SIZE INHERITANCE IN THE MOUSE

by

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I. INTRODUCTION

1. General Introduction.

In the wild, species of animals have a characteristic body size with limits which are rarely transgressed. As D'Arcy Thompson (1942) notes: "We are accustomed to think of magnitude as a purely relative matter. We call a thing big or little with reference to what it is wont to be, as when we speak of a small elephant or a large rat." Yet, absolute size is very important; all D'Arcy Thompson's work shows that it is not enough to measure the relative dimensions of organisms for it is differences in absolute, and not relative, size which can explain much of the diversity in form observed between different species. The study of allometry, as developed by Huxley (1932), has shown that differences in morphology used for the taxonomic purpose of separating species may result merely from changes of body size.

The genetic variability found within species is generally accepted as the variability which through the agency of natural selection ultimately produces divergent species. In the course of evolution many groups of animals and especially vertebrates show a more or less steady increase of body size (Cope's Rule cited by Rensch, 1948). An understanding of the manner in which size is inherited is therefore fundamental to a study of speciation. In domesticated species the replacement of natural selection by artificial selection has resulted in much larger intra-specific differences in size. An understanding of size can therefore explain much livestock improvement in the past and at the same time provide techniques for making possible much greater improvements in the future. The

investigation/

investigation of size inheritance is thus important both for fundamental theory and for its practical application.

Individuals in a population cannot usually be separated into distinct classes with different sizes: instead, variation is observed to be continuous about a mean value. Each individual is characterised by a measurement which, if the gradations of the measuring instrument are sufficiently fine, is potentially unique. Size has thus to be treated as a quantitative character subject to the laws of quantitative inheritance. It is therefore necessary to give a brief review of current theories of quantitative inheritance before proceeding to a consideration of our knowledge on size inheritance.

2. Quantitative inheritance.

The fundamental problem in quantitative inheritance is to find the relative importance of the various causes of the observed variation, and the first broad division which can be made is into heredity and environment. Prior to the rediscovery of Mendel's work this initial step was impossible owing to the prevailing theory of blending inheritance. In terms of this theory Galton formulated "Laws of Inheritance" and illustrated these with observations on the metrical characters of human populations. This work was continued by Pearson and his pupils and resulted in the development of mathematical methods for the description of continuous variation which are still of great value. With the rediscovery of Mendel's work in 1900, there seemed at first to be no place for continuous variation in Mendelian inheritance. The success of the Mendelian scheme in explaining the inheritance of qualitative characters, such as the coat colours of mammals, caused the eclipse of the rival school of Biometry. There was no doubting that inheritance was particulate and this fact had to be reconciled with the continuous variation of some characters, such as stature in man, which were undoubtedly inherited. In 1909 Johannsen published his experiments carried out with pure lines of beans and showed that environmental agencies could cause continuous variation and that a distinction was necessary between phenotype and genotype. In the same year, Nilsson-Ehle and East, working independently, showed that a large number of Mendelian factors

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each with small effect but capable of acting cumulatively could give rise to continuous genotypic variability. This multiple factor hypothesis received increasing substantiation and in 1918 Fisher was able to show how even the human data collected by the Pearson school fitted in with this extension of Mendelian inheritance. The following decade saw the mathematical consequences of Mendelism examined by Fisher, Haldane and Wright, each using different methods for his investigation. The multiple factor hypothesis came to be generally accepted and forms the basis of present-day theory.

In his 1918 paper Fisher (1918) also attempted to partition the genetic and environmental sources of variation and, carrying the analysis a step further, divided the genetic component into a portion due to the additive action of genes and a portion due to dominance. This partitioning of the sources of variation is still a current problem in quantitative inheritance and the methods of analysis in use derive from those used in the first place by Fisher (1918) and Wright (1921). Fisher's approach has found its use in dealing with plant material and Fisher, Immer and Tedin (1932) showed how second and third degree statistics might be utilised. These statistics, especially those of the third degree require large numbers for their estimation and consequently are more useful for plants than animals. This method has been fully described by Mather (1949) and further extended to allow for linkage in accordance with his Polygene Theory. For crosses between pure lines of plants it appears to be a valuable

method/

method of analysis.

The sources of variation in animal material have been analysed by methods originally developed by Wright (1921) using his method of path coefficients. He was able to take into account the effects of assortative mating and thus his method could readily be applied to domestic animals. Lush and his pupils have adapted and extended Wright's methods in the course of a large volume of work on domestic animals. Particular attention has been given to the determination of "heritabilities" or the fraction of the total variance which can be attributed to the additive effects of genes. Following detailed statements by Wright (summarised 1942) on the evolution - ary consequences of Mendelian inheritance and natural selection, Lush and his pupils have examined breeds and herds of farm animals as populations subject to artificial selection for economic characters, and have brought to light the influence which breed structure and different methods of selection can have in the improvement of livestock. Apart from its economic value the work of Lush's school has a fundamental scientific value. The analysis of hereditary variation into its component parts is still a very real problem. The extent of variation caused by the dominance and epistatic interaction of genes are in general completely unknown and the measurement of these factors is a complex problem awaiting solution.

Two outstanding experiments in which a very full analysis/

analysis of the variability of a character has been attempted are those of Mather (1949) and Chapman (1946). Mather employed second and third order statistics to analyse the sources of variation in ear conformation found in a cross betwen two varieties of barley. He found that the variation due to dominance was important but that epistasis was negligible. Chapman, working with the response of rats to injections of gonadotrophins, used the differences between the correlations of different relations to separate maternal effects and variance due to dominance and epistasis. In his random-bred population he found that additive genetic variance could account for practically all the genetic variance present. This result has been confirmed by selection experiments (Kyle and Chapman, 1948). Both Mather's and Chapman's experiments were, of necessity, carried out on a very large scale and it is unlikely that many experiments on either plan will be possible in the future with laboratory animals, while with larger animals such an undertaking would be completely impracticable.

Full use, however, does not appear to have been made of the opportunities offered by laboratory animals for the study of quantitative inheritance. The rather anomalous result of this fact is that relatively more is known about the inheritance of quantitative characters in domestic animals than in laboratory animals. Since laboratory animals are so much better known in other respects, this deficiency requires rectification if only to prevent lack of knowledge in one field hindering progress in other/

other fields. It will, therefore, be convenient to review the literature on the inheritance of body size first of all for laboratory animals and then for domestic animals, mentioning work on the inheritance of other characters where this is relevant.

3. Review of literature on size inheritance.

a) The inheritance of body size in laboratory animals.

The multifactorial hypothesis of the inheritance of quantitative characters although first based on results with plant material was rapidly substantiated by experiments on animals. The quantitative character chosen for study was usually a linear measurement or body weight. In general the experiments consisted in crossing two breeds or varieties differing as widely as possible in the character selected for study to produce an intermediate F_1 generation. An F_2 generation was raised and an attempt made to demonstrate greater phenotypic variability in it than in the F_1 generation. Further, the underlying genotypic variability was shown by selecting animals from various parts of the F_2 distribution and mating them inter se or making backcrosses to one parent. In order to trace the developments from this period of experimentation onwards, it is convenient to make an artificial division and to treat the important laboratory animals separately.

Rats

In spite of their popularity as laboratory animals, rats have been little used in the study of size inheritance probably owing to the lack of very divergent strains. Classical inbreeding experiments by King (1918) studied the effect of inbreeding on various characters in albino rats and found no decline in size but the inconstancy of the environment in these experiments prevents the complete acceptance of King's results

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at their face value. The heterosis in size found on outcrossing King's rats as for instance in Livesay's (1930) experiments, also suggests that inbreeding depression was not absent in King's experiments but merely concealed by an improving environment since it is generally accepted that heterosis and inbreeding depression are merely different manifestations of the same phenomenon.

Rabbits

Rabbits appear to have been the favourite material for early experiments on size-inheritance. Use of breeds differing in size were made by Castle (1909), MacDowell (1914), Punnett and Bailey (1918), Kopec (1924), Pease (1928) and Robb (1929). The most important work emerging from these earlier experiments were studies by Castle and others on the differences in growth rate which produced adult size differentiation. Castle and Gregory (1929) were able to demonstrate that differences in the size of large and small races of rabbits were due to a greater rate of cell division in the former and could be detected in the morula stage. This difference was taken to the biochemical level by Gregory and Goss (1933) who found differences in the glutathione concentration in these large and small races at birth. Castle's work led him to question the nuclear control of size and to suggest cytoplasmic control. This suggestion was short-lived in the face of accumulating evidence for the multiple factor hypothesis of genic control. An allied view put forward previously by Castle (1914) that size genes were mostly general in their effects, affecting all

parts/

parts of the body, was also strongly challenged. Sumner (1923) from measurements of various species of Peromyscus held that the majority of differences in size were due to size factors affecting only localised parts of the body. Wright (1932) was able to show that a compromise between these two extreme views was indicated by Castle's data.

The Guinea Pig

Castle was also responsible for the initiation of work on size inheritance in guinea pigs. Detlefson (1914) reported on one species cross of guinea pigs and Castle (1916) on another species cross. The most important development with this animal was started in 1906 by the U.S. Bureau of Animal Industry. Twenty-three lines of guinea pigs were set up and each was continued by brother-sister mating. Of these twenty-three original lines five survived to be reported on by Wright in 1922. Subsequently a large volume of work has been published by Wright and his collaborators, notably Eaton and Haines. The quantitative characters such as growth rate, fertility etc. have been examined and described in great detail both for inbred lines and for crosses between them. These experiments were more important than indicated by their intrinsic value, because the statistical methods especially developed by Wright for the analysis of these results have found use in much wider fields of experimentation. In particular, the method of path coefficients developed by Wright clarified much confusion between correlation and causation.

Mice/

Mice

The earliest work on the inheritance of body size in mice after the rediscovery of Mendel's work was carried out by Sumner (1910) using a stock of non-inbred albino mice. He investigated the effects of two different environments, hot and cold temperatures, on various measurements of size and found that the cold temperature produced mice with shorter tails and feet. Furthermore, when these modified mice were returned to the hot environment they produced offspring also with shorter tails and feet, though the difference was smaller. Sumner, unlike some of his contemporaries, did not immediately regard the reappearance of such modifications in the next generation as proof of the inheritance of acquired characters, but as merely important maternal effects. This work was criticised on the grounds that the heterogeneous stock used by Sumner might have been subject to selection, an objection which Sumner was able to answer by pointing out the high intensity of selection which would have been necessary to produce the differences which he observed. Sumner's experiments would seem to deserve repetition using modern experimental design, since persistent maternal effects constitute possible factors causing variation in quantitative characters.

Sumner must also be credited with the first determination of the heritability of a character in the house mouse. In 1915 (Sumner,1915) he presented data to show a significant correlation of 0.14 between parent and offspring for the ratio of tail length to body length calling this correlation a "Coefficient of heredity" in Pearson's nomenclature. After

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these experiments Sumner took up the problem of speciation in Peromyscus, and, although his studies were quantitative they provide little evidence with any direct bearing on the problem of size inheritance.

Green (1931 et seq.) studied in detail a cross between Little's inbred dba Mus musculus and wild Mus bactrianus. The birth weight of the F_1 generation in reciprocal crosses seemed to be almost independent of the maternal constitution - an observation which was in agreement with the results of Vetulani (1930). Backcrosses were also made to each parent, and where this parent was Mus musculus segregation occurred for the colour genes, a, b and d, but there did not seem to be any association of birth weight with any colour gene. Green (1931) also investigated the inheritance of adult size as measured by weight at 180 days, and obtained the results expected for a multifactorial character. But in the backcross to the musculus parent he obtained a significant association between brown and greater body weight. Subsequent work (Castle, Gates, Reed and Law, 1936), (Feldman, 1935), made it appear probable that the greater size was due to a physiological action of the brown gene. The effects of colour genes on size will be reviewed in a later section. (Section VII).

Occasional differences in reciprocal hybrids occurred in the experiments mentioned above, and have been variously interpreted as being due to 'maternal effects', cytoplasmic inheritance, or sex-linked genes. Grüneberg (1943) protests against/

against these additional postulates to explain differences in one sex only, and one can but concur, although where both sexes are affected genuine 'maternal effects' must be allowed as an explanation. Nevertheless, differences in one sex may be marked (Marshak 1936) and clearly not due to chance.

Goodale (1938, 1941) described a selection experiment for body size of Mus musculus which is of great importance. With a stock of commercial albinos he was able to increase body size by 70% in twenty-eight generations of selection using a form of progeny testing, despite the fact that the whole stock derived from only sixteen foundation animals. The rate of progress showed no signs of diminishing, a most surprising result in view of the results of other selection experiments. But, more important than these results was the demonstration of a valuable method for tackling a problem of quantitative inheritance. The apparent motivation for this experiment is better stated by Goodale (1937) in connection with another selection experiment. Having obtained a recessive gene for head spotting in the house mouse, Goodale set up two stocks containing this gene in a homozygous condition. One stock was bred at random while the other was selected for large sized head spots starting with only five foundation animals. All the three possible crosses were made between the selected, the unseledted and an inbred stock. These showed that the inbred stock did not carry modifiers increasing head spotting and yet in a quite small F_2 of the cross selected by inbred the parental extremes were recovered. Since all the genes for a large size of spot were present in

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the five foundation animals (apart from favourable mutations in the intervening generations), Goodale was led to suggest that "in the foundation animals, a series of alternating but sometimes staggered plus and minus (or zero) genes were located in one pair (possibly more) of chromosomes and that the minus genes almost balanced the plus genes, thus giving rise to the limited areas described." The fact that the size of the spot increases under the pressure of selection is referred to the accumulation of cross-over classes containing more plus than minus genes. Goodale quotes in support of this view experiments by Payne with Drosophila, who, starting with a brother-sister mating, was able to increase the number of scutellar bristles far beyond the limits expected from the low initial variability. The consequences of such a system with a large number of plus and minus genes each with small effect have been elaborated by Mather (1941) in his "Polygene Theory".

Thus, Goodale's results with selection for large body size would lead to a similar conclusion in the absence of other explanations. The continued progress in this experiment, however, at once invalidates any conclusions on the number of genes involved, as arrived at by Goodale (1938) by consideration of the range of weight attained. Any such estimate of the number of genes based on so many assumptions and measured by a statistic with such a large standard error as that of the range can have little value.

MacArthur/

MacArthur (1944) has reported in much more detail upon another selection experiment, again on body size in the house mouse. Starting from crosses of six inbred strains, selection was made for both large and small body size. In twenty-six generations, no limit was reached in selecting for small size, but the rate of progress in the large line became very low. This experiment was intended to throw light on the problem of whether size genes acted additively or geometrically, and, from the early progress in this experiment MacArthur found evidence for a geometric action of size genes which was in accord with his earlier work on tomatoes (MacArthur and Butler, 1938). During his mouse experiment MacArthur (1944) was able to study changes correlated with changes in size, in particular changes in the allometry of the body (MacArthur and Chiasson, 1945). By various methods the heritability of body weight at 60 days was estimated to be about 25% in the earliest generations decreasing to about 10% at the end. This decline in heritability is a finding common to most selection experiments (though not to Goodale's) and yet it receives no special mention. The possible causes of this decline are not discussed although of such obvious importance. Both Goodale and MacArthur express the belief that it should be possible by selection in the laboratory to produce mice as large as rats but a declining response to selection would prevent the achievement of this goal. The causes of a diminishing response to selection clearly require elucidation.

b) <u>The inheritance of body size in farm animals</u>.

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The results of the large amount of analytical work done by Lush and his followers in determining the heritability of various characters in farm animals have been collected by Phillips (1947). No attempt at a critical review was made despite large differences between the values obtained by different workers for the same character. The reason for this omission is not far to seek. In a large number of cases, authors have not given the fiducial limits of their estimates or presented their results in sufficient detail to allow calculation of these limits. Where the estimate of heritability has been based upon a correlation or regression coefficient this omission is inexcusable. Further, estimates of heritability are presented without explicit statement that they are based on insignificant regression coefficients, e.g. Stonaker and Lush (1942).

There are, of course, notable exceptions to this general criticism and several authors have made valuable contributions to the understanding of size inheritance in farm animals. Studies by Hazel and co-workers (Baker et al., 1943) on the importance of heredity and environment in the growth of pigs are particularly illuminating. From an analysis of variance they were able to separate components of variance due to three sources: the heredity of the individual, the environment peculiar to the individual and the environment common to littermates. The relative importance of heredity (the variation due to this source as a fraction of the total) increased from 6% at birth to 15% at weaning and to 25% at 168 days. Accompanying this change there was a steady decline in the relative importance

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of the environment common to litter-mates while the environment peculiar to individuals showed an initial decrease followed by an increase. Analysis of the correlations between growth rates at different ages into genetic and environmental components showed that although the genetic component of variance was low, the genetic correlation was high indicating that the growth rate at different ages was due in large measure to genes with persistent effects. On the other hand environmental correlations were low, thus leaving little room for possible persistent maternal effects.

Substantially the same trends were obtained by Nordskog et al.(1944) who, however, found that heredity did not have any effect until after weaning. A more interesting confirmation of the trend of estimates of heritability with age was given by Krider et al. (1946). In this experiment estimates obtained from the resemblance between relatives were combined with estimates from the results of four generations of selection for slow and rapid growth, and the agreement was found to be good. The differences between the slowly and rapidly growing lines were however very erratic from generation to generation and the first generation of selection, in particular, produced a large difference between the lines. Another selection experiment with pigs, this time for efficiency of gain, has bee reported by Dickerson and Grimes (1947). The two lines separated sharply in the first generation of selection and four more generations of selection produced no more divergence.

Nevertheless/

Nevertheless, the results from selection are found to agree reasonably with the expectation from the resemblance between progeny and parents within lines. Part of the lack of progress is attributed to a negative genetic correlation between the lactation of females and economy of gain (Dickerson, 1947), any improvement in a pig's inherent economy of gain inherited from its mother being cancelled by her poorer lactation. Such situations might account for some of the differences in estimates of heritability obtained by various methods.

Poultry are the most favourable farm animals for the study of quantitative inheritance. The large number of progeny obtainable from a single mating and the absence of important maternal effects simplify breeding experiments. Despite these advantages much effort in this field has been ill-directed, the notable exception being the work of Lerner and his collaborators who have made full use of the methods employed by Lush. Lerner's book "Population Genetics and Animal Improvement" gives a full account of themanner in which Lush's methods may be used and at the same time points out possible sources of error. For body size as measured by shank length Lerner (1943) found a heritability of 38% in his flock of White Leghorns. The experiment on selection for egg production reported by Lerner and Hazel (1947) may also be mentioned since it provides one of the few cases in which a check is available on heritability determinations. In this experiment the results of eleven years of selection for egg production were found to agree with the expected gain calculated from estimates of heritability.

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This a posteriori analysis, however, is not altogether convincing in view of an outbreak of disease in the middle of the experiment which caused a great drop in average egg production.

To sum up the work on farm animals it may be pointed out that there is no lack of estimates of heritability of measurements of size and other economic characters but the nature of the material has prevented many checks on the accuracy of these estimates.

4. Methods of investigating quantitative inheritance.

In this section it is not intended to detail all the methods which have been used to determine heritabilities but merely to indicate the representative methods.

The oldest method of measuring the extent of genotypic variance is to measure the excess variance of an F_2 over the average variances of an F_1 and parent pure lines. Owing to the paucity of pure lines in animal material this method is of little use.

With animals practically all determinations of heritabilities are based upon the resemblance between relatives, and since some relatives tend to have similar environments it is necessary to avoid mistaking environmental correlations for genetical correlations. Thus, the use of identical twins which have had at least a uterine environment in common is liable to lead to an overestimate of heritability. Similarly, environmental correlations are liable to bias estimates of heritability based on the correlation between parent and offspring and that based on maternal sibs. With care, however, serious bias can be avoided as, for instance, in using the correlation between paternal half-sibs.

Even when environmental correlations have been discounted there are further sources of error to be taken into account. The resemblance between relatives besides being due to the additive action of genes may also be due to dominance and epistasis, the influence of these factors depending upon the/

the exact relationship. This provides a possible method of evaluating the importance of dominance and epistasis but in general the sampling errors of correlations between different relatives have been and are always likely to be too large to reveal any differences. A more promising method of finding the extent of non-additive genetic variance is by making di- or poly-allel matings, and estimating the variance due to interaction. Such experiments are, however, not easily carried out and require to be done on a large scale.

For estimating the extent of additive genetic variance selection probably provides the most powerful method of analysis and the method least liable to error. Adequate environmental control is necessary owing to the long period over which the experiment takes place and is best provided by selection in opposite directions. The estimate of heritability will be measured by the resultant divergence between the means of the two lines as a fraction of the divergence sought after. This method has the great advantage that the results are measured by first degree statistics (means) and, are accordingly, expected to be more accurate than determinations made using second degree statistics (variances, covariances etc.). The sampling variance of heritability estimates obtained by selection has not yet been calculated and until this is done it is not possible to assess the statistical precision of this method, although it is expected to be high. Nevertheless, the mere fact that the estimate is based on the results of selection makes it potentially more valuable for the practical purpose of improving farm animals than estimates extending over only one or two generations.

5. The improvement of farm animals.

Lush's book "Animal Breeding Plans" presents plans for the improvement of farm animals in accordance with the present knowledge of quantitative inheritance. Before complete faith is vested in these schemes it is necessary to note certain assumptions inherent in the basic deductive analyses of Fisher and Wright.

In his 1918 paper Fisher set out "to ascribe to the constituent causes (heredity and environment) fractions or percentages of the total variance which they together produce." It is a basic premise of quantitative inheritance (as for instance set out by Lerner, 1950) that the phenotypic variance of a character is wholely determined by genetic and environmental factors and by the interaction between them. Fisher's neglect of genotype-environment interactions evoked criticism from Hogben who went to the other extreme and despaired of measuring the relative importance of heredity and environment. The true situation for most characters probably lies between the two extremes and its elucidation is awaited. Meanwhile Lush (1945) has neglected genotype-environment interactions purely on empirical grounds. A paper by Haldane (1946) on the theoretically possible types of interaction shows that the problem is not being neglected despite the convenience of so doing.

Further detail in the balance sheet showing the causes of variation is desirable. The environmental components can usually be separated by an analysis of variance; the genotypic components are not so easily obtained. This is largely/

largely due to the fact that the complications involved in the action of numerous genes have to be forced into a formal analysis if the situation is to be analysed at all. If the formal analysis of genotypic variance into additive genetic variance, and variance due to dominance and epistatic deviations presents difficulties and anomalies this is hardly surprising for even our choice of scale is at best empirical. Yet the division of genotypic variance into at least additive and non-additive parts is of great importance in deciding upon the best method for the improvement of farm animals.

If the genotypic variance in the character to be improved is mostly additive genetic variance then improvement by selection will be the best plan. Should, in this case, the heritability be high, mass selection will be an efficient way of making improvement but should the heritability be low, selection will be more efficient if the performance of relatives is taken into account, as in family selection or progeny testing. If, on the other hand, there is little additive genetic variance but much non-additive genetic variance and more especially variance due to epistasis, then a system of inbreeding and crossing is called for.

At present the relative importance of additive and non-additive gene action in determining economic characters of farm animals is unknown. There are, however, several indications that non-additive effects are not important. Experiments, especially designed to measure interaction or "nicking" have failed/

failed to show that this is an important source of variation.

Lerner (1945) found in a flock of White Leghorns that non-additiveness contributed only about 1% to the total variance in age of sexual maturity. Hazel and Lamoreux (1947) also working with White Leghorns found a similar small contribution of nicking effect to the total variance in body weight at 22 weeks of age. In dairy cattle, Seath and Lush (1940) working on yield and Johnson, Bartlett and Copeland (1940) working on total butterfat production found little evidence for nicking. In pigs, the performance of inbred-crosses is not outstandingly superior to that of outbred pigs and this fact may also point to the small importance to be attached to non-additive effects, although Dickerson (1949) has come to the opposite view.

If non-additiveness is not important farm animals can best be improved by selection. In the past, experiments on selection with the larger farm animals have only occasionally been possible owing to the time and expense involved. But, they are nevertheless of great importance. Before increasingly complex breeding plans are made depending on estimates of the components of variation, more evidence is required as to the accuracy of these estimates. The number of links in the chain of deductive reasoning are increasing and verification of the original partitioning is needed. Selection experiments provide the critical test for the presence of additive genetic variance. Adequate corroboration has not been produced by Lush and his school. The two selection experiments with pigs have not been continued/

continued for a sufficient number of generations to make the results valuable and only the experiments of Lerner and Hazel (1947) and Kyle and Chapman (1949) can be accepted as providing satisfactory corroboratory evidence of internal consistency. Lerner and Dempster (1949) found it necessary to put forward a plea for the empirical verification of the calculated efficiencies of selection indices and have since published such a verification for family selection as compared with mass selection (Lerner, Cruden and Taylor, 1949). It would therefore appear that in the field of animal breeding emphasis is changing from the determination of estimates of heritability and calculation of the expected rates of progress with various breeding plans to the verification of these calculations by selection. In the future it is to be hoped that realisation of the practical value of the end-products of selection experiments will make it possible to carry these out on a more adequate scale than at present.

6. Present experiment.

The foregoing review has shown the extent of our knowledge of size inheritance. This knowledge derives from experiments with various species of animals, and providing that there are no unsuspected barriers to the integration of this fragmentary evidence from different species, the broad outlines of size inheritance appear to be well understood. The multiple-factor hypothesis forms an adequate explanation of size inheritance and is generally acceptable to scientists outside Russia. We know something of the mode in which size genes produce differences in adult size and of the extent and duration of their action. The extent of genetic variability for various measurements of size has been estimated in many species and suitable techniques for making these estimates have been developed.

Of the many problems connected with quantitative inheritance the one chosen for the present study concerns the long-term response to selection. The effects of selection over a period of a few generations are fairly well understood and present few difficulties. The results of long-term selection, on the other hand, have received little attention apart from theoretical discussions on the effects of natural selection (e.g. Fisher, 1930). Yet the practical importance of this knowledge to the improvement of the present breeds of livestock by selection can hardly be overrated. Most breeds have already been improved by many generations of selection and though

selection/

selection still appears to be the most promising method of making continued progress in the future, more experimental evidence is needed on this point.

The problem may be briefly stated thus : is the initial rate of response to selection maintained indefinitely, or is there a limit to the improvement which can be brought about by selection? If the rate of response falls off in later generations what is the cause of this? And, can this barrier to improvement be overcome?

The number of selection experiments which have been carried out must be very large but only a small percentage have been continued for a sufficient number of generations to show whether or not a declining response to selection was obtained. *

In Drosophila, where it is possible to obtain a very quick turnover of generations, there are several experiments which show that a selection limit is reached after a number of generations. Payne's (1918) selection for extra scutellar bristles was marked by periods of sudden response followed by stability which Payne attributed to mutation but which Mather (1941) ascribes to the breaking up of polygenic combinations by crossing over. But whatever the explanation of these irregularities a final limit appears to have been reached. Payne's later experiments on selection for bristle number in "reduced" flies (Payne, 1920) showed the attainment of a limit in both plus and minus lines after about seventeen generations. Selecting for high and low facet number in Bar-eye flies, Zeleny (1922) found no increase in the divergence between his lines after the twenty-eighth generation/

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* See review by Goodale (1938a)

generation. Two other workers, Sturtevant (1918) using bristle number in Dichaete flies, and MacDowell (1917) using bristle number in a stock homozygous for a recessive bristle increaser, also found that these quantitative characters could not be changed beyond certain limits by selection. More recent experiments by Mather (1941) in which selection was made for number of abdominal chaete show, in the majority of lines, if not a limit, a declining response to selection. Reeve and Robertson (1950) made selections for thorax length and wing length and found that although the response was at first slow and steady (see Reeve and Robertson, 1949) a selection limit was eventually reached.

Apart from the work on Drosophila little evidence is available. Most selection experiments with domestic animals cover too few generations but the selection of poultry for high and low egg number started at Cornell in 1913 (Hall, 1934) showed that while it seemed to be possible to increase egg number slowly over the nineteen generations reported, there was very little response to selection for low egg number. MacArthur's (1949) selection experiment for large and small size in the mouse showed a declining response to selection in both lines whilst Goodale's selection for large size in the same species showed no signs of a declining response. The results of both these experiments will be more critically examined in the next section but it is clear that Goodale's result is not a general finding and the majority of long-term selection experiments do show a declining response to selection and if continued for a sufficient/

sufficient period of time a selection limit is reached. This conclusion is of great importance in the field of animal breeding where the response to selection must be a primary consideration.

The main object of the present experiment is to investigate the causes of the falling off in response found in selection experiments. The possible causes of this falling off can be divided into two main categories :-

1) A loss of genetic variance produced by either inbreeding or selection, or by these two factors in combination;

2) An approach to a "physiological barrier" which could act either by preventing animals from exceeding a certain limit or by natural selection, those animals which approach nearer to the limit leaving fewer descendents than those further from the limit.

The first explanation, loss of genetic variance, can be investigated by the following method: crossing two long-selected strains in which the response to selection has declined or ceased altogether gives a cross-bred population which is expected to contain new genetic variance and so should show a renewed response to selection.

Two stocks of mice which had both been selected for large body size and which seemed well suited to this purp se were available, namely those of Goodale and MacArthur. The detailed reasons for considering these lines suitable will be presented in the next section and it is here only necessary to anticipate a conclusion drawn in that section which is that the crossing of these two lines would be expected to produce new genetic variance. The problem is thus studied by continuing selection/

selection in these two lines of mice and comparing the results with those obtained by selection in a new strain made by crossing these two lines.

These selection experiments were all carried out with the objective of producing the greatest change in body size in the shortest possible time. Theoretical considerations led to the adoption of certain methods of selection which were expected to give a maximum rate of progress under selection and so are called "efficient" hereafter, although less efficient methods from this point of view would have given more information on the relative importance of maternal and environmental factors for growth. When sufficient data had been accumulated it was hoped to be able to construct a more efficient index for selection and if necessary to alter the structure of the population in order to obtain the maximum increase of body size by selection.

Finally, a check on the mode of analysis by selection can be made. The individual weights of each mouse in the experiment are available for the calculation of correlations between relatives from which alternative estimates of heritability can be derived. Disagreement between these estimates may throw doubt on the basic assumptions of the analysis. Agreement, on the other hand, while not giving direct verification of these assumptions can at least demonstrate the internal consistency of this analysis.

II. SUITABILITY OF STOCKS FOR THE PRESENT EXPERIMENT.

Goodale's and MacArthur's results are of prime importance to the present thesis. It is necessary to show that both stocks were exhibiting a declining response to selection and also to give reasons for supposing that the crossing of these two stocks will produce new genetic variance.

MacArthur's results (Fig.9) show clearly a declining response to selection in his Large Line. By generation 18 it seemed that a ceiling had been reached since there had been no increase in mean weight for eight generations. Then, inexplicably, there was a sudden rise and fall but the general trend was once more upwards. The mice of the twenty-sixth generation, obtained from Professor MacArthur in 1948, were presumably a somewhat selected group but males and females had mean weights about equal to mice of the twenty-third generation, the last point on the graph. The diminishing response to selection is also brought out in Table 6 of MacArthur (1949). It can, therefore, be concluded that MacArthur's Large strain is near to a limit of selection and the response to further selection would be very small.

Goodale's results (Fig. 9) on the other hand do not show a declining response, but instead an almost linear increase in size. This, it should be pointed out, is a unique result for a long-term selection experiment. Three possible explanations may be offered. Firstly, the culling of unpromising young at weaning or the equivalent procedure of remating parents/

parents with large offspring at weaning, will tend to bias the mean 60-day weight upwards but clearly this will not affect the shape of the response curve providing the extent of these practices does not alter. Secondly, the use of the progeny test could be made to produce a steady response merely by waiting for animals which produce a certain increase over the current mean. A declining response to selection might be concealed by a lengthening generation interval, but there is no internal evidence in Goodale's papers for this suggestion. Thirdly, and most probably, Goodale's graph, although possibly biassed by the previous two considerations, gives a fair record of his progress up to 1942. But a declining response appears to have been encountered after this date. Dr. Goodale,in a private communication to Dr. Falconer, has given the mean weights for the thirty-eighth generation of his stock and these are approximately the same as those for the twenty-eighth generation given on the graph. Selection for size over this period has not been as intense as previously as it was necessary to intercalate a period of selection for fertility, but, nevertheless, it is clear that the response was less rapid than before.

Thus, both Goodale's and MacArthur's mice appeared to be approaching a selection limit. This conclusion was substantiated by the continued selection of these stocks described later and their use for an investigation into the causes of a declining response to selection was therefore justified.

The/

The second requirement for the success of the present experiment is that the two stocks should produce new genetic variance when crossed. For this to occur the two lines must contain different size genes and the evidence on this point must be examined. The two stocks are of independent origin, Goodale's Large Line originating from sixteen albino mice obtained from a commercial breeder, and MacArthur's from crosses between six inbred strains of laboratory mice. Provided that body size is not controlled by a small number of genes, each line can therefore be expected to carry different size genes. There is visible evidence of this since the two lines differ in conformation, and, although of approximately the same weight, differ in the proportions of muscle, fat and skeleton which go to make up this weight. Goodale's mice are long and lean compared to MacArthur's which are shorter and very much fatter. This difference is apparent in Fig. 5 where the great girth of MacArthur's Large Line male can be seen. There is therefore good reason to suppose that the two lines owe their size to different genes and will produce new genetic variance when crossed. These long selected stocks of mice are thus very suitable for the present experiment.

III. METHODS

It has already been pointed out in the introduction that the value of selection as a tool in the investigation of quantitative inheritance depends upon the efficiency of the method of selection employed. The theoretical considerations involved are best discussed in concrete terms with reference to the method of selection actually employed. It will, therefore, be convenient to describe briefly the practical procedure adopted before giving all the reasons why this particular scheme was adopted.

The biometrical analysis of a laboratory population of mice is greatly facilitated by ruling that successive generations are kept quite distinct and that even outstanding animals are not used in more than one generation. This rule was adhered to with one exception in the fourth generation of MacArthur's large line when, owing to sterility, five animals which had been mated in the previous generation were used again to maintain the line. At first an attempt was made to raise generations in all lines contemporaneously so that seasonal differences might be discounted. This, however, entailed a long delay in mating the more fertile cross-bred lines and as there was little indication of any regular seasonal effect these two lines were allowed to forge ahead together from the fourth generation onwards. The parent lines were mated contemporaneously throughout the experiment.

The/

The parent lines were maintained for the first three generations by six matings of one male and one female, raising two litters from each. Sterility made it necessary to put up more matings and the number was increased to twelve. At the same time it was decided to raise only one litter from each mating since this had the theoretical advantage of giving a greater expected gain per annum and a practical advantage in requiring less space. In the cross-bred lines the first eight matings to breed were used; in the parent lines, all matings that produced and raised litters, the number of these never being more than eight.

In the first three generations of the parent lines females were allowed to suckle all the young they produced up to a total of twelve - above this number newly born young were removed. The cross-bred lines proved to be so fertile that nearly all litters had to be reduced and so litters born of F_1 parents were reduced to ten. In subsequent generations this number was altered to eight and not only were litters reduced to eight but also, where possible, litters of less than eight were made up to eight by fostering extra young. This procedure was adopted in the parent lines at the same time.

Weights and measurements were taken as follows:

1) Collective birth weight of all living young (prior to any reduction);

2) Collective 12-day weight of litter (as a measure of the mother's lactation and so including fosterlings);

3) Individual 3-week weight and tail length (fosterlings discarded);

4)/

4) Individual 6-week weight and tail length;

5) Individual 60-day weight and tail length of animals not already mated.

Selection for body size was made solely on 6-week weight and not as in Goodale's and MacArthur's experiments on 60-day weight. This change gave a quicker turnover of generations and made full use of the early sexual maturity of the mouse. In addition it was hoped that mating at an earlier age might combat the sterility encountered in MacArthur's mice which became excessively fat with age. Selection was limited to a choice of the largest animals within litters and within sexes, i.e. in mathematical terms, an animal was selected on the deviation of its weight from the litter mean for that sex. No attention was paid to the level of the litter mean and an attempt was made to take an equal number of animals from each litter. Any conscious differential selection between matings was thus based on fertility and not on the mean weight of offspring.

Selected males and females were paired according to relationship, those which were least related being mated together. The labour involved in computing coefficients of relationship of animals from all matings was lessened by the use of a method similar to that described by Cruden (1949). The genetic co-variance between all animals was calculated for the first generation and tabulated. If in this generation two matings, of A with B and C with D, produce offspring X and Y respectively, then the genetic covariance of X and Y may be simply/

simply obtained from the relationship :

$$\text{cov}_{XY} = \tfrac{1}{4}(\text{cov}_{AC} + \text{cov}_{AD} + \text{cov}_{BC} + \text{cov}_{BD})$$

This simple arithmetic procedure was carried out for each generation. To convert the tabulated values of genetic covariance into genetic correlations it is merely necessary to know in addition the genetic variances of individuals since, from Wright (1921) :

$$\text{Coefficient of relationship} = \text{Genetic correlation}^{*} = \frac{\text{cov}_{XY}}{\sqrt{(\text{var } X)(\text{var } Y)}}$$

In practice the covariance of individuals were used in planning matings since changes in variance brought about by inbreeding were fairly uniformly distributed within any one generation. The inbreeding coefficient of any animal could be simply obtained since it is one half of the genetic covariance of its parents. The method described by Cruden involves calculating the inbreeding coefficients of young from all possible matings and so is formally identical with that described here.

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* This term has also come to be used in another sense. Here it refers to the genetic resemblance between individuals but it is also used to describe the genetic variability common to two characters (see Section VII).

IV. THEORETICAL CONSIDERATION OF THE METHOD OF SELECTION EMPLOYED.

1. Types of selection.

a) Mass selection.

The simplest type of selection is mass selection in which individuals are selected from the entire population solely on the basis of individual phenotypic merit. The expected advance under selection is found from theequation :

$$\Delta I = h^2 \Delta P$$

where ΔI = average superiority of offspring

h^2 = heritability = $\frac{\text{genetic variance}}{\text{phenotypic variance}} = \frac{\sigma_G^2}{\sigma_P^2}$

ΔP = average superiority of selected parents, i.e. the selection differential.

Provided the population is not increasing or decreasing in numbers an average value for the selection differential can be obtained from Fisher and Yates (1949) Table XX which gives the average deviation from the mean of the largest observation in a sample of size n drawn from a normally distributed population. This deviation, i, is given in standard deviation units and so has to be multiplied by the standard deviation of the phenotype, σ_p, to obtain the value of the expected selection differential in ordinary units.

$\Delta I = h^2 \Delta P$ becomes

$\Delta I = h^2 \, i \, \sigma_p$ and since $h = \frac{\sigma_G}{\sigma_P}$ this equation can conveniently be expressed as

$\Delta I = i \, \sigma_G \, h$

In/

In a given situation σ_G will be constant and i fixed by the necessity of keeping the population at a constant size, so that h is the only variable. In comparing the expected rates of progress under various systems of mating, therefore, the variable of most immediate interest is the square root of the heritability. The possibility of varying the selection differential is considered in a later sub-section.

b) Selection within and between families.

When the population is made up of a number of families it may be advantageous to give consideration to family merit in making selections. In order to do this it is necessary to carry out the partitioning of the total variance which it is possible to make - into variance within families and variance between family means. These two component variances are further divisible into genetic and non-genetic parts. Thus, the ratio of the genetic variance within families to the total variance within families is the heritability of phenotypic differences observed between the members of a family while the corresponding ratio for the variances between families is the heritability of family averages. A comparison of the square roots of the two heritabilities will indicate whether it is more advantageous to select between the members of a family or between families as a whole.

In the present experiment the family unit is the litter since in general there was only one litter per mating. The total variance of six week weight has been partitioned within and between litters by the analysis of variance and the

results/

results are shown in Tables I, II and III. The pooled values for females of the large cross-bred line from generations 1 - 7 will be used by way of illustration, namely :

Variance within litters - $\sigma^2_W = 3.69\ gm^2$
Variance between litter means - $\sigma^2_L = 3.15\ gm^2$

The heritability of phenotypic differences within litters is easily deduced. If the genetic correlation between litter mates is r and the phenotypic correlation t, then the genetic variance within litters is $(1 - r)\ \sigma^2_G$ and the phenotypic variance $(1 - t)\ \sigma^2_P$.

Therefore - $$h^2_W = \frac{(1-r)\ \sigma^2_G}{(1-t)\ \sigma^2_P}$$

Litter mates are all full sibs and have a genetic correlation, r, of 0.5 as shown by Fisher (1918). The phenotypic correlation, t, can be calculated as an intraclass correlation :

$$t = \frac{\sigma^2_L}{\sigma^2_L + \sigma^2_W} = \frac{3.15}{3.69 + 3.15} = 0.46$$

$$h^2_W = \frac{(1 - 0.50)\ \sigma^2_G}{(1 - 0.46)\ \sigma^2_P} = 0.93\ h^2$$

The expected progress under mass selection is given by :

$$\Delta I = i\ \sigma_G\ h$$

and, therefore, for selection within litters :

$$\Delta I = i\ \sigma_{G_W}\ h_W = i\ \sqrt{0.5\ \sigma^2_G}\ h_W = i\ \sigma_G\ h \times 0.68$$

Selection within litters is thus 68% as efficient as mass selection.

The efficiency of selection solely on litter mean can be deduced similarly:

h^2_L/

$$h^2_L = \frac{(1 + (n-1)\ r)\ \sigma^2_G}{(1 + (n-1)\ t)\ \sigma^2_P}$$ where n = average number in litter = 3.8 (females only)

$= 1.05\ h^2$

The expected progress by selecting between litters on their mean performance is, therefore :

$$\Delta I = i\ \sigma_{G_L} h = i\ \sqrt{0.5}\ \sigma_G\ h = i\ \sigma_G\ h \times 0.72$$

Selection on litter average is thus 72% as efficient as mass selection and practically no better than selection within litters.

c) Combination selection.

In theory it is possible to make a combination selection considering each animal on its own merit and on the performance of the litter to which it belongs. This is achieved by splitting the deviation of its weight from the population mean into a deviation from its litter mean and a deviation of the litter mean from the population mean and weighting each deviation by its appropriate heritability. Calculation of the efficiency of this method using a formula given by Lush (1947) shows it to be only 0.2% more efficient than mass selection. For all practical purposes mass selection would, therefore, seem to be the best method to employ. However, with a small population in the laboratory there is another important consideration which must be discussed, namely the rate of inbreeding.

2. Inbreeding.

With a small laboratory population made up of relatively few matings the rate of inbreeding becomes an important consideration. The effect of inbreeding on a single stock is to decrease the amount of genetic variance which it was shown above enters directly into the formula for predicting improvement.

But, apart from this long-term effect, inbreeding of itself causes a degeneration of many characters. In most animals, including the mouse, inbreeding without selection causes a decrease in body size. The causes of this degeneration is open to debate but the reality of the degeneration is beyond dispute. Fortunately the effect of inbreeding on size can probably be neglected in considering the divergence between two lines selected in opposite directions, the assumption being that the effects of inbreeding are the same on both lines. Inbreeding degeneration of reproductive performance cannot, however, be neglected. Reduction of fertility reduces either the rate at which the population can be replaced or the selection differential which can be obtained leading, in either case, to a diminished rate of progress. Moreover, there is no prospect of this situation improving.

Selection within litters as used in the present experiments results in a minimal amount of inbreeding since the full number of matings from which exigencies of space permit litters to be raised are represented in the next generation.

Any/

Any selection between matings would result in a higher rate of inbreeding. It will be remembered that the least related animals were always mated together so that there was a maximum avoidance of inbreeding. Wright (1921) showed that a regular system of matings between quadruple second cousins (eight in a group) produced 3.5% inbreeding per generation while matings between octuple third cousins (sixteen in a group) produced 1.7% per generation. In the present experiment the increase in the inbreeding coefficient per generation was often higher than 1.7% owing to the number of unproductive matings. The average inbreeding coefficient for each generation is given in Table IV. It will be seen that in the cross-bred lines the increase in the inbreeding coefficient in the later generations is close to/1.7% expected with sixteen parents.

the

3./

3. Breeding system and the time factor.

It is now possible to consider in more detail a factor which up till now has been considered as a constant, namely the selection differential. This is dependent upon the reproductive performance of the population and the number of animals saved must be sufficient to maintain the size of the population. With mice, a possible method of increasing the selection differential would be to raise more than one litter. Two litters, for example, would be expected to produce individuals with a greater deviation from the mean than one litter, and so would have the advantage of giving a greater rate of progress. But this is progress per generation and as the interval between generations will be increased by waiting for two litters the result may be a decrease in the rate of progress per unit of time. The importance of the time factor in deciding upon the merits of various systems of selection has been fully discussed by Dickerson and Hazel (1944) for farm animals and their methods are readily applicable to the present problem. Their basic equation is formulated as

$$\Delta G = \frac{\Delta I}{T}$$

where ΔG = average annual gain
ΔI = average genetic gain per generation
T = average age of parents when their offspring are born, or the average interval between generations.

The choice/

The choice between one or two litters may be discussed in terms of this equation.

The raising of a second litter will increase T so that if ΔI is increased relatively more than T, ΔG will increase. If, on the other hand, ΔI is increased relatively less than T, then ΔG will decrease. The following figures may be taken for T: they are close to those observed in the cross-bred lines. T may be either :

1) 6 weeks (growth) + 2 weeks (to keep all matings in step) + 3 weeks (gestation) = 11 weeks,

or

2) 11 weeks + 5 weeks (to produce second litter) = 16 weeks.

In the first case :

$$\Delta G_1 = \frac{\Delta I_1}{T_1} = \frac{h^2 \Delta P_1}{T_1} = \frac{h^2 \Delta P_1}{11}$$

and in the second :

$$\Delta G_2 = \frac{\Delta I_2}{T_2} = \frac{h^2 \Delta P_2}{T_2} = \frac{h^2 \Delta P_2}{16}$$

Therefore :

$$\frac{\Delta G_1}{\Delta G_2} = \frac{16}{11} \times \frac{\Delta P_1}{\Delta P_2}$$

ΔP, the selection differential, depends upon the average number of young per litter which can be raised. If this number is x, then the average value of ΔP in standard units can be read directly from Fisher and Yates (1949) Table XX entering the column headed n with $x/2$ to obtain ΔP_1 and with x to obtain ΔP_2 *.

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* This value will be slightly too high since what is strictly required is the greatest mean deviation found in two samples of $x/2$. Tables of average value of this deviation do not appear to exist.

Interpolations are necessary when x is an odd integer and these have been calculated allowing for the random distribution of sexes within litters. This effect reduces all the values in Fisher and Yates table slightly. For an example, consider litters of size 5 : to keep the population in equilibrium one male and one female have to be selected from each but the number of each sex within litters of five will be distributed according to the expansion of the binomial $(\frac{1}{2} + \frac{1}{2})^5$ and $(\frac{1}{2})^5$ or 1/32 of the litters would be expected to contain no males, an equal number no females. The best animals to maintain the size of the population would usually be the second best in litters all of one sex which are expected with an equal frequency. Corrected selection differentials calculated on this basis are given below. The corrections are small and other factors such as the variation in litter size which are not so easily allowed for are probably more important.

No. n.	3	4	5	6	7	8	9	10	11	12	13	14	15	16
Expected S.D.	-	.56	-	.85	-	1.03	-	1.16	-	1.27	-	1.35	-	1.42
Corrected	.32	.51	.70	.82	.92	1.01	1.08	1.14	1.19	1.25	1.29	1.33	1.37	1.41

The approximate values of ΔP can now be substituted in the equation $\frac{\Delta G_1}{\Delta G_2} = \frac{16}{11} \times \frac{\Delta P_1}{\Delta P_2}$ so that it is now possible to tabulate the ratio of the expected rates of improvement with one and two litters directly against litter size.

Litter size.

/

	Litter Size						
	4	5	6	7	8	9	10
$\frac{\Delta G_1}{\Delta G_2}$	0.73	.89	.95	1.01	1.04	1.05	1.08

These figures show that when the average litter size is seven or greater ΔG_1 exceeds ΔG_2 so that it is more efficient to raise only one litter before making a selection. The average litter size in all lines in the present experiment being greater than seven, each mating was generally only kept until it had produced one litter.

4. Comparison with Goodale's and MacArthur's Methods.

The system of breeding and selection employed in the present experiments differsconsiderably from those employed by Goodale and MacArthur. The procedure adopted by these workers will be described briefly and an attempt made to evaluate the efficiency of their methods.

In Goodale's experiment, the criterion of size used throughout was 60-day weight although there was some selection of litters when weaned at the age of one month. The procedure (Goodale, 1938) was as follows :- once a week three males, not previously mated were each put with several females (usually five) and when the females had become pregnant the males were stored until their mates had weaned their litters. If any litters were of outstanding size at weaning the mating was reconstituted at once; if not, the male was stored until his litters had been weighed at 60 days and then remated or discarded according to the performance of his progeny. Males used for progeny testing in this manner were usually the heaviest available at that time, but sometimes preference was given to the members of an outstanding family. The females were selected in a like manner, younger animals, other things being equal, being chosen in preference to older ones. Parents who had proved their worth remained in service until outmoded.

The method of selection MacArthur used was very similar to that used by Goodale (MacArthur, 1944). In general one male was mated with four to eight females and when they had become pregnant the male was stored until his litters had been weighed/

weighed at 60 days. An attempt was made to obtain ten young from each female, i.e. usually one litter in the Large Line. MacArthur gives in general terms his criteria for selection. They are based on the progeny testing of parents. If all or nearly all the progeny had 60-day weights above the mean of the previous generation then these progeny were selected. Some selection was made on individual performance and the selection was kept on a broad basis by choosing at least one female from families of only moderate performance. Selection was also made between the matings within a given generation. Progeny below standard on weighing at 30 or 60 days were discarded and the parents culled or remated to another partner. If the progeny were outstanding at 30 or 60 days then the parents were remated.

From the foregoing summaries of the methods used by Goodale and MacArthur it will be seen that great stress is laid upon the use of the progeny test. Despite this emphasis the term "progeny testing" is used very loosely. To avoid confusion it is best to restrict the term to the selection of parents by the performance of their progeny, this being the manner in which it is used for most farm animals. But, applied to poultry and plants it usually refers to selection between progenies and is therefore better called for the purposes of clarity, "family selection". Both Goodale and MacArthur use the words "progeny testing" to cover both processed which they use in combination. Family selection has already been discussed for families of one litter using figures for the 6-week weight of the cross-bred line.

Lush/

Lush (1947) gives the ratio of the expected progress under family selection to that under mass selection as

$$\frac{1 + (n-1)r}{\sqrt{n\left[1 + (n-1)t\right]}}$$

where

r = genotypic correlation
t = phenotypic correlation
n = family size.

Family selection is more efficient than mass selection when this ratio exceeds unity, i.e. where $t < r^2 - \frac{(1-r^2)}{n}$. Wherever t is as large as r^2, therefore, family selection will be less efficient than mass selection. With one family of full sibs $r = \frac{1}{2}, r^2 = \frac{1}{4}$ and since with one litter per family t is likely to be greater than $\frac{1}{4}$, family selection will be less effective than mass selection.

The 60-day weights of an unselected sample of Goodale's males raised in the present experiment were analysed and showed that the intra-litter correlation was as high as 0.6. Thus, with a litter size averaging ten, family selection on the basis of one litter would only be 69% as efficient as mass selection. With more than one litter in each family t will be smaller owing to the elimination of environmental correlations peculiar to individual litters. In the absence of data on this point, it is only possible to set a lower limit of $\frac{1}{2}h^2$ to t, which shows that when heritability is less than 50% family selection could be more efficient than mass selection. While no general conclusion on the value of family selection as used by Goodale and MacArthur can be reached it is clear that

selection/

selection between single litters was practised and this procedure was disadvantageous. "Progeny testing" in the strict sense must now be examined.

The advantage, if any, of a progeny test depends on the average performance of several progeny providing a more accurate measure of the individual's genotype than its own phenotype. Rendel and Robertson (1950) have shown that the regression of the average performance of future daughters of a bull on the average of n test daughters is $\frac{\frac{1}{4} n h^2}{1 + (n-1)\frac{1}{4}h^2}$ By applying their argument to the present problem it can be shown that the regression of future offspring on the average of a litter of size n is $\frac{\frac{1}{4} n h^2}{1 + (n-1)t}$. The regression of future offspring on an individual's own phenotype is $\frac{1}{2}h^2$ so that the relative accuracy for the prediction of breeding value of the progeny test, compared to the individual's own phenotype, is $\frac{\frac{1}{2}n}{1 + (n-1)\ t}$. With one litter t = 0.6 and supposing as before that n (the average litter size) = 10 this ratio becomes 0.78, i.e. the progeny test of an individual is only 78% as good a guide to its breeding value as its own phenotype. Little is therefore gained in judging a parent on one litter. Lack of data again prevent the assessment of the value of two or more litters by the same mate. It is, however, possible to calculate the relative accuracy of the progeny test of a male mated to m females each of which produces one litter. The relati e accuracy of this test is $\frac{\frac{1}{2} mn}{1 + (n-1)t + n(m-1)\frac{1}{4}h^2}$

and/

and using by way of illustration the figures t = 0.6, n = 10, m = 5, and h^2 = 0.2, this equals 2.98. Varying m and h^2 have relatively little effect on the magnitude of this ratio so that there is a definite advantage in progeny testing males by this system. The mean generation length with progeny testing will be approximately twice that without, but even so the advantage still rests with a system of progeny testing provided that the population is sufficiently large to avoid inbreeding difficulties.

The rate of inbreeding during Goodale's and MacArthur's experiments may be roughly estimated, MacArthur using the formula (Wright, 1931) :

$$\Delta F = \frac{1}{8M} + \frac{1}{8F}$$

where ΔF = increase in the inbreeding coefficient per generation.

M = number of male parents.

F = number of female "

states (MacArthur, 1944) that the rate of inbreeding in his experiment was approximately 1-3% per generation. However, he does not appear to have taken into account that the above formula is for random mating only and that M and F should be the effective numbers of male and female parents when there is any deviation from random mating or any differential reproduction (Wright, 1940). Owing to the use of progeny testing the number of effective parents will be far less than the actual number so it is probably best to use MacArthur's upper limit of 3% for inbreeding per generation in his experiment. When obtained in this/

this laboratory these mice had had 26 generations of selection. The remaining heterozygosity can therefore be estimated as $(0.97)^{26}$ = 37% or the coefficient of inbreeding 63%.

Goodale gives no figures for the rate of inbreeding in his population and it is only possible to make a very rough estimate of what it was. Goodale (1941) gives the total number of parent mice in approximately 28 generations as 1,200 males and 6,000 females so the average number per generation is 43 males and 214 females. Of these males each of which has on the average five females, Goodale says "Few return to the breeding pens, the remainder being discarded." It is these few males which thus make the largest contribution to the next generation and will largely determine the rate of inbreeding. A reasonable figure for their number seems to be ten - more would hardly be called "few" and less could hardly serve a sufficient number of females. The rate of inbreeding is on this figure roughly $\frac{1}{8M} = \frac{1}{80} =$ 1.25% (the fraction $\frac{1}{8F}$ being negligible.) When obtained in this laboratory Goodale's mice had been subject to approximately 38 generations of selection and would thus have left $(0.9875)^{38}$ = 62% of their original heterozygosity, i.e. they would be 38% inbred.

In summary, a general assessment of the methods employed by Goodale and MacArthur may be attempted. It seems that these workers erred in giving too much weight to the family in making their selections. This is especially so when the family consists of one litter only, in which case the

phenotypic/

phenotypic correlation is likely to be of the same size as the genotypic correlation ($\frac{1}{2}$) and mass selection by giving equal weight to individual and family performance is optimal (Lush, 1947). On the other hand the progeny testing of males mated to several females would seem to be an advantageous procedure even when the increase in generation length is allowed for. The rate of inbreeding, taking into account the progeny testing of males, is estimated at 3% per generation in MacArthur's experiment and 1.25% per generation in Goodale's.

V. RESULTS

In this section the bare results of the whole experiment will be given, their genetic interpretation being deferred till the next section.

1. Selection in parent stocks.

a) Goodale's Large Line (designated GL).

Mice from Goodale's Large Line were obtained in the spring of 1948, and mated together at random. The progeny from these mice form the initial generation which was designated GL_1. Eight generations of selection for weight at six weeks were made by the methods already described. The mean weights of all males and females are given in Table V together with the numbers on which these means are based. In generation 7 there was an outbreak of heptatitis which reduced the numbers drastically. Quite apart from this incident, however, later generations are represented by fewer mice. This decrease in numbers is not due to a reduction in fertility or litter size at birth (see Fig. 7) but to the poor mothering abilities of females. In later generations this line became remarkable for the number of females which did not lactate following parturition. The mammary glands of these females were highly developed and it would seem that some factor responsible for the initiation of lactation was missing.

Since the numbers of each sex in a generation are so often small and unequal it is desirable to calculate a weighted/

weighted mean for the two sexes in order to obtain a better estimate of the generation mean. The method of weighting used was to find the ratio of the mean weight of all males to that of all females and to use this ratio to "neutralise" the sex difference. It was found empirically by comparing the GL line and the smaller GLS line that the ratio of the sex means remained approximately constant at 1.2 and therefore a geometric mean seemed preferable to an arithmetic mean. Consequently, the weights of females were multiplied by $\frac{1}{2}(1 + 1.2)$ and males by $\frac{1}{2}(1 + 1/1.2)$ to obtain the general mean. Any errors in this method of weighting are negligible in comparison with other irreducable errors. The means calculated by this method are plotted in Fig. 1(a).

Fig. 1(c) shows the selection differentials applied in the production of each generation. These selection differentials were calculated for each mating as the average deviations of each parent from its litter mean, this being the criterion for selection. Combination of the selection differentials for individual matings was carried out by weighting according to the number of progeny measured in the next generation.

In order to show sampling errors of each mean short vertical lines have been drawn through each point extending to one standard error above and below the mean. The value of the standard error has been calculated from the equation :

$$\sigma^2_N = \tfrac{1}{4}(\sigma^2_M + \sigma^2_F + 2\sigma_M\sigma_F r_{MF})$$

where σ^2_N = variance of sex mean
σ^2_M = " " weights of males
σ^2_F = " " " " females
r_{MF} = correlation between weights of males and females

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The value r_{MF} used was obtained by correlating the mean weight of females with the mean weight of males of the same litter. This gave $r_{MF} = 0.59$.

Fig. 1(a) shows that in the GL line 8 generations of selection for large size has been ineffective and though generation means are very erratic it is clear that there is no upward trend with time. The amount of variation from generation to generation is so large that it calls for special explanation.

Seasonal effects are the first consideration, but in this laboratory there appears to be no regular cyclical effect of season upon weight. The 6-week weights of several inbred lines were examined but any seasonal variation was swamped by differences in litter size. The fluctuations in the other lines in this experiment were, within a short period of time, so frequently in opposite directions that it seemed impossible to make any correction for season.

Variations in litter size and the lactation of mothers are important factors. Owing to the small number of matings in each generation variations in the pre- and post-natal nourishment which individual mothers give their young become important. Maternal effects - using this phrase to cover post- as well as pre-natal effects - largely determine the mean 12-day weight of the litter (Falconer, 1947). It is, therefore, possible to make a correction for variation in maternal/

maternal effects by calculating the regression of 6-week weight on 12-day weight. Means corrected in this manner are plotted in Fig. 1(b). Although variations from generation to generation are reduced in amplitude they are still large differences to have to ascribe to intangible environmental factors.

Correction for maternal effects does not change the conclusion drawn above that eight generations of selection for large size have been ineffective. A selection for small size was made in the fourth generation initiating a line (designated GLS) selected for small size. This line suffered from all the defects of the GL line and after four generations it was dropped so that more space could be made available for the maintenance of the GL line. The progress in this line is also shown in Fig. 1(a) together with the selection differentials realised on each generation. The standard errors of each mean are large but it is safe to conclude that selection produced a large divergence between the lines in the first generation followed by little further change. Correction for 12-day weight as shown in Fig. 1(b) does not alter this conclusion.

b) MacArthur's Large Line (designated ML).

Mice from MacArthur's Large Line were also obtained in the spring of 1948, and after an initial generation bred at random from these mice selection for large size at six weeks was carried out for seven generations by the methods already outlined. The mean weights of all animals are given in Table VI together/

together with the numbers on which these means are based. With the passage of time there is a fairly regular decline of the numbers in each generation due to an increase in sterility and a decrease in litter size (see Fig. 7). Mice in this line tend to become excessively fat, a characteristic often associated with sterility in domestic animals.

General means neutralising the sex difference, selection differentials, and standard errors, were calculated as before and are plotted in Figs. 2(a) and 2(c). Means corrected for 12-day weight are shown in Fig. 2(b). These two graphs both show an upward trend of the means with successive generations. The measurement of the improvement produced by selection presents difficulties. Obviously little reliance can be placed upon the mean of individual generations and the measurement of improvement by the difference in mean level of the first and last generation would be liable to sampling errors and errors due to intangible environmental factors. The calculation of a regression line minimises these sources of error but is not ideal because of variations in the size of the selection differential. Thus, if errors due to sampling and environmental factors could be abolished the response curve would still not be a straight line, but, nevertheless, a regression line would,even in this case, provide a good estimate of the average response per generation. In the present experiment in which there are large errors due both to sampling and/

and to environmental factors the fitting of regression lines can be carried out without further hesitation.

The regression line fitted to the uncorrected means has a slope of 0.26 grams per generation.* Correction of the means for 12-day weight reduces the slopeof the regression line to 0.23 grams per generation. This reduction can be attributed to the progressive increase in mean 12-day weight resulting from the decrease in mean litter size, there being an inverse relationship between these two variables. Since Falconer (1947) has shown that 12-day weight is mostly determined by the mother and not the litter the value 0.23 grams/generation obtained after correction for 12-day weight will be used to give an estimate of the rate of genetic improvement.

In order to estimate heritability it is now necessary to know the selection differential. The error involved in measuring the selection differential in any one generation is due to the sampling error of the mean if it can be assumed that the individual is measured without error. Over a number of generations these sampling errors should balance out if they are of equal magnitude and it should suffice to take a simple average of the selection differentials applied to all generations. This has been done in the present case with rather uncertain justification owing to the variable size of the selection differential and gives 1.39 grams per generation. The estimate of heritability is therefore $\frac{0.23}{1.39}$ or 16.5%.

From/

* For tests of significance see Table II.

From the fourth generation a line (designated MLS) was started in which selection was made for small size. This line was only carried for four generations at the end of which period it was crossed to the ML line to prevent the total extinction of the stock. Fig. 2(a) shows the progress of this line. The standard errors of generation means are large but it would appear that there was a steadily increasing divergence between lines and no sudden separation with the first generation of selection as found with Goodale's Large Line. The average increase in divergence calculated from a regression line is 0.61 grams per generation. The average of the selection differentials producing this divergence is 2.49 grams so that the estimate of heritability given by these figures is $\frac{0.61}{2.49}$ = 24.5%. Considering the small number of generations on which this estimate is based it must be judged to be in good agreement with the previous estimate of 16.5%.

2. Results of Cross-Breeding Experiment.

A cross between Goodale's and MacArthur's Large Lines was made in the Autumn of 1948.

Animals for the cross were chosen from generations GL_2 and ML_2. Mice could not be taken at random since the best were required for the next generation of their own lines. Males could possibly have been used for their own lines and for crossing, but for the sake of uniformity it was decided to use two unselected parents in making the cross. Those animals nearest in weight to the mean for their litter were chosen and mated together at random, each male having two females.

Reciprocal crosses were made in approximately equal numbers. It was not the intention to alter litter size, but litters of more than twelve were reduced in number either to twelve or ten depending on the vigour of the newly born young when first recorded. Otherwise the methods used were identical with those in the other large lines.

a) Results of the cross.

Since the two parent lines were partially inbred it was not surprising to find that the cross between them showed heterosis. The measurement of the amount of heterosis observed depends upon the values which are taken for the parent lines. If the values indicated by fitted regression lines were used, heterosis would be indicated, though not if the values for the actual parent generations were used. The best comparison is probably with the contemporary generations of the parent lines, ignoring/

ignoring the changes in these due to selection, such changes being small and only likely to lead to a decrease in the estimate of heterosis. The F_1 generation (which was called GM_o) had a mean 6-week weight 2% above the better parent line and 6% above the mid-parent point.

The reciprocal crosses differed in mean weight at six weeks. Mice from Goodale's line mothers were significantly larger than those from MacArthur's line mothers. Most of this difference can be ascribed to maternal effects, and, in particular, to differences in lactation. The mean 12-day weight of the mice with Goodale's line mothers was 6.27 gm. compared with 5.43 gm. for the reciprocal cross. Since there was not a great variation in litter size this mean weight of individuals is a fairly accurate measure of lactation. This conclusion was confirmed by analysis of variance of each reciprocal cross. These were carried out separately for males and females and the results are presented in Table VII There are highly significant differences between litters with the same sire. If the components of variance are obtained the variance due to sires comes out negative in three out of the four cases and so its true value is probably zero. The intra-litter correlation is high (circa 0.6) indicating strong maternal effects.

Though the increase in weight due to heterosis was marked, it was small compared to the increase in other characters which may collectively be called vigour. Thus, disease was almost non-existent and there were no losses after weaning/

weaning and the F_1 females were outstanding in fertility and lactation (see Section VII and Fig. 7)

The initial results of the cross were most promising and provided material on which a large amount of selection could be practised.

b) Results of selection of cross-breds.

Two lines, GML selected for large size and GMS selected for small size, were started by selection of the largest and smallest mice from the cross-bred generation GM_0. These lines were kept quite distinct with no interchange of mice. The results of seven generations of selection are shown in Table VIII and Fig.3. Means, selection differentials, and standard errors were calculated as before. Corrections have not been made for 12-day weight since in these lines there are more productive matings and litter-size which affects 12-day weight is more nearly constant.

Individually both lines appear to have made progress under selection. The first generation of selection on GM_0 mice produced no divergence between the lines. In considering progress in each line it is probably safer to exclude the heterotic GM_0 generation and to start from GML_1 and GMS_1. From GML_1 to GML_7 the large line has improved fairly regularly and a fitted regression line has a slope of 0.46 grams per generation. The small line, on the other hand, shows no smooth trend from generation to generation, almost all of the response having occurred in one generation from GMS_2 to GMS_3 when the mean weight fell by about 3 grams.

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The two lines originating from a common stock were intended to prpvide adequate mutual controls and the regularity of the GML line and the irregularity of the GMS line might both be exaggerated. If the divergence is plotted, however, (Fig.4) no irregularities are removed. The first two generations of selection produced little divergence; in the next generation the divergence increased to 4 grams and subsequently has increased more slowly.

Estimates of heritability can be calculated as before, by fitting regression lines to the means of subsequent generations and averaging the selection differentials applied to each generation. For the GML line this method gives h^2 = 24.0% and for the GMS line h^2 = 40.5%. Using the divergence between the lines as a measure of progress and pooling the selection differentials gives h^2 = 31.1%.

Further estimates of heritability can be obtained from the resemblance between parent and offspring. Selection of parents biasses the correlation between parent and offspring but does not affect the regression of offspring on parents, except to increase its fiducial limits. The regression of 6-week weight of offspring on that of each of its parents and on the parental mean have been calculated and are presented in Table IX The regression on each parent is a measure of $\frac{h^2}{2}(1 + r_{pp})$ where r_{pp} is the correlation between parents, and that on the parental mean is a measure of h^2 (Wright, 1921).

Each/

Each regression can therefore be expressed as an estimate of h^2 to allow direct comparison with each other. This has been done in Table X and at the same time the estimates from the offspring of both sexes have been averaged. These combined estimates still have large standard errors owing to the small variance of parents but they are of use because, coming from regressions calculated within generations, they are independent of estimates from the results of selection which rely on differences between generations. The comparison of estimates from these two sources will be made in the next section, when possible causes of difference have been discussed.

VI. ANALYSIS OF GENETIC SITUATION.

An attempt will be made by using the results of selection to analyse the genetic situation in each line. If, at times, the interpretation given pays little attention to the fiducial limits of various estimates it is not done from disregard for the element of chance but merely to lay more stress on the mode of interpretation, the validity of which is a limiting factor even when all statistical requirements have been met.

1) Goodale's Large Line.

The results show that no response to selection for large size has been obtained. There is a possibility that selection was in fact effective but that the improvement due to selection was counterbalanced by the inbreeding degeneration produced by the 2-3% of inbreeding per generation. The variation in inbreeding among the matings of a single generation is too small to give any useful estimate of the rate of decline of size produced by inbreeding. In the absence of such an estimate the possibility of this balance between selection and inbreeding should be revealed by selection for small size when these two factors would be expected to reinforce each other. Four generations of selection are hardly adequate to provide an answer to this problem but the fact that the largest divergence between large and small lines was obtained in the first generation makes it appear probable that if selection and inbreeding degeneration are acting in unison they cannot be important factors.

If, then, selection produces no response is there a lack of genetic variance? The separation of the large and small lines at once shows that this is not so. The nature of the response to selection, accepting it at its face value, reveals the nature of this genetic variance. The sudden divergence of the lines in the first generation shows non-additive action of genes. A sudden decrease in size would be produced by the breaking up of combinations of genes which together produce large size but separately have very small effects. Such non-additive gene action is accommodated in a formal analysis under the heading of "Variance due to epistatic deviations." If all the genetic variance is due to this form of epistasis it would at once account for the lack of continued progress in either direction.

The conclusion to be drawn is that Goodale's Large Line still possess genetic variance but this is apparently mainly due to epistasis. This finding is not unexpected in a long-selected line since in time additive genetic variance will be fixed by selection alone and will leave only the non-additive variance. Sprague and Tatum (1942) working with lines of maize found this expectation was realised, new lines possessing additive genetic variance, old lines less additive and more non-additive genetic variance. Further selection in Goodale's Large Line cannot, therefore, be expected to produce any further progress, but must be carried on to maintain the favourable gene combinations and prevent a decrease in size.

2) <u>MacArthur's Large Line</u>.

MacArthur's Large Line responded to selection for

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large size. Heritability was estimated by the ratio of the progress made to the selection applied and gave a value of 16%. This estimate agrees well with the 24% that was obtained by four generations of two-way selection. Unfortunately, there is at present no method of calculating the standard errors of these estimates but since the first estimate is based on more generations it is probably more accurate. The important point is that in MacArthur's Large Line about 16% of the total variance used for selection is estimated to be additive genetic variance. However, the method of selection employed in the present experiments is selection within litters which utilises only half the genetic variance in the whole population so that it is desirable to convert the foregoing estimate to the heritability for mass selection. This can be done by using the formula given in Section IV.

$$h^2_w = \frac{1 - r}{1 - t} h^2$$

where h^2_w = heritability for selection within litters

h^2 = heritability for mass selection

r = genetic correlation between litter mates

t = phenotypic correlation between litter mates.

The average value of t observed for males and females is 0.68 and r is 0.50 so that

$$h^2_w = \frac{16}{100} = \frac{1 - 0.50}{1 - 0.68} h^2$$

$$h^2 = 10\%$$

It was shown in Section IV that MacArthur's mice were probably about 63% inbred when obtained and it is therefore desirable

to/

to make allowance for this inbreeding and to relate the abo estimate to what it would be expected to be in MacArthur's foundation generation.

When heritability is 10% the observed phenotypic variance (see Table II) may be partitioned as follows :-

Observed phenotypic variance	= 8.40 gm^2	= 13.77 gm^2
Estimated genetic variance	= 0.84 gm^2 for	= 1.38 gm^2 for
Estimated environmental variance	= 7.56 gm^2 females	= 12.39 gm^2 males

With 63% inbreeding the loss of heterozygosity is 37% so that the genetic variances above are expected to be 37% of the original, which would thus be:

$$\text{Estimated original variance} = 0.84 \times \frac{100}{37} = 2.27 \text{ gm}^2 \text{ for females}$$

$$= 1.38 \times \frac{100}{37} = 3.73 \text{ gm}^2 \text{ for males.}$$

If it is now assumed that the environmental variance does not change - an assumption which may not be completely valid - the original heritability may now be calculated :

$$h^2 = \frac{\text{Genetic variance}}{\text{Total variance}}$$

$$= \frac{2.27}{2.27 + 7.56} = 23\% \text{ using variances of females}$$

$$= \frac{3.73}{3.73 + 12.39} = 23\% \text{ using variances of males.}$$

Small changes in the environmental variance would not alter this estimate greatly so that this estimate of 23% for the heritability for mass selection of 6-week weight in the original population may be used with a fair degree of assurance for comparison with other estimates of heritability.

MacArthur (1949) gives estimates of the heritability of 60-day weight which decline from about 25% to 10% at the end.

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The figure given here for 6-week weight is very close to the first estimate. Larger differences might be expected owing to the different characters selected because, although the relative size of the genetic variance of each is likely to be comparable owing to the high genetic correlation between them, there is no assurance that the environmental variances at these ages will be comparable.

If reliance can be placed upon the results of selection for small size, the nature of the response is informative. Unlike Goodale's lines the two lines selected for large and small size in MacArthur's Large Line diverged steadily. There was no sudden response in the first generation so there is reason to suppose that epistasis is not important in this line. MacArthur's Line thus forms a complete contrast to Goodale's in showing the presence of additive genetic variance and the absence of variance due to epistasis.

3. Cross-bred Lines.

a) General Conclusion.

The most important result from the selection for large and small size made on the cross-breds is that an increasing divergence between the lines can be obtained and this response can be attributed to the creation of new genetic variation. Using the lines as mutual controls heritability within litters was estimated at 31% so that the estimated additive genetic variance within litters is 0.97 gm^2 for females and 1.62 gm^2 for males. In one parent line, heritability within litters was estimated at 16% or the additive genetic variance within litters at 0.53 gm^2. and 0.54 gm^2; in the other line there was little if any additive genetic variance. The crossing of the two lines must therefore be held responsible for the observed increase in genetic variance. This conclusion is borne out by the high heritability in the cross-bred stock; the estimate of 31% is for selection within litters and using $h^2_w = \frac{1 - r}{1 - t} h^2$ where r = 0.50 and t = 0.45 (average value for males and females) gives an expected heritability for mass selection of 6-week weight of 34% which is larger than the estimate for the heritability of 6-week weight in MacArthur's original stock arrived at in the previous section. There can, therefore, be no doubt that crossing of the two lines produced the expected increase in genetic variance.

b) Early generations.

The response of the first few generations to selection/

selection show an unexpected feature. The ineffectiveness of selection on the F_1 generation merely serves to confirm the absence of any large amount of additive genetic variance in the parent lines but the ineffectiveness of selection on the F_2 is quite unexpected. In Section II evidence was produced to show that Goodale's and MacArthur's Large Lines carried different size genes. After a cross, segregation should occur in the F_2 generation making possible a rapid response to selection through the combination of these genes. The results show, however, that this expected rapid response did not occur until a generation later. This lack of response between the F_2 and F_3 followed by a large response between the F_3 and F_4 may be merely the result of sampling errors or by unexplained fluctuations such as were found in the parent lines. Yet it is so striking that any possible genetic explanation must be examined. The first is linkage.

Mather (1943) has given an explanation which will account for an F_2 variance being equal in size to that of the F_1. The absolute linkage of + allelomorphs in repulsion would prevent recombination and so produce no increase in the size of the F_2 variance. With looser linkage the variance of the F_2 increases. The two lines of mice in this experiment, because they owed their size to different genes when crossed would give an F_1 with size genes in repulsion. But, since the chiasma frequency in the mouse is high, being on the average 2.8 per bivalent in males and 2.4 in females (Crew and Koller, 1932) linkage between plus and minus allelomorphs cannot on the

average/

average be very tight. Nevertheless such a situation could reduce the variance of the F_2 to the fiducial limits of the observed variance. The likelihood of this explanation is increased by the behaviour of the F_3 generation. The increase in variance was small (see Table VIII) but the fact that most of this was genetic was shown by the large response to selection for small size on the F_3. This large and sudden response could again be explained by linkage. Crossovers between the plus and the minus allelomorphs which would be quite frequent owing to the high chiasma frequency will change the phase of plus allelomorphs from repulsion to coupling resulting in a segment of chromosome with major effect which could more easily be isolated by selection. Some such crossovers would be selected from the F_2 generation but would have little effect on the mean of the F_3 since they would be comparatively rare. But, in the next generation a segment with major effect could be represented in two or three families owing to the breeding structure of the population. Thus with the addition of fresh crossovers in the previous generation there should be an accelerating response to selection until the selected line became homozygous for that segment. In fact, there was a sudden response for one generation only so that there can be no invocation of linkage of polygenes in explanation of the results.

The second possible explanation is that there is a major factor affecting size which by the process described for the segment of chromosome does not become widespread enough/

enough to produce a large divergence between the lines until the F_4 generation. Such a major factor would be detected in two ways, either by its linkage with a marker gene or by its effect on the frequency distribution of weights. In the two cross-bred lines there were four colour genes segregating and an analysis of weights of mice manifesting each gene was carried out. The results of this analysis are presented in Section VII and show no association of a size difference with any of the colour genes. Since there are twenty pairs of chromosomes and only a fairly tight linkage would manifest itself by this method of analysis, the chances were against finding any linkage of a marker gene and a major size factor. This method of approach is therefore unlikely to be useful.

On the other hand, detection of departures from normality of the frequency distribution of weight require independent evidence on the adequacy of the scale and even then the method is not very sensitive. As an example suppose that the difference between the large and small lines was due to a single recessive gene which came from one parent line. The distribution of recessive and dominant phenotypes can be symbolised by *

$$(pa + qA)^1 \quad \text{with variance } \sigma^2_A = pq\,(m_A - m_a)^2$$

where p = frequency of recessive phenotype, a with mean size m_a.

q = frequency of dominant phenotype, A with mean size m_A.

If the variance of all other factors affecting size is σ^2_R the moments of the observed variable can be obtained :

μ_2'

* The following argument was given by Professor Wright during lectures delivered in the University of Edinburgh 1949-50.

$$\mu_2 = pq\,(m_A - m_a)^2 + \sigma^2_R$$

$$\mu_3 = pq\,(p - q)(m_a + m_A)^3$$

Asymmetry will be measured by the value of the form index

$$\gamma_1 = \frac{\mu_3}{(\mu_2)^{3/2}}$$

In the F_2 generation $p = \frac{1}{4}$, $q = \frac{3}{4}$ and $(m_A - m_a) = 4gm.$ as indicated by the difference in levels of the two lines after selection. So we have

$$\mu_2 = \text{observed variance} = \sigma^2_T$$

$$\mu_3 = \tfrac{1}{4} \times (\tfrac{1}{4} - \tfrac{3}{4})(-4)^3$$
$$= 6$$

and $$\gamma_1 = \frac{6}{(\sigma^2_r)^{\frac{3}{2}}}$$

The standard error of γ_1 for a normal curve

$$= \sqrt{\frac{6}{n}} \quad \text{where } n = \text{number of observations.}$$

To establish asymmetry at the 5% level of probability

$$\frac{6}{(\sigma^2_r)^{\frac{3}{2}}} \not< 2x\sqrt{\frac{6}{n}}$$

$$n \not< \frac{2}{3}(\sigma^2_T)^3$$

for mice in the GML line the average values of σ^2_T were $\sigma^2_T = 6.89$ for females and $\sigma^2_T = 10.15$ for males. Therefore, to establish asymmetry, n is not less than 218 and 697 for females and males respectively. To detect kurtotic deviations from normality by use of the form index requires even more observations. With more than one major size factor segregating it would therefore be impossible with a small population to detect such

factors/

factors by deviations from normality of the frequency distribution of weights.

Neither explanation of the anomalous behaviour of the first generations has proved satisfactory and further speculation would be unwarranted without repetition of this experiment to remove the suspicion that it is not after all a chance of sampling.

c) Selection up and down.

Heritability in the small line was apparently almost twice as great as that in the large line, viz. 40.5% as against 24%. A simple explanation of this difference can be given since both lines are being slowly inbred at approximately 1.7% per generation, and any inbreeding depression should hasten selection downwards and hinder selection upwards. Assuming that this depression acts equally and additively on each line, it is possible to calculate the size of this effect which would cause the observed difference on heritability. The true heritability is estimated by the divergence between the lines and is 31.1%. If the inbreeding depression per 1% increase in inbreeding coefficient is x gms. then since :-

$$\frac{\text{Response}}{\text{Sel.diff.}} = h^2$$

for the large line

$$\frac{0.46 + x1.7}{1.92} = 0.311$$

or $$x = \frac{1.37}{1.7} = 0.081$$

and for the small line

$$\frac{0.58 - x1.7}{1.43} = 0.311$$

$$x = \frac{.135}{1.7} = 0.079$$

An inbreeding depression of about 0.08 grams per 1% increase in the inbreeding coefficient, F, would account for the observed results. Values for the regression of 6-week weight on the inbreeding coefficient for mice from the cross-bred stock are available from experiments on inbreeding without selection (Parvaneh - unpublished*). The average value of this regression is -0.073 grams per 1% increase in F for the range F = 0 to F = 54.4%. There is a suggestion of curvilinearity in this regression so that for small amounts of inbreeding this value may be a slight underestimate. The agreement with the calculated 0.08 grams per 1% increase in F is therefore excellent and it must be concluded that inbreeding depression can account for the observed differences in heritability on selecting in two directions.

d) Offspring-parent regressions.

It has already been pointed out that offspring-parent regressions calculated within generations provide an independent estimate of heritability. The estimates in Table X must be compared with the estimate of 34% for mass selection obtained from the results of selection and the agreement in some cases is poor even taking into account the large standard errors. To discuss these discrepancies it is necessary to examine the type of bias to which each of the three regressions (on sire, dam and mid-parent) is subject.

Dickerson/

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* Acknowledgements are due to Mr. P. Parvaneh for the use of these results.

Dickerson and Grimes (1947) have discussed the various sources of bias. They considered that next to a controlled selection experiment, regression on the parental mean provides the best estimate of heritability. This estimate is subject to two sources of bias. Firstly, the resemblance between offspring and parent may in part be due to epistasis. If the character is dependent upon gene combinations these may be transmitted by the immediate parents to their offspring, but no progressive change is possible, and if this were the only type of genetic variance selection would do no more than maintain the gene combinations. Thus, the offspring mid-parent regression will overestimate heritability when epistatic genetic variance is important. The second source of bias depends upon the maternal environment being of importance in the determination of the character as it is for 6-week weight. The regression will be biased if the maternal environment which a dam provides is correlated with her phenotypic size. If this correlation is positive, which seems most likely, heritability will be overestimated, if it is negative heritability will be underestimated.

When these two sources of bias are negligible the regression on dam and sire may still be biased. Doubling the regression of offspring on dam is liable to overestimate the actual heritability which would be obtained by selection if the

maternal/

maternal environment is subject to heritable differences. Doubling the regression of offspring on sire would in this case underestimate the actual heritability since it neglects the improvement which can be made in the maternal environment. Further complications arise when there is a genetic correlation between a dam's genotype and the maternal environment which she provides. If this correlation is negative, as Dickerson and Grimes (1947) found for economy of gain in the pig, a genotypically superior dam may more than offset the genetic improvement of her offspring by the poor environment which she provides. In this way doubling the regression of offspring on dam may overestimate actual heritability and vice versa for the regression on sire. The sources of bias in offspring-parent regressions can thus become complex and while there will be rarely sufficient data to show which source is important valuable indications may be obtained by comparing the regressions.

The heritability estimates for the GML line obtained from offspring-sire and offspring-dam regressions (Table X) are in reasonable agreement with each other, but in the GMS line the agreement between these estimates is not good. The standard errors of these estimates are large and the results for the offspring of each sex are not very consistent so that no great weight can be placed upon the observed differences. Nevertheless, the regression on the dam is larger than that

on/

on the sire, though not significantly so. This difference, if real, would indicate that there are heritable differences in the maternal environment. Examination of heritability estimates for 3-week weight obtained by offspring-parent regressions does not, however, confirm this suspicion. The weaning weight should provide a better estimate of the maternal environment and yet the regression on the dam is actually less than that on the sire (Table X). There is therefore no reason to suppose that heritable differences in maternal environment are important in determining 6-week weight for if they exist they are uncorrelated with the 6-week weight of the dam.

The regressions of offspring on mid-parent give a higher estimate of heritability in the GML line and a lower estimate in the GMS line than the estimate of 34% obtained by selection. Epistasis can only lead to an overestimate of heritability but a possible explanation of both differences might be found in differing correlations between the phenotype of the dam and the environment which she provides. As far as these two characters are measured by the 6-week weight of the dam and the 12-day weight of her litter there is no sign of any large difference since in the GML line the correlation (calculated within generations) is 0.36 and in the GMS line, 0.31. It seems better, therefore, to attempt no better explanation of this discrepancy than is provided by "the errors of sampling".

To sum up - the genetic situation in the cross-bred lines/

lines appears to conform to the simplest model. Additive genetic variance is estimated at 34% and other genetic sources of variance are apparently unimportant. An additive scheme involving improvement by selection offset by some decline due to inbreeding depression provides an adequate explanation of the observed results of selection. The results are on the whole consistent and the examination of several minor discrepancies have not revealed any other agency which requires to be taken into account.

VII. CORRELATED RESPONSES

Changes in characters other than the one subject to selection are important because of the hidden correlations which they may reveal. If chance changes can be ruled out, then the presence of genetic correlations between characters can be established. Such genetic correlations may be due to linkage, i.e. the spatial relationship of the genes which can alter with crossing over or to properties of individual genes and therefore unalterable within a given set of genes. The latter type is usually known as a physiological correlation and assumed to be unalterable, although there is no reason to suppose that it could not be altered to some extent by selection. Such correlations are of great importance in animal breeding. Besides providing an explanation for the lack of progress in some selection experiments, they may be of practical value. Genetic correlation between two characters may make it possible to improve one character with a low heritability by selecting on another more highly heritable character which is genetically correlated with the first.

1. Changes in gene frequency of colour genes.

a) Review of literature.

There is a large body of evidence to show that many colour genes have a pleiotropic effect on body size. The association between colour genes and body size was first reported by Green (1931) in crosses of Mus musculus and M.bactrianus./

M.bactrianus. The Mus musculus parent carried the recessive colour genes non-agouti, brown and dilute so that when F_1 mice were back-crossed to this parent segregation of the three colour genes occurred. Green observed that there was a clear association between brown colour and a larger body size, and possible associations of body size with non-agouti and with dilute. These associations could be explained either by a pleiotropic effect of the colour gene or by linkage (in coupling) between the colour gene and a size factor coming from the larger parent. Green chose the latter explanation but subsequent experiments by other workers make pleiotropy a more likely explanation. Grüneberg (1943) has pointed out that critical evidence for linkage requires matings in the repulsion phase in addition to those in coupling made by Green. Grüneberg quotes the experiments of Law (1938) as being a case in which the gene b was introduced by the smaller parent but he ignores the fact that both Green and Law used Little's dba strain and so both would have matings in coupling between b and the postulated size factor. Unless it is assumed that large and small strains have different alleles at the locus of this factor, Law's experiment would only seem to support Green.

Green's explanation cannot be ruled out in his experiments but all the evidence from other workers working with different strains is consistent in showing the association between colour genes and body size so that there is strong circumstantial evidence for pleiotropy. Feldman (1935)

showed/

showed that in two partially inbred lines and the cross between them, brown animals were on the average heavier than blacks. A paper by Castle and his co-workers in 1936 (Castle, Gates, Reed and Law, 1936) is the first of a series in which the effects of various colour genes on body weight, body length and tail length were described. It will suffice to list the effects of the colour genes (Castle, 1941).

No effect:	albino, extreme dilution and non-agouti.
Increase of size:	brown, dilute, yellow.
Decrease of size:	pink-eye, pallid, leaden.

There was a complicated interaction between these genes. Even when the action was in the same direction there were differences between genes in their relative effects on body size and linear dimensions. But even more remarkable are the changes of direction of action found in various combinations. Brown increases body size when alone or in combination with dilute, but decreases it in the presence of leaden or pallid,

A stock of mice in which colour genes are segregating is of interest from the point of view of size inheritance. Those colour genes with any physiclogical action on size provide identifiable examples of the numerous and in general unidentifiable size factors assumed to be segregating. MacArthur who introduced several colour genes into his foundation stock was able to follow the changes in frequency of these genes in both his selected lines. Genes with no action on size alter in frequency by the chances of sampling. This "drift" in a small population was held responsible for the disappearance/

disappearance of albino, c, from the small line and recessive spotting, s, from the large line. Brown and dilute were found in Castle's experiments to increase body size both alone and in combination and MacArthur reported, in agreement with expectation, that these two genes had disappeared from the small line and had become nearly homozygous in the large line. One inbred line used to start the lines was homozygous for short-ear, se, and dilute, d, which are very closely linked (Snell, 1931). Short-ear decreases body size and therefore MacArthur states that crossovers would be favoured by selection. This undoubtedly is true but it is difficult to see why such an explanation, depending on a low frequency of crossing over, is invoked when another of the foundation inbred lines carried dilute without short-ear. If crossovers were sufficiently frequent to be of any importance it is relevant to ask why his small line did not become homozygous for short-ear. It is, in fact, reasonable to assume that this did not occur owing to the rarity of crossovers. Homozygous se d/se d animals are reported to be only slightly smaller than normals so perhaps the fact that the small line did not acquire this genotype may be ascribed to chance.

A mating made for another purpose involving one of MacArthur's mice resulted in an unexpected discovery which affects the interpretation of MacArthur's results. This mating showed that the diluting factor present was not closely linked to short-ear, and further tests showed that the diluting factor was/

was not dilute (d) but leaden (ln), which is phenotypically indistinguishable from dilute. Since the mice obtained from MacArthur were nearly all homozygous for a diluting factor there is no possibility of any great change in gene frequency having taken place and it must be concluded that the phenotype which MacArthur describes as "dilute" was leaden. This discovery at once reveals a serious discrepancy between expectation and results. Castle's results showed that brown and leaden in combination decreased body weight at maturity by about 5.5% so that the small and not the large line would have been expected to become homozygous for these genes and not vice versa as MacArthur found. Since no attention was paid to the phenotype for colour in selecting the large and small lines the results are completely contrary to expectation. Drift seems a hardly adequate explanation in view of the relatively large effect of brown and dilute in combination reported by Castle and another explanation must be considered.

It is necessary to establish that a particular gene does have its alleged effect in the particular stock with which one is working. Reliance on the published results of other workers might lead to error through two causes. Firstly, the colour gene may be linked to a size factor in one particular stock. Secondly, the presence of a specific modifier in the stock might give a "neutral" gene some effect, or reverse the direction of action of a gene not assumed to be neutral. For instance, pallid might be considered to be a specific modifier

of brown causing this latter gene to decrease body size instead of increasing it. Therefore, if changes in the frequency of colour genes resulting from selection for size are to be related to the effects of those colour genes on size, these effects must be determined in the stock which is subject to selection and not in another.

b) Results.

Goodale's large line was homozygous albino, and so there were no colour genes which could be followed. The cross to MacArthur's large line showed that the line was apparently homozygous for albino and non-agouti but lacked any other mutant colour genes.

MacArthur's large line was homozygous brown but segregated for agouti, black-and-tan and leaden. Albino was also present at a low frequency but did not segregate and was only revealed by the cross to Goodale's large line. Since the number of matings in some generations was so small no analysis of gene frequency has been attempted in this line since any directed changes which occurred could not be distinguished from drift.

The two cross-bred lines (GML and GMS) showed segregation of colour genes at four loci. MacArthur's large line was responsible for the introduction of A, a^t, b, ln and some c genes, and Goodale's for a and the remaining c genes. Estimates of the frequency of recessive genes are given in Table XI and were obtained (generation GM_o excepted) from the formula: /

the formula :

$f = \sqrt{r/n}$ where f = frequency of the recessive gene.
r = observed no. of recessive phenotypes.
n = size of population.

This formula will only hold if there was random mating with respect to that gene and if it has any effect on size this assumption will not be valid. But since the standard error of these estimates must be very large owing to the small number of parents of each generation, no more accurate mode of estimation was attempted.

Table XI shows that after 7 generations of selection none of the four genes is approaching fixation or extinction in either line. All the observed changes could almost certainly be accounted for by drift but two trends in the GMS line may be noted. Firstly, non-agouti shows an abrupt fall in frequency in the third generation when there was a sudden response to selection for small size. Such an association is perhaps suggestive of linkage of a with a major size factor, but the subsequent rise in frequency would not be expected and it may be a mere accident of sampling. Secondly, leaden may be increasing in frequency in the GMS line as the average of the last 5 generations is high. These results, therefore, appear to be conforming with the change in frequency expected from the alleged pleiotropic effect of the leaden gene on size but not with the change expected for brown.

The desired measurement of the effect of colour

genes/

genes on size was not possible in MacArthur's large line owing to a lack of numbers but was attempted by pooling the two cross-bred lines. A comparison was made within litters of the weights of albino and non-albino mice. Similar comparisons were made for non-agouti, brown and leaden mice among the non-albinos. Weighted mean differences were calculated (Snedecor, 1946, p.289) and are given in Table XII. Standard errors were calculated from the intra-litter variance calculated for all cross-bred mice.

It will be seen that none of the differences are significant and yet the standard errors are quite small. The standard errors expressed as a percentage of the mean weight of all cross breds are also given in Table XII and it is clear that differences of the order of 2% should be detectable. There is therefore little evidence for any of these mutants having any effect on size of the magnitude found by Castle and his collaborators.* It is true that Castle was dealing only with adult weight and not weight at 6 weeks but the generality of the growth process makes it unlikely that these mutants affect weight only after 6 weeks, and it is more probable that a difference due to the genetic milieu exists. The causes ascribed to such a difference can only be speculative and more work is needed on the action of colour genes on size. In particular/

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* The comparison here is of mutant homozygotes with heterozygotes and normal homozygotes together, whereas in Castle's experiments usually only the heterozygotes and mutant homozygotes are compared. Therefore, if dominance is absent, the differences calculated should be greater than Castle's.

particular mutations in inbred lines could be collected and utilised to establish beyond doubt whether or not that mutant has a physiological action on size in a stock which has a definite genetic entity. The importance of linkage with size factors and of the genetic milieu could thus be evaluated.

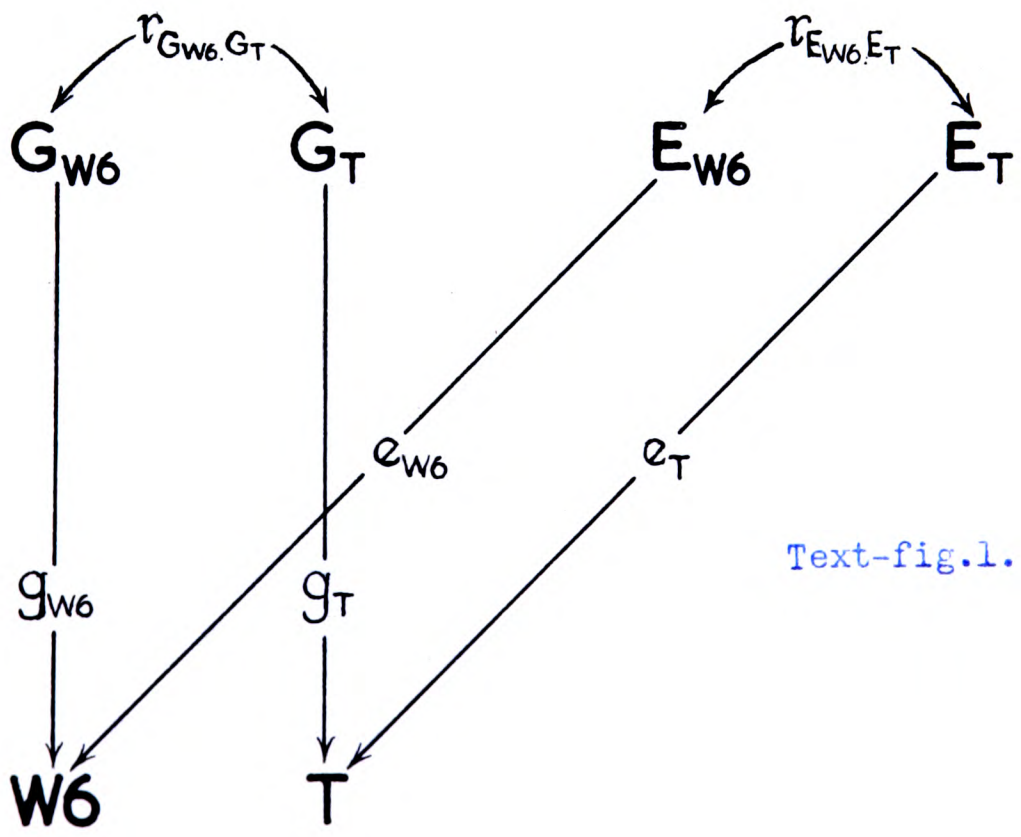
The discovery that colour genes have very small if any, association with size in the crossbred lines, resolves many difficulties. All changes in gene frequency in the cross-bred lines can be safely ascribed to drift and the same explanation is available for MacArthur's results. To conclude this section it can be stated that, contrary to expectation, the changes in gene frequency have turned out to be uncorrelated responses.

2. Other measurements of size.

a) Tail length at 6 weeks.

Tail length provides a measurement of a part of the skeleton and is expected to be correlated with general body size and body weight, and, in fact, the observed correlation between tail length and body weight is 0.48 in the cross-bred stock. This observed phenotypic correlation is due to two causes. Individuals may be alike with respect to tail length and weight either because they have genes which affected both, or, because they have been subjected to an environment which affected both. The phenotypic correlation may therefore be broken down into genetic and environmental components.

This relationship is shown as a scheme of cause and effect in Text-fig. 1 :



Text-fig.1.

The system of representation adopted is that of Wright (1923). The phenotype for 6-week weight, W6, is represented as being completely determined by the additive genotype for 6-week weight, GW6, and the environmental component* of 6-week weight, EW6, which make respective contributions measured by the path coefficients g_{W6} and e_{W6}. Tail length, T, is similarly determined by G_T and E_T. The genetic correlation is the correlation, $r_{G_{W6}\ G_T}$, between the additive genotypes and the environmental correlation is similarly $r_{E_{W6}\ E_T}$.

The observed phenotypic correlation $r_{W6.T}$ can be calculated from Text-fig.1 above. Summation of all paths connecting W6 and T gives the equation :

$$r_{W6.T} = g_{W6}.g_T\ r_{GW6.G_T} + e_{W6}\ e_T\ r_{E_{W6}.E_T} \qquad \text{(i)}$$

Also by the principle of complete determination,

$$g^2_{W6} + e^2_{W6} = 1$$

$$g^2_T + e^2_T = 1 \qquad \text{(ii)}$$

The path coefficients, g_{W6}, g_T, are related to the heritability of each character. Heritability can be defined as the regression of genotype on phenotype. Thus :

$$h^2_{W6} = \frac{\text{cov}\ (W6.G_{W6})}{\text{var}\ (W6)}$$

$$= \frac{\text{cov}\ (W6.G_{W6})}{\sqrt{\text{var}\ (W6) \times \text{var}\ (G_{W6})}} \times \sqrt{\frac{\text{var}\ (G_{W6})}{\text{var}\ (W6)}}$$

$$= r_{W6.G_{W6}} \times h_{W6} \quad \text{since} \quad h^2_{W6} = \frac{\text{var}\ (GW6)}{\text{var}\ (W6)}$$

Therefore, $h_{W6} = r_{W6.G_{W6}}$.

But/

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* Including non-additive gene effects.

But, in Text-fig. 1 there is only one path connecting G_{W6} and W6 so that the correlation between these two variables becomes equal to the path coefficient

$$r_{W6.G_{W6}} = g_{W6}$$

The path coefficient g_{W6} is thus the square root of the heritability of 6-week weight and g_T similarly the square root of the heritability of tail length. Providing estimates of these parameters are known e_{W6} and e_T can be obtained from equation (ii). Substituting in equation (i) the only remaining unknowns are the genetic and environmental correlations so that if either of these correlations can be measured then the other may be quite simply calculated.

The genetic correlation can be measured by relating one character in one animal to the other in a relative. Hazel (1943) has given a formula using this method for the calculation of genetic correlations but Dickerson and Grimes (1947) found that this formula gave unreasonable correlations greater than one when maternal effects were important. The offspring-parent regressions for tail length given in Table IX show a higher regression on the dam than on the sire in the GML line. This is interpreted as an indication of a heritable maternal effect correlated with the dam's genotype. Hazel's method would therefore be unsuitable and an alternative method is desirable. This is provided by the results of two-way selection for weight since tail length is affected by the divergence in weight to the extent to which it is determined by the same genes

The/

The divergence in tail length produced by the selection for weight can be used to find the genetic correlation between these two characters. If the divergence in tail length is regressed on the divergence in weight this will give the genetic regression of tail length on weight. The genetic correlation is obtained straightforwardly from the regression coefficient by multiplying and dividing by the appropriate genetic standard deviations.

This method can be applied to the results of selection in the cross-breds. The mean tail length of successive generations of the GML and GMS lines are shown in Fig. 6(b). Following a large divergence in the first generation the divergence has slowly increased in subsequent generations with some irregularities. The regression of divergence in mean tail length on the divergence in mean 6-week weight was 0.349 mm/gm.

The genetic correlation

$$r_{G_{W6}.G_T} = b_{G_T.G_{W6}} \times \frac{\sigma_{G_{W6}}}{\sigma_{G_T}}$$

$$= b_{G_T.G_{W6}} \times \frac{h_{W6}\,\sigma_{W6}}{h_T\,\sigma_T}$$

where $\sigma_{G_{W6}}$ = genetic standard deviation of 6-week weight.

σ_{G_T} = genetic standard deviation of tail length.

h^2_{W6} = heritability of 6-week weight etc.

Now $b_{G_T.G_{W6}} = 0.349$ mm/gm.

and averaging values for the GML and GMS lines,

$h^2_{W6} = 0.342$

$h^2_T = 0.439$

σ_{W6} = 2.511 gm) for females and = 3.093 gm) for

σ_T = 3.643 mm) = 3.940 mm) males.

hence/

hence

$$r_{G_{W6}.G_T} = 0.349 \times \sqrt{\frac{0.342}{0.439}} \times \frac{2.511}{3.643} = 0.212 \text{ for females}$$

and 0.242 for males.

The genetic correlation between tail length and 6-week weight averages 0.227.

The environmental correlation can now be calculated.

$$g_{W6} = \sqrt{h^2_{W6}} = 0.585$$

$$g_T = \sqrt{h^2_T} = 0.663$$

and from equations (ii)

$$e_{W6} = \sqrt{1 - g^2_{W6}} = 0.811$$

$$e_T = \sqrt{1 - g^2_T} = 0.749$$

substituting in equation (i)

$$r_{W6.T} = g_{W6}.g_T.r_{G_{W6}.G_T} + e_{W6}\, e_T\, r_{E_{W6}\, E_T}$$

$$0.476 = 0.585 \times 0.663 \times 0.259 + 0.811 \times 0.749\, r_{E_{W6}\, E_T}$$

$$r_{E_{W6}\, E_T} = 0.626$$

The analysis is now complete, the observed phenotypic correlation of 0.476 between tail length and 6-week weight having been broken down into a genetic correlation of 0.227 and an environmental correlation of 0.626. The association between these two characters is therefore due more to the common action of environmental differences on both than to the common action of genes.

b) Weight at 3 weeks.

Three week weight is also correlated with 6-week weight. Fig. 6(a) shows the mean 3-week weight in successive generations. There are some irregularities but the trend is for/

for the two lines GML and GMS to diverge. This increasing divergence is proof of a genetic correlation but the evaluation of this correlation presents a new difficulty. In the GMS line the heritability of 3-week weight is estimated from offspring-parent regressions to be 45%, in the GML line as zero. Since this difference is significant it is not legitimate to pool these estimates in order to arrive at the genetic standard deviation of 3-week weight. It is, therefore, only possible to state that 6-week weight and 3-week weight are genetically correlated without setting a figure to this correlation.

3. Fertility.

Owing to the small number of matings made in each generation absolute fertility cannot be studied adequately It is only possible to follow litter size in successive generations as shown in Fig. 7. Only living young were used in the calculation of the mean litter size since stillborn young are liable to be eaten by the mother before they can be counted.

In Goodale's large line there was considerable variability from generation to generation but there was no tendency for the mean to change either with selection for large or small size.

In MacArthur's large line there was a fairly regular decline in litter size. Since this occurs also in the line selected for small size this change can hardly be correlated with the increase in body size. It is probable that this decline was caused by inbreeding which occurred at a fairly high rate owing to the small number of fertile animals.

When the two parent lines were crossed, the average litter size was approximately that of contemporaneous mice in the parent lines. The cross-bred females were very fertile and showed a large heterotic effect. The females of the next generation were only slightly less fertile and the fertility of the cross-breds has remained high. There is little indication of the GML and GMS lines diverging in litter size despite the quite large divergence in body size. If then there is a genetic correlation between body size and litter size it cannot/

cannot be very large. The association between these two characters found by MacArthur also does not appear to be very close since he obtained no great change in the divergence of litter size between his small and large lines after the fourth generation although the lines showed increasing differences in body size after this generation. Litter size does not therefore appear to be highly correlated with size although this may not apply to other measures of fertility.

VIII. THE IMPROVEMENT OF SIZE IN THE MOUSE.

In this section the observations and conclusions of previous sections will be used to deduce the best procedure for increasing 6-week weight by means of selection. This involves the choice of an optimal breeding structure and the construction of a selection index making use of concomitant measurements. It must be pointed out at once that the results of such planning are dependent on the characteristics of the particular population used, in this instance the GML line, and are not generally applicable. Nevertheless the final recommendations given for the improvement of size show the large increase in efficiency which is possible by the use of these methods.

1. Optimal breeding structure of the population.

a) With present size of population.

The method of determination of the optimal breeding structure of a population has been described by Dempster and Lerner (1947) and is basically a matter of computing expected gains per unit of time with various schemes. This method has already been employed in Section IV for a small population and little further discussion is needed. There is, however, the factor of inbreeding depression to be taken into account since although this could be ignored for two-way selection it demonstrably decreases 6-week weight.

If an estimate of the inbreeding depression is available it is possible to take this into account in predicting progress/

progress under selection (Comstock and Winters, 1944). the expected advance from selection being given by :

$$\Delta I = j \Delta F + h^2 \Delta P \qquad \text{(i)}$$

where ΔI = average superiority of offspring

h^2 = heritability

ΔF = increase in the inbreeding coefficient

j = regression of the character on the inbreeding coefficient

ΔP = selection differential.

Values of j for the 6-week weight of the cross-bred stock are available from experiments on inbreeding without selection (Parvaneh - unpublished).

j = -0.071 gm. per 1% increase in F for females

j = -0.075 gm. per 1% increase in F for males.

The linearity of this regression is open to doubt and the values quoted cover the range F = 0 to F = 59.4% so that for small amounts of inbreeding j may be underestimated. However, the order of magnitude of this regression is sufficiently well defined to warrant further discussion using an average for both sexes of j = 0.073 gm. per 1% increase in F.

It is now possible to calculate the expected advance per generation using the values of j and ΔF which were observed for the cross-bred mice. The decrease in size will be $j . \Delta F$ grams = 0.073x 1.7 = 0.124 gm. The increase in size due to selection is $h^2 \Delta P$ and the expected values of ΔP for various litter sizes are given on p. 46 in standard deviation units.

In/

In order to obtain the answer in grams it is therefore necessary to multiply by the average for the two sexes of the standard deviation of 6-week weight observed within litters. The results of the complete calculation for various litter sizes and heritabilities is given below.

Expected improvement in grams per generation.

Heritability (within litters) h^2 =	Litter size.								
	5	6	7	8	9	10	12	14	16
0.10	.027	.052	.074	.093	.108	.121	.149	.162	.179
0.20	.177	.229	.272	.311	.341	.366	.414	.448	.482
0.30	.328	.405	.470	.527*	.572	.612	.683	.734	.786
0.40	.478	.582	.668	.745	.805	.857	.952	1.020	1.089

With the above figures it is now possible to take the time factor into consideration and, as in Section IV, to decide whether one or two litters will give the greatest improvement per unit of time. The expected rates of progress with one and two litters, ΔG_1 and ΔG_2, can be calculated assuming a generation interval of eleven weeks for one litter and sixteen weeks for two, which were close to the figures observed in the GML line. To compare these expected rates the ratio of ΔG_1 to ΔG_2 is tabulated below /

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* This improvement has not been realised in the GML line chiefly because the average litter size is less than eight at 6 weeks. Such discrepancies should not alter appreciably the relative efficiencies of different breeding systems.

Heritability (within litters) h^2 =	Litter size			
	5	6	7	8
0.10	0.32	0.51	0.66	0.76
0.20	0.70	0.80	0.88	0.94
0.30	0.78	0.86	0.93	0.98
0.40	0.81	0.89	0.95	1.00

From the above figures it can be seen that it will generally be better to rear two litters than one, and that the advantage will increase as heritability decreases. With litters of eight the raising of two litters ceases to be advantageous only when the heritability exceeds 40%.

This conclusion is the opposite to that arrived at for two-way selection in which the effects of inbreeding could be neglected. There is a further difference in that the heritability is now an important variable when deciding between one and two litters. Since heritability is expected to decline during the course of selection the advantage in raising two litters will increase so that a change to this practice is very desirable for the production of larger mice.

b) <u>With increased population size</u>.

Increasing the size of the experimental population would permit the use of the more efficient method of mass selection. In small populations this would not be possible owing/

owing to the resulting high rate of inbreeding. The calculation of the probable rate of inbreeding when applying mass selection and at the same time trying to avoid inbreeding is not easily calculated. It is, however, possible to give an approximate solution which is provided by a calculation due to Professor Wright * of the rate of inbreeding in a population consisting of N pairs in which brother-sister mating is avoided. With a litter-size of 8 it should be possible to meet this proviso when there are five or more matings. As the number of matings increases this solution will progressively overestimate the minimum rate of inbreeding since matings of more distant relatives will also be avoidable, but nevertheless it will provide a convenient figure for use with small populations.

The inbreeding coefficient under this system is given by the formula :

$$F = F^{1} + \frac{1}{4N}\left(1 - 4F^{1} + 2F^{11} + F^{111}\right)$$

where F = inbreeding coefficient of the present generation.

F^{1} = " " " parental "

F^{11} = " " " grand-parental "

F^{111} = " " " great-grand-parental "

The limiting rate of decrease of heterozygosis, z is given by solving the equation

$$z^{3} + z^{2}\left(2 + \frac{1}{N}\right) + z\left(1 + \frac{3}{2N}\right) + \frac{1}{4N} = 0$$

The largest root of this equation is the rate of increase in the inbreeding coefficient, ΔF. A few values have/

* Lectures delivered in the University of Edinburgh 1949-50.

have been calculated for various values of N.

N = 5 ΔF = 0.041

N = 10 ΔF = 0.023

N = 15 ΔF = 0.016

N = 20 ΔF = 0.012

With an increase in the number of matings the inbreeding coefficient decreases rapidly at first and then more slowly until it becomes approximately $\frac{1}{4N}$. A comparison of selection within litters and mass selection can now be made.

If now the population size were doubled making 16 matings, ΔF under mass selection would be 1.5% and for selection within litters ΔF = 0.8%.

Using the indicated heritabilities of 34% for mass selection and 31% for selection within litters the expected improvements can be calculated for a litter size of eight.

The value of ΔP for a litter size of eight is given on p. 46 and is 1.01 standard deviation units. This figure is multiplied by the average for the two sexes of the standard deviation calculated overall and within litters for use in the respective instances.

For mass selection -

$\Delta I = -0.073 \times 1.5 + 0.34 \times 1.01 \times 2.802$

$= 0.853$ gm.

For selection within litters -

$\Delta I = -0.073 \times 0.8 + 0.31 \times 1.01 \times 2.151$

$= 0.615$ gm.

Mass/

Mass selection now has a 39% advantage over selection within litters and an advantage of 55% over the present system of selection within litters using eight matings.

The question of whether to raise one or two litters has to be raised again, the preceding results being for one litter. For mass selection with two litters -

$$\Delta I = -0.073 \times 1.5 + .34 \times 1.41 \times 2.802$$

$$= 1.234 \text{ grams.}$$

The ratio of expected rates of improvement with one and two litters is thus :

$$\frac{\Delta G_1}{\Delta G_2} = \frac{.853}{11} \times \frac{16}{1.234} = 1.01$$

Once again the raising of two litters is slightly better than raising one for the purpose of increasing size, and as a declining heritability will increase this advantage it is desirable to raise two litters.

Still larger increases in the population size would make it possible to progeny-test males and increase the rate of improvement still further. Such schemes are best tailored to individual requirements and since they involve no new principles they are not discussed further.

2. Construction of a selection index.

From the point of view of the animal breeder the best method of selection is that which gives the greatest rate of improvement in overall merit. The various traits which contribute to merit have to be weighted according to economic value. In selecting for a single character this step is not necessary. The problem is to make use of concomitant measurements to improve the efficiency of selection by combining all measurements into a selection index, which is intended to maximise the genetic differences between individuals. Smith (1937) was the first to develop a selection index and used Fisher's discriminant functions to find the weights to be given to several observable variables in order to maximise the genetic differences between various lines of plants. This method has been applied to egg production by Panse (1946) but otherwise the development of selection indices in this sphere of animal breeding has followed the methods of Wright and Lush. Hazel and Lush (1942) compared three methods of selection for a character compounded of various traits with different economic values. They found that use of a total score method would be expected to give the greatest improvement, but they did not formulate any method of obtaining the best combination of traits to be used as the score. This need was supplied by Hazel (1943) who presented a genetic basis for the construction of such a score or index. Since it was a multiple correlation method intended to give the maximum correlation between genotype and

the/

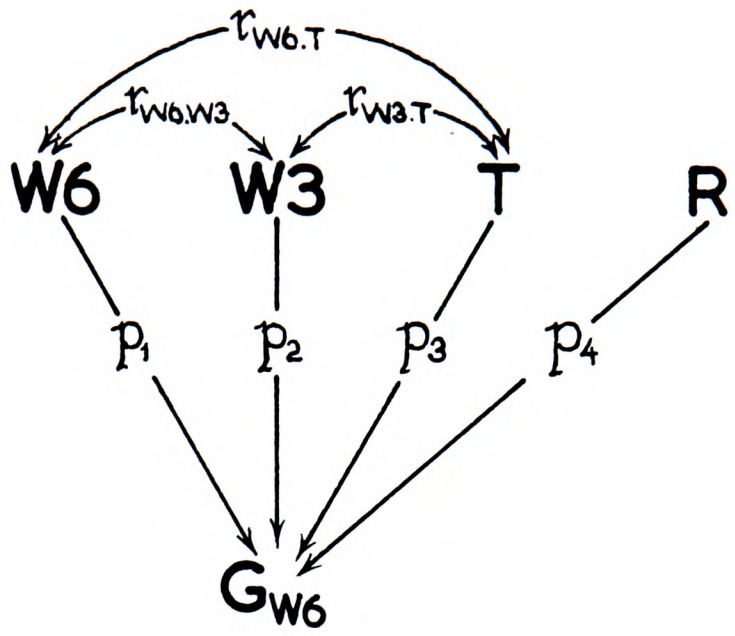
the index it would seem to be identical in principle with the method used by Smith.

The present purpose in this section is to derive a selection index combining the measurements of 6-week weight, 3-week weight and tail length which will give the maximum progress in 6-week weight. The form of the desired index is :

$$I = b_1W6 + b_2W3 + b_3T$$

where b_1, b_2 and b_3 are concrete regression coefficients giving the optimum balance between 6-week weight, (W6), 3-week weight, (W3), and tail length, T. In Hazel's method further weighting coefficients have to be introduced to allow for the different economic importance of component traits in deciding overall merit, but since for the present purpose two component traits, W3 and T, have zero "economic importance" they would vanish from the selection index. A modification of Hazel's method is therefore required, which differs chiefly in the path coefficient diagram.

The phenotypic measures, weight at 6 weeks (W6), weight at 3 weeks (W3) and tail length (T) are to be used to predict the individuals additive genotype for weight at 6 weeks (G_{W6}). The path coefficient diagram for the development of this prediction formula is shown in Text-fig.2 using the symbolism of Wright (1923). A residual factor, R, is inserted to complete the determination of G_{W6}.



Text-fig.2.

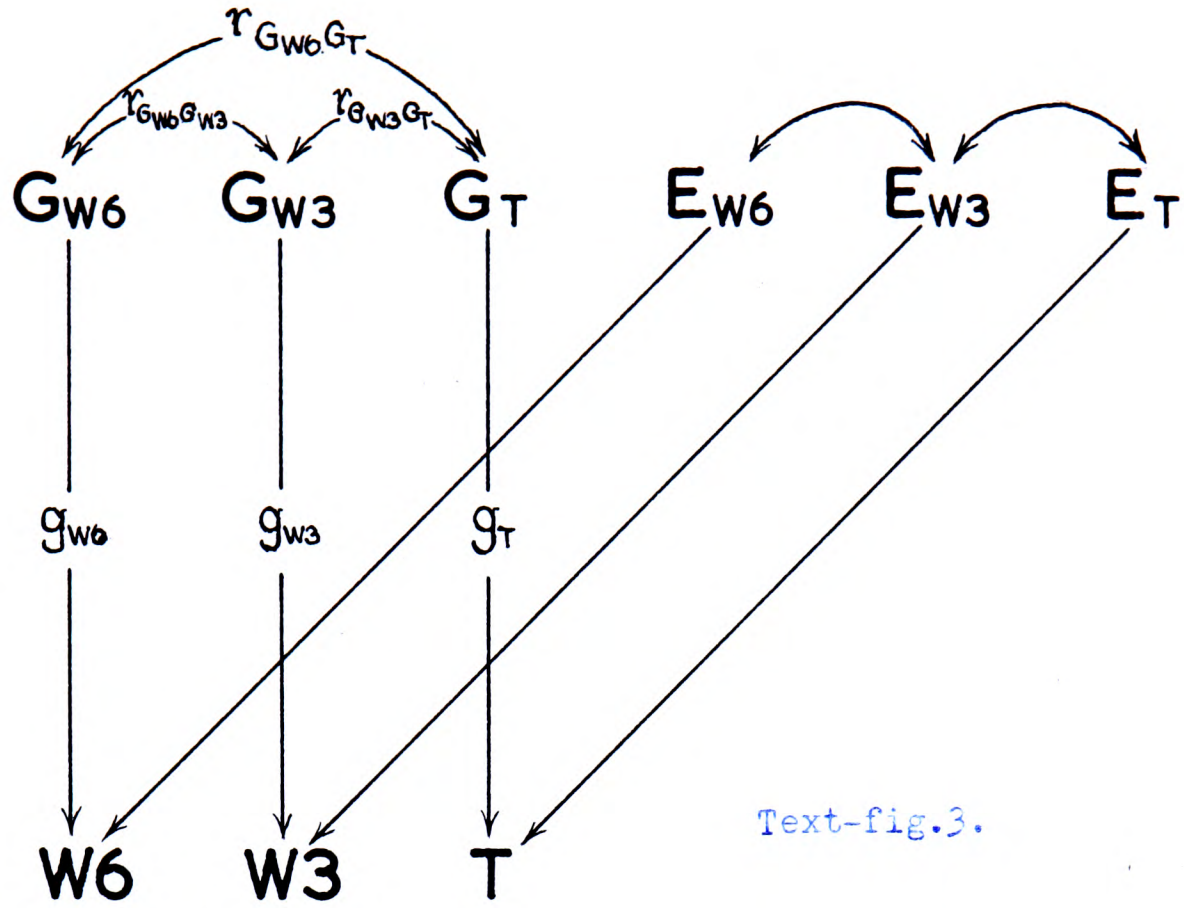
From this diagram three simultaneous equations may be obtained by summing all paths between two variables in order to get the correlation coefficient between these variables.

$$r_{W6.G_{W6}} = p_1 + p_2 \cdot r_{W3W6} + p_3 \cdot r_{W6T}$$

$$r_{W3.G_{W6}} = p_1 . r_{W3W6} + p_2 + p_3 \cdot r_{W3T} \qquad \text{(i)}$$

$$r_{T.G_{W6}} = p_1 r_{W6T} + p_2 r_{W3T} + p_3$$

The three path coefficients, p_1, p_2 and p_3 give the optimum weighting factors to be attached to each measurement; they are the concrete regression coefficients b_1, b_2 and b_3 converted to standard measure. The phenotypic correlations r_{W6T}, r_{W3T} and r_{W3W6} in the above equations can be obtained by straightforward calculation but the remaining correlations are not so easily obtained and require another path coefficient diagram (Text-fig. 3)/



Text-fig.3.

This diagram differs from that given in Section VII only in the addition of an extra variable. By the argument used there it follows that :

$$r_{W6.G_{W6}} = g_{W6} = h_{W6}$$

$$r_{W3.G_{W6}} = g_{W3}\, r_{G_{W3}.G_{W6}} = h_{W3}\, r_{G_{W3}G_{W6}}$$

$$r_{T.G_{W6}} = g_T\, r_{G_T.G_{W6}} = h_T\, r_{G_T.G_{W6}}$$

A method of calculating genetic correlations has also been given in Section VII so that the remaining unknowns $r_{G_{W3}.G_{W6}}$ and $r_{G_T.G_{W6}}$ can be estimated. Substitution can be made in the simultaneous equations (i) enabling p_1, p_2 and p_3 to be calculated, and hence b_1, b_2 and b_3 giving the required index.

The efficiency of the index is measured by the size of the correlation between the index and the genotype for 6-week weight. This correlation can also be calculated from Text-fig. 2,/

2, since by the principle of complete determination,

$$1 = \sum pr$$

$$1 = p_1 r_{W6.G_{W6}} + p_2 r_{W3.G_{W6}} + p_3 r_{T.G_{W6}} + p_4 r_{R.G_{W6}}$$

But, since there is only one path between R and G_{W6},

$$p_4 = r_{R.G_{W6}}$$

$$p_4 r_{R.G_{W6}} = r^2_{R.G_{W6}}$$

and, therefore -

$$r^2_{R.G_{W6}} = 1 - (p_1 r_{W6.G_{W6}} + p_2 r_{W3.G_{W6}} + p_3 r_{T.G_{W6}})$$

This expression measures the degree of determination by residual factors and so to find the degree of determination by the index it is merely necessary to subtract this expression from one.

$$r^2_{I.G_{W6}} = 1 - \left[1 - (p_1 r_{W6.G_{W6}} + p_2 r_{W3.G_{W6}} + p_3 r_{T.G_{W6}})\right]$$

The required correlation is therefore :

$$r_{I.G_{W6}} = \sqrt{p_1 r_{W6.G_{W6}} + p_2 r_{W3.G_{W6}} + p_3 r_{T.G_{W6}}}$$

This correlation may be compared with the correlation for selection on 6-week weight alone, $r_{W6.G_{W6}} = h_{W6}$ and it will be seen that it is a measure of the progress which can be made by selection since this depends upon the square root of the heritability. (Section IV.).

The foregoing method will be used to calculate a selection index for the GML stock. The necessary estimates of the phenotypic and genotypic constants of that stock are given in Table XIII.

All the constants are calculated within generations for all mice and are appropriate for mass selection. For an

index/

index to be used for selection within litters the only alteration necessary is to calculate all constants within litters within generations.

The three correlations between the three measurements of size and the additive genotype for 6-week weight are first calculated.

$$r_{W6.G_{W6}} = h_{W6} = \sqrt{0.342} = 0.585$$

$$r_{W3.G_{W6}} = h_{W3}\, r_{G_{W6}.G_{W3}} = 0$$

$$r_{T.G_{W6}} = h_{T}.r_{G_{W6}G_{T}} = \sqrt{0.439} \times 0.227 = 0.150$$

It is now possible to substitute in the simultaneous equations (i). Using the figures for females this gives :

$$0.585 = p_1 + p_2 \times 0.545 + p_3 \times 0.476$$

$$0 = p \times 0.545 + p_2 + p_3 \times 0.440$$

$$0.150 = p \times 0.476 + p_2 \times 0.440 + p_3$$

the solution of which is

$$p_1 = 0.854$$

$$p_2 = -0.437$$

$$p_3 = -0.064$$

The path coefficients are converted to concrete regression coefficients for use in the selection index.

$$b_1 = p_1\frac{\sigma_{GW6}}{\sigma_{W6}} = 0.499 \qquad b_2 = p_2\frac{\sigma_{GW6}}{\sigma_{W3}} = -0.400 \qquad b_3 = p_3\frac{\sigma_{GW6}}{\sigma_{T}} = -0.026$$

The efficiency of the selection index is unaffected by dividing all weighting coefficients by a constant, since this does not alter the correlation $r_{I.G_{W6}}$, so that dividing by

0.499/

0.499 gives the index in the form :

$$I = W6 - 0.80\ W3 - 0.05\ T$$

Similar calculations with the figures for males gives :

$$I = W6 - 0.93\ W3 - 0.01\ T$$

The efficiency of these selection indices is measured by their correlations with the genotype for 6-week weight :

$$r_{I.G_{W6}} = \sqrt{p_1 r_{W6.G_{W6}} + p_2\ r_{W3.G_{W6}} + p_3\ r_{T.G_{W6}}}$$

For females this multiple correlation coefficient

$$r_{I.G_{W6}} = \sqrt{0.854 \times 0.585 + 0 - 0.064 \times 0.150}$$

$$= 0.700$$

and for males = 0.676

The correlations are 19.6% and 15.6% larger for females and males respectively than the correlation of 6-week weight with the additive genotype for 6-week weight which is 0.585. Selection using the calculated indices is therefore predicted by averaging the improvement in each sex to give $\frac{1}{2}$ (19.6 + 15.6) = 17.6% improvement over selection on 6-week weight alone.

The general form of these indices can be rationally explained. The 3-week weight of an individual provides a measure of the environment which the individual has experienced up to that age. Since the 3-week weight contributes to the individual's phenotype at 6 weeks it is possible to subtract this contribution in order to get a better measurement of the individual's genotype at 6 weeks. Three-week weight is thus

a/

a good environmental indicator and therefore receives negative attention in the selection index. The differing amount of attention given to 3-week weight in males and females arises from the observed correlation between 6-week and 3-week weight being lower in males than in females. This difference is due to males growing more than females in the period from three to six weeks and so being less dependent on weaning weight.

In both sexes the selection index shows that very little attention should be paid to tail length. The size of the genetic and environmental correlations between tail length and 6-week weight are such that tail length is neither a good genetic indicator or a good environmental indicator and its exclusion from the index would therefore make very little difference to the efficiency of the index. Thus, for females the simplified index is

$I = W6 - 0.85\ W3$ with $r_{I.G_{W6}} = 0.698$ giving an expected 19.3% improvement, over straight selection; for males the index is

$I = W6 - 0.94W3$ with $r_{I.G_{W6}} = 0.676$ giving an expected 15.6% improvement. Tail length can therefore be eliminated from the selection indices with a negligible loss in precision.

A further simplification of these indices is possible without a great loss in precision if the selection is made on gain from three to six weeks, which is in fact equivalent to using the index $I = W6 - W3$ in both sexes. The correlation of gain with the genotype for 6-week weight comes to 0.693 for females/

females and 0.676 for males giving expected improvements over straight selection of 18.5 and 15.6% respectively. From the small changes in precision produced by quite large changes in the selection indices it is clear that the general form of the index is well defined and precise weighting coefficients are unnecessary.

To conclude this section the recommendations for the improvement of size may be briefly summarised. Firstly, selection should be based on the gain from three to six weeks; secondly, two litters should be raised from each mating; thirdly, if it is possible to double the size of the population a change should be made to mass selection. The first recommendation is expected to give an improvement, taking the average of the two sexes, of 17%; the second a negligible improvement at present but considerable advantage when heritability declines; the third an improvement of 39% over selection within litters in a population of the same size. Thus, with mass selection on gain with sixteen matings gives an expected improvement per generation of

$$\Delta I = j\,\Delta F + h^2\,\Delta P$$
$$= -0.073 \times 1.5 + (0.43 \times 1.01 \times 2.802) \times \frac{117}{100}$$
$$= 1.016 \text{ gm.}$$

Selection within litters on 6-week weight with eight matings (the present system) gives an expected improvement of

$$\Delta I = -0.073 \times 1.7 + 0.31 \times 1.01 \times 2.151$$
$$= 0.549 \text{ gm.}$$

The increased improvement in 6-week weight which can be

expected/

expected from the simple changes recommended is therefore 85% - a not inconsiderable gain.

IX. GENERAL DISCUSSION AND CONCLUSIONS.

It is now possible to discuss the more general aspects of the results obtained in the present experiment and to examine some of the consequences of these findings, pointing out their implications in the field of animal breeding.

1. Causes of a declining response to selection.

In the introduction the possible causes of a declining response to selection were divided into two main categories :

(1) A loss of genetic variance,

(2) An approach to a physiological barrier.

The first possibility was put to a direct test by crossing two strains of mice which had been subject to many generations of selection for large size, one strain being apparently at a selection limit and the other showing only a small response to selection. This cross produced new genetic variance and selection for large size became more effective than in either parent strain. The conclusion to be drawn is therefore that, in the strains used, the declining response to selection can be attributed to a loss of genetic variance. Alternative explanations for a declining response which fall into the second category have not been subject to experimental test but reasons for believing that they are not important will now be given.

The term "physiological barrier" or "physiological limit" is used to denote a hypothetical limit set by the physiology of the animal beyond which it is supposed either that the internal organs could not meet all metabolic

requirements or, that the appendages could not provide the movement necessary for survival. This concept of a ceiling set by physiological factors divorced from the character selected may or may not be a useful one, but accepting it as a reality for the purposes of argument the consequences are clear. The size of the internal organs in proportion to the size of the body will determine the physiological barrier. This proportion is determined by allometric constants and since these constants appear to be unaltered by selection for size (MacArthur and Chiasson, 1945) or even by direct selection (Lerner,1943a) the ceiling must be at the same level for all genotypes in the population. Having fixed the ceiling the problem now resolves itself into one of scaling and can be decided by seeing whether the upper end of the scale requires any damping as suggested by Rasmussen (1933) to allow for the approach to a physiological barrier. The problem of scaling is discussed in Appendix I where it is decided that a linear scale is adequate for the range of size in the present experiment. In addition the frequency distribution of weights shown in Fig. 8 shows no signs of curtailment at the upper end of the scale. It can, therefore, be concluded that if a physiological barrier to size increase exists in the mouse it has not yet been encountered.

An allied explanation for a declining response to selection is provided by supposing that natural selection reduces the "fitness" of animals which approach nearest to the physiological/

physiological barrier (or furthest from the original mean). Evidence for the occurrence of this process has been produced in a selection experiment with Drosophila (Mather and Harrison, 1949), in which a selection for the number of abdominal chaetae had been made. When mass matings were made in the selected line the mean number of chaetae fell in subsequent generations owing to the elimination by natural selection of flies with the highest number of chaetae. In mice the same possibility must be considered since natural selection, acting against the heaviest phenotypes, might be counteracting the artificial selection for large size. This possibility is at once discounted by the small response to selection for small size in Goodale's and MacArthur's Large Lines when natural selection should be assisting the artificial selection.

Further, it is difficult to see how natural selection could operate in the stocks used in the present experiment. The progress made is measured in relation to the amount of selection which is applied and the method of weighting the selection differential according to the number of offspring measured eliminates the possible effects of differential fertility. Differential mortality is also unlikely, since although post-natal mortality exists those animals which die are almost invariably the lightest in the litter. Possibly this could merely result from disease being the agency of natural selection but this is very unlikely since these ailing mice are at no time markedly larger

than/

than their litter-mates. Any large pre-natal mortality can also usually be ruled out by the large litter size characteristic of large mice. It is, therefore, safe to assume that natural selection is not important in the present experiments. Having also decided that a physiological barrier is also not operative it is possible to return to the first explanation for a declining response to selection, namely a loss of genetic variance, and to examine this in more detail with the knowledge that if other factors are operative, at least they are not important.

The two processes which are known to reduce genetic variance are inbreeding and selection. It is therefore desirable to attempt to assess the importance of each in causing homozygosity. Early experimenters on quantitative inheritance tended to overestimate the importance of selection in causing homozygosity. Thus Payne (1920) comes to the conclusion that his lines of Drosophila selected for high and low bristle number "have not only separated by selection but have become fixed by selection," despite the fact that during selection these lines had been subject to sixty generations of brother-sister mating. Wright's (1921) papers, however, removed such misconceptions when he calculated the loss in heterozygosis due to selection for a character depending on n pairs of equal allelomorphs and possessing a heritability, h^2. With a selection differential of s standard deviation units the loss of heterozygosis expected per generation is $\frac{s^2h^2}{2n}$ of the total heterozygosis present. Clearly for a polygenic character influenced/

influenced by the environment this fraction will be very small indeed. Such a simple model, as Mather has pointed out, requires modification when the concept of linkage is introduced since the effective number of factors is reduced. In a species such as the mouse with many chromosomes and a high chiasma frequency, this modification is probably negligible. Since the fraction $\frac{s^2h^2}{2n}$ depends directly upon the amount of genetic variance the fixation produced by selection will continually decline. Theoretical considerations therefore would point to the unimportance of selection as the cause of the loss of genetic variance for a polygenic character and enable the rate of decline to be compared with the rate of inbreeding.

Unfortunately, there is no data available in which the decline in the response to selection can be accurately compared with the calculated rate of inbreeding. MacArthur (1949) observed a fall in the heritability of sixty-day weight from approximately 25% to 10% in twenty generations, but the rate of inbreeding was only specified by "1-3% per generation." In Section IV the actual rate of inbreeding was roughly placed at an average of 3% per generation when allowance was made for the effects of progeny testing. Using this figure the loss of heterozygosity in twenty generations would be $(0.97)^{20} = 54\%$. The decline in heritability, however, indicates a larger loss of heterozygosity for the following reason. Assuming that the environmental variance is constant the genetic variance falls from 1/3 to 1/9 of the environmental variance, which is a 67% reduction. In view of the great uncertainty with which the

rate/

rate of inbreeding is estimated this discrepancy cannot be considered serious.

The results of the present experiment provide a little information on the agreement between inbreeding and decline of response. In MacArthur's Large Line a heritability within litters of 16% was observed for 6-week weight which it was calculated corresponded with an initial heritability in MacArthur's foundation generation of 23%. This figure is reasonable and shows that there cannot be a gross error in the computed reduction in heterozygosity. In Goodale's Large Line no response to selection for 6-week weight was found although the very rough estimate made in Section IV showed a computed reduction in heterozygosity of only 38%. It appears that there may be a real discrepancy here and yet the uncertainty of the estimate of inbreeding makes it impossible to be certain on this point. Selection in the cross-breds will have to proceed for many more generations before a comparison can be made in this stock but when this does become possible the knowledge of the exact degree of inbreeding (see Section III) will make the comparison of great value.

Two-way selection in the cross-breds to date has, however, revealed a factor which can cause a selection limit before all genetic variance is exhausted. This is a phenomenon known as "inbreeding depression" or "inbreeding degeneration." This degeneration observed in various characters on inbreeding is the complement of the heterosis observed on outcrossing.

It/

It is, therefore, relevant to draw attention to the heterosis observed on crossing Goodale's and MacArthur's stocks. Although the increase in size on crossing these two partially inbred stocks was not large it was as large as those observed by Eaton (1941) in crosses between long-inbred strains of mice. The occurrence of heterosis in size on crossing both selected stocks and unselected inbred strains of mice shows that inbreeding depression must be expected and therefore if possible incorporated into an additive scheme.

This has been done by assuming that the change in size produced by additively acting genes can simply be added to the change in size due to the non-additive action of genes in causing an inbreeding depression. The validity of this assumption has yet to be rigorously tested and is urgently needed, for, if this scheme is adequate, then it has important implications. Continued selection in a stock which has reached a limit or is even declining in merit owing to inbreeding depression will still be undergoing improvement which will be revealed on crossing. Thus, the improvement found in the first generation of a cross should be maintained without selection until inbreeding depression sets in once more.

2. The prediction of progress under selection.

It is desirable to decide the value of heritability estimates based on one or two generations for predicting the future progress which can be made by selection. In the present experiment heritability estimates based on the resemblance between offspring and parent were not very precisely determined since the structure of the population was not arranged for this purpose but as far as they go these estimates were not in disagreement with the progress obtained by selection. The hazards in using heritability estimates for prediction are, however, great.

The first requirement which has to be met is that the estimate does in fact measure the extent of additive genetic variance. The resemblance between two relatives may also be due to non-additive gene action or a common environment. Even when a true measure of additive genetic variance has been obtained there are further dangers in predicting progress.

The additive genetic variance is probably due to numerous genes some with large effect, some with only small effect. The major factors will make the most important contribution to variance and to progress in the first few generations but they will rapidly become fixed by selection and further response to selection will depend upon genes with smaller effects. Long-continued progress, which may be more important eventually than a rapid response, will be due to numerous genes with small effects. It would therefore be helpful to know the frequency distribution of size of effect

in/

in the gene complement. Unfortunately, there does not appear to be much possibility of doing this. The difficulties of detecting a major factor have already been discussed and it was decided that such a factor would probably segregate unnoticed. A single major factor might be isolated by repeated backcrossing on to an inbred line but its identification in outbred material is thereby made no easier. There is a further complication at this point in that at the level of the individual gene interactions become important. For instance, the data of the present experiments suggest that several colour genes have no action on size in the cross-bred stock whereas other workers have found a measurable effect. The only method of treating genes influencing a quantitative character would, therefore, appear to be to treat them en masse. There is an alleviating feature in the calculations of Panse (1940) which show that the effects of very different gene complements should not differ greatly so that the loss of predictive value in treating all genes as polygenes should also not be too great.

There now remain two further mechanisms liable to cause a gross deviation of the progress realised under selection from that predicted by heritability estimates. The first is inbreeding depression which needs no further comment here except to point out that the size of this effect is unpredictable. The second is a possibility of a buffering system provided by negative genetic correlations. For instance, the 6-week weight of the mouse is influenced by the maternal environment/

environment and if this environment which the dam provides were negatively correlated with her genotype for 6-week weight progress under selection would be hindered. There was no evidence that this mechanism was operative but it is to be expected in selection experiments in which the measured character has two components. As Dickerson (quoted by Lerner, 1950) has pointed out genes affecting both components favourably or unfavourably will rapidly be fixed or eliminated by selection leaving heterozygous those genes acting favourably on one component and unfavourably on the other. Thus, negative genetic correlations are to be expected.

Heritability estimates therefore require supplementary data on the genetic situation before they can safely be used for predicting the progress under selection.

3. The improvement of farm animals.

The present experiment has raised several points of relevance to the improvement of farm animals. It must be pointed out that although it may be convenient to regard the present experiment as a "model" of a situation which may arise in agricultural practice the dangers of so doing are not to be overlooked. Nevertheless laboratory mammals can bridge the wide gap between purely academic research on quantitative inheritance and the practical improvement of farm animals. With increasing knowledge of the genetics and physiology of different mammalian species it should become possible to know where results from one species may be applied to another and where they may not. Meanwhile experiments with laboratory mammals can at least suggest fruitful lines of research for the experimenter with farm animals. Having made these qualifications it now appears legitimate to draw analogies and to make suggestions for the improvement of farm animals.

a) Cross-breeding.

The present experiment may be regarded as a "model" experiment paralleling the situation which would arise by the crossing of two distinct breeds of livestock. Almost all long-term selection experiments have shown that a declining response is found after a number of generations of selection and eventually a selection limit is reached. The continued selection within breeds of farm animals would thus appear to produce, sooner or later, no more response and the problem arises as to the manner in which further improvement may be made. How

far/

far removed the present breeds of farm animals are from their selection limits for economic characters it is impossible to say but at some time in the future this problem will apply with full force to farm animals. The two long-selected stocks of mice in the present experiment are analogous to breeds of live-stock and the crossing of them illustrates a method which may be useful when the need arises in farm animals.

The increased improvement which could be made by crossing the mice was due to two causes, both important. The first cause which was more important in the present experiment was the creation of new genetic variance which became available for selection. The second cause, subsidiary here, but possibly of greater importance in other situations, was the increase in fertility which made possible intense selection and hence the best use of the new genetic variance. The outstanding difference between the cross-bred and the parent stocks was the superiority of the cross-breds in fertility, lactation and general health; in fact, superiority in those characters which constitute the rather ill-defined term of the animal breeder "vigour". A decline in fertility appears to be a general occurrence in long-term selection experiments, being found for example by Zeleny (1920) and Mather (1941) in Drosophila and by Goodale and MacArthur (MacArthur, 1949) in mice. Part of this decline may be due to the inevitable inbreeding in a closed population, but in addition Mather (1941) has suggested a negative genetic correlation between the selected character and fertility may be responsible. He visualises genes for fertility being scattered/

scattered through the chromosomes and liable to fixation or elimination owing to their linkage with genes affecting the selected character. But whatever the cause of the decline in fertility it is removed by cross-breeding. Thus, in every respect the "model" experiment with mice suggests that when selection within breeds of farm animals becomes ineffective crossing of two or possibly more breeds will make further improvements possible.

b) Choice of methods for the improvement of farm animals.

In the introduction it was pointed out that the two main methods of improvement open to the animal breeder were selection without intense inbreeding or inbreeding followed by crossing, the choice depending on whether additive or non-additive gene action was more important. At the commencement of any scheme of animal improvement relatively little will be known about the exact partitioning of the genetic variance into additive and non-additive components, and so the choice of schemes will of necessity be somewhat arbitrary. However, an initial trial of improvement by selection without inbreeding would have advantages over the alternative method, since it would provide an indication of the initial improvement which can be made, and would give some indication of the potentialities of the method. Should additive genetic variance be low then any remaining gene differences with large effect would be fixed and a change could then be made. Inbreeding and crossing is, at the moment, a matter of trial and error with no guides to the amount/

amount of inbreeding desirable before crossing or any indication of the future progress which can be made. In fact, until more is known of the causes of heterosis such a method of animal improvement appears unpromising. In the U.S.A. the extensive scheme of inbreeding and crossing pigs, started as a result of the success of hybrid maize, is beginning to produce results. Improvements may be expected; but could not greater improvements have been made by selection without inbreeding? This question remains unanswered and the alternative method of improvement may still prove the better.

It is true that selection has been employed in the past and yet there is still room for the improvement of our livestock. This is hardly surprising when all the factors which reduce the intensity of selection are considered. For instance, in dairy cattle the short average life of herds (Donald and El-Itriby, 1946) and the small amount of selection for milk yield even in the best herds (Rendel and Robertson, 1950) show that intense and prolonged selection awaits a proper trial. But even if inbreeding and crossing are desirable, selection during this process is still desirable. The results of the present experiment suggest that a hidden improvement may be made by selection when a character is regressing owing to inbreeding and that on crossing this improvement will become manifest. In addition calculations on the methods for improving size in the mouse have shown that changes in the structure of the breeding population and the use of a selection index could give a not inconsiderable gain in efficiency. These methods can likewise/

likewise be expected to hasten the improvement of farm animals.

Finally, the experiment described in this thesis provides evidence, though of necessity only corroborative, on the validity of the methods employed by Lush and his school in that these methods provided a satisfactory interpretation of the experimental results. With the accumulation of similar evidence from other experiments some faith may therefore be vested in breeding plans based on these methods.

SUMMARY

1. The importance of a knowledge of size inheritance for an understanding of the evolutionary tendency for size to increase and of the improvement of farm animals is pointed out. The literature on size inheritance is reviewed for both laboratory and farm animals. Methods of measuring sources of variation are examined and the unexploited method of selection is discussed. The results of such analyses of variation in farm animals are shown to be important in determining the appropriate method of improvement.

2. The present experiment was designed to investigate by a study of size inheritance the causes of a declining response to selection and the attainment of a selection limit which from a survey of long-term selection experiments appears to be a universal phenomenon. The two categories of explanation offered for this phenomenon are either a loss of genetic variance or the existence of a physiological barrier to further improvement. The first explanation was tested by crossing two lines of mice long-selected for size. One line, selected by Goodale, was found to be at a selection limit, with most of the remaining genetic variance being apparently non-additive; the other, selected by MacArthur, showed a small response to selection, heritability being estimated at 10%. The crossing of these two lines created new genetic variance and lines selected for large and small size showed a renewed response to selection,

heritability/

heritability being estimated at 34%.

3. The conclusion is drawn that the loss of genetic variance was responsible for the declining response to seledtion and the selection limit found in the parent lines. The possibilities of a physiological barrier and of cpposing natural selection when the physiological barrier is approached are examined and discounted. From the data available it is not possible to decide the relative importance of inbreeding and selection in causing a loss of genetic variance.

4. It was found that it was easier to decrease the size of cross-bred mice by selection than to increase it. This difference is attributed to inbreeding depression and it is shown that the observed inbreeding depression found on inbreeding the cross-bred mice without selection could account for this difference.

5. An anomalous result was found in the first few generations of selection in the cross-bred mice. The rapid response to selection expected in the F_3 as a result of segregation in the F_2 did not occur until the next generation. No genetic mechanism could be found which could give an adequate explanation of this result.

6. Correlated responses to selection for 6-week weight were found in 3-week weight and tail length at 6-weeks. Tail length/

length is calculated to have a genetic correlation of 0.23 and on environmental correlation of 0.63 with 6-week weight. No correlated responses in the size of the first litter were observed.

7. The frequency of colour genes in the cross-bred mice showed no pronounced changes and it was discovered that the colour genes brown, b, and leaden, ln, (mistakenly called dilute by MacArthur) apparently did not have their reported pleiotropic action on size in this stock.

8. Various types of selection are considered for the purpose of obtaining the required maximum divergence in 6-week weight per unit of time between lines selected for large and small size. With a small population size (8 matings) it is found that selection within litters minimises inbreeding and so is most suitable. It is also found that only one litter should be raised from each mating.

9. The best method for the improvement of 6-week weight in the cross-bred line is investigated and it is recommended that :-

(a) Selection should be made on gain from 3-6 weeks.

(b) Two litters should be raised from each mating.

(c) If possible, the number of matings should be increased from 8 to 16 and mass selection adopted.

If all these recommendations were carried out the predicted improvement in the increase of size per unit of time is 85%.

10./

10. The importance of the present experiment is discussed in relation to animal breeding. The limitations of heritability estimates as a means of predicting future progress by selection are pointed out. An analogy is drawn with the declining response to selection which might be encountered in breeds of farm animals. Cross-breeding and selection are suggested as a means of improvement which, although of considerable antiquity, could by due attention to the genetical principles involved give far greater improvements in the future.

APPENDIX I

CHOICE OF AN ADEQUATE SCALE

APPENDIX I

CHOICE OF AN ADEQUATE SCALE

The first step in the analysis of quantitative variability is the choice of a scale which will make genetic and environmental factors as nearly additive as possible. This is done in an attempt to minimise interactions between genotype and environment or between loci. If the scale does not minimise these interactions it cannot be considered adequate for a biometrical analysis.

Mather (1949) has pointed out that the choice of an appropriate scale for a genetical analysis must be arrived at empirically. Even when, as in some data on size, there is some *a priori* reason to suppose that a logarithmic transformation is needed it is necessary to check this assumption. This point is well brought out by work on the size (weight per locule) of tomatoes by Powers (1950) who found that although in many varieties a logarithmic scale was better than an arithmetic scale this was not true for all varieties or for the same variety in different years. It is therefore necessary to attempt to find empirically the best scale for the data on hand. This may be attempted in various ways.

Mather (1949) bases his scaling tests on the relation between the means of parent lines, F_1, F_2 and backcrosses. With many genes having additive effects it is expected that the means will have the following values.

$\bar{F}_2$/

$$\bar{F}_2 = \tfrac{1}{4}(\bar{P}_1 + \bar{P}_2 + 2\bar{F}_1)$$

$$\bar{B}_1 = \tfrac{1}{2}(\bar{P}_1 + \bar{F}_1)$$

$$\bar{B}_2 = \tfrac{1}{2}(\bar{P}_2 + \bar{F}_1)$$

These tests can be quite sensitive with certain material but in the present experiment the means of parents, F_1 and F_2 are all quite close and the variance between generation means is so great that this method could not detect gross departures from additiveness.

A suitable scale should also make the frequency distribution of weight normal unless there are a few major factors regregating and causing shewness. The frequency distribution of weights for GML animals is shown in Fig.8, and it appears to be normal. The detection of departures from normality require large numbers (See Section VI) and when major factors are a real possibility the criterion of normality does not help and it is therefore necessary to use other methods of scaling.

Systematic changes of variance when the mean measurement changes indicate that re-scaling is needed. MacArthur (1944) found that selection for large size in mice increased the variance measured in gm^2 and selection for small size decreased it so that an arithmetic scale was clearly not adequate over his range of size. Coefficients of variation, on the other hand, remained approximately constant showing that a logarithmic scale gave a better fit. This was in agreement with the early results of selection which were approximately equal on a logarithmic/

logarithmic scale.

Complete reliance cannot, however, be placed on the magnitude of variances in determining the most appropriate scale. The continual selection of extreme individuals may result in the production of animals especially sensitive to environmental agencies and so showing an increased variance. This mechanism could explain why MacArthur using a logarithmic scale found no decrease in total variability despite a loss of genetic variability. While this mechanism may or may not operate, different stocks of mice have different variances at approximately the same mean. Table XIV shows the intra-litter variances for the GL, ML, GML and GMS lines and their overall means. The probable amount of genetic variance decreases in the order GML and GMS, ML and GL and if all lines reacted equally to the common environment of the laboratory the environmental variance should be the same for each; so, assuming additiveness, the phenotypic variance of the lines should also decrease in the same order. In fact, the observed order is almost the reverse of this, the line which is expected to have the smallest phenotypic variance having the largest. Examination of the means shows that no rational transformation could place these variances in the correct order, thus indicating that there are real differences in reactivity to the same environment. If this is so, little reliance can be placed on variance in determining the appropriate scale.

There is another method of arriving at an adequate scale which depends on a suitable transformation making a

given/

given genetic or environmental difference constant over all parts of the scale. This criterion of scaling was recognised by Zeleny (1920) who found a suitable scale for facet number in Bar-eyed Drosophila by measuring the effect of different strains and temperatures on facet number and then making these differences uniform by the use of "factorial units". Since the experimental results are based on means and not variances the method is quite sensitive. The major objection to this method is that it is merely a transformation of the scale to make one effect additive - a very valid objection but one which may be met by the use of several genetic or environmental effects and seeing if they agree.

In determining the appropriate scale for 6-week weight of the mouse use may be made of the sex difference in mean weight. If the arithmetic scale is suitable the difference in mean weight of males and females should be constant irrespective of the mean but, if, on the other hand, a geometric scale is better the difference between the means of the logarithms of weights should be constant. The computation of these means is unnecessary as it is shown later that a very good approximation is made by comparing the differences in the logs of the mean weights. Table XV shows the sex difference on these two scales for several lines of mice including in addition to the lines used previously, three inbred lines and a line selected for small size. On the arithmetic scale the sex difference decreases fairly regularly with mean size. The fit on the geometric scale is clearly better; with the one exception of MacArthur's Large

Line/

Line the agreement is good and there is no remaining trend for the difference to decrease with the mean. This use of the sex difference in determining scale is open to the criticism that, depending on the importance of sex-linked genes, the sex difference which is measured is not the same for all lines. Since the differential segments of the sex chromosomes are only a very small fraction of the total chromosome material in the mouse this criticism would not seem to be important but in view of the aberrant sex difference observed in MacArthur's Large Line, the use of a known single gene difference would be preferable.

The gene pygmy (Appendix II) seems very suitable for this purpose since when homozygous it reduces size to rather less than half the size of normals. Moreover, investigation of the endocrine organs of pygmy homozygotes has revealed no abnormalities suggesting that the size difference is produced in the same way as it is by minor size genes. If this is so this major size gene should act on the same scale as these minor genes which we are trying to fit a scale to. Table XVI shows the mean weights of pygmies and normals in the line selected for small size where the mutation was found and after three generations of back-crossing to the cross-bred line. The means given are based on about ten pygmies of each sex and in each stock and on a largernumber of normal litter-mates. Comparison of the difference in mean weight between normals and pygmies on an arithmetic and a logarithmic scale shows that the

latter/

latter again gives the better agreement.

All the available evidence from several stocks of mice and covering a wide range of size agrees in showing that a logarithmic scale is appropriate for the analysis of weight measurements. If the logarithmic transformation is the correct one the distribution of the logs of weights will be normal except for the effects of any major factors. Assuming normality on this transformed scale the parameters of this distribution can be calculated from those of the distribution on the original scale. Where v represents the variable and c its coefficient of variation both measured on the original scale, *

$$\text{Logarithmic mean} = \overline{\log_e v} = \log_e \bar{v} - \tfrac{1}{2}\log_e(1+c^2)$$

$$\text{Variance on log scale} = \sigma^2_{\log_v} = \log_e(1+c^2)$$

$$\text{Form index} \qquad \gamma_1 = c\,(3+c^2)$$

The second relationship leads to a commonly used test for a logarithmic scale. If the logarithmic scale is adequate, the variance $\sigma^2_{\log v}$ should be constant irrespective of the mean and this will be so if c^2, the coefficient of variation is constant. The formula for the form index γ_1 shows that if c is small the skewness on the original scale is negligible. For the 6-week weight of the mouse c is of the order of 12% and consequently it is possible to work in grams with little loss/

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* Equations given by Professor Sewall Wright during lectures in the University of Edinburgh, 1949-50.

loss of precision either in the determination of the mean or variance,or through skewness. For the analysis of size differences within a stock the use of an arithmetic scale is thus permissible but when crosses are made between two stocks differing greatly in mean weight, the geometric mean must be used for the mid-parent point since this will be overestimated on an arithmetic scale. Similarly, when measuring progress due to selection for weight the use of a geometric scale is desirable once the change in size becomes an appreciable fraction of the mean. For the range of size described in this thesis a transformation to the correct logarithmic scale is however unnecessary.

APPENDIX II

PYGMY, A DWARFING GENE IN THE HOUSE MOUSE.

The following account of a single-factor size difference in the mouse takes the form of a paper accepted for publication by the Journal of Heredity.

PYGMY, A DWARFING GENE IN THE HOUSE MOUSE

In the course of his selection of mice for small size MacArthur[1] noticed that certain matings produced a proportion of undersized animals or "runts". These individuals were noticeably smaller than their normal sibs at 12 days and their growth was so retarded that, at 60 days, they were less than half the weight of normals and seldom weighed more than 9 gm.

In 1948, Professor MacArthur kindly sent Dr. Falconer of this laboratory some of his selected small mice to form the basis of another "small" stock. In addition, mice of the highly inbred C57 (black) and Strong-A strains were used as foundation animals. Two matings in the third generation of selection for small size produced a few animals distinctly smaller than their litter-mates and agreeing with the description of "recessive-like runts". The present author is indebted to Professor MacArthur for permission to investigate this condition further.

MacArthur's suggestion that these "runts" were produced by a recessive gene has been vindicated. The homozygote, the "runt", is sterile, but matings between heterozygotes have produced the expected proportion of "runts". Further, the condition can be recovered after outcrossing to various other stocks. Having established that these "runts" were produced by a mutant gene it seemed desirable to give the gene a definitive name, and to reserve the name "runt" for cases of undersized animals due to unknown causes. The name "pygmy",

symbol/

symbol pg, is proposed.

This is the third hereditary dwarfism to be reported in the house mouse. Snell[2] reported a form which subsequently proved to be due to an anterior pituitary deficiency and, more recently, Strong[3] has described the occurrence of a hereditary dwarfism in the descendants of mice treated for many generations with methylcholanthrene, a carcinogenetic agent.

Genetics.

Those matings in the Small stock which have produced pygmies have given a total of 331 normals to 105 pygmies when scored at 21 days. To test the goodness of fit to a 3:1 ratio a correction must be made for the bias introduced by the automatic exclusion of matings between heterozygotes which, by the chances of sampling, produced no pygmies. The average number of progeny per mating is 15.6 so that an approximate correction can be made by using

$$\tfrac{1}{4} \times \frac{1}{(1-(3/4)^{15.6})}$$

as the expected proportion of pygmies, and then using a $\hat{\chi}$ test.

	Normal	Pygmy	Total
Observed	331	105	436
Expected	325.8	110.2	
Deviation	5.2	5.2	

$$\hat{\chi}(1) = 0.33 \quad P > 0.5$$

The defieiency of pygmies, though not statistically significant, is probably real as some animals diagnosed as pygmies at 12 days died before 21 days, when all were finally scored. No estimate of the viability can yet be made because the/

the data are too meagre. It seems that deaths are due more to the inability to compete with their much larger litter-mates than to any inherent weakness.

Heterozygotes from the Small stock were outcrossed to stocks of large mice and also to various inbred lines. Intercrosses were set up from the progeny of these matings and some produced pygmies which segregated in a clear cut manner. These intercrosses have produced a total of 251 normals to 76 pygmies. The expectation is corrected as before for the average number of progeny per mating.

	Normal	Pygmy	Total
Observed	251	76	327
Expected	243.0	84.0	
Deviation	8.0	8.0	

$\chi^2_{(1)} = 1.03 \quad P > 0.3$

There is no indication that the recognition of pygmies is dependent on the genetic milieu of the Small stock. When the two sets of data given above are compared in a 2 x 2 contingency table they give a χ^2 for 1 d.f. of only 0.07 ($P > 0.7$).

The gene pygmy (pg) has been tested for identity with Snell's dwarfism (dw). Through the kindness of Professor T. Kemp a male heterozygous for Snell's dwarfism was obtained. This male (genotype +dw) was mated to two females both proven heterozygous pygmy (genotype +pg). These two matings produced 16 and 20 progeny respectively, none of which was noticeably smaller than its sibs. Since, if the two genes were identical, the probability of obtaining no homozygote in a progeny of 36 is/

is only $(3/4)^{36}$ or 0.000032, it must be concluded that the two genes are not identical.

It has not been possible to test pygmy for identity with Strong's hereditary dwarfism. Phenotypically, however, the two conditions seem quite distinct since Strong reports that two of his dwarfs were mated together and produced two litters, whereas pygmies are sterile in both sexes.

Description

The growth pattern of pygmies differs considerably from that of Snell's dwarfs. The latter cannot be distinguished with any certainty before 12 days, although Francis[4] could trace the retardation in growth back to 4 days. Pygmies on the other hand are markedly smaller than normals at birth. Their identification at this age, however, is not certain as the birth weights of pygmies and normals overlap.

By about 4 days the pinna of the ear has become free and has flattened out. At this age the relatively smaller size of the pinna in pygmies provides an additional and more reliable criterion for classification. By 12 days the difference in ear size is pronounced and classification on this basis is easy and certain (Fig. 1).

The growth of pygmies follows a very similar course to that of normals, though at a much reduced rate (Fig.2). A noteworthy feature is the negligible increase in the average weight of pygmies from 12 to 21 days. This stationary period can probably be ascribed to the intensity of competition with normals/

normals for milk. Daily weighings of individual pygmies in litters which were progressively reduced in size to give them more milk showed a steady increase in weight over this period. This is unlike the definite loss in weight shown by Snell's dwarfs in the period 17 to 25 days which Boettiger and Osborn[5] found to be independent of the nutritional status.

The adult pygmy differs in proportions as well as size from a normal mouse (Fig.1). Relative to the body length, the ears are short, the feet rather short and the tail long, as reported by MacArthur and Chiasson[6]. The coat appears to be as long as that of normals and so appears relatively longer. It is kept well groomed so that the general appearance is that of a sleek, streamlined animal. Quite unlike Snell's dwarfs, adult pygmies are at least as active as normals. Aided by their small teeth, they quite often demonstrate their activity by enlarging the ventilation holes of the aluminium cages in which they are kept, and so escaping.

Both sexes have so far proved to be sterile, but the cause of the sterility is not known. In the male, the testes are small and do not always descend into the scrotum. Most males develop priapism, usually by the age of 6 weeks. The priapism does not seem to be correlated with the descent of the testes. A few males have been obtained with descended testes and without priapism, but their sexual activity has not been observed to pass the stage of interest in oestral females.

Pygmy/

Pygmy females have open vaginae, often showing traces of blood. The uterus and ovaries are small, but the latter contain ripening follicles. There is evidence of an irregular oestrous cycle. A normal male, known to be fertile, has copulated with several large pygmy females and produced vaginal plugs, but none of the pygmy females showed any signs of pregnancy.

Further experiments are being made with the object of finding the physiological basis for the dwarfing and sterility of pygmy homozygotes.

Summary

1. Undersized animals segregated in a stock of mice derived from MacArthur's selected small mice. These "runts", in MacArthur's terminology, are produced by a recessive gene which has been given the name pygmy, symbol pg, to preserve the usual sense of the word "runt".

2. The gene pygmy is not allelomorphic with Snell's anterior pituitary dwarfism.

3. Pygmies differ phenotypically from Strong's hereditary dwarfism.

4. The growth and adult features of pygmies are described.

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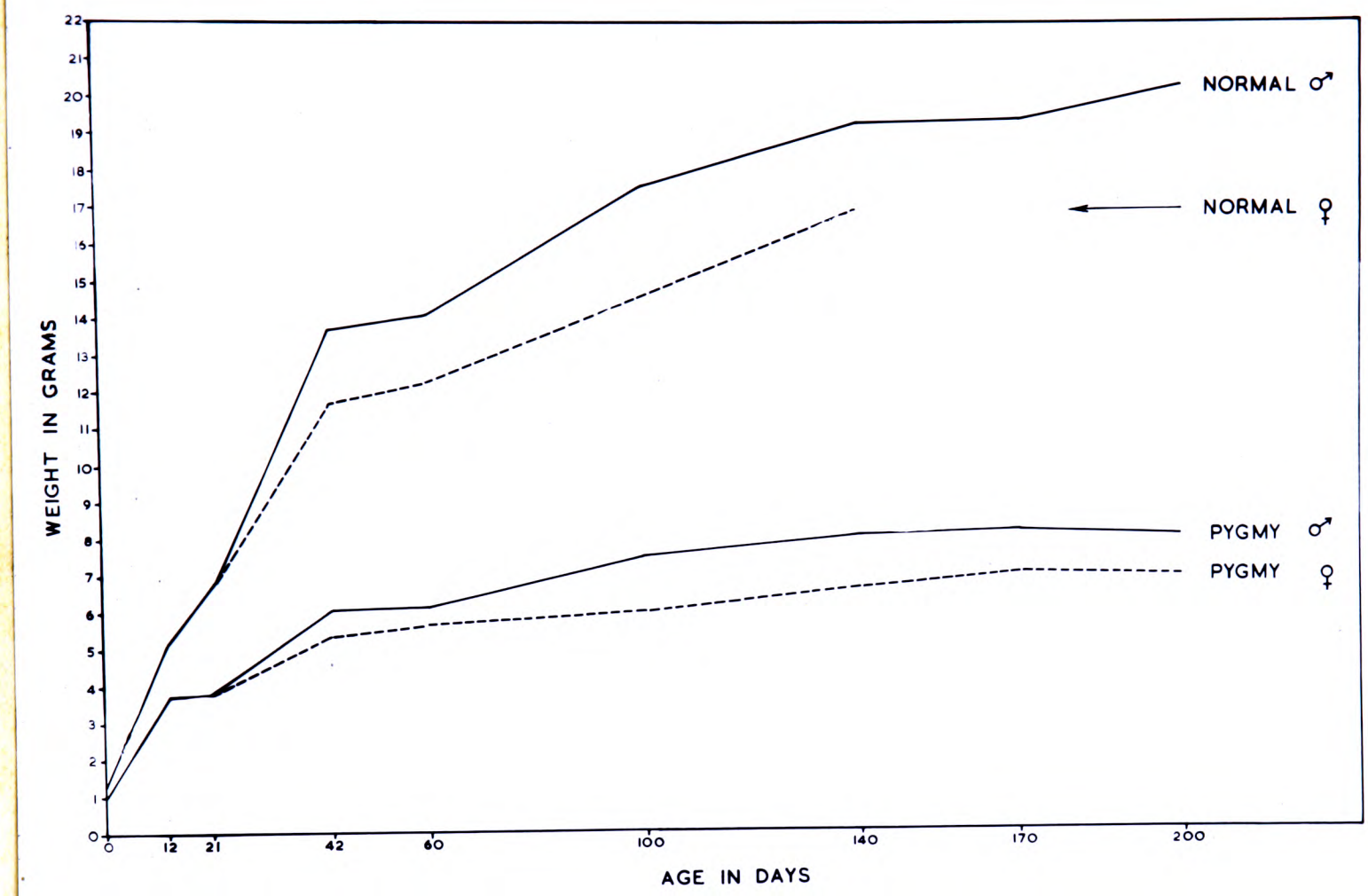
PYGMY AND NORMAL LITTER MATES

Fig.1.

A photograph of dead animals from the Small stock. On the left, pygmies and on the right, normals; above, adults and below 12 days old.



GROWTH CURVES OF PYGMY AND NORMAL MICE.

Fig. 2.

Compound growth curves of pygmies and normals from the Small stock. Weighed at the ages shown on the graph.

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FIGURES AND TABLES

Fig.1

Results of Selection in Goodale's Large Line.

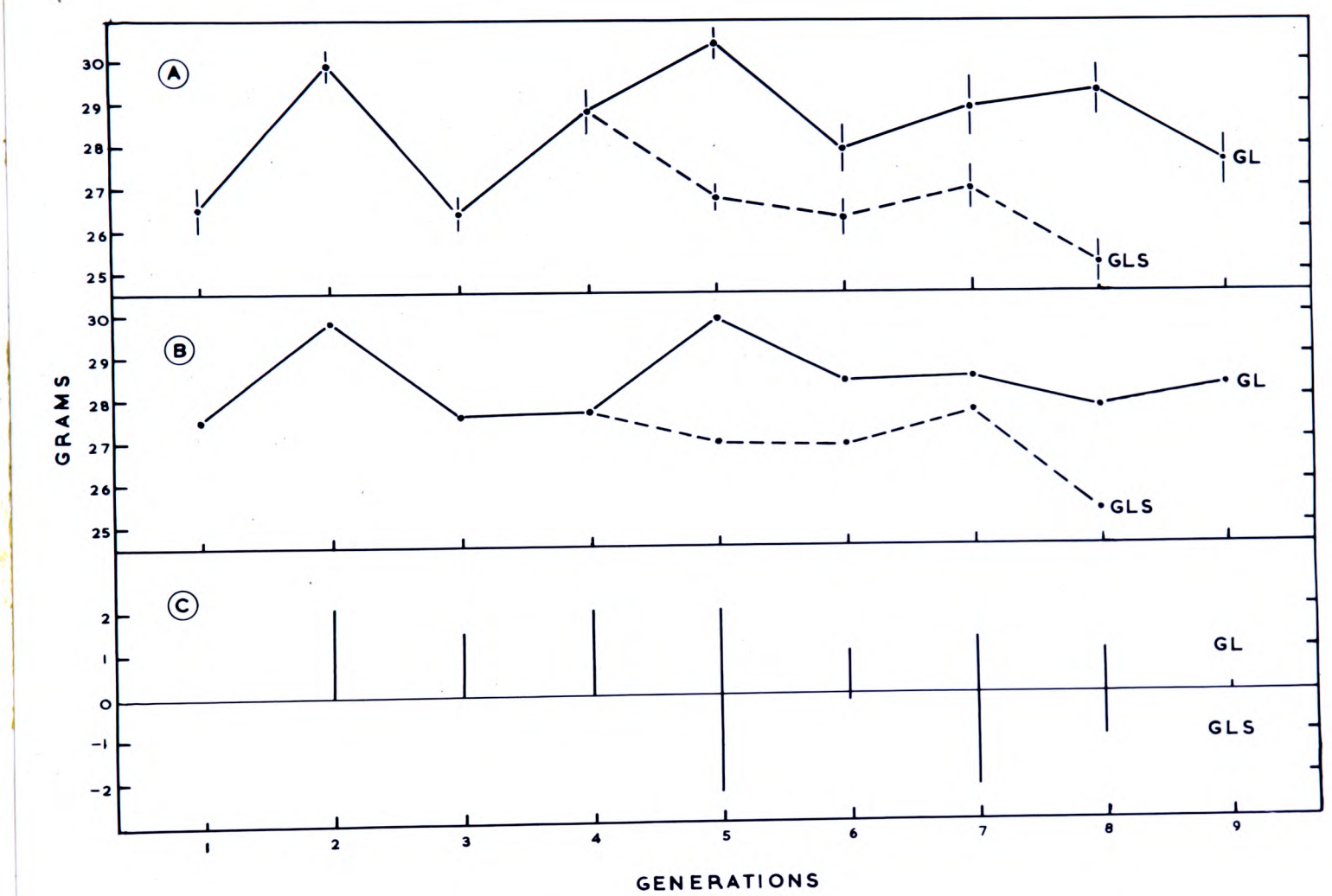


Fig. 1(a) Weighted sex means of 6-week weight in successive generations. The short vertical lines extend one standard error above and below the mean.

1(b) Weighted sex means of 6-week weight corrected for 12-day weight.

1(c) Selection differentials (weighted according to number of offspring) applied in producing each generation.

Fig.2.

Results of Selection in MacArthur's Large Line.

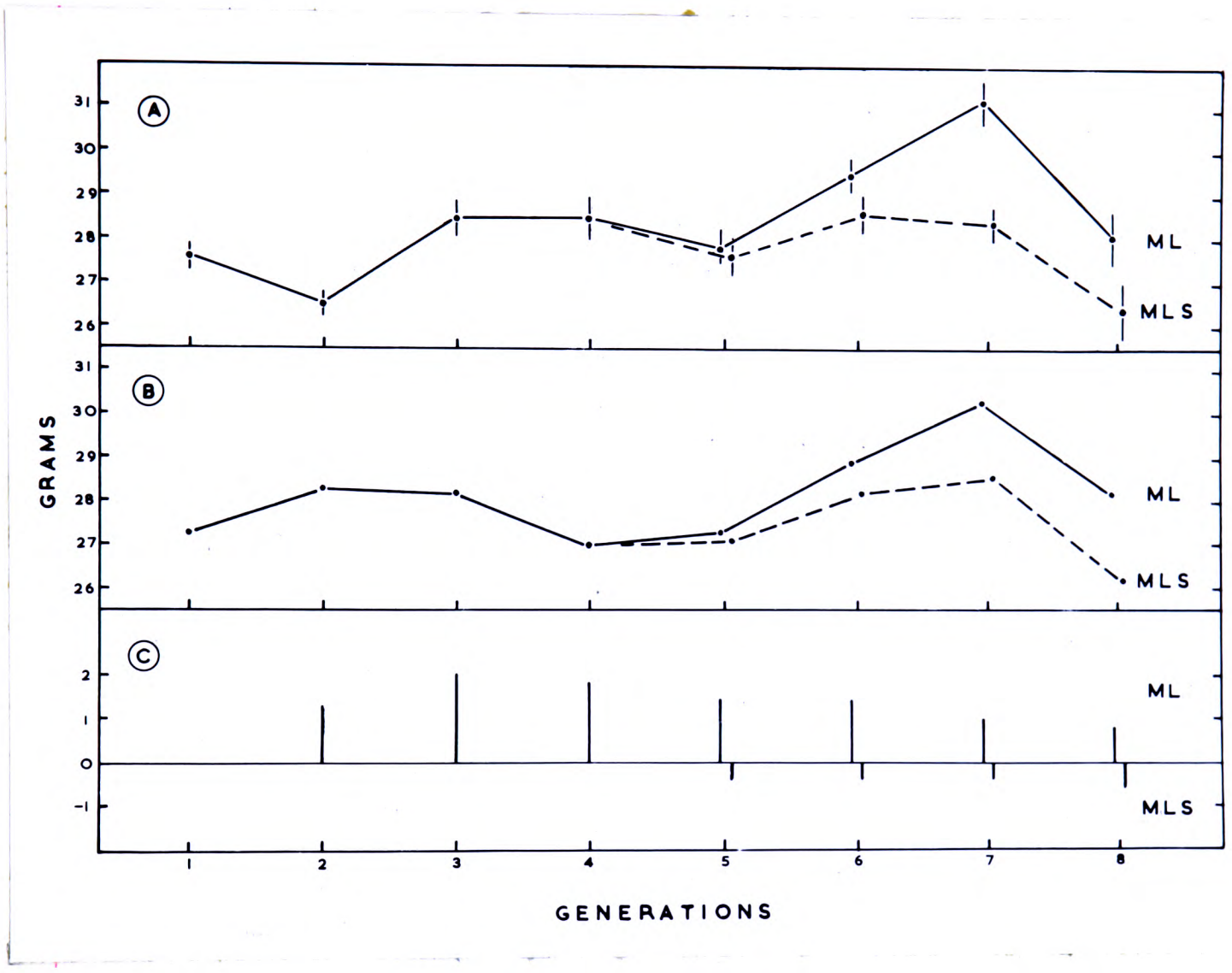


Fig. 2(a) Weighted sex means of 6-week weight in successive generations. The short vertical lines extend one standard error above and below the mean.

2(b) Weighted sex means of 6-week weight corrected for 12-day weight.

2(c) Selection differentials (weighted according to number of offspring) applied in producing each generation.

Fig.3.

Results of Selection of Cross-bred Mice.

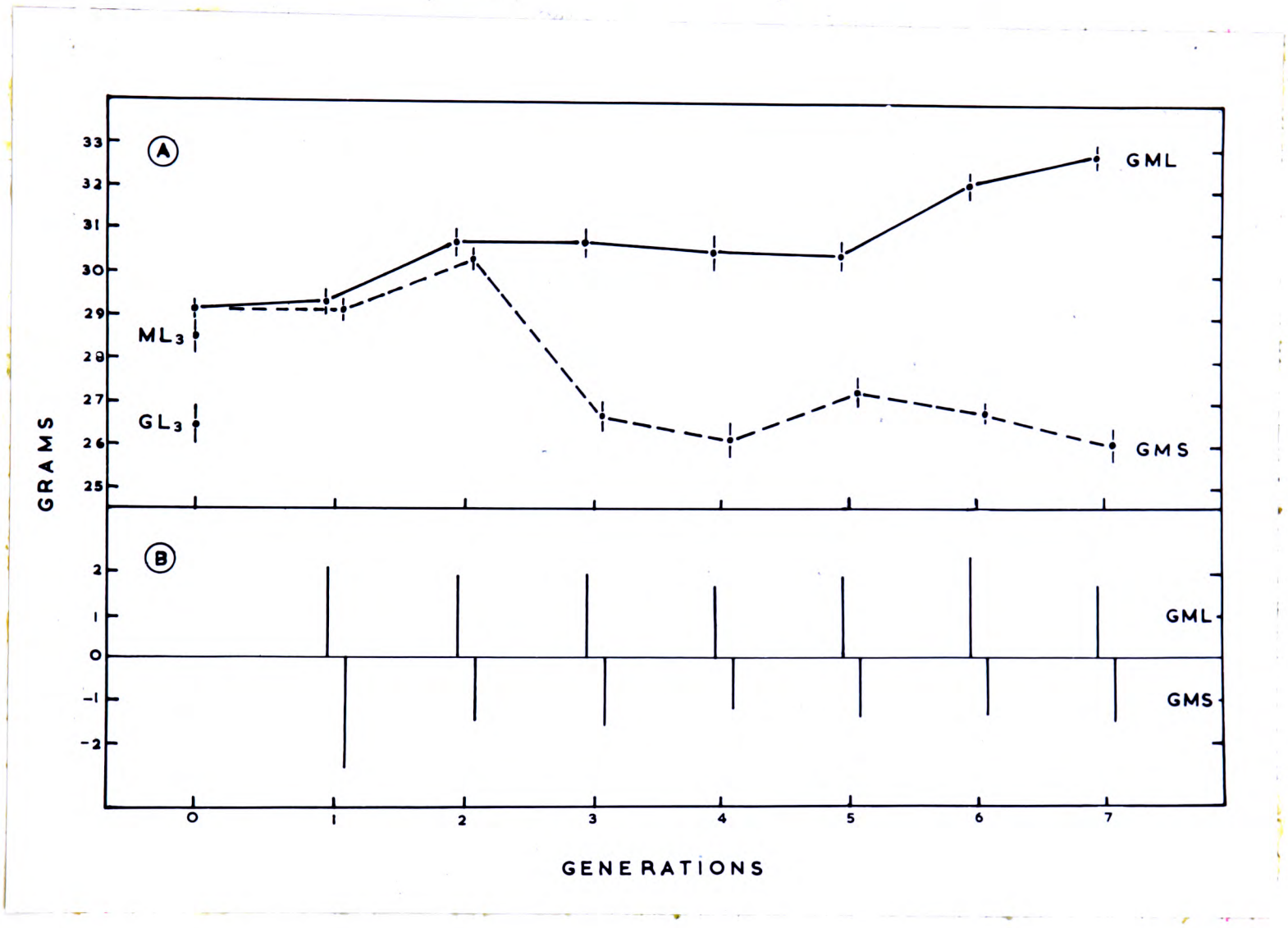


Fig. 3(a) Weighted sex means of 6-week weight in successive generations. The short vertical lines extend one standard error above and below the mean. Means of contemporary parent lines also shown for the first generation.

3(b) Selection differentials (weighted according to number of offspring) applied in producing each generation.

Fig.4.

Divergence in Size in the Cross-bred lines.

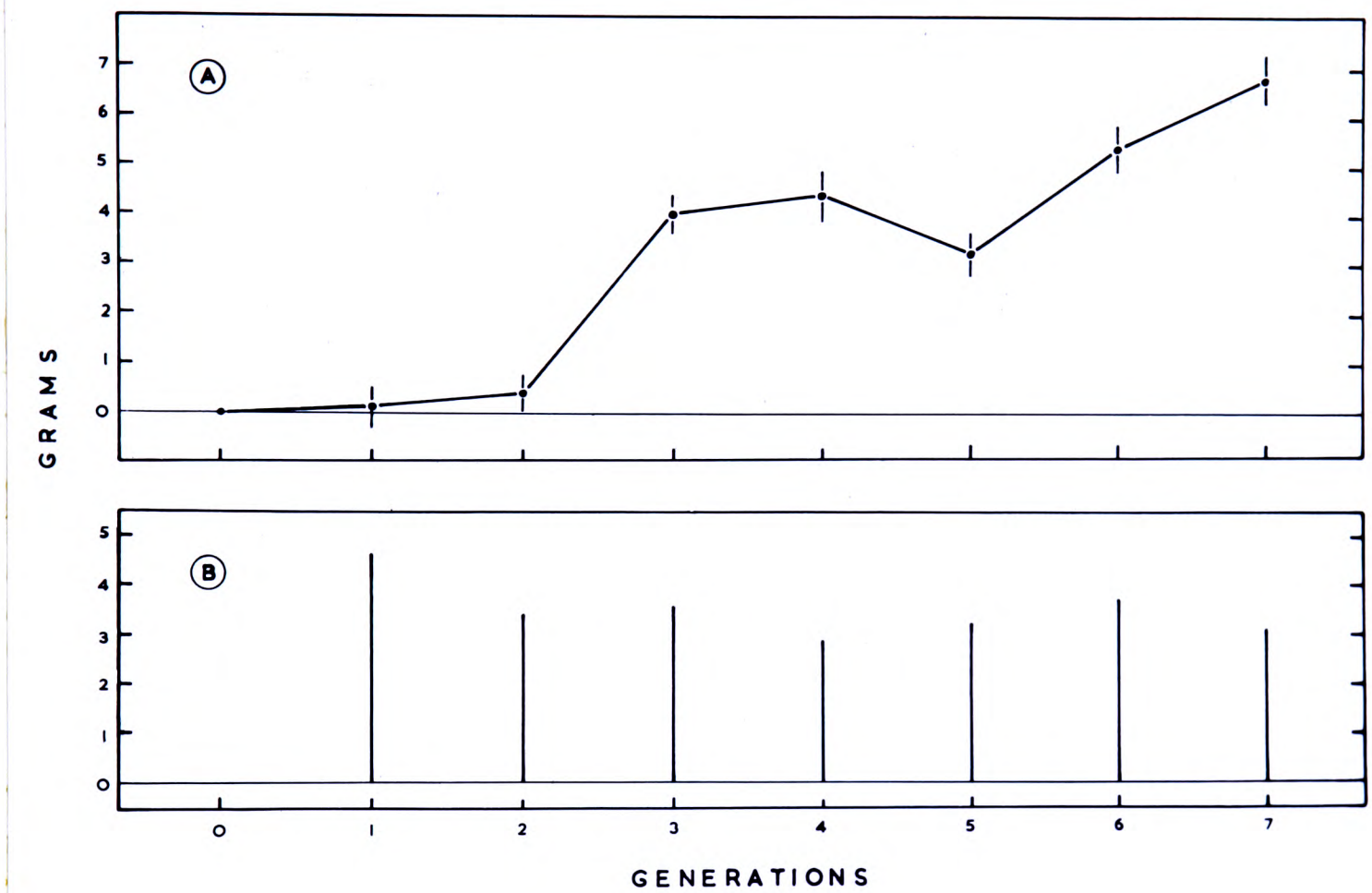
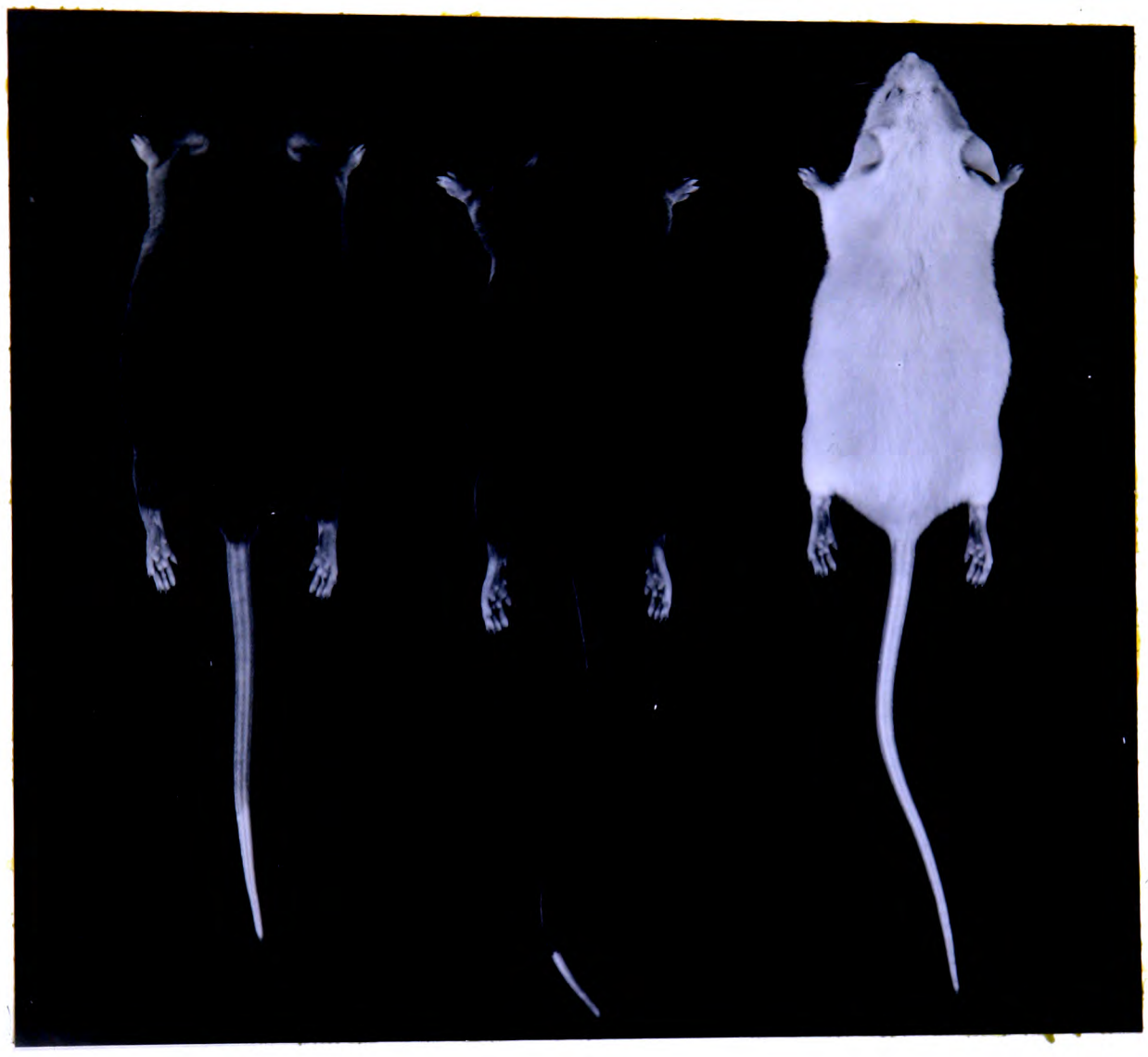


Fig. 4(a) Divergence of 6-week weight between the lines selected for large and small size.

4(b) Selection differentials in the large and small lines summed for each generation.

Fig.5.

Differences in Conformation of the Lines.



A photograph of adult males (dead) to show the different conformation of the parent lines and that of the cross-bred mice. On the left a MacArthur's Large Line male, showing the fatness and relative shortness of the tail in this line; on the right a Goodale's Large Line male, which is much thinner and has a longer tail; in the centre a cross-bred male with a conformation intermediate between that of the parent-lines.

Fig.6.

Correlated Responses to Selection for 6-week Weight in the Cross-bred Mice.

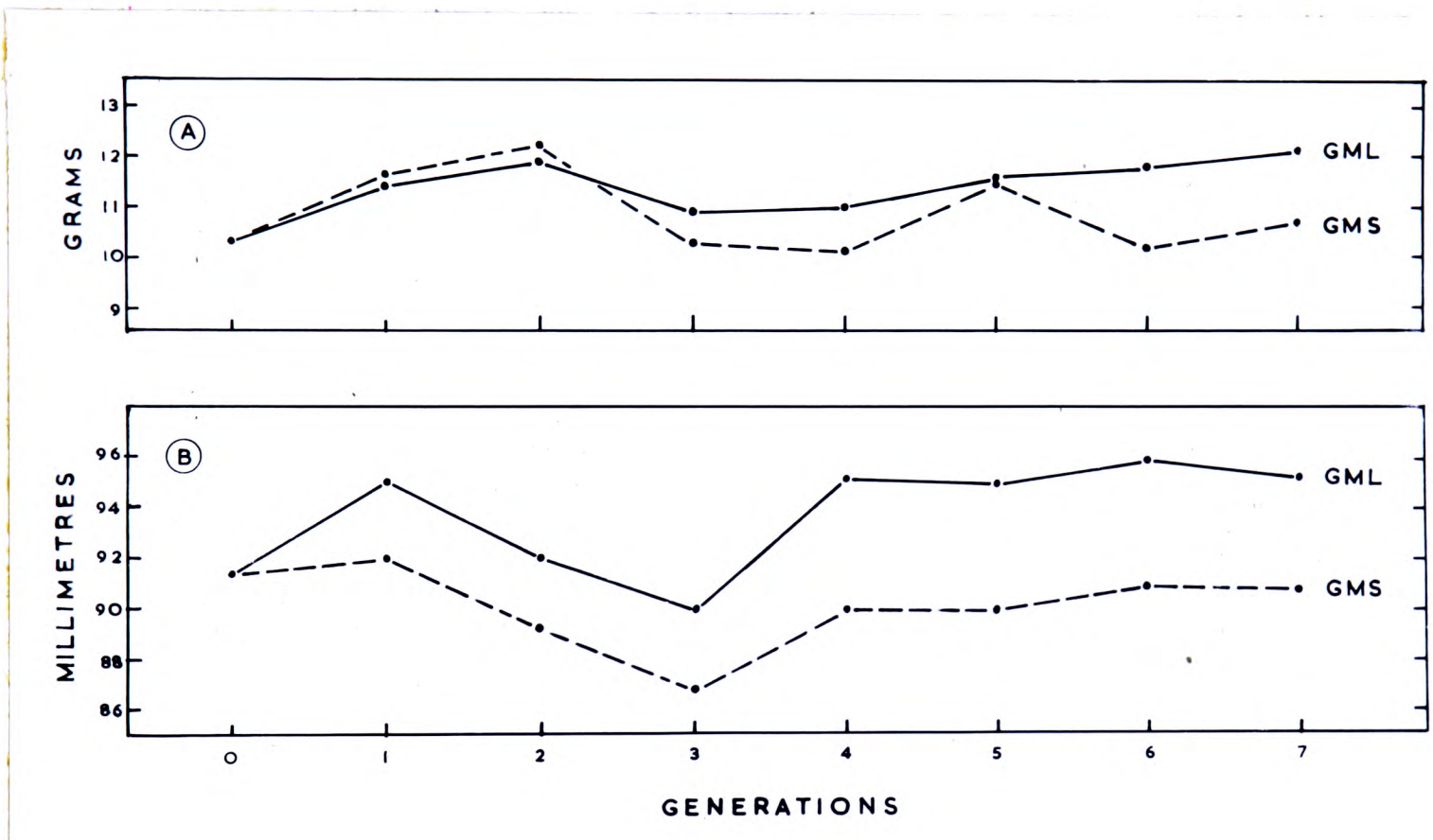


Fig. 6(a) The changes in mean 3-week weight produced by selection for 6-week weight.

6(b) The changes in mean tail length produced by selection for 6-week weight.

Fig.7.

Litter Size of all Lines.

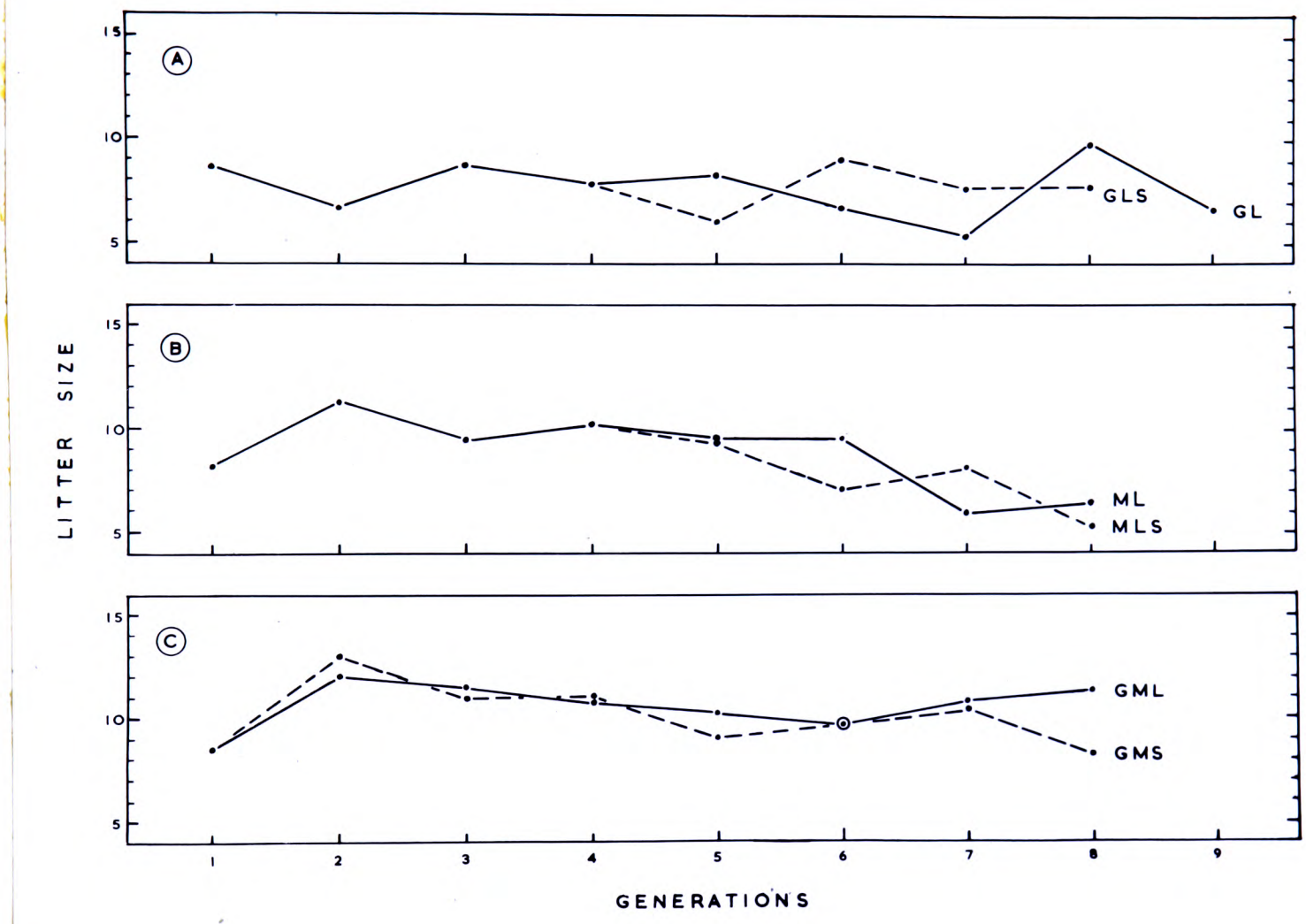


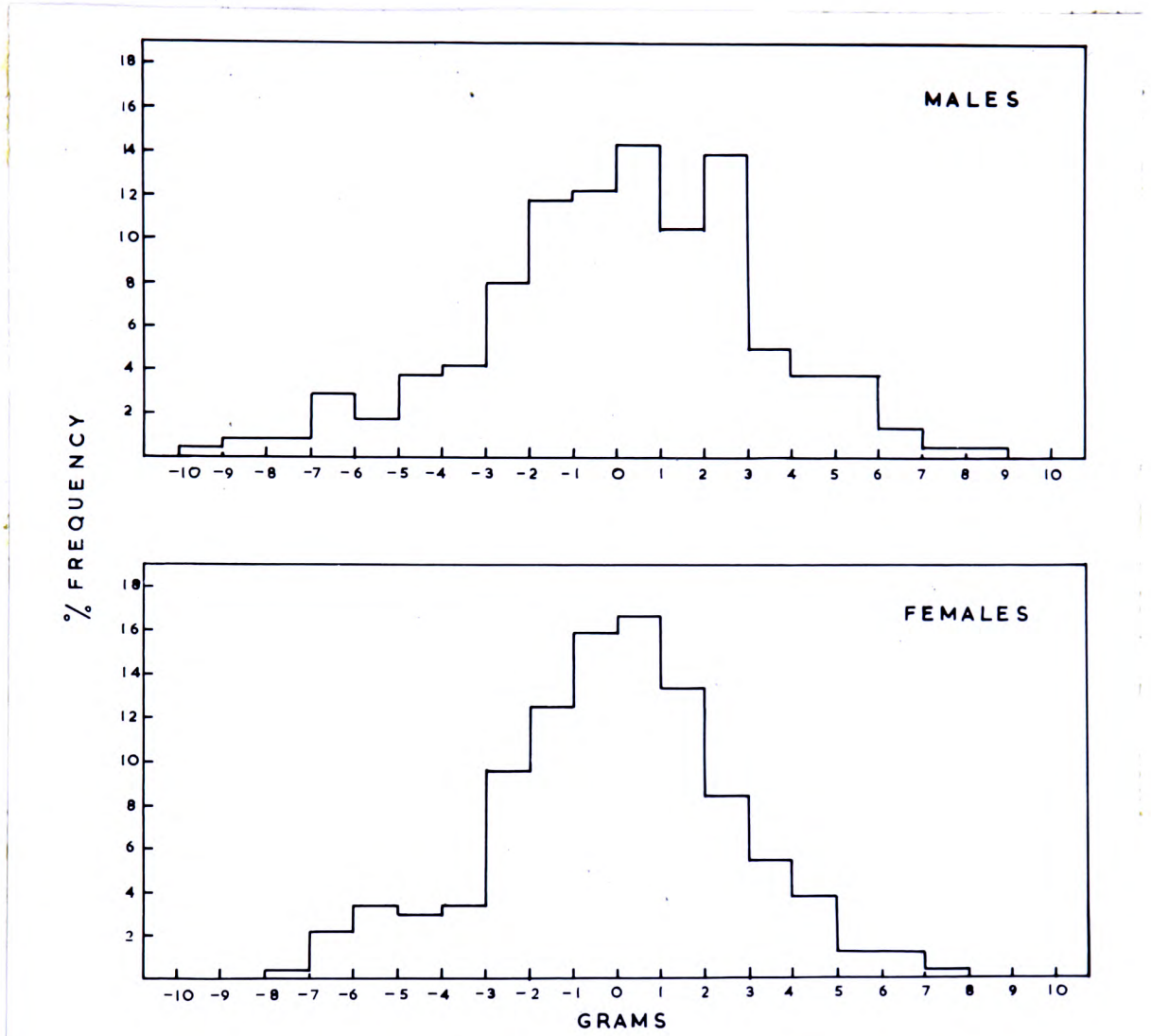
Fig. 7(a) Mean number of living young in the first litters of Goodale's Large Line.

7(b) Mean number of living young in the first litters of MacArthur's Large Line.

7(c) Mean number of living young in the first litters of cross-bred mice.

Fig.8.

Frequency Distribution of 6-week Weights.

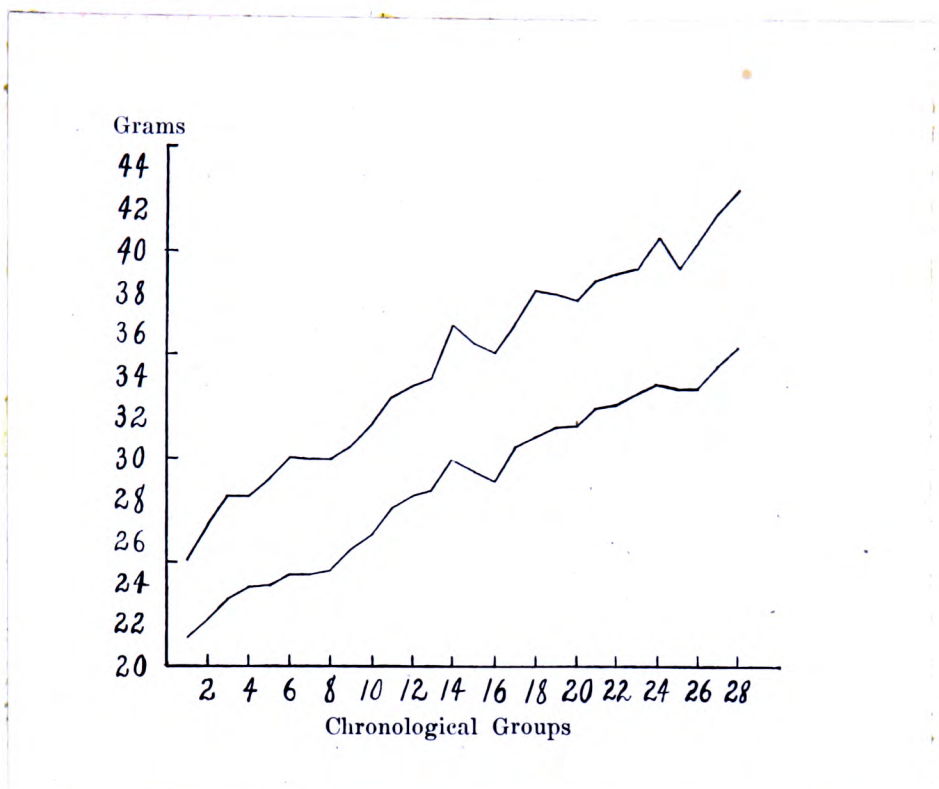


Frequency distributions of 6-week weights of 238 males and 241 females from the GML line. The observed weights in seven generations are pooled by measuring them as deviations from the generation mean.

Fig.9.

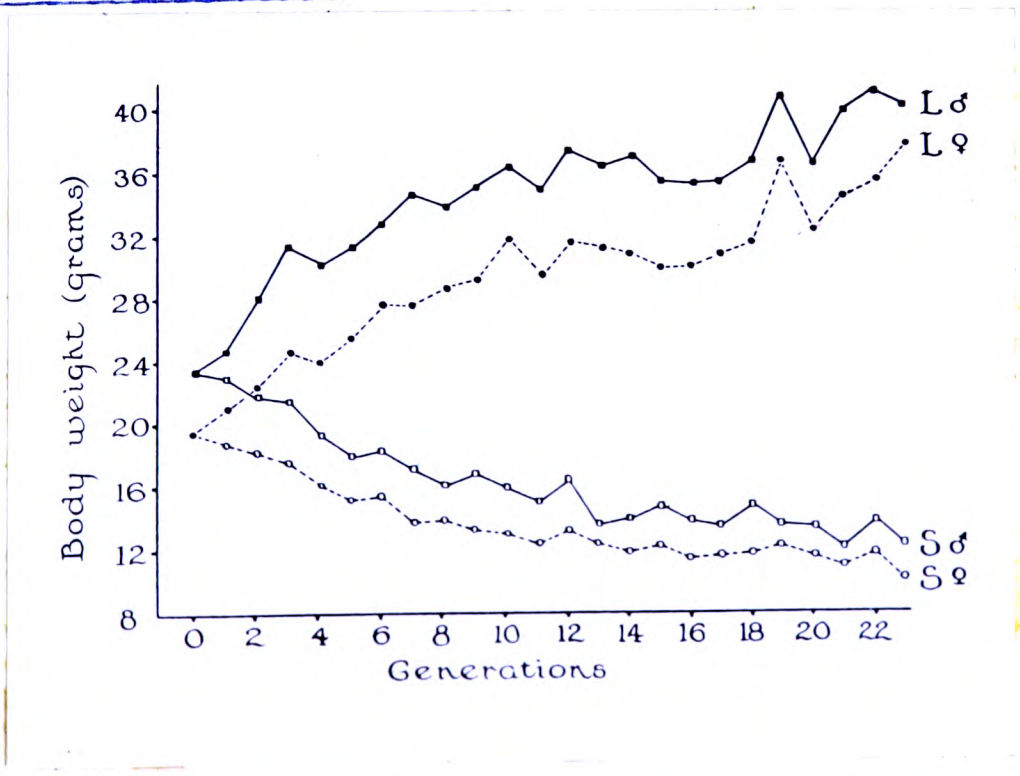
Previous Selection of the Parent Lines.

Goodale's Large Line.



The upper line shows the mean 60-day weight of males, the lower line the mean 60-day weight of females. Males are arranged chronologically in groups of 500 and females grouped correspondingly. The number of generations is approximately the same as the number of groups. (Fig.1 of Goodale, 1941).

MacArthur's Large Line.



The upper lines (L ♂ and L ♀) show the mean 60-day weight of males and females selected for large size. The lower lines (S ♂ and S ♀) show the results of selection for small size. (Fig.1 of MacArthur, 1949).

TABLE I.

Analysis of Variance of 6-week Weight for Goodale's Large Line.

Females

Source of variation	D.F.	S.S.	M.S.	M.S. is an estimate of	Intra-litter corr.
Within generations - Between litters	61	1243.87	20.39	$6^2 + 2.996^2_L$	0.42
Within litters	140	890.92	6.36	6^2	

Males

Source of variation	D.F.	S.S.	M.S.	M.S. is an estimate of	Intra-litter corr.
Within generations - Between litters	61	2982.05	48.89	$6^2 + 3.286^2_L$	0.56
Within litters	160	1496.03	9.35	6^2	

TABLE II

Analysis of Variance of 6-week Weight for MacArthur's Large Line.

Females

Source of variation	D.F.	S.S.	M.S.	M.S. is an estimate of	Intra-litter corr.
Between generation means	7	362.45	-		
Regression	1	207.30	207.30		
Deviations from regression	6	155.15	25.86		
Within generations,					
Between litters	55	1359.50	24.72	$\sigma^2 + 4.286\sigma^2_L$	0.60
Within litters	177	590.33	3.34	σ^2	

Test for significance of regression -

$$F = \frac{207.30}{25.86} = 8.02 \quad P < 0.05$$

Males

Source of variation	D.F.	S.S.	M.S.	M.S. is an estimate of	Intra-litter corr.
Between generation means	7	543.39	-		
Regression	1	175.97	175.97		
Deviations from regression	6	367.42	61.24		
Within generations,					
Between litters	53	2564.90	48.39	$\sigma^2 + 4.406\sigma^2_L$	0.75
Within litters	177	600.96	3.40	σ^2	

Test for significance of regression -

$$F = \frac{175.97}{61.24} = 2.87 \quad P < 0.20$$

TABLE III

Analysis of Variance of 6-week Weight by Generations for Cross-bred Lines.

Generation		Females				Males			
		Between litters		Within litters		Between litters		Within litters	
		D.F.	M.S.	D.F.	M.S.	D.F.	M.S.	D.F.	M.S.
GM_o*		16	16.44	63	2.51	16	34.98	57	4.27
GML	1	7	18.51	29	2.62	7	28.20	32	6.67
	2	9	38.84	21	3.18	9	9.77	32	5.52
	3	10	12.82	32	2.84	10	45.47	32	2.89
	4	7	7.54	24	5.63	7	28.43	20	6.98
	5	8	12.86	26	5.00	6	8.21	16	11.61
	6	7	10.18	21	2.59	7	10.91	24	3.86
	7	7	13.80	20	4.30	7	38.56	23	5.25
Total GML		55	16.92	173	3.69	53	25.18	179	5.71
GMS	1	7	11.44	33	1.76	7	21.49	30	4.27
	2	8	6.82	24	1.97	8	13.69	27	5.29
	3	10	30.56	29	1.34	10	28.69	28	4.24
	4	6	10.40	13	4.10	7	53.81	23	6.39
	5	8	11.52	22	2.84	8	11.79	24	5.12
	6	8	15.91	24	4.02	8	15.48	25	3.98
	7	7	17.00	23	3.04	7	11.78	16	3.67
Total GMS		54	15.57	168	2.54	55	22.32	173	4.73

		Components of variance		Intra-litter corr.
		σ^2	σ^2_L	
GM	females	3.69	3.15	0.46
	males	5.71	4.40	0.44
GMS	females	2.54	3.13	0.55
	males	4.73	4.19	0.47

* Calculated within reciprocal crosses.

TABLE IV

Average Inbreeding Coefficient (F) of Mice in each generation.

(These are calculated relative to the first generations in this laboratory where only sibships were known.)

Goodale's Large Line

Generation	F %	No. of parents
GL 1	0	10
2	0	12
3	2.5	10
4	5.8	14
5	8.6	14
6	12.3	16
7	13.7	12
8	16.1	14
9	19.4	16
GLS 1	10.7	10
2	12.8	16
3	15.7	12
4	17.2	16

MacArthur's Large Line

Generation	F %	No. of parents
ML 1	0	12
2	6.2	12
3	7.4	6
4	7.8	10 *
5	13.2	14
6	14.3	15
7	18.0	12
8	23.3	6
MLS 1	13.1	16
2	14.6	12
3	16.9	18
4	19.2	4

* Includes five also used in the previous generation.

Cross-bred mice

Generation	F %	No. of parents
GM_o	0	32
GML 1	0	16
2	0	20
3	0.3	22
4	2.5	16
5	3.7	18
6	5.4	16
7	7.5	16
GMS 1	0	12
2	0	18
3	0.6	22
4	2.4	16
5	4.1	16
6	5.7	18
7	7.4	16

TABLE V

Results of Selection in Goodale's Large Line (all measurements in grams)

a) Selection for large size.

Generation GL		1	2	3	4	5	6	7	8	9	Total
Females	Mean 6-week wt. Number	23.82 20	27.47 46	24.71 31	26.11 31	27.97 18	25.60 24	27.17 9	27.51 18	26.01 13	26.24 210
Males	Mean 6-week wt. Number	29.11 30	32.46 34	27.96 30	31.54 29	32.84 26	30.20 17	30.78 14	31.13 24	29.05 26	30.57 230
Weighted mean of sexes		26.53	29.43	26.37	28.77	30.40	27.87	28.90	29.28	27.36	28.40
Weighted mean corrected for 12-day weight		27.52	29.81	27.60	27.74	29.89	28.39	28.51	27.78	28.33	-
Standard error of mean		0.52	0.35	0.37	0.51	0.35	0.57	0.69	0.63	0.63	-
Selection differential		-	2.06	1.50	1.99	2.01	1.02	1.30	1.03	0.12	-

b) Selection for small size.

Generation GLS		1	2	3	4	Total
Females	Mean 6-week wt. Number	24.21 16	24.02 30	24.73 19	24.04 19	24.22 84
Males	Mean 6-week wt. Number	29.56 14	28.60 26	29.21 18	26.41 22	28.30 80
Weighted mean of sexes		26.80	26.27	26.95	25.22	26.26
Weighted mean corrected for 12-day weight		27.04	26.90	27.74	25.38	-
Standard errors of mean		0.33	0.39	0.49	0.51	-
Selection differential		2.28	0.14	2.26	1.12	-

TABLE VI

Results of Selection in MacArthur's Large Line (all measurements in grams).

a) Selection for large size.

Generation ML		1	2	3	4	5	6	7	8	Total
Females	Mean 6-week wt. Number	25.89 58	25.00 47	26.74 18	27.65 24	25.95 25	27.86 29	29.16 18	27.37 21	26.57 240
Males	Mean 6-week wt. Number	29.40 47	27.99 62	30.24 19	29.28 24	29.82 23	31.11 27	33.30 21	28.41 15	29.60 238
Weighted mean of sexes		27.61	26.49	28.44	28.51	27.84	29.48	31.23	28.13	28.09
Weighted mean corrected for 12-day weight		27.29	28.27	28.21	27.03	27.34	29.42	30.29	28.17	-
Standard error of mean		0.30	0.24	0.37	0.48	0.47	0.41	0.48	0.65	-
Selection differential		-	1.24	2.00	1.84	1.44	1.45	0.95	0.78	-

b) Selection for small size.

Generation MLS		1	2	3	4	Total
Females	Mean 6-week wt. Number	26.10 38	27.23 18	26.43 29	25.66 5	26.41 90
Males	Mean 6-week wt. Number	29.14 31	29.73 13	30.31 33	26.50 3	29.62 80
Weighted mean of sexes		27.62	28.56	28.37	26.41	28.01
Weighted mean corrected for 12-day weight		27.62	28.16	28.63	26.17	-
Standard error of mean		0.37	0.40	0.41	0.66	-
Selection differential		1.17	1.34	0.25	2.57	-

TABLE VII

Analysis of variance of 6-week weight of GM_o mice (Reciprocal crosses analysed separately)

		Sources of Variation	D.F.	S.S.	M.S.	M.S. in an Estimate of	Components of variance	Intra-litter Corr.
GL females x ML males	Female Offspring	Sires	4	46.96	11.74	$\sigma^2 + 4.45\sigma^2_L + 7.98\sigma^2_S$	$\sigma^2_S = -0.42$	
		Litters	4	60.45	15.11***	$\sigma^2 + 4.45\sigma^2_L$	$\sigma^2_L = 2.94$	0.59
		Individuals	32	65.32	2.04	σ^2	$\sigma^2 = 2.04$	
	Male Offspring	Sires	4	178.24	44.56	$\sigma^2 + 3.46\sigma^2_L + 6.29\sigma^2_S$	$\sigma^2_S = 1.42$	
		Litters	4	142.99	35.75**	$\sigma^2 + 3.46\sigma^2_L$	$\sigma^2_L = 8.71$	0.61
		Individuals	23	129.49	5.63	σ^2	$\sigma^2 = 5.63$	
ML females x GL males	Female Offspring	Sires	5	85.56	17.11	$\sigma^2 + 4.38\sigma^2_L + 6.47\sigma^2_S$	$\sigma^2_S = -0.96$	
		Litters	3	70.02	23.34***	$\sigma^2 + 4.38\sigma^2_L$	$\sigma^2_L = 4.64$	0.61
		Individuals	31	93.09	3.00	σ^2	$\sigma^2 = 3.00$	
	Male Offspring	Sires	5	128.88	25.78	$\sigma^2 + 4.71\sigma^2_L + 7.00\sigma^2_S$	$\sigma^2_S = -1.53$	
		Litters	3	109.56	36.52***	$\sigma^2 + 4.71\sigma^2_L$	$\sigma^2_L = 6.85$	0.67
		Individuals	34	114.00	3.35	σ^2	$\sigma^2 = 3.35$	

** Significant at 1% level of probability

*** Significant at 0.1% level of probability.

TABLE VIII

Results of Selection in Cross-bred Lines (all measurements in grams).

a) Selection for large size.

Generation		GM_o	GML_1	2	3	4	5	6	7	GML Total
Females	Mean 6-week wt.	26.57	27.26	27.94	27.58	27.85	28.65	30.14	30.18	28.40
	Number	81	37	31	43	32	35	29	28	235
Males	Mean 6-week wt.	31.77	31.37	33.54	34.01	33.20	31.79	34.09	35.47	33.39
	Number	75	40	42	43	28	23	32	31	239
Weighted mean of sexes		29.14	29.32	30.75	30.73	30.53	30.47	32.13	32.82	30.89
Standard error of mean		0.20	0.29	0.33	0.28	0.39	0.29	0.30	0.29	-
Selection differential		-	2.08	1.95	1.97	1.70	1.88	2.39	1.63	-

b) Selection for small size.

Generation		GMS_1	2	3	4	5	6	7	GMS Total
Females	Mean 6-week wt.	26.41	27.43	24.94	24.85	24.89	24.67	23.56	25.32
	Number	41	33	40	20	31	33	31	229
Males	Mean 6-week wt.	32.02	33.25	28.34	25.57	29.62	28.83	28.59	29.88
	Number	38	36	39	31	33	34	24	235
Weighted mean of sexes		29.16	30.36	26.68	26.09	27.24	26.75	25.99	27.59
Standard error of mean		0.23	0.24	0.31	0.38	0.33	0.24	0.37	-
Selection differential		2.59	1.47	1.63	1.22	1.37	1.35	1.51	-

TABLE IX

Regressions of offspring on parent (calculated within generations of parent and offspring).

Variable		D.F.	Regression of offspring on			Correlation between parents
			Dam	Sire	Mean of parents	
6-week Weight						
GML line	♀ offspring	233	0.13 ± 0.07	0.27 ± 0.07	0.47 ± 0.09	- 0.12
	♂ offspring	235	0.23 ± 0.10	0.18 ± 0.09	0.44 ± 0.14	- 0.08
GMS line	♀ offspring	181	0.17 ± 0.10	0.10 ± 0.08	0.21 ± 0.11	0.15
	♂ offspring	190	0.20 ± 0.11	0.01 ± 0.08	0.06 ± 0.12	0.12
Tail Length						
GML line	♀ offspring	233	0.31 ± 0.07	0.20 ± 0.05	0.58 ± 0.08	- 0.02
	♂ offspring	235	0.37 ± 0.07	0.14 ± 0.06	0.46± 0.09	0.02
GMS line	♀ offspring	176	0.10 ± 0.09	0.34 ± 0.07	0.44 ± 0.11	0.00
	♂ offspring	185	0.27 ± 0.09	0.06 ± 0.08	0.26 ± 0.11	0.06
3-week Weight						
GML line: all offspring		483	-0.05 ± 0.05	-0.04 ± 0.05	-0.10 ± 0.07	- 0.15
GMS line: all offspring		371	0.15 ± 0.06	0.26 ± 0.05	0.43 ± 0.08	- 0.01

TABLE X

Heritability estimates from regression of offspring on parents.
(Estimates from male and female offspring combined)

		Heritability estimated from regression of offspring on		
		Dam	Sire	Mean of parents.
6-week wt.	GML line	0.40 ± 0.16	0.50 ± 0.16	0.45 ± 0.10
	GMS line	0.33 ± 0.16	0.09 ± 0.12	0.11 ± 0.10
Tail length	GML line	0.68 ± 0.12	0.35 ± 0.10	0.52 ± 0.08
	GMS line	0.39 ± 0.16	0.41 ± 0.14	0.35 ± 0.10
3-week wt.	GML line	-0.12 ± 0.12	- 0.09 ± 0.12	- 0.10 ± 0.07
	GMS line	0.30 ± 0.12	0.52 ± 0.10	0.43 ± 0.08

TABLE XI

Estimated gene-frequency of colour genes in subsequent generations of the cross-bred lines. (Frequency as a percentage.)

	GENERATION							
	GM_o	GML						
		1	2	3	4	5	6	7
Non-agouti	50	53	30	61	48	32	66	64
Brown	50	38	23	29	39	35	45	43
Albino	65	55	51	55	49	53	63	62
Leaden	47	41	40	29	25	55	59	64

	GENERATION							
	GM_o	GMS						
		1	2	3	4	5	6	7
Non-agouti	50	40	53	13	15	25	21	34
Brown	50	40	53	69	65	59	46	41
Albino	65	61	56	53	54	54	56	50
Leaden	47	47	47	71	58	70	53	79

TABLE XII.

The weighted mean differences in 6-week weight associated with colour genes calculated within litters for all cross-bred mice.

	Wt. of Mutant Phenotype - Wt. of Wild-type			
	in grams		% of mean wt.	
	Females	Males	Females	Males
Non-agouti	-0.05 ± 0.33	0.49 ± 0.43	-0.18 ± 1.25	1.55 ± 1.35
Brown	0.31 ± 0.30	-0.24 ± 0.42	1.15 ± 1.11	-0.76 ± 1.33
Albino	0.26 ± 0.24	-0.07 ± 0.31	0.90 ± 0.89	-0.23 ± 0.98
Leaden	0.39 ± 0.32	-0.30 ± 0.40	1.46 ± 1.19	-0.94 ± 1.27

TABLE XIII

Data for the Calculation of the Selection Index.

		Females	Males
σ_{W6}	=	2.666 gm^2 *	3.175 gm^2
σ_T	=	3.795 mm^2	4.231 mm^2
r_{W6W3}	=	0.545	0.502
r_{W6T}	=	0.476	0.475
r_{W3T}	=	0.440	0.551

Sexes Combined

σ_{W3}	=	1.703 gm^2
h^2_{W6}	=	0.432
h^2_T	=	0.439
h^2_{W3}	=	0
$r_{G_{W6}.G_T}$	=	0.227

* The figures given in this table are based on mice of the GML stock and include in addition to the selection line some mice of this stock used for other purposes. For this reason these figures do not agree exactly with those which may be calculated from Table IV.

TABLE XIV

Mean 6-week Weights of all Lines (in order of magnitude) and Intra-Litter Variances.

	Mean (gm.)	Intra-litter variance (gm^2)
GML males	33.39	5.71
GL "	30.57	9.35
GMS "	29.88	4.73
ML "	29.60	3.40
GML females	28.40	3.69
ML "	26.57	3.34
GL "	26.24	6.36
GMS "	25.32	2.58

TABLE XV

Differences between Sex Means

Line	6-week weight ♂ mean gm	6-week weight ♀ mean gm	Difference in gm	Difference in $logs_{10}$
GML	33.39	28.40	4.99	.070
GMS	29.98	25.32	4.56	.072
GL	30.57	26.24	4.33	.066
ML	29.60	26.57	3.03	.047
CBA inbred	20.97	17.94	3.03	.068
A inbred	19.61	17.03	2.58	.061
C57 inbred	18.62	16.15	2.47	.066
Small stock	13.27	11.39	1.88	.066

TABLE XVI

Differences between Normals and Pygmies.

	Mean 6-week weight.		Difference in	
	Normal	Pygmy	gm	$\log_{10}$
Large stock - males	25.8	12.3	13.5	.322
females	22.2	11.2	11.0	.297
Small stock - males	13.7	6.1	7.6	.351
females	11.7	5.4	6.3	.336

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