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Effect of heavy metals on SDH and LDH enzymes activity of Bivalve Lamellidens marginalis…..

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Abstract: *Enzymes are referred as biological, catalysts which obey certain general rules. . The literature shows investigative lacunae with regard to effects of heavy metals on SDH and LDH activity of aquatic animals especially where fresh water molluscs are concerned. Hence an attempt has been made to study the effect of heavy metals $CuSO_4$, $HgCl_2$ and $CdCl_2$ on fresh water bivalve, Lamellidens Marginalis with respect to change in the level of SDH and LDH enzymes. The depletion in the enzymatic activity may be due to the damage caused by the heavy metal to the cells of the alimentary canal. The important metabolic enzymes in the Kreb's TCA cycle like SDH and LDH were studied with respect to the heavy metal stress. It was found that the activity of SDH and LDH was depleted significantly after the heavy metal stress.*

Keywords*: Heavy metals $CuSO_4$, $HgCl_2$ and $CdCl_2$, L. marginalis, LDH, SDH, enzymes, acute, chronic.*

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I. INTRODUCTION

Enzymes are referred as biological, catalysts which obey certain general rules. The enzyme catalyzed reactions take place at physiologically low temperatures (37^0C) and require extremely small amounts of enzymes. Chemically, enzymes are complex protein molecules synthesized in the cells where they act as biocatalysts in carrying out various physic-chemical reactions. These proteins have their own specificity and kinetics. The enzymes help in attaining a reaction in a state of equilibrium. An enzyme recognizes its specific substrate and reacts with it to form products and gets regenerated at the end of the reaction.

The usual measure applied for the assessment of any environmental effect of a pollutant on animal is mortality. However, other effects which are gradual and are indicative of physiological change can be as detrimental as mortality to the animal's survival. Although instantaneous effects of heavy metal poisoning may be physical such as retraction of animal in the shell, loss of locomotory activity, changes is normal behaviour, reproductive disorders, suffocation, coating of respiratory surface with mucus, long term effect would be exclusively due to physiological alterations.

The most fundamental effects would be the study of changes in enzyme levels, since these organic cellular catalysts control the formation of biochemical intermediates which are indispensable to all normal physiological processes.

Molluscs have exhibited ability to adapt themselves to life in so many different types of habitats. It is not surprising that they have learnt to feed in different ways. They show a variety of digestive patterns. The correlation between digestive enzymes and diet has been established but specific characterization of different enzymes of different animals presents many interesting and puzzling questions (Prosser 1973). Synergistic effect of heavy metals present in the electroplating industry waste water on the enzymatic activity in the vital organs of fish, Oreochromis mossambicus, (Navaraj Permalsamy, Kumaraguru Arumugam 2013).

Heavy metals are pervasive components of the aquatic environment, including fresh waters. The gross effects of these components have been substantially documented in both marine and fresh water. Yet the extent of physiological damage resulting from high and low level heavy metal contamination of fresh water and the harm that is done to aquatic animals, has not been adequately described, though there are few studies which indicate the effect of heavy metals on aquatic organisms (Hewitt and Nicholas, 1963). A change in enzymatic pathway is another approach which though yet untried could potentially be used in toxicant analysis. Exposure to metals might in some way alter the enzyme and change its response in co-factors, temperature, pH and also its micheli's constant (Jackim, 1974). LDH activity was found to be enhanced by copper treatment, while lead treatment inhibited the LDH activity. All the enzymes showed high activity in the intestinal diverticula, (Aanand S. Purushothaman C.S, Pal A.K.2010)

The mollusc has surprising enzyme equipment. Indeed there seems to be no other group in the animal kingdom with such an array of digestive enzymes. Particularly the carbohydrases.

Kreb's citric acid cycle is the final and common pathway for the oxidation of carbohydrates, proteins, lipids, since glucose, amino acids and fatty acids are metabolized to acetyl Co-A and then acetyl Co-A is oxidized to Co_2 and water through a series of metabolic steps. The oxidation of acetyl Co-A reduces equivalents in the form of electrons. These electrons are released due to activity of a group of specific enzymes known as dehydrogenases. This dehydrogenase catalyses the formation of high energy phosphate bond through a process of oxidative phosphorylation (Campbell, 1973). The energy, thus generated is most important for the organism. Any disruption or alteration in the activity of these enzymes of the citric acid cycle may therefore, disturb the entire physiological equilibrium resulting in complications of various natures.

Poison of any nature, whether pesticides, salts of heavy metals can cause ultra structural change in the mitochondria, endoplasmic reticulum etc. and inhabit the enzymes of TCA cycle. Miroslaw (1973) reported depletion in the level of hepatic SDH in rats when poisoned with Malathion. The depletion of SDH and increased LDH activity were reported by Bhagyalaxmi (1981) in the crab, Oxiotelphusa Senex Senex when treated with sumithion, Shrivastava et al. (1977) in the fish Colisa fusciatus and Kaundinya and Ramaurthi (1978) in Tilapia mossambica observed altered oxidative metabolism after exposure to organophosphate pesticides. Kabeer Ahmed (1979) in the snail, Pila Globosa observed changes in oxidative metabolism after exposure to organophosphate pesticide. Alam (1984) in the snail. V. Bengalensis observed alteration in oxidative metabolism after exposure to salts of heavy metals like Cu, Hg and Zn.

II. MATERIAL AND METHODS

The bivalves Lamellidens Marginalis were collected from the Godavari river at Paithan. After bringing the bivalves to the laboratory, they were cleaned thoroughly and placed in plastic troughs. They were acclimatized to the laboratory conditions for 5 to 6 days prior to subjecting them to experiments. The water in the troughs was changed every day. Only active and healthy animals were chosen for experiments. During chronic treatment the animals were fed on crushed fresh water algae and Hydrilla.

The bivalve was exposed to median lethal concentration and sub lethal concentration of pollutant as acute and chronic treatment respectively.

The acclimatized bivalves were divided into four groups, of ten each. The first groups of bivalves were kept as control. The remaining groups were exposed to 1.6 ppm $Cuso_4$, 0.6 ppm $HgCl_2$ and 3.9 ppm cadmium chloride for 72 hours, for acute treatment. The concentrations used for chronic exposure were 0.82 ppm copper sulphate, 0.32 ppm mercuric chloride and 1.95 ppm cadmium chloride. The chronic treatment was given upto 20 days. The control and treated bivalves were fed on freshwater crushed algae and Hydrilla during exposure period.

Estimation of Succinic dehydrogenase (SDH) :

SDH was determined by the method of Nacholas et al; (1960), 2% (weight / volume) homogenate was made in 0.25 M ice cold sucrose solution and centrifuged at 2500 rpm for 15 minutes. The clear supernatant fraction was used for the assay of the enzyme activity.

The reaction mixture is a final volume of 2.0 ml which contained, 40 u moles of sodium succinate, 100 u moles of sodium phosphate buffer (pH 7.4) and 4 u moles of INT. The reaction was initiated by the addition of 0.5 ml of homogenate. The incubation was carried out at 37^0C for 30 minutes in a thermostat, after which 5.0 ml of glacial acetic acid was added which stopped the enzyme reaction. The formazon formed was extracted into 500 ml of toluene overnight at 5^0C. The colour of the formozan was measure at 495 um in a colorimeter against a toluene blank. SDH activity was expressed as u moles of formazon formed/mg protein/hour.

Estimation of lactate dehydrogenase (LDH) :

The LDH activity was determined by the method of Necholas et al. (1960). The tissue was homogenized (5% weight / volume) in 0.25 ml ice cold sucrose solution centrifuged at 2500 rpm for 15 minutes and the supernatant was used for enzyme assay. The reaction mixture is a final volume of 2.0 ml which contained 10 u moles of phosphate buffer (pH 7.4), 40 u moles of INT. The reaction was started by the addition of 0.4 ml of homogenate. The reaction mixture was incubated at 36^0C for 30 minutes and the reaction was stopped by the addition of 5.0 ml of glacial acetic acit. The formazon formed was extracted into 5.0 ml of toluene against a toluene black of zero hour control. The enzyme activity was expressed as u moles of formazon formed/mg protein/hour.

III. OBSERVATIONS AND RESULT-

The activities of different enzymes were studied in the normally fed and pollutant (heavy metals) treated freshwater bivalve, Lamellidens marginalis. The experimental findings obtained are summarized in Tables.

Metabolic enzymes:

Succinate dehydrogenase (SDH) - The SDH activity in control bivalves was found to be 13.302, 12.452 and 10-789 n moles of formazon /mg protein/hr in digestive gland, foot and mantle respectively. A significant depletion in SDH activity was observed after all the heavy metal exposures.

Acute exposure to cuso4 also exhibited SDH activity in all tissues of the bivalve L. marginalis. The values of SDH activity were found to be 8.830, 10-002, and 9.00 u moles of formazon/mg protein/hr after 72 hours exposure in digestive gland, foot and mantle respectively. Similar trend was observed in chronic treatment also in the three tissues. The values were 9.813, 9.303 and 7.832 u moles l of formazon/mg protein/mg protein/hr respectively at the end of 20 days.

$HgC1_2$ acute stressed showed greater decrease in SDH activity in mantle and foot than digestive gland.

In mantle the activity was found to be 9.000, 7-003, 7-862 U moles of formazon/mg of formazon /mg protein/hr after 72 hrs. Of $CuSO_1$, $NgCl_2$, respectively. But in chronic exposure of $HgC1_2$, SDH activity decreased more in foot than digestive gland and mantle. The values were 5.12 u 6.675 moles of formazon/mg protein/hr of formazon/mg protein/hr in mantle at the end of 20 days.

In acute treatment. $CdC1_2$, reduced the SDH activity in digestive gland, foot, and mantle. The values of SDH activity were 8.503, 8.150, and 7.862 u moles of respectively after 72 hours. Similar reduction was also values were 11.113. 8.930 and 8.123 u moles of formazon/mg protein/hr after 20 days in digestive gland foot and mantle respectively.

The maximum percentages decrease was upto 53.12% (72 hrs $HgC1_2$ stress) and 57.50% (20 days $HgC1_2$ stress). All 0.01 and $P < 0.001$ levels.).

Lactate dehydrogenase (LDH) :

In the control animals LDH activity observed was 16.521, 6.692 and 10.421 u moles of formazon/mg protein/hr in digestive gland, foot and mantle respectively. Acute stress of $Cuso_4$ drastically altered the activity in foot mantle compared to digestive gland. In digestive gland the activity, was found to be 6.432 u moles of formazon / mg protein/hr at the end of 72 hrs. exposure. In foot, the activity was found to be 2.132 u moles of formazon/mg protein/hr after 72 hours and in mantle 4.923 u moles of formazon/mg protein/hr at the end of 72 hours exposure. Similar trend was also observed in chronic exposure. The value were 14.540, 2.780 ad 10.180 u moles of formazon/mg protein/hr in foot, digestive gland and mantle respectively after 20 days.

$HgCl_2$ stress in acute treatment showed a drastic decrease in activity in foot, compared to digestive gland and mantle. The values were 7.430, 1.532 and 3.792 u moles of formazon/mg protein/hr in digestive gland, foot and mantle respectively. Chronic treatment of mercuric chloride showed more decreased activity in mantle than digestive gland and foot. The values were 12.523, 2.202 and 4.913 u moles 1 of formazon/mg protein/hr in digestive gland, foot and mantle respectively, mercuric chloride showed a drastic decrease in activity in mantle after chronic exposure.

Treatment of $CdCl_2$, also greatly reduced the activity of LDH in foot compared to digestive gland and mantle. The activities were 7.600, 1.823, 5.113 u moles of formazon/mg protein/hr in digestive gland, foot and mantle respectively after 72 hours of exposure chronic treatment of $CdCl_2$ also exhibited decrease in LDH activity in foot and mantle of the bivalve L.marginalis . But in digestive gland LDH activity increased after 5 and 10 days exposure to $CdCl_2$ and then it went on decreasing after 15 and 20 days. The values were 17.780, 17.530 u moles of formazon/mg protein/hr after 5 to 10 days exposure respectively. The maximum percentage increase was upto 0.4035% (5 days exposure of $CdCl_2$ Stress). The increased activity at 10 and 20 days exposure to $CdCl_2$ was not significant while all other values were significant at $P<0.05$, $P<0.01$ and $P<0.001$ level

Table – 1 Level of SDH activity in the bivalve, L.marginalis after acute treatment of heavy metals Copper sulphate, Mercuric chloride and Cadmium chloride

Pollutant	**Exposure Period (in hrs)**	**Digestive**		**Foot**		**Mantle**	
1	**2**	**3**		**4**		**5**	
Control	0	13.032±0.0024		10.456±0.0042		9.120±0.0071	
$CuSO_4$	24 hrs	11.500±**0.0052** **P<0.001**	-19.80	10.456±**0.0042** **P<0.001**	-30.32	9.120±**0.0091** **P<0.001**	-30.17
	48 hrs	9.976±**0.0070** **P<0.05**	-40.40	10.036±**0.0051** **P<0.001**	-39.20	9.003±**0.0041** **P<0.001**	-33.11
	72 hrs	8.830±**0.0040** **P<0.001**	-60.88	10.002±**0.0082** **P<0.001**	-47.16	9.00±**0.006** **P<0.001**	-40.13
$HgCl_2$	24 hrs	12.600±**0.0042**	-20.40	9.432±**0.0052**	-30.40	7.231±**0.0091**	-30.41

		P<0.001		**P<0.001**		**P<0.001**	
	48 hrs	10.212±**0.0052** **P<0.05**	-36.21	8.732±**0.0010** **P<0.001**	-40.21	7.120±**0.0045** **P<0.001**	-40.13
	72 hrs	8.172±**0.0030** **P<0.001**	-62.11	8.000±**0.0032** **P<0.001**	-53.12	7.003±**0.0030** **P<0.001**	-40.50
$CdCl_2$	24 hrs	10.104±**0.0032** **P<0.001**	-70.90	8.784±**0.0038** **P<0.001**	-19.20	7.345±**0.0012** **P<0.001**	-22.00
	48 hrs	9.560±**0.0020** **P<0.05**	-78.23	8.993±**0.0029** **P<0.001**	-12.92	7.642±**0.0051** **P<0.01**	-28.21
	72 hrs	8.503±**0.0025** **P<0.001**	-85.20	8.150±**0.0028** **P<0.001**	-20.23	7.862±**0.0032** **P<0.001**	-30.22

SDH activity is expressed as ug of Pi/mg. protein/hr
Each value is the mean of five observations ± S.D.
Values are significant at P<0.001 and P< .5 level
-ve sign indicate % inhibition

Table – 2 Changes in the LDH activity in the bivalve, L.marginalis after acute exposure of heavy metals, Copper sulphate, Mercuric chloride and Cadmium chloride

Pollutant	**Exposure Period (in hrs)**	**Digestive gland**		**Foot**		**Mantle**	
1	**2**	**3**		**4**		**5**	
Control	0	16.521±0.2432		6.692±0.005		10.42±0.0042	
$CuSO_4$	24 hrs	11.232±**0.020** **P<0.05**	-30.08	3.345±**0.0042** **P<0.001**	-50.72	7.651±**0.0042** **P<0.001**	-40.20
	48 hrs	10.242±**0.22** **P<0.001**	-40.22	2.523±**0.0043** **P<0.05**	-60.76	6.242±**0.0023** **P<0.05**	-56.30
	72 hrs	6.432±**0.21**	-72.52	2.132±**0.0021**	-92.45	4.923±**0.0022**	-79.15
$HgCl_2$	24 hrs	10.300±**0.22** **P<0.05**	-25.10	3.000±**0.0062** **P<0.001**	-52.11	7.002±**0.0050** **P<0.001**	-45.13
	48 hrs	9.502±**0.27** **P<0.001**	-40.00	2.001±**0.0042** **P<0.05**	-80.13	5.812±**0.038** **P<0.05**	-70.90
	72 hrs	7.430±**0.25** **P<0.001**	-78.12	1.532±**0.0062** **P<0.001**	-90.01	3.792±**0.0029** **P<0.05**	-80.20
$CdCl_2$	24 hrs	13.923±**0.21** **P<0.01**	-3.40	4.120±**0.0040** **P<0.001**	-30.18	9.342±**0.0032** **P<0.05**	-30.17
	48 hrs	11.430±**0.20** **P<0.05**	-19.20	3.430±**0.0072** **P<0.05**	-60.30	7.430±**0.0021** **P<0.001**	-40.61
	72 hrs	7.600±**0.22** **P<0.001**	-50.13	6.823±**0.0032** **P<0.001**	-77.19	5.113±**0.0030** **P<0.001**	-63.30

LDH activity is expressed as u moles of formazon formed/gm protein/hr
Each value is a mean of five observations ± S.D.
Values are significant at P<0.01, P<0.05 and P<0.001 level

Table – 3 Level of SDH activity in the tissues of bivalve L.marginalis after chronic exposure of heavy metals copper sulphate, Mercuric chloride and Cadmium chloride

Pollutant	**Exposure Period (in Days)**	**Digestive gland**		**Foot**		**Mantle**	
1	**2**	**3**		**4**		**5**	
Control	5 10 15 20	12.785±0.0008 12.832±0.00122 12.987±0.0087 12.982±0.0074		11.997±0.0013 11.884±0.0019 11.885±0.0020 11.887±0.0023		10.991±0.0037 10.997±0.0012 10.972±0.0047 10.845±0.0042	
$CuSO_4$	5	10.512±**0.0021** **P<0.05**	-12.65	10.532±**0.0017** **P<0.05**	-10.31	8.823±**0.00293** **P<0.05**	-13.65
	10	10.421±**0.0052** **P<0.05**	-17.21	10.443±**0.0015** **P<0.05**	-15.20	8.338±**0.0019** **P<0.05**	-18.42

	15	9.903±**0.0020** **P<0.05**	-22.13	9.930±**0.005** **0.001**	-21.11	7.997±**0.0032** **0.001**	-28.20
$HgCl_2$	5	8.234±**0.0010** **P<0.05**	-30.13	8.210±**0.0032** **P<0.05**	-30.71	8.310±**0.0037** **P<0.001**	-20.68
	10	8.203±**0.0020** **P<0.05**	-35.30	8.190±**0.0052** **P<0.05**	-40.30	8.199±**0.0017** **P<0.001**	-30.13
	15	8.001±**0.0017** **P<0.05**	-40.45	7.789±**0.0019** **P<0.05**	-51.32	7.799±**0.0011** **P<0.05**	-37.1
	20	6.123±**0.0015** **P<0.05**	-57.50	6.630±**0.0023** **P<0.05**	-53.10	0.675±**0.0034** **P<0.05**	39.18
$CdCl_2$	5	11.832±**0.0012** **P<0.001**	-89.08	9.732±**0.0011** **P<0.001**	-17.92	9.422±**0.0032** **P<0.001**	-9.123
	10	11.829±**0.0022** **P<0.001**	-90.14	9.145±**0.0020** **P<0.05**	-20.39	8.777±**0.0016** **P<0.001**	-10.987
	15	11.430±**0.0032** **P<0.001**	-90.23	8.889±**0.0017** **P<0.001**	-27.38	8.234±**0.0034** **P<0.05**	-15.68
	20	11.113±**0.0009** **P<0.05**	-93.34	8.530±**0.0012** **P<0.001**	28.13	8.23±**0.0023** **P<0.001**	-20.13

SDH activity is expressed as u moles of formazon formed/mg protein/hr.
Each value is a mean of five observation ± SD.
Values are significant at P<0.05, P<0.001 level
-ve sign indicates % inhibition.

Table -4 Level of LDH activity in the tissue of bivalve L.marginalis after chronic exposure of heavy metals, copper sulphate, Mercuric chloride and Cadmium chloride

Pollutant	**Exposure Period (in Days)**	**Digestive gland**		**Foot**		**Mantle**	
1	**2**	**3**		**4**		**5**	
Control	5 10 15 20	17.413±0.0032 17.424±0.0019 17.420±0.0018 17.630±0.0015		5.530±0.0013 5.528±0.0012 5.529±0.0014 5.522±0.0009		13.334±0.0004 13.413±0.0012 13.379±0.0008 13.383±0.0025	
$CuSO_4$	5	15.532±**0.0005** **P<0.05**	-8.80	3.390±**0.0015** **P<0.01**	-30.08	10.500±**0.0020** **P<0.05**	-5.30
	10	15.503±**0.0012** **P<0.001**	-15.30	3.340±**0.0028** **P<0.001**	-39.30	10.490±**0.0016** **P<0.05**	-7.80
	15	14.892±**0.0018** **P<0.001**	-23.20	2.999±**0.0018** **0.001**	-45.42	10.900±**0.0011** **P<0.001**	-14.55
	20	14.540±**0.0022** **P<0.001**	-28.18	2.780±**0.0012** **0.001**	-47.38	10.180±**0.0025** **P<0.05**	-18.30
$HgCl_2$	5	13.340±**0.0023** **P<0.05**	-20.40	2.789±**0.0017** **P<0.001**	-42.30	9.189±**0.0017** **P<0.05**	-28.30
	10	13.309±**0.0011** **P<0.05**	-25.55	1.630±**0.0031** **P<0.05**	-52.39	8.889±**0.0011** **P<0.05**	-40.34
	15	12.980±**0.0050** **P<0.05**	-32.01	2.221±**0.0021** **P<0.05**	-60.42	6.332±**0.0010** **P<0.05**	-61.92
	20	12.523±**0.0010** **P<0.05**	-32.70	2.202±**0.0007** **P<0.05**	-69.71	6.332±**0.0010** **P<0.05**	-61.92
$CdCl_2$	5	17.780±**0.0037** **P<0.001**	+.4035	4.730±**0.0019** **P<0.05**	-1.089	11.357±**0.0002** **P<0.001**	-0.1743
	10	17.530±**0.0027** **NS**	+.03256	4.550±**0.0023** **P<0.05**	-3.783	11.289±**0.0016** **P<0.001**	-8.521
	15	16.247±**0.0015** **NS**	-.05846	4.330±**0.0012** **P<0.001**	-5.941	11.180±**0.002** **P<0.05**	-8.973
	20	16.090±**0.0035** **NS**	-2.0423	4.129±**0.0010** **P<0.001**	-8.830	10.990±**0.0015** **P<0.05**	-9.549

LDH activity is expressed as u moles of formazon formed/mg protein/hr.
Each value is a mean of five observations ± SD.
Values are significant at P<0.01, P<0.001, P<0.05 level or NS = Not Significant
-ve sign indicates % inhibition.

IV. DISCUSSION

Out of the many devices which can be applied to investigate the physiological alterations made by the pesticide and heavy metal treatment, the most fundamental one would be the study of changes in the enzyme activities, since these organic cellular catalysts control the formation of biochemical intermediates which are indispensible to all the normal physiological processes.

There is no doubt that the effect of certain metals is profound on the enzyme activity in aquatic organisms though the mode of action of these heavy metals has not been clearly outlined with regard to their mechanism of action in certain key enzymes which are responsible for the general energetic of animals. There is a considerable amount of literature devoted to the study of organic pesticides concerning the enzyme system of various animals (Yap et al., 1975; Koundinya and Ramamurthi, 1978; Natrajan, 1981; Shastry and Malik, 1981 and Dalela et al., 1982). However, information regarding the effect of heavy metals on the enzyme action is restricted (Hewitt and Nicholas 1963 and Jackim, 1974).

The succinic and lactate dehydrogenase are important enzymes of Kreb's cycle whose qualitative changes are significant during certain pathological conditions (Hari et al., 1970 and Harper et al., 1978). The percentage of alteration produced in the activity of these enzymes can be a physiological measure of the degree of inhibition of the glycolytic pathway and hence the normal metabolism of the animal. It is evident from the Table 5, 6, 7 and 8, 9, 10 that the activity of the enzymes is greatly inhibited in all the tissues of Lamellidens Marginalis, maximum inhibition being observed in the digestive gland which indicates its greater vulnerability to pollutant stress. Similar observations have been reported by Alam (1984) in Viviparous Bengalensis when exposed to copper, mercury and cadmium.

SDH is the oxidative enzyme which was drastically affected by the action of heavy metals. Inhibition in the activity of succinic dehydrogenase is incubation, the shift of the metabolic pathway towards the anaerobic direction to meet the increased energy demands during pollution stress. Depletion level of SDH activity usually denotes the various pathological conditions (Mairo et al., 1973) since the osmoregulatory mechanisms are also dehydrogenase (Bashamohideen and Parwateshwar Rao, 1979), it's disruption may be responsible for osmoregulatory failure also.

The inhibitory effects of heavy metals on SDH activity also point to the degree of disturbances of mitochondrial integrity. Miroslaw (1973) reported the alteration in the mitochondrial structure after pesticide exposures. Similar results were also obtained by Koundinya and Ramamurthi (1978) and Shivprasad Rao and Ramana Rao (1979) in fishes. Swami et al. (1983) in the bivalve, Bhagyalakshmi (1981) in the crab and Alam (1984) in the snail, V. Bengalensis when exposed to the pesticides.

Decrease in SDH activity indicates a generally inhibited mitochondrial oxidation of succinate, which may lead to a drop in energy production. Since heavy metal exposure is a very serious kind of adverse effect and is classified as stress, mitochondrial alterations are bound to take place on a very large scale which are exhibited in the total disrupted of all biochemical processes including enzymatic reactions. The inhibition of SDH activity observed in case of L. Marginalis after heavy metal exposure is similar to the findings of Kabeer et al., (1978) in fresh water mussels exposed to organophosphates and to those of Natarajan (1981) in the fish, Channa Striatus and Alam (1984) in case of Viviparous Bengalensis.

The enzyme LDH plays an important role in the carbohydrate metabolism. In the present study LDH activity of the digestive gland, foot and mantle exposed to $CuSO_4$, $HgCl_2$ and $CdCl_2$ showed a significant decrease which was time dependent. Similar observations were also reported by Alam (1984) in V. Bengalensis, when exposed to different heavy metal salts. Inhibition of LDH activity ultimately results in the accumulation of lactic acid which is an end product of anaerobic glycolysis. It is logical to assume that heavy metal stress inhibits cellular oxidation which is evident from the suppression in SDH and LDH activity. Reduced mitochondrial activity was reported as a cause of inhibition of oxidoreductases in fish blue gill liver mitochondria by Hiltibran (1974). Such a mechanism might have occurred in Lamellidens also, after heavy metal treatment. During chronic exposure, the activites of SDH & LDH did not show a vast difference from the control in all tissues after 5, 10, 15 and 20 days. This indicates that, the anaerobic activity of the cell due to pollution stress has been reversed, an example of physiological and biochemical adaptation. This decrease in activity might be suggestive of weakening of biochemical difference which in turn could be the result of nervous disorder that has been reported in many findings with organic pesticides (Bhagyalakshmi, 1980; Kabeer Ahmed et al., 1980; Rajender 1981; and Bodkhe, 1983). These pesticides inhibit the activity of the enzyme acetylcholine terase and nervous transmission is retarded. Heavy metal might can be a similar way and since all the activities are controlled by the nervous system in an animal, it is possible that fall in the level of enzyme

activity might be the result of the nervous weakening. Dragomiresceu et al. (1975) have suggested that inhibitory effect of organophosphate compounds on certain dehydrogenase may be due to the repressor effect on their synthesis of due to the formation of enzyme inhibitor complexes which are not easily dissociable thereby reducing the activity of enzyme. Such a mechanism can also occur in the case of heavy metal toxicity. The depletion in LDH and SDH activity obtained after heavy metal stress in the bivalve L. Marginalis is similar to the finding of Alam (1984) in the snail, V. Bengalensis exposed to heavy metal salts $CuSO_4$ and zinc sulphate.

V. SUMMARY

.The influence of different heavy metals, copper sulphate, mercuric chloride and cadmium chloride on the enzymatic activity of the bivalve, Lamellidens Marginalis was observed.

.The depletion in the enzymatic activity may be due to the damage caused by the heavy metal to the cells of the alimentary canal.

.The important metabolic enzymes in the Kreb's TCA cycle like SDH and LDH were studied with respect to the heavy metal stress. It was found that the activity of SDH and LDH was depleted significantly after the heavy metal stress.

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