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PJBS

ISSN 1028-8880

Pakistan Journal of Biological Sciences

ANSInet
Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

Pakistan Journal of Biological Sciences 6 (9): 821-825, 2003
ISSN 1028-8880

Effect of Agricultural Chemicals on Reptiles: Comparison of Pyrethroid and Organophosphate with Phytopesticide on Cholinesterase Activity

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Abstract: Experiments were carried out to find the effect of three pesticides cypermethrin (pyrethroid), malathion (organophosphate) and biosal (phytopesticide) on wildlife species of reptile *Calotes versicolor*. Two different concentrations i.e. 0.1 and 1% for cypermethrin and malathion and 25 and 50% for biosal were used. It was observed that cholinesterase activity decreased upto 27 and 54% in kidney and 20 and 35% in liver under the effect of cypermethrin. Under the effect of malathion cholinesterase decreased upto 58.46 and 65.09% in kidney and 30.27 and 66.97% in liver while under treatment of biosal cholinesterase activity decreased upto 13.06 and 18% in kidney and 39.52 and 52.61% in liver, respectively. The results are compared with those reported in the literature todate.

Key words: Pyrethroid, organophosphate, phytopesticide, cholinesterase

Introduction

Pesticides can change almost all aspects of both natural and managed ecosystems, such as agroecosystems, patterns of energy flow (via effects on food chains) nutrient cycling species diversity interspecies interactions soil, water and air quality. Pesticides destroy and reduce the diversity and abundance of biodiversity with cascading effects at higher tropic levels. Threats to biodiversity are posed by deforestation, overgrazing, soil erosion, rampant hunting and fishing and agricultural practices. A number of wildlife species can be affected when agricultural pesticides are used because of their effect on physiological function. Cholinesterase occures in a number of animals and reduction can result in sublethal toxicity and death (Cooper, 1991). Many amphibian and reptile populations are presently in decline. Environmental contaminants have been implicated as a possible cause of some of these declines. There has been a great deal of interest in reptile and amphibian ecotoxicology (Pauli *et al*., 2000). Some work have been reported about reduction of cholinesterase activity and protein contents on reptiles and amphibians (Balasundaram and Selvarajan, 1990; Berril *et al*., 1993; Khan, 2002; Khan *et al.,* 2002a; Khan *et al*., 2002 b; Khan *et al*., 2003a; and Khan *et al*., 2003b). Some work has been done on the effects of pesticides on amphibians and reptiles by Ascher, 1981 and 1993, Bradbury and Coats, 1982; Hill and Fleming, 1982; Grue *et al*., 1983; Henny *et al*., 1987; Delany *et al*., 1988; Hill, 1989; Clark *et al.,* 1990 and 1995; Cooper, 1991; Mineau, 1993; Burn and Leighton, 1996; Garcelon and Thomas, 1997; Kegley *et al.,* 1999; Larson *et al*., 1998; O'Hara *et al.,* 1999; Bishop *at el*., 2000; Fatima, 2002; Khan and Fatima, 2002; Khan *et al*., 2002a; Khan *et al*., 2002b and Khan *et al*., 2003a and b reported effect of phytopesticide and pesticides on different non-target species of wildlife. In the present investigation the comparison of cypermethrin and malathion with biosal on cholinesterase of wildlife species of *Calotes versicolor* were studied.

Material and Methods

The experimental work was carried out on the wildlife species of *Calotes versicolor* Two different concentrations i.e. 0.1 and 1% for cypermethrin and malathion and 25 and 50% for biosal were used . The cypermethrin, malathion and biosal were injected one µl per lizards. A batch of untreated (Lab standard) was also kept for comparison. After 24 hours of treatment, lizard kidney and liver were taken out as per Shakoori and Ahmad's (1973) techniques for cholinesterase estimation. Cholinesterase activity was estimated by Randox Kit No. CE-190. This method is based on Knedel and Boetteger (1967).

Results

Under the effect of cypermethrin cholinesterase was decrease decreased upto 27 and 54% in kidney (Table 1) and 20 and 35% in liver (Table 2). Under the effect of malathion cholinesterase decreased upto 58.46 and 65.09% in kidney (Table 3) and 30.27 and 66.97% in liver (Table 4) while under treatment of biosal cholinesterase activity decreased upto 13.06 and 18% in kidney (Table 5) and 39.52 and 52.61% in liver (Table 6), respectively.

Discussion

Agricultural pesticide use in lesser developed countries

Table 1: Activity of cholinesterase in kidney of *calotes versicolor* treated with cypermethrin

	Time (sec.)	Mean (U/l)	Mean×11730	S.D. (±)	S.E. (±)	Range at 95% confidence limit	%Inhibition
	00	0.00	00.00	0.00	0.00	0.00	
Control	30	0.36	4269.72	0.00	0.00	0.36204-0.36596	
	60						00%
	90						
	00	0.00	00.00	00.00	00.00	0.00	
0.1%	30	0.26	3061.53	0.00	0.00	0.25748-0.264528	27%
	60	0.26	3120.18	0.02	0.01	0.239932-0.292068	
	90	0.25	3043.93	0.02	0.01	0.04704-0.23348	
	00	0	00.00	00.00	00.00	0.00	
1%	30	0.16	1911.99	0.00	0.00	0.154376-0.171624	54%
	60	0.16	1923.72	0.01	0.00	0.15028-0.17772	
	90	0.16	1888.53	0.27	0.15	0.15064-0.47264	

Table 2: Activity of cholinesterase in liver of *calotes versicolor* treated with cypermethrin

	Time (sec.)	Mean (U/l)	Mean×11730	S.D. (±)	S.E. (±)	Range at 95% confidence limit	%Inhibition
	00	0.00	00.00	0.00	0.00	0.00	
Control	30	0.48	5677.32	0.02	0.01	0.4588-0.5092	
	60	0.49	5747.71	0.02	0.01	0.4652-0.5148	00%
	90	0.48	5700.78	0.03	0.01	0.44974-0.522226	
	00	0.00	00.00	0.00	0.00	0.00	
0.1%	30	0.38	4492.59	0.04	0.02	0.33675-0.42925	
	60	0.39	4586.43	0.00	0.00	0.38316-0.39884	20%
	90	0.38	4473.04	0.02	0.01	0.35846-0.40354	
	00	0.00	00.00	0.00	0.00	0.00	
1%	30	0.31	3683.22	0.03	0.01	0.27670-0.35124	
	60	0.31	3683.22	0.00	0.04	0.2356-0.3924	35%
	90	0.31	3671.49	0.01	0.00	0.30124-0.32476	

Table 3: Activity of cholinesterase in kidney of *calotes versicolor*, treated with malathion

	Time (Sec.)	Mean sample	Reagent blank Sample x 131.6 =	S.D.	S.E.	Range	% Inhibition
Untreated	00	0.361	13.8180	0.00251	0.00145	0.3581-0.3638	00.00%
	30	0.364	13.9496	0.00173	0.00100	0.3544-0.3659	
	60	0.365	14.0812	0.00360	0.00208	0.3609-0.3690	
	90	0.367	13.2916	0.00378	0.00218	0.3627-0.3712	
Treated (0.1%)	00	0.286	5.6588	0.00264	0.00152	0.283-0.288	58.46%
	30	0.289	7.2380	0.00115	0.00066	0.287-0.290	
	60	0.295	6.3168	0.00472	0.00273	0.289-0.300	
	90	0.301	6.9748	0.00200	0.00115	0.298-0.303	
Treated (1%)	00	0.270	4.8692	0.00754	0.00436	0.2614-0.2785	65.09%
	30	0.273	4.8692	0.00854	0.00493	0.2633-0.2834	
	60	0.275	5.0008	0.00971	0.00561	0.2640-0.2859	
	90	0.280	5.3956	0.00960	0.00555	0.2691-0.2908	

Table 4: Activity of cholinesterase in liver of agama lizard *calotes versicolor*, treated with malathion

	Time (Sec.)	Mean sample	Reagent blank Sample x 131.6 =	S.D.	S.E.	Range	% Inhibition
Untreated	00	0.483	28.5572	0.00208	0.00120	0.4806-0.4853	00.00%
	30	0.485	28.6888	0.00208	0.00120	0.4826-0.4873	
	60	0.485	28.2940	0.00251	0.00145	0.4821-0.4878	
	90	0.488	28.4256	0.00152	0.00088	0.4862-0.4897	
Treated (0.1%)	00	0.378	20.1348	0.00208	0.00120	0.4807-0.4853	30.27%
	30	0.381	20.0032	0.00251	0.00145	0.4878-0.482	
	60	0.387	20.5296	0.00568	0.00328	0.478-0.491	
	90	0.391	20.6612	0.00854	0.00493	0.480-0.499	
Treated (1%)	00	0.337	9.0804	0.00503	0.00290	0.331-0.343	66.97%
	30	0.342	9.4752	0.00776	0.00448	0.333-0.3507	
	60	0.346	9.2120	0.00665	0.00384	0.338-0.353	
	90	0.349	9.3436	0.00608	0.00351	0.3421-0.3558	

Table 5: Activity of cholinesterase in kidney of *calotes versicolor* treated with biosal

Hours	Treatment	Time sec.	Mean (U/I)	S.D ±	S.E ±	Range at 95% confidence limit	% Inhibition
24	Untreated Control	30	2920.77	0.022	0.012	2920.74-2920.79	
		60	2897.31	0.015	0.0091	2897.29-2897.32	
		90	2873.85	0.005	0.003	2873.84-2873.85	-
24	Treated 25%	30	2616.01	0.015	0.009	2615.99-2616.02	
		60	2557.35	0.002	0.001	2557.34-2557.35	13.06
		90	2498.70	0.004	0.002	2498.69-2498.70	
24	Treated 50%	30	3460.64	0.025	0.041	3460.61-3460.66	
		60	3448.91	0.015	0.009	3448.89-3448.92	
		90	3425.45	1.001	1.001	3423.48-3427.41	18

Table 6: Activity of cholinesterase in liver of *Calotes versicolor* treated with biosal

Hours	Treatment	Time sec.	Mean (U/I)	S.D ±	S.E ±	Range at 95% confidence limit	% Inhibition
24	Untreated Control	30	4504.32	0.03	0.017	4504.28-4504.35	
		60	4480.86	0.04	0.02	4480.82-4480.89	
		90	4433.94	0.05	0.03	4433.88-4433.99	-
24	Treated	30	2709.86	0.516	0.298	2709.27-2710.44	
	25%	60	2709.86	0.022	0.012	2709.83-2709.88	
		90	2698.13	0.022	0.012	2698.10-2618.15	39.52
24	Treated	30	2134.86	0.025	0.014	2134.83-2134.88	
	50%	60	2123.13	0.01	0.005	2123.12-2123.13	
		90	2111.58	0.02	0.015	2111.55-2111.60	52.61

also has increased because they are increasingly growing fruits and vegetables for sale to more developed countries, but these pesticides has produced some harmful effects to non-target wildlife including reptiles and amphibians. In the present study it was observed that the activity of cholinesterase of kidney and decrease after cypermethrin, malathion and biosal treatment. Mineau (1993) reported that post exposure of carbamat and organophosphate cholinesterase activity reduced in wild birds. Shakoori *et al.* (1995) reported 84% inhibition in *Tribolium castaneum* due to sublethal doses of cypermethrin. Gard and Hooper, 1995 reported the organophosphorus and carbamate exert their effects by binding to and inhibiting acetylcholinesterase enzyme at nerve synapses. Azmi *at el.* (1999) studied the effect of tetranortriterpenoids (neem product SDS) and deltamethrin (pyrethroid) on phosphomonoesterase activity in *Cyprinus carpio* (common carp) and reported that enzyme inhibition under the effect of these pesticides. Burgees *et al.* (1999) observed that organophosphate insecticide decreased cholinesterase activity in birds. Parson *et al.* (2000) observed the effect of organophosphate and carbamate on non-target wild animals, these pesticides inhibited cholinesterase activity. Khan (2002) studied the effect of permethrin and biosal in Indian Garden Lizard and reported that after the treatment with permethrin cholinesterase activity was decreased upto 17 and 19% in kidney and 18 and 24% in liver respectively, in the case of biosal treatment, decrease of cholinesterase was found as 13.06 and 18% in kidney and 39.52 and 56.21% in liver respectively. In the present finding also, it was observed that cypermethrin caused upto 54%, malathion upto 65.09% and biosal upto 24% reduction in the activity of cholinesterase. The present findings are generally in accordance with the earlier reports. In the present work cypermethrin, malathion and biosal reduced cholinesterase activity of kidney and liver of *C. versicolor.* On the basis of the present studies it is concluded that agricultural pesticides malathion is most toxic among the pesticides tested presently. The phytopesticide biosal could be a better pesticide if used at lower doses.

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