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A quantitative genetic analysis of fitness and its components in *Drosophila melanogaster*

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Summary

Forty-one third chromosomes extracted from a natural population of *Drosophila melanogaster* were assessed for net fitness and for the quantitative characters viability, net fertility, female productivity, male weight, abdominal bristle number, and sternopleural bristle number. Net homozygous and heterozygous fitness of the third chromosomes was estimated by competition against a marked balancer third chromosome. Average fitness of the homozygous lines relative to wild-type heterozygotes was 0·13, indicating substantial inbreeding depression for net fitness. All significant correlations of quantitative characters with fitness and with each other were high and positive. Homozygous fitness is strongly correlated with net fertility, viability, and female productivity, moderately associated with male weight, and not significantly associated with bristle traits. The combination of metric traits which best predicts homozygous fitness is the simple multiple of viability and female productivity. Heterozygous fitness is not correlated with homozygous fitness; furthermore, the relative contribution of metric traits to fitness in a heterozygous population is likely to be different from that deduced from homozygous lines. These observations are consistent with a model of genetic variation for fitness in natural populations caused by segregation of rare deleterious recessive alleles.

1. Introduction

The theory of natural selection describes the process of adaptive evolution in terms of variation between individuals in their capacity to contribute progeny to future generations. Individuals in a population may be assigned empirical fitness values which are the numbers of offspring represented in the following generation. Darwin postulated that if individuals vary in their fitness values, and if part of this variation is inherited, the population will evolve to be comprised of disproportionately more of the fitter types by the process of natural selection. This theory of evolution is, however, somewhat tautological, since fitness values are determined retrospectively, after the individuals have contributed to the following generation. For evolutionary theory to be predictive we need to examine further the relationship between individual fitness and the genetic variation underpinning it.

Immediately complications arise. Fitness of an individual, although it can in principle be assigned a single value, may be considered also as an assemblage of its component metric characters, each of which may be separately evaluated (Falconer, 1981). Natural selection for fitness is thus analogous to artificial selection for a multiple-trait selection index, constructed such that several characters are simultaneously selected, with appropriate weightings. Prediction of response to natural selection then requires identification of all the characters which together comprise fitness, their relative weightings (which may be determined by the particular environment), and the phenotypic and genetic variances and covariances of these characters with fitness and with each other. Construction of such an index would enable the prediction of not only the response to natural selection of the population in different habitats, among which the relative weighting of the components may vary, but also the evolution of the component characters as a correlated response.

The first stage in the construction of the index is identification of the components. Robertson (1955) has suggested that the spectrum of quantitative traits may conceptually be divided into three broad categories, based on the magnitude of their genetic correlation with the composite character, fitness. Characters which are major components will have high positive genetic correlations with fitness, since genes that increase the value of the character also increase fitness. At the other extreme, genetic variation for neutral characters is unrelated to genetic variation for fitness; genetic correlations between these traits and fitness will be zero. Many characters may exhibit a

third pattern of relationship to fitness, that of an intermediate optimum, whereby intermediate values of the character confer maximum fitness.

The shape of such fitness functions for a variety of metric traits has largely been inferred from three lines of indirect evidence: magnitude of the heritability of the character, pattern of response to artificial selection, and change of mean value on inbreeding. The continued action of natural selection on any trait will ultimately result in the reduction of genetic variance of that trait, as selection fixes favourable and eliminates undesirable alleles. Therefore traits which are major components of fitness will typically exhibit low levels of genetic variability because of the past action of natural selection; conversely, neutral characters will retain relatively greater amounts of additive genetic variance. The magnitude of the ratio of the additive genetic to total phenotypic variance (heritability) of a trait may thus indicate any past association of the character with fitness, but caution need be exercised when making this inference, as selection is not the only force which can reduce genetic variance. The response to artificial selection experiments may also be informative. For the reasons given above, major components of fitness will not readily respond to artificial selection. However, mean values of characters which are either neutral or have an intermediate optimum with respect to fitness will alter under selection pressure. If artificial selection is suspended prior to the elimination of all additive genetic variance, only natural selection then operates on the trait, and any subsequent change of mean is diagnostic. If the relation of the character to fitness is one of an intermediate optimum, and the artificial selection has perturbed the population mean away from that optimum, on relaxation of selection the population mean should revert towards the original optimum value under the influence of natural selection. The mean of a neutral character, however, would not change from the artificially selected value. Finally, observations of the change in mean value of a character under inbreeding are useful. Inbreeding increases homozygosis, and thus reduces fitness, either by unmasking rare deleterious recessive alleles previously protected from the action of natural selection, or by breaking apart favourable heterozygous associations. The proportionate reduction of mean of a character on inbreeding is therefore an indication of the association of the character with fitness, since major components will be greatly reduced, whereas neutral characters will show little or no change.

How may fitness profiles be determined directly? Any of the standard techniques of quantitative genetics enables the estimation of phenotypic and genetic variances and covariances among any number of measurable metric traits; the problem lies in independently measuring the composite trait, fitness. The difficulty is apparent if we regard fitness as the simple product of the two major components, viability and fecundity; variation in these components accounts for all the variation in fitness. How do we measure variation in viability among individuals, when we can only observe the survivors? The solution generally adopted has been to consider fitness differences with respect to a single locus, so that specific genotypes may be replicated and many individuals of each genotype examined to obtain average viabilities and fertilities for the different genotypic classes. Therefore, to assess variation in total fitness, we need to replicate whole genotypes of a number of individuals, so that each entire genotype is represented by an average measure of viability and fertility. Such clones are difficult to obtain in sexually reproducing organisms, but techniques of chromosomal manipulation in *Drosophila* allow at least a partial resolution of the problem. By crossing wild-caught males to females of specially constructed balanced marker strains, it is possible to replicate single whole chromosomes derived from natural populations. This technique has been extensively applied to the study of the viability component of fitness in *Drosophila* (for a comprehensive review of this work see Simmons & Crow, 1977), and has been modified to provide an elegant multi-generation measurement of total fitness by Sved & Ayala (1970), Sved (1971).

The objectives of the following experiment are therefore to utilize the technique of Sved and Ayala for the measurement of fitness in order to assess the nature of genetic variation for fitness in *Drosophila*, to determine directly the relationship of a number of quantitative characters to fitness, and finally to predict fitness values as a function of easily measured metric traits.

2. Materials and Methods

(i) Drosophila *Stocks*

A sample of a *D. melanogaster* population collected from Death Valley was kindly donated by Dr L. Nunney of the University of California, Riverside. Approximately 200 males and females were used to initiate a population cage, which was allowed to reach an equilibrium population density before the commencement of the experiments. These animals did not cause hybrid dysgenesis (Kidwell, Kidwell & Sved, 1977) when crossed to a standard M (Canton S) strain, provided by Dr M. Kidwell; therefore crosses of the wild strain to the standard laboratory strain (also of M cytotype) to replicate individual chromosomes were not complicated by dysgenic phenomena.

The chromosome replication procedure involves crossing individual males of the wild-caught population to females which are heterozygous for two different markers, one on each homologue of the chromosome of interest. The marker stock used for these experiments was the third chromosome balancer, *TM3/ruse*. The *TM3* complex (Lindsley & Grell, 1968)

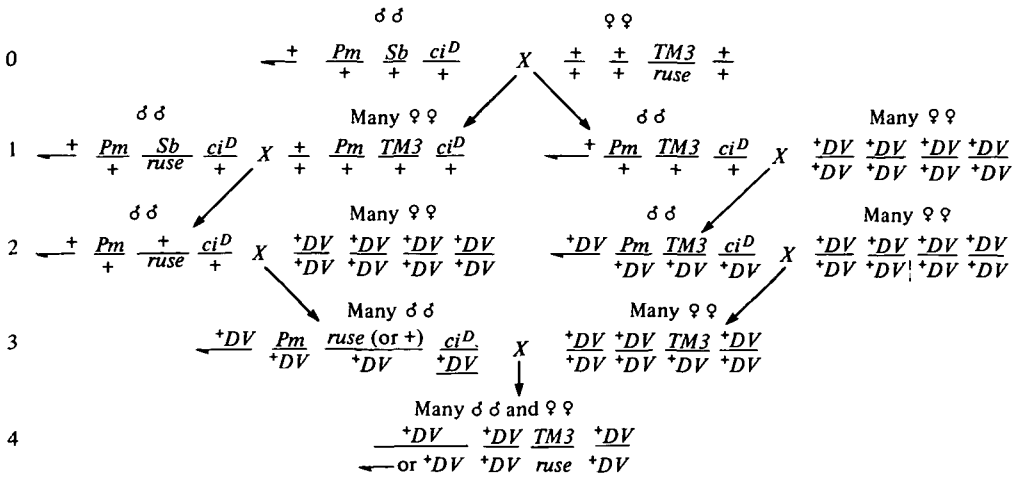

Fig. 1. Crossing scheme employed to obtain a third chromosome marker stock in the Death Valley (*DV*) background. For a description of the mutant phenotypes, see Lindsley & Grell (1968).

has a single long inversion covering much of the chromosome, and carries the dominant markers *Sb* (stubble bristles) and *Ser* (serrate wings), as well as the recessive markers *e* (ebony body) and *ru* (rough eyes), and is lethal when homozygous. It is balanced against the multiply-marked *ruse* chromosome, synthesized by A. Robertson, which contains the markers *se*, *st*, *e*, *ca* and *ru*.

The first series of crosses, illustrated in Fig. 1, were designed to create a stock which incorporates the third chromosome markers in an otherwise wild-type Death Valley background. In the second series of crosses, individual males from the Death Valley population were crossed to two females of this stock. A single *TM3*/+ son from each cross was then backcrossed to three females of the marker stock. Six *TM3*/+ male and female progeny of the backcross (of phenotype *Sb*, *Ser*, not *ru*, not *e*) were used to create homozygotes of a single third chromosome descending from the original wild-type male, while preserving as much as possible of the rest of the Death Valley genome, given the constraint of two single male bottlenecks. At this point a second replicate was made of each third chromosome line, by transferring the *TM3*/+ heterozygous parents to a fresh culture after the initial period of egg laying. In the following generation 20 *TM3*/+ males and females were collected from each replicate of each line to be the parental generation of the fitness determination experiment, thereafter population size was kept constant at 40 pairs of parents/replicate/line/generation, as described below. A total of 41 third chromosome homozygote lines, with two replicates of each, were analysed.

(ii) *Culture conditions*

Flies were reared in $\frac{1}{3}$ pint milk bottles, with approximately 100 ml of Edinburgh cornmeal-molasses-agar medium (UFAW Handbook, 1967), unless otherwise specified. All cultures were kept at a constant temperature of 25 °C.

(iii) *Measurement of homozygous fitness*

The logic underlying the technique devised by Sved & Ayala (1970; Sved, 1971) to estimate homozygous fitness of a chromosome is pleasingly simple, and relies on the performance of +/+ homozygotes relative to *TM3*/+ heterozygotes when the *TM3* and + chromosomes are allowed to compete in a population over several generations. Zygotes produced will be of three genotypes, *TM3*/*TM3*, *TM3*/+, and +/+, but since the homozygous *TM3* combination is lethal, only two genotypic classes are observed in the adults. If the wild-type homozygotes are less fit than the marker heterozygotes, fitness is overdominant, so an equilibrium frequency of the + chromosome will be attained, and the observed value of the equilibrium is a function of the fitness of the wild-type homozygote relative to the marker heterozygote. If, on the other hand, the +/+ homozygotes are more fit than the marker heterozygotes, the *TM3* chromosome will be eliminated from the population at a rate dependent on the fitness of the heterozygote relative to the wild-type homozygote.

For the case of overdominant fitness, the function which relates fitness (w) of the homozygote +/+ relative to the heterozygote *TM3*/+, to the observed proportions of adult heterozygotes (h) and homozygotes ($1-h$) at equilibrium, is $w = (r-h)/r(1-h)$ (Sved, 1971). The observed proportion of heterozygotes, r, emerging from a cross of two heterozygotes, enters into the formulation to account for the observations being made on viability-selected adults, rather than zygotes.

Experimental values of h and r were obtained separately for each replicate of each homozygous line according to the following procedure. The parental generations consisted of 20 *TM3*/+ males and females, which were allowed to mate and oviposit for four days, and then removed from the culture. Thirteen days after the parents were first introduced, all the adults in the culture were counted and classified

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according to genotype. The proportion of heterozygotes, h, was calculated for males and females separately, then 40 males and 40 females were selected as parents of the following generation, keeping the value of h for each sex in the selected group the same as in the total sample for that generation. These parents were introduced to fresh cultures exactly 2 weeks after the initiation of the previous generation, so that all lines were maintained as discrete generations with constant generation interval throughout the course of the experiment. Results were recorded for a total of 15 generations. Viability determinations were made separately, at Generations 8 and 9. Twenty virgin $TM3/+$ males and females were collected from each population bottle at Generation 8, and treated according to the regime outlined above. At day 13 all adults in the bottle were counted and classified, and the proportion of heterozygotes, r, calculated. This procedure was repeated at Generation 9. The choice of twenty pairs of parents for the viability test, as opposed to the forty pairs used each generation in the fitness bottles, was governed by the practical consideration of producing sufficient $TM3/+$ animals from many of the cultures. The viability measurements were therefore made under slightly less stringent competitive conditions than those used to estimate h.

(iv) *Measurement of heterozygous fitness*

If the wild-type genotype is more fit than the marker-bearing heterozygote, the marker will be eliminated from the population, at a rate which depends on the relative fitness of the marked to wild-type individuals. This will generally be the case if the wild-type individuals are heterozygous. The rate of elimination can thus be used as a measure of fitness using the method of Anderson (1969), designed specifically for the case in which the marker chromosome is lethal when homozygous.

The recurrence relationship, $Q_{t+1} = Q_t w/ (1+ Q_t(f-2)+2Q_t w)$, expresses the frequency of the lethal chromosome, Q, in two successive generations in terms of the fitness (w) of the lethal-bearing heterozygotes relative to the wild-type heterozygotes. (f is the fertility of the marker heterozygotes, relative to wild-type). Inverting both sides of this expression, $1/Q_{t+1} = ((f-2+2w)/w)+(1/w)(1/Q_t)$ which is of form $Y = a+bX$. If we plot $1/Q_{t+1}$ *vs.* $1/Q_t$. for several generations, the reciprocal of the slope of the regression line is the estimate of fitness. Note that if h is the observed proportion of $TM3$ heterozygotes, $Q = (\frac{1}{2})h$.

Two separate heterozygous fitness determinations were made. To estimate 'total' heterozygous fitness, single $TM3/+$ males and females were collected from each of the 41 homozygous lines, and crossed in pairs according to a mating design which ensured all progeny would be heterozygous for two different homozygous chromosomes. Lines were labelled 1–41 in the order in which they were extracted, i.e. random with respect to homozygous fitness. Then the female of line 1 ($\female_1$) was crossed to the male of line 2 ($\male_2$), $\female_2 \times \male_3$, $\female_3 \times \male_4$, et cetera, the final cross being $\female_{41} \times \male_1$. The 41 pairs of flies were then placed in a culture bottle, and subsequently treated in the same manner as the homozygous fitness lines. The proportion of $TM3/+$ adults emerging each generation was recorded until the $TM3$ chromosome was eliminated from the population, after which 40 wild-type males and females were randomly selected each generation to maintain the culture. Four replicate total heterozygous fitness experiments were set up, two from the first and two from the second replicates of the homozygous lines.

The second heterozygous fitness experiment was designed to assess the relationship between mean homozygous fitness of a set of lines, and the fitness of the same set when heterozygous. The 41 homozygous lines were ranked according to their fitness, and subdivided into six groups of similar homozygous fitness; five groups of seven homozygous lines, and one of six. Within each group, the lines were re-ranked in random order, and crossed as described above for the measurement of total heterozygous fitness. Eight replicate 'partial' heterozygous fitness experiments were set up, four from each of the first and second homozygous line replicates.

(v) *Measurement of quantitative characters*

Values of six metric traits were measured on each replicate of the 41 homozygous lines, and the four replicate 'total' heterozygous lines.

(1) *Viability*. Viability of $+/+$ relative to $TM3/+$ is easily estimated from the observed proportion of heterozygotes, r, emerging from a cross of two heterozygotes. The expected ratio of heterozygotes to homozygotes, $r:(1-r)$, is $\frac{1}{2}:(\frac{1}{4})v$, from which v, the viability, is $2(1-r)/r$. Two separate viability determinations were made at equilibrium for each of the two replicates of each homozygous line, as described above. Heterozygous viabilities were estimated from the proportions of heterozygotes in the first generation of the heterozygous fitness estimation experiments.

(2) *Fertility*. Since total fitness can be resolved into the product of the two major components, viability (v) × fertility (f), an estimate is $f = w/v$, or $(r-h)/2(1-r)(1-h)$. A single estimate of fertility was calculated for each replicate of the homozygous lines.

(3) *Female productivity*. Fertilized wild-type females were collected from each of the replicates after they had attained equilibrium, at Generations 12 and 13 for the homozygous lines, and 8 and 9 for the heterozygous lines. Groups of three females were placed in a culture vial, allowed to lay eggs for four days, and then removed. The total number of progeny which had emerged 13 days after the parents were first introduced was used as the measure of female productivity; 45 such cultures were set up for each replicate. This is not an ideal measure of female fertility, since it confounds

egg production with egg-to-adult survivorship, but it has the advantage of technical convenience over the more appropriate method of enumerating eggs laid during the test period.

(4) *Male weight.* At Generation 14 for the homozygous lines, and 10 for the heterozygous lines, 20 wild-type males were collected from each replicate, aged for three days, and individually weighed to the nearest μg on a Cahn microelectrobalance, while under ether anaesthesia.

(5) *Abdominal bristle number.* Wild-type males and females were collected from each replicate at Generation 11 for the homozygous lines, and 7 for the heterozygous lines. The numbers of bristles on the last abdominal tergite (segment six of females, five of males) was recorded for 20 individuals of each sex.

(6) *Sternopleural bristle number.* Wild-type males and females were collected from each replicate at Generation 10 for the homozygous lines, and 6 for the heterozygous lines. The total number of sternopleural bristles on the left and right sides was recorded for 20 individuals of each sex.

3. Results

(i) *Homozygous fitnesses*

Sixty-one third chromosomes were extracted from the Death Valley population, of which 20 (33%) were lethal when homozygous. The remaining 41 third chromosome homozygotes were measured for fitness and the other quantitative characters.

Fig. 2 depicts the weighted average frequency of *TM3*/+ heterozygotes (h) in successive generations for the homozygous and 'total' heterozygous lines. Weighted averages were computed by pooling observed numbers of *TM3*/+ heterozygotes and +/+ homozygotes each generation across each of the two replicates of the 41 homozygous lines, and across the four replicates of the heterozygous lines, then recalculating the value of h using those total numbers. By Generation 6, the homozygous lines had attained an average equilibrium frequency of *TM3*/+ of approximately 0·52, while the *TM3* chromosome was virtually eliminated from the heterozygous populations.

To calculate homozygous fitnesses for each of the lines separately, equilibrium was therefore conservatively judged to be reached by Generation 8. A weighted average value of h was computed for each replicate of each line by pooling the numbers of *TM3*/+ and +/+ flies emerging over Generations 8–15, and using these total numbers to estimate the equilibrium frequency of heterozygotes. Similarly, weighted average values of r were computed from the total numbers emerging in both viability experiments. These values of h and r were used to estimate homozygous fitness for the replicates of each line, and then pooled across replicates to give a total weighted average fitness

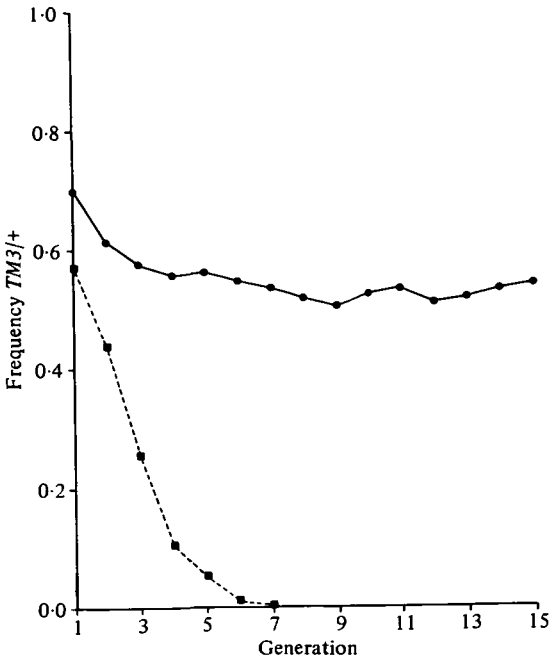

Fig. 2. The solid lines and circles depict the frequency of *TM3*/+ each generation averaged over both replicates of all 41 third chromosome homozygous lines, and the broken lines and squares the frequency of *TM3*/+ each generation averaged over the four 'total' heterozygous lines.

for each of the 41 lines. The final weighted average estimates of r, h, and w for the 41 homozygous lines are given Table 1, with standard errors computed from the variation between replicate estimates.

Major features to note include the good agreement between replicate estimates, as indicated by the small standard errors, and the wide range of the fitness estimates across lines. The precision is partly because of convergence to a deterministic equilibrium value of h characteristic of each chromosome, and partly because the estimates are based on large samples (Table 1 represents over 340000 flies). Nevertheless, four estimates of w were small and negative, which happens when $h > r$. These chromosomes were later shown to cause infertility when homozygous, as were three other chromosomes with small, positive values of w. The seven fertility lethal lines were therefore assigned fitnesses of 0. The distribution of homozygous fitness is markedly different from the commonly observed bimodal distribution of homozygous viabilities, with one mode at lethality and the second for chromosomes of quasi-normal viability (Lewontin, 1974). In contrast, homozygous fitnesses have an inverse 'J' shaped distribution, with a large peak at lethality (44% of the chromosomes extracted were either inviable or infertile), and all other values equally frequent.

The average fitness of the total population of third chromosome lines is 0·41 (excluding viability lethals), relative to *TM3*/+ heterozygotes. Since the fitness of *TM3*/+ heterozygotes compared to wild-type

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Table 1. *Weighted average estimates of* h, r *and* w *for homozygous third chromosome lines, and also for the total heterozygous population (Het)*

Line	*h*	*r*	*w*	Line	*h*	*r*	*w*
1	0·31 (0·06)	0·64 (0·01)	0·76 (0·08)	22	0·77 (0·03)	0·79 (0·01)	0·15 (0·07)
2	0·26 (0·06)	0·67 (0·00)	0·82 (0·06)	23	0·25 (0·08)	0·62 (0·02)	0·79 (0·11)
3	0·81 (0·05)	0·80 (0·01)	0	24	0·27 (0·06)	0·64 (0·00)	0·80 (0·06)
4	0·54 (0·03)	0·68 (0·04)	0·44 (0·04)	25	0·44 (0·05)	0·66 (0·02)	0·59 (0·04)
5	0·44 (0·02)	0·64 (0·00)	0·57 (0·05)	26	0·60 (0·04)	0·64 (0·03)	0·16 (0·18)
6	0·65 (0·01)	0·68 (0·01)	0·14 (0·01)	27	0·21 (0·05)	0·62 (0·01)	0·83 (0·06)
7	0·53 (0·01)	0·70 (0·04)	0·52 (0·07)	28	0·10 (0·01)	0·62 (0·02)	0·93 (0·00)
8	0·90 (0·02)	0·84 (0·01)	0	29	0·12 (0·01)	0·64 (0·00)	0·92 (0·01)
9	0·72 (0·01)	0·75 (0·00)	0·13 (0·03)	30	0·25 (0·03)	0·68 (0·02)	0·85 (0·03)
10	0·49 (0·01)	0·67 (0·04)	0·52 (0·06)	31	0·50 (0·04)	0·66 (0·00)	0·47 (0·09)
11	0·48 (0·00)	0·67 (0·01)	0·54 (0·02)	32	0·65 (0·00)	0·72 (0·01)	0·29 (0·02)
12	0·75 (0·02)	0·76 (0·02)	0·04 (0·12)	33	0·82 (0·01)	0·82 (0·01)	0
13	0·43 (0·01)	0·64 (0·01)	0·59 (0·03)	34	0·64 (0·02)	0·70 (0·01)	0·23 (0·11)
14	0·91 (0·00)	0·92 (0·03)	0	35	0·38 (0·06)	0·67 (0·02)	0·69 (0·05)
15	0·42 (0·03)	0·66 (0·01)	0·63 (0·05)	36	0·21 (0·04)	0·65 (0·01)	0·86 (0·03)
16	0·89 (0·03)	0·87 (0·04)	0	37	0·20 (0·01)	0·61 (0·02)	0·84 (0·01)
17	0·74 (0·00)	0·78 (0·01)	0·20 (0·07)	38	0·71 (0·01)	0·75 (0·03)	0·14 (0·07)
18	0·86 (0·02)	0·90 (0·02)	0	39	0·67 (0·00)	0·72 (0·01)	0·19 (0·06)
19	0·61 (0·05)	0·69 (0·00)	0·31 (0·13)	40	0·38 (0·11)	0·61 (0·04)	0·62 (0·13)
20	0·89 (0·02)	0·88 (0·02)	0	41	0·71 (0·00)	0·74 (0·00)	0·13 (0·02)
21	0·77 (0·01)	0·78 (0·01)	0·03 (0·04)	Het	0	0·57 (0·01)	0·31 (0·07)

Standard errors (±S.E.) are empirical, calculated from variation between replicate means. Fitness of each +/+ homozygote is expressed relative to the *TM3*/+ heterozygote; heterozygous fitness is of *TM3*/+ relative to +/+ heterozygotes.

heterozygotes is 0·31, the fitness of the population of homozygous lines relative to wild-type heterozygotes is 0·127 (assuming independence of the fitness measurements).

Previous estimates by this method of the fitness of third chromosome homozygotes derived from natural populations were not based on many chromosomes. Both Tracey & Ayala (1974) and Sved (1975) estimated the average fitness of fourteen chromosomes extracted from indiginous American and Australian populations, respectively, and obtained fitnesses of chromosomal homozygotes relative to heterozygotes of 0·32 (Tracey & Ayala, 1974) and 0·10 (Sved, 1975). These estimates are all in good qualitative agreement, especially considering experimental differences in base populations, sample sizes, marker chromosomes and culture conditions.

(ii) *Quantitative characters*

Average values of six quantitative characters measured on each of the homozygous lines, as well as on the total heterozygous population, are listed in Table 2. Replicate means agree well for all characters, with the exception of fertility, which is represented by only one estimate per replicate. Otherwise, variation between replicates accounts for only a small proportion of the total variation, with the bulk of the variation attributable to differences among the lines (Table 3). The component of variance among lines is attributable to genetic differentiation, and is a function of the genetic variance in the base population and the inbreeding coefficient. However, it is not possible to deduce what proportion of this is additive, and what proportion is non-additive genetic variance, as there is no general solution to the redistribution of genetic variance in the base population within and among inbred lines, when there is dominance (Robertson, 1952).

An indication of the extent to which dominance contributes to the differentiation among the lines for the various characters is the magnitude of the inbreeding depression, since a change of mean on inbreeding can only occur if there is net directional dominance at the relevant loci. Inbreeding depressions, calculated as the difference between the mean of all homozygous lines from the mean of the total heterozygous population per 10% increase in F, and scaled relative to the heterozygous mean, are presented in Table 4. The average inbreeding coefficient of the homozygous lines was estimated in the following manner. The third chromosome is completely inbred ($F = 1$), but is only 40% of the *Drosophila* genome. The remaining 60% of the genome is inbred as a joint result of the chromosome extraction procedure, finite numbers of parents per generation, and variable family size. Since the numbers of male and female parents was known each generation, the effective population number could be estimated, from which the average inbreeding coefficient of chromosomes 1, 2 and 4 was calculated to be 0·42. The average inbreeding coefficient of the homozygous lines is therefore

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Table 2. *Means of quantitative characters measured on third chromosome homozygote lines and the total heterozygous population (Het), averaged over replicates (and sex, where appropriate)*

Line	Viability	Fertility	Female productivity	Male weight	Abdominal bristles	Sternopleural bristles
1	1·105	0·685	60·50	0·849	17·24	20·16
2	0·998	0·823	49·48	0·914	21·75	19·09
3	0·515	0	0	0·773	22·40	14·38
4	0·940	0·466	51·98	0·877	20·73	17·38
5	1·115	0·509	39·49	0·853	17·68	18·69
6	0·939	0·154	34·50	0·896	19·28	19·09
7	0·846	0·621	54·89	0·809	23·30	19·04
8	0·384	0	0	0·682	19·86	14·69
9	0·670	0·197	25·88	0·749	13·31	18·04
10	1·000	0·516	44·85	0·878	17·29	19·84
11	1·066	0·504	62·76	0·848	21·78	14·80
12	0·630	0·063	15·92	0·739	21·03	22·01
13	1·119	0·524	45·11	0·877	17·14	19·24
14	0·177	0	0	0·577	20·73	24·28
15	1·042	0·601	66·74	0·846	18·71	19·80
16	0·305	0	0	0·660	16·01	14·80
17	0·552	0·362	42·18	0·812	18·04	18·58
18	0·218	0	0	0·659	7·21	22·09
19	0·879	0·358	59·35	0·877	22·18	24·30
20	0·276	0	0	0·693	18·64	14·01
21	0·572	0·050	2·40	0·635	19·35	18·99
22	0·521	0·282	6·85	0·841	18·10	17·86
23	1·222	0·649	85·07	0·956	19·11	17·20
24	1·116	0·716	61·35	0·974	18·56	17·95
25	1·050	0·558	49·95	0·915	16·98	16·89
26	1·125	0·143	34·76	0·764	18·38	18·63
27	1·221	0·683	57·71	0·858	21·01	16·53
28	1·237	0·754	75·34	0·829	20·03	17·21
29	1·123	0·821	69·69	0·863	18·86	16·93
30	0·939	0·902	73·83	0·814	18·94	18·64
31	1·044	0·449	47·63	0·849	18·31	18·84
32	0·761	0·378	53·01	0·771	21·86	20·46
33	0·425	0	0	0·703	20·43	18·81
34	0·868	0·268	47·18	0·871	17·95	14·28
35	0·989	0·699	63·85	0·890	18·85	17·25
36	1·060	0·809	73·61	0·900	20·74	20·65
37	1·284	0·656	55·43	0·812	19·94	15·28
38	0·684	0·210	47·93	0·828	20·94	18·81
39	0·795	0·241	48·76	0·932	21·13	18·15
40	1·255	0·492	55·58	0·800	23·09	23·64
41	0·715	0·181	23·40	0·765	15·44	20·03
Het	0·664	0·469	92·37	0·904	20·09	18·24

Homozygous viability and fertility are expressed relative to *TM3*/+ heterozygotes, whereas heterozygous viability and fertility are of marker heterozygotes relative to +/+ heterozygotes.

Table 3. *Variance components of metric characters from analysis of variance of third chromosome homozygote lines*

	Variance components (percent of total variance)			
Character	Among lines	Between replicates within lines	Within replicates	Total
Fitness	0·0989 (92·09)	0·0085 (7·91)	—	0·1074
Viability	0·0949 (73·00)	0 (0)	0·0351 (27·00)	0·1300
Fertility	0·0667 (74·78)	0·0225 (25·22)	—	0·0892
Female productivity	627·6840 (66·44)	28·2669 (2·99)	288·7924 (30·57)	944·7433
Male weight	0·0080 (59·70)	0·0004 (2·99)	0·6050 (37·31)	0·0134
Abdominal bristles	7·9195 (62·35)	0·1845 (1·45)	4·5985 (36·20)	12·7025
Sternopleural bristles	6·3946 (69·52)	0·2092 (2·27)	2·539 (28·20)	9·1977

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Table 4. *Inbreeding depressions, expressed as decrease in mean per* 10% *increase in* F, *as a percent of the non-inbred mean*

Character	Mean of all homozygous lines ($\bar{X}_I$)	Mean of total heterozygous lines ($\bar{X}_H$)	Depression $\frac{0{\cdot}1(\bar{X}_H-\bar{X}_I)}{F\bar{X}_H}\times 100$
Fitness	0·1270	1·0000	13·43
Viability	0·5648	1·0000	6·70
Fertility	0·1867	1·0000	12·51
Female productivity	41·0716	92·3705	8·54
Male weight	0·8154	0·9038	1·50
Abdominal bristles	19·0768	20·0875	0·77
Sternopleural bristles	18·4701	18·2375	0 (−0·20)

The average inbreeding coefficient of each third chromosome line was estimated to be approximately $F = 0{\cdot}65$, as explained in the text.

approximately 0·65. The data presented in Table 4 are consistent with earlier work (Kidwell & Kidwell, 1966; Tantawy & Reeve, 1956) – fitness, viability, fertility and female productivity are severely impaired on inbreeding, whereas body weight and bristle numbers are little affected. Dominance is therefore implicated in the differentiation among the inbred lines for the former characters, but not the latter ones.

The fitness profiles (Figs 3–8) and correlations among line means for pairs of characters (Table 5) are in accord with the above results. The relationship between homozygous fitness and fertility (Fig. 4) is most striking, as the correlation between these characters is indistinguishable from unity. Viability and female productivity are strongly associated with total fitness, male weight and fitness are moderately associated, whereas abdominal bristles and fitness are weakly associated and sternopleural bristles are not detectably related to fitness. It is interesting that all significant correlations of the metric traits with fitness and with each other are positive; this is further evidence concerning the nature of genetic variation for fitness, and will be addressed in the Discussion. The combination of metric traits which best predicts homozygous fitness is the simple multiple of viability and female productivity; the correlation of this multiple with fitness, 0·93, is higher than that between fitness and either viability or female productivity separately, and cannot be improved by including information from the other characters (Fig. 9).

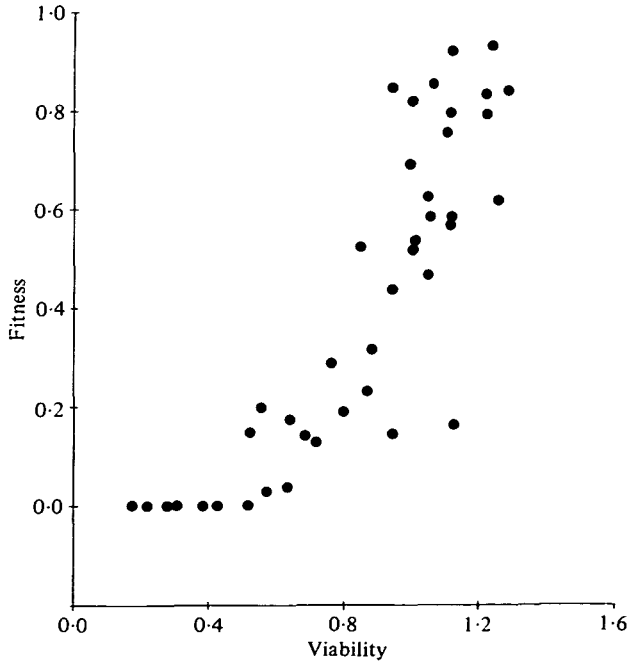

Fig. 3–8. Fitness profiles. Each point represents the mean score of a quantitative character averaged over both replicates of a homozygous line, plotted against the homozygous fitness of that line.

Fig. 3. Relationship of viability to fitness.

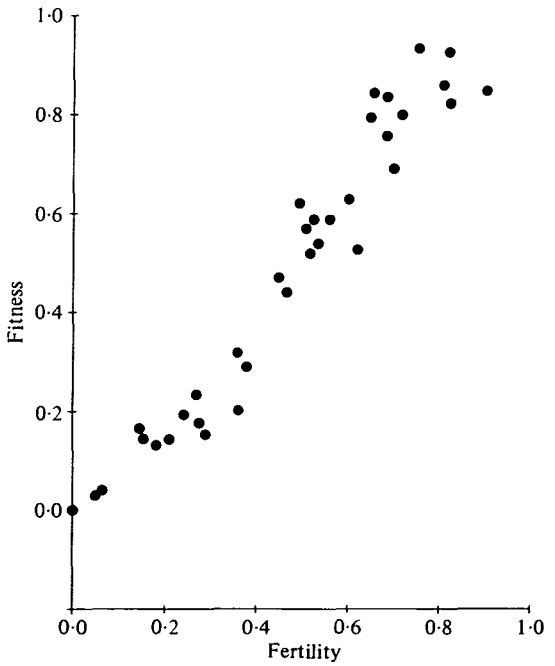

Fig. 4. Relationship of fertility to fitness. The seven infertile lines are represented by a single point at the origin.

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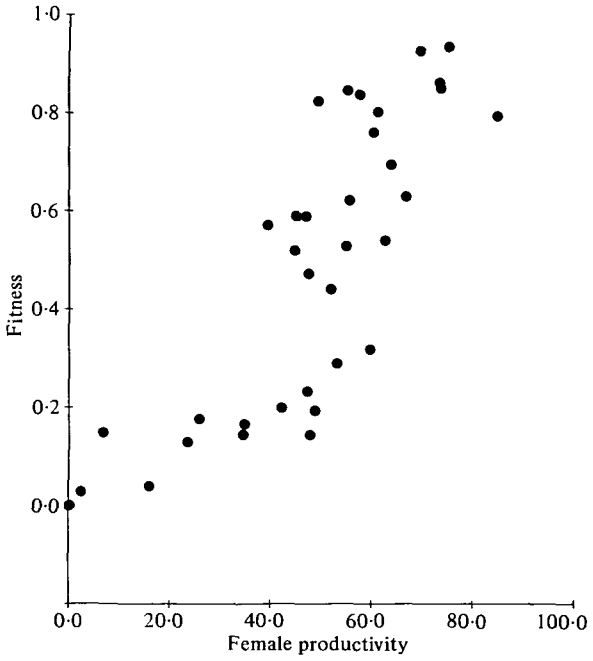

Fig. 5. Relationship of female productivity to fitness. The seven infertile lines are represented by a single point at the origin.

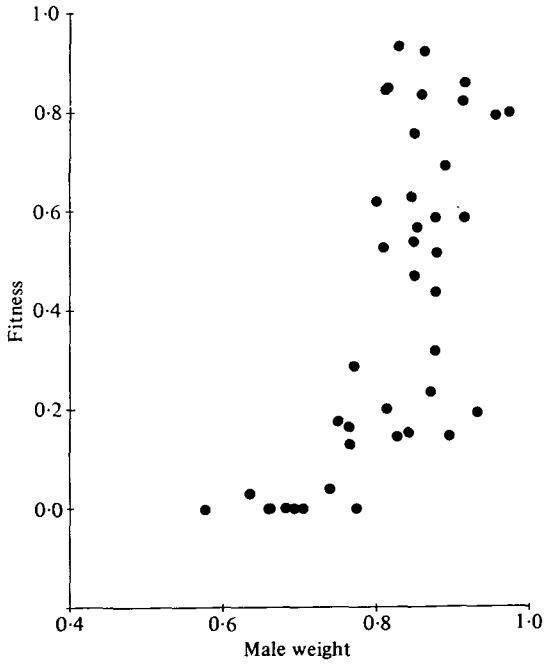

Fig. 6. Relationship of male weight to fitness.

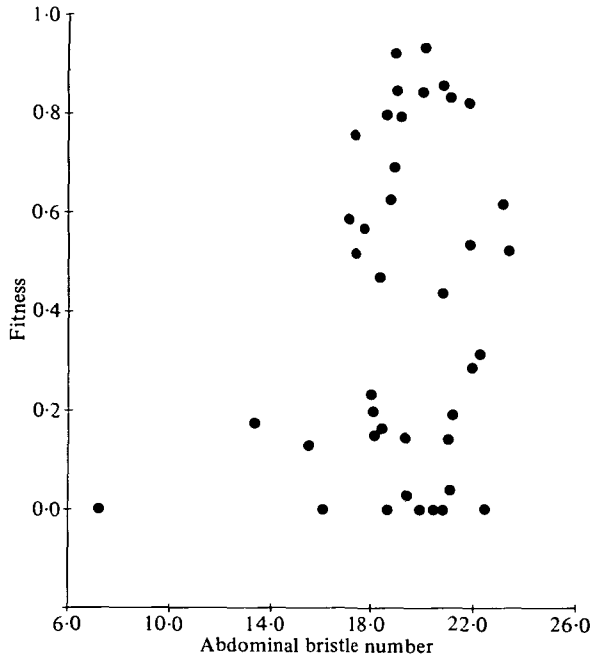

Fig. 7. Relationship of abdominal bristle number to fitness.

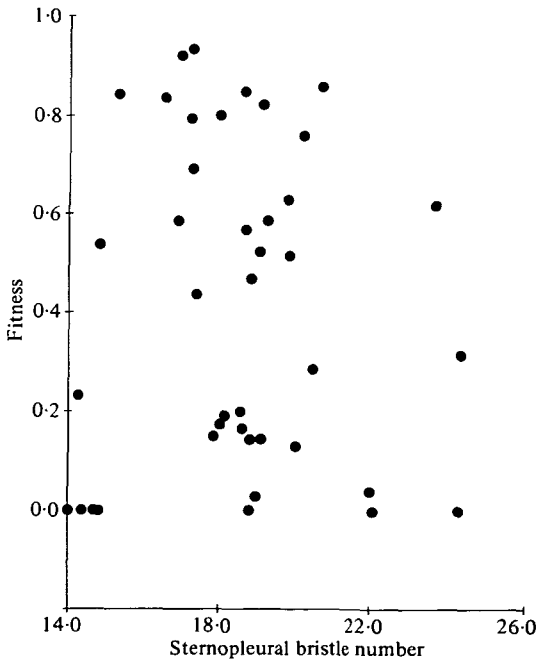

Fig. 8. Relationship of sternopleural bristle number to fitness.

(iii) *Relationship between homozygous and heterozygous fitness*

Although one can predict homozygous fitness well by the product of homozygous female productivity and homozygous viability, extension of the prediction to real populations depends on the relationship of homozygous fitness and its components to heterozygous fitness and its components. Determination of heterozygous fitness by competitive elimination of a marked chromosome is not straightforward for crosses between pairs of homozygous lines, because the heterozygous combination is not preserved intact each generation, but recombines and segregates. In order to ensure that wild-type genotyes competing against *TM3*/+ were mostly heterozygous, homozygous lines were pooled in the manner described in *Materials and Methods*, and heterozygous fitness and viability estimates were made from the resulting six 'partial' heterozygous lines.

Comparison of average fitness and viability of the component homozygous lines with the values from the

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Table 5. *Product-moment correlations of means of quantitative characters among third chromosome homozygote lines* (±S.E.)

	Viability	Fertility	Female productivity	Male weight	Abdominal bristles	Sternopleural bristles
Fitness	0·852 (0·024)	0·975 (0·037)	0·866 (0·048)	0·677 (0·019)	0·193 (0·005)	−0·037 (0·026)
	Viability	0·809 (0·079)	0·853 (0·022)	0·760 (0·055)	0·260 (0·015)	−0·014 (0·038)
		Fertility	0·886 (0·000)	0·713 (0·004)	0·198 (0·022)	−0·010 (0·004)
			Female productivity	0·778 (0·027)	0·285 (0·009)	0·050 (0·021)
				Male weight	0·211 (0·002)	−0·107 (0·003)
					Abdominal bristles	0·050 (0·047)

Correlations were computed from means averaged over replicates (and sex, where appropriate). Standard errors are empirical, calculated from variance between correlation coefficients obtained for each replicate separately.

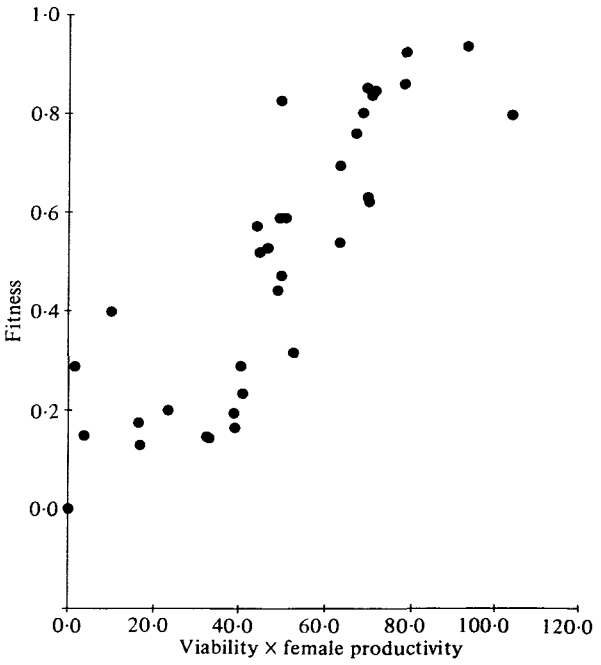

Fig. 9. Prediction of fitness from component metric traits. Each point represents viability × female productivity of each of the 41 third chromosome homozygous lines, averaged over both replicates, plotted against the homozygous fitness of that line.

corresponding 'partial' heterozygous line gives an indication of the nature of genetic variance for fitness (Table 6). There is no significant correlation of homozygous and heterozygous fitness: heterozygotes derived from crosses of the seven homozygous fertility lethals were as fit as heterozygotes derived from the six lines of greatest homozygous fitness. This is consistent with a model of genetic variation for fitness due to dominance, but not with one of additive variation for fitness. However, one cannot infer correlations among homozygous and heterozygous fitness components from the correlation between net homozygous and heterozygous fitness. The correlation between homozygous and heterozygous viability (0·74) is significant, and points to some additive genetic variance for viability. The discrepancy arises because homozygous viability is a major component of homozygous fitness (correlation 0·9), but heterozygous viability is unimportant as a determinant of heterozygous fitness (correlation of 0·2 not significantly different from zero).

4. Discussion

What is the nature of genetic variance for fitness in *Drosophila*? The observations of substantial inbreeding depression for total fitness and lack of correlation between homozygous and heterozygous fitness are consistent with a model of genetic variation for fitness caused by segregation of rare deleterious recessive alleles in natural populations. The observed fitness variation among homozygous third chromosome lines extracted from a natural population can then be attributed to the unmasking of different recessive alleles which impair fitness to varying degrees. These deleterious recessives appear to fully complement each other no matter how severe the homozygous effect, so heterozygous combinations of the homozygous lines are all of equal fitness. This is consistent with earlier indirect calculations that the heterozygous effect on fitness of mutations with both drastic and mildly deleterious homozygous effects are approximately the same (the argument is summarized in Simmons & Crow, 1977). (In contrast, the dominance of viability mutations varies inversely with their homozygous effect, so that lethal mutations are nearly completely recessive, while heterozygotes for slightly deleterious

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Table 6. *Fitness and viability* (±S.E.) *of the partial heterozygous populations, and the homozygous lines of which they are composed*

Partial heterozygous population	Fitness		Viability	
	Homozygotes	Heterozygotes	Homozygotes	Heterozygotes
1	0 (0)	0·496 (0·092)	0·218 (0·030)	0·683 (0·023)
2	0·034 (0·006)	0·410 (0·131)	0·446 (0·034)	0·909 (0·106)
3	0·081 (0·011)	0·362 (0·128)	0·562 (0·044)	0·832 (0·067)
4	0·168 (0·005)	0·513 (0·018)	0·681 (0·023)	0·842 (0·075)
5	0·222 (0·010)	1·491 (0·159)	0·745 (0·028)	0·878 (0·095)
6	0·270 (0·005)	0·559 (0·347)	0·746 (0·038)	0·890 (0·096)

Heterozygous fitnesses and viabilities are weighted averages over replicates, and standard errors are empirical, calculated from variation between replicate estimates. Homozygous fitnesses and viabilities are unweighted averages of the values previously obtained for the homozygous lines, for the lines which were crossed to form the corresponding partial heterozygous population. Standard errors are computed from variation among means of the homozygous lines within each group. All estimates are expressed relative to the fitness and viability of the total heterozygous population.

genes have a reduction in viability 30–50% of the homozygous effect (Simmons & Crow, 1977)).

Correlations among homozygous line means for major components of fitness were highly positive; this is also consistent with a hypothesis regarding genetic variation for fitness as largely due to dominance variance. One may expect that the past action of natural selection operating simultaneously on all major components of fitness will have fixed those pleiotropic alleles having a positive joint effect on the components, and eliminated those with a detrimental joint effect. However, pleiotropic alleles with a positive effect on one major fitness component, yet a negative effect on another may be less influenced by natural selection and so remain longer at intermediate frequencies, so that negative additive genetic correlations among fitness components are expected in random breeding populations (Falconer, 1981). However, positive genotypic correlations among fitness components may be generated by inbreeding if rare deleterious recessive alleles have negative pleiotropic effects on all fitness components. The word 'genotypic' is used to describe such correlations to indicate that they include correlations among dominance deviations as well as correlations among breeding values of the fitness components. The relative contribution of the two sources of covariance is not known in general and is not a simple function of the inbreeding coefficient, just as the redistribution of genetic variance for a single trait among inbred lines is not known when there is dominance variance. This may explain the high positive genotypic correlations of fitness components among the third chromosome homozygous lines observed here and in other inbred stocks (Giesel & Zettler, 1980; Rose, 1984).

The implication of these results regarding observations of fitness components in natural populations is that the genetic variance–covariance matrix of fitness components will strongly depend on the geographic structure of the population concerned. A single large panmictic population at selective equilibrium may be expected to exhibit a pattern of negative genetic correlations among major components of fitness, but any local population subdivision and hence inbreeding will cause positive correlations.

It cannot be overemphasized that any measurement of fitness and its resolution into components is relevant only to the exact experimental conditions of measurement. Not only are fitness rankings of different lines likely to alter when biotic or abiotic features of the environment are changed, so will the relationship of quantitative characters to fitness as well as the prediction of fitness as a function of component metric traits. The very nature of the genetic variation underpinning fitness may alter as the environment changes, for previously selectively neutral variants segregating in the population may become components of fitness in a novel environment, creating additive genetic variation for fitness and hence response to selection in the new conditions (for an example see Dykhuizen, De Framond & Hartl, 1984). Under the conditions of temperature, media composition, density and mating used in this experiment, total homozygous fitness of any third chromosome line could be well predicted by the product of that line's viability and female productivity. These conditions were very lenient regarding male mating competition, as mating could occur over a period of at least 4 days, and adult longevity was unimportant, as the populations were censused every 14 days. It is easy to envisage other conditions, such as decreasing the time allowed for mating, increasing the census interval, or increasing culture density, for which a premium would be set on other fitness components.

P. Pignatelli (1983, unpublished Honours thesis) has shown for a subset of the third chromosome lines described above that there is substantial genotype X environment interaction for fitness across a range of

temperatures. The genetic correlation of fitness measured across this environmental spectrum is thus low; different constellations of genes determine the character, fitness, in different environments. It follows that knowledge of rank order fitness of a series of lines, relationships of quantitative traits to fitness, and predictive fitness functions in a single environment are irrelevant insofar as fitness in different environments is concerned. Comparison of the results of this experiment with those of other recent experiments measuring fitness and its components in *Drosophila melanogaster* reinforces this assertion. Yamazaki & Hirose (1984) measured net fitness and the components viability, productivity and development time of 50 homozygous lines from a natural population of *D. melanogaster* by competition with *D. hydei*. They report a high genotypic correlation of net fitness and population productivity under this regime, but net fitness is poorly correlated with viability, as is productivity with viability. These results are inconsistent with those presented here; it appears natural selection weights components of fitness differently in interspecies competition than in intraspecies competition. Haymer and Hartl also compared net fitness estimates of several strains of *D. melanogaster* by five different inter- and intra-specific competitive techniques (1982) and two non-competitive measures (1983). They found that the fitness rankings of the lines varied across this battery of measurement techniques, again presumably because of variation in the relative weighting of fitness components in each competitive environment.

The combination of poor association between homozygous and heterozygous fitness and components, and overwhelming genotype × environment interaction for both the composite trait, fitness, and its components renders forever elusive the search for generalized relationships of individual quantitative traits to fitness and prediction of fitness by a function of its components. Perhaps this is not entirely unexpected in a world in which the survival of the 'fittest' individuals each generation has produced such amazing biological diversity.

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