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Testing chemical repellency in cockroaches (Blattodea) based on feces dispersion

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With 2 figures and 1 table

Abstract

A new method of evaluating repellency of chemicals to Blattella germanica is described. The method is based on recording the number of feces left in areas of a glass arena treated and untreated with an insecticide. The increased accumulation of feces indicates an area where adults spend more time over 24 hours testing period.

1 Introduction

Cockroaches are able to avoid places contaminated by toxic deposits. This phenomenon is known as a behavioural resistance which has been shown an important factor of reducing cockroach mortality in the presence of pesticide (Ebelling et al., 1966). Investigation of repellent properties of different substances is generally based on recording of distribution of individuals on treated/untreated surfaces. However, the resulting pattern of cockroach aggregation could easily be disturbed by any accidental movement of air or surface vibration and the result of experiment becomes invalid. Several methods for testing chemical repellency in cockroaches has been proposed so far. They include harbourage-choice method (Goodhue and Tissot, 1952), multiple harbourage-choice arena method (Zungoli et al., 1988), slant-board method (Goodhue, 1960), and choice-box method (Ebelling, 1966). These methods are based on direct observation of animals, require special equipment and do not evaluate data with respect to the role of aggregation pheromone (Schneider and Bennett, 1985) which is also present in cockroach feces (Ishii and Kuwahara, 1967; Rust and Reijnders, 1976).

The aim of our work was to develop simple method which would enable a quick screening of repellency of chemicals to various strains of Blattella germanica. The innovated approach is based on the observation that the increasing cumulation of feces mark a safe area and appropriate resting sites (Denzer et al., 1988; Stejskal, in press).

2 Materials and Methods

The tests were conducted under laboratory conditions of 25 °C and 60–65% RH. For experiments we used 7–14 days old adult males of the German cockroach of an insecticide susceptible strain maintained in the Food Research Institute of Prague. Two days before experiments, an ample amount of fresh food (apples and mice pelleted food) and water was provided to the experimental individuals. For the repellency test commercially available formulations of pesticides diluted in acetone were used. The tested blatticides contained active ingredients applied in following concentrations and doses: permethrin – 0.08%, 0.03%, 0.008%, 0.003% and 0.01 ml/cm² of Coopex 25 WP (Ag-REVO), pirimiphos-methyl – 0.08%, 0.03%, 0.008% 0.003% and 0.01 ml/cm² of Actellic EC 50 (Zeneca), bendiocarb – 0.08%, 0.03%, 0.008%, 0.003% and 0.004 ml/cm² of Ficam 80 W (AgREVO). Each chemical was applied at lower than the manufacturer’s recommended dosage in order to obtain low mortality rate and, consequently, high numbers of feces.

A 4-choice arena was set up in which the insects could be either on the treated or untreated quarters of equal size. The repellency tests were carried out on filter papers put on the bottom of petri dishes of 24 cm diameter. Inner edges of dishes were spread by vaseline to prevent the escaping of the cockroaches. Round filter papers of 24 cm diameter were treated with an insecticide or acetone, dried and cut into 4 parts of an even size. Treated and untreated parts were arranged symmetrically so that opposite areas were treated with insecticide or acetone. The papers were joined and mounted on the bottom of the cover lid of petri dish by a sticky tape. A piece of apple was placed into the center of each petri dish. Before tests cockroaches were cooled rather than anesthetised with CO2 to avoid hyperactivity (Zungoli et al., 1988). Ten males of Blattella germanica were confined to the arena by closing the dish with the narrower bottom part of petri dish. For comparison of distribution of feces on insecticide-free surface 10 replicates with acetone treated and untreated quarters were set up in the same way. All petri dishes were covered by a nontransparent plastic foil to avoid interference with negative phototaxis. A number of feces at each quarter of filter paper was recorded after 24 h. Each test was repeated 10 times.

The numbers of feces in untreated (UT) or treated (T) areas were added and than a UT/T ratio of feces in each replication was calculated. The marginal values of UT/T were excluded and the results were subjected to ANOVA and Duncan’s multiple range test to separate means. Another criterion for repellency evaluation was the percentage of petri dishes where UT > T. The latter criterion did not depend on absolute numbers of feces excreted by cockroaches.

3 Results and discussion

The repellency results obtained by this method are similar to the published data obtained by other methods. The results of 24 h counts of feces, expressed as UT/T ratios, are summarized in table 1. Fig. 1 and 2 show decreasing trends in repellency with decreasing concentration of insecticides. As expected, the significantly highest repellency was found in permethrin. The repellency of other pyrethroid insecticides to cockroaches was...
already documented (Schneider and Bennett, 1985). Although UT/T ratio increased also for Actellic and Ficam, the differences to control were not significant at the 95% level. The reasons obviously were a lower repellent effect of both chemicals compared with permethrin and a large individual variation of cockroach behaviour resulting in unequal aggregation of feces in the control tests (tab. 1 - acetone). The variation of aggregation behaviour was also observed in adults (Schneider and Bennett, 1985).

Tab. 1. A distribution ratio (UT/T) of Blattella germanica feces between untreated (UT) and treated (T) areas of the arena in the upper concentrations of three blatticides used, and acetone

<table>
<thead>
<tr>
<th>Active Ingredient</th>
<th>Formulation (concentration)</th>
<th>Average UT/T ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>Permethrin</td>
<td>Coopex 25 WP (0.08%)</td>
<td>3.8a ± 0.5</td>
</tr>
<tr>
<td>Pirimiphos-methyl</td>
<td>Actellic 50 EC (0.08%)</td>
<td>2.5ab ± 0.5</td>
</tr>
<tr>
<td>Propoxur</td>
<td>Ficam 80 WP (0.08%)</td>
<td>1.5 b ± 0.3</td>
</tr>
<tr>
<td>Acetone</td>
<td>Acetone</td>
<td>1.3 b ± 0.2</td>
</tr>
</tbody>
</table>

The advantage of our approach is that the distribution of feces reflects average distribution of animals over a 24 h testing period. Additionally, it allows interpretation of data with respect of the role of aggregation pheromone. It should be kept in mind that aggregation pheromone contained in feces can itself affect the spatial distribution of cockroaches. On the other hand, our method is not suitable for screening quickly killing concentrations of pesticides. However, the feces distribution approach can be modified or combined with other methods. For example, while testing repellency in well established Ebeling choice boxes one should take into account not only mortality and distribution of individuals, but also the amount and dispersion of their feces. We would like to stress the importance of using insect of the same nutritional status which allows strict comparison results of vario-