Laboratory evaluation of a new insect growth regulator
pyriproxyfen, as a cockroach control agent

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(Received: February 20, 1989)

Key words: juvenile hormone mimic, pyriproxyfen, hydroprene, cockroach.

Abstract: Pyriproxyfen (S-31183), a pyridyl ether compound, which shows
high juvenile hormone mimic (JHM) activity against insects, was evaluated for
activities against the German cockroach, Blattella germanica. Topical application
of pyriproxyfen to last instar female nymphs induced higher inhibition of emergence
than that by hydroprene. Last instar nymphs were the most sensitive to pyriproxyfen.
High correlation was observed between the degree of inhibition of emergence of
females and the inhibition of reproduction. Topical application or voluntary contact
with pyriproxyfen caused the death of younger instar nymphs and supernumerary
nymphs. Under laboratory conditions, a population of the German cockroach con-
sisting of various nymphal stages and adults was exposed to harborage treated with
pyriproxyfen at a rate of 3.8 mg/m². Growth of the experimental population was
suppressed, and the insects died off in less than a year.

INTRODUCTION

Cockroaches are one of the most important public health pests. They are hemimetabol-
ous, and often live in inaccessible places.

There have been a number of reports on
the use of insect growth regulators (IGRs)
as cockroach control agents (Riddiford et al.,
1975; Staal et al., 1985; Bennett et al., 1986;
King and Bennett, 1988; Tsuji and Taneike,
1988). Pyriproxyfen (S-31183, 4-phenoxy-
phenyl (RS)-2-(2-pyridyloxy)propyl ether)
has a high juvenile hormone mimic (JHM)
activity, and its high efficacy as a fly and
mosquito control agent has recently been
reported (Estrada and Mulla, 1986; Mulla
et al., 1986; Hatakoshi et al., 1987; Kawada
et al., 1987, 1988; Langley et al., 1988). In
the present work, the activity of pyriproxyfen
against the German cockroach was evaluated
and the possibility of its use as a cockroach
control agent is discussed.

MATERIALS AND METHODS

Test compounds. Pyriproxyfen: 4-pheno-
xyphenyl (RS)-2-(2-pyridyloxy)propyl ether
(purity 97.2%). Hydroprene: ethyl-3,7,11-
trimethyl-2,4-dodecadienoate (purity 98.0%).
Test insects. Adults and nymphs of the
German cockroach, Blattella germanica, were
used. They were reared for successive genera-
tions in the laboratory at 25±1°C and 60%
relative humidity with a 16L-8D photoperiod
regime.

Evaluation methods. 1) Inhibition of
adult emergence: One microliter acetone
solution of the test compound was topically applied to the sternal mesothorax of each last instar female nymph. Treated nymphs were reared in plastic cups with food, water and harborage at 25±1°C with a 16L-8D photoperiod regime, and were observed at 1- or 2-day intervals. The degree of inhibition of adult emergence was scored according to the following system: score 0, normal adult; score 1, adult with melanic coloration or with abnormal wings; score 2, supernumerary nymph (Fig. 1). Forty nymphs were used in each dosage treatment, and obtained scores were averaged.

2) Inhibition of reproduction: A 0.5 μl acetone solution of the compound was topically applied to the sternal mesothorax of each of the nymph of different ages (3-, 4-, 5-, 6- and 7-week-old).

Treated nymphs were kept under the rearing conditions noted above. Inhibition of adult emergence was scored, and the number of progeny hatched from the first oothecae produced by females of the same dose group was counted weekly. The number of progeny per female was corrected for untreated control to get the inhibition of reproduction.

3) Efficacy by voluntary contact with pyriproxyfen: A 5% emulsifiable concentrate (EC) of pyriproxyfen was diluted with deionized water to the desired concentration. The solution was applied by a pipet to the surface of an overlaid plywood panel (15 ×15 cm) at a rate of 50 ml/m². The panel was placed at the bottom of a plastic container (300 cm²) with food, water and harborage. Ten or 20 nymphs in each age (0-, 1-, 2-, 3-, 4- and 5-week-old) were released into the container and reared under the conditions noted above. Inhibition of adult emergence and mortality were checked 18 weeks after release of the nymphs, and the number of progeny was counted weekly.

4) Inhibitory effect on experimental population: A plywood panel was treated in the same manner as described above. Three panels were stacked 1 cm apart to make a shelter. The shelter was placed at the bottom of a plastic container (700 cm²). Fifteen female and 10 male adults, and 50 younger-, 40 middle- and 30 older-instar nymphs were released into the container. They were provided with food and water, and reared under the conditions noted above. The total number of cockroaches was checked at 2- or 3-week intervals.

RESULTS AND DISCUSSION

1. Inhibition of emergence and reproduction of the German cockroach by topical application of pyriproxyfen

Inhibition scores of adult emergence by pyriproxyfen topically applied to last instar
female nymphs are shown in Table 1. The average scores for pyriproxyfen treatment exceeded 1.0 at all the dosages tested, including 0.1 μg/nymph. While 5 or more μg/nymph was needed for hydroprene to show the same activity as pyriproxyfen. Patterson and Koehler (1985) reported that last instar German cockroach nymphs (0 to 4 days before becoming adults) were relatively insensitive to hydroprene exposure. This phenomenon, which seems to be common in JHM, was also observed in our experiment, i.e., the average score increased when the results of insects that molted within 7 days after treatment were excluded. The score for treatment with pyriproxyfen was 2.0 in this case, at dosage of more than 0.5 μg/nymph, while 10 μg/nymph was needed for hydroprene to give the same score. Nymphs become more sensitive as they grew older (Table 2). The dosage of pyriproxyfen required to score more than 1.0 was 16 μg/nymph for 3 to 4-week-old nymphs, 4 μg/nymph for 5-week-old nymphs and 1 μg/nymph for 6 to 7-week-old nymphs, respectively. This tendency was similar to that of holometabolous insects, such as the housefly or mosquito (Hatakoshi et al., 1985, 1987). Relatively high mortality was observed at 16 μg/nymph treatment (Table 3). When treated in younger nymphal stages, the deaths were observed primarily before imaginal molting; while in older stages, the deaths were observed mainly in supernumerary nymphs. Mortalities were low at dosage of less than 4 μg/nymph. JHMs at high dosage are known to cause the death of

Table 1  Inhibitory effect of topically applied pyriproxyfen on adult emergence of the German cockroach.*

<table>
<thead>
<tr>
<th>Compound</th>
<th>Dosage (μg/nymph)</th>
<th>% individuals in each score**</th>
<th>Average score</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Score 0</td>
<td>Score 1</td>
</tr>
<tr>
<td>Pyriproxyfen</td>
<td>10</td>
<td>0 (0)</td>
<td>53.6 (0)</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>0 (0)</td>
<td>61.5 (0)</td>
</tr>
<tr>
<td></td>
<td>1</td>
<td>2.9 (0)</td>
<td>70.6 (0)</td>
</tr>
<tr>
<td></td>
<td>0.5</td>
<td>6.7 (0)</td>
<td>73.3 (0)</td>
</tr>
<tr>
<td></td>
<td>0.1</td>
<td>9.7 (0)</td>
<td>74.2 (54.5)</td>
</tr>
<tr>
<td>Hydroprene</td>
<td>10</td>
<td>0 (0)</td>
<td>66.7 (0)</td>
</tr>
<tr>
<td></td>
<td>5</td>
<td>6.5 (0)</td>
<td>77.4 (28.6)</td>
</tr>
<tr>
<td></td>
<td>1</td>
<td>23.1 (0)</td>
<td>65.4 (57.1)</td>
</tr>
<tr>
<td></td>
<td>0.5</td>
<td>32.4 (0)</td>
<td>55.8 (66.7)</td>
</tr>
<tr>
<td></td>
<td>0.1</td>
<td>54.1 (46.2)</td>
<td>43.2 (46.2)</td>
</tr>
<tr>
<td>Control</td>
<td></td>
<td>97.3 (100)</td>
<td>2.7 (0)</td>
</tr>
</tbody>
</table>

* Sixth instar female nymph. ** Figures in parentheses indicate the value when the insects molting within 7 days after treatment were excluded.

Table 2  Changes in the average score of the German cockroach responding to pyriproxyfen topically applied at different ages.

<table>
<thead>
<tr>
<th>Nymphal age when treated (in weeks)</th>
<th>Average score at different dosage (μg/nymph)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.0625</td>
</tr>
<tr>
<td>3</td>
<td>0</td>
</tr>
<tr>
<td>4</td>
<td>0</td>
</tr>
<tr>
<td>5</td>
<td>0.10</td>
</tr>
<tr>
<td>6</td>
<td>0.25</td>
</tr>
<tr>
<td>7</td>
<td>0.84</td>
</tr>
</tbody>
</table>
Table 3 Mortality* of nymphs topically treated with pyriproxyfen.

<table>
<thead>
<tr>
<th>Nymphal age when treated (in weeks)</th>
<th>Mortality as nymphs (%)</th>
<th>Mortality as supernumerary nymphs (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>4 µg</td>
<td>16 µg</td>
</tr>
<tr>
<td>3</td>
<td>0</td>
<td>55.0</td>
</tr>
<tr>
<td>4</td>
<td>0</td>
<td>45.0</td>
</tr>
<tr>
<td>5</td>
<td>10.0</td>
<td>25.0</td>
</tr>
<tr>
<td>6</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>7</td>
<td>0</td>
<td>10.0</td>
</tr>
</tbody>
</table>

* Fourteen weeks after hatching.

Table 4 Changes in the average number of progeny* per female topically treated with pyriproxyfen at different nymphal age.

<table>
<thead>
<tr>
<th>Nymphal age when treated (in weeks)</th>
<th>No. of progeny at different dosage (µg/nymph)**</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0.0625</td>
</tr>
<tr>
<td>3</td>
<td>32.6 (8)</td>
</tr>
<tr>
<td>4</td>
<td>32.1 (12)</td>
</tr>
<tr>
<td>5</td>
<td>32.9 (12)</td>
</tr>
<tr>
<td>6</td>
<td>21.7 (15)</td>
</tr>
<tr>
<td>7</td>
<td>22.2 (13)</td>
</tr>
</tbody>
</table>

* No. of nymphs hatched from the first ootheca. ** Figures in parentheses indicate the number of females used.

nymphs (Vogel et al., 1979; King and Bennett, 1988). King and Bennett (1988) reported that nymphal mortality produced by topically applied JHM (fenoxycarb) was higher in younger stages. Pyriproxyfen appears to act in a similar mode to fenoxycarb rather than to hydroprene. The death of supernumerary nymphs, however, was not noted. Such mortality seems to be caused by failure of successive molting of supernumerary nymphs. The average progeny numbers per female topically treated with pyriproxyfen at her different nymphal age are shown in Table 4. The number of progeny hatched from the first oothecae ranged from 30 to 40 for untreated females. High correlation \((r=0.91)\) was seen between the average score of females and the inhibition rate of reproduction, that is, inhibition of reproduction could be parallel to inhibition of adult emergence (Fig. 2).

Pyriproxyfen was topically applied at 7-week-old (●), 6-week-old (■), and 5-week-old (▲) nymphs, respectively.

![Fig. 2. Correlation between average score of female German cockroach and inhibition rate of reproduction.](image-url)
2. Effect by voluntary contact with pyriproxyfen

The average scores obtained by voluntary contact with pyriproxyfen are shown in Fig. 3. The age-dependent increase in the score obtained by this method was slightly higher as compared with that by topical application of the compound. Dosage of 112.5 or less μg/container (300 cm²) was less effective against nymphs in all instars, while the dosages of 225 or more μg gave high scores in each stage. Average scores were more than 1.5 at 225 μg, more than 1.9 at 450 μg and 2.0 at 900 μg, respectively. Figure 4 shows the changes in number of progeny from the first ootheca per female. The number of progeny from females treated with 56.25 μg was nearly the same as that from untreated females. The decrease in number of progeny became noticeable at the dosage of 112.5 μg or more. Insects were sterilized at the dosage of 225 μg or more. A slightly negative correlation was observed between the average score and reproductive inhibition at 112.5 μg dosage. This phenomenon was probably due to a side effect of pyriproxyfen, i.e., prolonged contact with pyriproxyfen in younger-instar nymphs induced physiological changes in adults of normal appearance. The same phenomenon by JHM was previously noted by Staal et al. (1985) who reported that morphogenetically normal adults were found occasionally that were unable to reproduce. Cumulative mortality of nymphs 18 weeks after exposure is shown in Table 5.

High mortality was observed at higher dosages. Mortality reached to more than

![Graph](image_url)

Fig. 3 Changes in the average score of the German cockroach by dosage of pyriproxyfen and the nymphal age at which voluntary contact was started.

Figures indicate pyriproxyfen dosage (μg/container).

![Graph](image_url)

Fig. 4 Changes in average number of progenies per female by dosage of pyriproxyfen and the nymphal age at which voluntary contact was started.

Average number of progenies was indicated by the number of nymphs hatched from the first oothcae. Figures indicate pyriproxyfen dosage (μg/container).

<table>
<thead>
<tr>
<th>Nymphal age in weeks when exposure started</th>
<th>Mortality (%) * at different dosage (μg/container)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>112.5</td>
</tr>
<tr>
<td>0 and 1</td>
<td>0</td>
</tr>
<tr>
<td>2 and 3</td>
<td>16.7</td>
</tr>
<tr>
<td>4 and 5</td>
<td>15.0</td>
</tr>
</tbody>
</table>

* Eighteen weeks after exposure of insects to pyriproxyfen.
70% at 900 μg treatment and to more than 50% at 450 μg. There was no significant difference in mortality between the different ages exposed to the same dosage of the compound. Almost all of the dying insects in this experiment were supernumerary nymphs failed to molt.

3. Effect on experimental populations

Figure 5 shows the difference in the population growth between experimental German cockroach populations reared in containers in which the harborage was treated with pyriproxyfen or not. The population of the untreated group grew more than 20 times as high as the initial level after 5 months. In contrast, the level of the treated population was less than twice as high as that of the initial population when treated at 3.8 mg/m², and decreased to 70% level at 38 mg/m². By the 9th month at 3.8 mg/m² and by the 7th month at 38 mg/m², the number of insects reduced to 73 and 29, respectively, of which all remaining were supernumerary nymphs having no reproductive ability. The treated population became extinct within 1 year. Staal et al. (1985) reported that treatment with hydroprene by a total release fogger at a rate of approximately 6.2 mg/m² had little effect on the population of the German cockroach after 1 year. They also reported that a residual treatment with hydroprene EC at a dosage of 44 mg/m² reduced the population to nearly zero by the 10th month. In all the tests they conducted, however, no significant reduction of population was observed in the earlier months. Our results indicate that a significant reduction of population was induced in less than 2 months after treatment with pyriproxyfen. The difference could be attributed to nymphal death caused by pyriproxyfen.

4. Conclusion

The results of our experiment seem to show the possibility that pyriproxyfen can be used as one of a cockroach control agents. Population growth of the German cockroach is suppressed or maintained at a low level by the inhibition of reproduction and nymphal deaths caused by contact with pyriproxyfen.

Further examinations of different factors, such as dosages and methods of application including combination with adulticides under actual conditions, will be needed for the practical use of the compound against cockroaches.

Acknowledgements

The authors wish to express their thanks to Dr. E. J. Gerberg of Insect Control and Research, Inc., U.S.A. for his invaluable suggestions.

References


American Chemical Society, Bethesda.


### 摘要

**昆虫成長制御剤ビリブロキシフェンのゴキブリに対する効力評価**

昆虫に対して高い幼若ホルモン様活性を示すピリテルミカル系化合物、ビリブロキシフェン（S-31183）のチャパネゴキブリに対する活性を評価した。雌終齢幼虫に対する局所施用法により、本剤はハイドロプレンに勝る羽化阻害効果を示した。幼虫の本剤に対する感受性は、終齢幼虫の羽化1週間前までの期間に最も高かった。また、雌の羽化阻害の程度と次世代の産仔数の減少との関には高い相関がみられ、羽化阻害の程度が高いほど産仔数が減少する傾向が得られた。さらに、本剤の局所施用や本剤に対する非強制的な接触により、若齢幼虫あるいは過剰脱皮幼虫の脱皮失敗による死亡が観察された。チャパネゴキブリ各ステージの幼虫および成虫を、ビリブロキシフェンにより処理したシェルターで飼育したところ、床面積あたり3.8 mg/m²の薬量で個体数の増殖を抑え、1年以内に個体群は消滅した。以上の結果より、ビリブロキシフェンはゴキブリ防除剤として有効な成分と考えられ、実用的には成虫防除剤との組合せによって高い防除効果が期待される。