Meiosis is the process whereby our genes are shuffled – between maternal and paternal chromosomes – and reduced from a diploid to a haploid state. In this activity, you will have the opportunity to model the molecular mechanism of this shuffling process...otherwise known as “crossing over”.

Note to teachers: Why would you want to teach molecular meiosis? Two reasons:

1. **Because it is hard.** When we simply say to our students “genetic information is swapped between paternal and maternal chromosomes during meiosis in a process called crossing-over” – we reduce this amazingly complex molecular process to an almost trivial piece of information. On the other hand, the molecular mechanism of crossing-over is complicated,...and therefore it is understandable that we have not tried to teach it. With this molecular meiosis activity, we believe it is now possible to teach this hard topic to high school students.

2. **Because this will afford you the opportunity to spiral back** to some basic aspects of DNA biology that you have previously taught your students....and allow them to apply their prior knowledge to this complicated molecular process. In particular, the previous knowledge your students will apply to this process includes:

   i. A DNA strand has a polarity, i.e. a 5’ end and a 3’ end.

   ii. The two strands of DNA are antiparallel – with AT and GC base pairs.

   iii. DNA polymerase synthesizes DNA in the 5’ to 3’ direction.

The crossing-over process that we want to model is an example of a more general process known as “**homologous recombination**”. Homologous recombination was initially understood as a series of events that led to the repair of double-stranded breaks in DNA. We now recognize that a specific variation of this process allows for the deliberate exchange of genetic information between maternal and paternal chromosomes during meiosis.

*This activity is based on the information in Molecular Biology of the Cell, 6th edition by Alberts et.al. – Figure 5-54 and accompanying text.*
It is possible to teach a simplified version of “crossing-over during meiosis” using the foam chromosome models (without the added nucleotides). In this simplified approach, you can start with the two non-sister chromatids, one from Dad (blue) and one from Mom (red) as shown in the left image below. A single crossing-over event – as modeled in the activity that follows – results in the swapping of one end of the blue paternal chromatid with the end of a red maternal chromatid (middle image). Two crossing-over events can result in a paternal chromatid with a swapped section of the maternal chromatid ... and visa versa (right image).

But crossing-over is not simple. It is amazingly complicated. And now there are tools you can use to expose your students to the complexities of crossing-over. So,...here we go. We will begin with foam models of two non-sister chromatids with a specific sequence of nucleotides inserted in the model.

1. Imagine that the model shown below represents paternal (blue) and maternal (red) copies of human chromosome 11.

2. The homologous recombination that results in crossing-over during meiosis begins with a bold step – a double-stranded cut is introduced into one of the non-sister chromatids... the paternal (blue) copy in this example.
3. Following the double-stranded cut, a 5’ to 3’ exonuclease digests away one strand of DNA from each cut end – one nucleotide at a time – moving in the 5’ to 3’ direction. The result of this exonuclease action is a short stretch of single-stranded DNA...representing 3’ extensions on the cut paternal chromosome. At the same time, the homologous DNA sequence on the maternal chromosome is denatured...in preparation for strand invasion.

** Emphasize that strand invasion is only possible as a result of the concerted action of the endonuclease that makes the first double-stranded cut and the exonucleases that generate the 3’ single-stranded ends. Once that is done,.....it is simply the formation of Watson/Crick base pairs between complementary sequences that results in strand invasion....and eventually, crossing-over

4. During strand invasion, the protruding 3’ ends of the paternal DNA form Watson/Crick base pairs with their homologous sequences on the maternal chromosome.

5. DNA polymerase then extends the 3’-OH primers of the paternal chromosomes, synthesizing new DNA (grey nucleotides in this model) in the 5’ to 3’ direction.
6. To resolve these intertwined chromosomes, single-stranded cuts are introduced into the maternal (red) DNA at the sites of the green arrows.

7. Ligation reactions then form the final phosphodiester bonds that seal up the single-stranded nicks in the chromosomes....generating the chromosomes shown below.

Because multiple crossing-over events occur along two homologous non-sister chromatids during meiosis, the red maternal chromosome will have patches of blue paternal chromosome along its length, and visa versa.

All models are wrong – and one thing that is wrong with the above model of crossing-over is that it does not include any of the many proteins that are required to make this happen, like (i) Spo11 that cuts the DNA to initiate the process, (ii) the 5' to 3' exonuclease that nibbles away the ends to create the 3' single-stranded extensions, (iii) Brca1 that loads Rad52 onto the homologous DNA in preparation for strand invasion, and (iv) the large complex of proteins (eukaryotic homologs of E. coli RuvA and RuvB) that provides the scaffolding that holds all the DNA together throughout the process.
Molecular Meiosis Teaching Points

1. Crossing-over is only one of several different ways in which genetic variation is introduced in each new generation. Another mechanism of introducing variation in offspring is the independent assortment (random segregation) of maternal and paternal chromosomes in germ line cells.

2. Crossing-over is a critical event in meiosis in that it ensures that homologous chromosomes have correctly paired up at the metaphase plate....and will therefore be accurately partitioned to daughter cells during meiosis I.

3. Crossing-over is a relatively rare event...with only 2 to 4 events per chromosome.

4. All models are wrong...some are useful. This model is useful in that it highlights some of the molecular events that that occur during crossing-over. But in many ways, it is an over-simplification of the “real” process. Encourage your students to analyze the model....and list ways in which the model fails to represent the process of chromosomal crossing-over. Critiques might include:
   - No proteins (like Spo11, exonuclease, Brca1, Rad50 and the scaffolding complex that organizes and stabilizes the Holiday structure) were included in the model.
   - The model suggested that this process occurs on naked DNA....when in fact this process involves condensed chromosome, with nucleosomes, cohesin, condensin, and etc...

5. Multiple “foundational concepts of DNA structure” provide the molecular basis of the DNA transactions that occur during crossing-over. These foundational concepts include:
   - Each strand of DNA has a polarity,...from 5’ to 3’
   - The two strands of DNA in double-stranded DNA are anti-parallel
   - The two strands of DNA in double-stranded DNA are complementary to each other, i.e., connected by AT and GC base pairs.
   - During strand invasion, simple base pairing rules (AT and GC) insure that the crossing-over event occurs between two homologous chromosomes.
   - All DNA polymerases synthesize new DNA in the 5’ to 3’ direction...by adding new nucleotides to the 3’-end of a “primer” DNA strand.

6. Evolution can be thought of as natural selection acting on a diverse population composed of individuals with slight variations in their genomes. Meiosis is only one of several mechanisms whereby diversity (variation) is introduced into our genomes...as we pass our genes along from one generation to the next.