ABSTRACT. Acute toxicity tests were conducted to measure the response of first instar Toxorhynchites splendens to commonly used mosquito adulticides: malathion, naled and resmethrin. The concentrations of pesticide causing 50% mortality (LC50) after 24 h was 2.87, 69.1 and 623 ppb for resmethrin, malathion and naled, respectively. Naled was determined to be the least toxic of the 3 compounds tested for integrated use with Tx. splendens. The latter assessments were based on comparisons between laboratory-derived dose-response curves and maximum concentrations reached in standing water calculated using standard application rates.

Interest in using Toxorhynchites mosquitoes as biological control agents has hinged upon the possibility of reducing populations of container-developing mosquitoes. In the proper setting, Toxorhynchites species may be efficacious control agents for mosquitoes (Focks 1985, Focks et al. 1986). Toxorhynchites amboinensis (Theobald) was successfully employed for integrated control of Aedes aegypti (Linn.) in New Orleans in conjunction with ULV malathion treatments (Focks et al. 1986). Other species, such as Tx. splendens (Wiedemann), have received interest for use in low-income areas of Florida (E. T. Schreiber, unpublished data). Toxorhynchites splendens is of particular interest due to its preference to oviposit into ground-level containers (Yap and Foo 1984), as typically found in urban environments.

As true for many biological control agents, Toxorhynchites-induced reductions of pestiferous mosquitoes are not expected to reach 95%, but instead are based on establishment of equilibrium between predator and prey (Focks 1986). For this reason, Toxorhynchites must be integrated into existing mosquito abatement programs as demonstrated by Focks et al. (1986). However, before further integration is attempted, an evaluation of this Toxorhynchites predator's susceptibility to commonly used toxicants is warranted. This study investigates the acute toxicity of 3 mosquito adulticides to immature Tx. splendens.

The Toxorhynchites splendens used were reared at the John A. Mulrennan Sr. Research Laboratory (JAMSRL), but were originally obtained from the New Orleans Mosquito Control District, New Orleans, Louisiana. Colony maintenance was described in Focks and Boston (1979). Eggs were oviposited by Tx. splendens into black cups (150 ml) and upon hatching, individually placed into 10 ml cells on a plastic multicell sheet to avoid injury or loss due to cannibalism.

Bioassays were conducted in accordance with the American Society for Testing and Materials (ASTM 1980). The compounds tested were malathion (Cythion*), naled (Dibrom* 14) and resmethrin (Scourge*: 18% resmethrin + 54% piperonyl butoxide). Standard methods and equipment developed at JAMSRL (Tietze et al. 1991) were employed for the conduct of bioassays. Deviations from these methods are described below. Larvae were singly transferred from the cells into 50-ml Pyrex beakers containing 40 ml of aged well water. Each test consisted of 30 beakers per concentration and 6 or 7 concentrations in addition to controls. The results of 3 "valid" tests (i.e., control mortality < 5%; dissolved oxygen > 40%; and chi-square test for homogeneity result in P > 0.01), were combined

Table 1. Toxicity of mosquito adulticides to <24 h old Toxorhynchites splendens.

<table>
<thead>
<tr>
<th>Compound</th>
<th>Test length (hours)</th>
<th>No. tests</th>
<th>Lethal concentration ng AI/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Malathion</td>
<td>24</td>
<td>3</td>
<td>69.1 (63.2-76.5)</td>
</tr>
<tr>
<td>(Cythion*)</td>
<td>48</td>
<td>3</td>
<td>49.8 (44.8-55.9)</td>
</tr>
<tr>
<td>Naled</td>
<td>24</td>
<td>3</td>
<td>623 (554-696)</td>
</tr>
<tr>
<td>(Dibrom* 14)</td>
<td>48</td>
<td>3</td>
<td>488 (431-548)</td>
</tr>
<tr>
<td>Resmethrin</td>
<td>24</td>
<td>3</td>
<td>2.87 (2.65-3.15)</td>
</tr>
<tr>
<td>(Scourge*)</td>
<td>48</td>
<td>3</td>
<td>2.07 (1.95-2.20)</td>
</tr>
</tbody>
</table>

*Note: The compounds tested were malathion (Cythion*), naled (Dibrom* 14) and resmethrin (Scourge*: 18% resmethrin + 54% piperonyl butoxide).
Fig. 1. Dose-response curves of first instar *Toxorhynchites splendens* to adulticides, malathion, naled and resmethrin after 24 and 48 h of exposure. Arrows denote theoretical concentrations in 15.2 cm of water based on maximum ground application rate (solid arrow) and maximum aerial application rate (open arrow). Theoretical aerial and ground rates for resmethrin are equal.

and analyzed using probit and chi-square tests to determine LC$_{50}$ and percent mortality. Mortalities were corrected using Abbott's formula (Abbott 1925).

Resmethrin was the most toxic compound to first instar *Tx. splendens*, followed by malathion and naled (Table 1). Theoretical concentrations were based on maximum labeled ground and aerial application rates (volume per acre) assuming 100% of the material deposited on water 15.2 cm (6 in) in depth. In contrast to malathion and naled, labeled rates for ground and aerial application of resmethrin were the same. These worst-case-scenario concentrations predicted for ground and aerial ultra-low volume sprays indicated resmethrin and malathion to exceed levels deemed toxic to *Tx. splendens* (Fig. 1). This was not the case for naled, where both theoretical exposures were below the dose-response curve (Fig. 1). Naled quickly hydrolyzes in water (Chen 1984) which may be the reason for its relatively low toxicity there.

Rawlins and Ragoonansingh (1990) compared the susceptibilities of fourth instar *Tx. moctezuma* (Dyar and Knab) and *Ae. aegypti* to organophosphates. They found that, while *Tx. moctezuma* was more tolerant to temephos than *Ae. aegypti*, both species had similar susceptibilities to malathion, fenthion, fenitrothion and chlorpyrifos. They concluded that temephos was the only organophosphate tested that would be useful in an integrated management program against these species. In comparison to the reported value for *Tx. moctezuma*, we found the toxicity of malathion to *Tx. splendens* to be 5x greater (Table 1). This discrepancy may largely be caused by differences in larval instars tested (i.e., fourth vs. first instars), which is directly related to differences in larval size and esterase content (M. S. Mulla, unpublished data) or due to species differences.

Further testing in the field is warranted to assess *Tx. splendens* mortality in their natural habitat. Such studies should focus on whether field populations of larval and adult *Tx. splendens* are affected by these adulticides. These studies should simulate operational application techniques in typical urban environments and determine what concentration of adulticide in water results from such applications.

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REFERENCES CITED
