The CRISPR in the Classroom Network: A Support System for Instructors to Bring Gene Editing Technology to the Undergraduate Classroom

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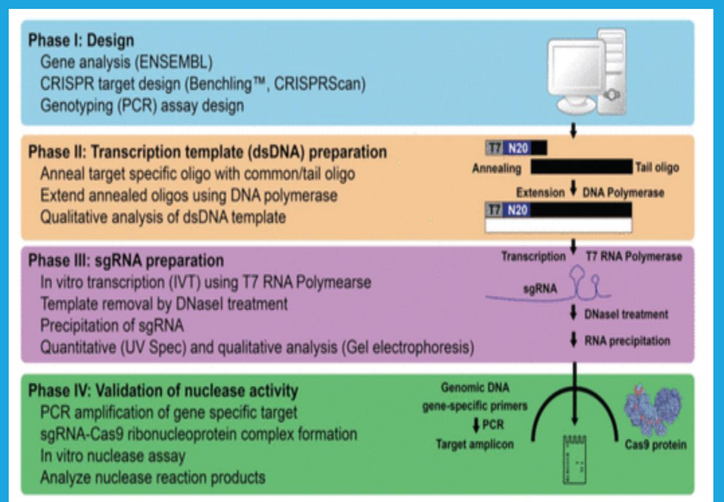
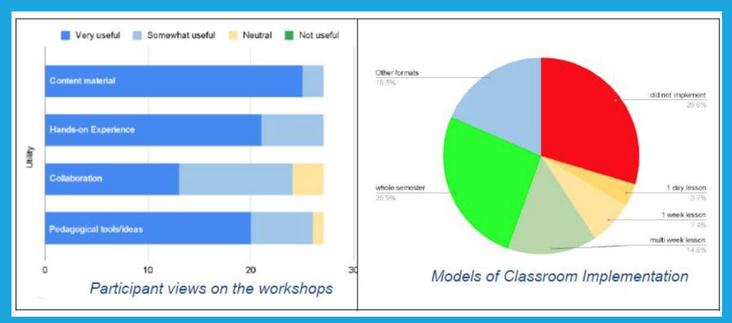
CRISPR-Cas9 technology represents a once-in-a-generation advance in molecular biology that allows precise gene editing and has become a mainstream technique in research. However, as is often the case with new technology, most undergraduate laboratory instructors do not have the training or support to integrate CRISPR-Cas9 into their courses. To remedy this, we have formed the "CRISPR in the Classroom" Network and are facilitating a series of workshops and mentoring activities designed to provide instructors, postdocs, and graduate students the skills, support, and confidence needed to introduce and implement CRISPR-Cas9 technology in undergraduate laboratory classrooms (NSF RCN-UBE #2120417). Our summer workshops provide participants with flexible, easily-adapted curriculum and start-up kits to overcome the hurdles associated with implementing a new technology. Assessment data from a previous online workshop and two NSF-sponsored in-person workshops (Awards #1823595 and 1916486) show most workshop participants develop the skills and confidence necessary to implement CRISPR-Cas9 modules into their laboratory courses within one year of the workshop. The CRISPR in the Classroom Network represents a dynamic community of practice dedicated to providing undergraduate life science instructors with the tools and support needed to integrate CRISPR-Cas9 technology in their courses and across model systems.

CRISPR-Cas9 as a tool to teach molecular biology



The CRISPR in the Classroom workshop is designed to take instructors through the bioinformatics and molecular biology necessary to generate a specific guide RNA that can be used to target a gene of interest via in vitro nuclease activity

Workshops provide the support and confidence to build CRISPR into the undergraduate curriculum



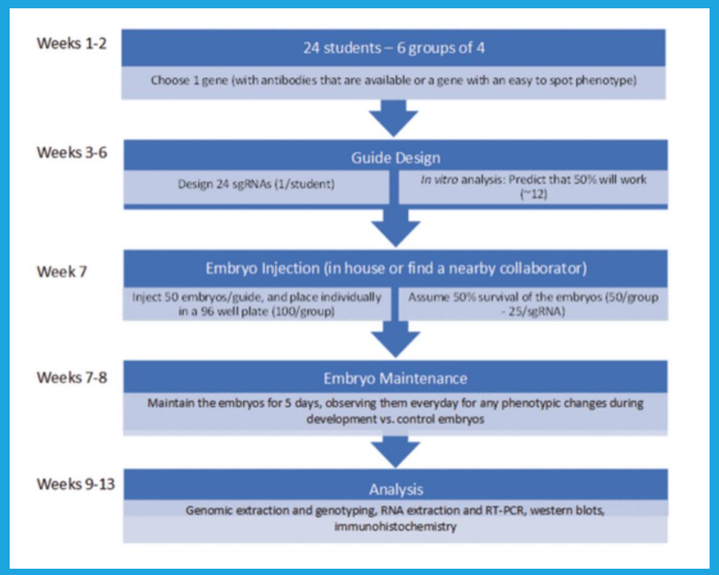
Participant satisfaction and implementation plans in central offerings from the 2019 virtual CRISPR in the Classroom workshop

The modular design of the CRISPR in the Classroom workflow allows for adaptability to any class setting

Module	Outcome/output	Suggested time
Bioinformatics	• Analyze gene sequences • Identify regions that can be effectively targeted based on available high-scoring guides and biological relevance • Primer design for PCR genotyping (on-target and off-target)	3–4 hours
sgRNA synthesis	• ssDNA → dsDNA template → ssRNA lab workflow • Compare how ssDNA, dsDNA, and ssRNA behave during electrophoresis • Basic qualitative (gel) and quantitative (spec) analysis of sgRNA	6–8 hours
DNA target synthesis	• PCR optimization (gradient PCR)	2–3 hours
In vitro nuclease activity	• *In vitro* validation of sgRNA • Functional QC of sgRNA • Off-target efficiency	2–3 hours
Embryo injections	• Observe or perform microinjection into 1–2 cell stage embryos	4–5 hours
Phenotypic analysis (of injected embryos)	• Scoring embryos injected with sgRNA-Cas9 RNP complex • Identification of potential phenotypes	2–3 hours
Basic genotyping (of injected embryos)	• Analyze presence of potential indel mutations (on target and off-target)	2–3 hours
Advanced genotyping (to detect mutations present in injected embryos)	• Cloning of PCR products • Sequencing cloned PCR products • Analyze mutations	6–8 hours

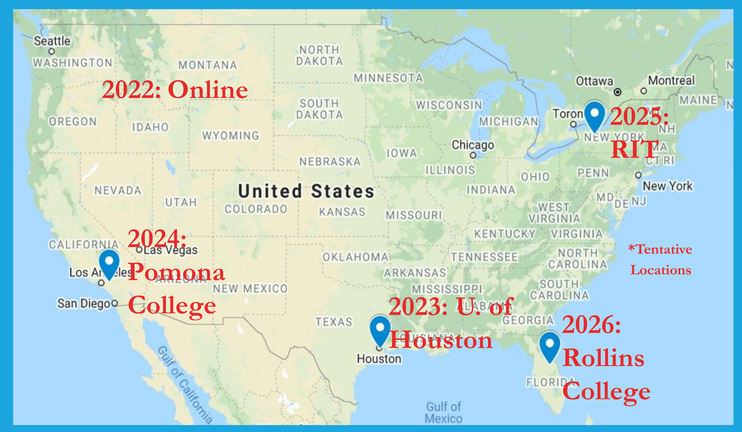
CRISPR = clustered regularly interspaced short palindromic repeats; ssDNA = single-stranded DNA; dsDNA = double-stranded DNA; sgRNA = single guide RNA; QC = quality control.

An instructor may choose to use all or selected portions of the workflow's components based on the needs/level of their class



A sample semester schedule for using the CRISPR in the Classroom workflow.
While the workflow was designed for work with zebrafish DNA, it is easily adapted to any other model system

Locations of the Summer Workshops



We are seeking current and future undergraduate instructors from all types of institutions to join our network. Instructors from minority-serving institutions are especially encouraged to apply.

Workshop participants are provided a free **CRISPRIN Classrooms™ kit** to help with immediate implementation of CRISPR technology into their curriculum.

For information about kits, please contact Dr. Tiffany Hoage.

Contact and Grant Information

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1-Univerity of Houston, Houston, TX, 2-Hampden-Sydney College, Hampden-Sydney, VA, 3-CRISPRIN Classrooms™, 4-University of North Carolina at Pembroke, Pembroke, NC, 5-Rochester Institute of Technology, Rochester, NY, 6-Georgia Gwinnett College, Lawrenceville, GA, 7-Pomona College, Claremont, CA, 8-Rollins College, Winter Park, FL, 9-Randolph-Macon College, Ashland, VA, 10-University of Alabama at Birmingham, Birmingham, AL