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### RESEARCH ARTICLE

## CHANGES IN TOTAL LIPID CONTENT IN THE LIVER OF THE FRESHWATER FISH, MYSTUS SEENGHALA (SKYES 1839), EXPOSED TO SUBLETHAL CONCENTRATION OF METHOMYL 40 % SP CARBAMATE INSECTICIDE.

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Sublethal concentration, Methomyl, Liver, Lipid, Toxicant.

### Abstract

This study has been undertaken to investigate the impact of sub lethal concentration of Methomyl 40 % SP Carbamate insecticide on total Lipid content in the liver of freshwater fish *Mystus seenghala* (Skyles 1839). During our present investigation fishes were exposed to the sublethal concentration (1/10th of 96hr LC<sub>50</sub> concentration) of Methomyl 40 % SP a carbamate insecticide for 10, 20 and 30 days respectively. The total Lipid content in the liver tissue was determined by the standard procedure from control fish and the fish exposed to sublethal concentration of Methomyl 40 % SP a carbamate insecticide. The results showed a significant decrease in the total Lipid content in liver tissues exposed to Methomyl a carbamate insecticide. The reduction in total Lipid content indicates that the test fish response to the toxicant stress.

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### Introduction:-

The use of insecticides has been recently increased as compared to organochlorine pesticides due to their high selectivity and lower persistence in the environment. Insecticides enter surface water via air drift, leaching from agricultural land and surface runoff during or after the application of pesticides (Goulding et al., 2013).

Fishes are in direct contact with the surrounding water through their gills. Pesticides have an effect on non-target animals like fish. Pesticides are extremely harmful, not only to fishes but also to different organisms. The intake of pesticides affects the biochemical composition of fishes (Shoaib N. et al. 2013).

*Mystus seenghala* (Skyles 1839) is found in large rivers, reservoirs, canals, flooded fields and ponds. The vernacular name of *Mystus seenghala* is "Shingta" in Maharashtra; *Mystus* is a predatory fish, feeds on other fishes. It is one of the largest edible fish. It is considered to be a very important food fish. *Mystus seenghala* fish species belong to the Cyprinidae family that is found in most rivers in India (Verma P. S. 2010). The wide distribution of the freshwater fish *Mystus seenghala* makes this species a potential and useful biomarker for monitoring aquatic ecosystems. Therefore, the aim of the present study was to investigate the changes in total Lipid content in the liver of the

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freshwater fish *Mystus seenghala* (Skyles 1839) after exposure to sublethal concentration of carbamate insecticide (Methomyl 40% SP).

## Material and methods:-

### Experimental Test Fish

For the present study, the live freshwater fish *Mystus seenghala* (Skyles 1839) ( $13 \pm 2$  cm) were collected in polythene bags from river Bhima of Pune district (M.S.) India, and brought to the laboratory. Care was taken to reduce hyperactivity and physical injuries to the fish. They were screened for any possible pathological symptoms. They were then kept in large Aquarium tank of 20 litre capacity and then acclimated in chlorine free water for 10 days under normal temperature. Before stocking, the tank was washed with 1 % KMnO<sub>4</sub> to avoid the fungal infection. Water was changed in alternate days. The fishes were fed a commercial fish diet.

### Experimental design

Well acclimatized freshwater test fish *Mystus seenghala* (Skyles 1839) approximately length ( $13 \pm 2$  cm) and average weight ( $17 \pm 2$  gm) were selected from the stock and exposed to sub lethal concentration of Methomyl 40 % SP for 10, 20, 30 days respectively to investigate impact of sub lethal concentration of Methomyl on total Lipid content in the liver. In the present study 1/10th of 96hr LC<sub>50</sub> concentration of Methomyl 40 % SP carbamate insecticide was selected as sub lethal concentration. The experiments were carried out in glass aquarium with 10 fishes each. The experimental medium was renewed daily till the end of the experiment. The experiment was repeated five times and the mean values recorded separately for test fish. Simultaneously 10 fishes were reared in pesticide-free medium and are treated as control for the experiment.

### Estimation of Lipid content

Estimation of total lipid was carried out by Barnes and Blackstock (1973) method.

100 mg wet tissue was homogenized in 20 ml chloroform – methanol mixture using a glass homogenizer, filtered through Whatman No.1 filter paper and to this was added 4 ml of 0.9% NaCl solution. This mixture was shaken well and transferred to a separating funnel and was allowed to stand overnight at 4 ° C. A clear biphasic layer was formed with the lower phase containing all the lipids. The upper phase was discarded and lower phase was collected and the volume was made up to 10 ml by the addition of chloroform. This was transferred to a 50 ml beaker and the solvent was allowed to evaporate at 50-60 ° C for five hours. Then 5 ml of concentrated H<sub>2</sub>SO<sub>4</sub> was added to it, mixed well, and placed in boiling water bath for 10 minutes, then cooled to room temperature. 0.2 ml of this sample was taken in a test tube and 5 ml of phosphovanillin reagent was added. Mixed well, and was allowed to stand for 30 min. Standard was prepared by mixing 0.2 ml of standard cholesterol and 5 ml of phosphovanillin reagent, allowing it to stand for half an hour. Blank was prepared by taking 0.2 ml of chloroform and 5 ml of phosphovanillin reagent. Absorbance of test and standard samples against the blank were measured at 520 nm by calorimeter.

The total Lipid content expressed as in mg/ 100 mg wet weight of tissue.

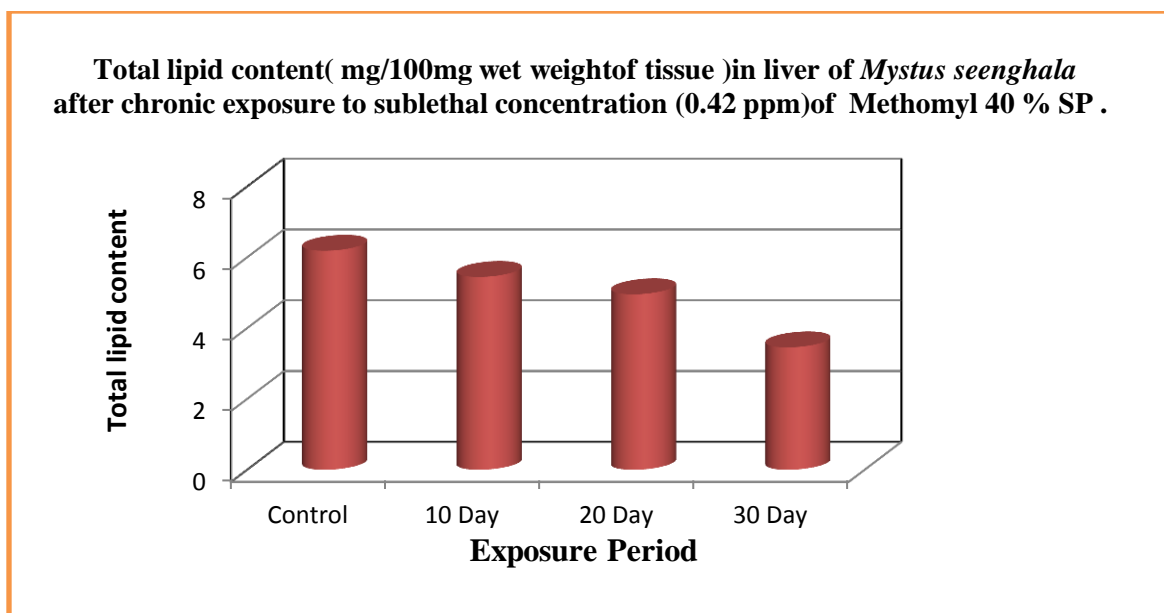
## Results and Discussion:-

The total lipid content in the liver of the test fish *Mystus seenghala* treated with the sub lethal concentration of Methomyl 40 % SP for different exposure period interval showed that reduced trend. The changes in the level of total Lipid contents in the liver of the fish, *Mystus seenghala*, were given in the Table No. 1 and Figure No. 1.

**Table No 1:-**Effect of Sub lethal concentration of Methomyl (0.42 ppm) (1/10th of 96hr LC<sub>50</sub> value i.e. 4.2 ppm) on total Lipid content (mg/100mg) in the liver tissues of *Mystus seenghala* after chronic exposure.

Organ	Control	Exposure Period		
		10 Days (0.42 ppm)	20 Days (0.42 ppm)	30 Days (0.42 ppm)
Liver	06.19 ± 0.12	05.45 ± 0.32 (11.95)	04.96 ± 0.21 (19.87)	03.46 ± 0.12 (44.10)

1. Each values are expressed in mg /100mg wet wt. of tissue.
2. Each value indicates the mean ( $X \pm SD$ ) of five estimations.
3. Figures in bracket indicate difference in percentage over control.



**Figure No 1:-**Effect of Sub lethal concentration of Methomyl 40% SP (0.42 ppm) on total Lipid content (mg/100mg) in liver tissues of *Mystus seenghala* after chronic exposure

Total Lipid content in liver showed a decline trend to  $05.45 \pm 0.32$  mg/100mg wet tissue on 10th day. It was found to have been further reduced to  $03.46 \pm 0.12$  mg/100mg wet tissue after 30 days of exposure period at the 0.42ppm sub lethal concentration of Methomyl 40 % SP, which was equivalent to 44.10 % reduced from the control value ( $06.19 \pm 0.12$  mg/100mg wet tissue).

In the test fish, total Lipid contents in the liver recorded were  $05.45 \pm 0.32$ ,  $04.96 \pm 0.21$  and  $03.46 \pm 0.12$  mg/100mg wet tissue at 0.42 ppm sublethal concentration of Methomyl 40 % SP for 10, 20 and 30 days of exposure respectively. The percentage of reduction in liver total Lipid content over the control was 11.95%, 19.87% and 44.10% after 10, 20 and 30 days of exposure respectively.

In the present study there is decline in total Lipid content in the liver tissue was recorded in the freshwater fish *Mystus seenghala* exposed to toxicant methomyl 40% SP.. Decrease in total lipid in tissues might be due to drastic decrease in glycogen content in the same tissue which is an intermediate source of energy during toxic stress conditions. After glycogen, lipid content may be used for energy production to overcome toxic stress. Some workers support these results in which lipid content decreased in fishes after exposure to pollutants. Tripathi and Verma (2004) reported changes in protein, glycogen and lipid content in the liver and muscle tissues of the freshwater fish, *Clarias batrachus* L treated with endosulfan. Singh and Singh (2006) reported changes in tissue lipids, phospholipids and protein profile in the male *Heteropneustes fossilis* Bloch exposed to sub lethal concentrations of endosulfan. Amudha et al. (2002) investigated that the effect of dairy effluent on *O. mossambicus* and reported that lipid content was decreased. Similarly decreases in lipid content level were observed by Swapna et. al., (2006) in *Cyprinus corpio* exposed to textile effluent. Shruti S. Gijare et. al., (2011) also reported that decrease in total lipid content in the liver in freshwater fish *Ophiocephalus punctatus* exposed to synthetic pyrethroid cypermethrin. Israel stalin S. and Sam Manohar Das. S. (2012) reported that maximum decline in total lipid content in the liver tissue of fish *Cirrhina mrigala* (Hamilton) exposed to Fenthion after 30 days.

### Conclusion:-

In the present study, the significant depletion in Lipid content was observed in liver tissue of test fish *Mystus seenghala* after chronic exposure to sub lethal concentration of methomyl 40 % SP a carbamate insecticide. A decrease in the total lipid content of the liver exposed to carbamate insecticide suggests that lipid might have been utilized for energy production for other metabolic functions during toxic stress conditions.

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