

## **Zinc Metal Caused Hexokinase Variations In Different Brain Regions Of Teleosts And Influence Of *Chlorella Vulgaris* With The Special Reference Of Detoxification.**

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**Abstract:** *The aquatic autotrophs extended protection to significant extent against to the sub-lethal zinc induced enzyme variations but still there is a serious innovative need to protect the economically, Culturally and nutritionally important fish species for protection. The sub - lethal Zinc metal concentrations in presence of *Chlorella vulgaris* caused significant variations in brain (cerebrum, diencephalons, cerebellum & medulla oblongata) enzyme hexokinase in *Labeo rohita*, *Clariasbatrachus* and *Channa punctatus*, in microbe presence to a lesser extent than metal exposure directly. Under detoxification studies the impact of *Chlorella vulgaris* on sub-lethal Zinc toxicity on hexokinase in various brain regions, *Chlorella vulgaris* may have potential as a precipitation agent. *Chlorella vulgaris* rapidly adsorbed appreciable amount of Zinc from the aqueous solutions. The autotroph *Chlorella vulgaris* has the detoxification ability and the present change of enzyme levels in different brain regions of three fish species.*

**Key words:** *Chlorella vulgaris, Hexokinase, Teleosts, Zinc.*

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### **I. Introduction**

Heavy metals are dangerous because they tend to bioaccumulate (Barron,2003 and Boyd, 2004). Toxic effects of heavy metal exposure are severe visceral damage and testicular atrophy, renal dysfunction, hepatic damage, hyper-tension, gas exchange breakdown at lamellar regions, central nervous system injury, anemia, bio-chemical and physiological variations, less fecundity alterations in abiotic & biotic factors of the habitat and even in generic disorder are well established. Heavy metals further affect organisms directly by accumulating in their body or indirectly by transferring to the next trophic levels of the food chain. Heavy metal poisoning could result, for instance, from drinking-water contamination (e.g. lead pipes) high ambient air concentrations near emission sources, or intake via the food chain (Barron et al.,2003; Dosi et al., 2007 & Das et al., 2008). Hence the need of the man is to innovate some alternative technologies and devices to protect the nature gifted consumables and to boost the yield from natural water bodies. In the present investigation the author made an attempt to study the influence of on *Chlorella vulgaris* sub - lethal concentration of Zinc caused marked change in hexokinase in cerebrum, diencephalons, cerebellum and medulla oblongata in *Labeo rohita* (Ham.), *Clariasbatrachus* (Linn.) and *Channa punctatus* (Bloch) under acute studies.

### **II. Materials And Methods**

Alive, healthy, mature, disease-free & active *Labeo rohita* (Ham.), *Clariasbatrachus* (Linn.) and *Channa punctatus* (Bloch.) 120-130 gm. of 18-20 cm. (standard length) were obtained from few selected local ponds to avoid ecological variation and acclimatized in the laboratory condition. Period of seven days were given for various exposures and investigations. Determination of safety & sub-lethal concentrations. Safety & sub-lethal concentrations of Zinc was determined on *Labeo rohita*, *Clariasbatrachus* and *Channa punctatus* by the Probit Analysis Method. Higher concentration of Zinc was used and slowly reduced the amount of concentration to know the Lc 50/100 value for 96-hour exposure. Acute studies : The *Labeo rohita*, *Clariasbatrachus* and *Channa punctatus* (120-130 gm) of 18-20 cm (standard length) were taken separately and kept in twenty groups and each group consist of forty eight fish species. No food was given to the above fish species during this period (08, 16 & 24hrs). The first set of *Labeo rohita*, *Clariasbatrachus* and *Channa punctatus* were exposed to sub-lethal concentration of zinc and the detail were described [Shaffi & Kakaria 2006]. The termination of the experiment, preparation of tissue extract and enzyme assays were described [Colowick & Kaplan, 1975; Shaffi & Habbibulla, 1977]. The experiments with acute studies were repeated at least seven times separately to subject the data for analysis of variance.

### III. Result

Exposure to sub-lethal concentration of Zinc caused marked changes in hexokinase in cerebrum, diencephalons, cerebellum and medulla oblongata of *Labeorohita* (sub-lethal concentration of Zn- 0.72 mg/ltr.), *Clariasbatrachus* (sub-lethal concentration of Zn- 2.75mg/ltr.) and *Channapunctatus* (sub-lethal concentration of Zn- 2.90mg/ltr.) under acute studies. Safety level concentrations of zinc metal was determined for *Labeorohita* (Zn-0.10 mg/ltr.), *Clariasbatrachus* (Zn-0.14 mg/ltr.) and *Channapunctatus* (Zn- 0.18 mg/ltr.). The influence of *Chlorella vulgaris* on sub-lethal concentration of zinc was investigated in table No. 1 and Fig. No 1

- please see Table no. 1 and digram no.1.

The exposure to sub-lethal concentrations of Zinc in presence of *Chlorella vulgaris* led to highest fall in diencephalons, hexokinase in comparison to cerebrum, medulla oblongata and cerebellum in *Labeorohita*. The maximum fall in hexokinase was in diencephalons followed by cerebrum, medulla oblongata and cerebellum at 08 hrs. exposure than at 16 & 24 hrs. exposure in *Clariasbatrachus*. The fall in hexokinase in *Channapunctatus* was optimum at 16 hrs. in diencephalon, in comparison to , medulla oblongata, & cerebellum than at 08 hrs. and at 24 hrs. in exposure to sub-lethal concentrations. The hexokinase fall was highest in diencephalon exposed to sub-lethal concentrations of Zinc in microbe presence at 08 hrs. than at 16 & 24 hrs. in comparison to cerebrum, medulla oblongata & cerebellum in *Labeorohita* than in *Clariasbatrachus* & *Channapunctatus* (Table-1 & chart digram no.1.).

### IV. Discussion & Conclusion

The sub-lethal Zinc concentrations in presence of *Chlorella vulgaris* caused significant variations in brain (cerebrum, diencephalons, cerebellum & medulla oblongata) enzymes hexokinase in *Labeorohita*, *Clariasbatrachus* & *Channapunctatus* in microbe presence to a lesser extent than metal exposure directly. Under detoxification studies the impact of *Chlorella vulgaris* on sub-lethal Zinc toxicity on hexokinase in various brain regions of brain i.e. cerebrum, diencephalon, cerebellum and medulla oblongata in three important inland teleost viz *Labeorohita*, *Clariasbatrachus* & *Channapunctatus* under short term exposure studies.

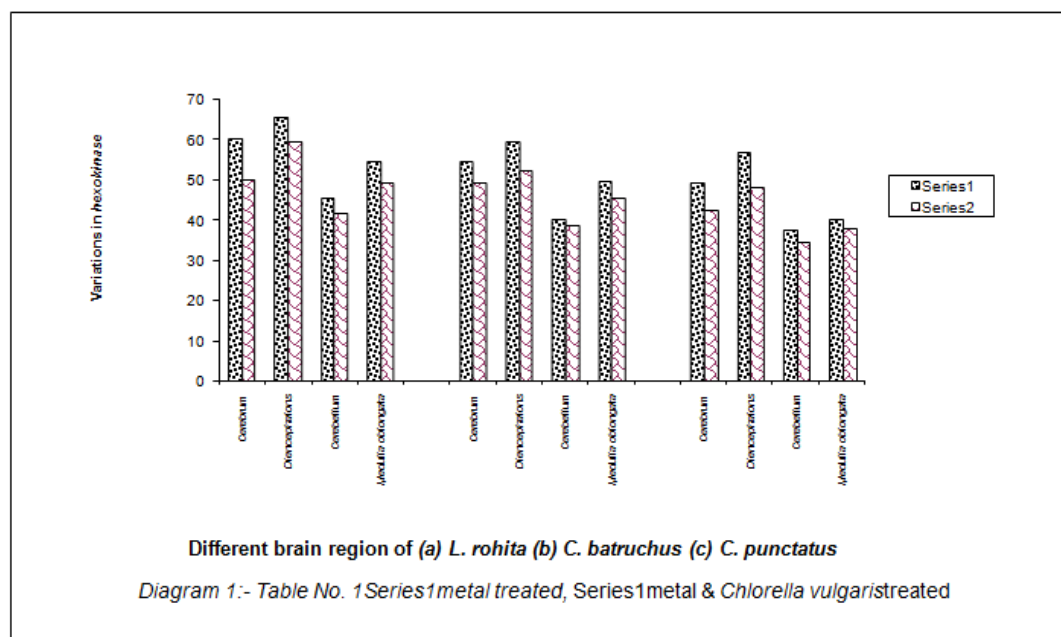
In the present investigation to a new strategy was adapted to detoxify the metal caused toxicity on brain enzyme compartmentation in three fish species. The sub-lethal Zinc concentration in presence of *Chlorella vulgaris* caused significant variations in brain (cerebrum, diencephalons, cerebellum & medulla oblongata) enzyme (hexokinase) in *Labeorohita*, *Clariasbatrachus* & *Channapunctatus* in microbe presence to a lesser extent than metal exposure directly.

**Table No.-01** : Influence of *Chlorella vulgaris* on zinc (sub-lethal) caused hexokinase variations in different brain regions of three fresh water fish species.

Regions of the Brain	Contr ol	Duration of sub-lethal Concentration exposure			% of fall/ Rise	Duration of sub-lethal concentration exposure with <i>Chlorella vulgaris</i>			% of fall/ Rise
		15 DAYS	30 DAYS	45 DAYS		15 DAYS	30 DAYS	45 DAYS	
<b>(a) Labeo rohita (Ham)</b>									
<b>Cerebrum</b>	0.352 ±.082	0.234 ±.019	0.196 c ±.026	0.179 c ±.019	49.14	0.264 c ±.032	0.224 c ±.041	0.211 c ±.024	40.05
<b>Diencephalon</b>	0.290 ±.046	0.264 ±.024	0.169 c ±.032	0.116 c ±.016	60.00	0.272 ±.042	0.184 ±.026	0.158 c ±.019	45.51
<b>Cerebellum</b>	0.218 ±.028	0.152 ±.032	0.149 ±.024	0.133 c ±.021	38.99	0.177 ±.024	0.164 ±.016	0.152 ±.014	30.27
<b>Medulla</b>	0.328	0.265	0.224 c	0.190	42.07	0.275	0.256		36.28

<b>oblongata</b>	±.036	±.042	±.032	c ±.028		±.035	±.032	0.209 c ±.024	
<b>(B) Clarias batrachus (LINN.)</b>									
<b>Cerebrum</b>	0.332 ±.042	0.244 ±.039	0.199 c ±.024	0.189 c ±.019	43.07	0.259 ±.029	0.224 ±.019	0.215 c ±.018	35.24
<b>Diencephalon</b>	0.241 ±.028	0.216 ±.024	0.148 c ±.019	0.120 c ±.016	50.00	0.221 ±.032	0.159 ±.028	0.135 c ±.021	43.98
<b>Cerebellum</b>	0.162 ±.019	0.152 ±.024	0.128 ±.032	0.113 ±.018	30.24	0.141 ±.019	0.132 ±.016	0.121 ±.016	25.30
<b>Medulla oblongata</b>	0.279 ±.032	0.254 ±.026	0.224 ±.036	0.211 ±.029	26.22	0.236 ±.024	0.204 ±.032	0.192 ±.019	22.58
<b>(C) Channa punctatus (BLOCH)</b>									
<b>Cerebrum</b>	0.286 ±.028	0.218 ±.021	0.198 ±.032	0.186 c ±.024	34.96	0.238 ±.019	0.209 ±.022	0.194 ±.020	32.16
<b>Diencephalon</b>	0.201 ±.034	0.184 ±.026	0.132 ±.016	0.112 ±.013	44.27	0.174 ±.021	0.138 ±.016	0.126 ±.026	37.31
<b>Cerebellum</b>	0.188 ±.024	0.189 ±.018	0.146 ±.021	0.135 ±.022	28.19	0.164 ±.022	0.159 ±.024	0.146 ±.032	22.34
<b>Medulla oblongata</b>	0.250 ±.029	0.208 ±.016	0.188 ±.032	0.175 ±.036	30.00	0.229 ±.032	0.198 ±.036	0.182 ±.042	27.20

Values are mean ± SDM of seven replicates . The data was subjected to test of ANOVA . The super scripts (a, b & c) indicates that P > 0.01, P > 0.02, & P > 0.05 respectively



The *Chlorella vulgaris* influenced the sub-lethal effect of Zinc caused variations in brain compartmentation (cerebrum, diencephalons, cerebellum & medulla oblongata) of hexokinase in *Labeorohita*, *Clarias batrachus* & *Channapunctatus* under acute or short term exposure. The sub-lethal levels of Zinc inhibited the hexokinase to a highest extent in diencephalon than in cerebrum, medulla oblongata & cerebellum in *Labeorohita* in comparison to *Clarias batrachus* & *Channapunctatus* but lesser than the fall of the enzymes in the above said fish species directly exposed to sub-lethal levels of Zinc directly without any microbe compelled us to develop an insight to understand the positive impact on important bio-chemical parameters like enzymes that are important to promote a variety of anabolic & catabolic processes in an organism effectively reflects that microbes act as antidote effect fall heavy metal toxicity and the less fall of the four enzymes under investigation may be that microbes has a soothing impact and hence the microbes are able to decrease the sub-lethal toxicity of sub-lethal level Zinc.

The following finding may help to understand the microbe-metal interaction and sub sequent detoxification of the metal to a less extent in a better way. The sub-cellular regions of Cyanobacteria and *Anabaena cylindrica* could trap the lead through its phosphate and precipitates in the form of lead phosphate on the cell wall inside the cell [Jayprakash et al. 2005, Bert et al., 2009]. *Chlorella vulgaris* has the detoxication ability and the present change of enzyme levels in different brain regions of three fish species. [Kushwaha et al. 2004, Cristina et al., 2005; Gelagutashvili 2006; Shaffi et al., 2007 & Page et al., 2009]. Similar kind of mechanism might have taken place in the present findings i.e. less fall of enzymes in which the cellular components of *Chlorella vulgaris* might have precipitated the metal into compound with the help of its cellular components and the present findings i.e. less fall of enzymes in presence of an autotroph than the enzyme fall when directly exposed to Zinc sub-lethal should understand on similar lines. In the present investigation the Zinc metal might have accumulated in *Chlorella vulgaris* and the less impact of Zinc noticed in the fall of hexokinase in various brain regions of *Clarias batrachus* & *Channapunctatus* in comparison to directly exposed to Zinc sub-lethal.

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