Activity: Building A paper model of CRISPR

Step 1: Cut out the Cas8 enzyme and two tabs from the sheet in Figure 1.

Step 2: Place the two tabs over the rectangles in white on the cas9 enzyme. Tape down the short edges of each tab, making sure not to put tape over the long edges. The blue outlines in Figure 2 show where the tape pieces need to go.
Step 3: Cut out the two target DNA molecules, the two guides RNA molecules, the donor DNA and the random nucleotide pieces from the sheet shown in Figure 3.

![Figure 3: The RNA and DNA model Sheet](image)

Step 4: To build the CRISPR-Cas9 model, you will need to model binding the guide RNA to Cas8. Attach guide RNA 1 to the Cas9 enzyme by sliding it under the tabs, as shown in Figure 4.

![Figure 4: Model of CRISPR-Cas9 using guide RNA 1](image)

The loop on the guide RNA should slide under the top tab, and the 3' end of the RNA should slide under the tab on the right.
Step 5: Model Targeting and Binding
Slide target DNA 1 under the guide RNA 1 through the rightmost tab on the Cas9 enzyme. Line up the red targeting sequence on the guide RNA with the complementary sequence on the target RNA, as shown in Figure 5.

Step 6: Model Cleaving
To model how Cas9 cleaves the DNA, use scissors to cut target DNA 1 along the dotted line, as shown in Figure 6. Do not cut the guide RNA.
Step 7: To model a possible outcome of NHEJ when CRISPR-Cas9 is used, tape the cut piece of target DNA 1 together with the “Random Nucleotides” piece between them. These random nucleotides represent a mutation that is likely to inactivate the target gene.
Cas9 protein

“Tabs”
RNA-DNA Model

Target DNA 1

Random Nucleotides

Guide RNA 1

PAM

DNA 1

Guide RNA 1

RNA 1