Principles and Applications of Omics Technologies to Advance Plant Biology HOS6932, 3 credits

Spring 2025

Meeting date and time:

Lectures: Each week two lectures will be made available online within the course Canvas page on Mon and Fri

Synchronous session: Every Thu a 50-minute synchronous Zoom session will be held from 1:00-1:50 pm https://ufl.zoom.us/j/2952105185

Prerequisites: None

Require undergraduate molecular and cellular biology, genetics, or biochemistry knowledge

Instructor:

Swathi Nadakuduti Environmental Horticulture Department 1519 Fifield Hall University of Florida <u>s.nadakuduti@ufl.edu</u> (352)-273-4575

Office hours – Immediately after the class every Thu until 3pm. Other appointments can be made on an individual basis.

Course description:

Recent advancement in technologies have revolutionized plant biology research especially in the fields of genome editing, high throughput sequencing, metabolomics etc., The main goals of this three-credit course are 1) To broadly review molecular technologies applied in plant science research emphasizing on recently evolving CRSIPR based gene editing applications and transformation advancements in plants and 2) Prepare graduate students to critically evaluate scientific literature and introduce the students to proposal writing.

Course expectations:

This course is designed to build on students understanding of basic principles and foundational knowledge on plant molecular biology. Students will critically evaluate the existing knowledge in plant science community as well as identify the gaps in plant research. This course is designed also to promote students' written and oral scientific communication skills. Each week will include two lectures on a given research topic, followed by active discussion of an assigned paper or an assignment on that topic. Students will review assigned literature and come prepared for the class discussion which includes critical analysis and dissection of the paper assigned. The lectures delivered along with assigned literature review would enable students to be current in the topics, critically be able to read and evaluate a scientific publication and be able to apply the knowledge gained to their own research. Students will also have opportunities to give oral presentations on student paper and defend their proposal. Examinations will be based on concepts developed in lectures and literature covered in the class, allowing students to independently think about a given research scenario and how to generate hypothesis and navigate to solve research questions. This course is designed to promote critical reading of manuscripts, think about

research, and practice their oral and written skills. Students considering careers in scientific research in plants in academia or R&D in private industry will benefit from this course.

Course learning objectives:

Students from this course will be able to:

- 1. Survey the plant molecular biology/genetics, functional genomics and metabolomics approaches emphasizing recent advances in plant biology. Evaluate and propose how you might use them for your own research?
- 2. Critically review, analyze, and evaluate scientific literature by justification, critique, and identification of gaps in the existing field of research.
- 3. Generate hypotheses for a given scientific problem and be able to apply and develop effective experimental approaches to test them.
- 4. Utilize publicly available data bases to build evidence and develop direction for your research question.
- 5. Gain experience in creating a research proposal, presenting, and defending it. Students identify a research problem, synthesize, and develop their original idea into an independent proposal and defend it in front of their class.
- 6. Practice and demonstrate effective written and oral scientific communication techniques.

Optional textbooks: Scientific literature WILL be provided by the instructor on every topic (Lecture + papers). For basic molecular biology concepts, students can refer to textbooks (available at UF libraries)

"*Lewin's Genes XI*" (Jocelyn E. Krebs, Elliott S. Goldstein and Stephen T. Kilpatrick, publication date 2012-12-31, Edition 11; eBook ISBN 9781449659066) or

"*Biochemistry & molecular biology of plants*" (edited by Bob B. Buchanan, Wilhelm Gruissem, and Russell L. Jones; publication date 2015-08-31; Edition 2; eBook ISBN 9781118502198)

Reading list: Two to four papers related to each topic being covered will be provided to students well in advance (at least a week prior to the class) out of which one will be chosen for in-depth discussion in the class. The list provided here includes background reading and papers for indepth discussion and might vary and will be updated depending on current literature and relevance to lecture topic.

classic papers:

- Jarvis P, Chen LJ, Li HM, Pete CA, Fankhauser C, Chory J (1998) An Arabidopsis mutant defective in the plastid general protein import apparatus. Science 282: 100–103
- Hamilton AJ, Baulcombe DC (1999) A species of small antisense RNA in posttranscriptional gene silencing in plants. Science **286**: 950–952
- Napoli C, Lemieux C, Jorgensen R (1990) Introduction of a Chimeric Chalcone Synthase Gene into Petunia Results in Reversible Co-Suppression of Homologous Genes in trans. Plant Cell 2: 279–289
- Jinek M, Chylinski K, Fonfara I, Hauer M, Doudna JA, Charpentier E (2012) A programmable dual-RNAguided DNA endonuclease in adaptive bacterial immunity. Science **337**: 816–821

LEC WK	LECTURE TOPIC	HMWK/READINGS	
15-Jan	Course organization and expectations	Zoom - introductions	
17-Jan	Basics of a Plant Cell	-	
22-Jan	Assignment # 1 - Lightening talks	Zoom - Lightening talks	
24-Jan	Structure of DNA and analysis	Eukaryotic gene structure	
27-Jan	Model organisms, public data bases - NCBI, TAIR	MSAs and phylogenetic trees	
29-Jan	Assignment # 2 + Class Discussion # 1	Zoom: Jarvis et al., (1998) Science 282: 100–103	
31-Jan	Forward genetics approaches for plant functional genomics		
3-Feb	Mutants in model plants, Identifying mutants, Gene discovery		
5-Feb	Class Discussion # 2	Zoom: Paper TBA	
7-Feb	Reverse genetics approaches for plant functional genomics		
10-Feb	RNA-mediated interference (RNAi), Virus Induced Gene Silencing		
12-Feb	Class Discussion # 3	Zoom: Paper TBA	
14-Feb	Gene-editing by CRISPR systems and evaluating mutagenesis		
17-Feb	Introduction to CRISPR/Ca9: Discovery and how CRISPR works? Variants of Cas9 and multiplexing		
19-Feb	Class Discussion # 4	Zoom: Paper TBA	
21-Feb	Guide RNA design and CRISPR constructs: plant systems		
24-Feb	Case studies of CRISPR success in Agricultural crop improvement		
26-Feb	Class Discussion # 5 + Assignment # 3	Zoom - Assignment discussion	
28-Feb	Base Editing - Cytosine, Adenine base editors, glycosylase editors		
3-Mar	Prime Editing - Case studies in plants		
5-Mar	MID-TERM EXAM	On Canvas	
7-Mar	Advances in plant genetic transformation technologies		
10-Mar	Agrobacterium mediated transformation - Case studies		
8-Mar	Class Discussion # 6	Zoom: Paper TBA	
12-Mar	Protoplasts: tool for plant research, transfection, and regeneration		
Mar 15 - 23	SPRING BREAK		
24-Mar	Delivery of DNA manipulation cargo bypassing tissue culture		
26-Mar	Class Discussion # 7	Zoom: Paper TBA	
28-Mar	DNA sequencing - Basics and advances		
31-Mar	Basics of Sanger sequencing, DNA sequencing by various platforms		
2-Apr	Assignment # 4	Zoom - Student oral presentations on assigned paper	
4-Apr	Transcriptomics and quantitative RT-PCR		

7-Apr	Transcription factors and regulatory proteins		
9-Apr	Class Discussion # 8	Zoom: Paper TBA	
11-Apr	Genomics enabled plant biochemistry research	protein- DNA and protein - protein interaction, Case studies	
14-Apr	Case studies: Metabolomics to study plant specialized metabolism	netabolism Metabolomics approaches	
16-Apr	Class Discussion # 9	Zoom: Paper TBA	
18-Apr	Single cell genomics		
21-Apr	Advances in imaging technologies for plant research		
23-Apr	Class Discussion # 10	Zoom: Paper TBA	
30-Apr	Student paper presentations		
1-May	FINAL EXAM	On Canvas	

Other papers to read:

Degao Liu, Evan E. Ellison, Erik A. Myers and Daniel F. Voytas etal., (2024) Heritable gene editing in tomato through viral delivery of isopentenyl transferase and single-guide RNAs to latent axillary meristematic cells. PNAS 121 (39) e2406486121

- Xiaoen Huang, Hongge Jia, Jin Xu, Yuanchun Wang, Jiawen Wen and Nian Wang (2023) Transgene-free genome editing of vegetatively propagated and perennial plant species in the T0 generation via a co-editing strategy. Nature Plants **9**: 1591–1597
- Atkins PAP, Voytas DF (2020) Overcoming bottlenecks in plant gene editing. Curr Opin Plant Biol 54: 79–84
- Dong OX, Yu S, Jain R, Zhang N, Duong PQ, Butler C, Li Y, Lipzen A, Martin JA, Barry KW, et al (2020) Marker-free carotenoid-enriched rice generated through targeted gene insertion using CRISPR-Cas9. Nat Commun **11**: 1178
- **Debernardi JM, Tricoli DM, Ercoli MF, Hayta S, Ronald P, Palatnik JF, Dubcovsky J** (2020) A GRF–GIF chimeric protein improves the regeneration efficiency of transgenic plants. Nat Biotechnol. doi: 10.1038/s41587-020-0703-0
- Gaudelli NM, Komor AC, Rees HA, Packer MS, Badran AH, Bryson DI, Liu DR (2017) Programmable base editing of A*T to G*C in genomic DNA without DNA cleavage. Nature **551**: 464–471
- Jin S, Zong Y, Gao Q, Zhu Z, Wang Y, Qin P, Liang C, Wang D, Qiu J-L, Zhang F, et al (2019) Cytosine, but not adenine, base editors induce genome-wide off-target mutations in rice. Science. doi: 10.1126/science.aaw7166
- Komor AC, Kim YB, Packer MS, Zuris JA, Liu DR (2016) Programmable editing of a target base in genomic DNA without double-stranded DNA cleavage. Nature **533**: 420–424
- Anzalone A V, Randolph PB, Davis JR, Sousa AA, Koblan LW, Levy JM, Chen PJ, Wilson C, Newby GA, Raguram A, et al (2019) Search-and-replace genome editing without double-strand breaks or donor DNA. Nature **576**: 149–157
- Lin Q, Zong Y, Xue C, Wang S, Jin S, Zhu Z, Wang Y, Anzalone A V., Raguram A, Doman JL, et al (2020) Prime genome editing in rice and wheat. Nat Biotechnol **38**: 582–585
- Maher MF, Nasti RA, Vollbrecht M, Starker CG, Clark MD, Voytas DF (2018) Plant gene editing through de novo induction of meristems. Nat Biotechnol. doi: 10.1038/s41587-019-0337-2
- Wang X, Ye L, Lyu M, Ursache R, Löytynoja A, Mähönen AP (2020) An inducible genome editing system for plants. Nat Plants 6: 766–772
- Zhao D, Li J, Li S, Xin X, Hu M, Price MA, Rosser SJ, Bi C, Zhang X (2020) Glycosylase base editors enable

C-to-A and C-to-G base changes. Nat Biotechnol. doi: 10.1038/s41587-020-0592-2

- Zong Y, Wang Y, Li C, Zhang R, Chen K, Ran Y, Qiu JL, Wang D, Gao C (2017) Precise base editing in rice, wheat and maize with a Cas9-cytidine deaminase fusion. Nat Biotechnol **35**: 438–440
- Misra BB, Assmann SM, Chen S (2014) Plant single-cell and single-cell-type metabolomics. Trends Plant Sci 19: 637–646
- Rich-Griffin C, Stechemesser A, Finch J, Lucas E, Ott S, Schäfer P (2020) Single-Cell Transcriptomics: A High-Resolution Avenue for Plant Functional Genomics. Trends Plant Sci 25: 186–197
- Wurtzel ET, Kutchan TM (2016) Plant metabolism, the diverse chemistry set of the future. Science 353: 1232–1236

Evaluation and Grade determination:

Class attendance: All students are required to attend the class and participate actively in class discussion for full participation credit.

Student Evaluation Basis	Number	Points/ event	Total
CLASS DISCUSSIONS ¹	10 Class Discussions	10	100
ASSIGNMENTS ²	4	25	100
MID-TERM EXAM ³	1	100	100
FINAL EXAM ³	1	100	100
PROPOSAL + ORAL PRESENTATION ⁴	1	100	100
TOTAL			500

¹<u>Class Discussions:</u> Will be based on journal articles provided to students on the topic being covered and will focus on: Problems addressed in the paper, research objectives, experimental design, critical data presentation (tables/figures), gaps or weaknesses, future work? Each student may be questioned on the literature reviewed for engagement. For full or partial participation credit, students are expected to come prepared to answer questions on the assigned paper and participate in the discussion.

**Assignments will be discussed in the class in their respective weeks.

 2 <u>Assignments</u>: Four assignments in total; Students will be given one-week time to turn in. Assignment # 1) A 5 min lightening talk on your graduate research, which must include a personal elevator pitch for 30sec, why your research is important (for broader audience), research objectives and your contribution

Assignment # 2) DNA analysis: Students will be given a gene identifier from each model organism, students will find: Gene structure, function, expression, CDS, cDNA, BLAST query, primary literature

Assignment # 3) CRISPR single guide RNA design: Students will use online programs to select spacer sequence for sgRNA design for a target gene and design strategies to evaluate mutagenesis

Assignment # 4) Student oral presentation: Students will give 10-minute oral presentation on an assigned paper of relevance to the topics being covered in the course

**Students may also be directed to participate in research seminars at UF and answering questions related to that seminar for extra credit.

³<u>Examination</u> questions will be drawn from concepts developed in lectures and discussions. All assigned reading (background & in class discussion), Material covered in lecture & and Material knowledge and application (problem solving). There will be review sessions before the exams.

⁴ <u>Proposal writing</u> Student assignment will be to write an original research proposal on a topic given/ student may select a relevant topic focusing on some aspect of plant molecular biology (can't be your graduate research project). Students will be provided guidelines for writing the proposal. In general, the proposal must include: Summary (1/2 page), Introduction/background and Rationale (2 pages), specific objectives, experimental design, and methodology (up to 4 pages) and references. Student's CV must also be provided for review. Proposal will be reviewed, and recommendations provided. Students gets to orally defend their proposal in the class in the final week. The grade for proposal will be based on both written proposal submission and oral presentation.

#points	% points	Passing Grade	Grade points
450 - 500	90 - 100%	А	4.0
425 - 449	85 - 89.9%	A-	3.67
400 - 424	80 - 84.9%	B+	3.33
375 - 399	75 - 79.9%	В	3.0
350 - 374	70 - 74.9%	В-	2.67
325 - 349	65 - 69.9%	C+	2.33
300 - 324	60 - 64.9%	С	2.0
275 - 299	55 - 59.9%	C-	1.67
250 - 274	50 - 54.9%	D+	1.33
225 - 249	45 - 49.9%	D	1.0
200 - 224	40 - 44.9%	D-	0.67
0-199	0-39.9%	E	0.00

For information on current UF policies for assigning grade points, see: https://catalog.ufl.edu/ugrad/current/regulations/info/grades.aspx (Undergrads) http://gradcatalog.ufl.edu/content.php?catoid=2&navoid=762#grades (Graduate students)

Class Policies that students MUST follow:

Attendance: Your attendance at all classes is a strong expectation, but if you are ill or an emergency occurs, contact your instructor PRIOR TO the scheduled class time. Participation credit can be earned in every class discussion. Student shall lose points for Class Discussion if absent for the class. Requirements for class attendance and make-up exams, assignments and other work are consistent with universitv policies that can be found at: https://catalog.ufl.edu/UGRD/academic-regulations/attendance-policies/.

Preparation for class: Please come to class prepared by reading the assigned literature. This can help you not only for this class but for your future semesters and your research as well.

Late Assignments: Assignments will NOT be accepted after the due date. If communicated ahead of time, late assignments may be considered with a late penalty of 25%.

Makeup Exams: Special permission is required ahead of time to accommodate makeup exam. This will be made possible only under reasonable circumstances but NOT for all requests.

Expectations for classroom behavior: Please do not talk privately during the lectures or multitask (i.e. doing work for other courses or research, using your computer, cell phone or other electronic devices for web surfing or email). Ask questions and be involved in the class.

COVID Response Statements:

Our class sessions may be audio visually recorded for students in the class to refer back and for enrolled students who are unable to attend live. Students who participate with their camera engaged or utilize a profile image are agreeing to have their video or image recorded. If you are unwilling to consent to have your profile or video image recorded, be sure to keep your camera off and do not use a profile image. Likewise, students who unmute during class and participate orally are agreeing to have their voices recorded. If you are not willing to consent to have your voice recorded during class, you will need to keep your mute button activated and communicate exclusively using the "chat" feature, which allows students to type questions and comments live. The chat will not be recorded or shared. As in all courses, unauthorized recording and unauthorized sharing of recorded materials is prohibited.

Online Course Evaluation Process:

Student assessment of instruction is an important part of efforts to improve teaching and learning. At the end of the semester, students are expected to provide feedback on the quality of instruction in this course using a standard set of university and college criteria. Students are expected to provide professional and respectful feedback on the quality of instruction in this course by completing course evaluations online via GatorEvals. Guidance on how to give feedback in a professional and respectful manner is available at: https://gatorevals.aa.ufl.edu/students/. Students will be notified when the evaluation period opens and can complete evaluations through the email, they receive from GatorEvals, in their Canvas course menu under GatorEvals, or via https://gatorevals.aa.ufl.edu/students/. Students will be given specific time in the class to fulfill this requirement.

Academic Honesty:

As a student at the University of Florida, you have committed yourself to uphold the Honor Code, which includes the following pledge: "We, the members of the University of Florida community, pledge to hold ourselves and our peers to the highest standards of honesty and integrity." You are expected to exhibit behavior consistent with this commitment to the UF academic community, and on all work submitted for credit at the University of Florida, the following pledge is either required or implied: "On my honor, I have neither given nor received unauthorized aid in doing this assignment." It is assumed that you will complete all work independently in each course unless the instructor provides explicit permission for you to collaborate on course tasks (e.g. assignments, papers, quizzes, exams). Furthermore, as part of your obligation to uphold the Honor Code, you should report any condition that facilitates academic misconduct to appropriate personnel. It is your individual responsibility to know and comply with all university policies and procedures regarding academic integrity and the Student Honor Code. Violations of

the Honor Code at the University of Florida will not be tolerated. Violations will be reported to the Dean of Students Office for consideration of disciplinary action. For more information regarding the Student Honor Code, please see:

http://www.dso.ufl.edu/sccr/process/student-conduct-honor-code.

** Please DO NOT borrow the writing of others – this is called Plagiarism. If you do not understand what this means, ask your instructor to clarify. If you are presenting others' thoughts or writing including diagrams/pictures you must cite them. Instructors will do the same!

Software Use:

All faculty, staff and students of the university are required and expected to obey the laws and legal agreements governing software use. Failure to do so can lead to monetary damages and/or criminal penalties for the individual violator. Because such violations are also against university policies and rules, disciplinary action will be taken as appropriate.

Services for Students with Disabilities:

The Disability Resource Center coordinates the needed accommodations of students with disabilities. This includes registering disabilities, recommending academic accommodations within the classroom, accessing special adaptive computer equipment, providing interpretation services and mediating faculty-student disability related issues. Students requesting classroom accommodation must first register with the Dean of Students Office. The Dean of Students Office will provide documentation to the student who must then provide this documentation to the Instructor when requesting accommodation.

0001 Reid Hall, 352-392-8565, https://disability.ufl.edu/

Campus Helping Resources:

Students experiencing crises or personal problems that interfere with their general wellbeing are encouraged to utilize the university's counseling resources. The Counseling & Wellness Center provides confidential counseling services at no cost for currently enrolled students. Resources are available on campus for students having personal problems or lacking clear career or academic goals, which interfere with their academic performance.

Health and Wellness

U Matter, We Care:

If you or a friend is in distress, please contact <u>umatter@ufl.edu</u> or 352-392-1575 so that a team member can reach out to the student. Dean of Students Office (CARE team) <u>https://care.dso.ufl.edu/</u>

Counseling and Wellness Center: http://www.counseling.ufl.edu/cwc 352-392-1575

Sexual Assault Recovery Services (SARS) Student Health Care Center, 352-392-1161

University Police Department at 352-392-1111 (or 9-1-1 for emergencies), or http://www.police.ufl.edu/

Academic Resources

E-learning technical support, 352-392-4357 (select option 2) or e-mail to Learningsupport@ufl.edu. https://lss.at.ufl.edu/help.shtml

Career Connections Center, First Floor JWRU, 392-1601, https://career.ufl.edu/

Library Support, http://cms.uflib.ufl.edu/ask. Various ways to receive assistance with respect to using the libraries or finding resources.

Teaching Center, Broward Hall, 392-2010 or 392-6420. General study skills and tutoring. https://teachingcenter.ufl.edu/

Writing Studio, **302 Tigert Hall**, 846-1138. Help brainstorming, formatting, and writing papers. https://writing.ufl.edu/writing-studio/

Student Complaints

Residential Course: https://sccr.dso.ufl.edu/policies/student-honor-code-studentconduct-code/ Online Course: http://www.distance.ufl.edu/student-complaint-process

University of Florida Complaints Policy

The University of Florida believes strongly in the ability of students to express concerns regarding their experiences at the University. The University encourages its students who wish to file a written complaint to submit that complaint directly to the department that manages that policy.

A student who is unsure as to the official responsible for handling his or her particular complaint may contact the Ombuds office or the Dean of Students Office. For complaints that are not satisfactorily resolved at the department level or which seem to be broader than one department, students are encouraged to submit those complaints to one of the following locations: Ombuds: http://www.ombuds.ufl.edu/ 31 Tigert Hall, 352-392-1308

The purpose of the Ombuds office is to assist students in resolving problems and conflicts that arise in the course of interacting with the University of Florida. By considering problems in an unbiased way, the Ombuds works to achieve a fair resolution and works to protect the rights of all parties involved. Dean of Students Office: http://www.dso.ufl.edu/ 202 Peabody Hall, 352-392-1261

The Dean of Students Office works with students, faculty, and families to address a broad range of complaints either through directly assisting the student involved to resolve the issue, working with the student to contact the appropriate personnel, or referring the student to resources or offices that can directly address the issue. Follow up is provided to the student until the situation is resolved. Additionally, the University of Florida regulations provide a procedure for filing a formal grievance in Regulation 4.012: http://regulations.ufl.edu/regulations/uf-4-student-affairs