FORCED CONTACT AND ARENA BIOASSAYS TO ASSESS THE PERFORMANCE OF A PYRETHROID WP DEPOSIT AGAINST ORIENTAL COCKROACHES

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Abstract—LT50s and LT95s were determined at 28°C for adult female *Blatta orientalis* over deposit concentrations varying from 15–200 mg AI m^{-2} on a plywood surface treated with a WP formulation of cypermethrin. Mortality was determined 3 days following removal from the deposits. Regression equations for these parameters were

LT50 (min) = 31.8–11.8 log [surface concentration (mg AI m^{-2})] (r^2 = 0.896)
LT95 (min) = 65.0–25.1 log [surface concentration (mg AI m^{-2})] (r^2 = 0.875)

These results were compared with mortalities obtained when the cockroaches were allowed free movement in arenas containing a harbourage and a treated strip of the WP deposit (40 mg/m^2) positioned either adjacent to the harbourage, across the centre, or at the far end of the arena with food/water stations placed on the deposit. The arenas were illuminated on a 12:12 photocycle. 50 adult females were introduced into each arena at the start of a light phase and allowed to condition it for 84 h. Treated strips were introduced prior to the start of the subsequent dark phase, and cockroach emergence, distribution and knockdown monitored during this dark phase. Mortality was recorded 3 and 6 days after introduction of the plates. Positioning the deposit either at the harbourage end or the food/water end of the arena gave >95% knockdown within 6–7 h and > 95% mortality at 3 days, consistent with the high proportion of the time which active cockroaches spent in these two locations during the first 3 h of the dark phase. Most of the cockroaches surviving the first dark phase also survived the subsequent 6 dark phases. In control arenas a maximum of 30% of the population was active in the arena at any time during the dark phase, whereas contact with treated strips placed at the harbourage/food and water ends produced a flushing action 2–4 h into the dark phase in which >95% of the population emerged over a period of approximately 1 h. The flushing effect produced faster knockdown and a greater percentage mortality than would have been anticipated on the basis of the forced contact bioassays and initial pattern of emergence.

INTRODUCTION

Formulations of residual deposits of blatticides are often compared during development by forced contact tests on treated surfaces. Such tests do not assess the effect of behavioural contributions to the performance of insecticides such as irritancy and avoidance behaviour, the aggregative behaviour of harbouring cockroaches which may lead to cross-contamination and activation, and the diurnal behaviour of cockroaches which determines their period of exposure to a deposit and provides an opportunity to recover from sub-lethal contacts. These limitations can be partly remedied by conducting laboratory tests using arenas with harbourages, although such tests normally enclose the cockroaches in a much smaller area than their typical foraging range, and the number of experimental variables (arena dimensions, width and position of deposit, photocycle, period of habituation to arena) is increased.

This paper describes a simple arena for determining the performance of a residual pyrethroid deposit to the Oriental cockroach *Blatta orientalis,* and relates 3 day mortalities resulting from exposure to treated plates at three positions in the arena with those produced in forced contact bioassays with varying periods of exposure to the deposit.

MATERIALS AND METHODS

The strain of *B. orientalis* has been cultured at Silwood Park for 5 years at 28°C on a diet of mouse pellets and water supplied *ad lib.* Forced contact and arena tests were carried out in a constant environment room at a temperature of 28° –1°C and an RH of 50% –10%. The room was set to a 12:12 L:D photocycle. Illumination in the light phase was provided by banks of 4 fluorescent

daylight tubes (58W, 5 foot), one bank positioned 90 cm above each arena; lighting in the dark phase was from 15W photographic development lamps with red filters, one above each arena.

Plywood plates were sprayed with cypermethrin as a 40% w/w WP (Demon 40) at 40 mg AI m^{-2} 24 h prior to use in bioassays, using a calibrated Mardrive track-sprayer. Forced contact bioassays were conducted by allowing individual anaesthetised cockroaches to recover for 2 h on untreated plywood plates, walking them onto the treated surfaces under a plastic beaker, exposing them for a timed interval and then walking them into 1 litre jars with slant board harbourage, food and water. Four replicates of 5 insects were exposed for each of five exposure periods at each surface concentration. LT50 and LT95 values (exposure times required to give 50% and 95% mortality after a 3 day holding period) were estimated by probit analysis

Arenas were 50×120×13 cm boxes with base and one end wall lined with filter paper, the remaining 3 walls lined with glass plates, and a melamine-faced partition with 4 exit holes at its base positioned 2 cm from the end wall lined with filter paper. The area between the filter paper wall and the melamine partition was covered with an opaque plastic sheet and formed the harbourage, and the remaining base area was available for foraging during dark phases. Food and water stations were positioned 5 cm from the end wall furthest from the harbourage.

Adult females (50) were added to the foraging area at the beginning of a light phase, and allowed to acclimate to the arena for 84 h. Treated and/or control plywood plates (50×11 cm) were added to each arena just prior to the beginning of the next dark phase with the minimum of disturbance. The experiment consisted of four replicates of four treatments, each treatment consisting of the acclimation period followed by a 6 day exposure to 3 plates, two unsprayed and one sprayed, with the sprayed plate either adjacent to the harbourage, across the centre of arena, or across the far end of arena, and the unsprayed plates occupying the other two positions. The control arena contained 3 unsprayed plates. The mean number of cockroaches in the foraging area of the arena and on each of the plates was monitored at hourly intervals during the first dark phase following introduction of plates as a series of 5 observations at 1 minute intervals. Knockdown was recorded at hourly intervals, and mortality recorded at 3 and 6 days after introduction of the plates.

RESULTS

Forced Contact Bioassays

The relationship between estimated LT_{50} (exposure time required to give 50% mortality after 3 days) and surface concentration in the forced contact bioassays is shown in Figure 1. The time to initiate hyperexcitation and knockdown in affected insects tended to decrease and the proportion of the insects showing initial knock-down followed by recovery within 3 days tended to increase with increasing surface concentration for a specific exposure interval. Knockdown generally occurred within 30 min of removal of cockroaches from the plates. The estimated LT_{50}s and LT_{95}s were fitted to logarithmic regressions, giving

LT50 (min) = 32.6–12.2 log [surface concentration (mg AI m^{-2})] (r^2 = 0.896)
LT95 (min) = 65.0–25.1 log [surface concentration (mg AI m^{-2})] (r^2 = 0.875)

within the surface concentration range 15–200 mg AI m^{-2}. At a surface concentration of 40 mg m^{-2} the experimental LT50, LT95 and corresponding 95% fiducial limits were 11.0 (9.2–12.9) and 19.1 (15.5–32.2) min respectively.

Arena Assays

Cockroaches were introduced into the arenas at the beginning of a light phase and established themselves on the end wall of the harbourage within approximately 30 min. The mean harbouring density was 770 insects/m^{-2}. Thereafter, emergence of the cockroaches occurred during dark phases, with only occasional cockroaches emerging to sample food and/or water during a light phase. Cockroaches in control arenas had a consistent pattern of emergence, with a peak of activity about 6 h into the dark period at which time approximately 30% of the population was mobile within the foraging area (Figure 2).

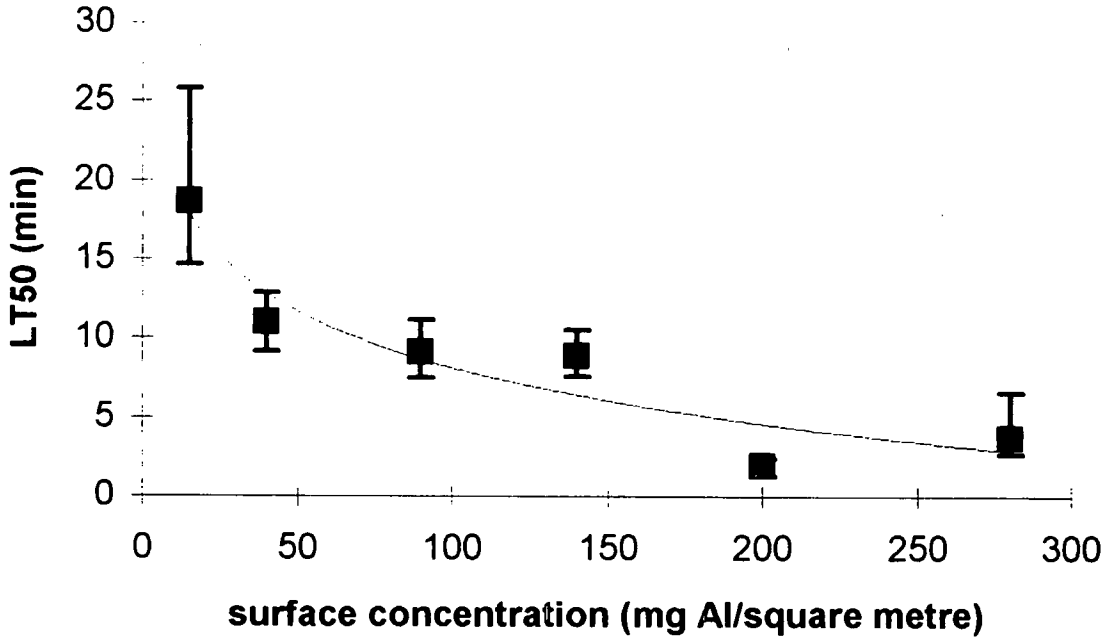

Figure 1. Effect of surface concentration on LT_{50} in forced contact bioassays

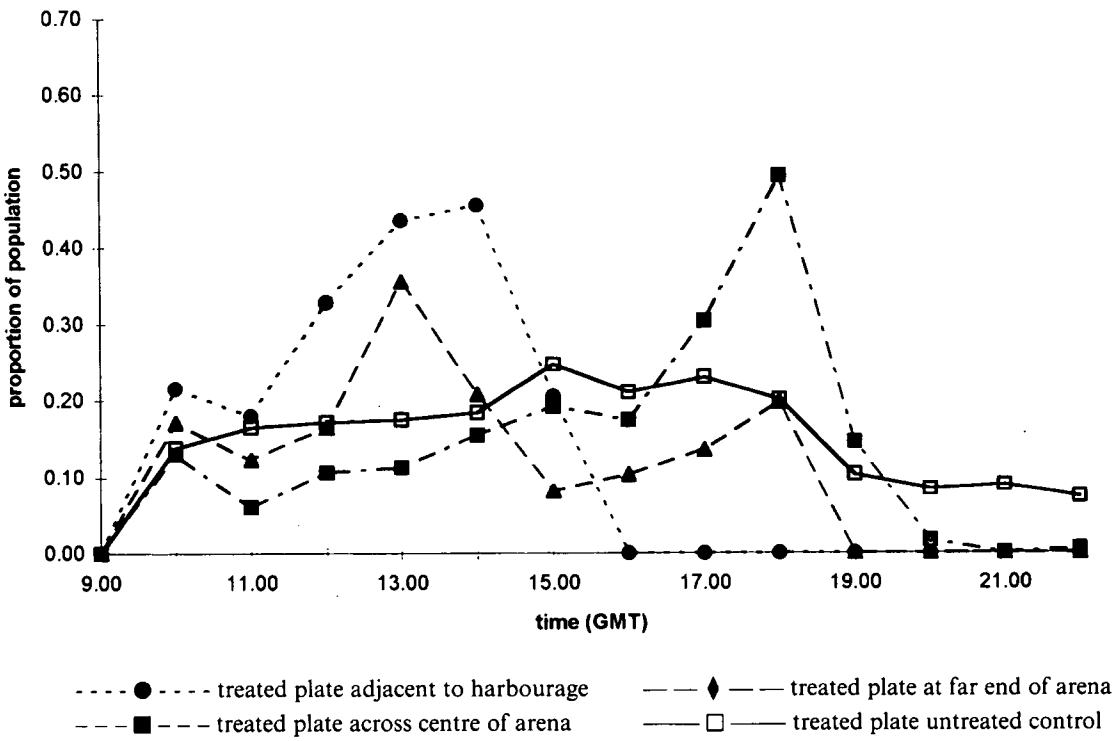

Figure 2. Proportion of cockroaches active in foraging area during first dark phase after introduction of treated plates.

Placement of the treated plate adjacent to the harbourage produced hyperexcited individuals within approximately 2 h of the start of the dark period, with > 95% expellency from the harbourage and knockdown of the population within 6–7 h. There was no significant recovery, and all knocked-down cockroaches were scored as dead after 3 days. When the treated plates were positioned at the far end of the arena and under the food and water stations, emergence and knockdown showed a similar pattern but the peak of emergence was delayed (Figure 2) and survivors (4–10%) were consistently found in the harbourage after the first dark period. Survivors remained unaffected by the insecticidal deposit over the subsequent 6 days. Results for arenas with treated plates in the central position were more variable, with 3 day mortalities varying from 16%–94%. In the cases where high mortality was observed, patterns of knockdown and expellency were similar to the two cases just described but peak emergence occurred 6–7 h into the scotophase; in one replicate no knockdown was observed during the first dark phase and the 3 day mortality was reduced to 16%. Survivorship in the other 3 replicates showed the same pattern as for plates in the two alternative positions. Mean mortalities at 3 and 6 days are summarised in Table 1.

Although the mean cockroach activity near the food/water stations and the harbourage during the first 3 h of exposure was lower when the arena contained a treated plate, the difference between the four treatments was not significant at $p < 0.05$ (Figure 3). In all treatments the mean cockroach activity on the centre plate was significantly lower than the mean activity on plates in either alternative position ($p < 0.05$).

DISCUSSION

Control of Oriental cockroaches in infested buildings may be frustrated by the difficulty of getting insecticidal treatments into complex harbourages, although it is usually possible to isolate populations from food and water sources by spraying into and around harbourage access points. The arena used in these tests, incorporating a toxic barrier between an untreated harbourage and a source of food and water, was designed to simulate this situation. The "choice" presented to the cockroaches was whether or not to emerge from the untreated harbourage and contact the deposit. The design therefore differed fundamentally from arenas in which irritancy or repellency of a residual deposit is assessed by measuring the extent to which cockroaches are deterred from entering a treated harbourage (Ebeling *et al.*, 1967; Rust and Reierson, 1978; Rust *et al.*, 1993;Hostetler and Brenner, 1994).

Emergence of the cockroaches from harbourages of untreated arenas started almost immediately after the onset of the dark phase, with the maximum number of cockroaches active in the foraging area after approximately 6 hours. Preliminary experiments in which cockroaches were individually tagged showed that this emergence pattern initially involved a few active cockroaches making repeated excursions to and from the harbourage, with these individuals being progressively joined/replaced by others which became active at a later time in the dark phase (le Patourel, unpublished results). At the peak of activity, approximately 30% of the population was active in the arena. The strong diurnal activity pattern of the Oriental cockroach was investigated by Gunn (1940) and Fuchs and Sann (1981), although its relationship to food and water availability or different light intensities during photophase has not been established. Likewise, it is not known whether the activity of gravid female Oriental cockroaches is lower than that of non-gravid ones, as

Table 1: Mean percentage mortality (±SD) in arenas following exposure to plates treated with cypermethrin WP (40 mg m^{-2})

Position of treated plate							
harbourage end of arena		centre of arena		food/water end of arena		control	
3 day	6 day	3 day	6 day	3 day	6 day	3 day	6 day
100	100	73±45	75±38	96±4	96±4	1±1	2±1

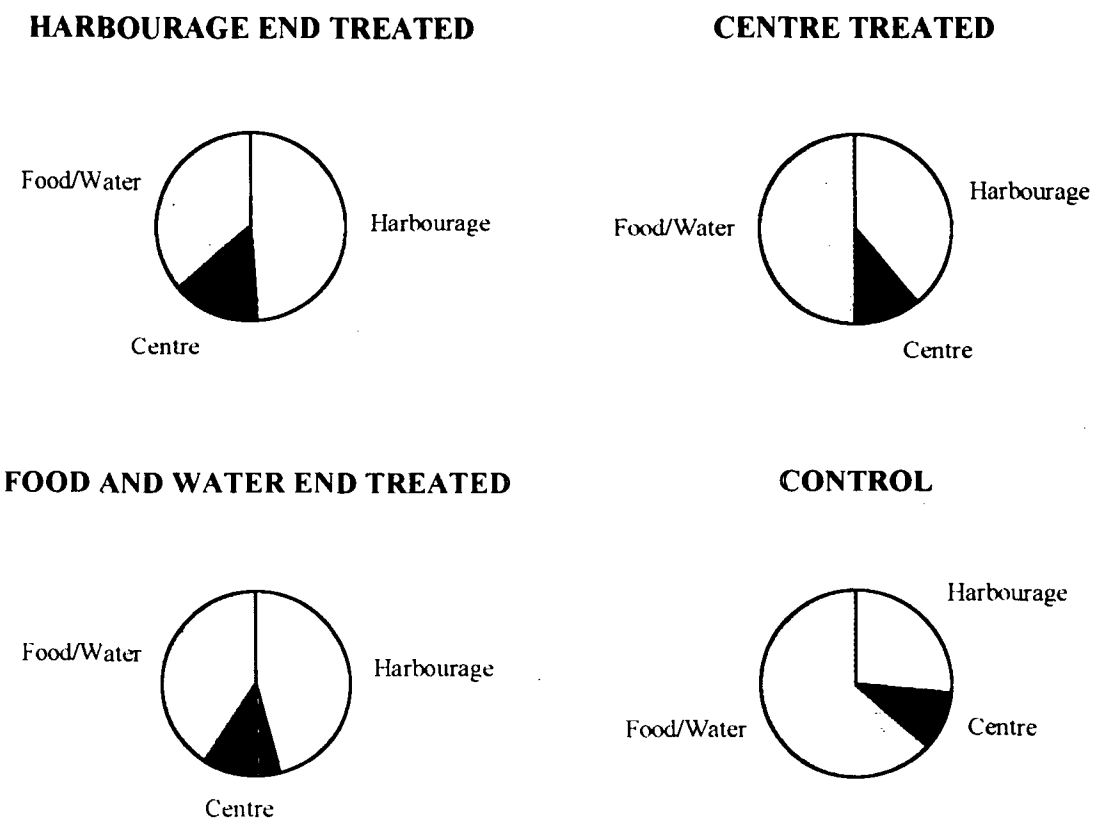

Figure 3. Relative cockroach activity on treated plates positioned either adjacent to harbourage, at far end of arena or across centre of arena during first 3 h of dark phase.

with German cockroaches (Silverman, 1986). No work has been reported on the comparative movement behaviour of field and laboratory strains of Oriental cockroaches, although in the case of German cockroaches Akers and Robinson (1983) reported a decrease in activity of a laboratory strain relative to two field strains. Denzer *et al.* (1985) investigated the activity of mixed populations of Oriental cockroaches under laboratory conditions and found both total and individual activity to increase with population density and time of conditioning of the arenas.

The initial pattern of emergence (0–3 h) into the foraging areas of arenas containing treated strips was not significantly different from controls, regardless of whether the strip was placed across the harbourage exits, under the food and water stations, or in an intermediate position. No evidence was obtained that initial sub-lethal contacts with the pyrethroid deposit deterred cockroaches from emerging or re-emerging from the harbourage and accumulating a lethal dose of insecticide during the first dark period. The activity of the cockroaches on the treated plates during the first 3 h of the first dark phase, greater for plates placed across the harbourage or under the food and water stations than for plates placed in an intermediate position across the arena, gave a good indication of the knockdown/mortality to be expected at the end of the dark phase.

The expellency from the harbourage induced by contact of a proportion of a population with a pyrethroid deposit has been commented on by Carter and Chadwick (1978) for both German and Oriental cockroaches following field application and by Schneider and Bennett (1985) for German cockroaches in arenas. The mechanism of the effect is unclear – it could be caused by contamination of inactive cockroaches in the harbourage by individuals returning after contacting a deposit, by disturbance of harbouring cockroaches as a result of increased activity of such intoxicated individuals, or by release of dispersion-producing chemicals in the harbourage. Ross (1992) has suggested that dispersion of German cockroaches can be caused by the vapour action of formulations of residual pyrethroids, but this seems unlikely in the present case given the relative

magnitude of the effect produced by deposits placed across the centre of the arena and at the far end under the food and water stations. The time-course of the expellency phenomenon, involving a lag period of 3–5 h after which most of the population emerged from the harbourage and were knocked down within 30–60 minutes, suggests prior contamination within the harbourage as the most likely cause.

It is not clear whether the few long-term survivors from treatments (Table 1) were physiologically tolerant of the deposit, had a modified behavioural pattern of foraging for food and water during subsequent dark periods, or – because they occupied a less crowded harbourage – were less susceptible to contamination by other cockroaches. They were observed to emerge from the harbourage and cross the treated strips during subsequent dark phases, but no detailed record was made of their activity in these experiments.

The forced contact bioassays, which indicated a continuous contact time of approximately 20 min on the 40 mg m^{-2} deposit to ensure >95% kill, are consistent with the hypothesis that mortality in the arena tests is due primarily to contact of active individuals with the treated plates at the start of the dark phase followed by a period of cross-contamination of inactive individuals in the harbourage and a resulting expellency which ensures that most or all of the population acquires a lethal dose within 6–7 h. Positioning the deposit in areas of the arena where cockroaches spent most time thus gave the greatest probability of producing the effect. If this mechanism is operative one would predict that mortality in the arena should decrease with decreasing population density (lower probability of cross-contamination in the harbourage) but be relatively insensitive to the size of the foraging area – provided the deposit is placed either adjacent to the harbourage or around the food/water stations.

The arena bioassays in which treated plates were placed either adjacent to the harbourage or at the far end of the arena produced a consistent expellent effect similar to that reported following field application of permethrin (Carter and Chadwick, 1978) and >95% mortality (3 day) at a dose (40 mg AI m^{-2}) equivalent to approximately 60% of the manufacturer's recommended rate for the formulation at a normal level of infestation.

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