

Six of 10 (60%) tree holes which had been positive mosquito breeding sites in 1977 (Welch, unpublished data) were now filled with nesting soil preventing the accumulation of water and making them incapable of supporting mosquitoes. Figure 1 shows a mosquito breeding site that fire ants have begun to fill with soil.

Literature reviews indicate that ants are often found in mosquito breeding sites. Buxton and Hopkins (1927) reported that ants removed mosquito eggs from their experimental plots in Samoa. Dunn (1926) surveyed tree holes in Lagos, Nigeria, for the yellow fever mosquito, *Aedes aegypti* (L.) and noted that ants were present in 85% of the holes. These investigators did not identify the ants or give details of their behavior in these habitats. James (1966) investigating possible predators of mosquito eggs in Ontario, Canada, found that ants were the most conspicuous insects collected from experimental plots. Three species of ants were suspected to be mosquito egg predators: *Myrma lobicornis fracticornis* Emery, *Lasius sikaensis* Pergande, and *Componotus herculeanus* (L.). These investigations involved the sampling of semi-permanent and transient pool sites.

There are apparently no published data to document the role of ants in the regulation of mosquito populations. The predacious habits of

S. invicta and its behavior in filling mosquito breeding sites with soil and incorporating the tree holes as nesting sites may make an important contribution to the natural control of certain tree hole mosquitoes.

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GENETICS OF RED-SPOTTED EYE IN *ANOPHELES CULICIFACIES*

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Four sex-linked mutants have been described in species A of the taxon *Anopheles culicifacies*. Giles: rose eye, *re* (Sakai et al. 1977), white eye, *w* (Sakai and Baker 1980), ventrally spaced eyes, *Vs*. (Sakai et al. 1981a) and golden body, *go* (Sakai et al. 1981b). The mutants *re* and *w* are allelic and codominant to each other but recessive to the wild type, *Vs* is dominant and lethal in males and *go* is recessive. This paper reports the genetic and linkage analyses of a new mutant, red-spotted eye, *rs*. The mutant was isolated from a gamma-irradiation-induced translocation strain, *T(Y; 2L;3)1* (Baker et al. 1978), and is characterized by red mottling in a white-eyed background. Females have darker eyes with more intense mottling than males.

The following strains were used in the crosses:

- 1) Sattoki—a wild type (+) laboratory strain.
- 2) Red-spotted eye (*rs*)—the red-spotted eye mutant strain.
- 3) Golden, red-spotted eye (*go/rs*).
- 4) Golden, rose eye, red-spotted eye (*go/re/rs*)—homozygous *re/rs* females and hemizygous *re/rs* males are phenotypically white-eyed.
- 5) Ventrally spaced eyes (*Vs*).

The methods of performing the crosses and the rearing and handling of the mosquitoes were as previously described (Sakai et al. 1977).

Table 1 summarizes the results of crosses



Fig. 1. Mosquito breeding site that fire ants have begun to fill with soil.

Table 1. Crosses to elucidate the inheritance of *rs* in species A of the taxon *Anopheles culicifacies*.

| Cross No. | Proposed Parental genotype | | | | Progeny phenotypes | | | | | | | | | | | | | | | | | | |
|-----------|----------------------------|---------------------|-------------------|---------------------|--------------------|-----|-----|-----|----|-----|----|-------|-------|-------|-------|-------|-------|----------|----------|----------|----------|-------------|---|
| | ♀ | | ♂ | | f* | sex | + | go | Vs | re | rs | go Vs | go re | go rs | Vs re | Vs rs | rc rs | go Vs re | go VS rs | go re rs | Vs re rs | go Vs re rs | |
| | | | | | | | | | | | | | | | | | | | | | | | |
| 1. | + | X | <i>rs</i> | X | ♀ | 475 | — | — | — | 0 | — | — | — | — | — | — | — | — | — | — | — | — | — |
| | + | X | — | Y | 11 | ♂ | 455 | — | — | 0 | — | — | — | — | — | — | — | — | — | — | — | — | — |
| 2. | <i>rs</i> | X | + | X | ♀ | 362 | — | — | — | 0 | — | — | — | — | — | — | — | — | — | — | — | — | — |
| | <i>rs</i> | X | — | Y | 9 | ♂ | 0 | — | — | 378 | — | — | — | — | — | — | — | — | — | — | — | — | — |
| 3. | <i>rs</i> | X | + | X | ♀ | 362 | — | — | — | 0 | — | — | — | — | — | — | — | — | — | — | — | — | — |
| | <i>rs</i> | X | — | Y (F ₁) | 8 | ♂ | 0 | — | — | 356 | — | — | — | — | — | — | — | — | — | — | — | — | — |
| 4. | <i>rs</i> | X | <i>rs</i> | X | ♀ | 0 | — | — | — | 362 | — | — | — | — | — | — | — | — | — | — | — | — | — |
| | <i>rs</i> | X | — | Y (F ₁) | 8 | ♂ | 0 | — | — | 403 | — | — | — | — | — | — | — | — | — | — | — | — | — |
| 5. | + | X | <i>rs</i> | X | ♀ | 264 | — | — | — | 247 | — | — | — | — | — | — | — | — | — | — | — | — | — |
| | + | X (F ₁) | — | Y | 9 | ♂ | 257 | — | — | 238 | — | — | — | — | — | — | — | — | — | — | — | — | — |
| 6. | <i>rs</i> | X | <i>rs</i> | X | ♀ | 272 | — | — | — | 244 | — | — | — | — | — | — | — | — | — | — | — | — | — |
| | + | X (F ₁) | — | Y | 9 | ♂ | 279 | — | — | 246 | — | — | — | — | — | — | — | — | — | — | — | — | — |
| 7. | <i>go rs</i> | X | <i>go rs</i> | X | ♀ | 135 | 42 | — | — | 48 | — | — | 110 | — | — | — | — | — | — | — | — | — | — |
| | + | X | — | Y | 6 | ♂ | 113 | 35 | — | 29 | — | — | 121 | — | — | — | — | — | — | — | — | — | — |
| 8. | <i>go re rs</i> | X | <i>go re rs</i> | X | ♀ | 173 | 43 | — | 3 | 4 | — | 3 | 0 | — | — | 52 | — | — | — | — | 151 | — | — |
| | + | X | — | Y | 7 | ♂ | 162 | 46 | — | 0 | 8 | — | 6 | 0 | — | 35 | — | — | — | — | 143 | — | — |
| 9. | + | Vs + X | <i>go + rs</i> | X | ♀ | 15 | 102 | 371 | — | 31 | 32 | — | 364 | — | 53 | — | — | — | — | — | — | — | — |
| | + | + | — | Y | 26 | ♂ | 0 | 92 | 0 | 44 | 0 | — | 391 | — | 0 | — | — | — | — | — | — | — | — |
| 10. | + | Vs + + X | <i>go + re rs</i> | X | ♀ | 2 | 35 | 178 | 0 | 0 | 15 | 8 | 0 | 1 | 3 | 9 | 0 | 0 | 209 | 34 | 0 | — | — |
| | + | + | — | Y | 10 | ♂ | 0 | 33 | 0 | 0 | 0 | 7 | 0 | 0 | 0 | 15 | 0 | 0 | 177 | 0 | 0 | — | — |

* Number of families tested.

done to investigate the inheritance of *rs*. In cross 1 between the +♀ and *rs* ♂, all the F₁ progeny were +, suggesting that *rs* is recessive. In the reciprocal cross (2) all the female progeny were + and all the males *rs* suggesting that *rs* is sex-linked as similar observations have been made from crosses with sex-linked mutants in *An. culicifacies* (Sakai et al. 1977, 1981a, 1981b; Sakai and Baker 1980) and these are the expected results for sex-linked traits when females are chromosomally XX and males XY. Backcrosses of the +(F₁) males from cross 1 to *rs* strain females gave + female and *rs* male progeny and the *rs*(F₁) males from cross 2 backcrossed to *rs* females gave all *rs* progeny. Backcrosses of the + females from crosses 1 and 2 with *rs* strain males resulted in + and *rs* female and male progeny in approximately

1:1:1:1 ratios. These results are consistent with the hypothesis that *rs* is sex-linked and recessive.

Crosses were made to investigate the linkage relationships between *rs* and *go*, *Vs*, and *re* (crosses 7–10). Highly significant (P<0.01) chi-squares were observed for linkage between all four mutants (Table 2). Highly significant departures from 1:1 ratios were observed for +:Vs and ♀:♂ in crosses 9 and 10 in agreement with previous observations that Vs is lethal in males (Sakai et al. 1981b). The observed recombination frequencies indicate that the gene sequence is *go-Vs-re-rs* with *go-Vs* = 7.1 ± 0.7%, *Vs-re* = 14.6 ± 1.6% and *re-rs* = 2.7 ± 0.4%. These results are in good agreement with previous experiments in which *go-Vs* = 7.8 ± 0.6% and *Vs-re* = 14.0 ± 0.8% (Sakai et al. 1981a).

Table 2. Observed recombination frequencies among *go*, *Vs*, *re* and *rs* in species A of the taxon *Anopheles culicifacies*.

| Cross no. | % recombination | | | | | |
|---------------|-----------------|--------------|--------------|--------------|--------------|--------------|
| | <i>go-Vs</i> | <i>Vs-re</i> | <i>re-rs</i> | <i>Vs-rs</i> | <i>go-re</i> | <i>go-rs</i> |
| 7. | — | — | — | — | — | 24.3 ± 1.7 |
| 8. | — | — | 2.9 ± 0.6 | — | 21.6 ± 1.4 | 24.5 ± 1.5 |
| 9.* | 8.1 ± 0.9 | — | — | 17.6 ± 1.2 | — | 25.7 ± 1.4 |
| 10.* | 5.3 ± 1.0 | 14.6 ± 1.6 | 2.4 ± 0.7 | 17.0 ± 1.7 | 19.9 ± 1.8 | 22.3 ± 1.9 |
| Combined data | 7.1 ± 0.7 | 14.6 ± 1.6 | 2.7 ± 0.4 | 17.3 ± 1.0 | 21.7 ± 1.1 | 24.4 ± 0.9 |

* Analysis for female progeny data only.

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INVESTIGATIONS ON THE MOSQUITO CONTROL POTENTIAL OF FORMULATIONS OF AROSURF®MSF AND CONVENTIONAL LARVICIDES¹

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Research by Levy et al. (1984) has indicated that formulations of Arosurf® MSF (Monomolecular Surface Film)² and commercial preparations of *Bacillus thuringiensis* var. *israelensis* (*B.t.i.*) (Teknar®, Bactimos® and Vectobac®) can significantly improve the mosquito-controlling efficacy when compared to the formulation components. Formulations of Arosurf MSF and Teknar, Bactimos or Vectobac were shown to kill larvae, pupae and emerging adults with application rates at or below label recommendations for each product. These Arosurf MSF-*B.t.i.* formulations are expected to be effective against ovipositing females of certain species and organophosphate and organochlorine resistant mosquitoes while producing no adverse impact upon the environment.

The following is a report of additional tests that were conducted to determine the potential of Arosurf MSF as a formulation component

for enhancing the efficacy of conventional chemical mosquito larvicides.

Laboratory bioassays were conducted against immature stages of *Culex quinquefasciatus* Say with formulations of Arosurf MSF and diesel oil No. 2, or diesel oil No. 2-isopropanol, or Abate®4-E (temephos) to determine the efficacy of each series of formulations when compared to the efficacy of the formulation components. Tests with diesel or diesel-isopropanol formulations were conducted against 10 third or fourth instar larvae while bioassays with Abate 4-E were conducted against mixed groups of 5 fourth instar larvae and 5 pupae.

Bioassays were performed in 400 ml glass beakers containing 250 ml of reverse osmosis (RO) water (3 replications/formulation). Diesel base formulations were applied to the surface of the water in each beaker with a microsyringe while formulations of Abate 4-E were applied with a glass pipette as an agitated (Bamix [M100] biomixer, Biospec Products, Bartlesville, OK 74003) water-base suspension.

Prior to application of the test formulations, larvae were fed 2 drops of finely ground rabbit chow-RO water suspension. All bioassays were conducted in a room maintained at 26-27°C (ambient) and 80% RH.

Cumulative mean percentage mortality of larvae or pupae was recorded at 24 hr post-treatment intervals and was the main criterion used to evaluate the efficacy of the formulations and their components. Results were statistically analyzed using "z" and "t" tests.

Additional tests were conducted to determine the mixing compatibility and sprayability of the various formulations at recommended application rates for each product. Preliminary field evaluations on the efficacy of several of the experimental formulations were also conducted on an operational basis to corroborate the laboratory evaluations.

Bioassays were conducted with formulations of Arosurf MSF and diesel oil No. 2 since diesel oil combined with spreading agents such as Triton®X-207 has been a standard larviciding/pupiciding formulation for many years (Hester et al. 1979). Tests against 3rd and 4th instar larvae of *Cx. quinquefasciatus* (Table 1) with formulations of Arosurf MSF and diesel oil applied at and below label recommendations for each product indicated that the 2 materials are compatible when mixed, and can produce a significantly higher rate of larval/pupal mortality in 24 hr, as well as effective delayed mortality, than either of the formulation components. Previously, Levy et al. (1982) showed that little or no Arosurf MSF-induced kill of 1st-4th instar larvae of *Cx. quinquefasciatus* would occur within

¹ Mention of a brand name or proprietary product does not constitute a guarantee or warranty by Lee County Mosquito Control District, and does not imply its approval to the exclusion of other products that may also be suitable.

² Arosurf®MSF (= ISA-20E = Arosurf®66-E2) is an EPA registered mosquito larvicide and pupicide produced by Sherex Chemical Company, Inc., P. O. Box 646, Dublin, OH 43017.