



## ENDOSULFAN MEDIATED IMPACT ON NUTRITIVE VALUE OF SNAKE HEADED FISH *CHANNA PUNCTATUS*

Dr. Garima Harit<sup>1</sup>, Dr. Neera Srivastava<sup>2</sup>

<sup>1</sup>Department of Zoology, The IIS University, Jaipur

<sup>2</sup>Department of Zoology, University of Rajasthan, Jaipur

### Abstract

**In the present investigation, *C. punctatus* was exposed to 15%, 25% and 50% doses of LC<sub>50</sub> value of endosulfan for 15 days. Muscle being the edible part of the fish, was evaluated for bioaccumulation of the endosulfan. Endosulfan content in muscle was observed to be dose and duration dependent. Treated fishes were subsequently transferred to endosulfan free freshwater for another fortnight to study the recovery response in fish. Gradual elimination of endosulfan residue was observed during depuration. Thus, uptake of endosulfan in the edible part of fish will not only influence its nutritive value but it might also affect the health of its consumers.**

**Key words:** *Channa punctatus*, Accumulation, Endosulfan, Recovery, Nutritive value.

### Introduction

Monitoring of pesticides in fish tissues is a measure to provide an early indication of the pesticide residue that accumulates within tissues & may be unsafe for man. Fish plays a vital role in food & nutritional aspects of the country due to the presence of high quality animal proteins, vitamins (A, B, D & E) and minerals essential for vitality & growth (Chitranshi, 2004). The ability of fish to metabolize organochlorines is moderate; therefore, contaminant loading in fish is reflective of the state of pollution in the surrounding environment (Guo *et al.*, 2008). This poses a great danger to fish, which constitute a major share in the aquatic environment & contribute to the economy of the nation. In addition, organochlorine pesticides have a tendency to accumulate in adipose tissues of the animals and also undergo food chain

amplification. Lipophilic pollutants are chemically very stable & resistant to microbial, photochemical, chemical & thermal degradation. The chemical stability of these compounds, their high lipid solubility & toxicity to human beings & animals has led government & researchers to feel concerned about their presence in the environment. Endosulfan is one of the most toxic organochlorine insecticides. Fishes are extremely sensitive to endosulfan; significant mortality has been reported as a result of endosulfan leakage into rivers (Naqvi & Vaishnavi, 1993).

*Channa punctatus* is consumed by the local residents of Jaipur city. Therefore, the aim of the present investigation was to observe the accumulation of endosulfan in the muscle which could reflect the nutritive value of the fish. Recovery study was carried out to observe the extent of reversibility.

### Materials & Methods

**Experimental animal:** Freshwater fish, *Channa punctatus* (Bloch) were procured from local freshwater bodies around Jaipur, averaged 22.41±1.38 cm in length & 66.12±0.31 gm in weight. To study the bioaccumulation & elimination of endosulfan in muscle, fish were exposed to endosulfan for 15 days after acclimatizing them for two weeks. Glass aquaria (500 L) were cleaned & rinsed with KMnO<sub>4</sub> solution before filling them with tap water.

**Acclimatization of fish:** To acclimatize the fish to laboratory conditions, they were kept in clean glass aquaria (500L capacity) for 15 days. After acclimatization they were distributed equally in separate aquaria. During acclimatization period, minced goat liver was fed once a week. Water of the aquaria was changed on the day after feeding.

Physico-chemical conditions of water i.e. pH, dissolved oxygen (D.O.), free carbon dioxide (CO<sub>2</sub>), hardness, alkalinity & chloride content were estimated prior to the experiment & at each autopsy interval by methods given by APHA, AWWA & WPCF (2005).

**Experimental design:** From acclimatized group one hundred & sixty fish were selected & distributed into four groups of 40 fish each in 500L glass aquaria. Fish were not fed during the experiment. Fish of group I were kept in normal tap water. They served as control. Group II treated with 15% LC<sub>50</sub> (0.006 ppm), group III treated with 25% LC<sub>50</sub> (0.01 ppm) & group IV treated with 50% LC<sub>50</sub> (0.02 ppm) of LC<sub>50</sub> value (0.04 ppm) of endosulfan. At day 5, 10 & 15, ten fish were autopsied from each group and muscle tissue was extracted to record the endosulfan uptake.

**Bioaccumulation of endosulfan in muscle:** Samples of muscle were taken from the same region in each fish; they were macerated with anhydrous sodium sulfate. The samples were then processed for extraction with acetone & hexane & estimated for endosulfan residue by the EPA method (1974). A glass column, packed with pre-washed glass wool, activated silica gel & anhydrous sodium sulfate was first rinsed with

hexane, followed by the sample extracts. All elutes obtained from muscle were completely evaporated with the help of a rotatory vacuum evaporator; the dried extracts were then dissolved in 2 ml hexane. Endosulfan residue was analysed with the help of a gas chromatograph. Bioaccumulation factor for pesticide was calculated as follows –

$$\text{Bioaccumulation factor} = \frac{\text{Concentration in Organism}}{\text{Concentration in water}}$$

## Results

**Experimental phase:** Results indicate that there is an increase in endosulfan concentration of the muscle with dose & duration of the experiment, in all treated groups (Table 1). Fish in group IV, however, survives only upto day 12, probably due to a higher dose (50% LC<sub>50</sub>) of endosulfan administered. In group I (control), however, the concentration of endosulfan is not within detectable limits. Bioaccumulation factor in all treated groups shows progressive increase from day 5 onwards. Percent accumulation of endosulfan by the end of the experiment is 42% in group II, 52% in group III & 72% in group IV.

**Table 1: Bioaccumulation of endosulfan (ppm) in muscle of *Channa punctatus* after sub-chronic exposure to endosulfan**

Groups	Parameters	Day 5	Day 10	Day 15	% Accumulation
I	Endosulfan content	BDL	BDL	BDL	-
	B.F.	-	-	-	-
II	Endosulfan content	0.00013±0.00010	0.00018±0.00016	0.00025±0.00005	42%
	B.F.	0.021	0.03	0.041	-
III	Endosulfan content	0.00022±0.00017	0.00036±0.0001	0.00053±0.00021	52.00%
	B.F.	0.022	0.036	0.053	-
IV	Endosulfan content	0.0035±0.0009	0.0048±0.00051	M	72.00%
	B.F.	0.175	0.24	-	-

B.F. = Bioaccumulation Factor; M= Mortality; BDL= Below detectable limit

**Table 2: Recovery response in endosulfan content (ppm) in muscle of *Channa punctatus* pre-exposed to endosulfan for 15 days**

Groups	Parameters	Day 0	Day 5	Day 10	Day 15	% Recovery at day 10	% Recovery at day 15
I	Endosulfan content	BDL	BDL	BDL	BDL	-	-
	B.F.	-	-	-	-	-	-
II	Endosulfan content	0.00025±0.0005	0.00015±0.00031	0.00002±0.00011	BDL	92%	-
	B.F.	0.041	0.025	0.0033	-	-	-
III	Endosulfan content	0.00053±0.0021	0.00040±0.00023	0.00026±0.00026	0.00017±0.00016	50.94%	67.92%
	B.F.	0.053	0.04	0.026	0.017	-	-

B.F. = Bioaccumulation Factor; M= Mortality; BDL= Below detectable limit

**Recovery phase:** Endosulfan content in the muscle decreases in both the pre-exposed groups at all autopsy intervals. Bioaccumulation factor also registers a decline in both the pre-treated groups (Table 2). In group II 92% recovery is recorded by day 10 & the values fall below detectable limits by the end of recovery period. However, in group III 50.94% recovery is noted at day 10 & 67.92% recovery is noted at day 15. It is thus evident that group II recovers completely by the end of recovery phase while group III will require a longer period.

### Discussion

Miranda *et al.* (2008) have suggested various factors (i.e. water solubility, degree of ionization, stability, molecule size or shape and lipid content of the species) to be responsible for endosulfan accumulation. Iyamu *et al.* (2007) have suggested lipophilic nature of OCPs to be an important factor. Organochlorine residue accumulates in aquatic biota as a result of its low solubility in water and high solubility in fats as suggested by Matthiessen *et al.* (1982). Endosulfan has been reported to accumulate in the muscle of various fish species (Malik *et al.*,

2007; Ackerman *et al.*, 2008 and Harit & Srivastava, 2009). Endosulfan content was recorded to be below detectable limits in the muscle of *C. punctatus* after 96 hr exposure (Srivastava & Kaushik, 2001). In the present study, however, marginal accumulation of endosulfan has been recorded in the muscle of *C. punctatus* which may be due to a longer period of exposure as compared to that observed by Srivastava & Kaushik (2001). Kumari *et al.* (2001) studied residual pattern of organochlorine pesticides in muscle of fishes collected from river Ganges; amongst DDT, HCH, endosulfan & aldrin, endosulfan was the least accumulated OCP in muscle of these fishes. They reported that carnivorous fishes had the highest organochlorine concentration followed by detritivorous, omnivorous & herbivorous fishes. *C. punctatus* is a carnivorous fish & it is probable that exposure of this fish to endosulfan has led to accumulation of endosulfan in the muscle. Accumulation of endosulfan in the muscle of *C. punctatus* in the present work reveals that edibility of the fish can be adversely affected by the dose & duration of the present investigation. This would not only lower the nutritive quality

of the fish but also pose a risk to the human population that consumes contaminated fish, especially if exposure is prolonged.

Tilak *et al.* (2007) reported that accumulation of butachlor (OCP) residue in several organs of *Channa punctatus*, could lead to mortality even at sub-lethal concentrations. In the present study too, total mortality in the group treated with a higher dose (50% LC<sub>50</sub>) of endosulfan, could be a result of high endosulfan accumulation. Perkins & Schlenk (2000) suggested that significant inhibition of muscle AChE may be a final step in acute cholinesterase poisoning; this cholinergic overload in muscle would lead to loss of muscular control & ultimate mortality of the fish. Inhibited AChE activity in the muscle, as suggested by Perkins & Schlenk (2000), could also be a cause for mortality in group IV fishes.

Matthiessen *et al.* (1982) recorded recovery response in endosulfan content of the muscle & viscera in fish pre-exposed for 12 weeks; they observed that endosulfan content returned to near normal within 3 months after cessation of aerial spraying, but residues were detectable even after 12 months. After 8 days of depuration, Ponmani & Dhanakkodi (1996) observed a slow & steady elimination of endosulfan from the body of *Cyprinus carpio* pre-exposed to the pesticide for 28 days & suggested a positive relation between the rate of elimination & the length of post exposure period. Present results support the above view, as an increase in endosulfan elimination is observed in muscle along with an increase in depuration period of *C. punctatus*. Berntssen *et al.* (2008) studied uptake of dietary endosulfan in the fillet of Atlantic salmon for 92 days, this was followed by a 56 day depuration period. They demonstrated that the uptake of endosulfan was higher in comparison to its elimination. Similarly, in the present study, uptake of endosulfan in the tissues of *Channa punctatus* is higher than its elimination during recovery phase. Palacio *et al.* (2002) exposed *Oreochromis niloticus* to diazinon for 96 hr & observed rapid elimination of diazinon from the whole body of fish on depuration for 10 days, by 6th day of recovery it fell to 0.29mg/kg from 28.45mg/kg. It continued to decrease towards non-detectable levels. A similar pattern of release is observed in the muscle of *Channa punctatus* pre-exposed to endosulfan for 15 days

in the present study; endosulfan content declines from 0.00025 ppm to 0.00002 ppm & reaches a non-detectable level by the end of the recovery phase.

### Conclusion

High accumulation of endosulfan in fish is detected after sub-chronic exposure. Accumulated endosulfan may biomagnify in man on consumption of the infected fish. Remarkable recovery is observed in the muscle of *C. punctatus* as the values fall within undetectable limits, which is similar to control. This study increases public awareness about toxicity levels of water bodies & suggests that stringent government regulations & monitoring of water bodies should be observed. Since application of pesticides in agriculture cannot be stopped, safety measures should be implied. Their effects on fish tissues should be monitored regularly, as the possibility of similar effects on human body cannot be ruled out.

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### References

1. Ackerman, L.K., Schwindt, A.R., Simonich, S.L.M., Koch, D.C., Blett, T.F., Schreck, C.B., Kent, M.L. & Landers, D.H. (2008) Atmospherically deposited PBDEs, pesticides, PCBs, & PAHs in Western U.S. National Park Fish: Concentrations & consumption guidelines. *Environ. Sci. Technol.*, 42: 2334-2341.
2. APHA, AWWA and WPCF (2005) Standard methods for examination of water and waste water. 21st Ed. (Eds: A.D. Eaton, C.L.S. Clesceri, W.E.W. Rice & A.E. Greenberg). Am. Publ. Hlth. Assoc., Washington, D.C.
3. Berntssen, M.H.C., Glover, C.N., Robb, D.H.F., Jakobsen, J.V. & Petri, D. (2008) Accumulation & elimination kinetics of dietary endosulfan in Atlantic salmon (*Salmo salar*). *Aquat. Toxicol.*, 86: 104-111.

4. Chitranshi, U.R. (2004) R & D priorities for sustainable development of inland capture fishery sector in India. In: Symposium on advances in fish physiology: ecological considerations, Varanasi, pp 12-14.
5. Environmental Protection Agency, EPA. (1974) Pesticide residue analysis in water. Method No. 43011-74-012.
6. Guo, Y., Meng, X.Z., Tang, H.L. & Zeng, E.Y. (2008) Tissue distribution of organochlorine pesticides in fish collected from Pearl River Delta, China: Implications for fishery input source & bioaccumulation. *Environ. Pollut.*, 155: 150-156.
7. Harit, G. and Srivastava, N. (2009) Bioaccumulation & elimination of endosulfan in muscle, gills & blood of *Channa punctatus*. *J. Environ. Sociobiol.*, 6: 13-20.
8. Iyamu, I., Asia, I.O. & Egwakhide, P.A. (2007) Concentrations of residues from organo-chlorine pesticide in water & fish from some rivers in Edo State Nigeria. *Int. J. Physical Sci.*, 2: 237-241.
9. Kumari, A., Sinha, R.K. and Gopal, K. (2001) Organochlorine contamination in the fish of the river Ganges, India. *Aquat. Ecosyst. Hlth & Management*, 4: 505-510.
10. Malik, A., Singh, K.P. & Ojha, P. (2007) Residues of organochlorine pesticides in fish from the Gomti River, India. *Bull. Environ. Contam. Toxicol.*, 78: 335-340.
11. Matthiessen, P., Fox, P.J., Douthwaite, R.J. & Wood, A.B. (1982) Accumulation of endosulfan residues in fish and their predator after aerial spraying for control of tsetse fly in Botswana. *Pest. Sci.*, 13: 39-48.
12. Miranda, A.L., Roche, H., Randi, M.A.F., Menezes, M.L. & Ribeiro, C.A.O. (2008) Bioaccumulation of chlorinated pesticides & PCBs in the tropical freshwater fish *Hoplias malabaricus*: Histopathological, physiological & immunological findings. *Environ. Int.*, 34: 939-949.
13. Naqvi, S.M. and Vaishnavi, C. (1993) Bioaccumulative potential & toxicity of endosulfan insecticide to non-target animals. *Comp. Biochem. Physiol. C*, 3: 347-361.
14. Palacio, J.A., Henao, B., Velez, J.H., Gonzalez, J. & Parra, C.M. (2002) Acute toxicity & bioaccumulation of pesticide diazinon in red tilapia (*Oreochromis niloticus* & *Mossambicus albina*). *Environ. Toxicol.*, 17: 334-340.
15. Perkins, E.J. & Schlenk, D. (2000) In vivo acetylcholinesterase inhibition, metabolism & toxicokinetics of aldicarb in channel cat fish: Role of biotransformation in acute toxicity. *Toxicol. Sci.*, 53: 308-315.
16. Poomani, R. and Dhanakkodi, B. (1996) Bioaccumulation and elimination of endosulfan in *Cyprinus carpio*. *J. Environ. Pollut.*, 3: 191-194.
17. Srivastava, N. & Kaushik, N. (2001) Use of fish as bioindicator of aquatic pollution. *Proc. Of ICCE*, pp 227-229.
18. Tilak, K.S., Veeraiah, K., Thathaji, P.B. and Bucthiram, M.S. (2007) Toxicity studies of butachlor to the freshwater fish *Channa punctata* (Bloch). *J. Environ. Biol.*, 28: 485-487.