SELECTION FOR AN INVARIANT CHARACTER, VIBRISSA NUMBER, IN THE HOUSE MOUSE

II. LIMITS TO VARIABILITY*

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Summary

A selection experiment for vibrissa number in tabby mice has been extended for 13 generations. Progress of selection for the first seven generations has already been reported. The results of the next six generations of selection are given and the whole experiment is considered with regard to (1) the shapes of the frequency distributions for the three genotypes (+, *Ta*+, and *Ta*·) for succeeding generations; (2) limits to phenotypic variation, i.e. canalization mechanisms; and (3) reproductive fitness.

I. INTRODUCTION

It is difficult to explain the absence of phenotypic variability on any basis other than genetic fixation, yet various workers have shown that lack of phenotypic variability does not necessarily signify an absence of genetic variability. Goldschmidt (1935), Landauer (1958), and others have found that the frequency and type of phenocopies produced by specific environmental shocks vary between different strains, which are, in the absence of such treatment, indistinguishable. Waddington (1953) extended their observations, by selecting on such exposed variability, showing that selection can, if maintained long enough, produce phenotypic variability in the absence of the variability-inducing treatment. Dun and Fraser (1959) have shown that a mutant gene, tabby, has a similar unmasking effect on genes modifying the numbers of facial vibrissae in the house mouse. This character is not completely invariant in wild-type mice (see Dun 1959) but it exhibits a very small amount of variability and is probably as invariable as most characters subjected to a sufficiently close and exhaustive examination. The tabby gene, a sex-linked partial dominant, causes, in addition to a wide range of other effects, a reduction of the number of vibrissae, and a marked increase of their variability. In the presence of the tabby gene, the number of vibrissae appears to be controlled by a simple polygenic system and has a heritability of the order of 40 per cent. (Dun and Fraser 1958). A selection experiment in which the *Ta* gene is kept segregating has been in progress for 13 generations. In this experiment selection has been practised on the vibrissa number of tabby segregants for both increased and decreased number of vibrissae. The + segregants have been examined to determine whether such selection can cause a breakdown of the invariability of vibrissa number in + mice. Dun and Fraser (1959) have reported on the first seven generations of the experiment. They

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showed that such a breakdown of the invariability of vibrissa number in normal mice had occurred. Their selection experiment has been maintained on the same basis for a further six generations. Data from these are given below and discussed with reference to (1) the breakdown of the invariance of normal mice; (2) the occurrence of limits to phenotypic variability; and (3) the existence of correlated effects on reproductive fitness.

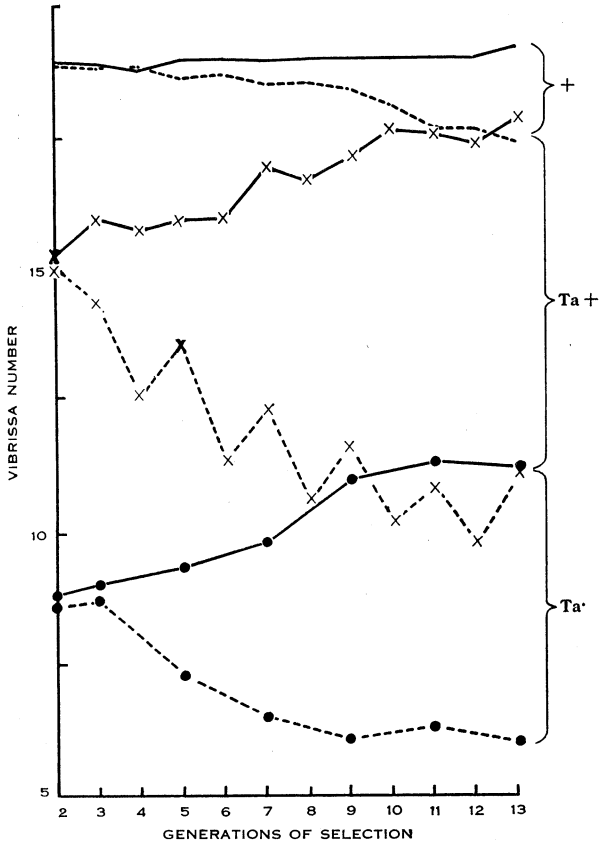

Fig. 1.—Mean vibrissa number at each generation of selection for the three basic genotypes. —— High selection line; - - - - low selection line.

II. Material and Methods

These are described in the introductory paper (Dun and Fraser 1959) except for the introduction of a new standard of scoring the postorbital vibrissae in tabby males. These vibrissae are absent in the majority of tabby males; when present they often occur as short, curled, vestigial hairs. Dun considered that such hairs were not true postorbital vibrissae and, therefore, always scored tabby males as lacking the postorbital vibrissae. We now consider that this decision was artificial and have scored all such fibres as true postorbital vibrissae.

An apparent anomaly in the number of generations of selection requires explanation. The experiment has been in progress for 13 generations, yet only the last 11 generations are described and discussed. The reason for this, namely a change in the basis of scoring vibrissa number, which was instituted at the third generation, is discussed in the introductory papers (cf. Dun 1959; Dun and Fraser 1959).

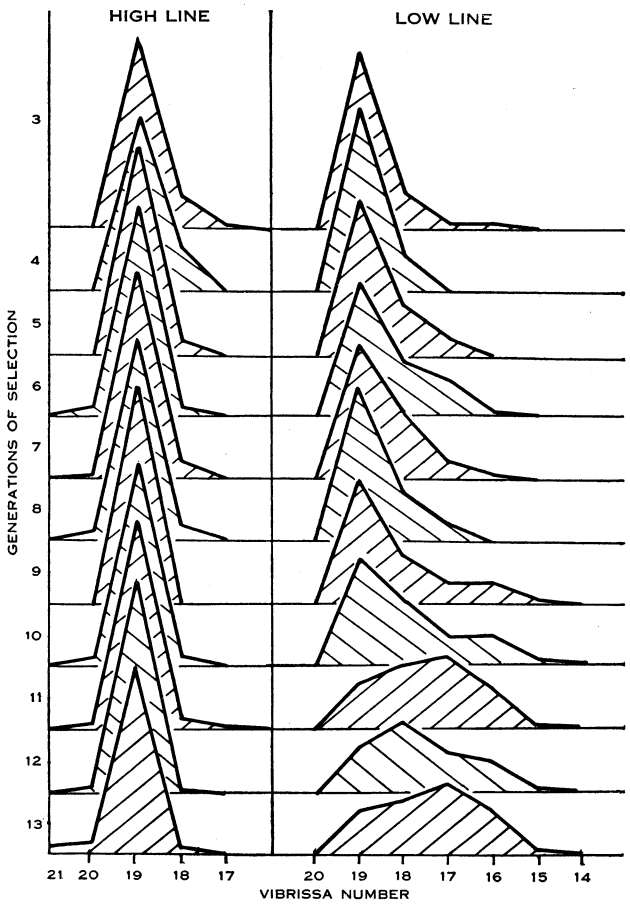

Fig. 2.—Frequency distributions of vibrissa number in + mice of the high and low selection lines.

III. Results

(a) *Effects of Selection on Vibrissa Number*

The segregation of the *Ta* gene produces three main genotypes: + males and females, *Ta*+ (♀), and *Ta*· (♂). The mating scheme is such that *TaTa* females are not produced. Since the tabby gene is a sex-linked partial dominant, this results in three levels of vibrissa number: the + level (*c.* 19 vibrissae in unselected mice), the *Ta*+ level (*c.* 15 vibrissae in unselected mice), and the *Ta*· level (*c.* 8–9 vibrissae in

unselected mice). The mean vibrissa numbers in the high and low selection lines (HST and LST) are shown plotted against generation of selection in Figure 1.

At the *Ta*+ level of expression, changes of vibrissa number found in the first phase of the experiment (generations 0–7) have continued in the second phase (generations 8–13). There are no indications of a decrease of the response to selection, except in the last two generations of the low selection line, and these are not sufficient to be more than a suggestion. There is a marked alternation of high and low response

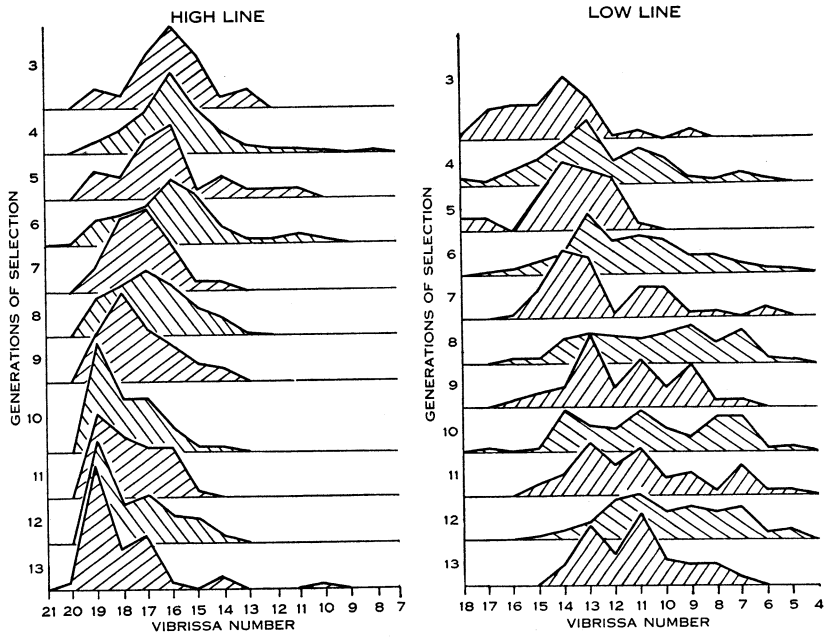

Fig. 3.—Frequency distributions of vibrissa number in *Ta*+ mice, illustrating the shift of the mode of the distribution towards high vibrissa numbers which occurs in the high selection line as this approaches the "normal" vibrissa number, and the absence of any change in the form of this distribution in the low selection line.

to selection in the low selection line. This is almost certainly caused by the mating system; female parents are ++ one generation, *Ta*+ the next. This indicates that a maternal effect is operating since *Ta*+ mice produced from *Ta*+ dams have higher vibrissa numbers than those produced from ++ dams.

At the *Ta*· level of expression, the response to selection has continued in the high line, but there is a definite decrease in the response to selection in the low line. This may be due to a threshold to vibrissa number at 5, which will be discussed below.

The + level of expression shows a definite divergence between the high and low lines, which is concluded to be a consequence of the differences of the underlying genotype determining vibrissa number which have been produced by the selection practised at the tabby levels of expression. The low line showed an initially slow response which has markedly increased over the last four generations. Response in the

high line has been slow, but mice have occurred with extra vibrissae at frequencies above those found in unselected normal mice. In the last generation, + mice with extra vibrissae have been frequent. The frequency distributions of vibrissa number of + mice in the high and low selection lines are shown in Figure 2.

These distributions show (1) that the distribution of vibrissa number in the LST line has changed markedly from the low variance, high-peaked distribution of the unselected distribution to an almost normal distribution; and (2) that no such change has occurred in the high line. In the high line the only noticeable effect is the formation of a longer "tail" to the higher number.

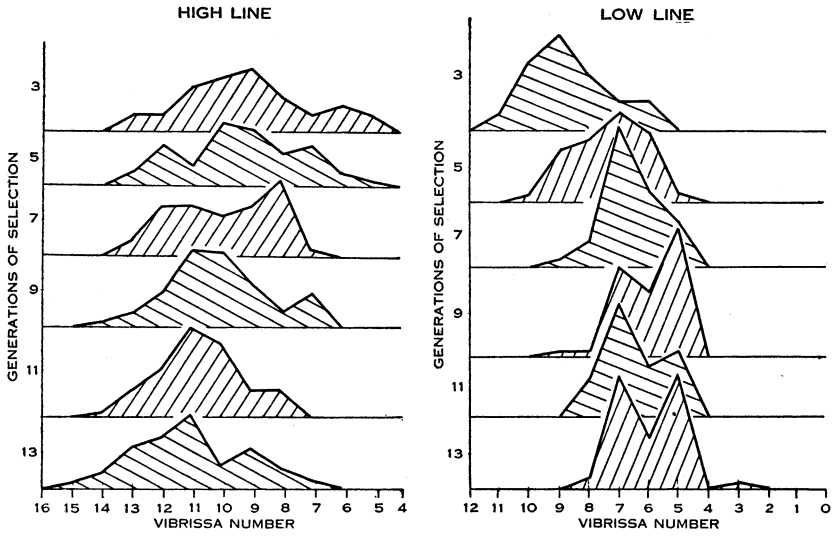

Fig. 4.—Frequency distributions of vibrissa numbers plotted against generation of selection for the *Ta·* level of expression.

The difference between the responses of the two selection lines at the + level of expression can be due:

(1) To the different rates of response which have occurred at the *Ta*+ level of expression. At this level selection for a decreased number of vibrissae has produced a greater response than selection for an increased number of vibrissae.

(2) To a stronger canalization of normal vibrissa number at the higher range than the lower. The canalization of normal vibrissa number could be weaker for + mice with 18–19 vibrissae, than for + mice with 19–20 vibrissae.

(3) To the underlying genotype determining vibrissa number being located closer to the lower than the higher edge of the canalization zone. The mean value of the underlying genotype could have a value closer to 18 than to 19 vibrissae. This latter possibility is illustrated in Figure 7.

(*b*) *Limits to Phenotypic Variability*

The existence of a zone of canalization at 18–19 vibrissae is indicated firstly by the invariance of + mice, and secondly, by the different rates of response to selection at the + and *Ta* levels of expression. Dun and Fraser (1959) suggested that the tabby gene acts on the canalizing mechanism reducing its limitation of phenotypic variability. On this hypothesis we would not expect any limitation of phenotypic variability to occur in *Ta*+ mice whose vibrissa number tends towards the normal number. The frequency distributions of vibrissa number of *Ta*+ mice show a definite shift of the mode of the frequency distribution of vibrissa number towards the high vibrissa numbers in the high selection line as this approaches the "normal" vibrissa number of 18–19. This is illustrated on the left of Figure 3. The analogous data from *Ta*+ mice of the low selection line (Fig. 3, right) shows that no similar shift of the mode occurs.

TABLE 1

NUMBERS OF *Ta*· AND +· MICE PRODUCED IN THE MATINGS OF *Ta*+ AND +· MICE OVER ALTERNATE GENERATIONS OF SELECTION

Generation	Low Selection Line		High Selection Line	
	Ta· (♂♂)	+· (♂♂)	*Ta*· (♂♂)	+· (♂♂)
3	29	41	34	41
5	34	39	41	48
7	34	53	37	58
9	38	40	58	40
11	47	46	44	56
13	46	48	59	43

Clearly, the original hypothesis, that the tabby gene reduces or removes canalization is not valid, since the above data demonstrate that the zone of canalization at 18–19 vibrissae can be detected in *Ta*+ mice when their mean vibrissa number is sufficiently high. The correct explanation is that the tabby gene reduces vibrissa number to a point where this is not affected by the zone of canalization, i.e. the explanation suggested by Rendel (1959).

A secondary zone of canalization at a low number of vibrissae is suggested by the decreasing rate of response to selection which occurs in the low selection line at the *Ta*· level of expression (see Fig. 1). The presence of such a secondary zone of canalization has been suspected from the absence of *Ta*· mice with less than five vibrissae. This deficiency has become increasingly apparent as the frequency distribution of vibrissa numbers in *Ta*· mice approaches the lower numbers, since a more and more marked skewing of the distribution occurs. This is shown in Figure 4.

This secondary zone of canalization is restricted to three types of vibrissa: the ulnar–carpals, the inter-ramals, and the supra-orbitals. At each of these positions it is extremely rare for all vibrissae to be absent. Complete loss of all postorals and postorbitals is not rare.

The decreasing rate of response, and the marked skewing of the frequency distribution of vibrissa number in $Ta\cdot$ mice of the low line could be due to inviability of $Ta\cdot$ mice with less than five vibrissae. That this is not so is shown by the relative frequencies of $Ta\cdot$ and $+$ mice in the various generations of selection (see Table 1). We would expect a decreased frequency of $Ta\cdot$ males in later generations of selection if there was a decreased viability of such males which had five or less vibrissae. The data show an opposite trend; the frequency of $Ta\cdot$ males increases in the later generations of selection.

TABLE 2

MEAN LITTER SIZES OF THE HIGH AND LOW SELECTION LINES AT EACH GENERATION OF SELECTION

Selection Line	Generation of Selection:										
	3	4	5	6	7	8	9	10	11	12	13
High	6·5	8·0	6·5	7·2	5·4	6·5	6·1	6·2	6·8	6·1	6·8
Low	5·9	7·2	5·9	6·7	5·3	7·0	5·6	5·5	6·4	5·2	5·7

(*b*) *Reproductive Fitness*

A usual concomitant of any long period of selection is decreased reproductive fitness. The mean litter sizes of the high and low selection lines are given in Table 2. There is no indication of a decrease of litter size in the later generations of selection. It is possible that a subdivision of the data on the basis of vibrissa number may show a correlation of reproductive fitness with vibrissa numbers.

TABLE 3

MEAN LITTER SIZES OF THE HIGH AND LOW SELECTION LINES, FOR THE $Ta+\times+\cdot$ MATINGS, SEPARATED ON VIBRISSA NUMBER OF THE $Ta+$ PARENT

Selection Line	Vibrissa Number of $Ta+$ Parent:															
	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
Low	3·5	4·8	5·9	5·6	6·6	5·7	6·8	6·0	6·4	4·6	6·2	6·0				
High											5·2	7·1	7·0	5·8	7·3	8·0

The mean litter sizes of $Ta+\times+\cdot$ matings are given in Table 3. Matings have been grouped according to the vibrissa number of the $Ta+$ parent. There is a slight indication of a decrease of litter size from parents with a very low vibrissa number, and of an increase of litter size from parents with a very high vibrissa number. This

indication is very slight and, without more extensive data, does not justify further discussion.

The mean litter sizes of $++ \times Ta\cdot$ matings are shown in Table 4. Litter size decreases as the vibrissa number of the $Ta\cdot$ parent increases and decreases away from the mean vibrissa number of nine vibrissae. A number of $Ta\cdot$ males from the high selection line were crossed to random-bred $++$ females of another stock. The mean litter sizes of these matings are given in Table 5. There is a far more marked

TABLE 4

MEAN LITTER SIZES FOR THE HIGH AND LOW SELECTION LINES FOR THE $++ \times Ta\cdot$ MATINGS, SEPARATED FOR THE VIBRISSA NUMBER OF THE $Ta\cdot$ PARENT

Selection Line	Vibrissa Number of $Ta\cdot$ Parent:									
	5	6	7	8	9	10	11	12	13	14
Low	5·6	6·5	6·4	6·8	6·3					
High					8·0	7·8	7·3	6·8	6·0	6·7

decrease of litter size in matings whose $Ta\cdot$ parent had a vibrissa number deviating from nine vibrissae. Although the relationship between vibrissa number and litter size requires further clarification, it is clear that there is an association between the vibrissa number of $Ta\cdot$ males and the size of their litters but no, or only a slight effect, in litters of $Ta+$ females.

TABLE 5

MEAN LITTER SIZES FOR THE MATINGS OF $Ta\cdot$ MALES FROM THE HIGH SELECTION LINES TO AN UNSELECTED SAMPLE OF $++$ FEMALES FROM AN INDEPENDENT RANDOM-BRED STOCK

Vibrissa number of $Ta\cdot$ parent	9	10	11	12	13	14
Mean litter size	6·9	6·2	4·1	4·7	4·3	2·1

IV. DISCUSSION

The results show the existence of three systems affecting vibrissa number. These are:

(1) The genotype on which our selection has been effective. This is termed the "basic" genotype.

(2) The mutants which modify the effect of the basic genotype. We have considered only the tabby gene. There are two other known mutants which affect vibrissa number. These are the crinkled and ragged mutants. It is

conceivable that these are a part of the basic genotype, and have been separated from it solely by their having marked effects on vibrissa number. They are termed the "mutant" genotype.

(3) The genetic system which determines the relationship of the basic and mutant genotypes to vibrissa number. This system by its variation of this relationship from simple linearity causes a canalization of vibrissa number, and is therefore termed, the "canalization" genotype.

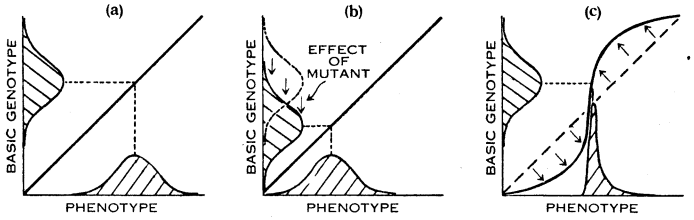

Fig. 5.—Relation of the "basic" genotype to vibrissa number: (*a*) in the absence of other genetic systems; (*b*) in the presence of the "mutant" genotype; (*c*) in the presence of the "canalization" genotype.

The basic genotype is assumed, as a starting point for the construction of our model, to be a simple additive polygenic system. In the absence of other genetic systems, the relation of the basic genotype to vibrissa number is assumed to be linear. This is illustrated in Figure 5(*a*). This can be extended to include the mutant genotype which causes a marked reduction of the number of vibrissae. This effect is assumed to be linear. It is illustrated in Figure 5(*b*).

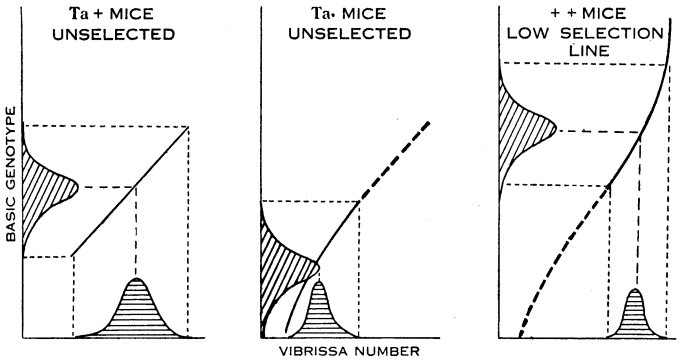

Fig. 6.—Relationship of the "basic" genotype to vibrissa number: (*a*) in unselected *Ta*+ mice; (*b*) in unselected *Ta*- mice; (*c*) in + + mice of the low selection line.

The canalization genotype determines a non-linear relationship of the basic genotype to vibrissa number, such that a large fraction of the possible genotypes produce the same vibrissa number. This is illustrated in Figure 5(*c*).

If we assume that the basic genotype and the mutant genotype are linear in their relation to vibrissa number, it is possible to estimate the curvilinearity of this

relation introduced by the canalization system. The distributions of vibrissa number in $Ta+$ mice at the commencement of selection are symmetrical, and extend over a greater range than any other genotype. It is assumed that the basic genotype is, in $Ta+$ mice, unaffected by the canalization system, i.e. this distribution is assumed to be the result of a linear relation between the basic genotype and vibrissa number. This is shown in Figure 6(a) for model distributions.

This relationship can be extended to include the $Ta\cdot$ genotype. At the commencement of selection, the distributions of vibrissa number are similar to those of $Ta+$ mice, being symmetrical and extending over a similar range. This extension is shown in Figure 6(b).

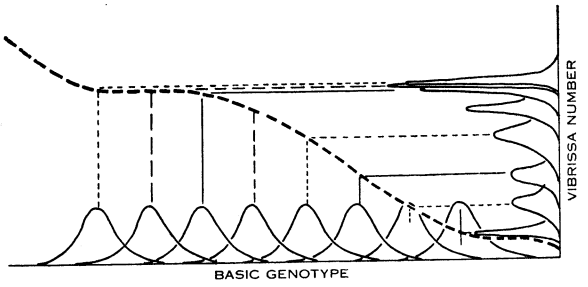

Fig. 7.—Relation of the "basic" genotype to vibrissa number which is concluded from our data, showing how a series of symmetric, equal-spaced distributions of basic genetic variation can result in asymmetric and unequal spacing of distributions of vibrissa number.

A further extension to include the $++$ genotype introduces the curvilinearity of the primary canalization zone centred on 19 vibrissae. Consider first, the last generation of selection in the low line. The $++$ mice of this generation have a distribution of vibrissa number which is symmetric, but extends over a much smaller range than of $Ta+$ and $Ta\cdot$ mice. This indicates that the relationship of the basic genotype to vibrissa number does not have any marked curvilinearity over this range, but has a steeper slope. This is illustrated in Figure 6(c).

Two other additions can be made. These both show marked canalization. They are of $++$ unselected mice and of $Ta\cdot$ mice from the low selection line. In the first, the distribution has a very small range (from 18 to 20) showing that the relation of the basic genotype to vibrissa number is very steep over this range. In the second, the distribution is markedly skewed and has a small range (from 5 to 8), indicating that the relationship is steeply sloped at 5 vibrissae. These aspects are included in the complete model shown in Figure 7.

Two assumptions have been made in the construction of this model which are difficult to demonstrate. These are (1) the independence, genetically, of the three genetic systems, i.e. the basic genotype and the mutant genotype are assumed to have no effect on the canalization of vibrissa number; and (2) that the form of the distribution of the basic genotype is unaffected by selection. This latter assumption is not unreasonable, and examination of the distribution of vibrissa number in $Ta+$

mice of the low selection line indicates that here, where the range of vibrissa number does not extend into a canalized zone, no marked changes of the form of the distribution have occurred even though a marked change of the mean has been produced by selection.

Our results with vibrissa number and the tabby gene are closely paralleled by Rendel's (1959) results with the number of scutellar bristles and the scute gene in *Drosophila*. One difference is our demonstration of a secondary zone of canalization at a lower vibrissa number. Rendel (1959) found that some degree of secondary canalization occurred at 2 and 6 scutellar bristles, but this was slight and minor compared to the primary canalization at 4 bristles. However, he has shown in a further experiment (Rendel, personal communication) that selection can produce at least a partial canalization around a bristle number of 2.

The existence of a canalized phenotype may be due (i) to a selective advantage of that phenotype; or (ii) to regularities in the basic pattern of development of the tissue concerned. Since our data do not indicate any marked selective advantage of particular vibrissa number, the first alternative does not seem very probable. A model for the second alternative can be found in the widespread effects of the tabby mutant, extending over many aspects of the development of vibrissae and coat hairs. If the basic genotype has similarly widespread effects, then it is feasible to suggest that the canalization of vibrissae is a secondary aspect of the regularity of epithelial development. Further work on this system of canalization therefore needs to be concentrated (*a*) on clarification of the relation of vibrissa number to selective advantage; and (*b*) on analyses of the pattern of development affected by the tabby mutant.

V. References

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