Center for BioMolecular Modeling

How are you going to connect CRISPR….to something you already teach?

By comparing CRISPR Cas9 and a restriction enzyme….

Both CRISPR Cas9 and a restriction enzyme are endonucleases… that make sequence-specific cuts in double-stranded DNA. So, what is so special about Cas9 that makes it possible for us to now to start thinking about using CRISPR technology to edit the human genome?

The answer to this question lies in the length of the DNA sequence that the two enzymes can recognize.

Restriction enzymes recognize short DNA sequences…usually only 4 to 6 base pairs (bp) in length. And these sequences are palindromes.

> The 6 bp palindromic cleavage sequence for the restriction enzyme Eco RI is GAATTC….
>
> You could expect to find one copy of this sequence in every 4,096 bp of a random sequence of nucleotides. DON'T TELL YOUR STUDENTS THIS! Instead, let them discover this for themselves in the activity described below.
>
> Since the human genome is 3.2×10^9 bp in length, you would expect to find 800,000 Eco RI sites scattered around the human genome. So, if you digested the human genome with Eco RI, you would expect to get a complex mixture of 800,000 different restriction fragments….with an average length of around 4000 bp.

In contrast to a restriction enzyme, ***CRISPR Cas9*** *has the ability to recognize a single "statistically unique sequence" in the human genome*….and cut the genome only once…at that site. This means that we can use Cas9 to "open up" a specific gene in the 3.2 billion bp human genome. And opening up the gene… by cutting the double-stranded DNA at that site… is the first step in editing that gene.

How long (in terms of bp) is that *statistically unique sequence* that can be recognized by Cas9? I am not going to tell you... because that is the point of the activity, "Cas9 cuts a Statistically Unique Sequence in the Human Genome."