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Research Paper

ACUTE TOXICITY OF ALPHA-CYPERMETHRIN TO OLIGOCHAETE WORM, BRANCHIURA SOWERBYI (BEDDARD, 1982) ALONG WITH THEIR BEHAVIOURAL RESPONSES.

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Acute toxicity of alpha-cypermethrin to Branchiura sowerbyi and their behavioural changes were evaluated in the present study. The 24, 48, 72 and 96 h LC50 values of alpha-cypermethrin to B. sowerbyi were 43.39, 39.75, 22.58 and 13.41 µg/l respectively. A significant variation (p<0.05) between mortality rate of worm and concentrations of the test chemical at all the exposure times (24, 48, 72 and 96 h) was observed. The mortality rate also significantly varies (p<0.05) with the exposure time at all the concentrations except 48h. In addition, a dose dependent change in the behavioural pattern of the worm was recorded. It was directly pro- portional to the increasing concentration of alpha-cypermethrin.

Acute toxicity, alpha-cypermethrin, Branchiura sowerbyi, behavioural

Initially, rough range finding tests were conducted for the test organism to determine the dose range at which mortality occurs (data not shown). The selected test concentrations of alpha-cypermethrin were finally used for the determination of LC50 values for . Ten test organisms (mean length 18.5 ± 6.22*Branchiura sowerbyi*the mm) were used in each replicate. The number of dead organisms was counted at every 24h during the experiment. The dead animals were removed immediately to avoid any organic decomposition. A constant amount of test medium was replaced every 24h by freshwater and the desired quantity of alpha-cypermethrin was immediately added to water to assure a fixed concentra- tion of the toxicant in solution and also to avoid other inter- fering factors affecting the test animals' performance. Water chemical analysis and the bioassays were done following the methods outlined in American Public Health Association (2012). Similar technique was also followed by previous work- ers (Badanthadka & Mehendale, 2005; Mukherjee & Saha,2012). The LC50 values for 24, 48, 72 and 96h along with 95% confidence limits were estimated by a computer program (US EPA, 1999).

The behavioural changes of the exposed worms at each concentration of alpha-cypermethrin were also recorded during the bioassay. The data on percent mortality was subjected to analysis of variance using the computer software provided by the R Devel- opment Core Team (2011) followed by Duncan's multiple range test to determine significant differences among means at different times of exposure and concentrations (Gomez & Gomez,1984).

Results and Discussion

Branchiura The lethal concentration of alpha-cypermethrin to was *B. sowerbyi* is summarized in Table 1. No mortality of*sowerbyi* recorded in control during the experiment. The mortality rate (%) of the test animals was significantly increased (p<0.05) from control with increasing concentration of the test chemical except 0.8 µg/l at all the exposure times (24, 48, 72, 96h). On the other hand, the mortality rate also varied significantly (p<0.05) with the progress of exposure time except 48h at all the concentrations (Table 2).

With the increasing concentration of test chemical and the progress showed different irregular be- haviors *B. sowerbyi* of exposure time, which are shown in Table 3. The control worms were active throughout the test period and showed clumping tendency with normal movement. The clumping tendency of the exposed worms was gradually reduced with increasing concentrations and times of exposure. The worms showed rapid movement at all the higher doses.

مقدمة

Alpha-cypermethrin, an active isomer of cypermethrin, is used to control the common domestic insects and pests belonging to families like Lepidoptera and Coleoptera infesting vegetables, citrus fruits, cotton, rice, etc. (Yordanova et al., 2009). Cypermethrin has been reported to be used against lice invasion in fishery (Das & Mukherjee, 2003). Alpha-cypermethrin is a synthetic pyrethroid which acts as a contact and stomach poison. It is classified as a Schedule 6 poison in the Standard for the Uniform Scheduling of Drugs and Poisons (Sarikaya, 2009). Alpha- cypermethrin acts on pre-synaptic neurons of organisms where it prevents the transmission of nerve impulses through the axon by blocking the passage of sodium ions across the channels in nerve fibers (Yordanova et al., 2009). It is non-systemic in nature and is moderate to highly toxic to diverse aquatic and terrestrial life (Mokry & Hoagland, 1990; Chamber, 1994). Very little information is available on the acute toxicity of alpha-cypermethrin on aquatic invertebrates (Yordanova -et al., 2009). It is highly toxic to fish. The 24 h LC50 values of alpha cypermethrin to silver barb and common mirror were 20.0 and 4.50 µg/l respectively (Grayson et al., 1990). There is no specific report on the lethality of alpha-cypermethrin on aquatic annelids particularly on the benthic ones that play a vital role in detritus food chains. This sprouted the present need to investigate the acute toxic effects of and their behavioural *Branchiura sowerbyi* alpha-cypermethrin to .responses

aterials and MethodsM

Test organism used in the bioassay was the benthic oligochaete (Class: Oligochaeta, Family: Tubificidae).*Branchiura sowerbyi* worm, The test organisms were collected from local non-polluted sources. The worms were allowed to acclimatize in the test wa- ter for 3 days before the experiment.

Commercial grade alpha-cypermethrin (10% w/w, EC) was col- *B.* lected from the local market. Static replacement bioassays with the was conducted in 500 ml Borosil glass beakers each *sowerbyi* containing 250 ml unchlorinated tap water (Temperature 28.3 ± 0.4 °C, pH 7.1 ± 0.2, Free CO2 9.9 ± 0.5 mg/l, Dissolved Oxygen 5.5 ± 0.3 mg/l, Total alkalinity 181 ± 8.6 mg/l as CaCO3, Hardness 119 ± 6.2 mg/l as CaCO3). A set of four beakers were exposed to each concentration of alpha-cypermethrin. Each set of tests was accompanied by four replicates of control.

Stock solution of the test chemical and its dilutions were made following the method of American Public Health Association (2012).

00am ± 0.43	00am ± 0.43	0.8
10bm ± 0.00	10bm ± 0.43	8
20cm ± 0.71	20cm ± 0.71	20
50dm ± 0.50	40dm ± 0.87	40
60emn ± 0.83	50em ± 0.50	60
70fm ± 0.00	70fm ± 0.43	80
80gm ± 0.00	80gm ± 0.50	100
90hm ± 0.00	90hm ± 0.43	120
100im± 0.00	100hm ± 0.43	150

able 3: Impact of alpha-cypermethrin on behavioural re- T (M: movement; CT: clump- ing *Branchiura sowerbyi* sponses of tendency; MS: mucus secretion; WE: wrinkling effect; –: none; +: mild; ++: moderate; +++: strong) exposed to various con*Branchiura sowerbyi* ntrations during different hours of exposure

						se Do (µg/l)	**vaheB aurio pl aramet er**	**ime T of expo sure**
		100	**80**	**60**	**40**	**20**	**8**
+++	+++	++	++	+	-	M	24h
	-	-	++	++	+++	+++	CT
	++	++	+	-	-	-	MS
	+++	+++	+	+	-	-	WE
+++	+++	++	++	+	-	M	48h
	-	-	+	++	+++	+++	CT
	++	++	+	+	-	-	MS
	+++	+++	++	+	-	-	WE
+++	+++	++	++	-	-	M	72h
	-	+	-	++	+++	+++	CT
	++	++	+	-	-	-	MS
	+++	+++	++	+	-	-	WE

The rate of movement was increased gradually with the increasing concentrations and time of exposures. The wrinkling effect and gradu- ally increased at 60, 80, and *B. sowerbyi* mucous secretion of 100 µg/l of alpha-cypermethrin at all the exposure times.

B. In the present study, the 96h LC50 value of alpha-cypermethrin to is 13.41 µg/l. The aquatic invertebrates and fish are most *sowerbyi* vulnerable to alpha-cypermethrin as its rate of metabolism and elimination are much slower in their body (WHO, 1992; Greulich & Pflugmacher, 2003). The 96h LC50 val- ue of cypermethrin *Tilapia nilotica, Cyprinus carpio, Salmo trutta, Salmo gairdneri* for were 2.20, 0.90-1.10, 1.20, 0.50 and *Scardinius erythrophthalmus* and 0.40 µg/l respectively (Sarikaya,2009).

- The present investigation indicates that the 96h LC50 value of alpha is slightly higher than the other aquatic *B. sowerbyi* cypermethrin to -invertebrates and vertebrates. The 48h EC50 value of alpha *Gammarus* was 0.8 µg/l and for *Daphnia magna* cypermethrin for 24h LC50 value was 0.3 µg/l (Yordanova et al.,2009). Yilmaz et *pulex* al. (2004) and Sarikaya (2009) reported that the 96h LC50 value of alpha-cypermethrin for guppy and nile tilapia were 9.43 µg/l and 5.99 µg/l respectively.

may *Branchiura sowerbyi* The LC50 value of alpha-cypermethrin to provide useful data in the determination of safe level for the agricultural effluents before their release to the natural water resources. The evaluation of the toxicity of alpha-cypermethrin is not sufficient for assuring safe level of the receiving water bodies. In addition, potential risk from alpha-cypermethrin metabolites should also be considered to get a more accurate picture in terms of toxicity.

) along with 95% 50able 1: Median lethal concentration (LCT *Branchiura* confidence limits of alpha-cypermethrin to the at different hours of exposure *sowerbyi*

ntration (µg/l)	**inagrot seT sm**
72h	**48h**	**24h**
39.75 (25.62-54.48)	43.39 (28.55-59.14)	Branchiura sowerbyi

***Branchi-* able 2: Mean values (± SD) of mortality rate (%) ofT exposed to different concentrations of alpha- *ura sowerbyi* 96h).cypermethrin at different hours of exposure (24h, 48h, 72h, Mean values within columns indicated by different superscript letters (a-h) and mean values within rows indi- cated by different superscript letters (m-o) are significantly different**

ours of exposureH	**(DMRT at 5% level)**
72	**48**	**24**	**se (µg/l)Do**
00am ± 0.00	00am ± 0.00	0.0

+++	+++	+++	++	+	-	M	96h
	-	-	-	++	++	++	CT
	+++	+++	++	++	+	-	MS
	+++	+++	++	+	+	-	WE

استنتاج

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