GENETIC CHANGE OF RECOMBINATION VALUE IN *DROSOPHILA MELANOGASTER*. I. ARTIFICIAL SELECTION FOR HIGH AND LOW RECOMBINATION AND SOME PROPERTIES OF RECOMBINATION-MODIFYING GENES[1]

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ABSTRACT

A high and low selection line were formed by individual selection of females on the basis of their recombination. In the high line, recombination between *Gl* and *Sb* was increased from 14.8 percent to about 30 percent in twelve generations, when a plateau was apparently reached. Realized heritability was 0.12. The absence of a response to selection for low recombination is attributed mainly to genetic random drift, and partially to directional dominance and directional gene frequencies. Natural selection was found to act against increases of recombination above a level of about twenty percent in the measured interval. High recombination tended to be recessive to low recombination. In both selected lines and unselected stocks, intervals proximal to the centromere tended to have a higher recombination variance than distal intervals.

THE possibility that linkage intensity between genes is modified by natural selection was first pointed out by FISHER (1930). He argued that "the presence of pairs of genes in the same chromosome, the selective advantage of each of which reverses that of the other, will always tend to diminish recombination," while high recombination is favourable in recombining two or more advantageous mutations in the evolutionary process. KIMURA (1956) developed a mathematical model of a genetic system which leads to closer linkage by natural selection. A similar model was also studied by BODMER and PARSONS (1962). These mathematical models may be important in explaining the evolution of widespread supergenes or gene complexes. Recently, NEI (1967) studied a more general model of linkage modification and reached the conclusion that natural selection will almost always result in the tightening of linkage between interacting linked loci in the presence of a recombination-modifying mechanism. NEI (1968) further postulated that the evolutionary increase of linkage intensity observed in terms of the number of nucleotide pairs per unit map length is the result of natural selection in the presence of epistasis.

There is a considerable amount of *indirect* evidence to support the possibility

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of linkage modification by natural selection. Structural changes of chromosomes such as inversions and translocations may directly affect recombination of the genes involved and those in adjacent regions of the same chromosome. It is also known with several species (Drosophila, Neurospora, Schizophyllum, etc.) that recombination frequency is under the control of single genes or is influenced by structural changes of another chromosome (e.g., GOWEN and GOWEN 1922; CATCHESIDE 1968; SCHAAP and SIMCHEN 1971; SCHULTZ and REDFIELD 1951). Selection for increased and/or decreased recombination has been successful in a number of experiments in Drosophila (DETLEFSON and ROBERTS 1921; PARSONS 1958; MUKHERJEE 1961), in *Tribolium* (DEWEES 1970) and in the lima bean (ALLARD 1963). Nevertheless, there is no *direct* evidence that linkage intensity has been changed by natural selection. As emphasized by NEI and IMAIZUMI (1968), natural selection is quite different from artificial selection in the mode of linkage modification.

The present study was initiated to investigate the genetic nature of linkage modification under both artificial and natural selection in *Drosophila melanogaster*. The scheme of natural selection is a simulated one with strong epistasis induced between the gene loci under investigation (c.f. KIDWELL 1972). As will be seen from the present and following papers, both artificial and natural selections were effective in changing recombination frequency. In the present paper the results of artificial selection and tests of recombination in the foundation stocks will be reported. The results of genetic tests of the variability of recombination among both selection and unselected lines will also be described.

MATERIALS AND METHODS

The third chromosome genes Glued, *Gl*(3-41.4) and Stubble, *Sb*(3-58.2) were used in the present study for the reasons that they are dominant, homozygous lethal, easily recognized and are separated by 16.8 cM which include the centromere. Dominant markers, that are homozygous lethal, made possible the use of unmarked males with a greater potential for genetic diversity. They were also an essential feature of the selection scheme in the second experiment which will be reported in the accompanying paper. A region of 15–20cM provided a potential for altering recombination in both directions. This interval also allowed a small percentage of double crossing over between the two markers. There is compelling evidence that the centromeric region tends to be most affected by genetic and environmental agents that alter recombination frequency (BODMER and PARSONS 1962; LUCCHESI and SUZUKI 1968).

Flies were kept in half-pint milk bottles containing a fixed volume of a conventional corn meal-molasses-agar medium. Proprionic acid was added as a mold inhibitor. Temperature was controlled at $25 \pm 1°C$ unless stated otherwise. Humidity was not controlled.

All bottles producing females for recombination tests were kept in the standard culture conditions described above. Virgin females were isolated between one and seven hours after emerging. They were individually mated with a male of appropriate genotype in a shell vial where they remained for 24 hr. Each pair was transferred to a milk bottle where the female was allowed to deposit eggs for five days, the first brood. In the two-way selection experiment the parents were transferred to new bottles to produce a second brood which was used to produce the next generation. In all other recombination tests the parent flies were discarded after the first brood of eggs was laid. Recombination estimates were made on the basis of the genotypes of the progeny hatching from the first brood.

The base population for the present experiment was formed early in 1967 from a marker

stock and five wild-type stocks. The marker stock carried *Gl* and *Sb* in coupling balanced over the LVM inversion and was obtained from the Oak Ridge National Laboratory in 1967. Three of the five wild-type stocks, Princeton, Ames and Oregon R-C were obtained from J. W. GOWEN in 1961. They had been maintained in mass culture in half-pint milk bottles for more than 30 years. One other stock, Nettlebed, was obtained from E. C. KELLER, JR., in 1964; and the remaining one, Cranston, was collected locally in 1963.

In order to obtain a broad genetic background the following procedure was adopted in the synthesis of the base population: the marker stock was crossed with a large sample of each of of the five wild stocks; *Gl Sb/Gl+ Sb+* males in the offspring generation were backcrossed to a large sample of their respective wild stocks for two generations, in order to reduce the genetic contribution of the marker stock; the five groups were then intermated in every possible combination and equal numbers of each resulting progeny class were taken to form one base population. This population was maintained by random mating for two generations in bottles prior to the start of the experiment with the restriction that only *Gl Sb/Gl+ Sb+* males and wild-type females were allowed to breed.

The selection and mating scheme is shown diagrammatically in Figure 1. A random sample of 25 *Gl Sb* females (in coupling) was taken from the base population and individually tested for recombination by mating to a wild-type male from the base population. On the basis of recombination testing the second brood progeny of the five highest and five lowest females in generation 0 were selected to establish the high and low lines respectively. Within each of the two lines, single pair matings among the selected five families were made in every possible combination, yielding a total of 25 types. Matings of each type were made in duplicate as insurance against failure of a single mating. The first brood progeny of each of the 25 matings were used to evaluate the frequency of recombination in their mothers. According to this test of recombination frequency the second brood progeny of the five highest females were selected in the high

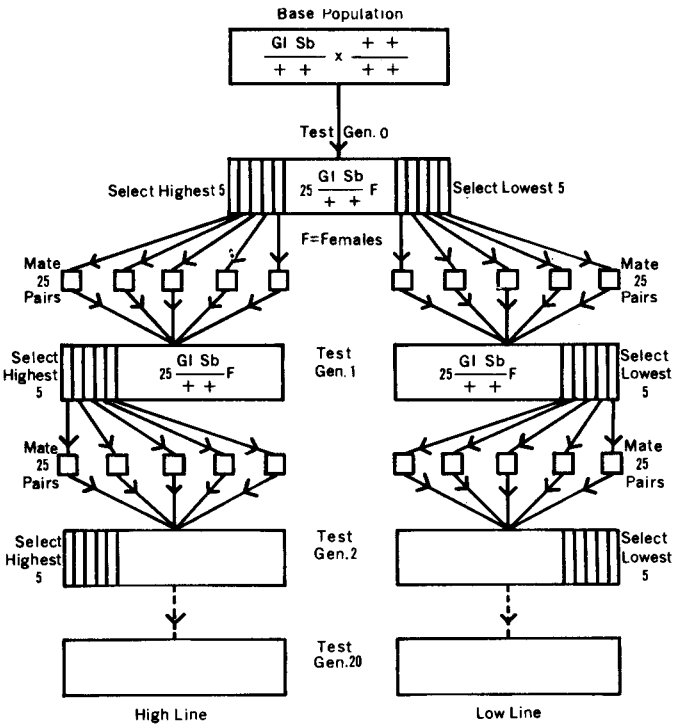

FIGURE 1.—Selection and mating scheme.

line, and those of the five lowest were selected in the low line. However, due to low fecundity, it was often necessary to increase the number of females selected, from five to a maximum of ten in some generations of the high line, particularly during the second half of the experiment. In the low line, only occasionally was it necessary to increase the number of females selected, from five to six or seven.

The cycle of selecting, mating and testing was repeated for a total of 20 generations. The two lines were continued independently, but their generations were kept in phase. Recombination testing was therefore carried out in an identical external environment for the same generation of both lines.

RESULTS

Response to selection: Figure 2 shows graphically the percent recombination value between *Gl* and *Sb* over 20 generations of selection. Solid lines indicate means of the 25 females tested each generation. Dotted lines indicate means of selected females, weighted according to the number of their progeny which were tested in the next generation. The mean percent recombination of the random sample of 25 females from the base population, measured in generation 0, was 14.8. In the same generation the mean percent recombination of the five high-selected females was 20.7 and that of the five low-selected females was 9.6.

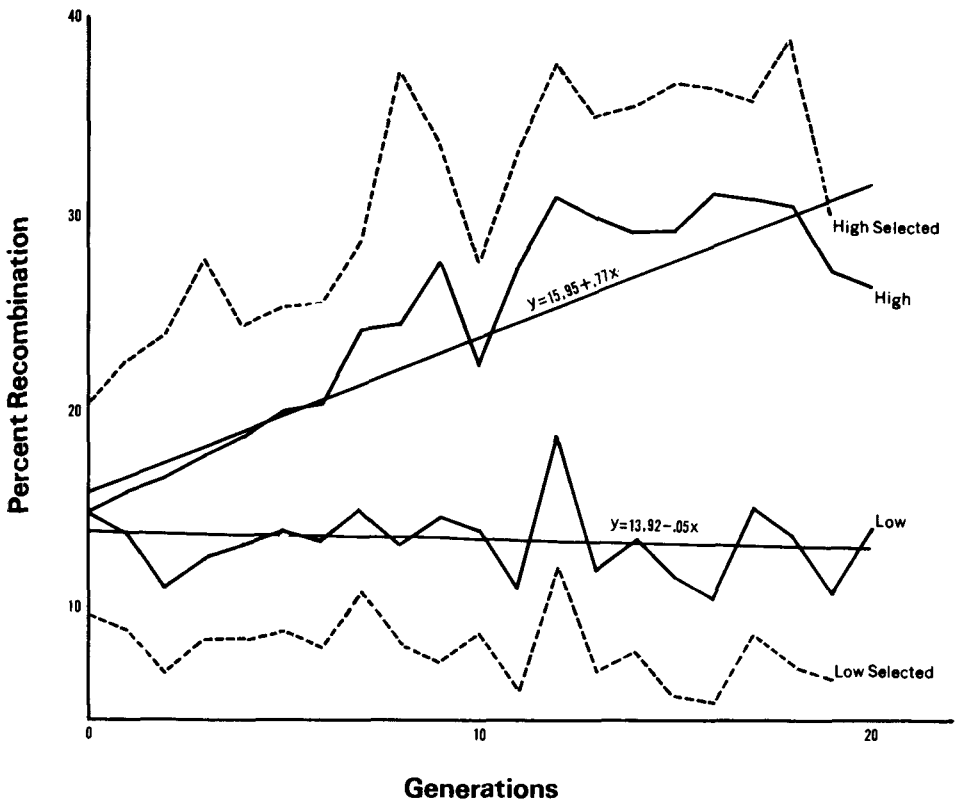

FIGURE 2.—Mean recombination between *Gl* and *Sb* in the high and low selection lines.

Recombination frequency in the high line increased by a total of about five percent at a remarkably steady rate between generations 1 and 6. The average rate of increase almost doubled in the next six generations, but changes from one generation to the next were less regular. From generations 12 to 18, recombination frequency did not vary more than one percent above or below 30 percent. It appears that a plateau was reached at this level. Interpretation of the apparent fall in recombination in the last two generations should be made with care. Fecundity became very low in both lines at generation 20, and recombination frequencies were estimated on the basis of smaller progeny groups. Over 21 generations the regression coefficient of recombination frequency on generation number was .775 ± .100, which was significantly different from 0 ($P < .001$). Realized heritability was .122 ± .015. The cumulated selection differential, over 20 generations, was 126.63 percent recombination units.

In contrast to the high line, response to selection for decreased recombination in the low line was not consistent in either direction or amount over more than two or three consecutive generations. The regression coefficient of recombination frequency on generation time was −.052 ± .072 which was not significant ($P > .05$). Realized heritability was .010 ± .013. There was therefore no significant response to selection, despite a cumulated selection differential over 20 generations of 109.35 percent recombination units.

Both the data on selection for increased and decreased recombination suggest that uncontrolled environmental effects and chance were together important in affecting the variation in recombination among generations. This variation increased in both lines after the first six generations of selection and may reflect an increase in the degree of inbreeding. The inbreeding coefficient at generation 10 is estimated to be .402 from the formula $F_t = 1 - (1 - 1/2N)^t$, where $t =$ generation number and $N =$ number of parents per generation. ROBERTSON (1961) showed that in a population under artificial selection, the effective population size may be less than the actual number of parents selected. However, in the present experiment the heritability of recombination value appeared to be too low for selection to have a significant effect on the inbreeding coefficient.

There was a marked decrease in fertility of selected females in the high line as the experiment proceeded. No significant change in fertility was seen in the low line. These observations were confirmed quantitatively by dividing the effective selection differential by the expected selection differential for each generation (FALCONER 1960). The mean value of this fraction over 20 generations in the low line is 0.97. In the high line the comparable means are 0.95 for the first seven generations and 0.63 for the remaining thirteen generations. These results suggest that natural selection had little or no observable effect below a level of about twenty percent recombination. Above that level natural selection appeared to act fairly strongly against selection for increased recombination.

After 20 generations, artificial selection was discontinued due to increasing difficulty in obtaining adequate numbers of progeny for testing and continuing the lines. They were, however, maintained by mass mating and retested after twenty generations of relaxed selection. Mean recombination was 20.5 percent

in the relaxed high line and 9.6 percent in the relaxed low line. Total numbers of progeny counted were 1305 and 1113 in the two lines respectively. From these limited data it would appear that recombination fell in both groups when selection was relaxed. The decrease in the high line was about twice that in the low line in terms of percent recombination, but the ratio of the means in the two lines was unchanged at approximately 2:1. Some part of the decline in the high line is probably due to natural selection acting against high recombination value, but the reduction in low line suggests that some unknown environmental changes are also responsible for the reduction.

Crosses between selection lines: At generation 17, reciprocal crosses were made between progeny of selected females of the high and low lines. In order to estimate the direction and magnitude of dominance of alleles influencing recombination, F_1 female progeny were tested for recombination in the *Gl–Sb* interval in comparison with daughters of unselected eighteenth generation matings. Twenty-five females were tested in each of the four groups. They were mated with wild-type brothers of the same parentage.

The genotypes of the four tested groups and their mean recombination values are given in Table 1. Both the reciprocal crosses had means below the midpoint between the high and low control levels, but there is a significant difference of 4.1 percent between the reciprocal crosses. The recombination value in the high male × low female cross did not differ significantly from that of the low control. The high control showed a significantly higher recombination value than all other groups.

Females of the reciprocal cross progeny differed only in the origin of the marked third chromosome and the cytoplasm. In the low male × high female cross the marked third chromosome and cytoplasm came from the high line. Conversely, in the high male × low female cross the marked third chromosome and cytoplasm came from the low line. The effects of the origin of the third chromosome and cytoplasm are thus confounded but the significant difference in recombination between reciprocal crosses does implicate one or both factors.

The present results suggest that the difference in recombination value between the high and low lines can be divided into two components. The first, the larger component, is attributable to alleles at one or more loci which are recessive for high recombination and whose location in the genome is unknown. The second,

TABLE 1

Mean recombination values and progeny sizes for Gl Sb/Gl+Sb+ *females from each of four genetic backgrounds*

Genotype	Low ♂ × high ♀ $\frac{H\ H}{L\ L}\ \frac{Gl Sb(H)}{Gl^+Sb^+(L)}\ \frac{H}{L}$	High ♂ × low ♀ $\frac{H\ H}{L\ L}\ \frac{Gl Sb(L)}{Gl^+Sb^+(H)}\ \frac{H}{L}$	Low ♂ × low ♀ $\frac{L\ L}{L\ L}\ \frac{Gl Sb(L)}{Gl^+Sb^+(L)}\ \frac{L}{L}$	High ♂ × high ♀ $\frac{H\ H}{H\ H}\ \frac{Gl Sb(H)}{Gl^+Sb^+(H)}\ \frac{H}{H}$
Recombination percent	16.2	12.1	10.6	25.4
Number of progeny per female	120.5	111.8	99.8	94.5

attributable to either the cytoplasm or to non-recessive, third-chromosome alleles, or both. is the smaller but yet also significant component.

Recombination tests with other markers: After thirteen generations of selection. when differences in recombination between the two lines were well established. recombination values in other third-chromosome intervals were tested. These tests were initiated to find out whether subregions of the *Gl–Sb* interval had been affected equally by selection with respect to recombination. Recombination values in adjacent chromosome regions were also examined to determine whether they had been affected by selection in the *Gl–Sb* region.

A marker stock, homozygous for the *res* chromosome, was obtained from J. W. GOWEN. This chromosome carries seven recessive markers: roughoid, *ru* (3-0.0); hairy, *h* (3-26.5); thread, *th* (3-43.2); scarlet, *st* (3-44.0); pink-peach, p^p (3-48.0); curled, *cu* (3-50.0); stripe, *sr* (3-62.0); ebony-sooty, e^s (3-70.7). Multiple-marked third chromosomes were introduced into samples of the two selection lines by a balanced inversion technique. Females were produced, heterozygous for a multiple-marked and an unmarked "selected" third chromosome. All other chromosomes were from the selection lines. Estimates of recombination values in six intervals were obtained from these females mated with males of the marker stock. Recombination in the *ru–h* interval was not recorded for technical reasons. Data for the *th–st* and *st–*p^p intervals were pooled due to the small size of the intervals.

For comparison, females heterozygous for the *res* marker stock and an unselected wild-type stock, Composite, were also tested. As the base population itself had been discontinued, the Composite stock was considered to be the best available control. This stock originated from the random mating of six wild stocks, including four of the five which contributed to the base population. It had been maintained in a population cage for over a year.

Estimates of recombination values in the five-intervals are presented in the third to fifth columns of Table 2. Recombination value in the high line was, with one exception, significantly higher in every interval than in the low line ($P < .05$). The exception was that male progeny did not show any significant difference in the p^p–*cu* region. Significant sex differences in the percentage of recombinants recovered were exhibited in progeny of high-derived females in the two regions *th–*p^p and p^p–*cu*. The reason for these differences is not known. The sex ratio was 50.51 percent males in the high-derived line, 46.27 percent males in the low-derived line and 52.03 percent males in the Composite line. No sex differences were found in the original selection lines carrying *Gl* and *Sb* marked chromosomes.

In the Composite background recombination values were intermediate between those of high and low lines in every interval except *sr–*e^s. A comparison of recombination value between the females carrying the *res* chromosome and those carrying *Gl* and *Sb* in the original selection lines is of interest. From the *res* data already presented, recombination value in the *th–sr* interval (18.8cM) is calculated to provide the most appropriate comparison, both in location and length with the *Gl–Sb* interval (16.8cM). Mean percent recombination between

TABLE 2

Mean percent recombination in 5 intervals of the res *chromosome against seven genetic backgrounds*

Interval	Standard map distance in cm*	High selection line		Low selection line	Composite	Ames	Oregon R-C	Princeton	Nettlebed
h – th	16.7	20.5		16.8	18.8	21.1	16.5	16.8	20.0
		♂♂	♀♀						
th – p^p	4.8	5.0	6.6	2.5	3.0	5.1	1.1	2.2	2.9
		♂♂	♀♀						
p^p – cu	2.0	1.2	2.8	0.9	1.1	1.7	1.0	0.5	0.9
cu – sr	12.0	14.1		8.9	11.7	13.1	8.6	9.5	9.0
sr – e^s	8.7	10.9		9.0	8.6	10.5	6.2	8.4	9.2
		♂♂	♀♀						
Total	44.2	51.6	55.1	38.1	43.2	52.1	33.4	37.4	42.0
		♂♂	♀♀						
n†		1835	1798	2516	963	1378	614	644	765

* Lindsley and Grell (1968).
†n = the number of progeny counted in each line.

th and *sr* was 20.2 in high-derived male progeny, 23.6 in high-derived female progeny, and 12.2 in low-derived male and female progeny. In comparison mean percent recombination between *Gl* and *Sb* for generations 16 through 19 in the selection experiment was 30.3 in the high line and 12.3 in the low line, there being no sex differences. Substitution of a *res* chromosome for a *Gl–Sb* chromosome therefore appears to have reduced recombination by 6% to 8% in the high selection line but had no effect in the low selection line.

The substitution of a *res* chromosome resulted in relaxation of selection for either one or two generations. A comparison of recombination frequencies in the control groups in Table 1 with those of the high and low lines in generation 19 provides an estimate of the magnitude of the effect of relaxed selection for one generation. In the high line, progeny of selected and unselected parents had a mean recombination value of 30.5 and 25.4 respectively. Comparable figures for the low line were 13.6 from selected parents and 10.6 from unselected parents. These data suggest that relaxed selection was an important factor in reducing recombination value when a *res* chromosome was substituted. However, the presence of recombination modifiers on unmarked third chromosomes may also have contributed to the lower recombination value in high-derived females, carrying a *res* chromosome, compared with their contemporaries in the high selection line itself.

Recombination values in the original wild-type stocks: The significant response to selection for increased recombination mentioned above suggests that the base population contained a considerable amount of genetic variation. Since this base population originated from the five wild-type stocks mentioned earlier, it is of interest to see how each of these stocks contributed to the genetic variation of the base population. To answer this question, the recombination values of four of the

five stocks, i.e., Ames, Oregon R–C, Princeton, and Nettlebed, were examined. The Cranston stock was not tested. For this test, the *res* marker stock was used, examining the recombination values in the six chromosome regions as before. The results obtained are given in the last four columns of Table 2.

Variation in recombination among unselected lines was unexpectedly large. Deviations from the overall mean were generally consistent in direction but not in magnitude for all intervals in any one line. There was at least as great an overall difference between the highest and lowest unselected lines, Ames and Oregon R-C, respectively, as between the two selected lines. Ames did not differ significantly from the high selected line in any interval. Differences among the low selected line, Nettlebed, Princeton and Oregon R-C were small in individual intervals but accumulated to become significant over all regions combined in some cases. In general the low-derived line lay close to the mean of these three unselected lines but was higher than Oregon R-C in most intervals. Recombination value in the Composite was near to the overall mean of the other six lines in every interval.

In order to test the repeatability of recombination values in the wild stocks Ames, Oregon R-C, Princeton and Nettlebed, another experiment using the markers *Gl* and *Sb* was carried out 18 months after the *res* tests. Chromosomes marked with *Gl* and *Sb* in coupling were backcrossed into these four wild stocks for seven generations. Females were then tested in two successive generations, the 8th and the 9th. In the 9th generation the females were inadvertently subjected to overcrowding in all preadult stages.

In Table 3 recombination data from these later tests are compared with those from the *res* experiment. As before, recombination in the *th–sr* interval of the *res* chromosome is presented in comparison with the *Gl–Sb* interval. The variation among lines in the *th–sr* interval is similar to that in the later tests. The rank order is almost identical in all three tests. Within any one line the recombination value is remarkably uniform when tested with different markers eighteen months apart. These results indicate that recombination values of the wild-type laboratory stocks tested are highly repeatable. It is therefore likely that these differences existed at the time of the formation of the base population.

TABLE 3

Mean percentage recombination between two sets of markers with four genetic backgrounds in the centromere region of chromosome three

Interval	Ames	Oregon R-C	Princeton	Nettlebed	Standard map length in cM
th – sr	20.5	10.8	12.1	12.8	18.8
	(1378)*	(614)	(644)	(765)	
Gl – Sb	17.8	7.4	12.0	15.4	17.2
(8th generation)	(1257)	(986)	(1569)	(1469)	
Gl – Sb	18.2	8.4	12.2	10.3	17.2
(9th generation)	(564)	(618)	(672)	(1148)	

* Figures in parentheses denote number of progeny counted.

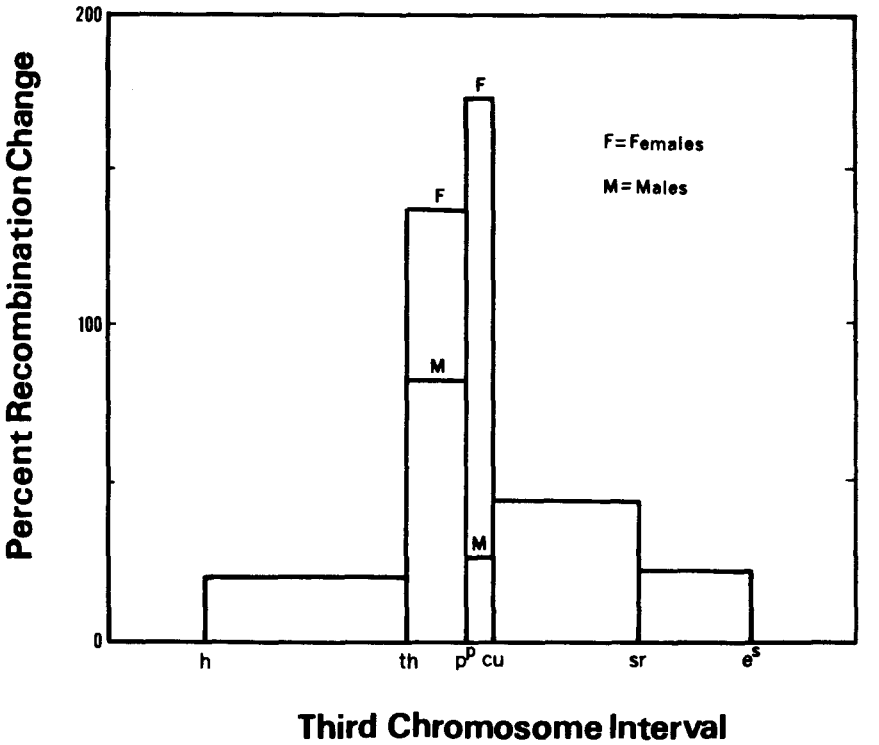

FIGURE 3.—Mean difference in percent recombination between the high and low selection lines expressed as a percentage of the Composite value.

Change in recombination value in relation to the proximity of the centromere: In both selected and unselected lines recombination values in intervals proximal to the centromere tended to be more variable than in distal regions. This is illustrated in Figure 3 for the selected lines. This figure shows the differences in recombination values between the high and low lines expressed as a percentage of recombination in the Composite stock. Computations were made from the data in columns 3, 4 and 5 of Table 2. The pattern of recombination differences between the highest and lowest unselected lines is very similar. These results appear to be another demonstration of the "centromere effect" which has been reported many times previously but not explained.

TABLE 4

Coincidence estimates for adjacent intervals of the third chromosome in lines heterozygous for the res *marker chromosome*

Region	High-derived	Low-derived	Pooled unselected
Left arm	0.49 ± .10	0.29 ± .16	0.38 ± .11
Right arm	0.39 ± .07	0.28 ± .11	0.36 ± .08
Across centromere	0.96 ± .04	1.11 ± .09	0.95 ± .05

Coincidence coefficients were computed from recombination data in the following pairs of intervals of the *res* chromosome: left arm, *h–th* and *th–p*^{p}; right arm, *p*^{p}*–sr* and *sr–e*^{s}; across the centromere, *h–p*^{p} and *p*^{p}*–e*^{s}. In the latter case the centromere and *p*^{p} are regarded as having the same location. Coincidence coefficients in the high-derived line, low-derived line and in the (pooled) unselected, wild strains are presented in Table 4. In all lines a fairly high order of positive interference is observed between adjacent intervals on the same arm of the chromosome. The low-derived line shows a smaller coincidence coefficient than the high-derived line, but the difference is not statistically significant. There is negligible interference between adjacent intervals across the centromere in all three groups. These results are in agreement with those from *Neurospora crassa*, yeast and *Chlamydomonas* as described by STRICKBERGER (1968).

DISCUSSION

In contrast to the case of reduced recombination there is only one known cytological mechanism which may strongly increase recombination frequency, i.e., the presence of either a heterozygous or homozygous inversion on a non-homologous chromosome. It was not possible to complete the cytological and genetic tests necessary to rule out the presence of an inversion. However, preliminary tests of recombination in the first and second chromosome provide no evidence of gross structural abnormalities. With these reservations, modifying genes rather than a cytological mechanism are assumed to be the recombination changing agent in the remainder of this discussion.

Considering the response to selection for increased recombination, the amount of crossing over in the *Gl–Sb* interval is likely to be increasingly underestimated as higher levels are reached because of multiple crossovers. Theoretically, the expected number of crossovers in a chromosome segment can be estimated by HALDANE's (1919) mapping function

$$x = (\log_e (1-2y))/2$$

where y is the recombination fraction. For example, the expected numbers of crossovers corresponding to the recombination fractions 14, 20, and 30 percent are 16.4, 25.5, and 45.8 percent respectively. HALDANE's formula depends on the assumption of no interference. Evidence from the *res* tests, however, suggests that interference across the centromere is small or nonexistent. These considerations are relevant to an assessment of the true magnitude of the difference in crossing over achieved by two-way selection.

The fact that maximum differences in recombination occurred in intervals close to the centromere in both selected and unselected lines suggests that the *Gl–Sb* interval chosen for selection was a good fit with the segment having the maximum potential variation. This must have had an important influence on the efficiency of selection. In the past it seems to have been often a matter of chance whether the interval chosen in a selection experiment was a good or bad fit with the natural units of recombination control. In addition the success of selection is dependent on the relevant modifying loci not being fixed in the population in

question. It would seem that a better knowledge of the nature of recombination-controlling mechanisms and more studies of the frequency of unfixed modifying loci in natural populations of the type made by SANDLER *et al.* (1968) will be essential to improve the predictability of selection experiments.

Previously reported selection experiments for increased and decreased recombination frequency in Drosophila have yielded widely differing results. DETLESON and ROBERTS (1921), MURKHERJEE (1961), MOYER (1964) and CHINNICI (1971) were successful in selecting for decreased recombination. DETLEFSON and ROBERTS reduced crossing over between two sex-linked mutants from 30 to 2 percent in one line and to 8 percent in another. They concluded that sex-chromosome crossing over was "controlled by multiple incompletely dominant factors." From an original population with a mean recombination of 11.39 percent between *px* and *pc* in males of *D. ananassae*, MUKHERJEE selected one strain with very low crossing over. MOYER achieved a significant separation in frequency of recombination by two-way selection but all the response was in the direction of reduced recombination. He used two recessives, marking a paracentric interval of chromosome II. CHINNICI reduced recombination between the sex-linked genes *sc* and *cv* from 15.4 to 8.5%.

In contrast, ACTON (1961) and GOWEN (1919) were not able to reduce recombination by selection. ACTON used recessive markers lying approximately ten units apart on the same side of the centromere of chromosome II. GOWEN employed six recessive markers spread throughout the length of chromosome III.

Successful selection for increased recombination in Drosophila has been less frequent than for decreased recombination. GOWEN (1919), DETLEFSON and ROBERTS (1921), MUKHERJEE (1961) and MOYER (1964) were all unable to increase recombination in their two-way selection experiments. However, PARSONS (1958) did obtain an increase in recombination from 4.93 to 7.38 between black and purple with a concurrent reduction from 8.77 to 6.94 between purple and vestigial. These two intervals lie on either side of, but close to, the centromere. There was little overall change in the segment between black and vestigial. CHINNICI (1971) observed an increase from 15.4 to 22.1% in a distal segment of the *X* chromosome. Also both DEWEES (1970), in Tribolium, and ALLARD (1963), in the lima bean, were able to increase recombination in two-way selection experiments. In general, therefore, selection for decreased recombination has been more successful than that for increased recombination. But, due to the lack of knowledge of the genetic mechanisms involved, it has not been possible to predict in advance either the direction or magnitude of response to selection for recombination change.

An important feature of the present experiment is the asymmetry of response to selection. The increase in recombination in the high line is of a magnitude not previously reported in a selection experiment in Drosophila, but there was no detectable response to selection in the opposite direction. The results establish that genetic variance with respect to recombination was present in the base population. The fact that response was in one direction only is worthy of some discussion.

Explanation of the asymmetry of response cannot be given in terms of factors acting through the selection differential; the asymmetry is still very marked when the response is plotted against the cumulated selection differential. Directional dominance and directional gene frequencies are probably involved to some extent. Evidence for the recessiveness of at least some of the alleles increasing recombination is provided by the reciprocal crosses between the two selection lines. Also, the pattern of response to selection in the high line is consistent with that expected of a recessive allele or alleles at a fairly low original frequency. However, FALCONER (1960) claims that both directional dominance and directional gene frequencies would not be expected to exert an effect in the early generations of selection.

The small population size suggests that genetic random drift may have been an important factor in the present results. Both the selection lines were originally descended from a maximum of five single pairs of flies in generation 0. The random drift explanation is also supported by the following informal observation: the production of Oregon R-C females was significantly below the mean of all wild lines tested. (As indicated earlier this line had the lowest recombination of all lines, both selected and unselected.) The frequency of "low recombination genes" from this source was therefore probably disproportionately low in the base population.

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Note added in proof: Examinations of salivary gland chromosomes were kindly made by Mr. OSAMU YAMAGUCHI at North Carolina State University. These revealed no evidence of structural abnormalities in the high selection line. A unique heterozygous inversion involving the distal half of the left arm of chromosome III was found in the low selection line. It is uncertain whether this inversion contributed to, or was incidental to the stability of recombination in this line. In any case, the conclusion was confirmed that the high selection line increase in recombination was not due to structural heterozygosity.

LITERATURE CITED

ACTON, A. B., 1961 An unsuccesful attempt to reduce recombination by selection. Am. Naturalist **95**: 119–120.

ALLARD, R. W., 1963 Evidence for genetic restriction of recombination in the lima bean. Genetics **48**: 1389–1395.

BOMER, W. F. and P. A. PARSONS, 1962 Linkage and recombination in evolution. Advan. Genet. **11**: 1–100.

CATCHESIDE, D. G., 1968 The control of genetic recombination in *Neurospora crassa*. pp. 216–226. In: *Replication and Recombination of Genetic Material*. Edited by W. J. PEACOCK and R. D. BROCK. Australian Acad. Sci., Canberra.

CHINNICI, J. P., 1971 Modification of recombination frequency in Drosophila. I. Selection for increased and decreased crossing over. Genetics **69**: 71–83.

DETLEFSON, J. A. and E. ROBERTS, 1921 Studies on crossing over. I. The effect of selection on crossover values. J. Exptl. Zool. **32:** 333–354.

DEWEES, A. A., 1970 (Abstract) Two-way selection for recombination rates in *Tribolium castaneum*. Genetics **64:** s16–s17.

FALCONER, D. S., 1960 *Introduction to Quantitative Genetics*. Ronald Press, New York.

FISHER, R. A., 1930 *The Genetical Theory of Natural Selection*. Clarendon Press, Oxford.

GOWEN, J. W., 1919 A biometrical study of crossing over. On the mechanism of crossing over in the third chromosome of *Drosophila melanogaster*. Genetics **4:** 205–250.

GOWEN, M. S. and J. W. GOWEN, 1922 Complete linkage in *Drosophila melanogaster*. Am. Naturalist **56:** 286–288.

HALDANE, J. B. S., 1919 The combination of linkage values, and the calculation of distance between the loci of linked factors. J. Genet. **8:** 299–309.

KIDWELL, M. G., 1972 Genetic change of recombination value in *Drosophila melanogaster*. II. Simulated natural selection. Genetics **70:** 433–443.

KIMURA, M., 1956 A model of genetic system which leads to closer linkage by natural selection. Evolution **10:** 278–287.

LINDSLEY, D. L. and E. H. GRELL, 1968 *Genetic Variations of Drosophila melanogaster*. Carnegie Inst. Wash. Publ. 627.

LUCCHESI, J. C. and D. T. SUSUKI, 1968 The interchromosomal control of recombination. Ann. Rev. Genet. **2:** 53–86.

MOYER, S. E., 1964 Selection for modification of recombination frequency of linked genes. Ph.D. Thesis, University of Minnesota.

MUKHERJEE, A. S., 1961 Effect of selection on crossing over in the males of *Drosophila ananassae*. Am. Naturalist **95:** 57–59.

NEI, M., 1967 Modification of linkage intensity by natural selection. Genetics **57:** 625–641.
——, 1968 Evolutionary change of linkage intensity. Nature **218:** 1160–1161.

NEI, M., and Y. IMAIZUMI, 1968 Efficiency of selection for increased or decreased recombination. Am. Naturalist **102:** 90–93.

PARSONS, P. A., 1958 Selection for increased recombination in *Drosophila melanogaster*. Am. Naturalist **92:** 255–256.

ROBERTSON, A., 1961 Inbreeding in artificial selection programs. Genet. Res. **2:** 189–194.

SANDLER, L., D. L. LINDSLEY, B. NICOLETTI and G. TRIPPA, 1968 Mutants affecting meiosis in natural populations of *Drosophila melanogaster*. Genetics **60:** 525–558.

SCHAAP, T. and G. SIMCHEN, 1971 Genetic control of recombination affecting mating factors in a population of Schizophyllum, and its relation to inbreeding. Genetics **68:** 67–75.

SCHULTZ, J. and H. REDFIELD, 1951 Interchromosomal effects on crossing over in Drosophila. Cold Spring Harbor Symp. Quant. Biol. **16:** 175–197.

STRICKBERGER, M. W., 1968 *Genetics*. The Macmillan Co., New York.