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SHORT PAPER

Order of loci on the X-chromosome of the mouse

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Phillips (1963), in reporting her discovery of the X-linked mutant striated (*Str*) in the mouse, *Mus musculus* L., attempted to locate its position on the X-chromosome by measuring its recombination with the genes *Ta* and *Bn* (tabby and bent-tail). She found 20% recombination between *Ta* and *Str*, and, considering male offspring only from a cross of *Str*+/+*Bn* × ++, she estimated the recombination between *Str* and *Bn* to be 34%. Since the recombination between *Ta* and *Bn* is 15% (Phillips, 1954) she concluded that *Str* was located on the side of *Ta* away from *Bn* and that the order of loci was *Bn–Ta–Mo–Str*. However, the estimate of the *Str–Bn* recombination, based on male offspring, was imprecise because of the low viability of *Bn* males, and, if one considers the female offspring, making due allowance for the incomplete penetrance of both genes in heterozygotes, Phillips' data are more consistent with a recombination of 5% between *Str* and *Bn* (indicating both genes to be on the same side of *Ta*) than with one of 34%. The position of *Str* therefore needed further study, together with the position of blotchy, *Blo* (Russell & Saylors, 1962), and the present paper reports investigation of these problems. Striated and blotchy have been located relative to *Ta*, *Bn*, Mo^{br} (brindled) and each other.

CROSSES AND RESULTS

(i) *Crosses involving striated and blotchy*

Russell & Saylors (1962) reported that blotchy showed about 4% recombination with *Ta* but did not find on which side of *Ta* it lay. Therefore, in the present work, two types of linkage test were made to test this. These were three-point tests using *Ta*, *Str* and *Blo*, and two-point tests using Mo^{br} and *Blo*.

Females of genotype *Str Ta*+/++*Blo* were bred by crossing *Str Ta*+/+++ females with ++*Blo* males and, later, *Str Ta Blo*/+++ females were obtained from these by crossing-over. These triply heterozygous females were then mated to normal males and their offspring were classified for *Str*, *Ta* and *Blo*, *Str* being distinguished from *Ta* by its patches of short fur, as described by Lyon (1963). (Striated males die *in utero*.) The results are shown in Table 1. In neither cross were there any offspring in which *Ta* had crossed over in relation to the two other genes though there were a number in which *Str* or *Blo* had crossed over. This implied that the *Ta* crossovers represented the double recombinant class and hence that the order of loci was *Str–Ta–Blo*. In view of the uncertainty of *Str*'s position, this was not sufficient to locate *Blo*.

Therefore, Mo^{br}+/+*Blo* females were prepared, by mating Mo^{br}+/++ females to +*Blo* males, and were mated to normal males. Their offspring were classified for curly whiskers at birth and for Mo^{br} and *Blo* by colour later. It is probable that Mo^{br}*Blo*/++ and

Mo^{br}*Blo* animals, if they occurred, would be indistinguishable from *Mo*^{br}+/++ and *Mo*^{br}+, and the results have therefore been grouped into only three classes (Table 2). A further, and unexpected, complication was that there was a marked shortage of offspring classified as carrying *Blo*. In addition, most of the offspring classified as carrying *Mo*^{br} were very severely affected, and many of the *Mo*^{br}+ females died, whereas typically females of this genotype have normal viability (Falconer, 1954). It is therefore thought that the probable

Table 1. *Results of three-point linkage backcrosses*

Heterozygous parent	Sex	Type of exchange and phenotype of offspring							
		None		*Str*		*Ta*		*Blo*	
Str Ta+/++*Blo*		*Str*	+	*Str*	+	*Str*	+	*Str*	+
		Ta	+	+	*Ta*	+	*Ta*	*Ta*	+
		+	*Blo*	*Blo*	+	+	*Blo*	*Blo*	+
	Female	43	64	0	4	0	0	5	5
	Male	—	55	—	3	—	0	—	2
Str Ta Blo/+++		*Str*	+	*Str*	+	*Str*	+	*Str*	+
		Ta	+	+	*Ta*	+	*Ta*	*Ta*	+
		Blo	+	+	*Blo*	*Blo*	+	+	*Blo*
	Female	48	42	3	3	0	0	3	1
	Male	—	39	—	1	—	0	—	0
		None		*Bn*		*Str*		*Ta*	
+*Str Ta*/*Bn*++		+	*Bn*	*Bn*	+	*Bn*	+	+	*Bn*
		Str	+	*Str*	+	*Str*	+	*Str*	+
		Ta	+	*Ta*	+	+	*Ta*	+	*Ta*
	Female	75	35	1	51	0	0	2†	4*
	Male	—	13	—	2	—	0	—	0
		None		T16H		*Ta*		*Blo*	
T16H *Ta*+/++*Blo*		T16H	+	T16H	+	T16H	+	T16H	+
		Ta	+	+	*Ta*	+	*Ta*	*Ta*	+
		+	*Blo*	*Blo*	+	+	*Blo*	*Blo*	+
	Female	48	41	1	0	0	0	4**	1†
	Male	30	32	0	1	0	0	2	1

* Phenotypically non-*Bn*, but genotype proved by test.
† Genotype proved by test.
** Two proved by genetic test.

explanation of the shortage of *Blo* offspring was misclassification of severely affected *Blo* and *Blo*+ as carrying *Mo*^{br}. The other main possible explanation is reduced viability of *Blo*-carrying young before the time of classification, but in either case there is no reason to suppose that wild-type young, if they had occurred, would not have been found. In fact, no wild-type young were seen, giving an observed recombination between *Mo*^{br} and *Blo* of 0% with an upper fiducial limit at the 5% probability level of 4·6%. If *Mo*^{br} and *Blo* were on opposite sides of *Ta* the recombination between them would be expected to be the sum of their individual recombination percentages with *Ta*, i.e. 8%. Thus, the observed figures in the present experiment are inconsistent with this arrangement, and consistent with *Mo*^{br} and *Blo* being on the same side of *Ta* and possibly allelic.

Table 2. *Phenotypes of offspring of* $Mo^{br}+/+Blo$ *females mated to normal males*

	Whiskers at birth		Colour at weaning		
Sex	Curly	Straight	*Mo^{br} Blo* & *Mo^{br}+*	*+Blo*	++
Female	126	0	77	17	0
Male	95	0	41	27	0

Upper fiducial limit of recombination $= 2 \times 3{\cdot}69/162$
$= 4{\cdot}6\%$

(ii) *Crosses involving striated and bent-tail*

Thus, the results of the first two series of crosses indicated the order of loci on the mouse X-chromosome to be *Str–Ta–*(*Mo^{br}–Blo*). This should mean that *Str* and *Bn* were both on the same side of *Ta*. Three-point backcrosses of *Ta*, *Str* and *Bn* were made next, in order to test this point. Females of genotype *Ta Str*+/++ *Bn* were crossed to normal males and their offspring were classified for *Ta*, *Str* and *Bn*. Linkage tests involving *Bn* are technically difficult owing to the low penetrance of *Bn*+ in females and the low viability of *Bn* in males. Therefore, in the present cross, no attempt was made to measure the recombination fractions. Instead, among the female offspring all the apparent crossovers between *Ta* and *Str* were kept and tested for *Bn*+. Then the frequencies of the various crossover types were compared. Out of six such animals tested all proved to be the result of crossing-over of *Ta* with the other two genes (Table 1). This was taken to show that *Ta* was not the middle locus. In addition, three *Bn* crossovers were found, one among the female and two among the male offspring. It was therefore concluded that *Str* was the middle locus.

This relative position of *Str* and *Bn* seems surprising in view of Phillips' estimate of 20% recombination between *Ta* and *Str*. However, throughout the present work the *Ta–Str* recombination has been consistently lower than this, and earlier work (Lyon, 1963) also gave a lower value. The final totals of the balanced two-point tests between *Ta* and *Str* mentioned in the earlier work are shown in Table 3. These indicate $8{\cdot}2 \pm 1{\cdot}3\%$

Table 3. *Results of two-point linkage backcrosses involving* Ta *and* Str

		Phenotypes of offspring			
Heterozygous parent	Sex	*Str Ta*	*Str+*	*+Ta*	++
Str+/+Ta	Female	6	117	106	16
	Male	—	—	113	10
Str Ta/++	Female	27	2	1	34
	Male	—	—	4	41

Recombination $= 39/484$
$= 8{\cdot}2\%$

recombination between the two loci. The figures from the three-point crosses give somewhat lower values, but the two-point crosses have been taken as the most accurate since in them there was less chance of disturbance of the observed ratios by reduced viability or penetrance of other genes. Thus the map of the relevant region of the mouse X-chromosome is thought to be

Bn—7–*Str*—8–*Ta*—4–(*Mo–Blo*)

(iii) *The position of the translocation break in Searle's translocation*

The relative positions of *Bn* and *Blo* having been established, it has been possible to confirm the position of the translocation break in Searle's translocation (T16H) (Lyon *et al.*, 1964). Two-point tests with the translocation and *Ta*, *Bn* or *Blo* had suggested that the break in the X-chromosome was between *Bn* and *Ta*. This was confirmed by three-point tests using T16H, *Ta* and *Blo*, together with a small amount of data from tests using *Bn*, T16H and *Ta*. Females of genotype T16H *Ta*+/++*Blo* were mated to normal males and their offspring were classified for *Ta* and *Blo*. Male offspring were also classified for T16H on the basis of their testis weight at 5–6 weeks; female offspring were classified for T16H either by their *Ta* phenotype or by genetic test where necessary (Table 1). No offspring were found in which *Ta* had crossed over relative to the other genes, although there were two T16H crossovers and 8 *Blo* crossovers. This indicated that *Ta* was the middle locus and some confirmation of this was given by the few male offspring of +T16H+/*Bn*+*Ta* females, which included one *Ta* crossover, thus suggesting that *Ta* was not the middle locus of this group. Thus the order of loci is

Bn–T16H break–*Ta*–*Blo*

with the translocation break 0·85% units from *Ta*, as reported by Lyon *et al.*

SUMMARY

The locus of the gene *striated*, *Str*, on the mouse X-chromosome, was previously reported to be on the side of tabby (*Ta*) away from bent-tail (*Bn*). Results given in the present paper show this report to have been incorrect, and that the order of loci is *Bn*–*Str*–*Ta*–(*Mo*–*Blo*). In addition, the position of the translocation break in Searle's translocation (T16H) has been confirmed, the order of loci with respect to T16H being *Bn*–T16H break –*Ta*–*Blo*.

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