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RESEARCH ARTICLE

Impact of Organophosphorus Nuvan on Some Aspects of Lipid metabolism in Fresh Water Teleost *Labeo rohita* (Hamilton).

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Abstract

Impact of sublethal concentration of nuvan (0.01µg/lt) were studied on total lipids, total fatty acids, lipase activity and free fatty acids in the fish *Labeo rohita*. The levels of total lipids and total fatty acids decreased on 1st day exposure and gradually elevated on 7 th day and 15th day. From 15th day onwards their levels gradually decreased and came nearer to control at 30th day exposure period. In contrast to this the levels of lipase activity and free fatty acids followed an opposite tend.

INTRODUCTION

Water the most abundantly available natural resource on the earth covering three fourths of the earth's surface. It is essential for existence and sustenance of all forms of life. Anything or process that causes or tends to cause a change in the state or nature or character of water in terms of colour, taste, texture(softness), temperature, pH value etc by any means is referred to as water pollution. Water pollution is posing a threat to the existence of many species of life in and around water and thereby causing imbalance in the biodiversity. In order to meet the global demand for increased food production a multitude fertilizers, chemicals and pesticides were put into use in ever increasing quantities.

Pesticides are the biological toxicants which are required by man to kill insects, pests and also man's fight against the spread of diseases (Gold et al., 1981). Pesticides usage became an indispensible and integral part of world agriculture. In addition to agriculture practices together with pest control programmes the surface runoff and aerial spraying forming the major source for translocating pesticides into aquatic ecosystems (Glotfelly, 1990; Roche et al., 2007; Joseph and Raj, 2011). Fishes represent most successful, the most remarkable, most massive and useful animals of aquatic environment. The contamination of water by pesticides may effect on non-target organisms like fish (Burkepile et al., 2000; Saxena and Gupta, 2005; Dutta et al., 2006, Ayoola, 2008; Franklin et al., 2010). The biochemical studies are good parameters which help to see the effects of toxicants on metabolism of fish (Kajare et al., 2000). So an attempt was made on sublethal impact of nuvan on some aspects of lipid metabolism in the fresh water fish Labeo rohita.

Materials and Methods

Test Chemical:

The pesticide selected for the present investigation was an Organophosphorus Nuvan. The active ingredient in Nuvan was Dichlorvos. It was widely used on diverse agricultural crops to control pests of crops, flies and mosquitoes. It has been widely used because of its degradability, non–persistent nature and low mammalian toxicity. Its commercial name was DDVP. Commercial grade was used and its effective concentration was 76%.

Experimental design:

Fresh water fish Labeo rohita, weighing 10±2 gm were procured from local fisheries department and stored in spacious aquaria. The water in aquaria was aerated twice a day, the fish were fed daily with groundnut cake and rice bran. The physico-chemical properties of water used for experiments had pH 7.4 ± 0.2, dissolved oxygen 6-7 ml /lt, hardness 160 ppm and temperature 28±1 ˚C. Before experimentation has been executed, the fish were acclimated to the laboratory conditions for a period of 10 days. Later groups of 10 fish were exposed to different concentration of Nuvan ranging from 0.7 mg/ml to 1.4 mg/ml. The mortality was observed during 96 hrs exposure period. The LC50 / 96 hrs was determined from the Percent and Probit mortality versus log concentration curve(Finney, 1964) and were subsequently verified by Dragstedt and Behrens method as given by Carpenter(1975). After determination of LC 50/96 hrs (0.11mg/ml), the fish were exposed to sublethal concentration of Nuvan (1/10th of LC50/96hrs i.e. 0.011 mg/ml) for four exposure periods i.e 1, 7, 15 and 30 day.

Methods:

In this study the levels of total lipids, total fatty acids, lipase activity and free fatty acids were estimated in the brain, liver, gill, intestine and muscle of fish. Each experiment was carried out in the organs of six individuals and the mean of six values were taken into consideration. The total lipids were estimated by the method of Folch et al (1957), total fatty acids and free fatty acids were estimated by method of Natelson(1971), lipase activity were estimated by the method of Colowick and Caplan (1965).

Results:

In this investigation the levels of total lipids, total fatty acids, lipase activity and free fatty acids were estimated in the brain, liver, gill, intestine and muscle of fish, on 1,7,15 and 30 days of exposure to sublethal concentration of Nuvan besides control levels were presented in tables 1, 2, 3 and 4. The levels of total lipids and total fatty acids were decreased relative to controls in all organs of fish at first day exposure and gradually elevated on 7 and 15 day exposure periods. From 15 day onwards their levels gradually decreased and came nearer to control at 30 day exposure period. The values were found to be significant (P<0.001). Whereas the levels of lipase activity and free fatty acids elevated in all organs of fish at first day exposure period, relative to controls. Their levels gradually decreased on 7 and 15 day exposure periods. From 15 day onwards their levels gradually elevated and came nearer to control on 30 day exposure period. The values were found to be significant (P<0.01).

Table 1: **Total lipids** (mg/gm wet wt.) in the organs of Labeo rohitha on exposure to sublethal concentration of Nuvan. Mean and standard deviation are a pool of six individual measurements. The percent changes in the total lipids at different periods were calculated in relation to the total lipids in the control medium. The differences between control and exposure period days were found to be statistically significant (P < 0.001).

Sl.No	Organs	Control	Exposure period in days			
			1 day	7 day	15 day	30 day
1	Brain SD± PC	85.5 1.10	58.2 1.83 -11.14	70.5 0.76 +7.63	76.4 1.52 +16.64	60.8 1.28 -7.17
2	Liver SD± PC	66.34 2.24	54.26 2.12 -18.20	42.35 1.80 -36.16	33.12 1.76 -50.07	58.32 2.14 -12.08

3	Muscle SD± PC	86.52 2.36	74.38 1.89 -14.03	61.75 2.52 -28.62	54.56 1.95 -36.93	78.24 2.26 -9.57
4	Intestine SD± PC	54.7 1.24	46.3 0.89 -15.35	53.2 1.20 -0.36	60.4 1.34 +10.42	52.6 0.66 -3.83
5	Gill SD± PC	41.6 0.58	33.5 1.57 -19.47	42.4 0.92 +1.92	48.9 1.36 +17.54	38.0 1.22 -8.65

SD – Standard Deviation; **PC** – Percent Change

Table 2: **Total fatty acids** (mg/gm wet wt.) in the organs of Labeo rohitha on exposure to sublethal concentration of Nuvan. Mean and standard deviation are a pool of six individual measurements. The percent changes in the total fatty acids at different periods were calculated in relation to the total fatty acids in the control medium. The differences between control and exposure period days were found to be statistically significant ($P < 0.01$).

Sl.No	**Organs**	**Control**	**Exposure period in days**			
			1 day	**7 day**	**15 day**	**30 day**
1	Brain SD± PC	18.54 0.42	17.36 0.58 -6.36	23.25 1.76 +25.40	26.82 1.80 +44.66	27.76 1.50 +49.73
2	Liver SD± PC	26.32 0.80	24.83 0.91 -5.50	28.17 0.68 +7.02	30.54 0.98 +16.03	27.98 1.50 +6.30
3	Muscle SD± PC	4.16 0.36	3.76 0.52 -9.61	5.83 0.40 +40.14	7.35 0.45 +76.68	6.25 1.12 +50.24
4	Intestine SD± PC	9.75 0.56	8.96 0.18 -8.10	10.84 0.16 +11.17	13.73 0.42 +40.82	11.25 1.20 +15.38
5	Gill SD± PC	4.86 0.36	4.05 0.20 -16.66	6.56 0.46 +34.97	7.84 0.49 +61.31	6.14 0.74 +26.33

SD – Standard Deviation; **PC** – Percent Change

Table 3: Lipase activity (Lipase units /gm wet wt.) in the organs of Labeo rohita on exposure to sublethal concentration of Nuvan. Mean and standard deviation are a pool of six individual measurements. The percent changes in the lipase activity at different periods were calculated in relation to the lipase activity in the control medium. The difference between control and exposure period days were found to be statistically significant (P < 0.05).

Sl.No	Organs	Control	Exposure period in days			
			1 day	7 day	15 day	30 day
1	Brain SD± PC	166.5 5.32	183.4 8.50 +10.15	168.5 8.22 +1.20	149.7 6.85 -10.09	158.6 12.43 -4.75
2	Liver SD± PC	342.6 11.32	362.5 6.57 +5.80	350.0 8.46 +2.15	328.5 8.12 -4.11	308.0 4.16 -10.09
3	Muscle SD± PC	82.5 5.54	112.2 6.24 +36.0	99.6 7.52 +20.72	68.4 4.56 -17.09	78.0 8.56 -5.45
4	Intestine SD± PC	276.4 7.46	290.6 7.52 +5.13	283.4 10.27 +2.53	265.2 9.38 -4.05	264.0 5.23 -4.34
5	Gill SD± PC	158.0 12.16	170.5 6.32 +7.91	164.2 4.67 +3.92	141.7 5.14 -10.31	152.5 7.85 -3.79

SD – Standard Deviation; **PC** – Percent Change

Table 4: **Free fatty acids** (mg/gm wet wt.) in the organs of Labeo rohita on exposure to sublethal concentration of Nuvan. Mean and standard deviation are a pool of six individual measurements. The percent changes in the free fatty acids at different periods were calculated in relation to the free fatty acids in the control medium. The differences between control and exposure period days were found to be statistically significant (P < 0.05).

Sl.No	Organs	Control	Exposure period in days			
			1 day	7 day	15 day	30 day
1	Brain SD± PC	6.68 0.09	6.42 0.26 -3.89	6.08 0.13 -10.02	5.85 0.40 -12.42	6.54 0.18 -2.09

2	Liver SD± PC	5.32 0.10	5.20 0.08 -2.25	5.08 0.13 -4.51	4.76 0.35 -10.52	5.25 0.08 -1.31
3	Muscle SD± PC	1.15 0.06	0.97 0.12 -15.65	0.84 0.16 -26.95	0.72 0.20 -37.39	1.08 0.05 -6.08
4	Intestine SD± PC	3.40 0.08	3.29 0.12 -3.23	3.14 0.16 -7.64	3.02 0.22 -11.17	3.32 0.06 -2.35
5	Gill SD± PC	2.18 0.07	2.05 0.15 -5.96	1.85 0.03 -15.13	1.72 0.26 -21.10	2.06 0.11 -5.50

SD – Standard Deviation; **PC** – Percent Change

Discussion:

The term lipids are heterogeneous group of complex macro molecules such as fatty acids, acylglycerols, phosphoglycerides, steroids, terpenes and prostaglandins. They are the components of living system, insoluble in water but soluble in polar solvents. They form energy rich reserves whose calorific values are twice that of carbohydrates or proteins. The mobilization of lipid reserves in an organism testifies the imposition of high energy demands (Sreenivasa Reddy and Ramana Rao, 1989). Observation on the involvement of lipid metabolism exposed to pesticides are many (Kaphlino et al., 1981; Swamy et al., 1983; Tewari et al., 1987; Singh and Singh, 2011; Stalin and Sam Manohar Das, 2012). All the above studies shows that shifts in lipid metabolism when animals are exposed to toxicants.

In the present study results also indicate the involvement of lipid metabolism in the fish Labeo rohita on exposure to sublethal concentration of Nuvan. In this study relative to controls the levels of total lipids and total fatty acids were decreased, where as the levels of lipase activity and free fatty acids were elevated on first day exposure. The decrease in total lipid and total fatty acid levels followed by elevation in the levels of lipase activity and free fatty acids on first day exposure indicates the high energy demand associated with imposed Nuvan stress. To overcome this animal tends to mobilize the lipid reserves by stimulating the lipase activity. Some of the observations were also coincides the present trend in the decrease of total lipids and these were as follows. It was reported decrease in total lipid content when Catla catla exposed to pyrethriod, fenvalerate (Susan et al., 1999). The reduction of lipid content in the freshwater crab Spiralothelphusa hydrodroma when it was exposed to a pesticide, chlorpyrifos (Senthilkumar et al., 2007). It was observed decline in lipid content in hepatopancreas of freshwater crayfish Cherax quadricarinatus following exposure to glyphosate (Frontera et al., 2011). It was reported similar decrease in lipid content in the fresh water shrimp Streptocephalus dichotomous on exposure to organophosphorus pesticides (Arun kumar and Jawahar Ali, 2013). It was also reported similar trend in decrease in lipids to meet the energy demands during stress condition (Padma Priya and Avasn Maruthi, 2013). It was also reported that decrease in lipid and cholesterol contents in fish Ophiocephalus orientalis on exposure to cypermethrin (Shruthi and Tantarpale, 2014). The decrease in total lipids, cholesterol, triglycerides were observed at initial exposure of Channa punctatus to sublethal concentration of endosulfan (Sarma et al, 2015). The decline in the lipids and elevation in free amino acids in the fish Clarias batrachus on exposure to Rogorin (Helan Chandra et al, 2015).

Lipase activity initially elevated on 1 day exposure followed by its inhibition on 7 & 15 day exposure periods. This was clearly evident by drastic decrease in free fatty acids and elevation in total lipids and total fatty acids up to 15 day. Thus maximum percent inhibition in lipase activity and free fatty acids were seen at 15 day exposure period

(Ganesan et al., 1989; Sivaprasad Rao and Ramana Rao, 1979; Kamble and Muley, 2000; Bhavan and Geraldine, 2002). It was observed metabolic shift from carbohydrate to lipid metabolism through acetyl-CoA barrier leading to an increment in lipids in the organs of fresh water mussel Lamellidens marginalis under pesticide toxicity (Swami et al., 1983). Similar significant decline in total lipids followed by elevation of lipase activity and free fatty acids has been reported in tissues of Labeo rohita exposed to sublethal concentration of Nuvan (Giridhar and Indira, 1997). A similar trend were also reported in Labeo rohita exposed to sublethal concentration of Deltamethrin (Neeraja and Giridhar, 2014). Furthermore it was also reported that the decrease in total lipids during initial exposure periods and gradually recovered on 30th day exposure period in fresh water fish Channa Punctatus on exposure to synthetic pyrethriod λ- Cyhalothrin (Sirohi and Saxena, 2006).

In later half of exposure the lipase activity and free fatty acid levels goes on elevation and came nearer to control on 30 day exposure period, where as the levels of total lipids and total fatty acids followed an opposite trend. Metabolic compensation involves break down and synthesis of products necessary to cope up with altered situations. Every living organism has its own so called detoxification mechanism to get rid of foreign substances entered in the body, however if toxic substances are encountered in higher concentration, they are bound to bring severe adverse effects (Satyavardhan 2010). In this study the alterations in lipid metabolism might to compensate with situation shown by the fish for its survival.

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