Developmental homeostasis of relative viability was examined for homozygotes and heterozygotes using second chromosomes from two populations of Drosophila melanogaster. One was a chromosome population in which spontaneous mutations were allowed to accumulate since it was begun with a single near-normal second chromosome. The second was a natural population approximately at equilibrium. For the estimation of relative viability, the Cy method was employed (Wallace 1956), and environmental variance between simultaneously replicated cultures was used as the index of developmental homeostasis. A new method was used in the estimation of sampling variance for relative viability that was employed for the calculation of environmental variance (error variance between simultaneously replicated cultures — sampling variance). The following findings were obtained: (1) The difference in environmental variance between homozygotes and heterozygotes could not be seen when a chromosome population with variation due to new mutations was tested. (2) When a chromosome group isolated from an approximate equilibrium population was examined, heterozygotes manifested a smaller environmental variance than the homozygotes if their relative viabilities were approximately the same. (3) There was a slight negative correlation between viability and environmental variance, although opposite results were found when the viabilities of individuals were high, especially when overdominance (coupling overdominance, Mukai 1969a, b) was manifest. On the basis of these findings, it was concluded that developmental homeostasis was a product of natural selection, and its mechanism was discussed.

Since the stability of heterozygotes over homozygotes in fitness or in its components for varied environmental conditions provides, under certain conditions, a basis for the maintenance of genetic variability in a random mating population, special attention has recently been paid to this phenomenon by several investigators (Powell 1971; Powell and Wistrand 1978; Gillespie and Langley 1974; Lewontin, Ginzburg and Tuiljapurkar 1978; Wills 1975).

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† On leave of absence from Ochanomizu University, Tokyo, Japan from November 1976 to March 1977.
Many population geneticists agree that the heterozygotes originating from a natural population are more stable with respect to viability, over a wide range of environmental conditions, than are the homozygotes (DOBZHANSKY and WALLACE 1953; DOBZHANSKY and LEVENE 1955). However, no consensus has been reached with respect to the stability of quantitative characters such as wing length and sternopleural bristle numbers in D. melanogaster (ROBERTSON and REEVE 1952; MATHER 1953; BEARDMORE 1960).

This type of stability, termed developmental homeostasis, has been considered to be one of the manifestations of co-adaptation of genic systems (DOBZHANSKY and LEVENE 1955) resulting from natural selection, although there is no decisive evidence supporting this understanding. On the contrary, there exists another explanation for developmental homeostasis in which "mere diversity of alleles at different loci," or higher heterozygosity, *per se*, may give sufficient basis for the stability of heterozygotes (LERNER 1954).

It is important to ask which of the two hypotheses is more appropriate or, perhaps, whether a new hypothesis is necessary to explain the nature of developmental homeostasis. This question may be partially answered if we compare the stability of heterozygotes originating from a natural population with that of heterozygotes arising from spontaneous mutation and a minimum of selection pressure. The results of such an analysis are presented in this paper.

**MATERIALS AND METHODS**

*Materials:* Spontaneous mutations affecting viability were accumulated in the second chromosomes of lines that had originated from a single normal second chromosome at minimum natural selection pressure (MUKAI 1964) for many generations. At certain generations, homozygous and heterozygous viabilities were estimated by the Cy method (WALLACE 1956). For the estimation of homozygous viability, the following crosses were made with simultaneous replications: $\text{Cy}+/+ (5\Omega \Omega) \times \text{Cy}+/+ (5\delta \delta)$ where Cy is the dominant marker gene affecting wing character and is included in $\text{In}(2LR)\text{SM1}$. + stands for the wild-type second chromosome of line i. Four days after crosses were made, all parental flies were transferred to second vials. Five days after the transfer was made, all parental flies were discarded. In the offspring, only two types of flies, $\text{Cy}+/+$ and $+/+\text{a}$, segregate since Cy homozygotes are lethal. The expected segregation ratio is 2 : 1. The numbers of Cy flies within the pair of vials were pooled and considered to be a single observation. The same thing was done for $+/+\text{a}$ flies. The relative viability of $+/+\text{a}$ individuals is expressed as $2a/(b+1)$ following HALDANE (1956) where $b:a$ is the ratio of Cy to wild-type phenotypes.

For the estimation of heterozygote viability, five pair matings between line i and line j were made with simultaneous replications $[\text{Cy}+/+ (5\Omega \Omega) \times \text{Cy}+/+ (5\delta \delta)]$. In the offspring, phenotypically Cy and wild-type flies segregate at the expected ratio of 2 : 1. Viabilities of heterozygotes were expressed in the same manner as for homozygotes.

For homozygotes, the results of viability estimation at generations 32, 52 and 60 were employed in the present analysis. For heterozygotes, the results at generations 32 and 52 were used. The analysis for the means and genetic variances for these generations have been reported in MUKAI and YAMAZAKI (1968).

In addition, special crosses made at generation 32 were used for examining the relationship between the environmental variance and genetic background. At this generation, $\text{Cy}+/+ (5\delta \delta)$ were crossed to $+/+\text{a}(7\Omega \Omega) \times +\text{a}+/+\text{a}(7\delta \delta)$ where + was supposed to be identical to the original $+/+$, except for one or two mutations, and $+/+\text{a}$ stands for a near-normal second chromosome that was extracted from the same population as $+/+\text{a}$. Thus, $+/+\text{a}$ is homozygous except
for the loci where new mutations accumulated. The heterozygosity of \( +_{\alpha}/+_{i} \) is expected to be higher than that of \( +_{\alpha}/+/ \) but probably less than the average heterozygosity of the population, since the relative viability of \( +_{\alpha}/+_{\alpha} \) was close to that of the best homozygote in the population (see Mukai, Chigusa and Yoshioka 1965). For the sake of simplicity, \( +_{\alpha}/+/ \) and \( +_{\alpha}/+/ \) will be called homozygous background and heterozygous background, respectively.

The relative viabilities of \( +_{\alpha}/+_{i} \) and \( +_{\alpha}/+/ \) were expressed by the ratio of \( a/(b+1) \) where \( b \) and \( a \) are the numbers of \( Cy \) and wild-type flies in the offspring of \( +_{\alpha}/+/ \) (or \( +_{\alpha}/+/ \)) \( \times Cy+/+ \). In both types of crosses eight simultaneous replications were made. The relative viabilities of \( +_{\alpha}/+_{i} \) heterozygotes were also estimated at generation 60, with six simultaneous replications. All the above experiments were done using 2.5 cm \( \times \) 10 cm vials. The original viability data were published in Mukai, Chigusa and Yoshioka (1965) and Mukai and Yamazaki (1968) where relative viability was expressed as the percentage of wild-type flies.

The second chromosomes extracted from the Raleigh, N. C. populations were also used. The original data were published in Mukai and Yamaguchi (1974). As in the case of chromosomes in which spontaneous mutations were accumulated, homozygote and heterozygote viabilities were estimated by the \( Cy \) method. Parental flies were transferred to the second vials four days after the crosses were made, and four days later the parents were discarded. Four simultaneous replications were made in 2 cm \( \times \) 10 cm vials.

Method for estimating environmental variance: In the early stages of these experiments, the viability of wild-type flies was expressed as a percentage of the total number of flies, and the environmental variance was estimated by the difference between the observed and expected variance among replicated cultures. The expected variance was calculated assuming that the numbers of adult \( Cy- \) and wild-type flies were binomially distributed (Dobzhansky and Levene 1955).

However, Professor C. C. Cockerham at North Carolina State University suggested to one of the authors (T. M.) that this assumption is not correct. Some proportion of the \( Cy \) (or wild-type) eggs will become \( Cy \) (or wild-type) flies following a binomial distribution. However, the viability index which we are going to use is actually a ratio of two terms from two independent binomial distributions. The distribution of this ratio is not binomial. Hence, the correct sampling variance for the present viability index was calculated.

A. Cases where \( Cy- \) and wild-type flies segregate at the expected ratio of 2 : 1: At first, it is assumed that the expected ratio of the number of wild-type to \( Cy \) eggs is 1 : 2 for the case of \( Cy+/+ \times Cy+/+ \). The assumptions are: (1) The original numbers of \( Cy \) eggs and wild-type eggs are \( M \) and \( M/2 \). Since \( M \) is a large number (more than 700) in the present experiment, the ratio of the numbers of \( Cy \) and wild-type eggs is assumed to be constant (2 : 1) and \( M \) is also constant as a first approximation. The validity of these assumptions will be discussed later. (2) The numbers of adult \( Cy \) and wild-type flies are \( m \) and \( n \), respectively. Let us equate \( p_1 \) to \( m/M \) and \( p_2 \) to \( n/(M/2) \) where \( p_1 \) and \( p_2 \) are distributed binomially. The sampling variances of \( p_1 \) and \( p_2 \) [\( V(p_1) \) and \( V(p_2) \), respectively] are expressed as follows:

\[
V(p_1) = \frac{m}{M} \left( 1 - \frac{m}{M} \right) = \frac{m(M-m)}{M^3} \tag{1}
\]

\[
V(p_2) = \frac{2n}{M} \left( 1 - \frac{2n}{M} \right) = \frac{4n(M-2n)}{M^3} \tag{2}
\]

The viability index \( (\nu) \) which has been employed is as follows:

\[
\nu = \frac{2n}{m} = \frac{2}{Mp_1} = \frac{p_2}{p_1} \tag{3}
\]
The following two cases will be considered: environmental effects act (1) additively and (2) independently at individual loci.

(1) Environmental effects act additively: Assuming that $p_1$ and $p_2$ are independent, the sampling variance of $u$ [denoted $V_u$] can be expressed as follows:

$$V_u = V(p_2) \left\{E \left( \frac{1}{p_1} \right) \right\}^2 + V \left( \frac{1}{p_1} \right) \{E(p_2)\}^2 + V(p_2) \cdot V \left( \frac{1}{p_1} \right)$$  \hspace{1cm} (4)

In order to simplify the above formula, the following calculation was made:

$$\left\{ E \left( \frac{1}{p_1} \right) \right\}^2 = \left( \frac{1}{\bar{p}_1} \right)^2 = \left( \frac{M}{\bar{m}} \right)^2$$  \hspace{1cm} (5)

where $\bar{p}_1$ and $\bar{m}$ are harmonic means of $p_1$ and $m$, respectively.

$$V \left( \frac{1}{p_1} \right) = V(p_1) \times \left[ \frac{d \left( \frac{1}{p_1} \right)}{dp_1} \right]^2 = \frac{m(M-m)}{M^3} \cdot \frac{M^4}{m^4} = \frac{M(M-m)}{m^3}$$  \hspace{1cm} \text{[using formula (1)]}

Using formulae (2), (5) and (6), $V_u$ becomes as follows:

$$V_u \approx \frac{4n(M-2n)}{M^3} \cdot \left( \frac{M}{\bar{m}} \right)^2 + \frac{M(M-m)}{m^3} \cdot \frac{4n^2}{M^2}
+ \frac{4n(M-2n)}{M^3} \cdot \frac{m}{m^3}
+ \frac{4n(M-2n)}{M^3} \cdot \frac{4n(M-2n)(M-m)}{Mm^3}$$

Since the coefficient of variation of $m$ is small, $\bar{m}$ may approximately be equated to $m$. Then,

$$V_u \approx \frac{4n}{M^2m^3} \left[ Mm(M-2n) + Mn(M-m) + (M-2n)(M-m) \right]$$  \hspace{1cm} (7)

In the actual analysis, for each replicate vial, $m$ and $n$ can be obtained, and $V_u$ can be estimated assuming a certain value for $M$. In fact, the number of eggs were counted recently under the same condition as the actual experiments described above. On the basis of this number, a reasonable value for $M$ was assumed. The average value of $V_u(\hat{V}_u)$ over all replicate cultures in a certain homozygote or heterozygote cross was calculated. Simultaneously, the error variance of viability indices within the homozygote or heterozygote was estimated (\hat{V}_o). The environmental variance ($V_{E\hat{u}}$) for this homozygote or heterozygote was estimated as follows:

$$V_{E\hat{u}} = \hat{V}_o - \hat{V}_u$$  \hspace{1cm} (8)

This is the basic statistic in which we are interested.

(2) Case where environmental effects act independently. In this case the logarithmic transformation of the viability index or formula (3) is appropriate. Then,

$$v = \ln p_2 - \ln p_1$$  \hspace{1cm} (9)
The sampling variance of $v(V_{st})$ can be calculated as follows, assuming that (1) $p_1$ and $p_2$ are independent and (2) sample size is very large:

$$V_{st} = V(p_2) \left[ \frac{d\ln p_2}{dp_2} \right]^2 + V(p_1) \left[ \frac{d\ln p_1}{dp_1} \right]^2$$

$$= V(p_2) \left( \frac{1}{p_2} \right)^2 + V(p_1) \left( \frac{1}{p_1} \right)^2$$

Using the relationships of formulae (1) and (2),

$$V_{st} = \frac{(1 - \frac{2n}{M})}{n} + \frac{(1 - \frac{m}{M})}{m}$$

$$= \frac{m + n - \frac{3mn}{M}}{mn} \quad \text{(10)}$$

In a similar manner, the environmental variance ($V_{EN}$) can be calculated.

(3) Traditional method: Assuming a binomial distribution of the numbers of Cy and wild-type flies in a vial without environmental variance, the sampling variance ($V_{sb}$) of the viability index $v = \frac{2n}{m}$ can be calculated as follows:

$$V_{sb} = V \left( \frac{2(1-p)}{p} \right)$$

where $p$ is the proportion of Cy flies. Assuming a large sample size, $V_{sb}$ can be simplified as follows:

$$V_{sb} = \frac{4n(m+n)}{m^3} \quad \text{(11)}$$

In a similar manner, the sampling variance for the viability index in the case of logarithmic transformation can be shown to equal:

$$V_{sbl} = \frac{m + n}{mn} \quad \text{(12)}$$

B. Cases where the Cy- and wild-type flies segregate at the expected ratio of 1:1. The formulae corresponding to (7), (10), (11) and (12) are obtained as follows:

$$V_s = \frac{n}{M^2m^2} [Mm(M-n) + Mn(M-m) + (M-m)(M-n)] \quad \text{(7')}$

$$V_{st} = \frac{m + n - \frac{2mn}{M}}{mn} \quad \text{(10')}$

$$V_{sb} = \frac{n(m+n)}{m^3} \quad \text{(11')}$

$$V_{sbl} = \text{the same as (12).}$$

We are aware that In(2LR)SM1 (Cy) chromosomes, in heterozygous condition, do not always suppress completely the effects of deleterious genes located in their homologous chromosomes (MUKAI et al. 1974; COCKERHAM and MUKAI 1978; MUKAI 1981). If developmental
homeostasis for viability is related to some of these deleterious mutant genes, there is a possibility that the result of the analysis assuming complete dominance of the Cy chromosomes might be biased. An analysis was done assuming additivity between homologous chromosomes with respect to the effects of environment. The results indicate that the assumption of complete dominance did not cause any serious bias, this point will be described further in the Discussion.

ANALYSES

Chromosomes from a natural population: The results of viability estimation performed for the chromosomes extracted from the Raleigh, N. C., population of *D. melanogaster* were employed for this analysis. First, approximately 700 second chromosomes were extracted by the marked inversion technique, and the viabilities of these homozygotes and the heterozygotes of their random combination were estimated by the Cy method (Wallace 1956). Four simultaneous replications were made (Mukai and Yamaguchi 1974). If there were one or more missing observations out of four, that homozygote or heterozygote was excluded from the analysis. Furthermore, if there was any homozygote whose viability index was less than 0.5, it was not employed in the analysis. The reason is that such a homozygote might have carried major-genic semi-lethal genes whose nature, especially in homozygous condition, is different from that of viability polygenes in which we are mainly interested. Corresponding to this restriction, only heterozygotes whose viabilities were larger than 0.5 were used for the analysis. In these heterozygotes, major-genic semi-lethal mutations were included in heterozygous condition, but their possible effects were disregarded.

By means of formulae (7), (10), (11) and (12), the expected sampling variances of each estimate of viability for both homozygotes and heterozygotes were calculated. The average value of the sampling variance among the simultaneous replications was also calculated. The observed variance for the simultaneous replicates was estimated \[\hat{V}_e\] for each of the homozygote lines and heterozygote crosses. Applying the above two estimates (observed variance and sampling variance) to formula (8), we estimated the environmental variance (\[\hat{V}_{en}\]) for each of the homozygote lines and heterozygote crosses. The same calculations were done after taking the natural logarithms of the viability indices. In this case we assume that there are many environmental factors, each with small effect, and that they act on the viabilities of individuals independently. In these calculations, the numbers of \(Cy/+\) and \(+/+\) eggs were assumed to be 700 and 350 per observation, respectively; i.e., \(M = 700\) in formulae (7) and (10). We have good reasons for making this assumption (Mukai, Schaffer and Cockerham 1972), and one of the present authors (S. K.) found the total number of eggs and larvae to be much more than 800 in the present experimental condition. Although there is variation in numbers of eggs laid among cultures, we assume that \(M\) is constant as a first approximation. Indeed, the genotypes of the parental flies were \(Cy\) heterozygotes and the variance of the number of eggs must be small. It is shown in the Discussion that the effect of variable \(M\) on \(\hat{V}_{en}\) is small.

In addition, the environmental variances were calculated using the traditional method in which it is assumed that the proportions of adult \(Cy/+\) and \(+/+\) flies follow the same binomial distribution.
The results of flies homozygous and heterozygous for chromosomes extracted from a natural population are shown in Table 1. Both homozygotes (part A) and heterozygotes (part B) were classified into several groups according to their relative viability. Within each group are presented the mean viability, the number of flies, and the environmental variance.

From Table 1, the following findings can be obtained: (1) When comparing heterozygotes and homozygotes of approximately equal viability, the environmental variance of heterozygotes is less than that of homozygotes regardless of how the environmental variance is expressed. This finding agrees with that obtained by Dobzhansky and Wallace (1953), Dobzhansky and Levene (1955), Wills (1975) and others. That is to say, heterozygotes are more homeostatic than the corresponding homozygotes for the existing microenvironmental variation. (2) The correlation coefficient between relative viability and environmental variance is generally negative, although its absolute value is close to 0. This small absolute value is partially due to a relatively large $\hat{V}_{EX}$ value when the mean viability is greater than one. This can be understood as follows: The extremely large viability (larger than one) is partially due to some replicate cultures in which environmental factors worked most favorably to increase the relative viability. On the contrary, in intermediate viability classes, some lines showing large environmental variances move to the adjacent classes, and simultaneously, approximately the same number of lines or crosses come from the adjacent classes. Thus, the average environmental variance is not influenced significantly. Hence it appears that the absolute values of the correlation coefficients between viability and environmental variance are slightly (negatively) underestimated. (3) The environmental variance based upon a single binomial distribution of $Cy/+\text{ and } +/+\text{ flies (traditional method) is smaller than that estimated using the new method.}$

Disregarding the correlation between the viability and environmental variance, the distribution patterns of environmental variances estimated by the new method without logarithmic transformation are graphically presented in Figure 1. From this figure, it is clearly seen that the environmental variances of heterozygotes are smaller than those of homozygotes.

Chromosomes with variation due to newly arisen mutations alone: Originally, 104 second chromosome lines were initiated from a single normal second chromosome, and mutations were accumulated at a minimum natural selection pressure. Homozygote viabilities estimated at generations 32, 52 and 60 were employed in the present analysis. The numbers of lines whose homozygous viabilities were larger than 0.5 were 80, 36 and 14 at generations 32, 52 and 60, respectively. The numbers of simultaneous replications were 8, 6 and 4 at generations 32, 52 and 60, respectively.

Heterozygote viabilities were estimated at generations 32 and 52. The viabilities of heterozygotes between successively numbered lines were estimated at generation 32, but at generation 52 those between successively numbered lines (RS) as well as between alternately numbered lines (RA) were estimated. The numbers of crosses whose viabilities were larger than 0.5 were 83, 45 and 42 at generations 32, 52 (RS), and 52 (RA), respectively.
### TABLE 1

*Environmental variance for second chromosomes extracted from a natural population of Drosophila melanogaster*

<table>
<thead>
<tr>
<th>Range of viability</th>
<th>Mean viability</th>
<th>No. of lines</th>
<th>Average No. of flies per observation</th>
<th>Ratio of two independent binomial terms*</th>
<th>Ratio of two terms of single binomial dist.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Standardized by $v$</td>
<td>Standardized by $\ln v$</td>
</tr>
<tr>
<td>0.5 — 0.6</td>
<td>0.5555</td>
<td>32</td>
<td>338.7</td>
<td>0.01430</td>
<td>0.04996</td>
</tr>
<tr>
<td>0.6 — 0.7</td>
<td>0.6605</td>
<td>54</td>
<td>334.4</td>
<td>0.01796</td>
<td>0.04683</td>
</tr>
<tr>
<td>0.7 — 0.8</td>
<td>0.7553</td>
<td>70</td>
<td>352.3</td>
<td>0.01437</td>
<td>0.02999</td>
</tr>
<tr>
<td>0.8 — 0.9</td>
<td>0.8475</td>
<td>92</td>
<td>351.5</td>
<td>0.01135</td>
<td>0.01848</td>
</tr>
<tr>
<td>0.9 — 1.0</td>
<td>0.9497</td>
<td>64</td>
<td>359.9</td>
<td>0.01326</td>
<td>0.01605</td>
</tr>
<tr>
<td>1.0 — —</td>
<td>1.0362</td>
<td>16</td>
<td>362.3</td>
<td>0.01873</td>
<td>0.01770</td>
</tr>
</tbody>
</table>

Mean environmental variance ($\hat{\nu}_{EN}$)  
$0.0141 \pm 0.0012$  
$0.0282 \pm 0.0028$  
$0.0112 \pm 0.0012$  
$0.0192 \pm 0.0027$

Correlation between $\hat{\nu}_{EN}$ and viability  
$-0.049 \pm 0.057$  
$-0.341 \pm 0.053$  
$-0.091 \pm 0.057$  
$-0.344 \pm 0.053$

Variance of $\hat{\nu}_{EN}$  
$0.000453$  
$0.002540$  
$0.000454$  
$0.002524$
TABLE 1—Continued

[B] Heterozygotes (number of crosses tested = 674)

<table>
<thead>
<tr>
<th>Range of viability</th>
<th>Mean viability</th>
<th>No. of crosses</th>
<th>Average No. of flies per observation</th>
<th>Ratio of two independent binomial terms*</th>
<th>Ratio of two terms of single binomial dist.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>standardized by ( \nu )</td>
<td>standardized by ( \nu )</td>
</tr>
<tr>
<td>0.5 — 0.7</td>
<td>0.7530</td>
<td>13</td>
<td>332.5</td>
<td>0.01109</td>
<td>0.02343</td>
</tr>
<tr>
<td>0.7 — 0.8</td>
<td>0.8581</td>
<td>73</td>
<td>337.7</td>
<td>0.00402</td>
<td>0.00337</td>
</tr>
<tr>
<td>0.8 — 0.9</td>
<td>0.9574</td>
<td>261</td>
<td>358.9</td>
<td>0.00565</td>
<td>0.00449</td>
</tr>
<tr>
<td>0.9 — 1.0</td>
<td>1.0462</td>
<td>233</td>
<td>357.9</td>
<td>0.00709</td>
<td>0.00476</td>
</tr>
<tr>
<td>1.0 — 1.1</td>
<td>1.1348</td>
<td>79</td>
<td>349.3</td>
<td>0.00710</td>
<td>0.00446</td>
</tr>
<tr>
<td>1.2</td>
<td>1.2407</td>
<td>15</td>
<td>378.2</td>
<td>0.02675</td>
<td>0.01404</td>
</tr>
</tbody>
</table>

Mean environmental variance (\( \hat{\nu}_{EN} \))

0.00671 ± 0.00058
0.00504 ± 0.00063
0.00288 ± 0.00058
0.00273 ± 0.00063

Correlation between \( \hat{\nu}_{EN} \) and viability

0.125‡ ± 0.039
-0.067 ± 0.039‡
-0.072 ± 0.039
-0.078*‡ ± 0.038‡

Variance of \( \hat{\nu}_{EN} \)

0.000230
0.000264
0.000225
0.000267

* The number of eggs per observation was assumed to be 1050 (\( Cy/+ : 700, +/+ : 350 \)).

‡ Correlation between \( \ln \nu \) and \( \hat{\nu}_{EN} \).

$\dagger$ Significant at the 1% level.

§ Highly significant.
Analyses were conducted independently for each generation and for each group (RS or RA). However, since it was likely that there was no significant difference between them, the results of all the homozygotes or all the heterozygotes were pooled. Therefore, although there are 130 homozygous lines and 170 heterozygote crosses, there is some correlation between lines or between crosses. Analyses were conducted in entirely the same way as in the case of chromosomes from the natural population. \( M \) was also assumed to be 700. The results are summarized in Table 2.

From Table 2, the following findings can be obtained: (1) Contrary to the results described above, the environmental variances for heterozygotes are not smaller than those for homozygotes. Indeed, the former is larger than the latter, but they are not significantly different from each other. (2) The correlation coefficient between relative viability and environmental variance is always negative as in the case of chromosomes extracted from the natural population. The characteristic described as (3) for the natural chromosomes can be applied to the chromosomes with variation due to new mutations alone. Disregarding the correlation between the viability and environmental variance, the distribution patterns of environmental variances estimated by using the new method without logarithmic transformation are graphically presented in Figure 2. From this figure, it can be seen that the distribution pattern of environmental variance for heterozygotes is quite similar to that for homozygotes.
TABLE 2

Environmental variance for second chromosomes within variation due to newly arisen mutations alone in Drosophila melanogaster

[A] Homozygotes (number of lines tested = 130)

<table>
<thead>
<tr>
<th>Range of viability</th>
<th>Mean viability</th>
<th>No. of lines</th>
<th>No. of flies per observation</th>
<th>Ratio of two independent binomial terms*</th>
<th>Ratio of two terms of single binomial dist.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Standardized by ( \bar{v} )</td>
<td>( \ln \bar{v} )</td>
</tr>
<tr>
<td>0.5 - 0.6</td>
<td>0.5594</td>
<td>24</td>
<td>610.3</td>
<td>0.00982</td>
<td>0.03119</td>
</tr>
<tr>
<td>0.6 - 0.7</td>
<td>0.6582</td>
<td>24</td>
<td>635.2</td>
<td>0.00547</td>
<td>0.01361</td>
</tr>
<tr>
<td>0.7 - 0.8</td>
<td>0.7540</td>
<td>37</td>
<td>651.5</td>
<td>0.00694</td>
<td>0.01220</td>
</tr>
<tr>
<td>0.8 - 0.9</td>
<td>0.8463</td>
<td>29</td>
<td>664.2</td>
<td>0.00781</td>
<td>0.01140</td>
</tr>
<tr>
<td>0.9</td>
<td>0.9664</td>
<td>16</td>
<td>725.8</td>
<td>0.00617</td>
<td>0.00660</td>
</tr>
</tbody>
</table>

Mean environmental variance (\( \hat{V}_{EN} \)) \[ 0.00730 \pm 0.00055 \]

Correlation between \( \hat{V}_{EN} \) and viability \[ -0.038 \pm 0.090 \]

Variance of \( \hat{V}_{EN} \) \[ 0.000039 \]
**TABLE 2—Continued**

[B] Heterozygotes (number of crosses tested = 170)

<table>
<thead>
<tr>
<th>Range of viability</th>
<th>Mean viability</th>
<th>No. of crosses</th>
<th>Average No. of flies per observation</th>
<th>Ratio of two independent binomial terms*</th>
<th>Ratio of two terms of single binomial dist.</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Standardized by $v$</td>
<td>Standardized by $v$</td>
</tr>
<tr>
<td>0.5 — 0.6</td>
<td>0.5517</td>
<td>31</td>
<td>616.0</td>
<td>0.01054</td>
<td>0.03779</td>
</tr>
<tr>
<td>0.6 — 0.7</td>
<td>0.6462</td>
<td>36</td>
<td>596.7</td>
<td>0.01202</td>
<td>0.03016</td>
</tr>
<tr>
<td>0.7 — 0.8</td>
<td>0.7559</td>
<td>38</td>
<td>643.6</td>
<td>0.00695</td>
<td>0.01228</td>
</tr>
<tr>
<td>0.8 — 0.9</td>
<td>0.8432</td>
<td>40</td>
<td>636.1</td>
<td>0.00801</td>
<td>0.01133</td>
</tr>
<tr>
<td>0.9 — 1.0</td>
<td>0.9587</td>
<td>13</td>
<td>694.4</td>
<td>0.00476</td>
<td>0.00526</td>
</tr>
<tr>
<td>1.0 — 1.1</td>
<td>1.0665</td>
<td>4</td>
<td>682.1</td>
<td>0.00622</td>
<td>0.00630</td>
</tr>
<tr>
<td>1.1 ——</td>
<td>1.1470</td>
<td>8</td>
<td>722.5</td>
<td>0.01224</td>
<td>0.00972</td>
</tr>
</tbody>
</table>

Mean environmental variance ($\hat{\nu}_{E_N}$)  

| Mean environmental variance ($\hat{\nu}_{E_N}$) | 0.00899 ± 0.00065 | 0.01970 ± 0.00184 | 0.00637 ± 0.00066 | 0.01541 ± 0.00184 |

Correlation between $\hat{\nu}_{E_N}$ and viability  

| Correlation between $\hat{\nu}_{E_N}$ and viability | $-0.134 \pm 0.157$ | $-0.348^{\dagger} \pm 0.073^{\dagger}$ | $-0.114 \pm 0.078$ | $-0.348^{\dagger} \pm 0.073^{\dagger}$ |

Variance of $\hat{\nu}_{E_N}$  

| Variance of $\hat{\nu}_{E_N}$ | 0.000071 | 0.000577 | 0.000073 | 0.000577 |

---

* The number of eggs per observation was assumed to be 1050 ($Cy/+/: 700$, $+/+ : 350$).

† Correlation between $\ln v$ and $V_{E_N}$.

‡ Significant at the 1% level.

§ Highly significant.
Comparison of the environmental variances for two types of chromosomes: natural chromosomes and chromosomes with variation due only to new mutations: The environmental variance for heterozygotes is significantly smaller than that for homozygotes in natural chromosomes, in contrast to the chromosomes with variation due solely to new mutations. Since there is a small correlation between the environmental variance and viability, the relation between the mean viability and the average of environmental variances for the same lines or crosses shown in Tables 1 and 2 are graphically presented in Figure 3. It can be seen from this figure that the above finding holds true for a large range of viability. Since the culture conditions of these two types of chromosomes (see MATERIALS AND METHODS) are different, their absolute values cannot be compared.

Effect of heterozygosity on the environmental variance in different genetic backgrounds: For the analyses of the homozygous background and the heterozygous background, environmental variances were estimated by the new method without logarithmic transformation. In the case of the homozygous background, the relationship between the environmental variances for heterozygotes (+o/+i) and the viabilities of the corresponding homozygotes (+i/+i) are given in Table 3. For the sake of reference, the heterozygote viabilities (+o/+i) are also pre-
Figure 3.—The relationships between viabilities and environmental variances for chromosomal homozygotes and heterozygotes. Left: chromosomes with variation due to newly arisen mutations alone. Right: chromosomes from a natural population.

Table 3

Environmental variance for relative viabilities of heterozygotes for newly arisen mutations alone

<table>
<thead>
<tr>
<th>Range of homozygous viability (+/+ and 0)</th>
<th>Mean viability Homozygotes</th>
<th>Mean viability Heterozygotes</th>
<th>No. of lines</th>
<th>Environmental variance for heterozygotes</th>
<th>Average No. of flies per observation</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 — 0.10</td>
<td>0.0934</td>
<td>1.0891</td>
<td>2</td>
<td>0.00734</td>
<td>838.8</td>
</tr>
<tr>
<td>0.10 — 0.20</td>
<td>0.1075</td>
<td>1.0947</td>
<td>14</td>
<td>0.00429</td>
<td>807.4</td>
</tr>
<tr>
<td>0.20 — 0.30</td>
<td>0.2427</td>
<td>1.1398</td>
<td>11</td>
<td>0.00538</td>
<td>788.5</td>
</tr>
<tr>
<td>0.30 — 0.40</td>
<td>0.3461</td>
<td>1.1221</td>
<td>18</td>
<td>0.00570</td>
<td>820.3</td>
</tr>
<tr>
<td>0.40 — 0.50</td>
<td>0.4488</td>
<td>1.1103</td>
<td>10</td>
<td>0.00346</td>
<td>802.9</td>
</tr>
<tr>
<td>0.50 — 0.60</td>
<td>0.5591</td>
<td>1.1077</td>
<td>6</td>
<td>0.00513</td>
<td>765.4</td>
</tr>
<tr>
<td>0.60 — 0.70</td>
<td>0.6543</td>
<td>1.0816</td>
<td>12</td>
<td>0.01027</td>
<td>682.0</td>
</tr>
<tr>
<td>0.70 — 0.80</td>
<td>0.7549</td>
<td>1.0718</td>
<td>35</td>
<td>0.00714</td>
<td>616.8</td>
</tr>
<tr>
<td>0.80 — 0.90</td>
<td>0.8441</td>
<td>1.0680</td>
<td>27</td>
<td>0.00500</td>
<td>633.0</td>
</tr>
<tr>
<td>0.90 — 1.00</td>
<td>0.9385</td>
<td>1.0224</td>
<td>18</td>
<td>0.00381</td>
<td>713.1</td>
</tr>
<tr>
<td>1.00 — 1.20</td>
<td>1.0591</td>
<td>0.9863</td>
<td>4</td>
<td>0.00136</td>
<td>710.0</td>
</tr>
</tbody>
</table>

Mean environmental variance 0.00563 ± 0.000451

Correlation between homozygote viability and corresponding heterozygote $\hat{\nu}_E$ $-0.168^* \pm 0.080$

* Significant at the 5% level.
The new method (without logarithmic transformation) used. The number of crosses = 157.
sent in Figure 4 and in Table 3. It should be noted here that, although it is an exceptional case, $+o/+i$ showed overdominance (Mukai 1969b and earlier). This has been called "coupling-overdominance" (Mukai 1969a), since newly arisen mutations were accumulated in only one of the homologous chromosomes ($+i$). For the homozygotes at generation 60, the lines that acquired a major genic semi-lethal mutation after the test at generation 32 were not excluded. Practically speaking, it was impossible to do so, since the homozygous viabilities of many chromosome lines fell below 0.5 due to the accumulation of polygenic viability mutations and synergistic interaction (Mukai 1969c). In this table, the pooled results of generations 32 and 60 are given, since the results of the two generations were very similar. The decrease in the homozygous viabilities ($+i/+i$) implies the increase in the degrees of heterozygosities of the corresponding heterozygotes ($+o/+i$). There is a small but significant negative correlation between the viabilities of $+i/+i$ and the $V_{EX}$ of the corresponding heterozygotes $+o/+i$ ($R = -0.168 \pm 0.080$) (see Table 3 and Figure 4). Furthermore, the correlation coefficient between the environmental variances and the viabilities of $+o/+i$ heterozygotes was calculated to be an $0.218 \pm 0.080$ which is significantly larger than 0 at the 1% level. On the basis of the above findings it may be concluded that overdominance and developmental homeostasis are not always the only aspects of co-adaptation of the genic system.

The heterozygous background data were available only for generation 32. The results corresponding to the homozygous background are given in Table 4 and Figure 4. Clear correlation cannot be seen between homozygous viabilities...
and the environmental variances for the corresponding heterozygotes ($+w/+j$) ($R = -0.103 \pm 0.113$). This implies that increases in the magnitude of heterozygosity near the average heterozygosity in the population do not necessarily increase the stability of heterozygotes.

From the results of the homozygous and heterozygous backgrounds, at least it may be concluded that developmental homeostasis is not caused by heterozygosity, per se. This is also consistent with the results of the chromosomes extracted from a natural population because there is a slight negative correlation between viability and environmental variance in heterozygotes. If heterozygosity, per se, increases the stability of individuals, there should be a positive correlation between homozygous viability and heterozygous environmental variance since the majority of mutations are harmful to the carriers in heterozygous condition in natural populations (Mukai and Yamaguchi 1974).

**DISCUSSION**

*Analytical method:* The method we used has some weak points: (1) The $Cy$ chromosomes are assumed to be completely dominant over their homologous chromosomes. (2) The value of $M$ was assumed to be constant. (3) The ratio of the numbers of $Cy/+\text{ and} (+/+\text{ genotypes was assumed to be 2 : 1 among the eggs laid. These assumptions are not realistic, and the effects of these three factors on the estimation of } V_s \text{ have been examined.}

First, the effect of the incomplete dominance of the $Cy$ chromosome on $V_{EN}$ was examined. With the $Cy$ method, in which viability was expressed as $2 \times (\text{the number of wild-type flies})/(\text{the number of } Cy \text{ flies} + 1)$, the denominator consists of one genotype ($Cy/+i$) in the estimation of homozygote viability, while of two genotypes ($Cy/+i$ and $Cy/+j$) in the estimation of heterozygote viability.

**TABLE 4**

Environmental variance for relative viabilities of heterozygotes between near-normal single second chromosomes and chromosomes with variation due to newly arisen mutations alone

<table>
<thead>
<tr>
<th>Range of homozygous viabilities ($+/i$)</th>
<th>Mean homozygous viability</th>
<th>Mean heterozygous viability ($+w/+i$)</th>
<th>No. of lines</th>
<th>Environmental variance</th>
<th>Average No. of flies per observation</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.5 - 0.7</td>
<td>0.6390</td>
<td>1.0063</td>
<td>9</td>
<td>0.00649</td>
<td>568.9</td>
</tr>
<tr>
<td>0.7 - 0.8</td>
<td>0.7555</td>
<td>1.0032</td>
<td>34</td>
<td>0.00598</td>
<td>579.3</td>
</tr>
<tr>
<td>0.8 - 0.9</td>
<td>0.8412</td>
<td>1.0113</td>
<td>25</td>
<td>0.00512</td>
<td>583.6</td>
</tr>
<tr>
<td>0.9 - 1.1</td>
<td>0.9768</td>
<td>1.0379</td>
<td>12</td>
<td>0.00603</td>
<td>591.6</td>
</tr>
<tr>
<td>Mean environmental variance</td>
<td>0.00578 ± 0.000561</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Correlation between homozygote viability and corresponding heterozygote $V_{EN}$</td>
<td>$-0.103 \pm 0.113$</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Correlation between heterozygote viability and their environmental variance</td>
<td>0.196 ± 0.111</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

The new method (without logarithmic transformation) was used. The number of crosses = 80.
The effects of environment on $Cy/+_i$ and $Cy/+_j$ are not always the same. This might be the cause of the small $V_{EN}$ of heterozygotes (an extension of one reviewer's comments). In order to examine this hypothesis, the following calculation was made: For the sake of simplicity, the viability of $+_i/+_j$ individuals is expressed as $\frac{1 + \alpha}{1 + \frac{\beta}{2} + \frac{\gamma}{2}}$, where $1 + \alpha$, $1 + \beta$, and $1 + \gamma$ stand for the relative numbers of $+_i/+_j$, $Cy/+_i$, and $Cy/+_j$ individuals, respectively. Assuming that $\alpha$, $\beta$ and $\gamma$ are much smaller than 1, the relative viability of $+_i/+_j$ can approximately be expressed as

$$v \approx 1 + \alpha - \frac{\beta}{2} - \frac{\gamma}{2}.$$  

Thus, the environmental variance ($V_{EN}$) may be expressed as follows:

$$V_{EN} = V(\alpha) + \frac{1}{4} V(\beta) + \frac{1}{4} V(\gamma) - \text{Cov}(\alpha, \beta) - \text{Cov}(\alpha, \gamma) + \frac{1}{2} \text{Cov}(\beta, \gamma).$$  

(14)

If a mutant gene ($A_2$) is completely dominant or recessive to its alternative allele ($A_1$) with respect to the effects of environment on viability or fitness in varied environments, it is impossible to maintain the mutant gene ($A_2$) together with the $A_1$ gene in polymorphic state (Levene 1953). If the effects of environment on the fitnesses or viabilities of genes are additive, then, under certain conditions, the $A_2$ genes can be maintained together with the $A_1$ genes, since the environmental variance of the heterozygotes becomes much smaller than that of either homozygote in varied environments taken into consideration simultaneously (Gillespie and Langley 1974). Thus, it is necessary to examine the difference in the following components ($V'_{EN}$) of formula (14) between homozygotes and heterozygotes under the assumption of additivity:

$$V'_{EN} = \frac{1}{4} V(\beta) + \frac{1}{4} V(\gamma) - \text{Cov}(\alpha, \beta) - \text{Cov}(\alpha, \gamma) + \frac{1}{2} \text{Cov}(\beta, \gamma).$$  

(15)

In the case of homozygotes where $\alpha = +_i/+_i$, $\beta = Cy/+_i$ and $\gamma = Cy/+_i$, $V'_{EN}$ (Homo) turns out to be:

$$V'_{EN} \text{ (Homo)} = V(Cy) - 2\text{Cov}(Cy, +_i) - 3V(+_i),$$

where $V(Cy)$ and $V(+_i)$ are environmental variance of $Cy$ and $+_i$ chromosomes, respectively, and $\text{Cov}(Cy, +_i)$ stands for the covariance between the environmental effects on $Cy$ and that on $+_i$ chromosomes.
In the case of heterozygotes where \( \alpha = +i/+, \beta = Cy/+, \text{ and } \gamma = Cy/+j, \) 

\[ V'_{EN} \text{(Hetero)} \] becomes as follows:

\[ V'_{EN} \text{(Hetero)} = V(Cy) - 2Cov(Cy, +) - \frac{3}{2} \left[ \text{Cov}(+i, +j) + V(+) \right], \]

where Cov\((+i, +j)\) indicates the covariance between the environmental effect on \(+i\) and that on \(+j\) chromosomes. It was assumed that \( V(+i) = V(+j) = V(+) \) and Cov\((Cy, +i) = \text{Cov}(Cy, +j) = \text{Cov}(Cy, +).\)

The difference between \( V'_{EN} \text{(Homo)} \) and \( V'_{EN} \text{(Hetero)} \) or \( D \) is:

\[ D = \frac{3}{2} \left[ \text{Cov}(+i, +j) - V(+) \right] \leq 0 . \] (16)

Thus, we must reject the hypothesis that the smaller environmental variance of heterozygotes is due to the incomplete dominance of \( Cy \) chromosomes. In fact, the effects of the denominator in the formula for viability act unfavorably to the detection of stability of heterozygotes \((+i/+j)\) in comparison with homozygotes \((+i/+i \text{ or } +j/+j)\) in varied environments. Thus, when the difference in environmental variance between homozygotes and heterozygotes is examined, the \( Cy \) method can be used.

Finally, it should be added that the additivity of environmental effects on genes makes the environmental variance of heterozygotes smaller than of homozygotes. The expected ratio is 1 : 2. The agreement of the observed ratio of environmental variances of heterozygotes and homozygotes for the chromosomes from the natural population (see Table 1) may indicate that the actual values of formula (15) are very small for both homozygotes and heterozygotes in comparison with \( V(\alpha) \) of homozygotes and heterozygotes or \( V(+i/+i) \) and \( V(+i/+j)\).

Second, the effect of constant \( M \) is considered. Under the condition of \( 3 \gg 1/M, \) formula (7), sampling variance in the case of the viability index based on the ratio of two independent binomial terms, becomes as follows:

\[ V_s \approx \frac{4n}{m^2} \left( m + n + 1 \right) \left[ 1 - \frac{1}{M} \left( 3mn + m + 2n \right) \right]. \] (17)

When \( M \) approaches infinity, \( V_s \) becomes \( 4n(m+n+1)/m^2, \) which is almost the same as formula (11), sampling variance in the traditional method without taking logarithms. Therefore, if there is a variation in \( M \) values, it affects the \( V_s \) value. This effect was numerically examined. Let us define \( R_A = \{1 - (3mn + m + 2n)/[M(m+n+1)]\}. \) Using this value, the effect of the variation of \( M \) on \( V_s \) as well as the ratio of \( V_s \) estimated by the new method to that of the old method can be estimated. Following the actual data, let us assume \( m = 250 \) and \( n = 100 \) (constant) and \( M \) is variable from 500 to 900, or 750 to 1350 in the total number of eggs per observation. The calculated \( R_A, \dot{V}_s \) and \( \dot{V}_{EN} \) values are shown below. The observed error variances for viability \( [\dot{V}_s] \) were assumed to be 0.020 and 0.012 for homozygotes and heterozygotes:
It may be concluded from the above results that the effect of variance of \( M \) (or the number of eggs produced by parental flies) on the estimate of \( \hat{V}_{EN} \) is not serious at all in the present experiment.

Incidentally, the table above shows the difference between the results estimated by the present method and the traditional method. The figures in parentheses show the ratios of the former to the latter. It is clear that the latter overestimates the sampling variance. Consequently, it underestimates the environmental variance \( (V_{EN}) \).

The assumption that the initial ratio of \( Cy/+ \) and \(+/+\) eggs was 2 : 1 was next examined. The square of this ratio (2) appears as the factor 4 in formula (7). If the numbers of the \( Cy/+ \) and \(+/+\) flies are approximately binomially distributed, then the ratio is also distributed in some way. Thus, the factor 4 in formula (7) is not a constant. In the actual experiment, there were four or more simultaneous replications. We calculated \( V_s \) for each replicate and after that the average of these \( V_s \) values was calculated, and used for that line or cross (see MATERIALS AND METHODS). Thus, the factor in formula (7) was effectively the average of the squares of the segregation ratios of \( Cy/+ \) and \(+/+\) eggs \( r \) within lines or crosses, so \( \bar{r}^2 = \bar{r}^2 + V(r) \) where \( V(r) \) is the variance of \( r \) and the expected value of \( \bar{r}^2 \) is 4. Since \( V(r) \) is positive, the factor in formula (7) becomes effectively larger than 4, but it is expected that \( V(r) \) is very small because the number of eggs for one observation is about 1000.

For the above reasons, the effects of these weaknesses are not serious at all in the present experiment. Thus, in general, if we can estimate the approximate number of eggs in a culture, the new method is much better than the traditional one.

**Developmental homeostasis and the maintenance of genetic variability in natural populations:** The main findings in the present investigation are as follows: (1) the difference in environmental variances between homozygotes and heterozygotes could not be seen when a chromosome population with variation due to new mutations alone was tested. (2) On the contrary, when a chromosome group isolated from an approximate equilibrium population was examined, heterozygotes manifested a smaller environmental variance than did homozygotes of approximately the same viability. (3) Although, in general, there is a
slight negative correlation between viability and environmental variance, the opposite results were found when viabilities were high, especially when overdominance (coupling overdominance, Mukai 1969a) was manifest.

On the basis of findings (1) and (2), it may be concluded that developmental homeostasis on viability due to heterozygosity is a product of natural selection as stated by Dobzhansky and Levène (1955). The analyses of the heterozygous background (intra-populationally heterozygous genetic background) showed that the addition of heterozygosity to a coadapted genic system with heterozygosity slightly less than the average heterozygosity in the population actually decreased the homeostatic power of the individuals (the correlation coefficient $= -0.103 \pm 0.113$). From these results and findings (3), it may be concluded that developmental homeostasis on viability is not necessarily due to heterozygosity, per se. Viability itself is one of the factors determining the homeostatic power of its carrier. Thus, when a comparison in developmental homeostasis is made between homozygotes and heterozygotes, individuals of these two types with approximately equal viabilities should be compared as in the present analyses.

What should be considered next is how natural selection induces the nature of developmental homeostasis in heterozygotes. Dobzhansky and Levène (1955) considered developmental homeostasis as one of the manifestations of the coadaptation of genic systems. However, the present experimental results clearly indicate that developmental homeostasis for viability and overdominance, which are said to be manifestations of coadaptation (Dobzhansky 1950), are not always two phases of the same coadaptation. In fact, when coupling overdominance was observed, the environmental variance increased (Table 3). However, it may be said that, regardless of the connection of coadaptation with developmental homeostasis, the frequencies of heterozygously homeostatic alleles should increase within loci after these alleles arise through spontaneous mutations, since the allele frequencies of newly arisen mutations in a population are low and since only a small fraction of them are supposed to be homeostatic in heterozygous condition. (Remember that newly arisen mutant alleles scarcely showed developmental homeostasis in heterozygous condition, either in a homozygous genetic background (Table 2) or in a heterozygous genetic background whose heterozygosity was a little below the average of the population heterozygosity (Table 4)]. Indeed, Levène (1953) and Gillespie and Langley (1974) theoretically showed that such heterozygously homeostatic mutant alleles have chances to increase their frequencies through natural selection and would be maintained in a population balanced with the other alternative allele. This is one type of diversifying selection (Dobzhansky 1970) due to the variation of microenvironmental conditions.

In an equilibrium population, the proportion of heterozygous loci where genetic variability has been maintained by the above type of diversifying selection is presumed to be low for the following reason. Using the formula of additive genetic variance ($\sigma^2_a$): $\sigma^2_a = 2pq(h(p-q)+q)^2s^2$, where $p$ and $q$ are the allele frequencies of $A$ and $a$ genes and $h$ and $s$ are the degree of dominance and the selection coefficient, respectively, the numbers of loci responsible for the observed additive variance for viability $[\hat{\sigma}^2_a = 0.01$ in the Raleigh, N. C. population
(Mukai et al. 1974) were estimated for $q = 0.10, 0.20$ and $0.30$; $s$ and $h$ were assumed to be $0.03$ and $0.5$ respectively (Mukai et al. 1972). The results are $105, 138$ and $246$ loci, respectively. Thus, a relatively small number of loci where diversifying selection is operating can explain the observed genetic variability in the N. C. population; but if the value of $s$ is small, the number of loci increases. It is a further problem to investigate whether or not the coadaptation of a genic system (or genetic background) is necessary for the manifestation of developmental homeostasis at a few loci.

All the above discussions were made assuming that developmental homeostasis for viability was induced entirely by the heterozygosity of viability polygenes, most of which are supposed to be different from structural genes (Mukai and Cockerham 1977). It may be proposed as an alternative hypothesis that viability polygenes are not concerned with developmental homeostasis for viability but the heterozygosity of structural genes is (Powell 1971; Powell and Wistrand 1978; McDonald and Ayala 1974; Takahata, Ishii and Matsuda 1975). The results in Tables 1 and 2 may support this hypothesis, since the heterozygosity in heterozygotes between chromosomes with variation due to newly arisen mutations alone did not manifest the nature of developmental homeostasis for viability, contrary to the results for heterozygotes between chromosomes extracted from a natural population. In the former heterozygotes, the average heterozygosity at the structural loci is much lower than that in the latter heterozygotes. In fact, the minimum spontaneous mutation rate of viability polygenes is $0.14-0.17/\text{second chromosome/generation}$ (Mukai 1964; Mukai et al. 1972), while the maximum mutation rate of structural genes was estimated to be $0.04/\text{second chromosome/generation}$ (Mukai and Cockerham 1977). The above hypothesis was examined by analyzing natural populations carefully and the result is not favorable to the above hypothesis. Details will be published in the following paper of this series (Mukai and Nagano unpublished results).

In conclusion, it can be said that developmental homeostasis for viability in heterozygotes in microenvironmentally varied conditions of random mating populations is a product of natural selection due to the increase in gene frequencies of heterozygously homeostatic genes, and it is speculated that developmental homeostasis is working at a relatively small fraction of the loci.

**LITERATURE CITED**


Corresponding editor: W. W. Anderson