

Course Syllabus
MBMB 631 CRISPR/Cas9 Genome Editing
Academic Year 2025

| | |
|-----------------------------|---|
| Course ID and Title: | MBMB 631 CRISPR/Cas9 Genome Editing |
| Course Coordinator: | Asst. Prof Alisa Tubsuwan, Ph.D. Institute of Molecular Biosciences, Mahidol University Tel: 0-2441-9003 Ext. 1366 Email: alisa.tub@mahidol.ac.th |
| Instructors: | Asst. Prof Alisa Tubsuwan, Ph.D. |
| Support Staff: | Miss Pirut Tong-Ngam |
| Credits: | 1(0-2-1) |
| Curriculum: | Master of Science Program in Molecular and Integrative Biosciences (Elective course) Doctor of Philosophy Program in Molecular and Integrative Biosciences (Elective course) |
| Semester offering: | First and second semesters |
| Pre-Requisites: | None |

Course Learning Outcome (CLOs):

By the end of the course, students should be able to:

1. Describe the molecular mechanisms underlying CRISPR/Cas9 genome editing
2. Demonstrate proficiency in the CRISPR/Cas9 genome editing tool to design and implement gene modifications for addressing scientific research questions
3. Demonstrate scientific integrity, responsibility, and safety practices
4. Communicate scientific concepts effectively through discussions and presentations

Alignment of Teaching and Assessment Methods to Course Learning Outcomes:

| Course Learning Outcomes | Teaching Method | Assessment Method |
|---|--|--|
| 1. Describe the molecular mechanisms underlying CRISPR/Cas9 genome editing and workflows (Knowledge - aligned with PLO1) | 1. Lecture: 2. Case studies | 1. Quiz |
| 2. Design an experiment using CRISPR/Cas9 genome editing tool to address a scientific research question. (Skills - aligned with PLO2) | 1. Lecture 2. Interactive discussion 3. Group activity 4. Hands-on lab practice | 1. Lab performance 2. Data analysis 3. Written report 4. Presentation 5. Class discussion |
| Demonstrate scientific integrity, responsibility, and safety practices (Ethics - aligned with PLO3) | 1. Laboratory and practical work 2. Writing lab report | 1. Regular attendance tracking 2. Direct observation and evaluation of students during lab sessions. 3. Submission lab report and tasks as per specified deadlines. 4. Evaluation of lab reports for plagiarism and quality of content. |
| Communicate scientific concepts effectively through discussions and presentations (Characters – Aligned with PLO4). | 1. Presentation 2. Group discussion and peer feedback sessions | 1. Presentation 2. Lab performance 3. Written report |

Course Description:

CRISPR/Cas9 Genome Editing; Guide RNA Design; Construction of Plasmid Expressing Guide RNA and Cas9; Delivery of CRISPR/Cas9 Components into Human Cells; Analysis of Gene Editing Outcomes by T7 Endonuclease I Assay; Sanger Sequencing and Computational Analysis

เทคโนโลยีตัดต่อยีนคริสเปอร์/คาส 9 การออกแบบไกด์อาร์เอ็นเอ การสร้างพลาสมิดสำหรับการแสดงออกของไกด์อาร์เอ็นเอ และโปรตีนคาส 9 การนำส่งส่วนประกอบของคริสเปอร์/คาส 9 เข้าสู่เซลล์ การตรวจสอบการแก้ไขจีโนมด้วยการทดสอบด้วยเอ็นไซม์ ที7 เอ็นโดนิวคลีเอส I และเทคนิคการวิเคราะห์หาลำดับนิวคลีโอไทด์ตามด้วยเครื่องมือทางคอมพิวเตอร์

Course Schedule:

Location: Classroom (Room C405) and Laboratory Classroom (Room D408), Institute of Molecular Biosciences.

| Unit | Time | Topic | Instructors and Assistants |
|------------------------|-------------|--|----------------------------|
| Day1 November 17, 2025 | | | |
| | 09.00-11.00 | Lecture: Overview and workflow for CRISPR/Cas9 genome engineering | AT |
| | 11.00-12.00 | Hand-on: Guide RNA design | AT |
| | 13.00-14.00 | Hand-on: Donor template design | AT |
| | 14.00-16.00 | Hand-on Genome editing validation method TIDE - rapid, powerful and easy analysis of CRISPR experiments | |
| Day2 November 18, 2025 | | | |
| | 09.00-12.00 | Hand on: Preparation of sgRNA expression construct I Activity: Preparation of the sgRNA oligos inserts) | AT |
| | 13.00-15.00 | Hand on: Preparation of sgRNA expression construct II Activity: cloning the sgRNA into spCas9 vector | AT |

| Unit | Time | Topic | Instructors and Assistants |
|------------------------|-------------|---|----------------------------|
| | 15.00-17.00 | Hand on: Preparation of sgRNA expression construct III Activity: Transform E. coli with the sgRNA expression plasmid, plate onto selective agar, and incubate for colony growth. | |
| Day3 November 19, 2025 | | | |
| | 09.00-12.00 | Hand on: Cell seeding Activity: Prepare and seed mammalian cells for genome editing | AT |
| | 13.00-16.00 | Hand on: Preparation of sgRNA expression construct IV Activity: Bacterial colony picking and culture for plasmid amplification | AT |
| Day4 November 20, 2025 | | | |
| | 09.00-12.00 | Hand on: Construction of plasmid expressing guide RNA and Cas9 IV Activity: sgRNA Plasmid isolation from cultured <i>E. coli</i> | AT |
| | 13.00-16.00 | Transfection Activity: Introduce CRISPR/Cas9 plasmid into mammalian cells | AT |
| Day5 November 21, 2025 | | | |
| | 09.00-12.00 | Hand on: Knockout validation by T7 endonuclease I assay I Activity: Cell harvesting, Genomic DNA extraction and PCR amplification of target regions. | AT |
| | 13.00-16.00 | Hand on: Knockout validation by T7 endonuclease assay II Activity: Digestion of PCR products using T7 Endonuclease I, followed by electrophoresis to analyze cleavage | AT |

| Unit | Time | Topic | Instructors and Assistants |
|-------|-------------|---------------------------------|----------------------------|
| Day 6 | | | |
| | 16.00-17.00 | Lab discussion and presentation | AT |

Assessment Criteria

| Assessment criteria | Rubric | Scoring rubric |
|------------------------------|----------------------------|--|
| Class attendance (5%) | Attendance | 4: points full attendance or received approval for all necessary absences 3: points-1 unexcused absence 2: points-2 unexcused absences 1: points-more than 2 unexcused absences |
| | Punctuality | 4: Punctual 3: Less than 5 min late 2: Less than 15 min late 1: more than 15 min late |
| Quiz (10%) | Correctness and completion | Raw scores will be adjusted to be in a range of 0-10% |
| Laboratory performance (35%) | Safety practice | 4: Strict adherence to safety protocols, exemplary safety practices 3: Few safety violations, generally adheres to safety protocols |

| Assessment criteria | Rubric | Scoring rubric |
|---------------------|---------------------------------|---|
| | | <p>2: Some safety violations, limited adherence to safety protocols.</p> <p>1: Frequent safety violations, disregard for safety protocols.</p> |
| | Experimental protocol adherence | <p>4: Strict adherence to experimental protocols.</p> <p>3: Generally, follows experimental protocols, moderate adherence.</p> <p>2: Occasionally deviates from protocols, limited adherence</p> <p>1: Frequently deviates from experimental protocols, lacks adherence.</p> |
| | Experimental technic adherence | <p>4: Executes laboratory techniques and procedures with precision and skill.</p> <p>3: Demonstrates good proficiency in laboratory techniques and procedures</p> <p>2: Shows basic proficiency but may lack precision.</p> <p>1: Struggles with basic techniques, leading to inconsistent results.</p> |
| | Laboratory equipment handling | <p>4: Handles laboratory equipment and instruments with expertise and care,</p> |

| Assessment criteria | Rubric | Scoring rubric |
|---------------------|-----------------------------|---|
| | | <p>preventing damage or accidents.</p> <p>3: Handles equipment competently but may occasionally mishandle or damage equipment.</p> <p>2: Shows a lack of proficiency in equipment handling, leading to frequent issues</p> <p>1: Frequently mishandles equipment, causing damage or delays.</p> |
| | Team work and collaboration | <p>4: Exceptional collaboration, seamless teamwork, excellent communication</p> <p>3: Effective collaboration, good teamwork, adequate communication</p> <p>2: Limited effectiveness in collaboration, some teamwork issues, minimal communication,</p> <p>1: Ineffective collaboration, poor teamwork, lack of communication</p> |
| | Time management | <p>4: Follows the experiment schedule closely, completing tasks within established timeframes.</p> |

| Assessment criteria | Rubric | | Scoring rubric |
|-------------------------|------------|--------------|---|
| | | | <p>3: Mostly adheres to the schedule but may occasionally fall slightly behind or ahead of the timeline.</p> <p>2: Often deviates from the schedule, leading to notable delays or rushed work.</p> <p>1: Consistently disregards the schedule, causing substantial delays or incomplete work.</p> |
| Laboratory report (25%) | Plagiarism | | <p>4: No evidence of plagiarism; all sources properly cited.</p> <p>3: Proper citation of sources, minimal plagiarism detected.</p> <p>2: Some minor issues with plagiarism or citation.</p> <p>1: Evidence of significant plagiarism or improper citation.</p> |
| | Contents | Introduction | <p>4: Excellent introduction that effectively sets up the study with clear objectives and hypotheses.</p> <p>3: Clear introduction with well-defined objectives and hypotheses.</p> <p>2: Basic introduction but lacks detail or clarity in objectives and hypotheses.</p> |

| Assessment criteria | Rubric | | Scoring rubric |
|---------------------|--------|----------------------|---|
| | | | 1: Inadequate introduction with unclear objectives and hypotheses. |
| | | Material and methods | 4: Methods section is detailed, concise, and replicable. 3: Methods section is present but may lack some details. 2: Methods section lacks detail. 1: Methods section is incomplete or confusing. |
| | | Results | 4: Results are accurately presented, with appropriate tables and figures. 3: Results are accurately presented. 2: Results are presented with limited clarity. 1: Results are poorly presented. |
| | | Discussion | 4: Discussion addresses significance and implications effectively 3: Discussion addresses some aspects of significance and implication 2: Discussion lacks depth and significance. 1: Discussion is minimal or absent. |

| Assessment criteria | Rubric | | Scoring rubric |
|---------------------|---------------------|------------|--|
| Presentation (15%) | | Conclusion | <p>4: Implications and relevance to the research question are discussed.</p> <p>3: Conclusions are drawn but may lack depth.</p> <p>2: Discussion lacks depth and significance.</p> <p>1: Conclusions are missing or entirely unsupported.</p> |
| | Writing Quality | | <p>4: Good writing quality with minor grammatical or spelling errors</p> <p>3: Writing quality is fair with noticeable grammatical or spelling errors.</p> <p>2: Writing quality is poor with frequent grammatical or spelling errors.</p> <p>1: Writing quality is extremely poor with numerous grammatical or spelling errors.</p> |
| | On-Time Submission: | | <p>4: Submitted on time or well before the deadline.</p> <p>3: Submitted close to the deadline but within an acceptable timeframe</p> <p>2: Submitted late but within a reasonable timeframe.</p> <p>1: Submitted significantly late or not submitted at all.</p> |

| Assessment criteria | Rubric | Scoring rubric |
|---------------------|-------------------------------|---|
| Presentation (15%) | Organization | <p>4: The presentation is exceptionally well-organized with a clear and logical structure</p> <p>3: The presentation is well-organized with a clear structure.</p> <p>2: The presentation is somewhat organized but may lack clarity or logical flow.</p> <p>1: The presentation lacks clear organization, making it difficult to follow.</p> |
| | Content | <p>4: The content is exceptionally clear, relevant, comprehensive, and effectively conveys key points.</p> <p>3: The content is clear, relevant, and covers necessary information.</p> <p>2: The content is somewhat clear and relevant but may lack depth.</p> <p>1: The content is unclear, irrelevant, or incomplete.</p> |
| | Knowledge/answering questions | <p>4: The presenter exhibits a deep understanding of the subject matter and answers questions with expertise.</p> |

| Assessment criteria | Rubric | Scoring rubric |
|---------------------|--------------------|---|
| | | <p>3: The presentation style is engaging and confident, maintaining the audience's attention.</p> <p>2: The presenter shows some understanding of the subject matter but may struggle to answer questions comprehensively.</p> <p>1: The presenter demonstrates a lack of understanding of the subject matter and is unable to answer questions effectively.</p> |
| | Presentation style | <p>4: The presentation style is highly engaging, confident, and dynamic, captivating the audience.</p> <p>3: The presentation style is engaging and confident, maintaining the audience's attention.</p> <p>2: The presentation style is passable but lacks strong engagement or confidence.</p> <p>1: The presentation style is ineffective, lacking engagement, and confidence.</p> |

Student's achievement will be graded using symbols: A, B+, B, C+, C, D+, D and F, based on the criteria as follows:

| Percentage range | Grade | Description |
|------------------|-------|-------------|
| 80-100 | A | Excellent |
| 75-79 | B+ | Very Good |
| 70-74 | B | Good |
| 65-69 | C+ | Fairly Good |
| 60-64 | C | Fair |
| 55-59 | D+ | Poor |
| 50-54 | D | Very Poor |
| 0-49 | F | Fail |