Recombination Models



A. The Holliday Junction Model

Fig. 1 illustrates the formation of a Holliday junction (I) and its branch migration (II). The branch point (the blue 'X') at the left in I has moved towards its right to give II. Note that symmetric heteroduplex (red/green) is formed during branch migration. The Holliday junction in III is identical to that in II, except that it is in the antiparallel geometry. In II, A-b and a-B are in the left to right orientation. In III, a-B is in the right to left orientation. This can be done by rotating the bottom DNA through 180 degrees about an axis at the junction, and in the plane of the paper. This operation changes the 'X' form junction in II to an 'H' form junction in III.

Color codes: Parental DNA molecules, Red and Green.

The Top strands (one pair of equivalent strands in the red and green DNA) are shown by thick lines. The complementary strands (the second pair of equivalent strands) are drawn as thin lines. The blue lines are phosphodiester bonds. Each blue line is a single phopshodiester bond regardless of how long or short it appears in the diagrams.



B. Structural Manipulations of the Holliday Junction

Fig. 2

Let us take junction III from Fig. 1 (top panel in Fig. 2), and stack the arms differently. Pull up arms A and b, and pull down arms B and a. The junction will now take up the form shown in the middle panel of Fig. 2. Arm A is stacked over arm B; arm b is stacked over arm a. If you like, rotate the whole junction through 90 degrees clockwise, so that it looks more like the junction at the top.

Notice that it is the thin strands that are crossed in the junction at the top; and it is the thick strands that are crossed in the junction at the bottom.

Resolve the junction <u>at the top</u> by cutting the crossed thin strands (or the phosphodiester bonds indicated by the long blue lines). Notice that each DNA product carries the heteroduplex (symmetric), and each has the flanking markers in the parental configuration (A over b and a over B).

Now, resolve the junction <u>at the bottom</u>, also by cutting the crossed (thick) strands (or the phosphodiester bonds indicated by the long blue lines). Each product has the same symmetric heteroduplex as before. However, the flanking markers are in the crossed-over or recombined configuration (A over B and a over b).

Here is a common sense rule:

If you form the Holliday junction by crossing one pair of strands (say, the thin strands in the diagrams), and if you resolve the junction by cutting the same pair of strands (again the thin strands), there will be no cross-over of the flanking markers. Remember, each DNA partner has two strands, and you have to cross both of them to get recombination (or all four strands for two duplexes involved in genetic exchange). If you cross a pair of strands (for Holliday formation), and cross the same pair again (for Holliday resolution), you have reversed the effect of the first crossing. Or, you go back to the parental state.

If you first cross the thin strands and then resolve the junction by crossing the thick strands, you get cross-over of the flanking markers. Notice that you have now crossed all four strands: first the two thin strands, and then the two thick strands.

C. One last gyration of the Holliday junction





Let us take the branch-migrated 'X-form' junction (II in Fig. 1) and place it alongside its isomerized H form at the bottom of Fig. 2 (see Fig. 3). Let us rotate the a-b cylinder (the bottom duplex) through 180 degrees about an axis at the junction and in the plane of the paper. The resulting Holliday junction has now the 'X-form' (IV).

Notice that the 'X' is formed by the thick strands in IV, whereas it is formed by the thin strands in II. Also, the flanking markers in the top duplex in II are A and b; they are A and B in II. Similarly, the flanking markers for the bottom duplex in II (B and a) have now switched to a and b in IV.

D. The Meselson-Radding Model



Fig. 4 illustrates the model. The strand nick is made in one of the two duplexes. The nicked strand (green), the donor of genetic information, is used to invade the red partner, resulting in a D-loop in the recipient duplex. With extension of the heteroduplex, the D loop is expanded. The gap in the green DNA is filled by repair synthesis. The D-loop is chewed away (black slashes), and the nicks are closed. A Holliday junction is formed at the end point of the heteroduplex. Note that the heteroduplex (red/green) is present only on one duplex but not the other (or it is asymmetric).

Note that branch migration of the Holliday junction can generate symmetric heteroduplex on either side of the Meselson-Radding region (asymmetric heteroduplex).

E. The Double Strand Gap Repair Model



The model is diagrammed in Fig. 5. The green DNA, which is the recipient of information, is broken and gapped. Unequal DNA degradation in the two strands generate 3' extensions which can invade the red duplex. The extruded strand (thin red) is a D-loop. Repair synthesis (dashed lines) and end joining results in two Holliday junctions. Since the missing green information is replaced by using the intact red DNA as template, the genetic information is all red in the gap-repair region. The borders of this region are indicated by the vertical dashed black lines.

Between the right border of gap repair and the right Holliday junction, you can see asymmetric heteroduplex (red/green on the top duplex). Similarly, between the left border of gap repair and the left Holliday junction, there is asymmetric heteroduplex (red/green on the bottom duplex).

Note that this model is inclusive of the Meselson-Radding model (asymmetric heteroduplex) and the Holliday model (symmetric heteroduplex).

F. Recombination Models and Segregation Patterns

Apply the models to the four chromatid stage of the meiotic cell. Remember that there are two chromosomes with red DNA in the region of interest to us, and two chromosomes with green DNA in the same region. We consider exchange between a red DNA duplex and a green DNA duplex as diagrammed in the models, and arrive at the consequences of the exchange on the segregation pattern.

1. Holliday model-----> Symmetric heteroduplex ----> Two colonies with half-sectors in yeast by germination of the four spores from a meiotic event; two pairs of non-identical sister spores in Ascobolus. 4: 4 aberrant segregation.

2. Meselson-Radding model-----> Asymmetric heteroduplex----> One sectored colony in yeast; one pair of non-identical sister spores in Ascobolus. 5:3 aberrant segregation.

3. Double strand gap repair model-----> Conversion of green DNA to red DNA in the region of repair. 6:2 aberrant segregation.