Teaching Genetic Linkage and Multiple Crossovers with Sets of Cards as Chromosomes†

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INTRODUCTION

Many undergraduate genetics courses feature genetic mapping, which is readily done in Drosophila melanogaster (1) and Sordaria fimicola (2). Mapping genes by observing phenotypic recombination illustrates the impact of the physical chromosome on inheritance. It also paves the way to an understanding of linkage disequilibrium, genome-wide association studies, and quantitative trait locus analysis.

Students have misconceptions regarding when crossovers happen (3), but misconceptions involving multiple crossovers have not been systematically investigated. Nevertheless, students appear to have difficulty reconciling the facts that a) genetic distance is defined in terms of recombination frequency (RF) and b) as RF increases it becomes an increasingly inaccurate predictor of genetic distance due to multiple crossovers. Students especially struggle with three-point test crosses (4), which require stepwise problem solving. Multiple external representations (e.g., physical models) may help students, in part by reducing cognitive load (5).

In the following exercise, students simulate these abstract topics with playing cards. In other card models of chromosomes, each card represents a chromosome (6); here, each card represents a gene, with a set of five cards representing a chromosome. Students manipulate model “chromosomes” by hand and work together to generate data that challenge the naive view that genetic distance and recombination frequency are equivalent. They also acquire skills for interpreting test crosses and comparing their results to published genetic distances.

PROCEDURE

Genetic linkage and RF should be introduced before or concurrently with this exercise. A student handout (Appendix 1) and answer key (Appendix 2) are provided; the instructor can let students do the handout on their own or walk students through, discussing each question in turn. The advantage to the latter approach is that many questions require data from the whole class to answer. The exercise takes approximately 60 minutes, though this varies, depending on whether the material has previously been covered. Appendix 2 also provides learning outcomes and additional assessment questions.

Students form pairs to share a deck of cards and a six-sided die. Each of the two students forms both a red and a black set of cards from ace to five (Fig. 1). They are told that each of their “poker hands” is part of a chromosome, and the two chromosomes are homologs of each other. Each card number represents a gene, with the two sets having different alleles represented by color. If there is complete linkage, all chromosomes will remain totally black or totally red; that is, there will be no recombination.

Students are told that between each pair of genes, e.g., ace and two, there is some chance of recombination caused by crossovers. Students simulate these recombination probabilities by rolling the die. For this exercise, we assume an RF of 1/6 (16.7%) between genes ace and two and between four and five (so recombination occurs on a roll of one), and an RF of 2/6 (33%) between genes two and three and genes three and four (so recombination occurs on a one or two). Recombination requires students to exchange each gene below the site of recombination between the homologous chromosomes (Fig. 1). Some students misunderstand the simulation, thinking they should roll the die for every individual gene and swap that particular gene if they roll 1, while leaving other genes unchanged. This will cause the simulation to fail, as distant gene pairs will not have higher RF. Thus the instructor may choose to draw two simulated chromosomes on the board, then carefully illustrate the effects of a single crossover.

Each student reports whether their simulated chromosomes are recombinant between ace and three, three and five, and ace and five. The class data are used to calculate RFs. The ace to five region should show less recombination than the RFs of ace to three and three to five combined. This is because a crossover between ace and three can be cancelled out by a crossover between three and five. This

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can be a difficult conceptual leap for students, who may require assistance.

Students are now introduced to genetic distance, a measure of crossover frequency related to but distinct from RF. As genetic distance increases, multiple crossovers are increasingly likely, and any even number of crossovers results in genes restored to their original configuration. Thus, when possible, genetic distance should be calculated using RF between closely linked genes.

However, multiple crossovers occur even between closely linked genes. Mapping functions estimate true genetic distance using RF (Appendix 1). I introduce Haldane’s mapping function:

\[
\text{Genetic distance in map units} = -50 \ln (1 - 2 \times \text{RF})
\]

(Equation 1)

where RF is a proportion between 0 and 1 (7). This function is simple, corrects for multiple crossovers, and results in additive map distances. Other more complex and more accurate functions can be used if the instructor wishes to focus on interference (8).

Finally, the model can optionally be extended to three-point test crosses by presenting simulation data differently. Each student reports on the alleles present on one of their chromosomes (determined randomly or by some other method; Appendix 1). Red alleles for *ace*, *three*, and *five* are dominant (and capitalized; *A*, *T*, and *F*, respectively) and black alleles recessive. Small lab sections may produce different results from expectations due to sampling error, so I provide a simulated data set that can be combined with student results (Appendix 3). Students discuss which combinations of alleles should be most common (parentals) and rarest (those requiring two crossovers, with the middle gene swapped relative to parentals). Finally, students calculate RF between more closely linked pairs of genes, find those genetic distances using the mapping function, and add those values together to estimate genetic distance between *ace* and *five*.

**CONCLUSION**

This method is a “handy” way to teach students about genetic mapping. I focus on the importance of multiple crossovers, but it could be modified to cover topics such as the role of homozygous recessive testers, or mismatch between genetic maps and physical maps, or simplified to simply explain crossovers. Upon questioning, students generally found this approach helpful.

Cards allow students to reproduce the mechanisms of crossing over conveniently, memorably, and kinesthetically. They are thus a useful alternative to writing out crossovers, which typically involves “before” and “after” pictures in which the connection may not be clear.

**SUPPLEMENTAL MATERIALS**

Appendix 1: Student handout
Appendix 2: Instructor notes and answer key
Appendix 3: Three-point test cross simulation

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