

lake front lights, the adults may disperse in greater numbers over larger areas of Sanford than observed in this study.

This study also shows that the quantitative composition of the midge species and the overall productivity of midges may change from year to year. These seasonal changes are caused by a complex interaction of intrinsic and extrinsic factors influencing the midge source. Among these factors, temperature is regarded as the most obvious, affecting seasonal cycles and abundance of some aquatic insects including chironomids (Ali et al. 1977, Elliott 1967). It was shown earlier (Ali and Baggs 1982) that water temperature in Lake Monroe and the reservoir had a positive correlation with density of larval *G. paripes*. However, a number of other factors in the midge larval habitats, such as the seasonal availability of the larval food, the nature and intensity of local oviposition by the emerging adults, presence or absence of natural enemies of midge larvae and pupae, and the prevailing favorable or unfavorable chemical conditions may also determine the larval population size of these pestiferous insects. Laboratory and field studies on the effects of nutrients on the biology and ecology of pest species of midges are presently being conducted to pro-

vide information for the eventual development of control strategies.

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EFFICACY OF *BACILLUS THURINGIENSIS* SEROTYPE H-14 AGAINST *PSOROPHORA COLUMBIAE* AND *ANOPHELES QUADRIMACULATUS* IN ARKANSAS RICELANDS^{1, 2}

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ABSTRACT. *Bacillus thuringiensis* serotype H-14 (*Bti*) was tested in small rice plots and in commercial rice fields. Application rates of 0.5 and 1.0 kg/ha in small plots resulted in 100% mortality of *Psorophora columbiae* at 24 hr posttreatment and slight residual activity 48 hr posttreatment (i.e., 13 and 27% respectively). It was also applied by airplane against 2 species at 1.0 kg/ha in 2 large field trials. Larval mortality of *Ps. columbiae* was 69 and 89% after 24 hr exposure in cups. Mixed natural populations of *Anopheles quadrimaculatus* larvae (mainly 1st and 2nd stages) were also reduced by 97%.

Intensive research has demonstrated *Bacillus thuringiensis* serotype H-14 (*Bti*) to be an effective control agent against many species of larval mosquitoes (Garcia and DesRochers 1979, Ig-

noffo et al. 1981) with no observable adverse effects on a wide range of associated nontarget organisms except some closely related Diptera (Undeen and Nagel 1978, Garcia et al. 1980a, Miura et al. 1980). Several *Bti* formulations have been tested in small plots against larvae of the dark rice field mosquito *Psorophora columbiae* (Dyar and Knab) and *Anopheles quadrimaculatus* Say in the rice-growing area of Arkansas (Hembree et al. 1980, Dame et al. 1981). Prior to this study no commercial rice field testing of *Bti* had been conducted in this region. We evaluated the efficacy of an experimental formulation of

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Bti in small plots against *Ps. columbiae* and in commercial rice fields against both *Ps. columbiae* and naturally occurring *An. quadrimaculatus* mosquito populations.

MATERIALS AND METHODS

SMALL PLOT TESTS. A wettable powder formulation of *B. thuringiensis* serotype H-14 ABG-6108 II WP (8278-77), labelled as containing 2000 ITU/mg (9×10^8 AA units/lb) was provided by Abbott Laboratory (N. Chicago, IL) for these experiments. In June 1981, small plot tests were conducted in rice field plots measuring 6.1×6.1 m from levee center to levee center, with a pan (drilled area) of 13.3 m^2 located at the Rice Branch Experiment Station, Stuttgart, Arkansas. Water depths were ca. 2.3 and 1.2 cm in the ditch and pan, respectively, with approximately 75% of the water volume contained in the ditch area. Starbonnet variety rice was grown on the levees and pan with only sparse plants in the ditches. At the time of the test the rice was ca. 40 cm tall.

Dosages of 0.125, 0.25, 0.5, and 1.0 kg/ha of *Bti* were each thoroughly mixed with 2 liters of well water immediately prior to use. Applications of the formulations to the small plots were made with a hand-pumped Hudson Thred-Lok® 3 gal utility sprayer (model no. 6031) equipped with a pressure gauge (40 psi) and a Hudson 8024 flat fan spray nozzle. Each application required spraying over the entire plot ca. 5 times for thorough distribution. Treatments and controls were replicated 3 times in randomized block design.

Late 3rd and early 4th stage *Ps. columbiae* larvae were collected with 450-ml dippers from flooded ditches. Ten larvae were placed in unwaxed 450-ml paper cups with a small amount of Tetramin® fish food and were transported to the plots.

Polyvinyl chloride (PVC) cylinders, ca. 30 cm tall, 10 cm diam, fitted with 35-mesh nylon screen bottoms and lower side vents, were banded to stakes at the edge of the rice pan. Following treatment, 10 larvae were transferred from the paper cups to each cylinder (1/plot) which was then covered with cheesecloth secured by a rubber band. All larvae were replaced with newly collected 3rd and 4th stage larvae at 24 and 96 hr posttreatment.

COMMERCIAL FIELD TESTS. Four, ca. 8.1-ha, commercial rice fields were each divided by the central-most levee into control and treatment plots of ca. 4-ha size. One field was Starbonnet rice variety; the other 3, Mars. Two aerial applications of the formulation to the commercial fields were made July 9 and 23, 1981, by a Piper Pawnee 260 at a rate of 1 kg *Bti*/16 liters

water/ha. Swaths were ca. 16.5 m wide flown at ca. 3-4.5 m above ground level at a speed of ca. 200 km/h.

The rice was an average of 50 and 70 cm tall at the 1st and 2nd applications, respectively. Approximately 30 min. elapsed following application to allow the spray to penetrate the rice canopy before *Ps. columbiae* larvae were introduced into the 8 field plots. The bioassay equipment, technique, and larval sources were the same as previously described for small plot testing.

After the first application (July 9), 3 PVC cylinders each with 10, 3rd stage *Ps. columbiae* larvae were placed about 15 m apart perpendicular to the direction of application in the interior of each plot. The same procedure was repeated following the second application (July 23), except that 4 cylinders with 10 larvae each were employed in each plot. Mortality was assessed 24 hr posttreatment. Data were analyzed by analysis of variance, and means were separated by Duncan's multiple range test.

Anopheles quadrimaculatus EVALUATION. In addition to introduction of *Psorophora* larvae, naturally occurring larval populations of *An. quadrimaculatus* were sampled 36 hr before and 24 hr after *Bti* applications. Sampling was performed by 2 people using 450-ml dippers; one sampling the levee ditches and the other the pan areas (2 samples/4-ha plot). Each sample consisted of 400 dips with a dip taken every 3-5 paces. As many randomly chosen pan areas and associated ditches were sampled as necessary to obtain 400 dips. Water from the dippers was poured through a nylon mesh bag (16 spaces/mm²) suspended from the rim of a perforated 4.5 liter plastic jar. This apparatus proved very effective for collecting larvae of all stages, including 1st instars. The larvae were identified to species and developmental stage. Data were analyzed by Student's "t" test, and means were separated by χ^2 test.

RESULTS AND DISCUSSION

SMALL PLOT TESTING. Small plot testing indicated excellent ABG-6108 activity against *Ps. columbiae* larvae. Mortality in small plots at 24 hr posttreatment ranged from 23% at 0.125 kg/ha to 100% at 0.4 and 1.0 kg/ha (Table 1). Only slight residual activity was exhibited at the 0.5 and 1.0 kg/ha doses (13 and 27% mortality, respectively) at 48 hr posttreatment. However, this level of control was not reflected against this species in commercial rice fields at 1.0 kg/ha.

COMMERCIAL FIELD TESTS. Sixty-nine percent *Ps. columbiae* mortality, corrected by Abbott's formula, was observed 24 hr after the first com-

Table 1. Percentage mortality in *Psorophora columbiana* larvae treated with *Bti* in small rice plots.

Dosage (kg/ha)	Percent mortality*		
	24 h	48 h	120 h
1.0	100.0 a	26.7 bc	0.0 c
0.5	100.0 a	13.0 bc	0.0 c
0.25	36.7 b	0.0 c	3.3 c
0.125	23.0 bc	3.3 c	0.0 c

* Means followed by same letter are not significantly different ($P = 0.05$), Duncan's multiple range test).

mercial field application. Variation among treatment plots was high, ranging from 57 to 100% mortality. The second application resulted in 89% control of *Ps. columbiana* with replicates varying from 75 to 100% mortality. Differences in *Ps. columbiana* mortality between the 2 applications were not significant. The reduction in control of *Ps. columbiana* between the small plot test and the commercial field applications may have been the result of differences in plot size and consequent practical methods of *Bti* application. Skips between swaths may have occurred in the first commercial application producing the observed variation between replicates. Though larvae were deliberately placed in relation to the swaths in both applications,

the smaller degree of variation between second application replicates may indicate more even coverage. Also, as Dame et al. (1981) pointed out, free-roaming larvae would be expected to encounter and ingest more *Bti* spores and crystals than caged larvae, and therefore skips in application would have less influence on the control of natural populations. Larvae contained in cups would not have freedom of movement and if placed in an application skip would certainly yield misleading results.

Poor penetration of plant canopies has been reported in field trials with several *Bti* formulations (Mulla et al. 1980, Washino and Garcia 1980). This does not appear to have been a significant factor involved in the first application to explain poor control against *Ps. columbiana* since good control was achieved against *An. quadrimaculatus* indicating the suspension reached the water. It is not known if the effectiveness of the second application was influenced by penetration.

Anopheles quadrimaculatus EVALUATION. *Anopheles quadrimaculatus* comprised 85% of the mosquito larvae present in the rice fields at the time of the first application. A 97% reduction of this species was observed in treatment plots 24 hr posttreatment (mean decrease from 39 to 1 larvae/replicate) compared to an

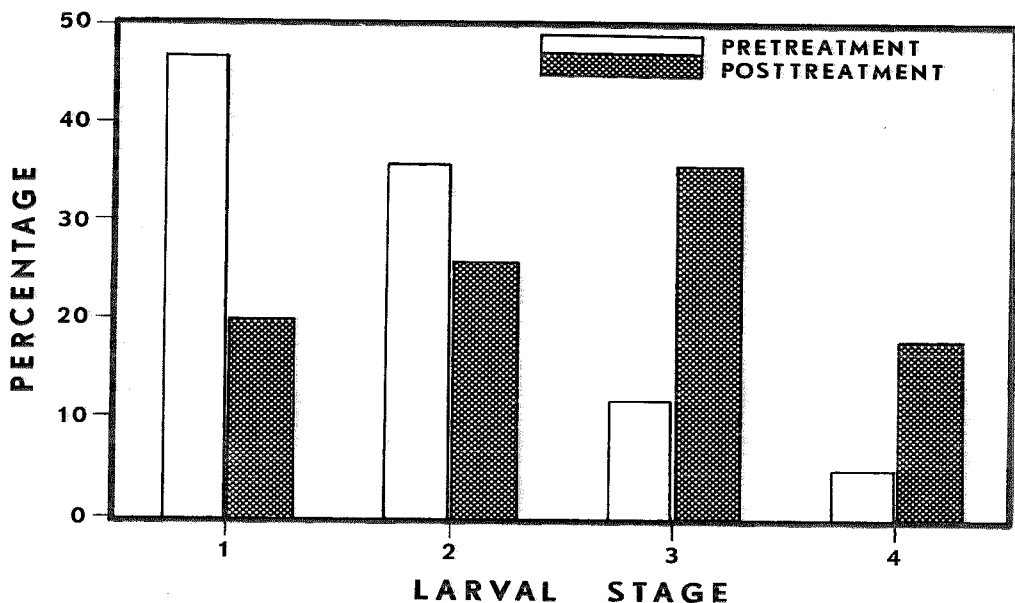


Fig. 1. *Anopheles quadrimaculatus* larval age distribution in control plots before and after first *Bti* application to commercial rice fields.

ca. 2-fold increase in control plots (mean increase from 39 to 77 larvae/replicate). Few larvae were found at the second application.

Anopheles quadrimaculatus larvae in control and treatment plots were similarly distributed with respect to developmental stage prior to the July 9 application. The control larvae as depicted in Fig. 1 were predominantly 1st and 2nd instars (47% 1st, 36% 2nd, 12% 3rd, 5% 4th). In contrast, the larval stages in control plots 24 h posttreatment were more evenly distributed with the highest percentage being 3rd stage larvae (20% 1st, 26% 2nd, 36% 3rd, 18% 4th stage).

Species of *Anopheles*, with the exception of *An. franciscanus* McCracken (Garcia et al. 1980b), have been reported to be less susceptible to *Bti* than larvae of other genera (Washino and Garcia 1980, Sun et al. 1980, Dame et al. 1981). It has been suggested that *Anopheles* may be intrinsically less susceptible, or that their surface feeding habits may not allow the ingestion of an appropriate amount of toxin since most formulations tend to sink rather quickly (Sun et al. 1980, Nugud and White 1982). A common characteristic of most anopheline larvae is their association with aquatic floatage and emergent vegetation (Horsfall 1955). In this study *An. quadrimaculatus* were readily observed atop floating algal mats in the rice fields. Some aspect(s) of *An. quadrimaculatus* utilization of the algal mat microhabitat may have been in part responsible for their apparently greater susceptibility (J. K. Olson, personal communication) or perhaps the density and cohesion of the algae acted to trap the *Bti* keeping the toxic spores accessible to these surface feeders. The extent of control achieved against *An. quadrimaculatus*, however, was most probably due to the age composition of the population since younger mosquito larvae have been reported to be more susceptible to *Bti* (Van Essen and Hembree 1980, Hembree et al. 1980, Dame et al. 1981).

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