INTERCHROMOSOMAL GENOTYPIC INTERACTIONS IN DROSOPHILA. II. AN ANALYSIS OF VIABILITY CHARACTERS[1,2]

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THIS is the second of a series of papers concerned with interchromosomal genotypic interactions, as measured by the interaction between the X chromosomes and the autosomal sets of three highly inbred lines. It has been previously demonstrated (KELLER and MITCHELL 1962) that there are interactions between products of genes on nonhomologous chromosomes, of *Drosophila melanogaster*, for the determination of various morphological characters; the present analysis was initiated to find out whether such interactions also occur between the gene products determining characters relating to viability.

REED (1941) studied interactions among the autosomes of *D. melanogaster*, in relation to viability and developmental period, using autosomal markers. He found positive additive effects in the interaction estimates for the second chromosome, and negative additive effects from interactions of the third and fourth chromosomes. When a "positive" or "negative" autosome was added to a given genotype it caused a change in viability in that respective direction.

It has generally been observed that high levels of heterozygosity are associated with a high reproductivity, whereas inbreeding tends to decrease viability in naturally outbred populations. Heterotic effects have been observed for the characters survival, fecundity, and fertility. Correlations have also been observed among some of the traits. BONNIER and JONSSON (1957) and BONNIER, JONSSON, and RAMEL (1959) found that low survival was associated with relatively high viability, and also that fast developmental rate was positively correlated with a high level of viability and autosomal heterozygosity. A negative correlation was found between developmental rate and egg productivity (ROBERTSON 1957) and developmental rate and female fertility (HIRAIZUMI 1960). An extensive discussion of the quantitative genetic control of viability is given by KELLER 1961.

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The literature pertinent to the theoretical aspects of this paper has been reviewed comprehensively (KELLER 1959; KELLER and MITCHELL 1962).

METHODS AND PROCEDURES

Description of the populations: The experimental methods and genetic stocks are identical to those described previously (KELLER and MITCHELL 1962). The viability data were obtained simultaneously with the morphological data. The types of populations compared in this experiment were (1) the parental *Inbreds* (I); (2) the *Transfers* (T). These consisted of an "isogenic" autosomal set from one inbred line combined with "isogenic" X chromosomes from another inbred line; (3) the X chromosomal *Heterozygotes* (H). These were the progeny of a backcross of a transfer-type female to the original inbred autosomal donor (X chromosome heterozygotes exist only in the female); (4) the *Recombinants* (R). These were the result of one generation of recombination in the X chromosome heterozygotes, with a subsequent backcross to the inbred line from which the autosomes were derived. Table 1 presents diagrammatically the various genotypes which were obtained for this experiment.

Detection of interactions: The mean value of a character in the inbred population can be partitioned into additive components (A) consisting of the X chromosome (x_ix_i) and autosomes (a_ia_i) respectively, and a nonadditive component resulting from the interaction between the two ($E\ x_ix_i,a_ia_i$). An Inbred can be represented by; $\bar{I}\ x_ix_i,a_ia_i = A\ x_ix_i,a_ia_i + E\ x_ix_i,a_ia_i$.

In a similar manner the mean for a Transfer population can be partitioned as follows: $\bar{T}\ x_jx_j,a_ia_i = A\ x_jx_j,a_ia_i + E\ x_jx_j,a_ia_i$. Estimates of these components can be obtained, being expressed as deviations from the grand mean, for all Inbred and Transfer populations. Deviations from the additive system will provide a measure of interaction between the two "isogenic" portions of the gneotype (the X chromosomes and the autosomes) plus some replicate effects. Only comparisons between the values of the Inbreds and Transfers were used to measure such deviations and are obtained here in the same manner as that described previously (KELLER and MITCHELL 1962).

The means of the characters in the Heterozygote populations will also include X chromosome interallelic interactions which would be part of the X chromosome additive component, as well as a component representing interaction effects between the heterozygous X chromosomes and the isogenic autosomes; i.e. $\bar{H}\ x_ix_j,a_ia_i = A\ x_ix_j,a_ia_i + E\ x_ix_j,a_ia_i$. A specific class of combinations (x_ix_j,a_ea_e or x_ix_i,a_ea_e) was not obtained from the present experiment. For this reason the effects of heterozygosity must be judged from overall comparisons of Inbreds, Transfer, and Heterozygotes, under the assumption that the values of interchromosomal interactions in the Heterozygotes are not excessively greater than those in the Transfers.

The Recombinant populations differ from the other three groups in that the genic linkages within a single recombined X chromosome have been rearranged. The average level of genic heterozygosity in these populations is expected to be one half that of the Heterozygotes. The interchromosomal interaction effect will vary accordingly, as will that component resulting from the changes in linkages. Therefore, the effects of disrupting an internal balance within the X chromosomes must again be judged from comparisons of the means of *all* of the populations. These X chromosomal disruptive effects should be more apparent in the Recombinant males where these effects are not confounded by genic heterozygosity.

The control for the effect of the breeding procedure is the comparison of the Inbred males with the males of the Heterozygote class, since these two groups should be genotypically identical; the only difference is that the Heterozygote males were the product of the breeding scheme, the Inbreds were not. Differences between these two groups would be the result of uncontrolled or spurious events such as fourth chromosome effects, failure of complete crossover suppression in the inversion systems, etc.

Description of characters: Data were obtained on five components of viability: (1) egg to pupal survival, (2) adult developmental period from egg to eclosion, (3) the percentage of females capable of laying eggs (percent fecund), (4) number of eggs laid, and (5) the percentage

TABLE 1

The genotypes of the inbred, the X-chromosome Transfer, the X-chromosome Heterozygote, and the X-chromosome Recombinant lines

Inbred lines				Transfer lines			
Females		Males		Females		Males	
X	II, III	X	II, III	X	II, III	X	II, III
X3/X3	A3/A3	X3/Y3	A3/A3	X6/X6	A3/A3	X6/Y3	A3/A3
				X17/X17	A3/A3	X17/Y3	A3/A3
X6/X6	A6/A6	X6/Y6	A6/A6	X3/X3	A6/A6	X3/Y6	A6/A6
				X17/X17	A6/A6	X17/Y6	A6/A6
X17/X17	A17/A17	X17/Y17	A17/A17	X3/X3	A17/A17	X3/Y17	A17/A17
				X6/X6	A17/A17	X6/Y17	A17/A17

Heterozygote lines				Recombinant lines			
Females		Males		Females		Males	
X	II, III	X	II, III	X	II, III	X	II, III
X6/X3	A3/A3	X3/Y3	A3/A3	X3–6/X3	A3/A3	X3–6/Y3	A3/A3
X17/X3	A3/A3	X3/Y3	A3/A3	X17–3/X3	A3/A3	X17–3/Y3	A3/A3
X3/X6	A6/A6	X6/Y6	A6/A6	X3–6/X6	A6/A6	X3–6/Y6	A6/A6
X17/X6	A6/A6	X6/Y6	A6/A6	X17–6/X6	A6/A6	X17–6/Y6	A6/A6
X3/X17	A17/A17	X17/Y17	A17/A17	X3–17/X17	A17/A17	X3–17/Y17	A17/A17
X6/X17	A17/A17	X17/Y17	A17/A17	X6–17/X17	A17/A17	X6–17/Y17	A17/A17

The first figures in each case represent the X chromosomes of the female, and the X and Y chromosomes of the males; the last figures indicate the derivation of the second and third chromosomes. The 3, 6, and 17 designate the inbred donor. Each recombinant line contains a mixture of the crossover and noncrossover chromosomes.

fertile matings. These data were obtained from the generation (in the breeding procedure) producing flies of the desired genotype. Survival and developmental periods were characters pertaining to pre-adult stages, while fecundity, fertility, and the number of eggs laid were adult characters. The overall procedure by which these various data were obtained involved the following steps:

1. Three to four pairs of young flies, of the desired genotype, were placed in a culture vial seeded with live yeast and allowed to deposit eggs for three days. On the fourth day, the flies were transferred to a nonyeasted fresh vial, placed at 26°C, and the original vial was discarded. Every 4 hours thereafter the eggs deposited in the new vial were counted, the vial seeded with live

yeast, and the adults transferred to fresh vial with both vials being returned to a 26°C incubator. 2. When pupation was complete, in a particular vial, the number of pupae present was counted and the vial was returned to a 26°C incubator. 3. The vials were checked periodically for eclosion. After emergence of the first adult, in a particular vial, the vial was checked every 4 hours (except for the early morning count) for the number of flies, their sex, and the interval of time elapsed. Some adults were chosen at random and placed in fresh vials and stored at 18°C for the morphological measurements. Other adults were randomly selected for use in the following step. 4. A series of pairs of adults, of identical genotypes, were sib-mated, upon eclosion, in fresh vials and put at 26°C. Every 24 hours thereafter the pair was transferred to a fresh vial. The eggs which were deposited during the 24-hour interval were then counted. Those vials containing eggs were placed at 26°C and were subsequently checked for fertile matings (presence of adult progeny).

From the above data the percent survival was determined as the ratio (pupae/eggs) × 100. Developmental periods were determined for the individual flies by calculating the elapsed period of time from the midpoint of the egg laying period to the midpoint of the eclosion period.

There were some vials which did not contain eggs from Step 4. These matings were scored as "nonfecund" matings. Other vials contained eggs but no larvae; these matings were scored as sterile pair-matings. Finally, the egg productivity was determined from the daily egg counts per pair per 24-hour period. Only data from vials containing at least five and less than 40 eggs were used in the determination of the survival estimates, since survival has been found to be density dependent beyond these limits (MITCHELL 1958).

RESULTS AND DISCUSSION

In Table 2 are presented the various autosomal (a), X chromosomal (x), and interaction (e) estimates relating to the equation $Y = \overline{Y} + x_i + a_j + e_{ij}$ (where $\overline{Y}$ = the grand mean for a character and Y = the value of a given character of the ith individual) as previously described by KELLER and MITCHELL (1962). In this analysis the interaction estimates (e's) are confounded with replicate effects, but are considered as estimates of the interaction of the (a) and (x) components (see DISCUSSION). This partitioning of variation was done with only the Inbred and Transfer groups. Under this model, the analysis indicated that a large percentage of the genetically controlled variation (with respect to the X chromosome) was due to interaction, especially for the characters percent fecundity, developmental period, and egg productivity. Only the Inbred and Transfer groups are represented here, and dominance effects do not exist. In the characters survival, fer-

TABLE 2

Partitioning of variation

Character	Variation due to			Percent variation due to e's
	a's	x's	e's	
Percent survival	125.5	27.1	28.4	16
Female developmental rate	31.8	11.3	49.8	54
Male developmental rate	9.1	2.3	46.3	80
Percent fecundity	31.1	8.3	254.7	87
Egg production	0.2	1.6	1.2	41
Percent fertility	16.1	5.6	6.6	23
Reproductive capacity	10.3	0.5	4.4	29

tility, and reproductive capacity, there are also contributions of the nonadditive component (*e* values), but to a lesser degree. It is apparent that there are different contributions of the autosomal and X chromosomal sets for each character. However, since other factors of genetic control are not included (e.g., dominance, or heterozygosity) Table 2 is used only to demonstrate the existence of "simple" interchromosomal effects not confounded with other genetic mechanisms, but possibly with some replicate interaction.

The viability components: (1) *Percentage of females capable of laying eggs (Fecundity).* The percentage of fecund matings was determined by taking the number of pair matings where no eggs were deposited throughout the egg counting period (144 hours), subtracting this number from the total number of crosses attempted, and multiplying by 100. Even a virgin female will usually deposit eggs after 144 hours (MAYNARD SMITH 1955). Therefore, the absence of oviposition can be taken as an estimate of an inability to produce or to deposit eggs. The percentage of fecund females is given in Table 3. These fecundity estimates exhibit a range of 56.9 percent, ranging from 37.7 percent for inbred Line 6 to 94.6 percent for recombinant Line X3-17/X17 A17/A17. Some of the Heterozygote populations might have had a higher percentage fecundity, but the small number of flies for the last two Heterozygote populations, Table 3, and for X6-17/X6 A6 A6 of the Recombinants leaves some doubt as to the reliability of these estimates. The high frequency of nonfecund females in all of the populations must be related to the original source of genetic material, i.e., inbred Line 6. The low fecundity of this line is based on the data from only *nine* pairs, which is a very low number for the estimation of this character. However, it is felt that it is a fairly accurate estimate, since the combinations of either X6 or A6 are usually below the grand average.

The highest fecundity values were obtained in the Recombinant and Heterozygote populations, with the Transfers being lower than the former two groups but slightly superior to the Inbreds (Table 4). Therefore, the effect of the Transfer of an "isogenic" X chromosome to an "isogenic" autosomal set was to increase the number of fecund females. Heterozygosity had a much greater beneficial effect (Table 4). The genetic nature of this character has not been further analyzed. However, it is clear that the Transfers resulted in an increase in the number of females capable of producing and/or depositing eggs. In general, the different X chromosomes showed no consistent pattern in the modification of fecundity; however, when the Line 3 autosomes were in combination with Line 6 X chromosomes there were consistent *increases* in the percentage of fecund females. When Line 17 X chromosomes were in combination with Line 3 autosomes there was a consistent *decrease* in the number of fecund females. The autosomes from Line 6 always resulted in a decrease in the percent fecund females when the X chromosome was heterozygous (Table 4); however, there was a definite increase in the overall percentage of fecund females for a given group only when epistatic (interchromosomal) effects were present. Line 17 autosomes were associated with increased values of fecundity with the exception of the X3/X3 A17/A17 combination, which was subnormal.

TABLE 3

Percent pair fertility and percent fecundity (females capable of laying eggs) and reproductive capacity over all populations

Population	Percent fertility	Percent fecundity	Reproductive capacity of progeny pairs (per pair/per 24 hr)	Total number of pairs
Inbreds				
$\frac{X3}{X3}$ $\frac{A3}{A3}$	91.67	66.20	5.95	31
$\frac{X6}{X6}$ $\frac{A6}{A6}$	83.61	37.71	1.51	9
$\frac{X17}{X17}$ $\frac{A17}{A17}$	96.97	80.81	10.46	17
Transfers				
$\frac{X6}{X6}$ $\frac{A3}{A3}$	74.89	73.62	5.32	34
$\frac{X17}{X17}$ $\frac{A3}{A3}$	72.22	43.33	2.00	13
$\frac{X3}{X3}$ $\frac{A6}{A6}$	78.69	75.22	3.85	33
$\frac{X17}{X17}$ $\frac{A6}{A6}$	90.26	83.59	2.36	28
$\frac{X3}{X3}$ $\frac{A17}{A17}$	75.00	55.77	4.69	8
$\frac{X6}{X6}$ $\frac{A17}{A17}$	92.42	79.29	11.30	28
Recombinants				
$\frac{X3\text{–}6}{X3}$ $\frac{A3}{A3}$	95.33	90.65	18.47	18
$\frac{X3\text{–}17}{X3}$ $\frac{A3}{A3}$	86.32	67.92	4.40	35
$\frac{X3\text{–}6}{X6}$ $\frac{A6}{A6}$	87.07	67.24	2.78	19
$\frac{X6\text{–}17}{X6}$ $\frac{A6}{A6}$	(100.00)	(69.56)	(6.57)	(4)
$\frac{X3\text{–}17}{X17}$ $\frac{A17}{A17}$	99.09	94.55	20.96	16
$\frac{X6\text{–}17}{X17}$ $\frac{A17}{A17}$	89.58	89.58	16.40	7
Heterozygotes				
$\frac{X3}{X6}$ $\frac{A3}{A3}$	90.93	81.86	9.19	58
$\frac{X3}{X17}$ $\frac{A3}{A3}$	91.66	78.79	22.69	38
$\frac{X3}{X6}$ $\frac{A6}{A6}$	86.67	77.11	16.54	12

TABLE 3—Continued

Population	Percent fertility	Percent fecundity	Reproductive capacity of progeny pairs (per pair/per 24 hr)	Total number of pairs
$\frac{X17}{X6}$ $\frac{A6}{A6}$	62.41	42.11	2.62	19
$\frac{X3}{X17}$ $\frac{A17}{A17}$	(100.00)	(100.00)	(44.72)	(3)
$\frac{X6}{X17}$ $\frac{A17}{A17}$	(100.00)	(100.00)	(32.03)	(2)

(2) *Egg productivity.* Egg productivity was estimated by the number of eggs deposited per fecund pair per 24-hour period between the 72nd and 144th hour after mating. During this interval a stable maximum in egg laying rate was observed at this temperature. The number of eggs oviposited per unit time is, in part, a function of whether the female has been fertilized, and of the extent of mating activity previous to and during the period of data collection. Since sterile matings will bias the estimate of the rate of egg deposition, the fecund but nonfertile pairs were excluded from the egg productivity analysis (see next section).

No significant differences existed in the egg production of the Inbreds, the Transfers, or the Recombinants. The Heterozygotes were significantly more productive than all of the other groups. It appeared that no *major* interchromosomal effects were present in the modification of egg productivity. However, egg production is frequently subject to modification by the influences of heterozygosity, as was the case in this experiment (Table 4). Line 3 X chromosomes, when in combination with Line 6 or Line 17 autosomes, resulted in the highest degree of heterotic effects. The presence of Line 6 X chromosomes had a negative effect (reducing egg production), with the Line 17 X chromosomes exhibiting intermediate effects. An identical pattern existed in the Transfer group, but to a somewhat lesser degree. The mean response in the Recombinants was reversed since

TABLE 4

Estimates of viability components for the genetic types

Populations	Inbreds	Transfers	Recombinants	Heterozygotes
Females capable of laying eggs (fecundity)	61.57%	68.47%	79.92%	79.97%
$\overline{X}$ 24-hr egg laying rate	10.93	12.72	12.90	21.22
Fertile matings	90.75%	80.58%	92.89%	86.33%
Egg to pupal survival	26.96%	23.63%	35.44%	36.66%
$\overline{X}$ developmental period egg to adult (female)	213.92 hr	201.10 hr	223.43 hr	195.89 hr
$\overline{X}$ developmental period egg to adult (male)	218.82 hr	209.11 hr	230.55 hr	203.55 hr
Reproductive capacity over 144 hr	4.94 pairs	4.97 pairs	10.18 pairs	16.43 pairs

TABLE 5

Means and standard deviations of the mean for survival and egg productivity

Population	Percent larval survival			Daily egg productivity		
	$\bar{X}\%$	$S_{\bar{X}}$	N†	$\bar{X}$	$S_{\bar{X}}$	N†
Inbreds						
$\frac{X3}{X3}$ $\frac{A3}{A3}$	27.85	3.69	24	10.76	1.44	48
$\frac{X6}{X6}$ $\frac{A6}{A6}$	15.75	5.43	3	10.10	2.70	10
$\frac{X17}{X17}$ $\frac{A17}{A17}$	37.29	6.04	17	11.93	1.55	25
Transfers						
$\frac{X6}{X6}$ $\frac{A3}{A3}$	29.48	4.64	22	10.91	1.45	42
$\frac{X17}{X17}$ $\frac{A3}{A3}$	15.88	8.94	6	13.40	4.70	11
$\frac{X3}{X3}$ $\frac{A6}{A6}$	17.90	2.93	24	12.13	0.82	50
$\frac{X17}{X17}$ $\frac{A6}{A6}$	7.50	2.74	17	13.95	1.05	45
$\frac{X3}{X3}$ $\frac{A17}{A17}$	25.33	5.95	7	14.74	4.04	11
$\frac{X6}{X6}$ $\frac{A17}{A17}$	46.04	3.33	35	11.16	1.22	40
Recombinants						
$\frac{X3\text{–}6}{X3}$ $\frac{A3}{A3}$	40.66	4.00	22	17.52	1.93	29
$\frac{X3\text{–}17}{X3}$ $\frac{A3}{A3}$	29.13	3.22	26	8.59	0.83	49
$\frac{X3\text{–}6}{X6}$ $\frac{A6}{A6}$	16.09	3.20	11	9.83	1.33	30
$\frac{X17\text{–}6}{X6}$ $\frac{A6}{A6}$	22.48	4.37	4	14.01	3.01	8
$\frac{X3\text{–}17}{X17}$ $\frac{A17}{A17}$	55.99	4.58	23	13.32	1.29	32
$\frac{X6\text{–}17}{X17}$ $\frac{A17}{A17}$	48.28	8.25	9	14.11	2.30	12
Heterozygotes						
$\frac{X3}{X6}$ $\frac{A3}{A3}$	35.23	3.27	47	11.69	0.96	93
$\frac{X3}{X17}$ $\frac{A3}{A3}$	46.62	3.26	44	22.47	1.86	61

TABLE 5—Continued

Population		Percent larval survival			Daily egg productivity		
		$\bar{X}$%	$S_{\bar{X}}$	N†	$\bar{X}$	$S_{\bar{X}}$	N†
$\frac{X3}{X6}$	$\frac{A6}{A6}$	23.41	2.90	13	35.24	3.24	16
$\frac{X17}{X6}$	$\frac{A6}{A6}$	24.42	3.88	6	13.62	2.30	18
$\frac{X3}{X17}$	$\frac{A17}{A17}$	53.62	4.47	5	27.80	...	5
$\frac{X6}{X17}$	$\frac{A17}{A17}$	64.71	0.00	1*	16.50	...	2

* $\frac{X6}{X17}\ \frac{A17}{A17}$ was excluded from the combined group estimates owing to insufficient data.
† Number of pairs tested.

the presence of portions of the Line 6 X chromosomes produced the highest amount of egg laying capacity. Since the mean egg productivity of the Recombinants did not significantly differ from that of the Inbreds and of the Transfers, this indicates that the effects of genic heterozygosity may be partially counterbalanced by the disruption of the internal balance of the X chromosome. The variability in egg production between the various populations and groups (Table 5) also exhibits a consistent pattern, for the Recombinants were the lowest in both the estimates of the coefficients of variation and variances. This was to some extent surprising, since the genotypic structure of the Recombinants was a type of modified backcross (Table 1). The comparable backcross X chromosome would be designated in our notation X3-6/X3-6 (for the females), whereas in this experiment the modified backcross is X3-6/X6.This possibly indicates that there is an additional mechanism involved, other than heterozygosity, for the reduction of the variability of this character. It might indicate that the relative constancy of the various groups was not only dependent on heterozygosity, but also upon the type of within-chromosome epistatic interrelationships.

(3) *Percent fertile matings:* The percentage fertile matings was expressed as the ratio *total fecund fertile matings: fecund nonfertile matings*, subtracted from unity and multiplied by 100. The percentage fertile matings is presented in Table 4 (row 3) to show the differences between the four types of populations. The Recombinants had the highest percentage fertility, then the Inbreds, Heterozygotes, and Transfers, in that order. Significant differences were found between the Inbreds and Transfers, and between the Recombinants and the Transfers. It appears that there is some type of negative interchromosomal effect in operation within the Transfers. This indicates that an additional interaction component is present, also of an epistatic nature. There was apparently no effect of the disruption of internal balance on this character.

From the data presented in Table 3 it appears that there is no *consistent* overall modification of the percentage fertile matings in the experiment. The percent fecundity data also exhibited the same pattern. However, there appears to be no

general correspondence between fecundity and fertility with the exception of the inbred Line 6, which was low for both characters. The estimates for the percentage fecundity and the percentage fertility in Table 4 were computed omitting the last two Heterozygote populations in Table 3.

The quantitative genetic modification of fertility in these experimental populations appears to be significantly influenced only by interchromosomal interactions. Also, the primary control appears to be in the autosomes.

(4) *Survival:* The estimates of survival were based on the percentage of eggs deposited which developed to pupation. Data were used from vials in which the eggs were deposited between 96 and 144 hours after mating (each sample included two 24-hour intervals). The deposition of sterile eggs was found to be at a minimum during this period. There were differences between the major groups (Inbreds, Transfers, Recombinants, or Heterozygotes) and there were also significant differences among some of the populations within the groups (Tables 3 and 4). For example, there was significant difference between inbred Line 3 and inbred Line 6 and between inbred Line 17 and 6, but not between Lines 6 and 17. Similar individual differences occurred between some populations within the Transfer, Recombinant, and Heterozygote groups (Table 3).

The autosomes of inbred Line 17, in all cases, were associated with the highest survival estimates, the autosomes of Line 6 were consistently low, and the autosomes of Line 3 showed a general pattern of epistatic modification. The consistency of this latter pattern, i.e., low survival when Line 3 autosomes were in combination with Line 17 X chromosomes, was reversed by heterozygosis in the Heterozygote group, which showed a low survival in the X6/X6 A3/A3 group and a high survival in the X17/X17 A3/A3 group. This, perhaps, indicates a similar genic complex for control of fecundity and survival, but not an identical one. There were also several significant differences between the Heterozygotes and the Recombinants and also between the Inbreds and Transfers, Table 4.

In this experiment the major genetic modification of survival, appears to be that of interallelic interaction (dominance). There was no general *major* modification due to interchromosomal interaction (16 percent due to *e*'s, Table 2). A few specific cases of epistatic modifications did occur; for example, a very large depressing effect of the 17 X chromosome was present when it was in combination with Line 6 autosomes. The relative constancy of the various genotypic groups, measured by the variances and coefficients of variation of the survival estimates, indicated that the stability (variability) of the survival of a given genotype is to a large degree controlled by the amount of heterozygosity present.

(5) *Developmental period:* The period of development from egg to adult was used as an estimate of the length of a reproductive generation. The means, variances, and N values (by sex) for the developmental periods are presented in Table 6. Except for the two autosomal Line 17 heterozygote populations, reliable estimates ($N > 56$) were obtained for the means and variances. Statistical interaction (measures of consistency within groups) were obtained by the method of analyses of variance (within a sex) used by KELLER (1959). The same probability levels ($P < .01$) were obtained for the interaction estimates in both female and

male developmental periods. These estimates indicated that the effects of the various X chromosome and autosomal combinations were highly inconsistent, i.e., very specific. Owing to these inconsistencies, only a few statistically significant differences were detected between the major groups (Inbreds, Transfers, Recombinants, and Heterozygotes) and no significant differences were found within most of the groups.

The general pattern of modification showed inbred Line 6 to be associated with a specific phenotype i.e., a shorter developmental period. A shorter developmental period was considered to be a favorable component of "fitness," since it would allow a greater turnover of the population, which would be of an advantageous nature during a period of rapid population expansion. Inbred Line 6 had the best potential of the inbreds in terms of developmental rate. However, there would probably be no adaptive advantage upon outcrossing since all other combinations of Line 6 chromosomes (with one exception) were found to be of a deleterious nature, i.e., they developed more slowly. In general, the Line 3 autosomes were found to be associated with superior developmental rate, again with only one exception. The data from the Heterozygotes indicate that the Line 17 autosomes were *also* inferior to the Line 3 autosomes, but not to the same extent as the Line 6 autosomes. Line 3 X chromosomes in combination with the other two autosomal backgrounds always developed more slowly while Line 3 autosomes in combination with the other two types of X chromosomes were *always* associated with shorter developmental periods. The developmental period of inbred Line 3 was intermediate to the other two inbred lines. Hence, the Line 3 genotype was not very well balanced and the autosomal contributions were, in some epistatic manner, suppressed by the Line 3 X chromosomes. Integration between the genotypic components was dependent upon interchromosomal genic continuity. There was very good agreement between the male and female estimates, with the females always having a shorter average developmental period.

Heterozygosis in the X chromosome was to some extent important in this character, but interchromosomal effects were of even greater significance, since no difference was noted between the Transfers and Heterozygotes. The effect of disruption of the internal balance of the X chromosomes was also apparent. In this character the average period of development of the Inbred males differed from the average developmental time of the Heterozygote males. This difference was an increase of 15.3 hours, and is attributed to the effect of the breeding scheme (Table 4). Therefore, the proper control group for comparative purposes was the Heterozygotes, not the Inbreds. The actual difference between the groups, relative to the Heterozygotes, was minus 27 hours for the effects of disruption of internal balance and minus 5.56 hours for the deleterious *interchromosomal* effects. Also, when using the Heterozygotes as a bases of comparison the average difference between the sexes was 7.66 hours. Correcting for this difference, the adjusted values for the female groups would be 208.78 hours for the Transfers; 231.09 hours for the Recombinants; and 203.55 hours for the Heterozygotes. No *relative* difference existed between these values and those of the males, for the same groups. In regard to the disruption of internal balance by recombination, there

TABLE 6

Means, variances, and number of individuals for periods of developmental in hours

Populations		Females			Males		
		$\bar{X}$	S^2	N	$\bar{X}$	S^2	N
Inbreds							
$\frac{X3}{X3}$	$\frac{A3}{A3}$	212.06	196.92	311	218.23	212.72	267
$\frac{X6}{X6}$	$\frac{A6}{A6}$	205.67	255.39	273	213.17	274.92	227
$\frac{X17}{X17}$	$\frac{A17}{A17}$	224.05	157.84	170	225.08	156.66	128
Transfers							
$\frac{X6}{X6}$	$\frac{A3}{A3}$	192.92	171.29	212	204.67	618.89	167
$\frac{X17}{X17}$	$\frac{A3}{A3}$	198.15	458.97	168	204.45	470.00	109
$\frac{X3}{X3}$	$\frac{A6}{A6}$	206.99	151.37	304	215.53	179.94	290
$\frac{X17}{X17}$	$\frac{A6}{A6}$	198.11	236.05	143	203.52	313.35	159
$\frac{X3}{X3}$	$\frac{A17}{A17}$	204.97	279.23	136	206.30	386.56	156
$\frac{X6}{X6}$	$\frac{A17}{A17}$	206.46	166.16	366	213.52	263.46	387
Recombinants							
$\frac{X3\text{–}6}{X3}$	$\frac{A3}{A3}$	210.05	316.97	264	219.26	288.51	268
$\frac{X3\text{–}17}{X3}$	$\frac{A3}{A3}$	224.44	168.01	87	234.16	404.37	70
$\frac{X6\text{–}3}{X6}$	$\frac{A6}{A6}$	225.05	159.50	129	228.93	215.32	107
$\frac{X6\text{–}17}{X6}$	$\frac{A6}{A6}$	229.27	191.53	122	238.58	313.28	88
$\frac{X17\text{–}3}{X17}$	$\frac{A17}{A17}$	231.25	263.00	222	237.30	232.44	200
$\frac{X17\text{–}6}{X17}$	$\frac{A17}{A17}$	220.53	202.18	205	224.76	153.24	187
Heterozygotes							
$\frac{X3}{X6}$	$\frac{A3}{A3}$	194.36	437.24	165	200.93	274.13	157
$\frac{X17}{X3}$	$\frac{A3}{A3}$	191.42	630.52	164	201.17	177.72	138
$\frac{X3}{X6}$	$\frac{A6}{A6}$	196.17	122.63	97	204.83	236.46	86

TABLE 6—Continued

Populations		Females			Males		
		$\bar{X}$	S^2	N	$\bar{X}$	S^2	N
X17/X6	A6/A6	201.64	222.68	62	207.30	202.14	56
X3/X17	A17*/A17	.....	.....	..	.....	.....	..
X6/X17	A17*/A17	.....	.....	..	.....	.....	..

* No data.

was a considerable effect on this character (plus 27 hours), and this was considered to be of a detrimental nature.

The genetic modification of developmental rate was appreciably affected by the disruption of the internal balance of the X chromosome. A small effect was also noted as a result of the inclusion of heterozygosity in the X chromosome.

Estimates of reproductive capacity: In the previous discussion some of the individual components of "viability" have been examined and various conclusions made as to their genetic modification. An index is presented in Table 3 for overall reproductive capacity per pair per 24-hour period, in terms of progeny pairs. This index of reproductive capacity was calculated using the following formula: (*egg laying rate* × *percent fertility* × *percent fecundity* × *percent survival*)/2, for a 24-hour period. This index was expanded to 144 hours (Table 4) so as to extend over the duration of the experiment, although for some characters data were gathered only over certain intervals this period. These indices are considered as relative estimates of the reproductive capacity of the respective populations. These estimates are probably *maxima*, and it should be noted that there is a possibility of some interrelationships between some of these components (to be reported later).

The reproductive capacity was the lowest in inbred Line 6, and the Line 6 autosomes, on the average, gave lower estimates of reproductive capacity than the other two types. Line 17 autosomes had the highest reproductivity, with Line 3 autosomes intermediate, with relatively inconsistent results.

The Inbred, Transfer, Recombinant, and Heterozygote totals for reproductive capacity (Table 4) indicate that there was no overall genetic effect of the Transfer group over and above the Inbreds. The Recombinants were characterized by a reproductivity about twice that of either the Inbreds or the Transfers, with the Heterozygotes being far superior to all of the other groups. The presence of X chromosomal heterozygosity apparently leads to gross heterosis in all components tested (except perhaps fertility).

GENERAL DISCUSSION

In the components-of-variance analysis, only the Inbreds and Transfers were considered. This might give a biased estimate of the overall variation due to interchromosomal effects (e's) in regard to varying the X chromosomes. It is felt that

these interaction estimates are probably overestimates due to the fact that there was a small amount of replicate error confounded with the interaction estimates. Under the model $Y_{ijk} = \bar{Y} + x_i + a_j + (ax)_{ij} + e_{ijk}$ it would have been possible to estimate the error and interaction components due to the replicate effects. However, this was not possible to do for all of the characters. The replicates were found to be fairly consistent and where they were estimated (in the developmental rates) they produced low experimental error estimates in the analyses of variance.

From the analyses of data collected in this experiment, it was possible to detect major genetic modifications due to interchromosomal interaction in the characters fecundity and developmental rate. The method of analysis of variance was used to demonstrate the presence of interchromosomal interaction in developmental rates (as in the morphological data previously reported). Only survival and fertility were not appreciably affected by interchromosomal effects in this experiment. The analysis of the components of variance indicated that interchromosomal effects were present to some degree in all of the viability traits. Reproductive Capacity, being a compound "character," showed no *mean* effect of interchromosomal interaction, but did exhibit a moderate amount of variation attributable to interactions between "isogenic" X chromosomes from one inbred line and "isogenic" autosomes from different inbred lines.

It is quite apparent, and not unexpected, that the principal determinant of the differences observed among the various populations in terms of overall reproductivity was the presence or absence of genic heterozygosity in the X chromosome. Another factor in operation in the Recombinant class (where one X chromosome was allowed to recombine) was the disruption of the internal balance of the recombined X chromosomes. As was expected, the males were affected to a greater degree in the comparisons between male and female developmental periods. The mean difference observed in egg laying rate between the Recombinant and Heterozygote classes was also attributed to this disruption, as well as the fact that the shorter developmental period of the recombinant class cannot be explained on any other basis.

The analyses of the viability characters provide somewhat different results from the previous analyses of morphological traits, and it seems that the viability traits are modified to a somewhat higher degree by interchromosomal interactions than are the morphological traits. This is entirely consistent with the concept of the integrated, well balanced genotypic structure being most important in characters related to viability. These inbred lines are genetically abnormal, but genotypically well integrated (to the specified experimental conditions). It was again noted that the modification, by genetic manipulation, which restored genic heterozygosity produced better adapted populations.

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SUMMARY

Three highly inbred strains of *Drosophila melanogaster* were derived from a sample obtained from a natural population and were tested to determine the

extent of genic balance and type of genetic control in operation within the lines. The chromosomal interrelationships were determined by estimating the response of five viability traits (larval survival, developmental period, egg productivity, fertility, and fecundity), in four types of populations: the parental inbred lines; (1) and lines having (2) X chromosomes from each inbred line combined with the autosomes of each of the other two lines; (3) X chromosomes combined in a heterozygotic phase with the autosomes from the other two inbreds; and (4) an X chromosome allowed to recombine for one generation with an X chromosome from a recipient inbred line and then combined with an unchanged X chromosome from the recipient line.

The genetic control of fecundity was significantly influenced by interchromosomal effects (but not always in the same direction). Also reintroduction of X chromosomal heterozygosity significantly and consistently increased the proportion of fecund females. Egg productivity was greatly increased by X chromosome heterozygosity, with an observable effect of intrachromosomal rearrangements (disruption of internal balance). Fertility, as measured by the percentage of fertile matings, was somewhat modified by interchromosomal interactions, but the same genetic manipulations showed no effect on the survival estimates. Interallelic interactions (dominance) affected the mean survival estimates to the greatest extent and heterozygosity also reduced their variability. Developmental periods were appreciably affected by the disruption of the internal balance of the various X chromosomes, and moderately affected by X chromosomal heterozygosity. General reproductivity was the component most affected by X chromosomal heterozygosity, which also modified all components (except perhaps fertility).

LITERATURE CITED

BONNIER, G., and U. B. JONSSON, 1957 Rate of development of viability mutants of *Drosophila melanogaster*. Evolution **11**: 271–279.

BONNIER, G., U. B. JONSSON, and C. RAMEL, 1959 Experiments on the effects of homozygosity and heterozygosity on the rate of development in *Drosophila melanogaster*. Genetics **44**: 679–704.

HIRAIZUMI, Y., 1960 Negative correlation between rate of development and female fertility in *Drosophila melanogaster*. Genetics **46**: 615–624.

KELLER, E. C., JR., 1959 Preliminary analysis of interchromosomal genotypic balance in inbred lines of *Drosophila melanogaster*. M.S. thesis. The Pennsylvania State University, University Park. —— 1961 Genetic-environmental interaction in inbred and hybrid strains of *Drosophila melanogaster*. Ph.D. thesis. The Pennsylvania State University, University Park.

KELLER, E. C., JR., and D. F. MITCHELL, 1962 Interchromosomal genotypic interactions. I. An analysis of morphological characters. Genetics **47**: 1557–1571.

MAYNARD SMITH, J., 1955 Mating behaviour and larval selection in *Drosophila subobscura*. J. Genet. **45**: 261–279.

MITCHELL, D. F., 1958 Inversion heterozygosity, recombination, and variability of quantitative traits. Cold Spring Harbor Symp. Quant. Biol. **23**: 279–290.

REED, S. C., 1941 Interaction between the autosomes of *Drosophila melanogaster* as measured by viability and rate of development. Can. J. Res., Sect. D **19**: 75–84.

ROBERTSON, F. W., 1957 Studies on quantitative inheritance. XI. Genetic and environmental correlation between body length and egg laying rate in *Drosophila melanogaster*. J. Genet. **55**: 428–433.