live fishes like *C. striatus, A. testudineus* and *H. fossilis,* not much break-through has been achieved in this direction. We also favour and recommend the mono or mixed culture of *Channa* spp., *H. fossilis, A. testudineus* and *C. batrachus* in ponds as well as pens and cages in open waters such as reservoirs, lakes, nallas, rivers and canals. This will help in augmenting fish production of the district as well as enhance economic status of the people.

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Note

Acute toxicity of selected pesticides on the prawn *Penaeus monodon*

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ABSTRACT

Static bioassay tests for acute toxicity of four pesticides on the prawn *Penaeus monodon* in brackishwater medium were conducted. The 96 h LC_{50} values of pesticides such as DDT, BHC(HCH), heptachlor and endosulfan were found to be 2.08, 4.10, 16.25 and 2.90 ppb respectively for prawns of size group 33-46 mm. Admissible safe concentration for DDT, BHC, heptachlor and endosulfan were 0.21, 0.41,1.63, 0.29 ppb respectively. LC_{50} values were also calculated for each time interval and toxicity of the pesticides was compared using toxicity curves. Pesticides were not detected in creek and coastal waters.

Indiscriminate use of highly persistent pesticides in agriculture and public health programmes has resulted in contamination of food environment causing hazards to wild and aquatic life (Matsumura *et al.*, 1972). The run off from agricultural land is one of the main sources of gradual pesticide pollution of aquatic environment. Hence, toxicological studies of these pesticides upon aquatic organisms are very important from the view point of environmental consequences. Toxic effects of organochlorine pesticides on fish have been reported by several workers (Konar, 1973; Rao, 1985; Bhatt, 1991). However, very little work has been done on the toxic levels of these insecticides on invertebrates (Fernandez *et al.*, 1996). The present study has been undertaken to determine the acute toxicity of four organochlorine pesticides on juveniles of *Penaeus monodon* in brackishwater. Further to assess pesticide residues in creek and coastal areas, water samples were collected once from coastal area of Tuticorin of Tamil Nadu and Kandaleru creek of Nellore District of Andhra Pradesh during March-April, 1993 and analysed for pesticides.

Chemicals and glasswares : Stock solutions of DDT (1,1,1 trichloro 2,2 bis p-chlorophenyl ethane) and BHC (1,2,3,4,5,6 hexachloro cyclohexane) were prepared by dissolving known quantities of technical grade pesticide in analytical grade acetone while endosulfan (6,7,8,9,10,10-hexachloro 1,5, 5a,6,9,9a-hexahydro-6,9-methano-2,4,3-benzodioxathiepin-3-oxide) and heptachlor(1,4,5,6,7,8,8-heptachloro 3a,4,7,7a-tetra-hydro-4,7-endo-methanoindene) were dissolved in distilled water to prepare stock solution. A range of test concentrations of each pesticide as indicated by preliminary

toxicity tests were prepared as shown below.

DDT : 0.5, 1.0, 2.0, 3.0, 4.0, 5.0 µg/l
BHC : 2, 3, 4, 5, 6, 7, 8 µg/l
Endosulfan : 0.5, 1.0, 2.0, 3.0, 4.0, 5.0, 6.0, 7.0 µg/l
Heptachlor : 5, 10, 15, 20, 25, 30, 35 µg/l

All pesticides (technical grade) were supplied by Hindustan Insecticide Ltd., Cochin. Pesticide standards were procured from E.Merck.

Rearing of animals : The test animals, *P. monodon* of size group 33-46 mm were collected from CIBA hatchery and were transferred into glass (100 l) aquaria containing clean water of similar temperature. They were acclimatised to laboratory conditions in brackishwater (salinity 15±1.0 ppt) recirculatory system for one week prior to the experiment. Animals were maintained in similar environmental conditions as present in the hatchery. Prawns were fed with boiled egg pieces during acclimatization to avoid cannibalism. Feeding by natural plankton and other food was suspended two days before the experiments.

Experimentation : Static bioassay test was employed with daily water exchange. The prawns were exposed to a wide range of test concentrations for 96 hrs. Duplicates were run for each pesticide. Ten prawns were used in each tank containing 75 l brackishwater with a separate set of controls. Observations for mortality were made every 8 hrs intervals upto 96 hrs of experimentation. The criteria employed to find out mortality was absence of any movement when proded gently with a glass rod. The 96 hr LC_{50} values were calculated using the method of Litchfied and Wilcoxon (1949). The data were subjected to probit analysis as suggested by Finney (1971). The physico-chemical characteristics of water during the experimentations were : water temperature 28±2°C, pH 8.5±0.2, dissolved oxygen 7.0±0.2 mg/l, salinity 15±1 ppt and total alkalinity 143±5 mg/l as $CaCO_3$.

Analysis of organochlorines : Three stations were selected from each site. One litre of composite water sample, which was formed by mixing three samples collected from soil-water interfaces of three different places of each station was stored in a 1.2 litre capacity glass container and was transported to the institute laboratory under standard condition at 4°C. Ice boxes were used to keep the samples at low temperature while transportation and protected against light. The determination of organochlorine pesticides in water samples was done following standard methods (APHA, 1989). One litre of unfiltered water sample was taken in separating funnel and extraction was completed with dichloromethane and hexane (15: 85 v/v). After clear separation of the aqueous and hexane layers, the aqueous layer was discarded and hexane layer was subjected to the demoisturing and clean up process using anhydrous sodium sulphate 60/100 PR grade florisil activated at 675°C, respectively. The cleaned up extract was concentrated by K.D. evaporator and was analysed by Gas Chromatograph (Chemito model 2865) equipped with electron capture detector having Ni^{63} isotope. The detection limit of the BHC and heptachlor is 0.003-0.025 ppb and 0.014-0.065 ppb respectively. The precision for the analysis of organochlorines studied ranged from 0.002 to 0.025 ppb. The glass column used was 6 m in length and had an inner diameter of 1/4", it was packed

TABLE 1. *96 hr LC_{50} values (ppb) with 95% confidence limits, safe concentrations and slopes of the individual regression lines of* P. monodon *for four pesticides*

Pollutant	96 hr LC_{50} value	Slope	95 % confidence limit		Derived safe level
			Lower	Higher	
DDT	2.08	3.0	1.9419	2.2181	0.21
Endosulfan	2.90	3.71	2.815	2.985	0.29
BHC	4.10	1.78	3.8789	4.3211	0.41
Heptachlor	16.25	0.915	15.8533	16.6467	1.62

with 1.5 % OV-17+1.95% OV-210 on 80/100 mesh chromosorb WHP. The analysis was recorded by the ORACLE-2 software programme. Pure and dry nitrogen gas was used as a carrier gas. The flow rate of the N_2 gas was maintained at 60 ml/min. The injector, column and detector (ECD-Ni^{63}) temperatures were maintained at 220, 200, 275°C respectively.

The 96 hr LC_{50} values with 95 % confidence limits, safe concentrations and slopes of individual regression lines of *P. mondon* for all the four pesticides are presented in Table 1. The 96 hr LC_{50} values for DDT, endosulfan, BHC and heptachlor were found to be 2.08, 2.90, 4.10 and 16.25 ppb respectively. Using the safe concentration factor of 0.1 (Murty, 1986), the allowable safe concentration of DDT, endosulfan, BHC and heptachlor for *P. monodon* are 0.208, 0.29, 0.41 and 1.62 ppb respectively.

The 96 hr LC_{50} values of BHC have been determined by various workers. Fernandez *et al.* (1996) reported a 96 hr LC_{50} value of 12.5 ppb for prawn *Macrobrachium idella idella.* The present value of 96 hr LC_{50} value of 4.1 ppb for *P. monodon* is very low compared to the value of *Macro-brachium.*

LC_{50} values were also calculated for different time intervals and are given in Table 2. Toxicity of four pesticides studied were compared using toxicity curves obtained by plotting changes in LC_{50} values with respect to different exposure times (Fig. 1). Significant differences in the toxicity were observed when different pesticides were employed. In general, the increase in percentage mortality was related both to time and pesticide concentrations.

TABLE 2. *LC_{50} values (ppb) of pesticides for* P. monodon *for different exposure periods and their concentrations in Kandaleru creek and Tuticorin coastal waters*

Pollutant	LC_{50} values			Water samples		
	24 hrs	48 hrs	72 hrs	96 hrs creek water (April 1993)	Kandaleru coastal water (March 1993)	Tuticorin
DDT	4.5	3.6	2.8	2.08	BDL	BDL
Endosulfan	6.0	5.0	4.1	2.9	BDL	BDL
BHC	7.0	6.0	5.1	4.1	BDL	BDL
Heptachlor	30	26	22	16.25	BDL	BDL

BDL - Below detection level

Fig. 1. LC_{50} values of pesticides for *P. monodon* for different exposure periods.

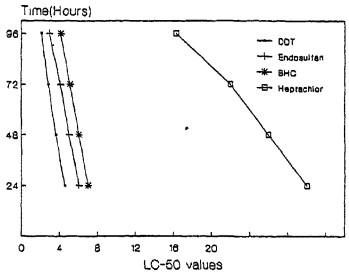

Fig. 1. LC_{50} values.

The highest mortality occurred between 48 and 96 hrs especially in higher concentrations of all pesticides. The toxicity curve for DDT is steeper than endosulfan followed by BHC whereas the curve for heptachlor is shallower than all the other pesticides studied. The order of toxicity of pesticides for *Penaeus monodon* juveniles was found to be DDT > endosulfan > BHC > heptachlor. It is evident that DDT is the potential pollutant for *P. monodon* juveniles as evidenced from the present study and previous reports for the other aquatic organisms (Selvakumar *et al.*, 1996). On the other hand heptachlor appeared to be somewhat safer to *P. monodon.*

According to U.S. EPA recommendations (Russell, 1979) the safe permissible level of BHC for aquatic organisms is 4 ppb, which is almost ten times higher than the safe permissible limit of BHC for *P. monodon,* derived from 96 hr LC_{50} value observed in the present study. However, this value was within the range reported for some other crustaceans (Murty, 1986).

In the present study, pesticides were not detected in Kandaleru creek water in Nellore district of Andhra Pradesh and coastal sea water near Tuticorin farm of Tamil Nadu during March-April 1993.

The experimental LC_{50} values obtained provide data on comparative effect of pollutants and are useful in screening potentially toxic substances. The present data indicate that among four pesticides studied, DDT is the most toxic and heptachlor is the least toxic for *Penaeus monodon* juveniles.

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