

INSECTICIDE RESISTANCE IN *BLATTELLA GERMANICA* (L.) (DICTYOPTERA: BLATTELLIDAE) FROM FOOD PRODUCING ESTABLISHMENTS IN DENMARK

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Abstract—A number of cases of *Blattella germanica* control failure were reported to the Danish Pest Infestation Laboratory from 1987 to 1991. A screening of the insecticide resistance in *B. germanica* in some selected locations was conducted with permethrin using tarsal contact tests to estimate KT_{50} (/WHO/VBC/75.593). Based on these data more detailed measurement of the resistance in the German Cockroaches from chosen locations was then assessed by topical application techniques; 2.5 μ l insecticide in acetone on the ventral sclerites of the cockroaches was used. Permethrin, deltamethrin, diazinon, and chlorpyrifos were used in these trials. Probit analysis was used with both methods, according to Finney (1971) and similar results obtained with the DPIL susceptible strain were used to calculate the resistance/ susceptibility ratio (R/S).

The cockroaches were mainly collected from mass food producing kitchens, bakeries, and a zoological garden in Copenhagen and North Zealand, but some were also collected in food producing factories in Aarhus, Jutland, and from residential buildings in Nuuk, Greenland.

In the topical application tests, female cockroaches from ten different locations were tested for their susceptibility to permethrin. The calculated R/S ratio based on the LC_{50} 's varied between 1 and 57. For deltamethrin from eight locations, the R/S ratio varied between 2 and 31, for chlorpyrifos from nine locations, the R/S ratio varied between 1 and 4, and for diazinon from six locations, the R/S ratio varied between 1 and 2.

Based on these findings, the reason for control failure in these particular locations could be ascribed to resistance to the two pyrethroids, permethrin and deltamethrin. Both insecticides were in use for cockroach control in Denmark at that time. The organophosphate, chlorpyrifos, with its low R/S ratio was then used in the locations, and with good results.

INTRODUCTION

In Denmark insecticide resistance in the German cockroach *Blattella germanica* became an established problem shortly after dieldrin was introduced for cockroach control in the late fifties. As early as 1962 dieldrin resistance was detected in a Danish cockroach strains with a resistance ratio of approximately 2000 (Keiding, 1963). In the late seventies dieldrin resistance caused so many problems that dieldrin based products could not be used any more. Chlorpyrifos and diazinon were introduced into the market in the late sixties and in the beginning of the seventies as, e.g. locally produced lacquer-formulations. In contrast to the report of chlorpyrifos resistance by other investigators (Milio *et al.*, 1987, Rust & Reiersen, 1991), this type of resistance has not until now caused any major control problems in Denmark although the chlorpyrifos have been and still are often used. In the early eighties, the pyrethroids permethrin and later deltamethrin based spray-formulations became commercially available and seemed to provide new possibilities for cockroach control in Denmark. These products were much easier to apply than the older lacquer-formulations, and they quickly obtained a widespread use by professional pest control operators.

A few years after the above-mentioned pyrethroids had been introduced for cockroach control, control failures were observed, a situation nearly parallel to the control failure with cypermethrin as described by Atkinson *et al.* (1991). In 1987 the Danish Pest Infestation Laboratory was contacted by a municipal pest control organization which had encountered control failure when using permethrin in mass food producing hospital kitchens. Based on some preliminary tarsal contact test, the present study was initiated with the objectives of characterizing the resistance profile of these and other field collected strains to the insecticides available for cockroach control in Denmark.

METHODS AND MATERIALS

The field strains used in this study were collected from mass food producing kitchens, bakeries, food and chocolate producing factories, a zoological garden, and a residential building (Nuuk,

Greenland). Apart from the zoological garden, only locations with a recognized control problem participated in the survey. The DPIL sus-strain was used as the susceptible reference-strain. The strain has been reared at DPIL since the fifties and is susceptible to a wide variety of insecticides (Jensen, 1987). All strains were reared at 27°C at 50% RH with a 12:12 (L:D) photo period on a diet of rat chow (Altromin no.1324) and water.

Tarsal contact test (WHO/VBC/75.593) was used to identify a potential permethrin resistance, and if a resistance problem was identified then new cockroaches were collected and the strain was reared in the laboratory to provide cockroaches enough for a topical application test. In the tarsal contact test, permethrin (Wellcome Res.lab.(UK)) diluted in acetone was used. 2.5 ml of the permethrin solution was applied to the inside of a 250 ml jar to produce a uniform surface residue of 20 µg/cm permethrin on the sides and bottom. No more than ten female cockroaches were released into a jar and the knock-down was registered with a given interval dependent on the strain and insecticide used. With the PI890211 strain males were only tested to spare the females, and with the two strains Ni881011 and Nu890711 no tarsal contact test was conducted.

In the topical application tests 2.5 µl of the insecticide solutions in acetone were applied to the ventral sclerites of CO₂ anaesthetized female cockroaches. The insecticides used were: permethrin (Wellcome Res.lab.(UK)), deltamethrin (Roussel-Uclaf), chlorpyrifos (Dow Chemicals), and diazinon (Ciba-Geigy). The doses used were found in a preliminary range finding test. Each concentration was replicated two or three times with upto 10 cockroaches from each strain dependent on the number of cockroaches available for the test. Treated cockroaches were transferred to clean jars and provided with water and food. It has been found that some recovery after treatment with synthetic pyrethroids is common, therefore, mortality was recorded after 48 hours instead of 24 hours as normally done.

Table 1. Response of field and laboratory strains of *B. germanica* females when exposed to 20 µg/cm² permethrin in glass jars.

Insecticide	N	Strain*	KT ₅₀	95% fiducial L.	HR		Locality	I.u.
					%	min.		
Permethrin	40	DPIL-sus	13	12 - 14	100	30	Laboratory	-
	20	Bi870911	!	-	55	1440	Hospital	P
	20	Ko871011	!	-	10	4320	Hospital	P
		NI881011	not tested				Factory	P/D
	20	Ho881115	!	-	50	1440	Bakery	P
	18	M PI890211	186	154 - 223	100	1440	Restaurant	not known
	20	Zo890611	17	15 - 20	100	35	Zool. garden	C
	6	Ni881111	39	32 - 48	100	80	Bakery	P
	20	Su880911	!	-	20	1440	Hospital	P
		NU890711	not tested				Res.buildings	not known
	20	Ar890411	!	-	0	5760	Restaurant	P

* The year in which the strain was collected is given by the two first digits after the letter code.

N= Number of *B. germanica* tested.

HR= Highest response given as per cent knock-down after a given time in minutes.

M= Males tested.

!= Probit analysis not possible

I.u.= Insecticide used in the nearest past as reported by the pest control operators.

P= Permethrin

D= Deltamethrin

C= Chlorpyrifos

Table 2. Dose-response data (48-h knock-down) for laboratory and field strains of female *Blattella germanica*. Tested against four topically applied insecticides. 2.5 µl of insecticide in acetone was applied to the ventral sclerites of the cockroach.

Insecticide	N	Strain	LC50 %w/v	95% fiducial limits	Slope	± s.e.	chi ²	d.f.	P	R/S ₅₀
Permethrin	236	DPIL-sus	0.013	0.012-0.015	4.63	0.49	6.8	4	0.15	-
	158	Bi870911	0.538	0.453-0.640	3.17	0.46	7.7	6	0.27	41
	161	Ko871011	0.522	0.408-0.669	2.06	0.37	1.7	6	0.95	40
	220	Ná881011	0.747	0.655-0.851	4.22	0.47	16.0*	5	0.01	57
	128	Hø881115	0.465	0.386-0.559	3.26	0.61	6.1	5	0.30	36
	115	PL890211	0.272	0.198-0.374	3.01	0.76	0.1	4	0.99	21
	109	Zo890611	0.018	0.016-0.021	5.91	1.01	9.4 *	2	0.01	1
	83	Ni881111	0.527	0.448-0.621	4.65	1.05	19.9 *	3	0.01	41
	140	Su880911	0.577	0.501-0.664	4.46	0.68	4.8	5	0.45	44
	80	Nu890711	0.040	0.032-0.051	3.58	0.70	2.1	2	0.37	3
150	Ar890411	0.752	0.606-0.932	2.70	0.45	14.6 *	5	0.02	57	
Deltamethrin	121	DPIL-sus	0.00081	0.0007-0.009	6.32	1.12	5.4	4	0.27	-
	100	Bi870911	0.017	0.013-0.021	3.09	0.62	1.2	3	0.75	21
	140	Ko871011	0.024	0.018-0.033	1.95	0.34	10.8	5	0.08	30
	140	Ná881011	0.016	0.013-0.019	3.56	0.60	8.7	5	0.15	20
	80	Hø881115	0.018	0.016-0.020	6.50	1.19	16.6 *	2	0.01	22
	120	PL890211	0.011	0.009-0.015	2.7	0.54	7.4	4	0.11	14
	140	Zo890611	0.001	0.001-0.002	3.23	0.48	1.8	5	0.88	2
	120	Ni881111	0.019	0.016-0.022	4.75	0.75	19.9 *	4	0.01	23
	140	Su880911	0.025	0.020-0.031	2.83	0.42	15.6 *	5	0.01	31
Chlorpyrifos	100	DPIL-sus	0.023	0.019-0.027	4.33	0.78	1.6	3	0.65	-
	119	Bi870911	0.041	0.035-0.048	4.19	0.64	13.6 *	4	0.01	2
	116	Ko871011	0.040	0.034-0.047	4.59	0.72	4.1	4	0.40	2
	132	Ná881011	0.031	0.027-0.035	5.66	0.81	6.6	5	0.24	1
	79	Hø881115	0.038	0.034-0.043	8.55	1.6	0.4	2	0.82	2
	140	PL890211	0.015	0.011-0.021	2.85	0.57	6.3	5	0.28	1
	122	Zo890611	0.029	0.026-0.033	6.00	0.91	1.4	4	0.87	1
	142	Ni881111	0.070	0.063-0.079	5.59	0.83	5.0	3	0.14	3
	170	Su880911	0.096	0.076-0.120	2.33	0.36	15.0 *	6	0.02	4
	140	Nu890711	0.022	0.018-0.028	3.08	0.52	7.2	5	0.20	1
Diazinon	150	DPIL-sus	0.016	0.014-0.018	4.86	0.56	3.1	4	0.55	-
	90	Bi870911	0.028	0.024-0.034	4.78	0.79	0.89	3	0.85	2
	100	Ná881011	0.027	0.022-0.033	3.62	0.65	5.5	3	0.14	2
	122	Hø881115	0.038	0.032-0.046	3.60	0.61	6.6	4	0.17	2
	180	PL890211	0.017	0.014-0.020	3.56	0.44	9.6	7	0.22	1
	139	Zo 890611	0.017	0.015-0.020	4.13	0.57	11.5 *	5	0.05	1
119	Ni881111	0.039	0.031-0.049	2.97	0.56	1.1	4	0.90	2	

* indicates heterogeneity.

N= number of tested cockroaches.

d.f.= degree of freedom.

P= goodness of fit.

R/S₅₀= resistance factor

Data was analysed using probit analysis according to Finney (1971). The LC₅₀'s were estimated for each strain tested against each insecticide. Goodness of fit was estimated by extrapolating between tabulated values. Test for statistical significance between LC₅₀'s was the failure of their 95% fiducial limits to overlap. The resistance ratios (R/S) was calculated as LC₅₀ resistant strain divided by LC₅₀ susceptible strain. R/S was considered significantly different from one when based on LC₅₀'s that was significantly different.

RESULTS

The results of the tarsal contact test are given in Table 1. Apart from the DPIL-sus strain, only strain Zo890611 turned out to be susceptible to permethrin, and all in all only three of the eight field

