



## Alterations of the Ionic Composition in Different Organs of Spotted Murrel (*Channa punctatus*) Exposed to Sublethal Concentration of Endosulfan

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### Abstract

The spotted murrel, *Channa punctatus* were exposed to sublethal concentration of endosulfan (8.1 µg/L) for 48 and 96 h to elucidate the impact of the pesticide on the ionic composition in different organs of the fish. After 48 and 96 h endosulfan exposure, fish were randomly selected from each control and experimental tanks, anesthetized, sacrificed and then different organs like liver, kidney, gill and muscle were dissected for mineral estimation. Results showed that liver Ca and Mg levels after 48 h increased significantly, however after 96 h, these two minerals showed reduction in their levels. The remaining minerals in the liver (P, Na, K, Fe, Zn, Cu and Mn) did not change significantly with increase in the exposure duration however, showed a decreasing trend after 96 h. Kidney Ca and Mg after 48 h exposure increased significantly by 89.5 and 79.8% and after 96 h the concentration of both these minerals were non-significantly higher by 23.2 and 27.8% from the control. Kidney Mn level after 96 h was also significant higher (by 31.7%) than the control. The remaining kidney minerals (P, Na, K, Fe, Zn and Cu) did not change significantly with change in the exposure duration, however, after 96 h some of them (P, Na, Zn) increased. Gill Ca level decreased significantly with the increase in the duration of endosulfan. Gill Fe and Cu levels also showed almost similar trend like gill Ca. The remaining minerals (P, Na, K, Zn, Mg and Mn) in the gill were not significant influenced by endosulfan exposure however, they (except Mn) showed a decreasing trend after 96 h. In muscle tissue, P and K increased significantly with the increase in the exposure periods whereas, muscle Zn showed a reverse trend. The remaining minerals (Na, Ca, Fe, Mg, Cu and Mn) did not change significantly with the change in the endosulfan exposure time however, they (except Mg and Mn) showed a decreasing trend at the end of 96 h. In essence, the present study showed that the concentration of majority of the minerals in the liver, gill and muscle after 96 h endosulfan exposure decreased, however, their level increased in the kidney. Thus, exposure of sublethal endosulfan concentration to *C. punctatus* caused significant alterations in the ionic composition in different organs of their body.

**Keywords** *Channa punctatus*, endosulfan, tonic composition, organs, toxicity.

### Ölümcül Olmayan Endosülfan Konsantrasyonuna Maruz Kalan Benekli Yılanbaşı Balığı (*Channa punctatus*)'nın Farklı Organlarında İyon Bileşimi Değişimleri

### Özet

Çalışmada Benekli yılanbaşı, *Channa punctatus*'un farklı organlarında pestisit iyon bileşimi üzerindeki etkisini göstermek amacıyla balıklar, 48 ile 96 saat arasında öldürücü olmayan endosülfan konsantrasyonuna (8,1 µg/L) maruz bırakılmıştır. 48 ile 96 saat süresince endosülfana maruz bıraktıktan sonra kontrol tankından ve deney tanklarından balıklar rastgele seçilmiş ve anestezi verilerek kesilmiştir. Daha sonra mineral oranları tahmini yapmak için karaciğer, böbrek, solungaç gibi organları ve kas dokusu ayrılmıştır. Sonuçlar; 48 saat sonra Ca ve Mg düzeylerinin anlamlı bir şekilde arttığını, fakat 96 saatten sonra bu iki mineral seviyesinde azalma olduğunu göstermiştir. Kalan karaciğer mineralleri (P, Na, K, Fe, Zn, Cu ve Mn), maruz kalma süresinin artmasıyla anlamlı bir değişiklik göstermemiş, fakat 96 saat sonra azalan bir eğilim göstermiştir. 48 saatlik sürede böbrekte Ca %89,5 ve Mg %79,8 oranında anlamlı bir şekilde artmış ve 96 saat sonra her iki mineralin konsantrasyonu %23,2 ile %27,8 oranında anlamlı olmayan oranda kontrol grubundan daha yüksek bulunmuştur. Böbrekte Mn seviyesi 96 saat sonra da anlamlı biçimde kontrol grubundan daha yüksek olmuştur (%31,7). Böbreklerde tespit edilen diğer minerallere (P, Na, K, Fe, Zn ve Cu) maruz kalma süresindeki değişikliklerle anlamlı olacak şekilde değişmemiştir. Fakat 96 saat sonra bazı mineraller (P, Na, Zn) artmıştır. Solungaçta Ca seviyesi, endosülfan süresindeki artışla birlikte anlamlı biçimde azalmıştır. Solungaçta Fe ve Cu seviyeleri de Ca'da olduğu gibi benzer eğilim göstermiştir. Solungaçta kalan mineraller (P, Na, K, Zn, Mg ve Mn) endosülfana maruz bırakılma sonrasında anlamlı şekilde etkilenmiş fakat (Mn haricindeki) mineraller 96 saatten sonra azalan bir eğilim göstermiştir. Kas dokusunda etki süresinin artmasıyla P ve K, anlamlı biçimde artmışken kasta Zn tam tersi bir eğilim göstermiştir. Esasen bu çalışma; 96 saatlik endosülfan etkisinden sonra karaciğer, solungaç ve kasta çoğu mineral konsantrasyonunun azaldığını ama bu minerallerin seviyesinin böbrekte arttığını göstermiştir. Dolayısıyla *C. Punctatus*'un letal sınır altında endosülfan konsantrasyonuna maruz kalması, balıkların vücutlarının farklı organlarındaki iyonik bileşimde anlamlı alterasyonlara sebep olmuştur.

**Anahtar Kelimeler:** *Channa punctatus*, endosülfan, iyon kompozisyonu, organ, zehirlenme.

## Introduction

The adoption of new technology in agricultural crop production and protection has increased the use of various pesticides among which endosulfan, an organochlorine pesticide is one. Although it is phased out in many western countries, still it is being used in tropical and sub-tropical regions (EFSA, 2005). The endosulfan used in agricultural fields enters the aquatic environment, generally as a consequence of runoff from the fields and/or accidental discharge and get dispersed widely throughout the water system. In addition, elevated residue levels of endosulfan in plant ingredients have also been reported (Lorenzatti et al., 2004). The increasing use of these plant ingredients in aqua-feeds in order to develop a more sustainable aquaculture has led to an increased exposure of endosulfan in fish. This compound has been found to be moderately persistent in the environment and has a tendency to bioaccumulate in various aquatic organisms (Naqvi et al., 1993). It is also reported to have a strong acute toxic effect on fish/shellfish (Naqvi et al., 1993; Petri et al., 2006) and cause a drastic decline of their population in the aquatic environment.

Fish live in intimate contact with the ambient water and so are very susceptible to aquatic pollutants. In freshwater fishes, blood and ionic concentration are regulated by interacting processes, such as absorption of ions from surrounding medium through active mechanisms predominantly at the gill, control of water permeability and selective reabsorption of ions from urine. Any alterations in one or more of these processes change the ionic composition in the fish body. These ions (e.g., sodium, potassium, chloride, calcium and magnesium) have been found to play a vital role in several body functions, viz. neuromuscular excitability, acid base balance, osmotic pressure, various enzymatic reactions and retention of membrane permeability (Verma et al., 1981). Further, inorganic phosphate acts as a major cytoplasmic buffer and is the basis of energy exchange (Aurbach et al., 1985).

The spotted murrel, *C. punctatus*, is a commercially important fish available throughout India. It is a natural inhabitant of stagnant muddy pond waters, paddy fields, weedy derelict swamps, canals, lakes, reservoirs, beels (Chondar, 1999). However, according to International Union for Conservation of Nature, it is at the present at near-threatened category. Among the various reasons of being threatened, one reason could be the effects of the pesticide runoff from agricultural fields onto the water bodies. So far, several biological effects of endosulfan in fish have been reported including growth retardation, respiratory and renal failure, oxidative stress, neurotoxicity, hepatic damage, haematological effects, biochemical and histological changes (Sarma et al., 2003; Sarma, 2004; Coimbra et

al., 2005; Glover et al., 2007). However, investigations specifically examining the changes in the ionic balance especially in different (osmoregulatory) organs of *C. punctatus* exposed to sublethal endosulfan concentration are limited. The present study was therefore conducted to study the changes in the ionic composition in different organs of *C. punctatus* exposed to sublethal endosulfan concentration.

## Materials and Methods

### Experimental Animals

*Channa punctatus* (average weight  $\pm$  standard deviation 35.6 $\pm$ 0.7 g) procured from the local fish market of Mumbai, India was transported with proper oxygenation to the wet laboratory of Central Institute of Fisheries Education, Mumbai, India. They were first given a prophylactic dip in salt solution (2%) and then were immediately stocked in a tank (1000 L) with proper aeration for about 30 days for acclimatization, during which they were fed 40% crude protein diet.

### Pesticide Used

Technical grade endosulfan (99%) (Shroff Research Institute, Mumbai, India) was used for the experiment. In our previous study (Sarma et al., 2003), the LC<sub>50</sub> value of the pesticide endosulfan for 96 h to the fish determined by probit analysis was found to be 24.3  $\mu$ g/L. Based on this value, the sublethal endosulfan dose of 8.1  $\mu$ g/L was chosen in the present study.

### Experiment Design

The experiment was carried out in 60 L identical plastic tanks. For the treatment (8.1  $\mu$ g/L of endosulfan) and control (without endosulfan), five replications were maintained. Each tank was stocked with 4 fish. The fish were exposed not to feed for 48 and 96 h, The water was replaced daily in all the tanks and fresh toxicant was added daily. The experiment was conducted in 12:12 h light-dark cycles. The water quality parameters were temperature 26-28°C, dissolved oxygen 6.5-7.1 mg/L, pH 7.2 to 7.5, alkalinity 48-58 mg/L, and hardness 48-60 mg/L.

### Sampling and Mineral Estimation

After the exposure period of 48 and 96 h, 20 fish (4 from each replicates) were selected from each control and experimental tanks, anesthetized with clove oil (50  $\mu$ l/L), and then sacrificed to dissect out different organs like liver, kidney, gill, and muscle for mineral estimation. Wet ashing method was followed to determine different minerals like phosphorus (P), sodium (Na), potassium (K), calcium (Ca), iron (Fe),

zinc (Zn), magnesium (Mg), copper (Cu), and manganese (Mn). Briefly, different tissues (liver, kidney, gill, and muscle) were weight and digested in nitric acid and perchloric acid mixture (1:1) in a Kjeldhal flask until the solution was colourless. The flask was then cooled and after appropriate dilution, different minerals (except P) were estimated by atomic absorption spectrophotometer (AAS 4129; Electronics Corporation of India Ltd., Hyderabad, AP, India). P was estimated spectrophotometrically using molybdovanadate method.

### Statistical Analysis

All the data were subjected to one-way ANOVA using statistical software SPSS version 11.0. Duncan's multiple range tests was used to determine the differences among treatment means at  $P < 0.05$ .

### Results

Data pertaining to the effects of sublethal endosulfan exposure on the liver mineral content of *C. punctatus* are summarized in Table 1. After 48 h endosulfan exposure, liver Ca and Mg levels increased significantly ( $P < 0.05$ ) by 43.1 and 65%,

respectively, however, after 96 h these two minerals showed a reduction in their levels by 33.2% and 8.6%, respectively as compared to the control [liver Mg, however, was not significantly ( $P > 0.05$ ) different from the control]. Although the remaining minerals (Zn, Cu, Mn, Fe, Na, K, and P) did not change significantly ( $P > 0.05$ ) with increase in the exposure time, a general decreasing trend was apparent after 96 h.

Sublethal endosulfan concentration exposure had a significant ( $P < 0.05$ ) impact on the kidney Ca, Mg, and Mn levels of *C. punctatus* (Table 2). Kidney Ca and Mg concentrations after 48 h exposure increased significantly ( $P < 0.05$ ) by 89.5 and 79.8%, respectively. After 96 h the concentrations of both these minerals decreased significantly ( $P < 0.05$ ) when compared to the values observed at 48 h. Kidney Mn level after 48 h exposure did not show any significant ( $P > 0.05$ ) change when compared to the control, however, after 96 h there was a significant ( $P < 0.05$ ) increment by 31.7%. The remaining kidney minerals (Zn, Cu, Fe, Na, K, and P) did not change significantly ( $P > 0.05$ ) with the change in the duration of exposure; however, after 96 h some minerals (Zn, Na and P) showed an increasing trend.

The concentrations of gill Ca, Cu, and Fe were

**Table 1.** Liver mineral content of *C. punctatus* exposed to sublethal endosulfan concentration for 48 and 96 h

Minerals	Liver		
	Control	48 h	96 h
Calcium <sup>x</sup>	618.54±22.38 <sup>b</sup>	885.07±75.14 <sup>c</sup> (+43.1%)	413.05±20.78 <sup>a</sup> (-33.2%)
Magnesium <sup>x</sup>	234.88±20.89 <sup>a</sup>	387.60±22.58 <sup>b</sup> (+65.0%)	214.75±10.66 <sup>a</sup> (-8.6%)
Zinc <sup>x</sup>	59.29±5.84	50.25±2.89 (-15.2%)	42.83±3.53 (-27.8%)
Copper <sup>x</sup>	66.46±5.09	58.46±5.39 (-12.0%)	47.56±4.18 (-28.4%)
Manganese <sup>x</sup>	24.46±2.37	19.45±1.17 (-20.5%)	18.44±1.49 (-24.6%)
Iron <sup>y</sup>	2.93±0.22	2.71±0.17 (-7.5%)	2.22±0.14 (-24.2%)
Sodium <sup>y</sup>	2.03±0.20	1.66±0.09 (-18.2%)	1.44±0.09 (-29.1%)
Potassium <sup>y</sup>	9.06±0.36	8.91±0.81 (-1.7%)	8.89±0.79 (-1.9%)
Phosphorous <sup>y</sup>	1.33±0.08	1.21±0.07 (-9.02%)	1.13±0.05 (-15.0%)

Values are means of five replicates±SE. Values with different superscripts in a row differ significantly ( $P < 0.05$ ).

Figures in parenthesis indicate percentage increase (+) or decrease (-) from the control.

<sup>x</sup> Mineral contents are expressed as µg/g of tissue. <sup>y</sup> Mineral contents are expressed as mg/g of tissue.

**Table 2.** Kidney mineral contents of *C. punctatus* exposed to sublethal endosulfan concentration for 48 and 96 h

Minerals	Kidney		
	Control	48 h	96 h
Calcium <sup>x</sup>	476.23±28.14 <sup>a</sup>	902.24±41.32 <sup>b</sup> (+89.5%)	586.48±43.99 <sup>a</sup> (+23.2%)
Magnesium <sup>x</sup>	286.57±14.04 <sup>a</sup>	515.46±48.72 <sup>b</sup> (+79.8%)	366.34±26.33 <sup>a</sup> (+27.8%)
Zinc <sup>x</sup>	60.51±2.23	52.07±4.86 (-13.9%)	68.02±6.03 (+12.4%)
Copper <sup>x</sup>	32.10±2.10	29.56±1.17 (-7.9%)	25.40±2.38 (-20.9%)
Manganese <sup>x</sup>	41.47±2.46 <sup>a</sup>	41.07±2.82 <sup>a</sup> (-0.97%)	54.62±3.19 <sup>b</sup> (+31.7%)
Iron <sup>y</sup>	3.59±0.69	2.74±0.35 (-23.7%)	2.07±0.53 (-42.3%)
Sodium <sup>y</sup>	2.97±0.33	2.76±0.26 (-7.1%)	2.99±0.41 (+0.7%)
Potassium <sup>y</sup>	9.20±0.61	7.37±0.66 (-19.2%)	6.98±0.67 (-24.1%)
Phosphorous <sup>y</sup>	1.32±0.01	1.38±0.01 (+4.5%)	1.42±0.01 (+7.6%)

Values are means of five replicates±SE. Values with different superscripts in a row differ significantly ( $P < 0.05$ ).

Figures in parenthesis indicate percentage increase (+) or decrease (-) from the control.

<sup>x</sup> Mineral contents are expressed as µg/g of tissue. <sup>y</sup> Mineral contents are expressed as mg/g of tissue.

significantly ( $P < 0.05$ ) influenced by endosulfan exposure (Table 3). Gill Ca level decreased significantly ( $P < 0.05$ ) with the increase in the duration of endosulfan exposure. Maximum decrease in the gill Ca level was noted at the end of 96 h, however, was not significantly ( $P > 0.05$ ) different from those noted at the end of 48 h. Gill Cu and Fe levels also showed almost similar trend like gill Ca. The levels decreased significantly ( $P < 0.05$ ) both at 48 and 96 h by 30.9 and 41.5%, respectively in case of gill Cu and by 39.2 and 1.1%, respectively in case of gill Fe. However, there were no significant differences ( $P > 0.05$ ) in the Cu level between 48 and 96 h endosulfan exposed fishes and also in Fe level between the control and 96 h endosulfan exposed fish. The remaining minerals (Mg, Zn, Mn, Na, K and P) in the gill tissue did not show any significant ( $P > 0.05$ ) change with change in the exposure time, however, after 96 h all the minerals except Mn showed a decreasing trend.

Significant ( $P < 0.05$ ) changes in the muscle mineral concentrations following endosulfan exposure were also noted (Table 4). Both muscle K and P concentrations increased significantly ( $P < 0.05$ ) due to endosulfan exposure. Maximum increase (15.8 and 42.6%, respectively in case of muscle K and P) was

noted at the end of 48 h, which however was not significantly different ( $P > 0.05$ ) from those noted at the end of 96 h. In contrast, muscle Zn concentration showed a reverse trend, maximum decrease (36.9%) was recorded at the end of 96 h. The endosulfan exposure had no significant ( $P > 0.05$ ) effect on the remaining minerals (Ca, Mg, Cu, Mn, Fe and Na), however, they (except Mg and Mn) showed a decreasing trend at the end of 96 h.

## Discussion

The results in the present study showed that the concentrations of majority of the minerals in the liver, gill and muscle of *C. punctatus* upon endosulfan exposure for 96 h decreased and was accumulated in the kidney. Raised level of kidney Ca along with its decreased concentration in the liver, gill and muscle after 96 h in the present study is almost in agreement with the findings of Swarnlata (1995), who found a decrease in the concentration of Ca in the liver, muscle, brain (however, not in kidney) of *Clarius batrachus* exposed to 2.03 mg/L and 0.24 mg/L of carbaryl and carbofuran, respectively, for 15 days. The probable reasons for such an observation in our study could be attributed to mobilization of minerals

**Table 3.** Gill mineral contents of *C. punctatus* exposed to sublethal endosulfan concentration for 48 and 96 h

Minerals	Gill		
	Control	48 h	96 h
Calcium <sup>x</sup>	5.84±0.39 <sup>b</sup>	5.14±0.24 <sup>a</sup> (-11.9%)	4.35±0.10 <sup>a</sup> (-25.5%)
Magnesium <sup>x</sup>	267.89±16.63	272.26±21.56 (1.6%)	255.04±15.99 (-4.8%)
Zinc <sup>x</sup>	54.48±6.25	47.12±2.49 (-13.5%)	39.65±2.46 (-27.2%)
Copper <sup>x</sup>	28.61±1.19 <sup>b</sup>	19.77±1.05 <sup>a</sup> (-30.9%)	16.74±1.01 <sup>a</sup> (-41.5%)
Manganese <sup>x</sup>	37.93±2.93	34.73±3.17 (-8.4%)	38.00±3.66 (+0.18%)
Iron <sup>y</sup>	931.17±47.46 <sup>b</sup>	564.14±48.54 <sup>a</sup> (-39.8%)	920.32±83.62 <sup>b</sup> (-1.1%)
Sodium <sup>y</sup>	1.38±0.01	1.33±0.01 (-3.6%)	1.28±0.01 (-7.3%)
Potassium <sup>y</sup>	6.40±0.11	5.91±0.17 (-7.7%)	5.82±0.17 (-9.1%)
Phosphorous <sup>y</sup>	6.02±0.44	5.83±0.48 (-3.2%)	5.02±0.43 (-16.6%)

Values are means of five replicates±SE. Values with different superscripts in a row differ significantly ( $P < 0.05$ ).

Figures in parenthesis indicate percentage increase (+) or decrease (-) from the control.

<sup>x</sup> Mineral contents are expressed as µg/g of tissue. <sup>y</sup> Mineral contents are expressed as mg/g of tissue.

**Table 4.** Muscle mineral contents of *C. punctatus* exposed to sublethal endosulfan concentration for 48 and 96 h

Minerals	Muscle		
	Control	48 h	96 h
Calcium <sup>x</sup>	440.60±23.31	451.33±42.99 (+2.4%)	335.93±37.48 (-23.8%)
Magnesium <sup>x</sup>	138.19±8.68	157.58±6.10 (+14.0%)	149.10±5.03 (+7.9%)
Zinc <sup>x</sup>	15.37±1.98 <sup>b</sup>	11.37±0.89 <sup>ab</sup> (-26.0%)	9.71±1.02 <sup>a</sup> (-36.9%)
Copper <sup>x</sup>	10.73±1.11	10.37±0.80 (-3.4%)	7.72±0.88 (-28.03%)
Manganese <sup>x</sup>	12.38±1.28	12.88±0.95 (+4.1%)	14.03±1.60 (+13.4%)
Iron <sup>y</sup>	898.90±44.21	663.93±33.54 (-26.14%)	647.06±52.42 (-28.02%)
Sodium <sup>y</sup>	560.81±32.42	587.93±18.92 (+4.84%)	524.55±26.18 (-6.5%)
Potassium <sup>y</sup>	7.89±0.41 <sup>a</sup>	9.14±0.26 <sup>b</sup> (+15.8%)	8.86±0.29 <sup>ab</sup> (+12.2%)
Phosphorous <sup>y</sup>	657.10±59.45 <sup>a</sup>	936.72±39.51 <sup>b</sup> (+42.6%)	880.55±33.62 <sup>b</sup> (+34.1%)

Values are means of five replicates±SE. Values with different superscripts in a row differ significantly ( $P < 0.05$ ).

Figures in parenthesis indicate percentage increase (+) or decrease (-) from the control.

<sup>x</sup> Mineral contents are expressed as µg/g of tissue. <sup>y</sup> Mineral contents are expressed as mg/g of tissue.

from various vital organs like liver, gill and muscle to kidney due to the toxic effect of endosulfan. Various other authors also observed similar decrease in Ca levels in *Cyprinus carpio*, *Labeo roita*, *C. batrachus*, and *Heteropneustes fossilis* exposed to dimethoate, fenvalerate, carbofuran, endosulfan, aldrin, formathion, propoxur, chlorpyrifos and deltamethrin (Singh et al., 1996; Srivastava et al., 1997; Singh and Srivastava, 1998; David et al., 2003; Logaswamy et al., 2007).

Magnesium is an essential element of animal cell involved in a variety of enzymatic reactions; however, information on how Mg ion is transported and regulated in fish is scanty. A significant rise in Mg level has been reported in *Mystus vittatus*, *H. fossilis* and *C. carpio* exposed to insecticides and their combinations (Logaswamy et al., 2007; Verma et al., 1979; Dalela, et al., 1981). Similarly, freshwater catfish, *H. fossilis* exhibited hypermagnesemia in serum on the 12th and 30th day and 4 weeks after exposure to sublethal concentrations of aldrin, propoxur and formothion (Singh et al., 1996; Singh and Srivastava, 1998). In our study, the Mg level also increased significantly ( $P < 0.05$ ) in the kidney and non-significantly ( $P > 0.05$ ) in the muscle, almost in accordance with the findings of the above authors. Gill et al. (1989) suggested that increase might be due to renal damage and dysfunction which in turn might impair the ability of the fish to actively excrete excess of these ions through kidney. Zn and Cu, two important constituents in many enzyme complexes also showed almost a decreasing trend in all the tissues under study, except in kidney where marginal increase in the Zn concentration was observed. In case of Zn also, it might have released from different tissues and were accumulated in kidney as the latter failed to excrete excess Zn due to cellular damage. From their studies on bioaccumulation pattern of Zn in *Channa punctatus*, Murugan et al. (2008) also opined that kidney is the target organ of Zn storage. Similarly Mn also showed its affinity to accumulate in the kidney like Mg and Zn.

Iron, an important ion related to respiratory activity of fish, decreased in all the organs irrespective of the exposure hours. However, in the gill tissue significant recovery was observed after 96 h. This might be due to the fact that fishes tried to reabsorb Fe into the blood through gill tissue mostly from environment. In our previous study (Sarma, 2004), haemoglobin concentration in *C. punctatus* also showed similar trend when exposed to sublethal endosulfan concentration, decreased initially and later increased almost near to control value as a compensatory mechanism to maintain homeostasis.

Sodium is one of the chief regulators of osmotic pressure of the body fluid (Singh et al., 2002). In the present study, Na ion concentration in the liver, kidney, gill, and muscle was not significantly affected. However, there was a non-significant ( $P > 0.05$ ) increment in the kidney and a decline in the

liver, gill and muscle. Logaswamy et al. (2007) observed a significant decline in Na level in the blood and liver of *C. carpio* exposed to dimethoate. David et al. (2003) also observed a significant decline in Na+ levels in the liver and gills of *L. rohita* exposed to fenvalerate. The Na content in the tissue mainly depends on the permeability functional efficiency of bio-membrane and efficient functional role of Na+ pump, which regulates ionic content of tissue. The declination in the major electrolyte Na might be due to histological alterations of Na+ pump in the gills or disturbances in the membrane permeability due to endosulfan toxicity. Bernabò et al. (2008) observed alteration in the morphology and function of gills in *Bufo bufo* exposed for short-term (96 h) to sublethal endosulfan concentration.

Potassium is the main intracellular cation involved in several physiological functions viz., nerve and muscle function, acid base balance and osmotic pressure. In our study, the muscle K levels showed a significant increment at both the exposure periods. The K levels in liver, kidney and gills were not significantly influenced by endosulfan exposure, however, these minerals showed a decreasing trend. Decreased in plasma Na and a shift of Na, K and water into muscle was reported by McCarty and Houston, (1976) and attributed these change to increase uptake from environment. An increased muscle K was also reported by Eddy et al. (1981) in rainbow trout after prolonged exposure of high carbon dioxide. In another study, Swarnlata (1995) reported an increase in the concentration of K in blood and spleen and decrease in kidney, liver, brain and also muscle of *C. batrachus* after 15 days treatment with carbaryl and carbofuran. Logaswamy et al. (2007) also found similar decrease of K content in the liver of *C. carpio* after exposure to dimethoate. In our study, increased muscle K level due to endosulfan exposure could possibly be attributed to disturbed K regulation which intern could be due to cell damage of gills and kidney, in agreement with the results of Lehtinon et al. (1990).

Phosphorus is the major basis of energy exchange. In teleost exposed to various pesticides, both hypo- and hyperphosphatemia have been recorded (Singh et al., 1996; Singh and Srivastava, 1998). In our study, the level of P non-significantly ( $P > 0.05$ ) decreased in the liver and gill whereas increased in the kidney. In the muscle also there was a significant ( $P < 0.05$ ) increment, in line with the results of Gill et al. (1991), who noticed a marked increase in inorganic phosphate in blood and skeletal muscle of *Puntius conchonius* following exposure to endosulfan for four weeks. Non-significant reduction in the liver and gill P levels in our study might result from decreased oxidative metabolism and lowered ATP production, whereas increase in the kidney and muscle P levels could be attributed to mobilization of ATP towards muscle and kidney tissue and subsequent enhanced breakdown of high energy

phosphates related to an overall hypermetabolic state (Gill *et al.*, 1991).

To sum up, the present study revealed that sublethal endosulfan exposure to *C. punctatus* caused significant alterations of the ionic composition in different (osmoregulatory) organs of this fish. Generally, maintenance of constant internal ion concentrations (e.g., Na, K, Cl, Ca and Mg) is essential for active regulation of water influx and ion efflux in aquatic organisms. Any imbalance in the levels of these ions in aquatic animals will lead to impairment of various physiological activities. Henceforth, minimum and judicious application of pesticide could be an alternative pest control measure to prevent excessive destruction of the aquatic organism.

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