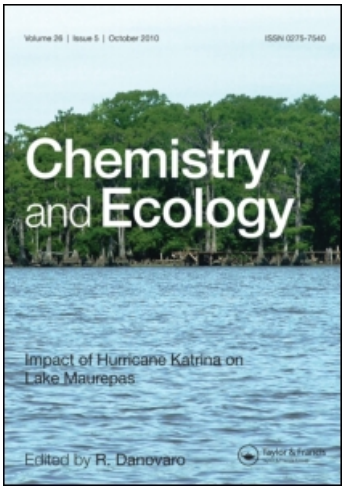

Chemistry and Ecology
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713455114

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To cite this Article Yellamma, K. , Ayyanna, K. and Reddy, A. T. V.(1995) 'Modulations in Protein Metabolism of *Oziotelphusa Senex Senex* During Cypermethrin Intoxication', Chemistry and Ecology, 11: 2, 113 — 116
To link to this Article: DOI: 10.1080/02757549508037691
URL: http://dx.doi.org/10.1080/02757549508037691

Chemistry and Ecology, 1995, Vol. 11, pp. 113–116
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MODULATIONS IN PROTEIN METABOLISM OF *OZIOTELPHUSA SENEX SENEX* DURING CYPERMETHRIN INTOXICATION

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(Received 4 November 1994; final form 6 March 1995)

Cypermethrin induced significant alterations in protein metabolic profiles in the central nervous system (CNS) and pedipalpal muscle (PM) of crab, *Oziotelphusa senex senex*, following ambient exposure. While total and soluble proteins decreased in CNS and PM, free amino acids, protease, alanine and aspartate aminotransferase (AlAT and AAT) were elevated at 24 h after exposure. These results thus confirm the prevailing protein hydrolysis and transamination in these tissues of crab, as a consequence of cypermethrin intoxication. Restoration of normalcy by 48 h demonstrates the importance of these metabolic events in counteracting the effects of cypermethrin. The results also suggests the safer utilization potential and ecological compatibility of cypermethrin.

KEY WORDS: *Oziotelphusa senex senex* (crab), cypermethrin, protein metabolism.

INTRODUCTION

In recent years, indiscriminate application of insecticides to agricultural crops has resulted in deleterious effects not only on the target pests, but also on non-target animals including man (Hill, 1985). In this context synthetic pyrethroids, analogues of natural pyrethrins, have gained prominence in the pesticide market due to their rapid degradability in physical as well as biological environments. But widespread and unscientific usage of these chemicals was found to affect several non-target animals, like crabs, inhabiting the rice field ecosystem. Hence in the present study, the extent of alterations induced by cypermethrin in various segments of protein metabolism and the possible duration for restoration of normalcy has been investigated, thereby suggesting safer application of cypermethrin in agriculture.

MATERIALS AND METHODS

Adult male crabs, *Oziotelphusa senex senex*, weighing 30 ± 2 g, were collected from local paddy fields and freshwater ponds and were acclimated to laboratory conditions for two weeks prior to experimentation. They were fed with pieces of meat *ad libitum* during this period.

Technical grade cypermethrin (90%), obtained from Bharat pulverizing mills, Bombay, India, was used as the toxicant. The crabs were divided into batches of 10 and were exposed to the ambient medium with different concentrations of cypermethrin dissolved in acetone for 48 h. Mortality at each concentration was recorded and

LC_{50} was derived by probit analysis (Finney, 1971). The LC_{50} calculated by this method was found to be 2ppm, and a level, a quarter of this (0.5 ppm), was chosen as a sublethal concentration.

Crabs, after division into 5 batches, were exposed to this sublethal concentration of cypermethrin for observation at 3, 6, 12, 24 and 48 h respectively. A further batch was maintained as a control. Total and soluble proteins (Lowry *et al.*, 1951), protease, free amino acids (Moore and Stein, 1954), alanine and aspartate amino transferase (Reitman and Frankel, 1957) were estimated in the central nervous system (CNS) and pedipalpal muscle (PM) of control and experimental (cypermethrin exposed) animals. The results were also analysed statistically for significance (Pillai and Sinha, 1968).

RESULTS AND DISCUSSION

Significant changes in protein metabolism in the central nervous system (CNS) and pedipalpal muscle (PM) of crab were observed during ambient exposure to a sublethal concentration of cypermethrin (Table I). While the levels of total and soluble proteins were decreased, an increase in free amino acid content, protease, alanine aminotransferase (AIAT) and aspartate aminotransferase (AAT) activity was recorded. Though the trend was the same for both CNS and PM, the changes were a little more evident in the case of CNS. While a peak decrease in total and soluble proteins, and maximum increase in free amino acids and protease activity was noticed at 12 h, this occurred at 24 h in the case of aminotransferases (AAT and AIAT) in both CNS and PM. However, all measured components showed a tendency towards recovery by 48 h.

The decrease in total and soluble proteins in the CNS and PM of crab could be attributed to increased proteolysis as a consequence of cypermethrin toxicity. Consistent with this assumption, an increase in free amino acid content was noticed in the present study. Moreover, the protein hydrolysing enzyme, protease, was also elevated, providing an increased hydrolysis of proteins. The most conspicuous aspect we have noticed in the present study was that all the events (proteolysis, transamination) culminating in the production of free amino acids (FAA), increased up to 12 h. Thereafter, a gradual decline in free amino acid levels was evident and by 48 h, they returned to the control level. The surplus of amino acids at 12 h and their subsequent decrease in later periods could be explained in terms of their role in restoring the physiological homeostatic mechanisms, as these amino acids may not only act as precursors for the synthesis of essential proteins, but also contribute towards gluconeogenesis, glycogenesis and also ketoacid synthesis (Swami *et al.*, 1983; Mayes, 1988). Since free amino acids are known to act as osmotic and ionic agents (Jurss, 1980) in establishing/maintaining ionic equilibrium between external and internal media, an increase in FAA levels in the present study could be explained.

At this juncture, the role of aminotransferases (AAT and AlAT) in restoring the metabolic harmony among various metabolic processes could be evident from the time of their peak elevation. Unlike proteins, free amino acids and protease (12 h), maximum increase in aminotransferases was found at 24 h. In the present study, the excess amino acids made available at 12 h might have been mobilized through increased transamination to restore the normal state in cypermethrin intoxicated crabs, as aminotransferases are known to act at key points of metabolism, as they supply keto acids to Kreb's cycle and also contribute towards fatty acid synthesis.

Table I Changes in protein metabolic profiles in the central nervous system (CNS) and pedipalpal muscle (PM) of crab, *Oziotelphusa senex senex*, during cypermethrin exposure (mean ± SD).

			Time elapsed after exposure				
Parameter	*Tissue*	*Control*	*3h*	*6h*	*12h*	*24h*	*48h*
	CNS	105.27	100.36	93.72	83.37	89.76	101.12*
		±5.09	±4.59	±5.17	±3.72	±4.87	±3.44
Total proteins			(−4.66)	(−10.97)	(−20.88)	(−14.73)	(−3.94)
(mg/g wet wt.)	PM	225.52	220.04*	204.60	180.80	198.94	218.33*
		±10.71	±14.52	±11.16	±13.78	±10.84	±12.16
			(−2.42)	(−9.27)	(−19.82)	(−11.78)	(−3.18)
	CNS	58.57	55.41*	51.85	41.94	48.26	54.28*
		±3.22	±4.42	±2.46	±3.78	±3.57	±2.33
Soluble proteins			(−5.39)	(−11.47)	(−28.39)	(−17.60)	(−7.32)
(mg/g wet wt.)	PM	105.05	102.27	95.72	81.80	92.47	104.30
		±6.04	±7.13	±3.64	±4.38	±4.48	±6.61
			(−2.64)	(−8.88)	(−22.13)	(−11.97)	(−0.71)
	CNS	103.81	105.35	112.85	124.84	118.89	103.80
		±4.79	±6.24	±6.35	±5.04	±9.43	±7.27
Free amino acids			(+1.48)	(+8.70)	(+20.25)	(+14.52)	(+0.1)
(μmoles of							
tyrosine g wet wt.)	PM	216.57	220.03	235.16	266.57	239.34	225.77
		±13.21	±11.29	±8.38	±12.72	±14.61	±13.44
			(+1.59)	(+8.58)	(+23.03)	(+10.51)	(+4.24)
	CNS	0.045	0.046*	0.050	0.061	0.051	0.046
		±0.002	±0.003	±0.003	±0.004	±0.003	±0.002
Protease			(+2.22)	(+11.11)	(+35.55)	(+13.33)	(+2.22)
(μmoles of							
tyrosine/mg	PM	0.104	0.109	0.114	0.135	0.124	0.107
protein/h)		±0.005	±0.003	±0.005	±0.007	±0.004	±0.003
			(+3.84)	(+9.61)	(+29.80)	(+19.23)	(+2.88)
	CNS	2.76	2.84*	3.06	3.59	3.77	2.88
		±0.14	±0.11	±0.13	±0.26	±0.23	±0.11
			(+2.89)	(+10.86)	(+30.07)	(+36.59)	(+4.34)
AlAT							
(μmoles of							
pyruvate/mg	PM	4.24	4.40	4.70	5.25	5.56	4.48*
protein/h)		±0.21	±0.39	±0.37	±0.24	±0.45	±0.36
			(+3.77)	(+10.84)	(+23.82)	(+31.13)	(+5.66)
	CNS	2.25	2.63	2.82	3.27	3.42	2.61
		±0.08	±0.08	±0.11	(±0.11)	±0.26	±0.22
			(+3.13)	(+10.58)	(+28.23)	(+34.17)	(+2.35)
AAT							
(μmoles of							
pyruvate/mg	PM	3.48	3.56*	3.75	4.11	4.51	3.61
protein/h)		±0.12	±0.18	±0.31	±0.34	±0.22	±0.15
			(+2.29)	(+7.75)	(+18.10)	(+29.59)	(+3.73)

Each value is mean ± S.D. of six individual observations
For each value, tissue from six animals was pooled.
Values in parentheses denote percent change from control.
Values are significant at $P < 0.05$ *Not significant

CONCLUSIONS

It may be concluded from the above results that cypermethrin-induced alterations in protein metabolism were restored to normalcy within 48 h duration. From an ecological point of view, also, these pyrethroid compounds thus may not pose long-lasting toxic effects on non-target organisms like crab. It is further reported that

cypermethrin is readily degradable by microorganisms in various soil types within 2–4 weeks by hydrolysis of ester bonds, thus showing no tendency to accumulate in soils (Elliott *et al.*, 1973, Mikami *et al.*, 1984). Hence it is suggested that application of cypermethrin in agricultural fields is relatively safer in the environment when compared to other synthetic chemical pesticides like organophosphates and organochlorides.

ACKNOWLEDGEMENTS

Authors are grateful to UGC, New Delhi, for financial assistance through a research project awarded to K.Y.

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