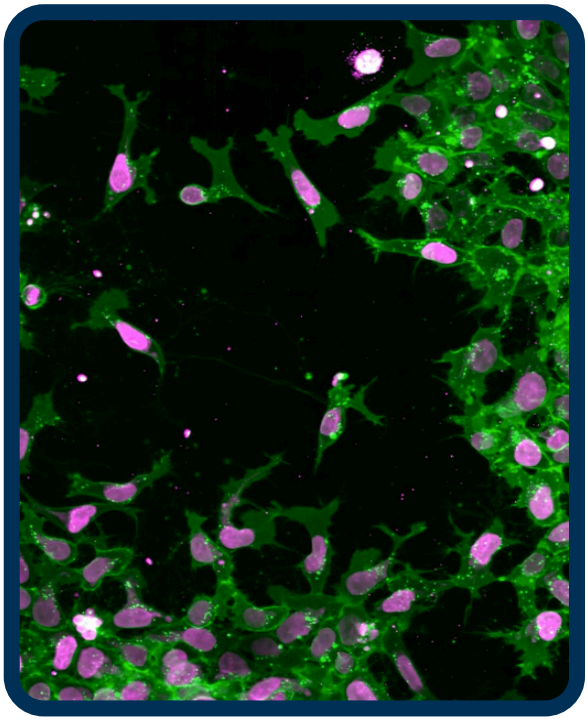
Chop & Glow

In this lab, students learn how and why scientists at the Allen Institute use CRISPR/Cas9 to generate populations of cells with fluorescent organelles and cell structures. Students walk through a hands-on lab protocol where they use the CRISPR/Cas9 machinery to cut or "chop" DNA samples. Students analyze their samples by using gel electrophoresis and observe how real populations of cells from the Allen Institute "glow" under fluorescent microscopes.

This activity includes a protocol developed by miniPCRbio.



Fluorescently tagged stem cells edited & imaged at the Allen Institute

Grade levels: 10th - 12th grade

Total field trip time: 3 hours 30 minutes

Recommended pre-lab teaching:

- An introduction to the steps of protein synthesis (transcription and translation)
- Difference between DNA & RNA
- DNA & RNA base pairing rules
- Note: It is not necessary to have covered CRISPR/gene editing prior to this lab

Learning objectives:

- **Determine** how gRNAs and Cas9 work together to target and cut specific DNA sequences
- **Test** how different guide RNAs affect the function of the CRISPR/Cas9 system
- **Apply** an understanding of the CRISPR/Cas9 system to how Allen Institute scientists generate populations of cells with different organelles fluorescently tagged
- **Analyze** real populations of stem cells from the Allen Institute using fluorescent microscopes

Lab skills:

- **Operate** a micropipette
- **Use** the real Cas9 enzyme to cut DNA
- **Load** the wells of an agarose gel and analyze the gel using electrophoresis
- **Follow** a lab protocol that uses multiple reagents and incubation steps

Next Generation Science Standards (NGSS)

Science and Engineering Practices	**Asking Questions and Defining Problems** Ask questions that arise from examining models or a theory to clarify relationships. **Developing and Using Models** Use a model based on evidence to illustrate the relationships between systems or between components of a system.
Disciplinary Core Ideas	**LS1.A: Structure and Function** Systems of specialized cells within organisms help them perform the essential functions of life. (HS-LS1-1) All cells contain genetic information in the form of DNA molecules. Genes are regions in the DNA that contain the instructions that code for the formation of proteins, which carry out most of the work of cells. (HS-LS1-1) **LS3.B: Variation of Traits** Each chromosome consists of a single very long DNA molecule, and each gene on the chromosome is a particular segment of that DNA. The instructions for forming species' characteristics are carried in DNA. All cells in an organism have the same genetic content, but the genes used (expressed) by the cell may be regulated in different ways. Not all DNA codes for a protein; some segments of DNA are involved in regulatory or structural functions, and some have no as-yet known function. (HS-LS3-1)
Cross Cutting Concepts	**Cause and Effect: Mechanism and Explanation** Events have causes, sometimes simple, sometimes multifaceted. A major activity of science is investigating and explaining causal relationships and the mechanisms by which they are mediated. Such mechanisms can then be tested across given contexts and used to predict and explain events in new contexts. **Structure and Function** The way in which an object or living thing is shaped and its substructure determine many of its properties and functions.