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LDH AND SDH ACTIVITY IN FRESHWATER FISH *CTENOPHYRYNGODON IDELLA* EXPOSED TO PHENTHOATE 50%EC (AN ORGANOPHOSPHATE)

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Abstract:

The pesticides and fungicides have been recognised as one of the serious pollutants of the aquatic ecosystems with deleterious effects on the living resources. Many pesticides and fungicides reporrted to produce a number of biochemical changes in fish both at lethal and sublethal levels. Present study phenthoate exposed to *Ctenophanrydon idella* in sublethal concentrations for 24h, 5and 10days of test fish in different tissues Under sub-lethal and lethal exposure to Phenthoate for 24h, the percentage of LDH depletion was found in all the tissues of test fish *Ctenopharyngodon idella*, maximum depletion was in liver (5.31),(5.46) muscle (3.02),(2.14) gill (2.80),(2.86) kidney (1.78)(,1.93) and brain(2.83)(2.90). tissues Under sub-lethal and lethal exposure to Phenthoate for 24h, the percentage of SDH depletion was found in all the tissues of test fish *Ctenopharyngodon idella*, was in liver (1.06),(0.93) muscle (1.18),(1.03) gill (0.67),(0.55) kidney (1.07)(,0.80) and brain(0.80)(0.82).

Keywords: Phenthoate, *Ctenophanrydon idella,* of LDH and SDH Lethal and sublethal concentrations

Introduction:

The field of ecotoxicology studies, the effect of anthropogenic chemicals on ecosystems at dissimilar levels of biological organization, from the molecular and cellular levels to whole ecosystems. Aquatic ecosystems are frequently impacted by water pollution, originating from runoff from urban and agricultural extents. Ultimately, the influence of toxic pollutant depends on the relative sensitivity of species, communal, ecosystem, the intensity and period of exposure. Acutely poisonous events most notably fish kills which were relatively mutual few decades ago are now rarely experiential in most industrialized countries; however, even sublethal toxicity could leads to plain impacts on entire

populations (Richard *et al.*, 2012).

Agriculture in Andhra Pradesh, India forms a main portion of the state economy. Large number of individuals of this state depends on agriculture provide with the required food grains. The production of paddy, chilies, tobacco, cotton, and other different kinds of crops are cultivated in wetland religions. Major part of cultivated lands is below Krishna River (Lat 15^0 18^1-16^0 50^1, month long. $70^0 10^1$-80^0 55^1east) area, which include Krishna and Guntur Districts of Andhra Pradesh. In the current past the local agriculture officers advise the farmers to decrease indiscriminate pesticide scattering and abate the usage of banned insecticides. From the upland areas of this locality, the pesticides are washed to the low land water bodies through surface runoff, where the aquaculture actions are taken up by the farmers.

Since both Krishna and Guntur Districts are maximum pesticides consuming places, the water is polluted by pesticides. It is important to know the impact of water qualities and the effect of pesticides to aquatic organisms, the more than a few pesticides used, the effect of Phenthoate on the non-target organisms are not so far investigation locally. Hence efforts have made to the toxicity of Phenthoate (50% EC) to Indian major carp *Ctenopharyngodon idella.*

Materials and Methods:

Fish *Ctenopharyngodon idella* of size 6±7 cm and 6.5± 2 g weight were brought from a local fish farm Kuchipudi, Guntur District of Andhra Pradesh, India and acclimatized at 28 ± 2^0C in the laboratory for 15days. Such acclimatized fish were exposed to sublethal and lethal concentrations of Phenthoate (50% EC)(20wg) commercial grade for 24h, 5 and 10 days. The vital tissues like muscle, brain, liver, gill and kidney of the fish were taken for the estimation LDH was estimated by the method srikanth and Krishnamurthy (1955) with slight modifications and SDH activity was estimated by the method of Nachlas et al.,(1960 were performed spectrophometrically

RESULT AND DISSCUSSION

The calculated value of lactate dehydrogenase (LDH) activity and the percent change over control along with standard deviation are given in the Table and Figure. The activity levels of dehydrogenase in *Ctenopharyngodon idella* exposed to Phenthoate 50% EC were expressed as micro moles of formazan/mg /protein/hr.

The LDH level of muscle, brain, liver, gill and kidney of control fish were almost stable. The control values of LDH in different tissues of the fish *Ctenopharyngodon idella* were in the order of:

Liver> Muscle > Brain > Kidney >Gill

Under lethal and sublethal exposure to Phenthoate (50% EC)for 24hr, the activity levels of LDH were found to increase in all the tissues of the fish *Ctenopharyngodon idella* the percent change in the activity levels of LDH, in the test fish were in the order of:

Phenthoate 50% EC lethal 24hr: Liver >Brain > Gill >Muscle > Kidney

Phenthoate 50% EC sub lethal 24hr: Liver>Muscle >Brain > Gill > Kidney

Under exposures to sublethal concentrations of Phenthoate 50% EC for 5th and 10th days. The percent depletion of LDH level in the test tissues of *Ctenopharyngodon idella* was in the order of

Phenthoate 50% EC controls of 5 days: Liver> Kidney >Brain >Muscle> Gill

Phenthoate 50% EC sublethal 5 days: Liver>Muscle >Brain > Gill > Kidney

Phenthoate 50% EC controls of 10days: Liver > Muscle >Brain > Kidney >Gill

Phenthoate 50% EC sublethal 10 days: Liver >Brain >Gill >Muscle > Kidney

Table: V.11. Changes in the specific activity levels of Lactate dehydrogenase (LDH) (µ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Ctenopharyngodon idella* exposed to sub-lethal and lethal concentrations of Phenthoate 50% EC**for 24hr:**

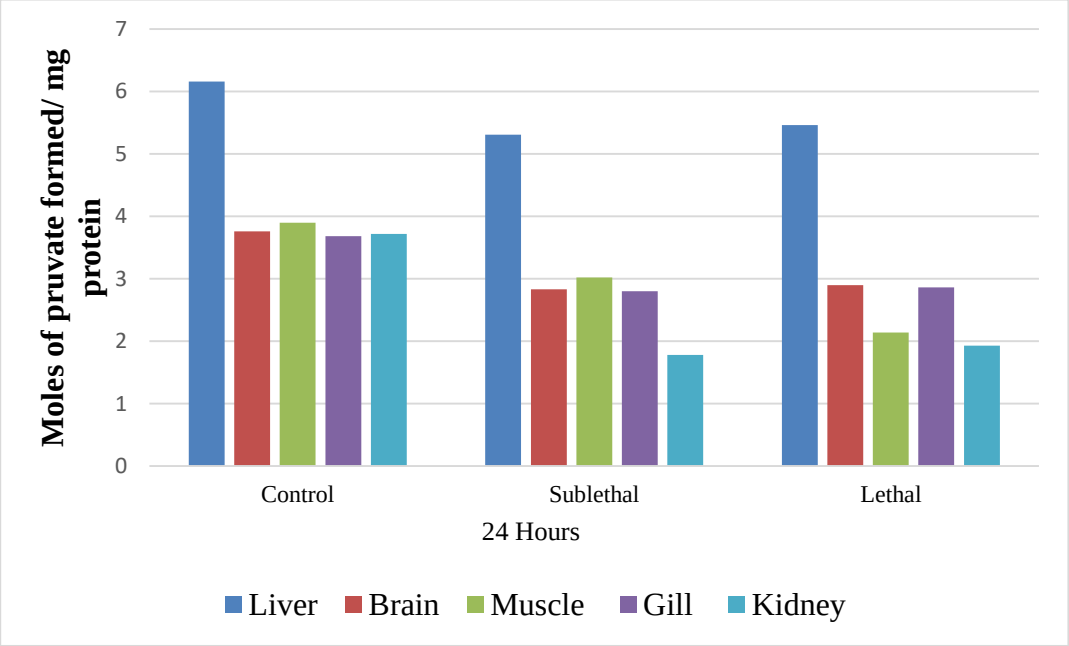
Values are the mean of five

LDH Tissues 24hr	Control	Sublethal(mg/g)	% Change	lethal(mg/g)	% Change
Liver	6.16±0.12	5.31±0.09	-16.72	5.46±0.01	-11.36
Brain	3.76±0.05	2.83±0.12	-24.73	2.90±0.14	-22.87
Muscle	3.90±0.16	3.02±0.01	-22.56	2.14±0.23	-45.13
Gill	3.68±0.20	2.80±0.07	-23.91	2.86±0.28	-22.28
Kidney	3.72±0.01	1.78±0.02	-52.15	1.93±0.05	-48.11

observations ;(±) indicates the standard deviation:

Values are significantly at P< 0.05

Fig: V.11. Changes in the specific activity levels of Lactate dehydrogenase(LDH) (µ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Ctenopharyngodon idella* exposed to sub-lethal and lethal concentrations of Phenthoate 50% EC for 24hr:



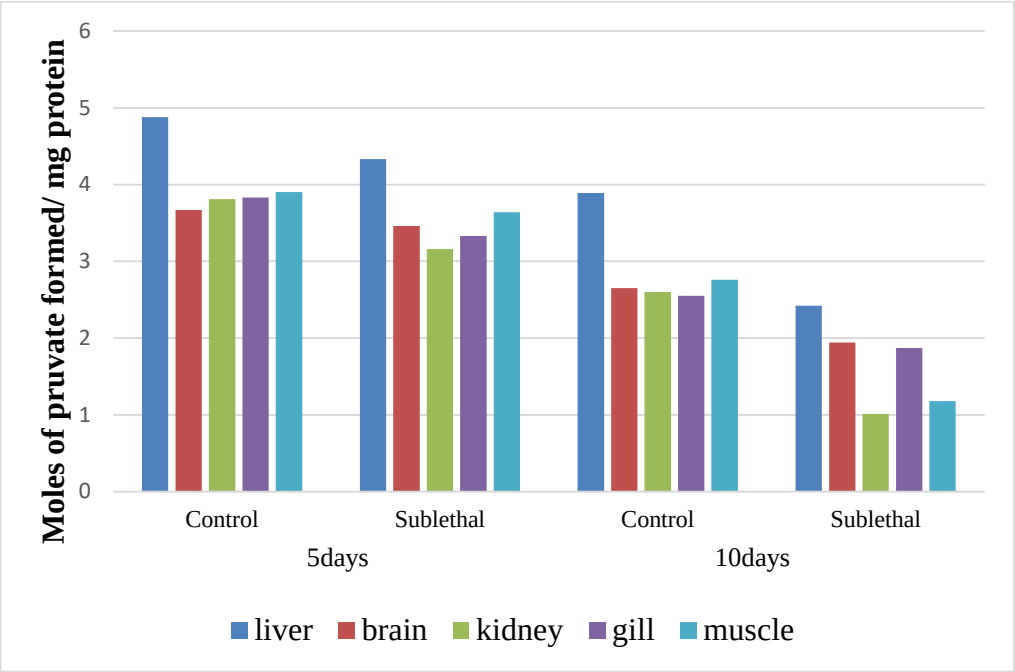
V. Table.12. Changes in the specific activity levels of Lactate Dehydrogenage (LDH) (µ moles of for mazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Ctenopharyngodon idella* exposed to sub-lethal concentrations of Phenthoate (50% EC)for 5 and 10 days:

LDH Tissues	5 days			10 days		
	Control	Sublethal (mg/g)	% Change	Control	Sublethal (mg/g)	% Change
Liver	4.88 ±0.01	4.33 ±0.04	-11.27	3.89 ±0.02	2.42 ±0.32	-37.59
Brain	3.67 ±0.05	3.46 ±0.08	-12.46	2.65 ±0.05	1.94 ±0.12	-26.04
Kidney	3.81 ±0.12	3.16 ±0.01	-16.97	2.60 ±0.24	1.01 ±0.05	-60.69
Gill	3.83 ±0.20	3.33 ±0.05	-12,98	2.55 ±0.13	1.87 ±0.07	-26.46
Muscle	3.90 ±0.15	3.64 ±0.32	-3.82	2.76 ±0.11	1.18 ±0.18	-56.83

Values are the mean of five observations ;(±) indicates the standard deviation

Values are significantly at P< 0.05

V. Fig.12. Changes in the specific activity levels of Lactate Dehydrogenage (LDH) (µ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Ctenopharyngodon idella* exposed to sub-lethal concentrations of Phenthoate (50% EC)for 5 and 10 days



Succinate Dehydrogenase (SDH) activity:

Succinate dehydrogenase (SDH) is a vital enzyme of citric acid cycle, catalysis the reversible oxidation of succinate to fumarate. In this present investigation it can be visualized that there is a rapid reflection of SDH activity in all tissues of fish *Ctenopharyngodon idella* treated with lethal and sublethal concentrations of Phenthoate (50% EC) compared with controls. The calculated value of SDH and standard deviation along with percent change over the controls is tissue specific viz., brain, liver, muscle, gill and kidney of fish *Ctenopharyngodon idella* exposed to lethal and sublethal concentrations of Phenthoate 50% EC for 24hr, 5 and 10 days.

In the tissue of 24hrs control fish, activity of SDH was in the order of:

Liver> Muscle >Kidney>Brain>Gill

The control fish, SDH activity was maximum in muscle followed by liver, kidney, gill and minimum in brain. The higher activity of SDH in liver and muscle suggests higher distribution of mitochondria in the tissues, since succinate dehydrogenase (SDH) is mitochondrially localized.

Phenthoate (50% EC) lethal 24hr: Muscle>Liver >Kidney > Brain > Gill

Phenthoate (50% EC) sub lethal 24hr: Muscle >Liver>Kidney >Brain > Gill

Phenthoate (50% EC) controls 5 days: Liver> Kidney >Muscle> Gill >Brain

Phenthoate (50% EC) sublethal 5days: Muscle>Kidney >Liver > Gill > Brain

Phenthoate (50% EC) activity of SDH at of 10days sublethal exposure to order of:

Phenthoate (50% EC) controls10 days: Liver>Kidney> Muscle>Gill>Brain

Phenthoate (50% EC) sublethal 10 days: Muscle>Kidney >Liver> Gill> Brain

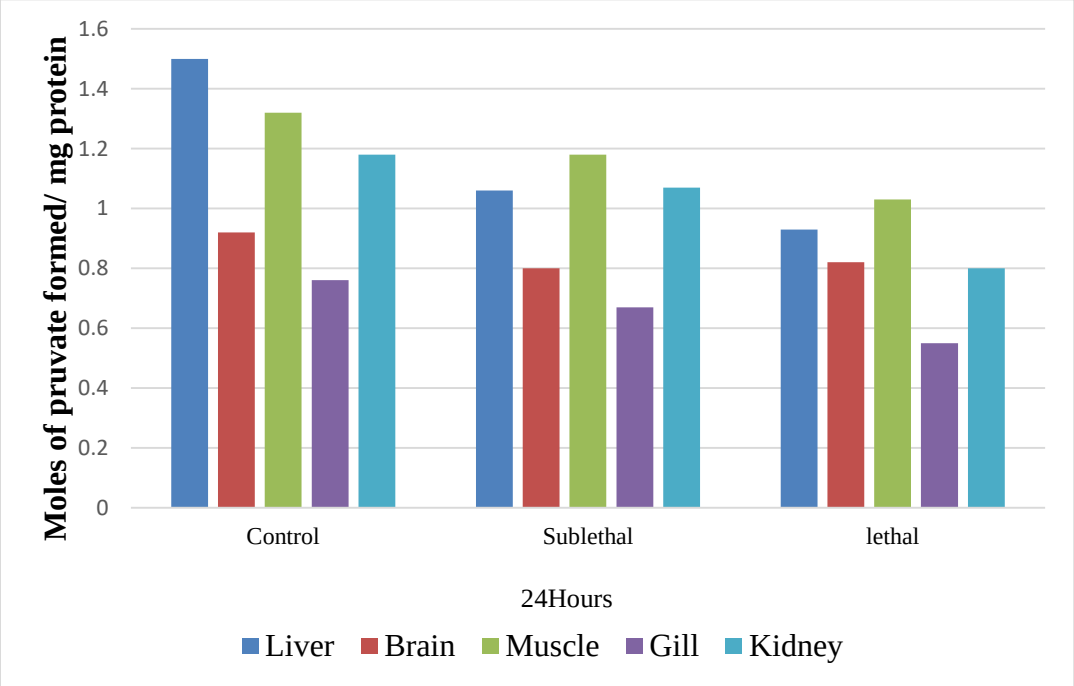
Table.13. Changes in the specific activity levels of Succinate dehydrogenase (SDH) (µ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Ctenopharyngodon idella* exposed to sub-lethal and lethal concentrations of Phenthoate 50% EC for 24hr:

SDH Tissues 24hr	Control	Sublethal (mg/g)	% Change	lethal(mg/g)	% Change
Liver	1.50±0.01	1.06±0.07	-28.95	0.93±0.03	-37.50
Brain	0.92±0.03	0.80±0.05	-14.58	0.82±0.20	-12.5
Muscle	1.32±0.18	1.18±0.01	-12.50	1.03±0.11	-21.64
Gill	0.76±0.05	0.67±0.08	-11.53	0.55±0.05	-26.92
Kidney	1.18±0.04	1.07±0.10	-9.61	0.80±0.02	-31.66

Values are the mean of five observations ;(±) indicates the standard deviation:

Values are significantly at $P< 0.05$

V. Fig .13. Changes in the specific activity levels of Succinate dehydrogenase (SDH) (µ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Ctenopharyngodon idella* exposed to sub-lethal and lethal concentrations of Phenthoate 50% EC for 24hr:



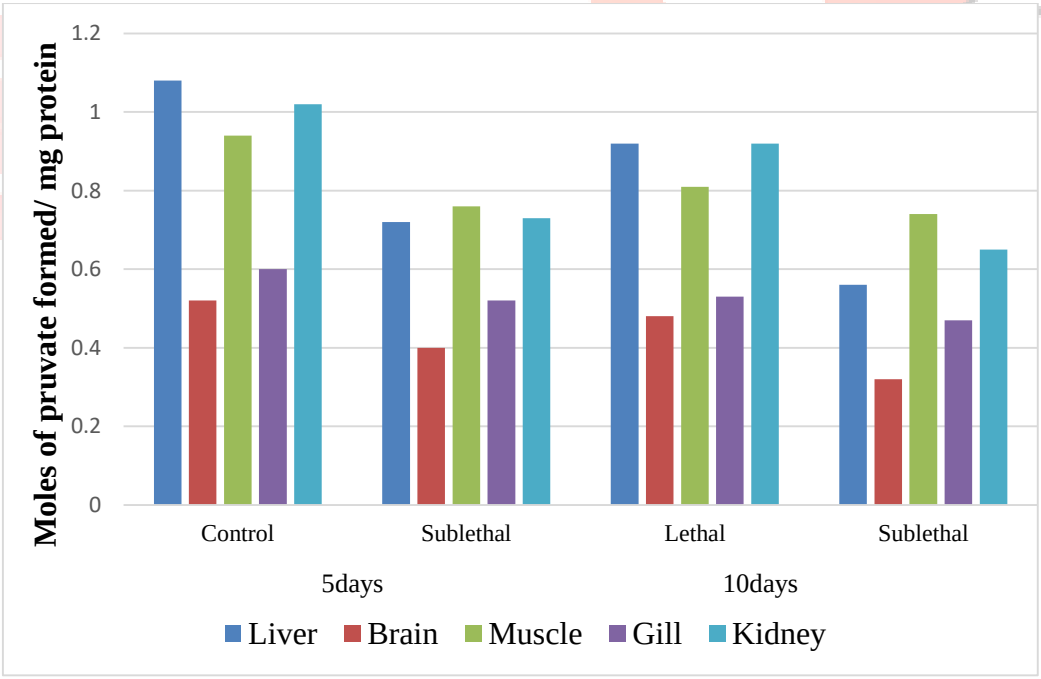
V. Table. 14. Changes in the specific activity levels of Succinate dehydrogenase (SDH) (µ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Ctenopharyngodon idella* exposed to sublethal concentrations of Phenthoate 50% EC **for estimation of SDH content in 5 and 10 days:**

SDH Tissues	5 days			10days		
	Control	Sublethal (mg/g)	% Change	Control	Sublethal (mg/g)	% Change
Liver	1.08±0.09	0.72±0.01	-32.72	0.92±0.05	0.56±0.04	-34.69
Brain	0.52±0.23	0.40±0.08	-22.41	0.48±0.01	0.32±0.12	-26.92
Muscle	0.94±0.07	0.76±0.06	-19.39	0.81±0.05	0.74±0.15	-5.88
Gill	0.60±0.01	0.52±0.15	-12.30	0.53±0.08	0.47±0.01	-10.16
Kidney	1.02±0.05	0.73±0.24	-31.59	0.92±0.01	0.65±0.03	-26.04

Values are the mean of five observations ;(±) indicates the standard deviation:

Values are significantly at $P< 0.05$

V.Fig.`14. Changes in the specific activity levels of Succinate dehydrogenase (SDH) (µ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Ctenopharyngodon idella* exposed to sublethal concentrations of Phenthoate 50% EC for estimation of SDH content in 5 and 10 days:



In the present study, it is observed that the activity of LDH was highly elevated in Phenthoate exposure indicating increased anaerobic respiration to meet the energy demands where aerobic oxidation is lowered (Gerringer *et al.*, 2017). Lactate dehydrogenase (LDH) converts the lactate to pyruvate and it plays very important role in carbohydrate metabolism (Khan *et al.*,2020). LDH involved in carbohydrate metabolism, any change in protein and carbohydrate metabolism might cause change in LDH activity.

Elevated LDH activity Phenthoate treated fish *Ctenopharyngodon idella* suggests that aerobic catabolism of glycogen and glucose has shifted towards the formation of lactate, which might have adverse effects on the organism.An increase in LDH activity might be due to stabilization of cytoplasmic membrane. that increased LDH activity in fish rainbow trout treated with carbamazepine Li *et al.,* (2009). Disturbances in their catalytic process due to xenobiotics compounds can cause cellular homeostasis affecting different enzymaticsystems.

Which can lead to effects at higher levels of biological organization such as tissues, organs, or individuals (Orrego *et al.*, 2011). Lethal and sub-lethal exposure to Phenthoate for 24h, the activity levels of LDH (Anitha *et al.*, 2018). The SDH and LDH enzyme activity levels increased (Pradeep Kiran JA and Bhaskar M.,2016). Tannery wastewater causes in *Chenna punctatus* fish effect of cellular hypoxia creating anaerobic condition and cellular damage. Investigation showed significant increase in LDH level with increase in concentration of tannery wastewater (5%, 10%, 15% and 20%TWW) (Rode *et al.*, 2016; Shahida Parveen and Ram Bharose *et al.*,2017; Khan *et al.*, 2020). The activity of LDH was highly elevated Profenofos and Carbosulfan exposure indicating increased anaerobic respiration to meet the energy demands where aerobic oxidation is lowered (Nagaraju Bantu, *et al.*, 2017). Changes in ion concentrations, organic constituents, enzyme activity and endocrinal activity as chemo regulators in fish have been attributed to pesticides (Moustafa *et al.*,2016). Increase of lactate dehydrogenase in all the tissues, (S. Murali Mohan *et al.*,2017).

Biochemical parameters are an important biomarker in determining the level of toxicity caused by the bio pesticide Derision (Tasneem and Yasmeen, 2020). It has been observed that the activity of the enzyme LDH has got increased due to the requirement of immediate oxygen when exposed to the toxicant, (V. Chaithanya Kumari, *et al.*, 2021). Metabolic enzymes such as citric acids synthesis and lactate dehydrogenase (LDH) are part of the respiratory enzymatic system, which can be affected by from the detoxification enzyme systems under stress conditions in fish (Rode *et al.*, 2016). Normal activity of LDH patterns was altered in situations of chemical stress. leakage of LDH is a marker of membrane permeability and cell death (Khan *et al.*,2020).

Tasneem and Yasmeen, 2018; Mishra and Banerjee, 2019; Thripathi and Singh.,2013; Gerringer *et al.*,2017, LDH activity in fish *Channa punctatus* significantly increased in skeletal muscle (2.2) fold followed by liver (1.8) fold, gill (1.6) fold and brain (1.4) in response to treatment with alpha methrion for 14days, due to an increase in anaerobic respiratory activity and production of more lactate for completion of metabolic process. As the rate if glycolysis increases, the pyruvate is not routed to Krebs's cycle, later catalyses to lactate; thereby shifting the respiratory metabolism from aerobiosis to anaerobiosis. Prakash *et al.*,(2020) increased LDH activity in brain and gill tissues of *Ctenopharyngodon idella* treated with cypermethrinfor 96h.

Due to disruption of respiratory epithelium might have caused tissue hypoxia resulting in oxidative metabolism which might responsible for in LDH activity in toxicant stress (Annamalai malarvizhi *et al.*,2012; Khan *et al.*, 2019). It was found that the activity of SDH and LDH was depleted significantly

increased after the heavy metal stress (Sandhya *et al.,* 2017**;** Prakash *et al.,* 2020). Similar observations on LDH activity were made under Deltamethrin stress in prawn by (Rode *et al.,*2016) leakage of LDH is a marker of membrane permeability and cell death. They also suggested that an increase in LDH activity might due to stabilization of cytoplasm membrane (Lalles, 2019).

A similar decrement in the SDH activity was observed by in fish exposed to Malathion and fenitrothion pesticides (Anitha *et al.,*2015). Decreased SDH activity might observe due to depletion in the oxidative metabolism at the mitochondria level (Pradeep Kiran and Bhaskar, 2016). The present study agrees with those of (Dong *et al.,* 2020) The inhibition of NAD dependent, SDH activity indicated a decreased pass of intermediates into the citric acid cycle (Khare *et al.,* 2019). Rather *et al.,* (2015) thoroughly investigated the biochemical changes induced by carbaryl, carbosulfan and parathion in *C. batrachus* Bhaskara and Vijaya (2016). Tasneem and Yasmeen,(2020) Butachlor and Machete induced biochemical alterations in *C. punctatus*. 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 was depleted significantly after the heavy metal stress (Sandhya and Sonawane, 2017). Effect of fenthion on enzymes of *C. carpio*, **(**Khalid., 2014). Tasneem and Yasmeen (2020) studied the effect of cypermethrin on enzyme activities of *C. carpio*. Effect of prorate on the level of total proteins in *C. carpio* by Lakshmaiah (2016). Effect of Triclosan on total protein content in *C. punctatus* was stress and decreased, (Ravi *et al*. 2015**).** Similar decrement in the SDH activity was also various workers in different species of fish exposed to different peptides. Al-Gham and Mahboob,(2012) decreased SDH activity in fish *Tilapia mossambica* and *Clarias gariepinus* exposed to different type of chemicals, due to depletion in the oxidative metabolism at the level of mitochondria leading to depression of TCA cycle. This might be responsible for suppression of oxidative phase of tissue metabolism under pesticidal impact showing a shift from aerobic metabolism to anaerobic metabolism under pesticidal impact showing a shift from aerobic metabolism to anaerobic metabolism under the pesticidal stress (Abdel-Salam Mohamed Ibraik Ohaida *et al.,* 2010; Rani *et al.,* 2016).

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