GENETIC ANALYSIS OF MAROON EYE IN *ANOPHELES STEPHENSI*

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ABSTRACT. Genetic analysis of *maroon eye* in *Anopheles stephensi* indicates that it is autosomal and recessive and is located in linkage group II approximately 44.4 map units from *Stripe*.

INTRODUCTION

Anopheles stephensi Liston is an important malaria vector in much of the Middle East and the Indo-Pakistan subcontinent. The evolution of insecticide resistance and the concomitant resurgence of malaria have stimulated research on this vector mosquito. Despite the large number of investigators working with *An. stephensi,* the genetic characterization of this species is still at a very preliminary stage. Only a few morphological mutations have been described (Mason and Davidson 1966, Aslamkhan, 1973, Sakai et al. 1974, Sharma et al. 1977, Subbarao and Adak, 1978, Sharma et al. 1979, Aslamkhan and Gul 1979), a number of enzyme polymorphisms subjected to genetic analysis (Bianchi 1968, Bullini et al. 1971, Iqbal et al. 1973a,b; Di Deco et al. 1978) and the inheritance of dieldrin resistance investigated (Davidson and Mason 1963, Bryan et al. 1976, Di Deco et al. 1978). The search for genetic markers continues, and this paper reports the genetic analysis of a new mutant *maroon eye (mar).*

The mutant was detected during routine handling of a laboratory colony originating from Khano Harni, Pakistan. A few individuals of both sexes were

found with maroon eyes in contrast to the wild type black eyes. The mutants were isolated, and in the following generation a true-breeding strain was established.

MATERIALS AND METHODS

The following strains were used in the experiment:

1) *Maroon eye (mar)*—This strain is homozygous for maroon eye, 2) Khano-Harni—A wild type laboratory colony, 3) *Maroon eye; Stripe (mar; Stp)*—This strain is homozygous for maroon eye and stripe larva (Sakai et al. 1974)

Mass matings were made for all crosses and beginning the third night after the initiation of the crosses and for 2 subsequent nights a restrained mouse was used as a blood source. Gravid females were individually isolated into cotton-plugged, filter paper-lined vials, one-third filled with water. Ovipositing females were removed and discarded. On the next day a small amount of liver powder was added to each vial, and on the following day the eggs were counted to determine hatch rate. Only egg batches showing nearly complete hatch were used for the experiment. The progeny from each female were reared as individual families. For the crosses involving *Stripe,* third and fourth instar larvae were classified as stripe or non-stripe and reared separately. Upon emergence, the adults were classified for sex and eye phenotypes.

RESULTS AND DISCUSSION

Table 1 summarizes the results of crosses to elucidate the mode of inheritance of *maroon eye* and also gives the chi-square analysis of the cross results. Although the data for the individual families were recorded separately, the results are pooled in the table, as there was no significant heterogeneity among families within a cross type.

Crosses 1 and 2 are reciprocal crosses between the *mar* and wild type strains. All the F_1 progeny were wild type, indicating that *maroon eye* is recessive. Moreover, these data also suggest that *mar* is not sex linked; studies with other sex linked mutants in *An. stephensi* resulted in the ap-

Table 1. Crosses to elucidate the mode of inheritance of maroon eye in *Anopheles stephensi.*

Cross no.	Parental Genotype ♀	Parental Genotype ♂	f*	Progeny phenotypes ♀ +	Progeny phenotypes ♀ mar	Progeny phenotypes ♂ +	Progeny phenotypes ♂ mar	Chi-square 1:1 ♀:♂	Chi-square 1:1 +:mar	Chi-square 1:1:1:1	Chi-square linkage mar-sex
1	+ X / + X	mar X / mar Y	9	573	0	543	0	0.81	—	—	—
2	mar X / mar X	+ X / + Y	7	464	0	449	0	0.25	—	—	—
3	+ X / mar X	mar X / mar Y	5	153	145	170	152	0.93	1.09	2.18	—
4	mar X / + X	mar X / mar Y	8	229	226	231	222	0	0.16	0.20	—
5	mar X / mar X	+ X / mar Y	10	312	315	351	305	0.66	1.44	3.97	1.87
6	mar X / mar X	mar X / + Y	5	109	98	115	88	0.04	3.52	4.18	0.62

* f = number of families tested.

pearance of the mutant phenotype among the F_1 male progeny of one of the reciprocal parental crosses (Aslamkhan 1973, Sharma et al. 1974, Aslamkhan and Gul 1979).

Crosses 3 and 4 are heterozygous females backcrossed to *mar* males, and crosses 5 and 6 are the heterozygous male backcrosses. There were no significant deviations from the 1:1 segregation of + : *mar* or ♀ : ♂ in these 4 crosses. In all these crosses, the wild type and maroon phenotypes segregated between the male and female progeny in a 1:1:1:1 ratio. Moreover, chi-squares testing for linkage between sex and maroon in the heterozygous male backcrosses (5 and 6) were not significant, confirming that maroon is autosomal.

As the data indicated that *mar* was autosomal, crosses were initiated to investigate the linkage relationship between *mar* and another autosomal mutant, *Stripe* larva (Sakai et al. 1974). The results of the backcross $(\frac{+}{+};\frac{mar}{mar}\times\frac{Stp}{+};\frac{mar}{+})$ are as follows: 106 wild type, 83 maroon, 85 stripe and 104 maroon, stripe. The linkage chi-square was 4.67 ($p = < 0.05$), and the percent recombination between the 2 markers was 44.4 ± 2.6. *Stripe* had previously been assigned to linkage group II; therefore, *maroon eye* is now also assigned to that linkage group.

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