# Quantitative Genetic Variation in Body Size of Mice From New Mutations

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ABSTRACT

To measure the amount of new genetic variation in 6-week weight of mice arising each generation from mutation, selection lines derived from an initially inbred strain were maintained for 25 generations. An analysis using an animal model with restricted maximum likelihood was applied to estimate a mutational genetic component of variance for the infinitesimal model of many genes of small effect. Assuming that the inbred base population was at a mutation-drift equilibrium, it is estimated that the heritability for body size has increased by 1.0% per generation, with lower and upper confidence limits of 0.6% and 1.6%, respectively. A model which includes a mutational genetic component of variance fits the data much better than one involving only base population genetic variance. A model with no genetic component fits the data very poorly. An environmental covariance of body size of mother and offspring was included in the model and accounts for 10% of the variance. By using information only from the observed response to selection, the estimated increase in heritability from mutation is 0.3% per generation. These values are higher than published estimates for the increase in variance from spontaneous mutations in bristle traits of Drosophila, for which there are extensive data, but similar to estimates for various skeletal traits in mice.

OBSERVATIONS in some selection experiments of responses which continue without appearing to plateau, together with empirical estimates of the amount of new variation arising per generation from mutation, imply that new mutations can make a significant contribution to long term selection responses (FRANKHAM 1980; HILL 1982). The role of mutation in maintaining the large amount of genetic variation commonly observed in natural populations (MOUSSEAU and ROFF 1987) is more controversial, but stabilizing selection or drift in a finite population will ultimately exhaust genetic variation, so the maintenance of genetic variation in the long term depends on an input of new mutations. Direct evidence that new mutations have affected responses to artificial selection comes from occasional observations of mutations of large effect in experimental selection lines (MACARTHUR 1949; YOO 1980; BRADFORD and FAMULA 1984; FRANKHAM 1988; CABALLERO, TORO and LOPEZ-FANJUL 1991). Effects of individual alleles causing variation in quantitative traits are usually difficult to detect, so the average amount of new genetic variance arising per generation in the trait from mutations of any type, $V_M$, is a pragmatic measure of the influence of mutation on a quantitative trait. This is commonly scaled by the environmental variance of the trait, $V_E$, and expressed as the "mutational heritability," $h_M^2 = V_M/V_E$, the increment in heritability per generation from mutation.

Information on mutational heritabilities comes mainly from the results of two types of experiment. (1) The rate of divergence between sublines from a highly inbred base population. The sublines are typically maintained by brother-sister mating, in which case drift dominates and the fixation rate can be assumed to equal the mutation rate. Similarly, the rate of divergence of the quantitative trait between sublines is proportional to the mutational variance (LYNCH and HILL 1986). (2) The response to artificial selection in inbreds. Theory is available to predict the rate of response from new mutations from additive genes (HILL 1982), but the fixation rate of recessive genes depends on the population structure (CABALLERO, KEIGHTLEY and HILL 1991). Because population sizes are larger than for inbred sublines and the majority of new mutations are likely to be deleterious for fitness, natural selection is likely to play a more important role by tending to eliminate genetic variance and hence reducing estimates of mutational heritabilities.

The most extensive data on mutational heritabilities come from experiments on various bristle number traits in Drosophila for which LANDE (1976) arrived at a consensus figure for $h_M^2$ of about $1 \times 10^{-3}$. Recently, two very large experiments which provide good estimates of $h_M^2$ for Drosophila bristle number have been reported. (1) CABALLERO, TORO and LOPEZ-FANJUL (1991) performed replicated selection on abdominal bristle number in an inbred strain, and the results confirmed the theoretical expectation that responses from fixation of new mutations would increase with population size (HILL 1982). The estimate from

this experiment of the mutational heritability using an additive model of many genes of small effect (the infinitesimal model) was $1.43 \times 10^{-3}$, but most of the new variance could be attributed to a few genes of very large effect. (2) MACKAY *et al.* (1992) maintained many replicated sublines derived from an inbred base for many generations and estimated the mutational heritability from the observed divergence among sublines and from the response to subsequent directional selection. These estimates were $3.3 \times 10^{-3}$ for abdominal and $1.5 \times 10^{-3}$ for sternopleural bristle number. In Drosophila, the spontaneous rate of accumulation of mutational variance in various quantitative traits can be inflated by as much as two orders of magnitude if systems of transposable elements (*e.g.*, *P*) are mobilized (MACKAY 1987; MORAN 1990; MACKAY, LYMAN and JACKSON 1992). As is the case for spontaneous mutational variance, much of the transposable element-induced mutational variance is associated with alleles of very large effect and these can be detected in an appropriately designed experiment (MACKAY, LYMAN and JACKSON 1992).

There is much less information on mutational heritabilities for vertebrate species and none for traits of economic importance in farm animals. There is no *a priori* reason to expect that mutational heritabilities in such traits are similar to those for bristle number traits of Drosophila because mutation rates per locus appear to be related to the generation interval (KIMURA 1983), and the number of loci that can affect a given trait may vary enormously between traits and species.

In mice there have been no selection experiments from an inbred base, but mutational heritability estimates have been obtained from the rate of divergence between inbred sublines for a range of skeletal traits (summarised by LYNCH 1988). These mutational heritabilities are around $10 \times 10^{-3}$, an order of magnitude higher than spontaneous mutational heritabilities for Drosophila bristles. Potential problems in such experiments relate, however, to the maintenance of many sublines for long periods without contamination, and the presence of residual variance in the base stocks which is expected to be substantial relative to the mutational variation. The experiments of BAILEY (1959), who measured the divergence in the length of two leg bones between inbred sublines of two inbred mouse strains, are of greatest relevance to the present paper because these traits are likely to be genetically correlated with body size. LYNCH (1988) reanalyzed the results of these experiments and obtained estimates for $h_M^2$ of $16 \times 10^{-3}$ and $31 \times 10^{-3}$ for ilium and ulna length, respectively. These estimates are surprisingly high and would imply that a response from new mutations would be detectable in the short term if selection were applied in lines of reasonable size using an initially inbred population.

The purpose of the present experiment, which involves selection in a population based on a long established inbred line, is to measure the amount of genetic variance arising per generation in 6-week weight in the mouse. Many selection experiments have been carried out on this trait, but all have involved outbreds or crosses between inbred strains (EISEN 1974). The data from the present selection experiment are analyzed using the standard method of equating the response to selection to the mutational heritability (HILL 1982). Alternative models are considered in which the variance is either generated by many genes of small effect (infinitesimal model), or a few genes of very large effect. In the former case, an allowance is made for the genetic variance which is expected to be present in the base population.

An analysis which uses only the response to selection ignores information on covariances between relatives within lines, a proportion of which can be genetic. In order to more fully use this information, a restricted maximum likelihood (REML) analysis (PATTERSON and THOMPSON 1971) with a modified version of the animal model is applied. In the animal model, the covariances between all the related individuals in the pedigree as well as the between line change in mean are used in the estimation of genetic components of variance, and a reduction in genetic variance induced by selection (BULMER 1971) is accounted for (SORENSEN and KENNEDY 1984). REML with the animal model allows, in principle, a separate estimation of the amount of genetic variance in the inbred base population and the increment in variance each generation from mutation. Environmental components, in this case a common environmental variance of full sibs and an environmental covariance of offspring and mother, are also simultaneously estimated. Preliminary results of this experiment were given previously (KEIGHTLEY and HILL 1990), but a more complete analysis of a much larger data set is presented here.

## MATERIALS AND METHODS

**Selection lines:** The experiment was initiated from individuals of inbred strain C3H/He obtained from Bantin and Kingman Ltd., England, in 1986. Within-family selection on body weight at 6 weeks was carried out in a high and low line for 25 generations. Only first litters were recorded in the experiment. For the first 14 generations, the lines were maintained at 12 pairs of parents each, and thereafter at 16 pairs. In order to maintain as high an effective population size as possible by reducing the overall level of inbreeding, while at the same time generating local inbreeding and increasing the fixation probability of recessive mutations, a circular mating scheme was initially adopted in which each selected female was mated to the selected male from the following family, and the circle completed by mating the female of the last family to the male of the first (KIMURA and CROW 1963). Theoretical analysis of this scheme indi-

cated that it is relatively inefficient for the purpose of fixing recessive mutations (CABALLERO, KEIGHTLEY and HILL 1991), so it was discontinued at generation 21 in favor of mating about half of the mice as full-sibs and the rest at random. The data set currently comprises 3142 individuals in 627 families.

The mean body size was also measured at generations 24 and 25 in offspring of parents selected in the reverse direction. This was done in order to estimate any effect of maternal body size on size of offspring, which would bias the estimate of the difference in genetic mean between lines.

**Estimation of mutational heritability from response to selection:** The mutational heritability can be estimated from the cumulative response to selection using adapted versions of formulas of HILL (1982), but the analysis is complicated because the genetic variance in the base population ($V_{g,0}$) is not expected to be zero. For example, the expected genetic variance in a full-sib line at mutation-drift equilibrium (assumed to be the base population for the present experiment), is about five times the mutational variance (LYNCH and HILL 1986). Consider two models which make different assumptions about the magnitude of effects of new mutant alleles:

*Infinitesimal model:* The effects of mutations are assumed to small and additive, and the effect of selection on the genetic variance is assumed to be small. Following HILL (1982) and FALCONER (1989, Ch. 13), it can be shown that the cumulative divergence between the high and low line from $t$ generations of within-family selection of intensity $i$ with random mating is approximately

$$D_t = 4N(i/\sigma)(1 - r)[((1 - (1/n))/(1 - \tau))]^{1/2}[V_{g,0}(1 - e^{(-t/2N)}) + V_M(t - 2N(1 - e^{(-t/2N)}))], \quad (1)$$

where $r$ is the correlation of breeding values of full-sibs (*i.e.*, one-half), $n$ is the mean number of each sex per family, $\tau$ is the observed correlation of phenotypic values of full-sibs, and $\sigma$ is the phenotypic standard deviation.

*Additive genes of large effect:* An extreme alternative to the infinitesimal model is a model of additive mutations of large effect fixed instantaneously by selection such that the asymptotic rate of response is reached immediately. In this case a term for the response as a function of the initial genetic variance cannot be included in a straightforward manner and has to be ignored. Again, following HILL (1982) and FALCONER (1989, Ch. 13), the cumulative divergence between high and low lines from $t$ generations of within-family selection is approximately

$$D_t = 4tN(i/\sigma)(1 - r)[((1 - (1/n))/(1 - \tau))]^{1/2}V_M. \quad (2)$$

**Estimation of mutational heritability using REML:** The application of REML to a mouse selection experiment is explained in detail by MEYER and HILL (1991). In a "standard" animal model, terms proportional to genetic covariances (relationships) between all individuals in the pedigree are specified in the numerator relationship matrix (NRM). The relationship between two individuals is defined as twice the inbreeding coefficient of a hypothetical offspring of the two individuals. In order to include mutation in the model, additional terms are added to the elements of the NRM to account for increased genetic covariances between related individuals. There are two genetic components, the genetic variance at generation 0, $V_{g,0}$, and the increment per generation from mutation, $V_M$, and these can be estimated separately by REML. The infinitesimal model is assumed for both genetic components. For example, after one generation of inbreeding and mutation, but ignoring the effect of selection on the genetic variance, the genetic variance at generation $t + 1$, $V_{g,t+1}$, is distributed as $(V_{g,t}/2)(1 - 1/N)$ between families and $V_{g,t}/2 + V_M$ within families, assuming the mutations occur in a late division of the parent's germ line. The data were analyzed with the REML computer programs of MEYER (1989) in which the inverse of the NRM with terms to include mutation was generated by the method of WRAY (1990). The algorithm was tested on simulated data generated with an infinitesimal model. A common environmental variance of full-sibs, $V_c$, and an environmental covariance between offspring and mother $\sigma_{ope}$ were simultaneously estimated. The former is an important source of variation in body size between litters (*e.g.*, FALCONER 1973). The latter was included because large mothers may tend to produce larger offspring than average because, for example, they provide them with more milk. A less biased estimate of the genetic component is therefore obtained by including it. This model of maternal effects is similar to that introduced by FALCONER (1965) except that here a covariance of mothers random environmental deviation and offspring is included rather than a phenotypic covariance term. It was computationally feasible to include an environmental covariance term in the REML procedure, but not so for a phenotypic covariance term. An analysis of simulated data generated with a phenotypic rather than environmental covariance between offspring and mother did not show substantial bias in the estimate of a genetic component. Fixed effects of sex, generation number and litter size at birth (six categories) were included in the model. The genetic variance component estimates are generally expressed as "heritabilities" by scaling by $V_E$, the environmental variance including litter and maternal effects.

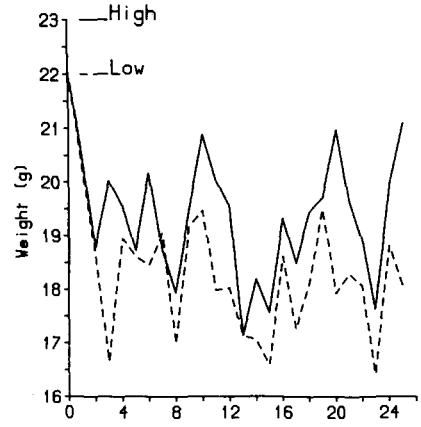


FIGURE 1.—Mean 6-week weight (g) corrected for litter size of high and low line plotted against generation number.

## RESULTS

The mean 6-week weights corrected for litter size of high and low lines plotted against generation number are shown in Figure 1, and the high-low divergence is shown in Figure 2. Because mean litter size is smaller in the low than high line and offspring in small litters tend to be larger than those in large litters, the effect of applying the litter size correction is to increase the average difference in mean body size between the lines. This difference does not, however, contribute to the estimate of the response to selection unless genetic changes in litter size occur as a consequence of the selection on body size. In Figure 2, a

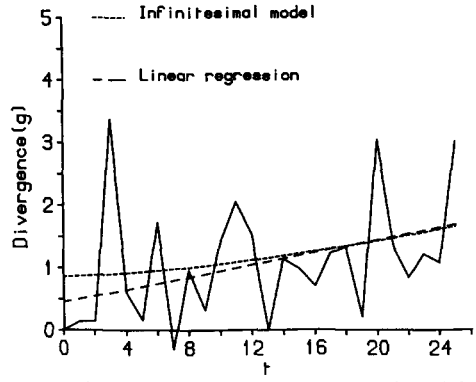


FIGURE 2.—Divergence between mean 6-week weight of high and low line (g) corrected for litter size plotted against generation number. Lines of best fit computed by linear regression and by fitting Equation 1 are also plotted.

line of best fit computed by linear regression, weighted simply by the numbers of animals recorded each generation with the point for generation 0 omitted, is also shown. The reason for omitting the point for generation 0 is that subsequent generations of selection induce a divergence between the lines because of the influence of maternal body size on size of offspring. (When the effects of litter size are removed, large mothers tend to have large offspring.) We assume that the divergence induced by selection has reached its maximum by generation 1. [See KIRKPATRICK and LANDE (1989) for a treatment of the effects of selection on maternally inherited characters.] The point of intercept of the regression line is positive and significantly different from zero, confirming that this maternal effect is positive, *i.e.*, there is a positive environmental covariance of body size of mother and offspring. The slope of the regression line is positive, $b = 0.05 \pm 0.025$ ($0.05 < P < 0.1$), suggesting that there has also been a response to selection. (This estimate of the standard error of the regression coefficient is biased downward because of autocorrelation of generation means and correlation of family members, but only trivially so because the heritability is small, selection was weak, and families were small.)

The mutational variance can be obtained from the response to selection using Equations 1 and 2.

**Infinitesimal model:** Assuming gene effects are small and additive, the cumulative between line divergence from selection of new mutations is given by Equation 1. The mutational variance was estimated by minimizing the sum of squares of the observed divergence each generation from an expected divergence obtained from Equation 1. A constant term ($k$) was also estimated (*i.e.*, $D_T = f(V_M) + k$) to account for a maternal environmental effect, and the neutral model was assumed for the base population variance such that $V_{g,0} = 5V_M$. Values of the other parameters required for (1) together with the methods used to estimate them from the data are given in Table 1.

TABLE 1

**Parameters used in computation of mutational heritability from the response to selection and how they are estimated**

| Parameter | Value | How computed |
|---|---|---|
| $N$, effective population size | 22.7 | From the mean relationship, $\rho$, at generation $t$: $N = [1 - (1 - \rho/2)^{1/t}]/2$ with $t = 25$. |
| $i$, within family selection intensity | 0.38 | From the observed within family selection differential divided by the within family phenotypic SD |
| $r$, correlation of within family phenotypic values | 0.41 | From REML analysis |
| $\sigma$, phenotypic sd | 1.91 | From REML analysis |
| $n$, mean number in each sex per family | 2.66 | |

TABLE 2

**Estimates of mutational heritability together with confidence intervals where available for the various methods of analysis**

| Method of estimation of $h_M^2$ | $h_M^2$ | Confidence interval |
|---|---|---|
| Response to selection, infinitesimal model | $2.7 \times 10^{-3}$ | |
| Response to selection, large gene effects | $1.4 \times 10^{-3}$ | |
| Animal model REML, global ML estimate | $16 \times 10^{-3}$ | $13 \times 10^{-3}$ to $24 \times 10^{-3}$ |
| Animal model REML with neutral expectation for $V_{g,0} = 5V_M$. | $10 \times 10^{-3}$ | $6 \times 10^{-3}$ to $16 \times 10^{-3}$ |

The least squares estimate of mutational heritability is $2.7 \times 10^{-3}$, which is not significantly different from zero. The expected divergence in phenotype given this model and fitting the least squares estimate of $h_M^2$ is shown in Figure 2. The point of intercept with the vertical axis gives the value of $k$, the maternal effect. Equation 1 was derived ignoring the loss of genetic variance which is expected to occur because of selection (BULMER 1971), but in this experiment the loss would be very small because the selection was very weak and the heritability very small. Mutational heritabilities obtained using this and other models are summarized in Table 2.

**Additive genes of large effect:** With this model the pattern of response is linear, so the best estimate of the cumulative divergence is 1.24 g (with a standard error of approximately 0.62 g) from the slope of the linear regression multiplied by the appropriate number of generations (25). Substituting this and the other parameters from Table 1 into (2), the estimate of mutational heritability is $1.4 \times 10^{-3}$.

**REML analysis with the animal model:** The animal model with mutation can be used to estimate

separately the base population genetic variance and the mutational variance, and it is possible to compute the likelihood of the data for any combination of the two parameters. Figure 3 shows the natural log likelihood of the data as a function of $h_M^2$ with the likelihood maximized with respect to all other parameters including $V_{g,0}$. The global maximum likelihood (ML) estimate of the mutational heritability is $16 \times 10^{-3}$, and the likelihood is about $e^{18} = 6.6 \times 10^7$ times higher than for $V_M = 0$. If confidence limits are defined such that values with a difference in log likelihood from the ML of greater than 2 are outside the limits (*i.e.*, roughly equivalent to 95% confidence limits), the confidence interval for the mutational heritability is $13 \times 10^{-3}$ to $24 \times 10^{-3}$. Global ML values of the random environmental effects are given in Table 3. About 10% of the variance is accounted for by including the environmental covariance of offspring and mother in the model, and with a likelihood $e^{3.2} = 25$ times higher that that for $\sigma_{ope} = 0$.

The likelihood of the data as a function of the base population heritability, with the likelihood of all other parameters including $h_M^2$ maximized, is shown in Figure 4. The global ML value is at $V_{g,0}/V_E = 0$ and an upper confidence limit as defined above is $34 \times 10^{-3}$. Presumably, the sharp drop in log likelihood occurs because there is a switch from explaining most of the genetic variance by mutation with little base population genetic variance to explaining most of the genetic variance by base population genetic variance and little mutational variance. A model with no genetic component gives a very poor fit to the data.

A REML analysis was also performed in which the genetic variance in the base population was set at the neutral expectation of $5V_M$. Figure 5 shows the likelihood of the data with the likelihood of the random environmental effects maximized. The ML value for $h_M^2$ is $10 \times 10^{-3}$, and the upper and lower confidence limits as defined above are $6 \times 10^{-3}$ and $16 \times 10^{-3}$.

**Heritabilities in early and late generations:** A possible nongenetic explanation for the large and highly significant estimated mutation heritability from REML is an increase in phenotypic variance due, for example, to some unaccounted for change in the environment in later generations. To ascertain whether this is the case, variance components were estimated by REML for five-generation intervals (Table 4). Genetic variance at the start of the interval and a line effect to account for genetic change resulting from selection prior to the start of the interval were estimated and there was assumed to be no mutational input. The results of this analysis suggest that the heritability has indeed increased during the period of the experiment, but that the phenotypic variance has also increase during the last five generations.

**Reverse selection:** The difference in mean performance of offspring of individuals selected in opposite directions within lines provides an alternative estimate of the maternal effect. Such data are given in Table 5 where, for example, $LH_{24}$ is selection downward in the high line at generation 24. The maternal effect is the average of $HH_{24} - LH_{24}$ (1.0 g), $HH_{25} - LH_{25}$ (2.2 g) and $HL_{25} - LL_{25}$ (1.5 g), which is 1.6 g with slight bias due to genetic change. Similarly, the divergence in genetic means between lines can be obtained from the difference between lines in mean weight of offspring of parents selected in the same direction, *i.e.*, the average of $LH_{24} - LL_{24}$ (0.1 g), $LH_{25} - LL_{25}$ (0.8 g) plus $HH_{25} - HL_{25}$ (1.5 g), which is 2.0 g. Both maternal effect and difference in genetic mean obtained in this way are rather higher than from the regression analysis (Figure 2).

## DISCUSSION

Alternative genetic analyses of the results of 25 generations of selection on body size in an initially inbred mouse line have been presented. With a model of additive genes of small effect (the infinitesimal model) utilizing information only from the observed response to selection, and assuming the neutral expectation for the base population genetic variance ($5V_M$), the mutational heritability estimate is $2.7 \times 10^{-3}$. This is biased upward if mutants of large effect are actually present, so it is an upper limit. If a model of large additive mutant allelic effects fixed instantaneously by selection is fitted, the estimate is $h_M^2 = 1.4 \times 10^{-3}$, but is likely to be an underestimate because the fixation rate of mutations with the breeding structure and selection intensity of this experiment is very slow. Neither of these estimates is significantly different from zero, however.

Using REML with the animal model, the global ML estimate of $h_M^2$ is $16 \times 10^{-3}$, and this is highly significantly different from zero. This analysis assumes the infinitesimal model, and uses all the information on covariances between relatives as well as the between line divergence from selection. If it is assumed that the initial genetic variance in the inbred base population is the neutral expectation of $5V_M$, the ML estimate is $10 \times 10^{-3}$. Sampling could explain the difference between this and the estimate based on the observed response to selection alone ($2.7 \times 10^{-3}$), because different information is used in the two analyses, but other possibilities are: (1) the environmental variance may have increased during later generations of the experiment and some of the increased phenotypic variance may have been estimated by REML as a mutational genetic component (*cf.* Table 4). (2) The genetic variance could be increasing within the lines from mutation, but if the mutations have side effects on fitness, the response would be less than predicted by an infinitesimal model (HILL and KEIGHTLEY

TABLE 3

**Global maximum likelihood estimates of random effects computed by REML**

| Effect | Symbol | ML estimate | ML estimate/$V_P$ | $L_{ML} - L_0$ |
|---|---|---|---|---|
| Base population additive variance | $V_{g,0}$ | 0 | 0 | 0 |
| Mutational genetic variance | $V_M$ | 0.060 | 0.016 | 18 |
| Common environmental variance | $V_c$ | 1.50 | 0.41 | |
| Offspring, mother environmental covariance | $\sigma_{opc}$ | 0.36 | 0.10 | 3.2 |
| Phenotypic variance | $V_P$ | 3.67 | 1 | |

The final column is the difference in log likelihood of the data with the parameter at its ML value ($L_{ML}$) and fixed at zero ($L_0$), with the likelihood of all other random effects maximized.

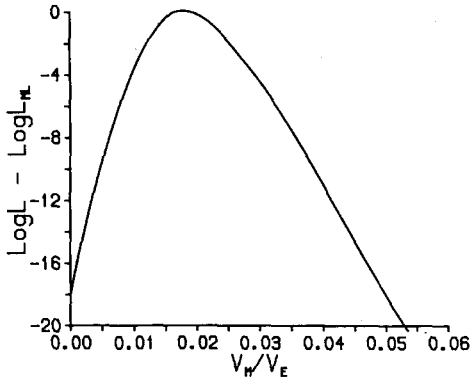


FIGURE 3.—Likelihood of the data expressed as the difference in log likelihood ($log_L$) from the global maximum as a function of the mutational heritability. The likelihood with respect to the variances of all the other random effects including $V_{g,0}$ is maximized.

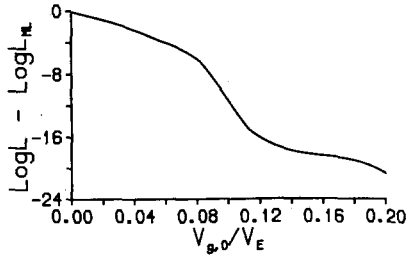


FIGURE 4.—Likelihood of the data expressed as the difference in log likelihood from the global maximum likelihood as a function of the base population genetic variance. The likelihood with respect to the variances of all the other random effects including $V_M$ is maximized.

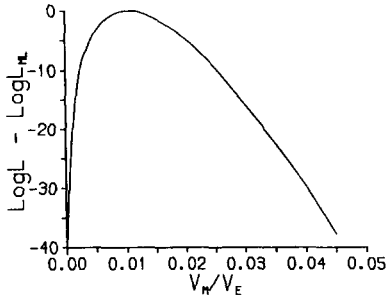


FIGURE 5.—Likelihood of the data expressed as the difference in log likelihood from the local maximum likelihood for $V_{g,0} = 5V_M$ as a function of the mutational heritability. The likelihood with respect to the variances of all the other random effects is maximized.

TABLE 4

**Estimates from REML with different generation intervals in the data set of heritabilities and variances at the start of each interval**

| Generations | $h^2$ | $V_P$ | $V_{EW}$ |
|---|---|---|---|
| 1–5 | 0.043 | 3.69 | 2.14 |
| 6–10 | 0.07 | 2.87 | 1.63 |
| 11–15 | 0.12 | 4.17 | 1.92 |
| 16–20 | 0.22 | 3.56 | 1.46 |
| 21–25 | 0.21 | 5.48 | 2.07 |

A line effect was included in the model to account for genetic differences between the high and low line at the start of the interval. $V_{EW}$ is the environmental variance not including litter and maternal effects.

TABLE 5

**Mean body size at generations 24 and 25 in offspring of parents selected in different directions within the high and the low lines**

| Direction | Line | Generation | Symbol | Mean (g) |
|---|---|---|---|---|
| High | High | 24 | $HH_{24}$ | 19.9 |
| Low | High | 24 | $LH_{24}$ | 18.9 |
| High | High | 25 | $HH_{25}$ | 21.1 |
| Low | High | 25 | $LH_{25}$ | 18.9 |
| Low | Low | 24 | $LL_{24}$ | 18.8 |
| High | Low | 25 | $HL_{25}$ | 19.6 |
| Low | Low | 25 | $LL_{25}$ | 18.1 |

The means are of about 16 litters with the same line and direction, and of about 10 litters with different line and direction. Data from individuals of the low line at generation 24 selected upwards were not obtained.

1988). MEYER and HILL (1991), who analyzed data form a selection experiment on food intake in mice, found that the infinitesimal model did not adequately account for changes in genetic variance induced by selection. The problems associated with applying the infinitesimal model to this type of data are illustrated by a new recessive mutation, *rimy*, which appeared in the high line in generation 14 (KEIGHTLEY and HAWKINS 1991). It was identified by its effect on coat color, but it also reduces body size by about 2 SD, and greatly reduces fitness. Because the mutant appeared in the high line, its incidence remained very low and has probably now been lost from the line.

The purity of the inbred strain presents problems

in this type of experiment. In an attempt to verify the degree of genetic homogeneity at the molecular level, individuals from the high and low line from generation 14 were "fingerprinted" using a molecular probe which hybridizes to various classes of nonecotropic endogenous retroviruses (HOLLAND, WOZNEY and HAWKINS 1983). These stable elements show a high level of polymorphism between inbred strains (STOYE and COFFIN 1988). About 15 loci could be distinguished in C3H/He, and the patterns for high and low line appeared to be identical. The C3H/He pattern was strikingly different from those obtained from a range of other inbred strains. It can be inferred that the mice were inbred at the start of the experiment and not contaminated up to generation 14.

Although the response observed has been very small, the achieved selection intensity has been correspondingly weak, equivalent to no better than selecting the best 80% within families. This is due to the poor breeding performance of the inbred line. For this reason, the effective population size was also greatly reduced. If there had been no breeding failures the predicted effective size averaged over the experiment would be about 54 for within-family selection with equal family size, but the observed effective population size was 22.7. Matings from an outbred mouse population would produce larger families and a smaller proportion would be infertile, and therefore a higher selection intensity would be possible. For example, in FALCONER's (1973) replicated selection experiment on 6 week weight, the within-family selection intensity was about 0.7, compared to 0.38 in the present case. For this reason responses from new mutations would tend to be higher in selection lines involving outbreds.

From a survey of diverse traits and species, LYNCH (1988) pointed out that mutational heritabilities tend to be higher in species with longer generation intervals. Although there are difficulties in comparing different traits across species, it is arguable that, for the present case of growth rate, mutational heritabilities in farm animals are higher than the present estimates for the mouse. The argument depends on a positive relationship between the mutation rate per locus and the generation interval, which would hold under the "molecular clock" hypothesis (ZUCKERKANDL and PAULING 1965). More recent data from the rate of "silent" (neutral) base substitution suggest, however, that the mutation rate per year in rodents is exceptionally high compared to other taxa (LI, TANIMURA and SHARP 1987).

With the mutation rate observed in the present experiment, very high heritabilities would be maintained if the genes involved were selectively neutral. The neutral model predicts that the equilibrium variance in a quantitative trait is $2NV_M$, so for example, with $N = 1000$ and $h_M^2 = 0.01$, the heritability of the trait would be about 95%. There is ample evidence, however, that new mutations almost invariably have deleterious effects on fitness. For example, in a highly replicated selection experiment on bristle score in an inbred Drosophila population (CABALLERO, TORO and LOPEZ-FANJUL 1991), few of the mutations that could be identified became fixed in the selection lines because they reduced fitness as well as changing bristle score. In most cases a reversal in the direction of response occurred on relaxation of selection. In mice, as many as 25% of the visible mutations in all classes compiled by LYON and SEARLE (1989) include an effect on body size (mostly downward) in their description, and most also reduce a major component of fitness.

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