A RED STRIPE CHARACTER AND ITS INHERITANCE IN ANOPOHELES ALBIMANUS WIED

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ABSTRACT. The description of a red stripe mutant (Rd) in larvae, pupae, and adults of Anopheles albimanus Wied. is presented. Despite incomplete penetrance, it was determined that Rd

is inherited as a single, dominant gene in the same linkage group as St at a distance of 15–28 units. A discussion of the penetrance of Rd when associated with either St or St' is also presented.

Georghiou et al. 1967). In this investigation we describe a new character, red stripe, and report on its mode of inheritance.

MATERIALS AND METHODS

Mosquito Strains. The following strains of A. albimanus were used:

(1) White stripe (St), a strain originating on Haiti and true breeding for a dorsal white stripe character in the larva (Georghiou et al. 1967)

(2) Non-stripe (St' ), was collected in Panama by L. E. Rozeboom in 1936 and provided by the Gorgas Memorial Laboratory, Balboa Heights, Panama.

(3) Red stripe (Rd), a mutant discovered in a collection obtained from Hacienda Melara, Department of La Libertad, El Salvador, in June, 1970 (Ariaratnam and Georghiou 1971).
Table 1. Crosses involving red stripe and segregation of phenotypes obtained.

<table>
<thead>
<tr>
<th>Cross</th>
<th>Parental phenotype</th>
<th>Total examined</th>
<th>$1^{e} Rd$</th>
<th>$2^{e} Rd$</th>
<th>$3^{e} Rd$</th>
<th>Total $Rd$</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>St* x Rd*</td>
<td>3015</td>
<td>18.8</td>
<td>74.1</td>
<td>7.1</td>
<td>92.5</td>
</tr>
<tr>
<td>2</td>
<td>St* x Rd*</td>
<td>3014</td>
<td>16.1</td>
<td>69.1</td>
<td>14.8</td>
<td>98.0</td>
</tr>
<tr>
<td>3</td>
<td>St* x Rd*</td>
<td>1017</td>
<td>10.4</td>
<td>62.2</td>
<td>27.4</td>
<td>98.5</td>
</tr>
<tr>
<td>4</td>
<td>St* x Rd*</td>
<td>1017</td>
<td>10.4</td>
<td>62.2</td>
<td>27.4</td>
<td>98.5</td>
</tr>
<tr>
<td>5</td>
<td>St* x Rd*</td>
<td>940</td>
<td>10.2</td>
<td>64.4</td>
<td>25.4</td>
<td>89.0</td>
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<tr>
<td>6</td>
<td>St* x Rd*</td>
<td>940</td>
<td>10.2</td>
<td>64.4</td>
<td>25.4</td>
<td>89.0</td>
</tr>
<tr>
<td>7</td>
<td>St* x Rd*</td>
<td>878</td>
<td>9.8</td>
<td>62.8</td>
<td>27.4</td>
<td>89.9</td>
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<tr>
<td>8</td>
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<td>9.8</td>
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<td>9</td>
<td>St* x Rd*</td>
<td>877</td>
<td>10.3</td>
<td>63.3</td>
<td>26.4</td>
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<th>Total $Rd$</th>
</tr>
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<tbody>
<tr>
<td>10</td>
<td>St* x Rd*</td>
<td>257</td>
<td>4.3</td>
<td>98.3</td>
<td>1.4</td>
<td>98.0</td>
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</table>

*Unclassified as to $1^{e} Rd$, $2^{e} Rd$, or $3^{e} Rd$.

Description of Mutant. The red stripe character, $Rd$, was found to be present in both larvae and pupae but with variable intensity of expression. In its full expression it is designated as $3^{e}Rd$. A slightly lighter red is referred to as $2^{e}Rd$, while the $1^{e}Rd$ variation differs from both $3^{e}Rd$ and $2^{e}Rd$ in that its expression may be limited to only the posterior portion of the 1st abdominal segment or can be faintly discernible on the lateral edges of, at most, four segments.

The red stripe character in the adult is not easily detectable because of metallic-gold hairs which cover the abdominal surface. Upon manipulation of the specimen in relation to the light source, one observes two parallel red lines which extend down the median dorsal surface of the abdomen. The lines were generally more evident in females, probably due to their larger size. No evidence of the red stripe character was seen on the thorax.

An initial check of the homogeneity of the red stripe character among offspring showed a 100% penetrance of $Rd$ ($3^{e}Rd$ and $2^{e}Rd$ only) in the colony.

Matings. Nine crosses were performed as indicated in Table 1. They involved either 100–200 or 5–20 individuals of each sex depending on whether a sample or all of the progeny were to be examined. Only larvae with the highest expression of $Rd$ were used for crossing. Sexing was facilitated by the isolation of pupae in 20 ml shell vials. The adults were introduced in a cage progressively over a period ranging from one day to two weeks.

Results and Discussion

Allelism between $Rd$ and $St$. Crosses 1, 2, 3 and 4 were made to test red stripe against the non-stripe and white stripe alleles of the $St$ locus. In Cross 1, 525 of 3015 larvae sampled (17.5%) did not show the red stripe character. Subsequently, Crosses 2 and 3 were a repetition of the $Rd$ x $St$ cross and showed only 3.8% and 0.7% larvae, respectively, without the red stripe. White stripe ($St$) was present in all offspring larvae examined in
each cross. This agrees with data reported by Georgiou et al. (1967), who showed St to be dominant to non-stripe (St+). In Cross 4 the red stripe was present in 73.5% of the larvae, the rest being non-stripe. Thus Rd is apparently dominant over St+. The incomplete penetrance noted above will be dealt with later in the discussion.

Before examining the possibility of allelism between the Rd and St loci, Cross 5 was performed to ascertain whether Rd is a monogenic or polygenic character. The results (Table 1, Cross 5) provided no evidence of polygenic inheritance. Since no gradations in the expression of the character occurred which could not be explained by biological variation (2° Rd) or reduced expressivity of the mutant character (1° Rd), it was felt that Rd is determined by a single, dominant gene. The possibility of Rd being allelic to the St locus may also be discounted, since the backcross (St♀ x Rd♂) to St+ (Cross 6) produced four classes instead of two that would be expected if the genes were allelic. These results differ from preliminary findings of Mason (1967), who believes that a red stripe mutant in Anopheles gambiae is of polygenic inheritance.

It will be noted that the data for segregation of phenotypes in Cross 6 (backcross to St+) showed 19%:47%:33%:1% segregation of red stripe, white stripe, non-stripe, and red and white stripe, respectively. Because of the deviation from a 1:1:1:1 segregation ratio, linkage of Rd and St was suspected.

**Linkage and Penetrance Tests.** Evidence of linkage between Rd and St was sought from an F2 progeny (Cross 5). It was felt that if Rd and St were indeed linked, one would expect 1:2:1 segregation of Rd:RdSt:St, as well as 3:1 ratios for Rd:rd and St:St+. Chi-square values for the 1:2:1 and the 3:1 (Rd:rd) showed highly significant deviations from the expected (χ² = 91 and 86, respectively). However, the Chi-square for 3:1 (St:St+) showed a good fit with a value of 0.842. What is unusual about the significant Chi-squares is the greater than expected percentage of Rd/St (~54%) and Rd (~26%) offspring. This is contrary to expectation based upon the F1 progeny in which only 96.2% showed both the Rd and St characters. However, two recombinant classes Rd/St and Rd could account for the observed deviations, since any expected reduction of Rd/St and Rd would be masked by recombinant larvae. If the expected for Rd/St is ~48% (i.e. 0.5 x 96%) and for Rd 25%, the percentage of recombinations is 7% (6% Rd/St and 1% Rd). Additional evidence for the occurrence of recombinants is the presence of 0.2% (16 of 7011) St+ larvae.

Incomplete penetrance of Rd when associated with St is indicated from Crosses 1, 2, 3, and 7. In the progeny of Crosses 1-3 the range of Rd penetrance is 82.5% to 99.3%. In Cross 7, a backcross to the Rd colony, since the recombinants were not differentiable from parental types, all larvae are expected to be either Rd or Rd/St. The observed 4.2% not belonging to either class must be a result of reduced penetrance of Rd. Since Crosses 2, 3 and 7 all indicate a penetrance of 96-99%, it is likely that these results better reflect the true degree of penetrance of Rd in association with St.

In the case of Rd vs. St+, a lower degree of Rd penetrance is noted. Cross 4 resulted in only 73.5% Rd individuals. Of these, 62.2% were 2° Rd with only 0.8% showing full expression (3° Rd) of Rd. A backcross to the St+ colony using 2° Rd larvae (Cross 8) yielded only 24.1% of progeny with the marker (no 3° Rd were found). A second backcross (Cross 9) utilizing 2° Rd types from Cross 8 yielded 35.4% Rd. Therefore the degree of penetrance of Rd when associated with St+ ranges from 48.2% (Cross 8) to 70.8% (Cross 9) to 73.5% (Cross 4).

The strength of linkage between St and Rd loci may be determined using backcross data from Cross 6. Approximately 52% of this population are Rd and St+ phenotypes, a percentage of which are Rd individuals misclassified as St+. Using
the various penetrance percentages, the calculated crossover values [e.g., 52% – (0.52 x 48% x 100) + 1%, Rd/St recombinants] show that St and Rd are 15–28 units apart.

Reasons for incomplete penetrance are largely speculative and in this case it might be attributed to modifiers affecting the pigment-producing pathways. Perhaps it is sex-related, since different levels of Rd penetrance occurred in the reciprocal backcrosses (Crosses 8 and 9).

References Cited

LABORATORY TRANSMISSION OF MERMITHIDS PARASITIC IN BLACKFLIES

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ABSTRACT. Laboratory studies indicate that control of blackflies with mermithid parasites may be technically feasible. The sole method by which *Neomesomeris flumenalis* paraparasites infected larvae of *Simulium vitatum* was by direct penetration of their integument. In tests in running water, simulating the natural habitat of immature blackflies, transmission rates of about 80% for first instar larvae, and 64% for second instar larvae were achieved. Infection rates were significantly lower for later instars and 6% for pupae. The presence of detritus was an important factor in obtaining high rates of parasitism as it allowed the preparasitic nemas to crawl about in search of blackfly larvae without being swept downstream.

A 1973 agreement between the governments of Dahomey, Ghana, Ivory Coast, Mali, Togo and Upper Volta and the World Health Organization to initiate a long term control program for onchocerciasis (river blindness) in the Volta River Basin has focused attention on this filarial disease and on the control of its vectors (family Simulidae). Although current plans call for the use of the larvicide Abate®, there has also been considerable interest in the possible use of parasites for biological control since blackflies are commonly infected with protozoans, fungi and nematodes (Jammback 1973).

The present report deals with laboratory studies on the technical feasibility of using mermithid parasites (*Nematoda: Mermithidae*) in such a control program. The primary objective of this study was the achievement of consistently high rates of mermithid parasitism under standardized laboratory conditions. Defining the exact mode of infection and the relative susceptibilities of individual blackfly life stages were critical aspects of this research.

Of the known blackfly pathogens, mermithids at present attract the most interest as potential biocontrol agents. Although the species found in blackflies are not found in other organisms, they appear to exhibit little host specificity among the species of Simulidae. The mermithid *Neomesomeris flumenalis* (Welch), for example, has been reported parasitizing at least 14 species of blackflies in 3 genera.