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COMPARISONS OF CAT FLEA (SPHONAPTERA: PULICIDAE) ADULT AND LARVAL INSECTICIDE SUSCEPTIBILITY

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ABSTRACT

The resistance levels of a Florida strain of cat fleas, *Ctenocephalides felis* (Bouche), were evaluated using nine chemicals after colonization for approximately one (1984) and

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two (1985) years. It was found that the fleas had lost some of their resistance to chlorpyrifos and malathion in 1985 compared to 1984. Moreover, higher resistance was detected to the three carbamates, bendiocarb, propoxur, and carbaryl. The increased carbamate resistance was attributed to the use of carbaryl dust to kill the fleas on cats after they were taken out of flea production. The resistance levels for propetamphos, diazinon, isofenphos and chlorfenvinphos did not change significantly.

Comparing the 24 h toxicity of the nine compounds against larval and adult stages in 1985, chlorpyrifos, diazinon, chlorfenvinphos, and propoxur were more toxic to adults than larvae and propetamphos, malathion, and bendiocarb were more toxic to larvae than adults. The tolerance to isofenphos was similar for larvae and adults. Both stages responded equally to carbaryl and were completely resistant.

All organophosphates in 1985 exhibited delayed larval mortalities and affected cocoon formation of the larvae that survived the 24 h exposure. However, none of the organophosphates, or the carbamates, appeared to have any effect on adult emergence indicating that the delayed toxicity does not extend beyond larval stage.

RESUMEN

Se evaluaron los niveles de resistencia de una raza de pulgas de gato, *Ctenocephalides felis* (Bouche), de la florida, usando nueve productos químicos después de la colonización, por aproximadamente un (1984) y dos (1985) años. Se encontró que las pulgas habían perdido algo de su resistencia a clorpirifos y malatión en 1985 comparado con 1984. También se detectó más resistencia a los tres carbamates, bendiocarb, propoxur, y carbaril. El aumento de la resistencia a los carbamates se atribuyó al uso de polvo de carbaril para matar las pulgas en los gatos después de que se sacaron de la producción de pulgas. Los niveles de propetamfos, diazinón, isofenfos y clorfenvinfos no cambiaron significativamente.

cuando se comparó la toxicidad de 24 h de nueve compuestos contra las etapas larvales y de adultos en 1985, clorpirifos, diazinón, clorfenvinfos, y propoxur, fueron más tóxicos a los adultos que a las larvas, y propetamfos, malatión, y bendiocarb fueron más tóxicos a las larvas que a los adultos. La tolerancia a isofenfos fue similar en larvas y adultos. Ambas etapas respondieron igual al carbaril y fueron completamente resistentes.

Todos los organofosfatos en 1985 tuvieron una reacción demorada en la mortalidad de las larvas en 1985, y afectó la formación del capullo de las larvas que sobrevivieron la exposición de 24 h. Sin embargo, ninguno de los organofosfatos o carbamates parecen tener ningún efecto en la salida de los adultos, lo que indica que la toxicidad demorada no se extiende más allá de la etapa larval.

Insecticide resistance had been reported in the literature as causing control failures in field populations of fleas (Brown 1958, Brown & Pal 1971, Kilpatrick & Fay 1952, McDuffie et al. 1953). Other studies (Collart & Hink 1986, El-Gazzar et al. 1986, Fox et al. 1968, Fox & de Leon 1984, Miller et al. 1977) have attempted to demonstrate the presence or development of insecticide resistance in the cat flea.

In 1984 after 9-12 months of colonization, a Florida strain of the cat flea, *Ctenocephalides felis* (Bouche), was tested for susceptibility to a number of chemicals commonly used for flea control in the U.S. and compared to a California strain which had been colonized for about two decades. The results of this study were reported by El-Gazzar et al. (1986) and clearly showed that the Florida strain exhibited a higher degree of resistance to all nine chemicals compared to the California strain.

The present study was conducted to determine whether the Florida strain had lost any pesticide resistance during colonization after the 1984 evaluation. The second objective of this study was to determine if insecticide susceptibility was the same for both the adult and larval stage of the cat flea.

MATERIALS AND METHODS

Cat fleas were reared at the USDA-ARS Insects Affecting Man and Animals Laboratory in Gainesville, Florida on domestic cats by the method described by El-Gazzar et al. (1986). Each cat was in flea production for 4 wk. During this period of production, 200 adult, mixed-sex fleas were placed on each animal per wk. After 4 wk, the cats were treated with a 5% carbaryl dust to kill fleas and to reduce the irritation and hair loss caused by the fleas. For the next 5 wk, animals not in production were housed in a separate animal building where no insecticides were applied. Insecticide susceptibility of adult fleas was first evaluated in 1984 after 9-12 months of colonization (El-Gazzar et al. 1986). A second evaluation with adult and larval fleas was carried out in 1985 after 21-24 months of colonization.

The insecticides used in this study were bendiocarb, carbaryl, chlorfenvinphos, chlorpyrifos, diazinon, isofenphos, malathion, propetamphos, and propoxur. The chemicals were tested as residues of technical materials dissolved in reagent grade acetone and applied to Whatman No. 1 filter paper. The range of concentrations tested was between 0.1 and 400 $\mu\text{g A.I./cm}^2$. Adult fleas were exposed to chemicals on treated filter paper strips (El-Gazzar et al. 1986). For each concentration, 100 adult fleas (1 to 6 h old) were exposed to treated filter papers in 80 ml test tubes. Mortality was determined after 24 h exposure.

Larvae (100 ca. 3.5 days old) were exposed on filter paper discs 11.0 cm in diameter. The discs were pressed inside 120 ml paper cups so that they had vertical edges to prevent larvae from escaping. Ca. 0.5 g of larval diet was placed on each filter paper disc. Control groups were exposed to filter paper treated only with acetone. After 24 h, numbers of dead and live larvae were counted. Surviving larvae were placed in 250-ml waxed paper cups containing 75 g of larval rearing medium. The medium consisted, by volume, of 5 parts of sand and part of food (20:3:2, by weight, pulverized dry laboratory rat chow, dried blood meal, and Brewer's yeast). The cups were covered with organdy cloth and held at $27 \pm 2^\circ\text{C}$ and $70 \pm 5\%$ RH. After 5 wk, the cups were checked for cocoons and adults. Each test was replicated two to four times.

Percent mortalities of both adults and larvae were corrected for control mortalities by Abbott's formula (1925). Probit analysis (Finney 1971) was applied to the corrected percent mortalities to estimate the lethal concentrations (LC_{50} and LC_{90}). In the larval study, percentage reduction in pupal cocoon formation (based on the number of larvae that survived the 24 h exposure) and adult emergence (based on the number of cocoons) were calculated for all treatments. Probit analysis (Finney 1971) was then applied for estimating the lethal concentrations for cocoon formation (delayed larval mortality) and adult emergence (pupal and unemerged adult mortality).

RESULTS AND DISCUSSIONS

The data on the nine chemicals tested are presented in Table 1. Based on the 1985 LC_{50} values for chlorpyrifos ($0.72 \mu\text{g/cm}^2$) and malathion ($37.72 \mu\text{g/cm}^2$), the Florida strain of fleas, had lost some resistance since 1984, requiring 39 and 38% less chemical, respectively, to achieve the LC_{50} . Meanwhile, this strain of fleas exhibited a greater degree of resistance to bendiocarb, propoxur, and carbaryl than in 1984, requiring 7.7, 2.7, and >11.9 times more material, respectively, to achieve the LC_{50} . The development of higher resistance to carbamates is probably attributable to the use of the 5% carbaryl dust on the cats after each flea production period. The high resistance developed by fleas to bendiocarb and propoxur indicated that there is probably cross-resistance between these two insecticides and carbaryl. The resistance level to diazinon, propetamphos, isofenphos, and chlorfenvinphos did not significantly change.

TABLE 1. LETHAL CONCENTRATIONS OF NINE INSECTICIDES TO CAT FLEA ADULTS IN 1985 COMPARED TO 1984.

Insecticides	<i>n</i>	1985 LC ₅₀ (μg/cm ²) (95% C.I.)	Slope	1985 LC ₅₀ / 1984 LC ₅₀ ^a
Chlorpyrifos	1,020	0.72 (0.60-0.87)	3.09	0.61
Diazinon	550	4.09 (2.39-6.97)	2.94	0.78
Propetamphos	580	6.24 (4.48-8.68)	3.09	0.93
Isofenphos	1,072	9.20 (6.68-12.49)	2.69	1.10
Malathion	669	37.72 (20.99-60.26)	1.74	0.62
Chlorfenvinphos	1,970	48.74 (41.21-56.61)	2.45	1.30
Bendiocarb	1,000	100.73 (31.36->500)	0.34	7.69
Propoxur	1,553	140.48 (23.07-unavail)	0.99	2.69
Carbaryl	1,000	>500 ^b		>11.86

Total number of adults used for each compound ranged between 1200 and 1800.

^a1984 values from El-Gazzar et al. (1986).

^bUnachievable within the range of concentrations applied.

Flea larvae were exposed to the same nine chemicals, and the data are shown in Table 2. The larval stage was more tolerant than the adult stage to four of the chemicals. At the LC₅₀ level, it took 1.7, 1.4, 1.7, and 1.5 times more chlorpyrifos, diazinon, chlorfenvinphos, and propoxur, respectively, to kill larvae than adults. However, of these four chemicals, only chlorpyrifos and diazinon differed significantly as determined by nonoverlap of the 95% C.I. Three compounds, propetamphos, malathion and bendiocarb were more effective against larvae than adults. At the LC₅₀ level, it took 35, 71, and 54% less propetamphos, malathion, and bendiocarb, respectively, to kill larvae compared to adults. Carbaryl was completely ineffective against both larvae and adults.

Of the nine compounds evaluated, all organophosphates, but no carbamates, resulted in delayed larval toxicity, expressed in Table 3 as a significant dose response on cocoon formation of larvae surviving 24 h of exposure. The greatest effect on cocoon formation (delayed larval toxicity) was by chlorpyrifos (LC₅₀ = 3.02 μg./cm²). Diazinon, malathion and chlorfenvinphos required higher concentrations (35.1, 195.4, and 263.8 μg/cm², respectively) to reduce cocoon formation by 50%. None of the chemicals caused delayed pupal toxicity as indicated by no significant dose response in those that survived to pupation.

TABLE 2. LETHAL CONCENTRATIONS OF NINE INSECTICIDES TO CAT FLEA LARVAE AFTER 24 H EXPOSURE.^a

Insecticides	<i>n</i>	LC ₅₀ (μg/cm ²) (95% C.I.)	Slope
Chlorpyrifos	2,080	1.25 (0.96-1.68)	1.09
Propetamphos	1,780	4.02 (2.48-6.09)	1.92
Diazinon	1,400	5.82 (5.01-6.93)	0.71
Malathion	1,900	10.74 (5.57-15.86)	0.74
Isofenphos	1,200	13.15 (10.52-10.98)	1.57
Bendiocarb	1,200	46.39 (35.02-68.74)	0.32
Chlorfenvinphos	1,700	80.65 (unavailable)	1.01
Propoxur	1,200	207.70 (78.93->500)	0.32
Carbaryl	1,200	— ^a	— ^a

^aTotal number of larvae used for each compound ranged between 1200 and 1800.

^bUnachievable within the range of concentrations applied.

TABLE 3. LETHAL CONCENTRATION OF NINE INSECTICIDES FOR CAT FLEA COOCON FORMATION (AS CRITERION FOR DELAYED LARVAL MORTALITY) AFTER 24 H EXPOSURE PERIOD.^a

Insecticides	<i>n</i>	LC ₅₀ (µg/cm ²) (95% C.I.)	Slope
Chlorpyrifos	2,080	3.02 (2.22-5.05)	0.87
Propetamphos	1,780	7.93 (3.03-97.84)	0.95
Isofenphos	1,400	17.81 (12.28-107.11)	1.39
Diazinon	1,900	35.10 (19.44-108.72)	0.43
Malathion	1,200	195.40 (76.19->500)	0.44
Chlorfenvinphos	1,200	263.80 (unavail.)	0.32
Bendiocarb	1,700	— ^b	0.16
Propoxur	1,200	— ^b	0.06
Carbaryl	1,200	— ^b	0.02

^aTotal number of larvae used for each compound ranged between 1200 and 1800.

^bUnachievable within the range of concentrations applied.

The results indicated that of the six organophosphates chlorpyrifos, propetamphos, and isofenphos were the most efficient chemicals against both immature and adult fleas. The results also indicated that cat fleas can develop high levels of resistance to carbamates after relatively short periods of exposure to these chemicals and the use of carbamates should be avoided by researchers raising fleas for experimental purposes.

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