ACTIVITY OF CHITIN SYNTHESIS INHIBITORS ON THE CAT FLEA, *CTENOCEPHALIDES FELIS BOUCHE*

Laila M. El-Gazzar, Richard S. Patterson, and Philip G. Koehler
Department of Entomology and Nematology
Institute of Food and Agricultural Sciences
University of Florida
Gainesville, FL 32611

Abstract: Three chitin synthesis inhibitors, alsystin, diflubenzuron, and cyromazine were tested against cat flea, *Ctenocephalides felis* Bouche, larvae. The chemicals were incorporated into the larval rearing media of 1.5, 2.5, and 3.5 day-old larvae. LC$_{50}$'s of 0.36, 0.09, and 0.94 ppm for alsystin, diflubenzuron, and cyromazine, respectively, were determined by probit analysis. As larval age increased, susceptibility to these chemicals decreased with diflubenzuron and cyromazine being toxic only when applied to the first two ages.

Key Words: *Ctenocephalides felis*, chitin synthesis inhibitors, diflubenzuron, cyromazine, alsystin.


Juvenile hormone analogs, such as methoprene, offer considerable potential for flea control, as has been demonstrated with the oriental rat flea, *Xenopsylla cheopis* Rothschild (Chamberlain 1975, 1979; Chamberlain and Becker 1977). Several insect growth regulators (IGRs) with juvenile hormone analog activity have been reported to kill the cat flea, *Ctenocephalides felis* Bouche, larvae and pupae effectively, when tested in the laboratory (El-Gazzar et al. 1986). They incorporated IGRs into cat flea larval rearing medium and reported 99% reduction in emerging adult fleas using 13.3 ppb of methoprene, 15.9 ppb of fenoxycarb, 410 ppb of hydroprene, or 700 ppb of Pro-drone. However, alsystin, diflubenzuron, and cyromazine, three other IGRs with chitin synthesis inhibitor activity, provided little or no mortality of cat flea larvae or pupae in concentrations up to 20 ppm.

Diflubenzuron and alsystin are benzoylphenyl urea chitin synthesis inhibitors that kill insects by preventing the normal deposition of the cuticle (Hazzar and Casida 1979; Kramer and McGregor 1979; Mass et al. 1981). Diflubenzuron, for instance, causes mortality of immature insects during ecdysis (Mulder and Gijswijt 1973) and kills dipterans by preventing pupal cuticular formation (Wright 1974). Cyromazine is a substituted melamine insect growth regulator that kills dipterans by preventing normal pupation (Bloomcamp et al. 1987; Williams and Berry 1980), but secondarily has been documented as having chitin synthesis inhibitor activity (Miller et al. 1981). Since chitin synthesis inhibitors were reported to be most effective against newly hatched larvae (Friedel and Mcdonnell 1985) and since all larvae used by El-Gazzar et al (1986) were ca. 3.5 d-old (late second or early third instar) when exposure to these chemicals was initiated, we re-evaluated alsystin, cyromazine, and diflubenzuron with younger ages of cat flea larvae.

1 Accepted for publication on 3 May 1988.
2 Current address: Department of Entomology, 114 Long Hall, Clemson University, Clemson, South Carolina 29631. On leave of absence from Middle Eastern Regional Radioisotope Centre, Cairo, Egypt.
3 Insects affecting Man and Animals Laboratory, ARS-USDA, P. O. Box 14565, Gainesville, Fla. 32604.
MATERIALS AND METHODS

Three IGRs, alsystin, diflubenzuron, and cyromazine, were evaluated against three age groups of cat flea larvae (1.5 [late first instar], 2.5 [second instar], and 3.5 [late second or early third instar] d-old). The chemicals were applied as technical active ingredients dissolved in reagent grade acetone. Serial dilutions were formulated to provide a range of concentrations between 0.01 and 20 ppm (5 discrete doses for cyromazine (0.125-20.00 ppm) and 6 discrete doses for diflubenzuron (0.01-6.00 ppm) and alsystin (0.25-10.00) for each stage) in the larval rearing medium. The composition of the larval rearing medium was 125:20:3:2 portions of sand, pulverized laboratory rat chow (Purina rodent chow #2), dried blood meal, and brewer's yeast. The chemicals were applied using the same method of El-Gazzar et al. (1986) by applying 3 ml of the appropriate dilution in 75 g of rearing medium and shaking the mixture in a 500 ml Mason jar. The mixture was transferred to 250-ml waxed paper cups, and the acetone was allowed to evaporate under a fume hood for 30 min. Appropriate-age larvae (25-200) were then placed on the medium in each cup, and the cup was covered with orange cloth secured with a rubber band. Treatments were replicated either 2 or 3 times to achieve numbers of >500 larvae per age group for each chemical.

After 5-6 wk of incubation at 27°C and 75 ± 5% RH, the cups were uncovered and the numbers of cocoons were counted, and pupae and adults were observed for morphological abnormalities. Data was adjusted for control mortality with Abbott's (1925) formula, and lethal concentrations were determined by probit analysis (Finney 1971).

RESULTS AND DISCUSSION

Table 1 presents the LC$_{50}$'s and LC$_{90}$'s for alsystin, cyromazine, and diflubenzuron when applied at different larval ages of the cat flea. Cyromazine was effective only when initially applied to media containing 1.5 or 2.5 d-old larvae. The LC$_{50}$ for the 3.5 d-old larvae was higher than 100 ppm compared to 0.94 and 5.46 ppm for the 1.5 and 3.5 d-old larvae, respectively. A range of 4-90% of the emerged adults from cyromazine treatments of 2.5-20.0 ppm were elongated; whereas, the untreated adults were normal morphologically. At the higher doses, adults reached a maximum length ca. 1.5 times the length of a normal flea. Degree of elongation appeared to be dose related as reported for cyromazine in house flies that produce elongated pupae that failed to eclose (Bloomcamp et al. 1987; Mulla and Axelrod 1983a, 1983b).

Diflubenzuron was active only when applied initially to media containing 1.5 and 2.5 d-old larvae. The LC$_{50}$'s were 0.09 and 2.22 ppm for 1.5 and 2.5 d-old larvae, respectively. Diflubenzuron was reported by Chamberlain and Becker (1977) to inhibit cocoon formation completely of oriental rat fleas at 5 ppm in the diet of second instar larvae. Our data indicate that diflubenzuron is similarly as active against cat fleas with 90% inhibition of cocoon formation at 8.58 ppm in the diet of equivalent stage larvae.

Alsystin was more active against young larvae than older larvae (Table 1). The LC$_{50}$'s were 0.36, 4.15, and 12.80 ppm for treatments made to media containing 1.5, 2.5, and 3.5 d-old larvae, respectively. In the case of 1.5 d-old larvae, complete inhibition of cocoon formation was achieved at doses >2.5 ppm.
Table 1. Lethal concentrations of three chitin synthesis inhibitors to cat flea larvae.

<table>
<thead>
<tr>
<th>Age (d)</th>
<th>LC\textsubscript{50} (ppm)</th>
<th>LC\textsubscript{90} (ppm)</th>
<th>95% C.I.</th>
<th>95% C.I.</th>
<th>Slope</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Alsystin</td>
<td>Cyromazine</td>
<td>Diflubenzuron</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1.5</td>
<td>700</td>
<td></td>
<td>0.36</td>
<td>0.20-0.53</td>
<td>2.15</td>
</tr>
<tr>
<td></td>
<td>Alsystin</td>
<td>Cyromazine</td>
<td>2.5</td>
<td>1.42-6.37</td>
<td>1.09</td>
</tr>
<tr>
<td>3.5</td>
<td>1,600</td>
<td></td>
<td>12.80</td>
<td>8.75-23.56</td>
<td>0.59</td>
</tr>
<tr>
<td>2.5</td>
<td>530</td>
<td></td>
<td>4.15</td>
<td>3.64-4.76</td>
<td>3.67</td>
</tr>
<tr>
<td>3.5</td>
<td>940</td>
<td></td>
<td>ineffective*</td>
<td>5.97</td>
<td>4.47-8.18</td>
</tr>
<tr>
<td>2.5</td>
<td>850</td>
<td></td>
<td>5.97</td>
<td>4.47-8.18</td>
<td>0.73</td>
</tr>
<tr>
<td>3.5</td>
<td>940</td>
<td></td>
<td>inefective*</td>
<td>5.97</td>
<td>4.47-8.18</td>
</tr>
</tbody>
</table>

* Probit analyses with LC\textsubscript{50} values > 100 ppm were considered ineffective.

The chitin synthesis inhibitors are potentially effective toxicants for control of the cat flea. However, bioassays of chitin synthesis inhibitors can be biased since first and second instar larvae are more susceptible than third stage larvae. Juvenile hormone analogs are most active just before pupation, and consequently, IGR's have been tested against late stage larvae regardless of whether the IGR is a juvenile hormone analog or a chitin synthesis inhibitor (El-Gazzar et al. 1986). The results of this study indicate that chitin synthesis inhibitors are most active against first instar larvae and bioassays of these compounds should be conducted with first instar larvae.

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REFERENCES CITED


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