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ACUTE TOXICITY STUDIES, DETERMINATION OF MEDIAN LETHAL DOSE AND BIOCHEMICAL CONTENTS IN FRESHWATER SNAIL *BELLAMYA BENGALENSIS (*LAMARCK, 1822)

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Abstract: *The Mollusca is the second largest phyla become an important aquatic creature that represents an enormous diversity, nutrition advantages, and its natural compounds with eight distinct classes. The freshwater gastropod snail Bellamya bengalensis are ecologically important for humans and are constantly exposed to pesticides. Acute toxicity studies were carried out in the freshwater gastropod molluscs Bellamya bengalensis exposed to sub-lethal concentrations of Endosalphan (Mera-71). Mortality of the experimental snail was assessed for LC_{50} value and assessment of the quantitative study of biochemical content (viz. protein, lipid, and carbohydrate) in Bellamya bengalensis was calculated. The LC50 values obtained with the snails exposed to Endosalphan (Mera-71) were 0.009, 0.008, 0.007, and 0.006 mg/l, for 24, 48, 72, and 96 hrs respectively and the water temperature was 30°C. LC_{50} value increased with the decrease in mean exposure concentrations and times respectively. The effects of sub-lethal concentrations of Endosalphan (Mera-71) on biochemical contain body parts hepatopancreas; gill, foot, and adductor muscle respectively have been studied in detail. The exposure of the snails to sub-lethal concentrations of the Endosalphan caused desquamation and breakdown of protein in all the body parts due to Endosalphan stress. From obtained results, the toxicity of Endosalphan (Mera-71) was concluded was responsible for the alterations in behavioural change.*

Key Words: *Acute toxicity, pesticide, Endosalphan Mera-71, biochemical content.*

1. INTRODUCTION:

Environmental pollution is one of the major challenges in the modern human society that can be severely contaminated the integrity and health of the aquatic ecosystem. These varieties of pollutants represent a potential risk to the freshwater biota (Khan and Chaudhuri, 1984; Benkendorff, 2010; Ali et. al., 2013, 2017; Hashem et al, 2017). Anthropogenic activities and influence on the aquatic environment is in the form of sub-lethal contamination which results in chronic stress conditions that have a negative effect on aquatic life. Concentration of toxic metals at higher level can cause adverse effects on aquatic organism at cellular or molecular level and ultimately leads to disorder in biochemical components to the organism (Banarescu and Petru, 1990; Appleton and Chris, 1996; Cordeiro, 2002; Guria et al. 2003; Russo and Lagadic 2004; Khangarot and Das 2010; Ray et al. 2013). Although several adverse health effects of contaminant have been known for a long time, exposure these pollutants continues, and is even increasing in some parts of the world (Järup, 2003; Bogan et. al., 2008; Solomon et. al., 2010; Wieczorek et al, 2013; Bhattacharya et. al., 2014).

Pesticides are commonly detected in freshwater ecosystems, yet there is considerable uncertainty over whether they are having any adverse impacts on aquatic communities where the Molluscan carry out multiple functional roles in the freshwater ecosystems. Identifying such effects requires aquatic community descriptors that are sensitive to changes in exposure and can indicate the level of exposure in a complex environment with diverse factors acting on communities (Gokhale and Mane, 1990; Camargo, 2003; Bogan and Arthur, 2008; Dhara et. al., 2017; Moura and Santos, 2020). Toxicity testing is an essential tool for assessing the effect and fate of toxicants in aquatic ecosystems. The evaluation of the acute toxicity of a particular pollutant to a representative species of molluscan snail in terms of mortality and time in a laboratory is necessary (Ernest Hodgson, 2004; Kaushik and Kaushik, 2006; Brix et. al., 2011; Raghava Kumari,

2013). This tool is useful to develop a tolerance level of contamination and to establish limits and levels of acceptability by the biotic components. The lethal concentration (LC) value is usually expressed for the results of acute toxicity. The LC50 effective dose is a concentration of which 50 % of mortality of exposed organisms occurs. For the study of short-term toxicity test, the period of exposure is generally 24, 48, 72, and 96 hrs and this is a convenient way of expressing acute lethal toxicity of a given pollutant to the average individual. The exposure time plays an important role in LC50 values. The toxicological study is not only protecting humans and the environment from the harmful effects of toxicants, but also facilitates the development of more selective toxicants with clinical drugs and pesticides (Kamble and Kamble, 2014).

Several studies on insecticides' effects on aquatic organisms demonstrated that concentrations that are not sufficient to control many species of crop pests, mosquitoes, and flies, nevertheless can kill eggs and larvae of molluscs (Morrissey et al, 2014; Rico et al, 2018; Raby et al, 2018; Saida et. al., 2020; Ortmann, 2021) immobilize of fishes, decreases the productivity of population of phytoplankton's and alter the tissue chemistry in molluscs and fishes, and the investigation shows the effects of pesticide contamination in the field (Camargo, 2003; Awati, 2004; Johnson et. al., 2009; Moura and Santos, 2020). There is a limited field studies report on a clear relationship between measured pesticide exposure and observed community response. Where effects have been described and studies have usually been concerned only with a small number of water bodies. Detecting the effects of pesticides in the aquatic environment is technically difficult and often costly. The present study was designed to investigate and identify the biological indicator that could be used to characterize the heterogeneity of pesticide contamination at larger scales.

2. MATERIALS AND METHODS:

The *Bellamya bengalensis* (Lamarck, 1822) molluscs (shell 1.2 ±0.5 cm to 2.9±0.5 cm in shell length; width 1.2±0.0.5 cm; weight 1.4±0.5 g) were collected from Sadatpur lake (19°34'56.91" N Latitude and 74°26'20.79" E Longitude) near Loni- Pravaranagar. The animals were soon brought to the laboratory and the shells were cleaned off encrusting epifauna and kept in glass aquaria containing 50L of de-chlorinated tap water for 7 days to acclimatize them to laboratory conditions. The animals were then grouped in three sets, each containing ten animals.

Photo Plate No. 1: Measurement of *Bellamya bengalensis* (Lamarck, 1822)

The first set served as control, the second set was exposed to predetermined nominal (LC0) and third set to lethal (96 h LC_{50}) concentrations of Endosalphan (Mera 71). The nominal and lethal concentrations of fenthion for I. viviparous were 7.2 and 13.6 ppm, respectively (Akarte and Mane, 1968). At the end of 96 h exposure, animals of each set 622 were sacrificed to isolate mantle, gills, hepatopancreas, foot, gonads and adductor muscle. Each body part of animals of each set was pooled separately and three samples were drawn for histopathological studies and total body for the biochemical analyses. Total protein was estimated according to the method of Lowry et al, glycogen according to De Zwaan and Zandee, (1972) and lipid was extracted according to Blig and Dyer (1959). Total cholesterol was estimated as given by standard method (Kolmer et al., 1951). Result is reported as the mean value of three analyses and expressed as mg biochemical content per 100 mg wet weight of tissue.

Classification

Kingdom : Animalia
Phylum : Mollusca
Class : Gastropoda
Super family : Viviparidae
Family : Viviparidae
Genus : *Bellamya*
Species : *bengalensis* (Lamarck, 1822)

The family Viviparidae is a family of large operculate freshwater snails, aquatic gastropod mollusks, sometimes known as the river snails or mystery snails. This family is classified in the informal group Architaenioglossa according to the taxonomy of the Gastropoda contains three subfamilies. This family occurs nearly worldwide in temperate and tropical regions, with the exception that they are absent from South America (Bouchet & Rocroi, 2005).

Pesticide: Mera-71 it technical grade synthetic ammonium glyphosphate 71 % EC/active, ammonium sulphate GS, Inert poly-oxyethylene Amine (12.50 %), Surfactant moisture (01.00 %) ingredient manufactured by Excel Crop Care. Ltd. 148/87, SV Road, Jogeshwari (West), Mumbai-400102, India manufactured at Bhavnagar, was used in the present toxicological and biochemical experiments.

Toxicity Experiments: Ten gastropod were kept in plastic tough containing 30 L of de-chlorinated tap water. Gastropod were exposed to four different concentrations of Endosulfan which were 0.005, 0.006, 0.007 and 0.008 mg/L. Synthetic pesticides Endosulfan was given as the final concentration (w/v) of aquatic ingredient in the test aquaria. Control gastropod was kept in de-chlorinated tap water only. Each set of experiment was replicated six times. Mortality was recorded every 24 h during the observation period of 96h. The LC values (LC, LC and LC), upper and lower 10 50 90 confidence limits (UCL, LCL at 95% confidence limits), slope values 't' ratio and heterogeneity were calculated by POLO computer programme**.** The regression coefficient was determined between exposure time and different values of LC_{50}.

Biochemical Experiment: Snail were exposed to sub-lethal concentration

Total Protein: Total Protein levels were estimated according to the method of Lowry *et al.* using bovine serum albumin as standard 10% trichloroacetic acid (TCA; 5 mg/mL, w/v) was prepared to homogenates tissue.

Total Lipid and glycogen: Estimation of total Lipid was estimated according to the method Baraes & Black stock (1973) and glycogen was made according to the method of Anthrone Method of Kemp &kits (1954)**.**

Statistical Analysis: Every experiment was replicated at least six times and the values are expressed as mean + - SE Students t-test was applied to determine significant ($P<0.05$) differences between treated and control animals. The product moment correlation coefficient was applied between exposure time and fecundity survival of the gastropod molluscs.

Photo plate No.2: Aerial view of Study Area

Aerial view of Sadatpur Lake (Source Google Maps 2022)

3. RESULT AND DISCUSSION:

Viviparidae commonly referred to as "mystery snails" have a worldwide distribution. Burch, (1982) list 17 total species from North America and some part of south Asian region. Preston, (1915) lists fourteen from India, Liu Yueyin (1979) includes twelve from China, and Zhadin, (1952) lists five from the territory of the former USSR. They are also present in Australia, Africa and Europe. The Viviparidae are noteworthy in their wide selection of foodstuffs. Brown (1991) indicates they can function both as grazers, consuming algae growing on any submerged surface, and detritivores, utilizing fine particulate organic matter and the bacteria and other microorganisms therein. They also filter feed on

suspended matter, competing with the clams and mussels. Dillon (2000) indicates that North American *Campeloma* can also be baited with carrion, and describes the process by which *Bellamya bengalensis* snails consume suspended particulate material. The gills of *Bellamya bengalensis* are characterized by unusually large triangular lamellae whose tips hang over a ciliated gutter or food groove running across the floor of the mantle cavity. Cilia direct mucus and entrapped particles to the food groove. Particles collected on the gill filaments are carried to the tip, where they also fall into the groove. A food / mucus string forms which is carried forward and collected into a ball or "sausage". Periodically the snail will turn its head and eat the collected food.

Principal genera, in three subfamilies:

Viviparidae; Viviparous is found in the Eastern North America and most of Europe and some part of India (Burch, 1982). So many species are abundantly found in Asia and other part of the world which are looking viviparous species but having different nomenclature. The mortality of freshwater gastropod molluscs *Bellamya bengalensis* (Lamarck, 1822) due to exposure to four different concentrations of Endosalphan (synthetic pyrethroids) for 24, 48, 72 and 96 hrs in different water concentration. The LC values of 50 Endosalphan *at* 24, 48, 72 and 96h were 0.009, 0.008, 0.007 and 0.006 mg/l, respectively and water temperature were 30°C. Endosalphan found more toxic in winter season than the summer season. The slope values were steep and the results were found to be within the 95% confidence limits of LC values. The 't' ratio was greater than 1.96 and the heterogeneity factor was less than 1.0. The 'g' value was less than 0.5 at all probability levels. Endosalphan (Mera 71) was used as synthetic pesticides for biochemical studies. Tables 3 and 4 indicated a significant ($P<0.05$) dose dependent decrease in total protein level in total body tissue of *Viviparous* exposed for 40 and 60% of 24 hrs LC of Endosalphan. Protein 50 % depletion was observed in both tissues while the maximum reduction in protein level was 65% of in control in total body; gastropod treated with 60 % of 24 hrs LC of Endosalphan at 30°C 50 % Changes induced by the sub-lethal concentration of Endosalphan in the level of lipid and glycogen in total body tissues of gastropod molluscs *Bellamya bengalensis.* Exposure to 40 and 60% of 24h LC of Endosalphan up to 50, 96 h resulted in an increase of total lipid and glycogen of gastropod molluscs *Bellamya bengalensis* to value of 120 and 142 % at 30°C water temperature and 116 and 137% of controls, respectively. The treatment of gastropod with sub-lethal doses for 96h, caused significant ($P<0.05$) inhibition of AChE activity in the nervous tissue of gastropod molluscs *Bellamya bengalensis* (Lamarck, 1822). Thus treatment with 40 and 60% of 24h, LC of 50. Endosalphan reduced the AChE activity, Exposure to 40 and 60% of the 24h LC of Endosalphan resulted 50 in a decrease of AChE activity in body tissue of gastropod molluscs *Bellamya bengalensis* (Lamarck, 1822) to 67 and 56% at 16°C and 72 and 59% of control at 30°C, respectively. Analysis of variance demonstrated that the inhibition of AChE was both time and dose dependent ($P<0.05$). After 96 h exposure to 40 and 60% of LC of 50 Endosalphan, lactic dehydrogenize (LDH) activity was increased to 130 and 154%. The maximum enhancement was observed (154% of control) in body tissue on exposure to 60% of 24h LC at 30°C. Data of biochemical section of the results clearly indicates that the sub-lethal exposure (40 and 60% of LC) of pesticides after 96h, significantly decrease 50 the level of total protein, lipid and glycogen levels decrease in total body tissues amino acids level and enzyme lactic dehydrogenize (LDH) in muscle, foot, gonad and body tissue of the freshwater gastropod molluscs *Bellamya bengalensis* (Lamarck, 1822) at both the water temperature. The rate of depletion in total protein lipid and glycogen level, were significantly ($P<0.05$) dose dependent.

Table 1: Changes in the protein content (mg/g of wet wt tissue and % changes over the control in different tissues of *Bellamya bengalensis* (Lamarck, 1822) gastropod exposed to sub-lethal concentration of Endosulfan for 96 h) small size

Sr. No.	**Tissue**	**Protein**		**% decrease**
		Control	Experiment	
1.	Hepatopancreas	10.2± 0.380	6.41± .460	40.12
2.	Gills	11.2± 0.463	5.42±.462	48.02
3.	foot	11.0± 0.431	6.45±.656	46.02
4.	Adductor muscle	10.2± 0.453	6.41±.404	40.2

Graph 1.: Changes in the protein content (mg/g of wet wt tissue and % changes over the control in different tissues of *Bellamya bengalensis* (Lamarck, 1822) gastropod exposed to sub-lethal concentration of Endosulfan for 96 h) small size..

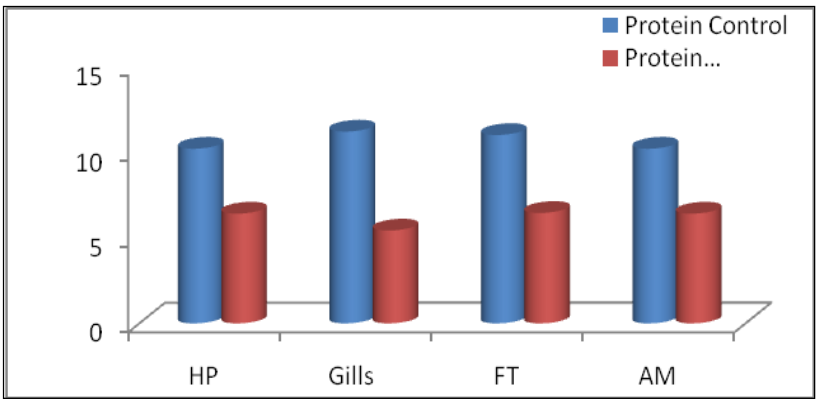


Table 2: Changes in the lipid content (mg/g of wet wt tissue and % changes over the control in different tissues of ***Bellamya bengalensis*** **(Lamarck, 1822)** gastropod exposed to sub-lethal concentration of Endosulfan for 96 h) small size

Sr. No.	Tissue	Lipid		decrease%
		Control	Experiment	
1.	Hepatopancreas	2.84± 0.231	0.170±.090	46.06
2.	Gills	2.90± 0.321	0.164±.040	46.2
3.	foot	3.09± 0430	0.153±.040	50.20
4.	Adductor muscle	3.22± 0.322	0.178±.050	44.08

Graph 3: Changes in the lipid content (mg/g of wet wt tissue and % changes over the control in different tissues of *Bellamya bengalensis* (Lamarck, 1822) gastropod exposed to sub-lethal concentration of Endosulfan for 96 h) small size

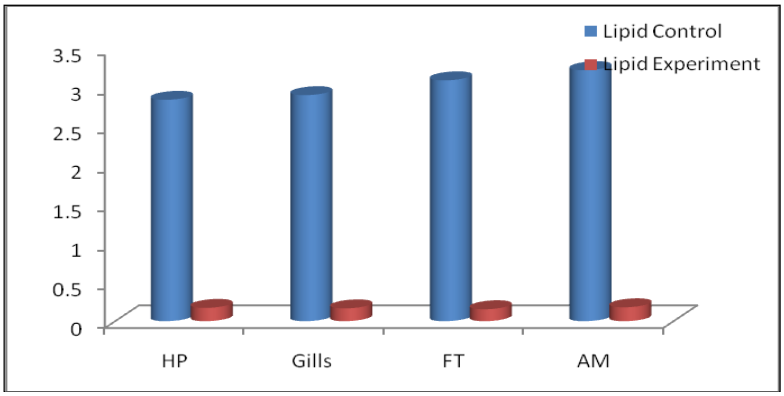


Table 3: Changes in the Glycogen content (mg/g of wet wt tissue and % changes over the control in different tissues of *Bellamya bengalensis* (Lamarck, 1822) gastropod exposed to sub-lethal concentration of Endosulfan for 96 h) small size

Sr. No.	Tissue	Glycogen		decrease%
		Control	Experiment	
1.	Hepatopancreas	4.32± 0.212	2.70±.050	44.02
2.	Gills	4.81± 0.214	2.85±.060	46.02
3.	foot	4.50± 0.232	2.88±.090	45.06
4.	Adductor muscle	4.42± 0.244	2.78±.080	44.02

Graph4: Changes in the Glycogen content (mg/g of wet wt tissue and % changes over the control in different tissues of *Bellamya bengalensis* (Lamarck, 1822) gastropod exposed to sub-lethal concentration of Endosulfan for 96 h) small size

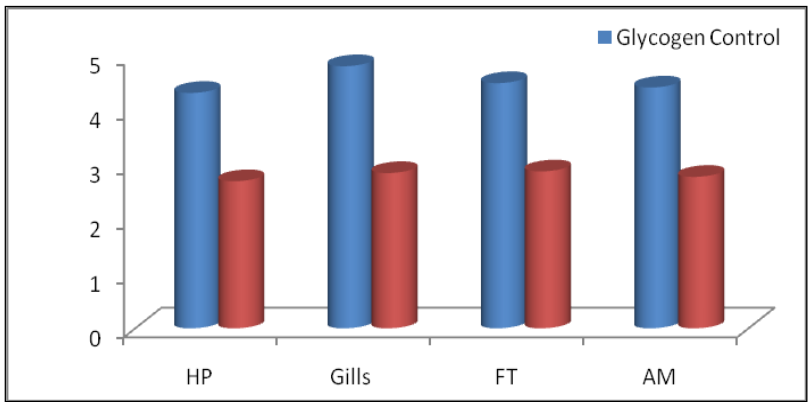


Table 4: Changes in the protein content (mg/g of wet wt tissue and % changes over the control in different tissues of *Bellamya bengalensis* (Lamarck, 1822) gastropod exposed to sub-lethal concentration of Endosulfan for 96 h) large size

Sr. No.	Tissue	Protein		decrease%
		Control	Experiment	
1.	Hepatopancreas	13.6±.221	7.03±.270	36.44
2.	Gills	12.5±.224	7.06±.030	36.22
3.	foot	16.5±.421	7.09±.060	36.05
4.	Adductor muscle	15.2±.420	5.02±.070	29.14

Graph 5: Changes in the protein content (mg/g of wet wt tissue and % changes over the control in different tissues of *Bellamya bengalensis* (Lamarck, 1822) gastropod exposed to sub-lethal concentration of Endosulfan for 96 h) large size.

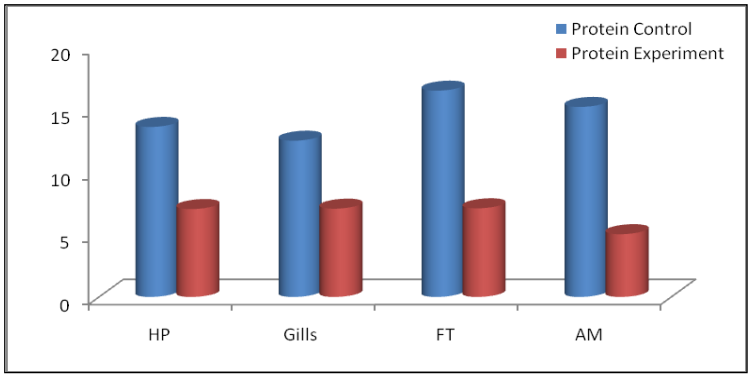


Table 5: Changes in the lipid content (mg/g of wet wt tissue and % changes over the control in different tissues of *Bellamya bengalensis* (Lamarck, 1822) gastropod exposed to sub-lethal concentration of Endosulfan for 96 h) large size

Sr. No.	Tissue	Lipid		Decrease%
		Control	Experiment	
1.	Hepatopancreas	2.53±.090	0.155±.120	41.55
2.	Gills	2.63±.060	0.174±.040	28.04
3.	foot	2.87±.202	0.186±.030	26.06
4.	Adductor muscle	2.63±.102	0.154±.040	29.06

Graph 5: Changes in the lipid content (mg/g of wet wt tissue and % changes over the control in different tissues of *Bellamya bengalensis* (Lamarck, 1822) gastropod exposed to sub-lethal concentration of Endosulfan for 96 h) large size

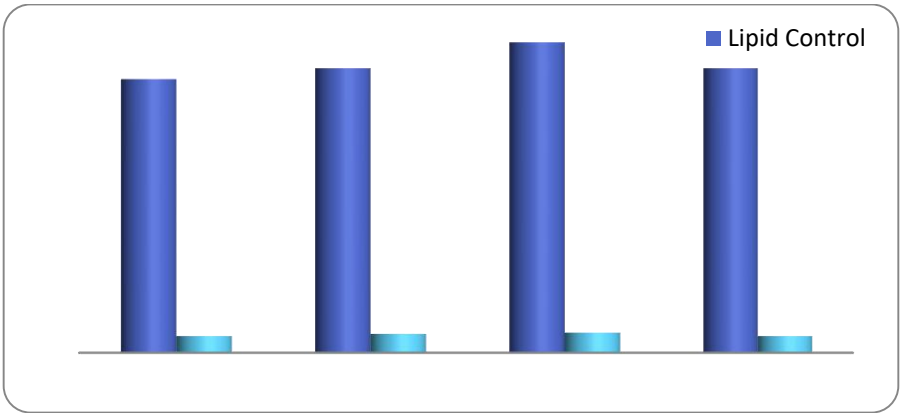
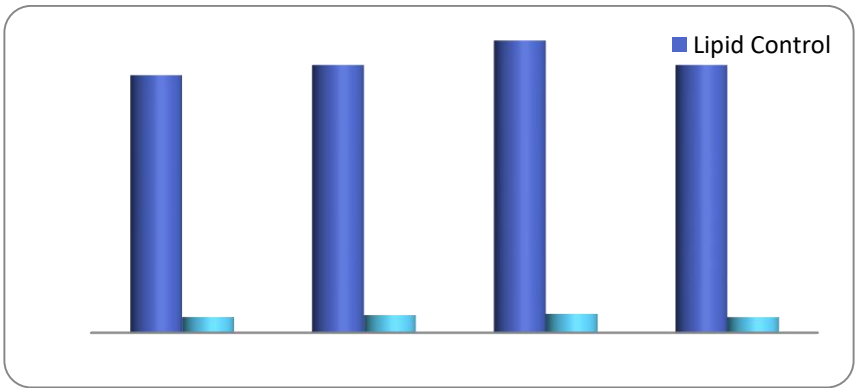


Table 6: Changes in the Glycogen content (mg/g of wet wt tissue and % changes over the control in different tissues of *Bellamya bengalensis* (Lamarck, 1822) gastropod exposed to sub-lethal concentration of Endosulfan for 96 h) large size

Sr. No.	Tissue	Glycogen		Decrease %
		Control	Experiment	
1.	Hepatopancreas	4.80±.201	2.40±.550	50.02
2.	Gills	4.40±.303	2.52±.460	48.06
3.	foot	4.50±.302	2.56±.320	49.05
4.	Adductor muscle	4.50±.320	2.41±.480	49.08

Graph 6: Changes in the Glycogen content (mg/g of wet wt tissue and % changes over the control in different tissues of *Bellamya bengalensis* (Lamarck, 1822) gastropod exposed to sub-lethal concentration of Endosulfan for 96 h) large size



The pattern of distribution of the metabolites in the control group is as follows – protein, glycogen, and lipid. In Viviparidae molluscs glycogen is stored in considerable amount in certain tissues, while in others it is insignificant (Hadfield and Paul, 2001; Colombo et. al, 2019). In the present study, it is observed that different portions of mantle of molluscs stored different amounts of glycogen, and middle mantle was found to be the most prominent glycogen storage. The two other portions of mantle, hepatopancreas and gonad also stored considerable amount of glycogen. Apart from glycogen in lamellibranch molluscs, lipids are generally considered as energy reserves during stress conditions. The stored lipid is used in preference to glycogen during such conditions. In caeruleus, the gonad is the most prominent organ to store lipid (Jimeno, 2002; Koueta and Boucaud-Camou 2003; Jansen and Groot, 2004; Benkendorff et. al., 2005; Ray and Sajal, 2018) .

Our histological observations revealed mature sex products in the follicles store more lipid. Hepatopancreas, middle mantle and foot also stored considerable amount of lipid. Nominal (7.2 ppb) and lethal (13.6 ppb) effected in changes in protein, glycogen, contents in *Bellamya bengalensis* exposure to fenthion lipid and cholesterol Animals exposed to 7.2 and 13.6 ppb Endosalphan showed significant decrease in protein content in body tissue as compared to control. The decrease was more in lethal exposure than nominal in body tissue. When compared which control, glycogen content in 7.2 ppb Endosalphan exposed animals decreased significantly in all of the body parts, except foot. In animals exposed to 13.6 ppb Endosalphan, the content decreased in all of the body parts, wherein the content increased than control. Compared to animals exposed to 7.2 ppb Endosalphan, glycogen content in those exposed to 13.6 ppb

Endosalphan showed a significant decrease in body tissue it showed a significant increase. Lipid content in body of animals subjected to nominal exposure decreased significantly, while in other body parts it significantly increased compared to control. In lethal exposure, other body parts showed significant increase in the content. Nominal and lethal exposure to Endosalphan caused changes in glycogen content of all the body parts of *Bellamya bengalensis*.

The breakdown of glycogen in most of the body parts is probably due to the inhibition of glucose-6-phosphatase or suppressed gluconeogenesis. This suggests that in these body parts glycogenolysis is inhibited or those gluconeogenesis and glycogen sis are increased to some extent resulting in an increased level of glycogen to keep the reserve energy in depot tissue and muscle (Capkin et al., 2006). Authors have shown that low levels of Endosalphan might usually stimulate certain physiological processes whereas higher dietary doses might cause inhibition or exhaustion (as related to an adaptive mechanism) in gastropod molluscs. Inhibition of glycogenolysis in rainbow trout exposed to Endosalphan (Mera71), and changes in serum protein and glycogen of rainbow trout exposed to Endosalphan (Grant and Mehrle 1973; Capkin et al., 2006) were reported earlier. In gastropod molluscs the conversion of glycogen into fatty acid or triglyceride reserves via trios phosphate entry in the glycolytic sequence and to the production of pentose sugars for nucleic acid synthesis as well as the necessary intermediates for lipogenesis is well documented (Gabbott 1976; Chaudhary et al., 1981; Rossi et al., 1993; Arakelova et al, 2004), while studying the long term exposure of technical grade Endosalphan on Heteropneustes fossils observed that the water and lipid contents of whole body decreased, while lipid content decreased. Recently, Swami et al. (1983) also suggested a shift in carbohydrate and protein metabolisms to lipid synthesis in freshwater gastropod exposed to pesticides/ herbicide.

The breakdown of protein increased more in lethal exposure of *Bellamya bengalensis* (Lamarck, 1822) in all the body parts due to Endosalphan stress. This breakdown along with the increase in glycogen content might be responsible for lipid alteration which might also be responsible for increased lipid content in hepatopancreas and both adductor muscles. A dual nature of disturbances in the reserves occurred in certain tissues. Thus, the possible mechanism for increase in lipid content in these body parts might be due to the increased lipid synthesis with changes in the type of synthesis, diminished degradation of lipid, increased mobilization of lipid and diminished transport of lipid away from these body parts. An alternative scheme for anaerobic metabolism in gastropod molluscs, in which redox balance is maintained by the simultaneous utilization of both carbohydrate and protein has been proposed by many researchers Krebs, (1972); Hochachka and Mustafa, (1972); Hochachka et al., (1973); Sadowska et al., (2003); Orban et al, (2007); Xie et al, (2012); Saito, (2014) and Tabakaeva, (2018) has shown that there is a significant loss of stored energy while converting pre-stored glycogen into lipid, lipid is a more concentrated form of energy and there is an increase in buoyancy due to its lower density when compared to carbohydrate and protein (Gabbott, 1976). The consistent decrease in glycogen and lipid in body tissues of Endosalphan exposed *Bellamya bengalensis* might account for their utilization for energy. Amongst lipids phospholipids are actively degraded (Harper et al. 1977; Saito,2014 and Tabakaeva, 2018). This is seen particularly in body tissue of *Bellamya bengalensis* exposed to both nominal and lethal concentrations of Endosalphan. Thus, the present study reveals dis-functions of physiological processes critical for the survival of *Bellamya bengalensis* exposed to Endosalphan. Certain body tissues that showed increase or decrease in glycogen and lipid contents due to Endosalphan toxicity may actually stimulate certain physiologic processes. However, special attention needs on the glycogen and lipid metabolic pathways disturbed due to pesticides in this gastropod mollusc like salinity temperature of the water in a freshwater and estuarine varies widely. As the estuarine is relatively shallow fluctuation of the atmospheric temperature affects the water. Over mud flats water is invariably shallow and the sun would heat up the water. As the river brings in a large load of suspended matter estuarine water is generally turbid. This turbidity affects the water circulation and feeding in filter-feeding animals, especially the sedentary species. Estuaries and backwaters are generally calm or still water areas and hence excessive sedimentation produces mud flats. These mud flats, particularly in the tropics, support of rich vegetation and mud-dwelling animals.

The pattern of distribution of the metabolites in the control group is as follows - protein : gonad > foot > anterior adductor muscle > hepatopancreas and posterior adductor muscle > posterior mantle > middle mantle > anterior mantle and gills; glycogen : middle mantle > posterior mantle and hepatopancreas > anterior mantle and gonad > anterior adductor and posterior adductor muscles > foot > gills; lipid: gonad > hepatopancreas > middle mantle > foot >~ anterior mantle > posterior mantle > anterior adductor muscle > posterior adductor muscle and gills; cholesterol: gonad > hepatopancreas > foot > gills > posterior mantle and anterior adductor muscle > anterior mantle and posterior adductor muscle > middle mantle

In Viviparidae molluscs glycogen is stored in considerable amount in certain tissues, while in others it is insignificant (Giese 1969). In the present study, it is observed that different portions of mantle of molluscs stored different amounts of glycogen, and middle mantle was found to be the most prominent glycogen storage. The two other portions of mantle, hepatopancreas and gonad also stored considerable amount of glycogen. Apart from glycogen in lamellibranches molluscs, lipids are generally considered as energy reserves during stress conditions. The stored lipid

is used in preference to glycogen during such conditions. In *Bellamya bengalensis,* the gonad is the most prominent organ to store lipid. Our histological observations revealed mature sex products in the follicles store more lipid. Hepatopancreas, middle mantle and foot also stored considerable amount of lipid. Nominal (7.2 ppb) and lethal (13.6 ppb) effected in changes in protein, glycogen, contents in *Bellamya bengalensis.* Exposure to fenthion lipid and cholesterol Animals exposed to 7.2 and 13.6 ppb Endosulfan n showed significant decrease in protein content in middle mantle, posterior mantle, gonad, foot and anterior adductor muscle compared to control. The decrease was more in lethal exposure than nominal in posterior mantle, gills, hepatopancreas and gonad. 623 When compared with control, glycogen content in 7.2 ppb fenthion exposed animals decreased significantly in all of the body parts, except foot. In animals exposed to 13.6 ppb fenthion, the content decreased in all of the body parts, except hepatopancreas and anterior adductor muscle, wherein the content increased than control. Compared to animals exposed to 7.2 ppb fenthion, glycogen content in those exposed to 13.6 ppb fenthion showed a significant decrease in gonad, foot and posterior adductor muscle; in anterior mantle, middle mantle, posterior mantle, gills, hepatopancreas and anterior adductor muscle it showed a significant increase. Lipid content in gonad and foot of animals subjected to nominal exposure decreased significantly, while in other body parts it significantly increased compared to control. In lethal exposure, gonad showed a significant decrease in the content compared to control. Other body parts showed significant increase in the content. Anterior mantle and middle mantle of animals exposed to 7.2 ppb fenthion showed a significant decline in lipid content, whereas posterior mantle, gills, hepatopancreas, gonad, foot and the adductor muscles showed a significant increase in the content than those exposed to 13.6 ppb fenthion. Compared to control, hepatopancreas and foot of the animals exposed to 7.2 ppb Endosulfan showed a significant decrease in cholesterol content, whereas middle mantle, posterior mantle and gills showed a significant increase. In animals exposed to 13.6 ppb fenthion, a significant increase in cholesterol content, than control, was observed in anterior mantle, middle mantle, gills, hepatopancreas, anterior adductor and posterior adductor muscles. Animals exposed to 13.6 ppb fenthion showed a significant increase in cholesterol content in anterior mantle, hepatopancreas, foot and posterior adductor muscle than those exposed to 7.2 ppb fenthion. In posterior mantle and gonad of the animals exposed to 13.6 ppb fenthion, cholesterol content decreased compared to 7.2 ppb Endosulfan exposed animals. Nominal and lethal exposure to enthion caused changes in glycogen content of all the body parts of I. caeruleus. The breakdown of glycogen in most of the body parts is probably due to the inhibition of glucose-6-phosphatase or suppressed gluconeogenesis. The body parts like hepatopancreas, anterior adductor muscle (in lethal exposure) and foot (in nominal exposure) showed increase in the glycogen content. This suggests that in these body parts glycogenolysis is inhibited or those gluconeogenesis and glycogensis is increased to some extent resulting in an increased level of glycogen to keep the reserve energy in depot tissue and muscle. These authors have shown that low levels of endrin might usually stimulate certain physiological processes whereas higher dietary doses might cause inhibition or exhaustion (as related to an adaptive mechanism) in gastropod. Inhibition of liver glycogenolysis in rainbow trout exposed to endrin (Menzie 1972; Wright et al., 1989; Rossi et al., 1993; Arakelova et al, 2004), increase in liver 624 glycogen of *Scorpaena porcus* exposed to lindane (Escoubet and Vincente 1975; Kirsten Benkendorff, 2010) and changes in serum protein and glycogen of rainbow trout exposed to endrin (Grant and Mehrle 1973) were reported earlier. In lamellibranch molluscs the conversion of glycogen into fatty acid or triglyceride reserves via triose phosphate entry in the glycolytic sequence and to the production of pentose sugars for nucleic acid synthesis as well as the necessary intermediates for lipogenesis is well documented (Gabbott 1976). Chaudhary et al. (1981) and Kirsten Benkendorff (2010), while studying the long term exposure of technical grade Malathion on Heteropneustes fossils observed that the water and lipid contents of whole body and ovary decreased, while liver lipid content decreased and liver water content increased. Swami et al. (1983) Ray, Sajal (2017) also suggested a shift in carbohydrate and protein metabolisms to lipid synthesis in freshwater mussels exposed to pesticides.

4. CONCLUSION:

The gastropod shell index values in Sadatpur Lake (from this study) are slightly contaminated. All the indications are strongly suggested that there had been some relatively recent inputs of Endosalphan (Mera 71) and Roger. This degree of contamination is very unlikely to have any effect on gastropod molluscs particularly *Bellamya bengalensis*. It is suggested that the possible source of contamination, though the farm and domestic need may also have contributed to this contamination. It is remarkable that a population of viviparous sp. survives in the reservoir of Sadatpur. The strong effluents of Canal of Pravara River in this region may rapidly disperse any possible contamination. Levels of contaminations are low. It is possible that there is good circulation of the water that results in a high dilution of Mera-71 and roger if present. Indications of contamination are low, with a possible small rise in the region Sadatpur. So the percussion must have to be taken on these pesticides.

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