab assignment
MAR 11	RNA transcription test RNA	• Pre-lab assignment	• Lab notebook • Post-lab assignment
MAR 16	RNA purification test RNA	• Pre-lab assignment	• Lab notebook • Post-lab assignment
MAR 18	Quantitative analysis of RNA folding and purity: native and denaturing PAGE	• Pre-lab assignment • Module 3 Report	• Lab notebook • Post-lab assignment
MAR 23	TBD		

MAR 25	RNA-ligand thermal analysis	• Pre-lab assignment	• Lab notebook • Post-lab assignment
MAR 30	RNA-ligand thermal analysis	• Pre-lab assignment	• Lab notebook • Post-lab assignment
APR 1	RNA-ligand thermal analysis	• Pre-lab assignment	• Lab notebook • Post-lab assignment
APR 6	TBD		
APR 8	Introduction to plate reader assay and 96-well screens	• Pre-lab assignment • Module 4 Report	• Lab notebook • Post-lab assignment
APR 13	RNA-ligand binding assays part 1: monovalent and divalent cation dependence	• Pre-lab assignment	• Lab notebook • Post-lab assignment
APR 15	RNA-ligand binding assays part 2: data processing and analysis	• Pre-lab assignment	• Lab notebook • Post-lab assignment
APR 20	RNA-ligand binding assays part 3: ligand variation	• Pre-lab assignment	• Lab notebook • Post-lab assignment
APR 22	RNA-ligand binding assays part 4: buffer dependence	• Pre-lab assignment	• Lab notebook • Post-lab assignment
APR 27	Binding assay wrap-up and analysis	• Pre-lab assignment	• Lab notebook • Module 5 Report
APR 29	In-class discussion / group summary		• Final report due