

medical facilities and care, which in turn could possibly increase our mutation load (Crow, 1997). In addition, somatic mutations, which are not scored in the assay of this study, could increase cancer rates and reduce lifespan (Yang *et al.*, 2003; Morley, 1995). At the same time, thousands of new industrial chemicals are introduced into our environment every day, most of them being potent mutagens (for example chemicals from tobacco use and some pesticides), thereby increasing the mutation rate in germinal tissues. These increases in mutations might, in turn, increase the genetic load in future generations.

References: Caceres, M., J.M. Ranz, A. Barbadilla, M. Long, and A. Ruiz 1999, *Science* 285: 415-418; Charlesworth, B., and C.H. Langley 1989, *Annu. Rev. Genet.* 23: 251; Crow, J.F., 1997, *Proc. Natl. Acad. Sci.* 94: 8380-8386; Crow, J.F., 2000, *Nat. Rev. Genet.* 1: 40-47; Drake, J.W., B. Charlesworth, D. Charlesworth, and J.F. Crow 1998 *Genetics*: 645-663; Eyre-Walker, A., and P.D. Keightley 1999, *Nature* 397: 344-347; Ewing, B., and P. Green 2000, *Nature Genet.* 25: 232-234; Keightley, P.D., 1996, *Genetics* 144: 1993-1999; Kondrashov, A.S., 1995, *J. Theor. Biol.* 175: 583-594; Kondrashov, A.S., 1997, *Annu. Rev. Ecol. Syst.* 28: 391-435; Kondrashov, A.S., 2001, *Trends Genet.* 17: 75-77; Lindsley, D.L, and G.G. Zimm 1992, *The Genome of Drosophila melanogaster*, New York: Academic Press, Inc; Lynch, M., J. Blanchard, D. Houle, T. Kibota, S. Schultz, L. Vassilieva, and J. Willis 1999, *Evolution* 53: 645-663; Lyttle, T.W., and D.S Haymer 1992, *Genetica* 86: 113-126; Mackay T.F., 1994, *Genetics* 111: 351-374; Morley, A.A., 1995, *Mutat. Res.* 338:19-23; Mukai, T., 1972 *Genetics* 72: 335-355; Muller, H. J., 1950, *Am. J. Hum. Genet.* 2: 111-176; Yang Z., R. Simon, and B. Rannala 2003, *Genetics* 165: 695-705; Yodder, J.A., C.P. Walsh, and T.A. Bestor 1997, *Trends Genet.* 13: 335-340; Woodruff, R.C., H. Huai, and J.N. Thompson, jr., 1996, *Genetica* 98: 149-160.



Recombination mapping of P-element transposon inserts: A new set of laboratory exercises for an undergraduate genetics course.

Marcus, J.M. Department of Biology, Western Kentucky University, 1 Big Red Way, Bowling Green, KY, 42101.

Despite the large number of P-element transposon insertion lines available to *Drosophila* researchers and the good location data associated with these insertions on the cytogenetic and genome maps (Spradling *et al.*, 1999), very few of these inserts have been formally placed on the recombination map (but see Marcus, 2003 for exceptions). FlyBase (2003) does provide a table for the conversion of cytological map position to recombination map position (<http://flybase.bio.indiana.edu/maps/lk/cytotable.txt>), but the resolution of this conversion table (generally 1 map unit) is rather coarse. Because of the high potential utility for conducting transposon mutagenesis of genes with unknown cytogenetic locations, it would be desirable to accumulate a set of transposon insertions with known locations on both the cytogenetic and recombination maps

This is not a difficult task because many transposon inserts are marked with easily traceable markers (such as w^+). In fact, there are hundreds of transposons that can be traced using the same eye color marker, but which exist in many different locations. The P-element insertion stock collection therefore represents a very useful opportunity to give undergraduate genetics students unique, but equivalent genetic unknowns that can be used for recombination mapping exercises. In some sense, this represents an advance over other types of eye color unknowns (*e.g.*, MacIntyre, 1974; Pye, 1980), because it eliminates a frustration that many genetics students express when doing *Drosophila*

genetics laboratory exercises: that they are re-mapping genes that have been mapped before hundreds of times, and that the labor-intensive exercises are a waste of time.

A series of *P[lacW]* transposon insertion lines (in a *w* background) were selected for this exercise that spanned the right arm of the third chromosome, and were mapped against a common mapping strain *w¹¹¹⁸; h¹ ri¹ e^s*. Insertion lines were selected such that the interrupted genes associated with the insertion had been identified, but not genetically mapped. Students created F1 hybrids by crossing males carrying *P[lacW]* with non-virgin females of the mapping strain. Then virgin females F1 hybrids with pigmented eyes were backcrossed to males from the mapping strain and the F2 progeny were scored for eye color, body color, and wing vein phenotypes. The *h¹* phenotype was not scored, because it was not easily visible through our student-grade dissecting microscopes, and was also more than 50 map units from most of the transposon inserts making it uninformative for recombination mapping. At Western Kentucky University, faculty proctor most teaching labs themselves, so the instructor was present to assist students in identifying phenotypes, and was able to enforce quality control over student data.

While the crosses were in progress, students used the FlyBase and NCBI databases to learn about their particular interrupted genes in a bioinformatics exercise and during the course of the experiments, students were asked to do three writing assignments related to these exercises: first to summarize what they were able to learn about their insertion from the bioinformatics exercise, second to summarize their own recombination data and calculate the map position of a single *P[lacW]* insertion, and finally to create and describe a map that summarizes all of the recombination data collected by the entire class. This approach to teaching genetics allows students to develop a strong identification with “their gene”, introduces them to some of the modern tools of genetics research, and allows students to make small but real contributions to our knowledge of the *Drosophila melanogaster* genome. By and large, the students seemed to be very enthusiastic about these exercises, including several students who were not generally enthusiastic about more typical laboratory and lecture experiences in my genetics course.

There were 26 students in my Fall 2003 undergraduate genetics course, and a total of 27 transposon insertion lines were mapped (one for each student, plus a spare, in case one of the crosses did not work and the instructor needed to provide a student with a replacement). This, coincidentally, represents all of the transposon insertion stocks currently available from chromosome arm 3R that fit the four criteria used to select lines: the stock carries a *P[lacW]* insertion, the insertion is associated with a particular open reading frame of known function, the stock is available from the Bloomington Stock Center, and the insertion has not been mapped by recombination before. All recombination map positions for P-element insertions that were obtained by students were verified in a parallel set of genetic crosses conducted by the instructor. Student data that was clearly faulty was discarded, but the majority of student data closely matched the data gathered by the instructor and the data sets were combined to generate the map positions presented here.

The recombination map positions that were obtained in these experiments are shown in Table 1. Maximum likelihood standard errors for each meiotic recombination map distance were calculated according to Weir (1996). Due to a minor inconsistency in how FlyBase (2003) reported the recombination map position for one insertion line *P[lacW]Pp1-87^{Bj6E7}*, this line was inadvertently included among the student unknowns even though its recombination map position ($3-51.1 \pm 0.5$, Reuter *et al*, 1986) was already established. As is typical of such occurrences, a student realized this before the instructor, but she completed the mapping experiment herself to see how her results compared to those published previously. Reuter *et al*. (1986) mapped their mutation in *Pp1-8* using a locus (*kar*) that was closer than the mapping loci used in our experiments, so their result is probably a better estimate of the actual genetic map position.

Table 1. Map positions of *P[lacW]* transposon insertions on chromosome arm 3R. Underlined entries indicate markers that had already been assigned meiotic map positions at the beginning of this study. The map position in brackets is a predicted map position, based on cytogenetic position. These data, and cytogenetic data for the markers used in this study, were obtained from Flybase (2003). SE is the standard error of each of the recombination frequency calculations, and N is the number of flies scored to calculate the map positions. The student who mapped each mutation is listed in the final column.

Mutation	Cytogenetic Positon	Meiotic Map	SE	N	Student
<u><i>ri</i>¹</u>	<u>077E03</u>	<u>46.8</u>			
<i>P[lacW]Karybeta</i> ^{3/3A4}	082D01-02	47.1	0.34	293	Thomas Thacker
<i>P[lacW]ksl</i> ^{5E2}	083A05-06	52.1	1.18	358	Nicole Weathers
<i>P[lacW]noi</i> ^{3E7}	083B01-02	47.0	0.17	575	Erica White
<i>P[lacW]Atus</i> ¹⁹³⁸	083B04-07	47.6	0.55	254	Amanda Maupin
<i>P[lacW]sec23</i> ^{13C8}	083B06-07	47.9	0.56	352	Shawn Peavie
<i>P[lacW]cas</i> ^{1C2}	083C01-02	58.5	1.36	556	
<i>P[lacW]Dhod</i> ⁶³⁵¹²	<u>085A05-07</u>	<u>48.0</u>			
<i>P[lacW]neur</i> ^{6B12}	<u>085C09-10</u>	<u>48.5</u>			
<i>P[lacW]pum</i> ^{hem}	<u>085C04-D01</u>	<u>48.5</u>			
<i>P[lacW]Tfllf-beta</i> ^{3C1}	086C03-04	59.1	2.10	244	Jaivonna Crook
<i>P[lacW]Vha</i> ^{55j2E9}	<u>087C02-03</u>	<u>51.7</u>			
<i>P[lacW]Pp1-87</i> ^{3j6E7}	087C11-13	57.2	2.27	182	Susannah Craig
<i>P[lacW]l(3)87Egs</i> ²¹⁴⁹	<u>087E10-11</u>	<u>[53]</u>			
<i>P[lacW]sqd</i> ^{6E3}	087F02-03	55.6	1.83	383	Lindsay Gardner
<i>P[lacW]fifl</i> ^{L4179}	087F07-08	57.0	1.86	342	Jaime Crocker
<i>P[lacW]B52</i> ^{s2249}	087F07-08	54.8	2.15	289	David Arboe
<i>P[lacW]trx</i> ^{14A6}	<u>088B01</u>	<u>54.2</u>			
<i>P[lacW]MRG15</i> ^{6A3}	088E11-12	62.1	2.00	197	James Heltsley
<i>P[lacW]CSN5</i> ^{L4032}	089D01-02	61.0	1.33	495	Kelly Bowersox
<i>P[lacW]Dad</i> ^{1E4}	089E10-11	59.0	1.50	460	Chris Carter
<i>P[lacW]Trap80</i> ^{s2956}	090F01-02	62.5	1.39	389	Matt King
<i>P[lacW]nos</i> ^{3B6}	<u>091F07</u>	<u>66.2</u>			
<i>P[lacW]bon</i> ^{S048706}	092E	70.7	0	140	Tommy Crockett
<i>P[lacW]Rab1</i> ^{12D}	093C01-02	61.3	1.70	297	Megan Jackson
<i>e</i> ^s	<u>093D01</u>	<u>70.7</u>			
<i>P[lacW]mod(mdq4)</i> ^{L3101}	<u>093D09-10</u>	<u>70.7</u>			
<i>P[lacW]how</i> ^{E7-3-4}	093F13	77.7	1.64	242	Joey Oliver
<i>P[lacW]Dph5</i> ^{L4910}	094B04-05	80.5	1.39	457	Margaret Au
<i>P[lacW]CycB3</i> ^{L6540}	096B03-05	84.3	1.78	369	Christina Archey
<i>P[lacW]OstStt</i> ^{3j2D9}	096B19-20	81.7	1.87	281	Alecea Davis
<i>P[lacW]scrib</i> ^{17B3}	097B08-09	81.1	1.38	490	Jennifer Dennison
<i>P[lacW]Takr99D</i> ^{s2222}	099D01-02	97.8	3.07	210	Cheri Watson
<i>P[lacW]hdc</i> ^{Fus-6}	099F	98.6	1.98	513	Janie Baxter
<i>P[lacW]dco</i> ^{3B9}	100B02-04	88.2	1.82	435	Kim Phillips
<i>P[lacW]S057302</i>	100C	100.5	2.59	312	Priscilla Hamilton
<i>P[lacW]awd</i> ^{2A4}	100E01-02	98.0	2.18	417	Kate Hertweck

In most cases, the 95% confidence intervals for the meiotic map positions of each locus calculated from the class data (calculated after Snedecor and Cochran, 1989; data not shown) overlapped with the map location predicted by FlyBase (2003). However, in other cases, our calculated map position differs significantly from what was expected. The observed disparities may be due to viability differences between F2 genotypes, the rather large distances between some of the transposon inserts and our mapping markers (which will tend to systematically underestimate recombination map distance (Haldane, 1919)), or to sampling errors caused by the small samples sizes of F2 progeny for some of the transposon insertion lines.

Overall, the data presented here are consistent with the meiotic recombination map positions predicted by FlyBase (2003) and confirm that students remain capable of making contributions to the

field of *Drosophila* genetics as a part of their undergraduate coursework (Wright, 1932). Future student laboratory exercises will extend these investigations to other chromosome arms.

References: FlyBase, 2003, *The Drosophila genetic database* (<http://flybase.bio.indiana.edu>); Haldane, J.B.S., 1919, *J. Genet.* 8: 299-309; MacIntyre, R., 1974, *Dros. Inf. Serv.* 51: 158; Marcus, J.M., 2003, *Genetics* 163: 591-597; Pye, Q., 1980, *Dros. Inf. Serv.* 55: 171; Reuter, G., R. Dorn, G. Wustmann, B. Friede, and G. Rauh 1986, *Mol. Gen. Genet.* 202: 481-487; Snedecor, G.W., and W.G. Cochran 1989, *Statistical Methods*, Iowa State University Press, Ames, Iowa, Eight Edition; Spradling, A.C., D. Stern, A. Beaton, E.J. Rhem, T. Lavery, N. Mozden, S. Misra, and G.M. Rubin 1999, *Genetics* 153: 135-177; Weir, B.S., 1996, *Genetic Data Analysis II: Methods for Discrete Population Genetic Data*, Sinauer, Sunderland, Mass., Second Edition; Wright, S., 1932, *Am. Nat.* 66: 282-283.

Call for Papers

Submissions to *Drosophila* Information Service are welcome at any time. The annual issue now contains articles submitted during the calendar year of issue. Typically, we would like to have submissions by 15 December to insure their inclusion in the regular annual issue. Submissions in Microsoft Word, which is now the program we use for our page setup, are especially helpful. Submissions by email are also possible, but if they are sent as attached files, we have greatest success using MS Word or Rich Text Format. Pictures and line drawings should be as sharp and high contrast as possible. Where tables are concerned, it is useful to have a paper copy to facilitate accurate formatting. Details are given in the Guide to Authors.

Standing Orders

Several years ago, formal standing orders were discontinued due to the need to obtain prepayment for issues. "Standing Orders" are now handled through a mailing list of active subscribers. All individuals on the active subscriber list will receive notices for forthcoming regular and special issues and a Standing Order Invoice to facilitate prepayment. If you would like to be added to the *Drosophila* Information Service mailing list, please write to the editor, Jim Thompson, Department of Zoology, University of Oklahoma, Norman, OK 73019.