GENETIC ANALYSIS OF NATURAL POPULATIONS OF *DROSOPHILA MELANOGASTER* IN JAPAN. II. THE MEASUREMENT OF FITNESS AND FITNESS COMPONENTS IN HOMOZYGOUS LINES

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ABSTRACT

Fifty lethal-free, sterility-free isogenic lines of *Drosophila melanogaster* that were randomly sampled from a natural population were tested for net fitness and other components of fitness by competition with *D. hydei*. Larval viability and developmental time were also measured using the balanced marker method. Distribution patterns of these fitness components were similar, but correlation between the fitness components varied depending on the combinations used. The highest correlations were obtained between net fitness and productivity (r_p = 0.6987, r_g = 0.9269). The correlation between net fitness and total larval viability was much lower (r_p = 0.1473 and r_g = 0.2171). These results indicate that measuring net fitness, not just a component of fitness, is necessary for the good understanding of the genetic structures of natural populations.

IN previous studies, the fitness of six different strains of *Drosophila melanogaster* was measured and strong evidence was obtained that the total competitive ability of *D. melanogaster* and *D. hydei* was a reliable estimate of net fitness (YAMAZAKI 1984). Laboratory strains of these fruit flies, which had been kept in captivity for many years, were used in these experiments, and, therefore, extrapolation from these results to natural populations was tenuous. In this study, net fitness and its components were examined using two different experimental approaches (interspecific competition and the balanced marker method) in 50 nonlethal, nonsterile, isogenic strains randomly sampled from a natural Japanese population of these flies.

In 1970 SVED and AYALA studied 18 strains homozygous for the second chromosome in *D. pseudoobscura* by mixing *Ba* and + chromosomes in populations. They traced the frequency change of + chromosomes. Their results showed that the frequency distribution of fitness was quite different from the bimodal distribution usually obtained from viability test measured by the balanced marker method. Similar results were obtained in *D. melanogaster* (SVED 1971, 1975).

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The main objective of the present experiments was to find whether there was a difference in the frequency distribution of fitness or fitness components obtained by different methods and to determine the extent of the correlation between these estimates in flies sampled from natural populations.

MATERIALS AND METHODS

In 1977, 430 wild-type adult male *D. melanogaster* were collected at Akayu, Japan, and 140 isogenic lines homozygous for both the second and third chromosomes were established by the balanced marker method. These lines carried neither lethal nor sterile genes on any of the major autosomes. The details concerning the establishment of these lines are described in YAMAZAKI *et al.* (1984). Fifty isogenic lines were selected at random from these lines for this experiment.

Both net fitness and components of fitness were measured by interspecific competition experiments using *D. hydei* as a standard. Details of procedures or definitions of these fitness or fitness components were described in a previous study (YAMAZAKI 1984). Two replication experiments were run with each strain. In total, 100 populations were established by the same method as the previous studies. Experiments were continued until 21 transfers were completed (84 days from the start of a population), regardless of the results.

Net fitness (or total competitive ability) and three fitness components, adult viability, productivity and developmental time, were measured by this competition experiment. At the same time, larval viabilities of both the second and third chromosomes were measured by the balanced marker method (WALLACE 1956); *Cy*/+ females and males obtained from the cross between *Cy*/*Pm* females and +/+ males were mated, and their progeny were scored for second chromosome viability. A standardized percentage of wild-type progeny (the percentage of observed wild type was divided by the expected wild-type percentage) was used as an index of larval viability. To compute third chromosome viabilities, *Sb Ser*/*Pr* third chromosome balancer was used. Total larval viability was obtained by multiplying the viability of the second and third chromosomes, although it is not well understood what kind of interaction exists in the larval viability of these two chromosomes. Several conflicting results are reported about the way different chromosomes interact and affect larval viability: positive synergistic interaction (SPASSKY, DOBZHANSKY and ANDERSON 1965; TEMIN *et al.* 1969; KOSUDA 1971) or negative synergistic interaction (SEAGER and AYALA 1982). In this analysis we simply multiplied the viability of each chromosome, as a first approximation, assuming that both viabilities act independently.

Flies were counted 10, 12, 14 and 17 days after the mating of the parental flies. Average developmental time of +/+ flies relative to that of the balanced marker *Cy*/+, in the case of the second chromosome, and *Sb Ser*/+, in the case of the third chromosome, was used as a developmental time index (MUKAI and YAMAZAKI 1971; YAMAZAKI 1971). Values of less than one indicate that the developmental time is faster than the standard marker flies, and a value larger than one indicates a slower developmental time. Two different estimates of developmental time were thus obtained in this study: one from interspecific competition experiments and one from the balanced marker method (*Cy* method).

RESULTS

Approximately 350,000 flies were counted to estimate these values. In most lines ten different estimates of net fitness or fitness components were measured, including four estimates for each of the major chromosomes. Larval viability and developmental time at each of the two chromosomes were measured in order to obtain total larval viability and total developmental time by multiplication, respectively. Therefore, a total of six fitness or fitness components were compared: net fitness, adult viability, productivity, developmental time, total larval viability and total developmental time.

Figure 1 shows the frequency distribution of each fitness or fitness compo-

TABLE 1

Statistics relating to fitness and its components of Akayu population: mean values for fitness and its components and their analyses of variances

Fitness or its components	Mean	Mean square			F	Genotypic variance
		Total (98)	Between-lines (49)	Within-line (49)		
Net fitness	−0.217	0.0238	0.0309	0.0168	1.84*	0.00705
Adult viability	0.837	0.0225	0.0257	0.0194	1.32	
Productivity	0.639	0.0854	0.1203	0.0504	2.39**	0.03495
Developmental time	0.714	0.0021	0.0017	0.0025	<1	
Total viability	0.664	0.0392	0.0691	0.0094	2.62**	0.02985
Total development	0.998	0.0009	0.0011	0.0007	1.57	

Numbers in parentheses are degrees of freedom.
* Significant at the 5% level.
** Significant at the 1% level.

nent of the 50 Akayu strains. The average values of each strain were used for the figure. Each distribution is positioned so that the amount of dispersion or range is approximately the same in the figure. The abscissa shows the fitness index. Thus, each abscissa is different in each fitness distribution. The ordinate is the number of strains for which fitnesses are contained within the fitness range specified in the abscissa. The other statistics necessary for the analysis of data are shown in Tables 1 and 2.

Several conclusions can be drawn from these results.

1. Distribution patterns of each fitness component are not far apart. They seem to be fairly similar in the sense that they are unimodal (see Figure 1). Therefore, no transformation of data was conducted. The unimodal distribution was obtained probably because all of the lines examined carried no lethal or sterile genes.

2. If the fitness estimated approaches the net fitness or includes more components of fitness, the distribution appears to scatter more and tends to shift to the left. Thus, a large percentage of strains approached the lower fitness classes, as the fitness estimated approached net fitness (Figure 1). A similar trend was observed by SVED (1971, 1975) from the competition of *Cy*/+ and +/+ in *D. melanogaster*. He concluded that most of the lines were lethal or semilethal when net fitnesses were measured.

3. Significant differences in fitness components, such as net fitness, productivity and total larval viability, were found among different strains. Developmental time, adult viability and total developmental time did not present significant differences among strains. The basic statistics for this analysis are shown in Table 1. These results indicate that there is little genetic variability for developmental time or adult viability. These results do not necessarily mean that adult viability or developmental time are generally not important for determining fitness. We can say, however, that these two factors are not important in determining net fitness in the present experimental system. The absence of significant line effects in developmental time and adult viability may

TABLE 2

Statistics relating to fitness and its components of Akayu population: analyses of covariances for fitness components and the estimates of their phenotypic and genotypic covariances and correlations

Fitness combinations	Covariance				Correlation	
	Total (98)	Between-lines (49)	Within-line (49)	Genotypic	Phenotypic	Genotypic
Net fitness—Productivity	0.0315	0.0461	0.0170	0.01455	0.6987**	0.9269 ±0.1264
Net fitness—Total viability	0.0045	0.0077	0.0014	0.00315	0.1473	0.2171 ±0.2288
Productivity—Total viability	0.0122	0.0219	0.0026	0.00965	0.2109*	0.2988 ±0.1940
Net fitness—Adult viability	0.0116	0.0153	0.0079	0.00370	0.5013**	
Net fitness—Developmental time	0.0005	0.0005	0.0005	0.00000	0.0707	
Net fitness—Total development	−0.0007	−0.0016	0.0002	−0.00090	−0.1512	
Adult viability—Productivity	0.0175	0.0242	0.0109	0.00665	0.3992**	
Adult viability—Developmental time	−0.0006	−0.0007	−0.0005	−0.00010	−0.0873	
Adult viability—Total viability	0.0011	0.0023	−0.0001	0.00120	0.0370	
Adult viability—Total development	−0.0007	−0.0012	−0.0001	−0.00055	−0.1556	
Productivity—Developmental time	0.0011	0.0012	0.0009	0.00015	0.0821	
Productivity—Total development	−0.0018	−0.0034	−0.0001	−0.00165	−0.2053*	
Developmental time—Total viability	−0.0011	−0.0018	−0.0003	−0.00075	−0.1212	
Developmental time—Total development	−0.0000	−0.0001	0.0001	0.00000	−0.0000	
Total viability—Total development	−0.0001	−0.0001	0.0000	−0.00005	−0.0168	

Numbers in parentheses are degrees of freedom.
* Significant at the 5% level.
** Significant at the 1% level.

be partly due to the small number of replications (two per strain) used in this experiment. In the previous experiments, in which eight replicate populations were run in each of six strains examined, significant line effects were obtained in adult viability (YAMAZAKI 1984).

4. Table 2 shows several phenotypic correlations and genotypic correlations between two fitness components together with other statistics necessary for the calculation of the earlier correlation. Interesting results emerge from these values. The method for calculating these correlations are shown in YAMAZAKI

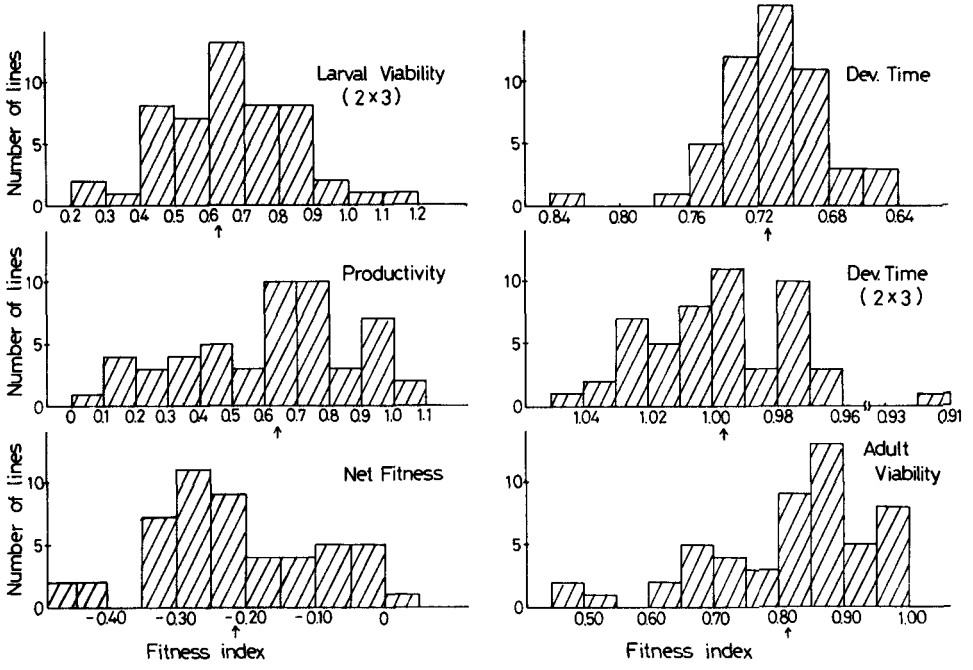

FIGURE 1.—Distribution patterns of fitness or fitness components of 50 isogenic lines in the Akayu population. Arrows indicate the mean of the distribution.

(1984). Although the distribution patterns of each fitness index were similar, as shown in Figure 1, the content was different. This is clearly shown in the low correlation coefficients between two fitness indices or fitness components (Table 2). Phenotypic correlations were as follows: between net fitness and productivity (0.70, Figure 2), between net fitness and adult viability (0.50), between productivity and adult viability (0.40), between net fitness and total larval viability (0.15, Figure 3), between productivity and developmental time (0.08), between net fitness and developmental time (0.07), between adult viability and developmental time (−0.09) and between net fitness and total developmental time (−0.15). Genotypic correlations were obtained only between fitness components with significant line effects: 0.93 between net fitness and productivity, 0.22 between net fitness and total larval viability and 0.30 between productivity and total larval viability.

DISCUSSION

All strains used in the present experiment were free from either lethal or sterile genes in both the second and third chromosomes. Therefore, apparent equality of the distribution between net fitness and components of fitness in Figure 1 does not necessarily mean that they are similar when lethal or sterile genes are included. In fact, viability tests of 360 isogenic lines (from which 50 lethal-free, sterility-free homozygous lines were chosen for this study) showed that 48% were lethal in either second or third chromosomes and the frequency

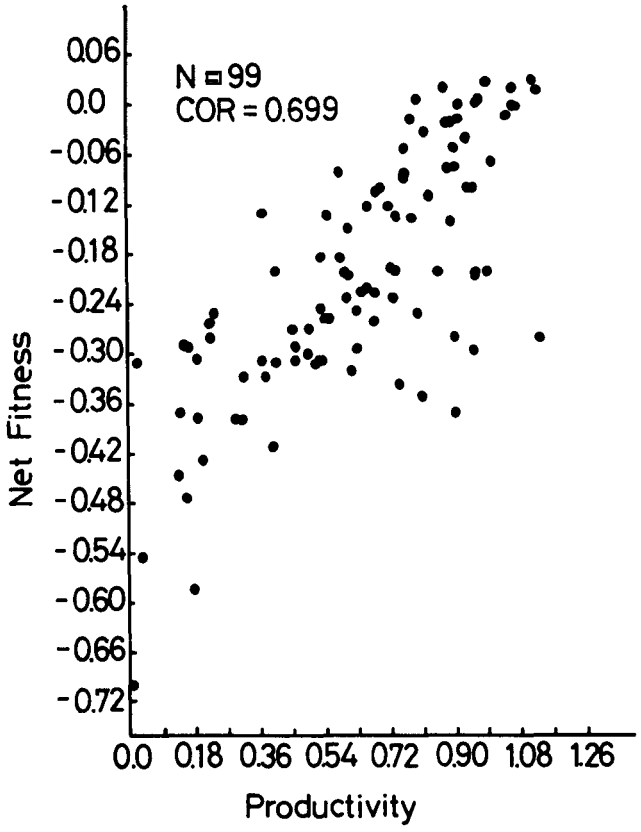

FIGURE 2.—Phenotypic correlations (COR) between net fitness and productivity of 50 isogenic lines in the Akayu population.

of sterility was 34% (only lethal-free strains were examined in the present study). The number of strains with effectively zero fitness (net fitness index < -0.4) was seven of 50 lethal-free, sterility-free strains in the interspecific competition experiments (13%). Therefore, approximately 70% ($1 - 0.52 \times 0.66 \times 0.86$) of the strains examined had zero fitness (in the sense that they left few progeny to the next generation) if all components of fitness or net fitness are taken into consideration. These results are reasonable considering that viability is only a part of net fitness.

However, the similarities in distribution of several fitness components are quite misleading. Both phenotypic and genotypic correlations are not as high as might be expected ($r_p = 0.1473$, $r_s = 0.2171$) between total larval viability and net fitness, as shown in Tables 1 and 2. Namely, these two distributions are superficially similar to each other but are different in content. The highest correlation was obtained between net fitness and productivity ($r_p = 0.6987$ and $r_g = 0.9269$). Productivity includes a larger part of net fitness (larval viability + female fecundity + male mating ability + adult viability) than simply total larval viability. Note that the comparison involving developmental times or

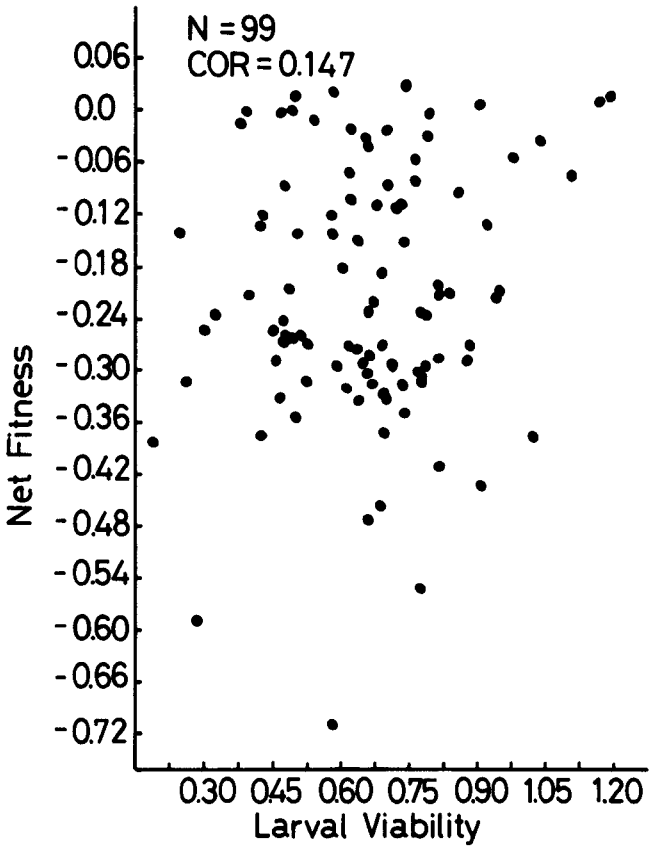

FIGURE 3.—Phenotypic correlation (COR) between net fitness and total larval viability of 50 isogenic lines in the Akayu population.

adult viability is not as meaningful as might be supposed from the values in the table, because the difference among strains was not significant.

These low correlations between net fitness and components of fitness may be interpreted by either one of the following models at the genic level.

1. Most genes that affect fitness have different effects on each of the fitness components (negative correlation): the genes beneficial to the carriers with respect to one fitness component, for instance, viability, may be detrimental with respect to, fertility for example (SIMMONS, PRESTON and ENGELS 1980).

2. Only a tiny fraction of genes influence more than one component of fitness. The majority affect only a simple component of fitness. In other words, there are viability genes or developmental time genes at different loci.

If the first model is true, measuring a component of fitness, such as viability or fertility, is not sufficient to understand the genetic structure of natural populations. Net fitness must be measured, since the genetic structure of equilibrium populations cannot be predicted from a component of fitness. Genetic structures or allelic frequencies of genes affecting one component of fitness, at equilibrium populations, can be estimated just from the knowledge of one

component of fitness if model 2 is correct. Under the latter assumption, estimates of one component of fitness can be extended rather safely to natural populations as far as that component of fitness is concerned.

The difference in models 1 and 2 cannot be distinguished from the present experiment. These results at least demonstrate that the majority of fitness-affecting genes are not pleiotropic in the same direction. It is safe to say that net fitness of individuals cannot be estimated accurately by extrapolating from the estimate of a single component of fitness.

To measure fitness accurately is a difficult task. Precision can be increased just by increasing the scale of the experiments. However, to increase the accuracy is a different and more complex problem. To increase accuracy, we must know the way selection works in nature. It is also known that the conditions under which fitness is estimated influence the results obtained (SNYDER and AYALA 1979; MUELLER and AYALA 1981; CLARK and FELDMAN 1981). Population geneticists, especially Drosophila population geneticists, often measure only a component of fitness with the hope that the component of fitness is positively correlated to net fitness. Most commonly, viability is used as an estimate. Fertility or sexual selection is reported to be very important to net fitness (SVED 1971; BUNDGAARD and CHRISTIANSEN 1972; BRITTNACHER 1981). In this experiment, it has been shown that net fitness could not be estimated from the measurement of only one component of fitness (PROUT 1971). Additional experimental evidence supports this conclusion; only the net fitness, but not the components of fitness, is tightly correlated with inducibility of the amylase locus, showing that components of fitness do not properly represent real net fitness (YAMAZAKI and MATSUO 1983, 1984).

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