ISSN: 0976-3031

Available Online at http://www.recentscientific.com

CODEN: IJRSFP (USA)

International Journal of Recent Scientific Research
Vol. 9, Issue, 3(G), pp. 25101-25104, March, 2018

International Journal of Recent Scientific Research

DOI: 10.24327/IJRSR

Research Article

IMPACT OF *SPIRULINA PLATENSIS* ON LONG-TERM ZINC METAL TOXICITY ON L*ABEO ROHITA* (HAM.), *CLARIAS BATRACHUS* (LINN.) AND *CHANNA PUNCTATUS* (BLOCH.)

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DOI: http://dx.doi.org/10.24327/ijrsr.2018.0903.1792

ARTICLE INFO

Article History:

Received 11th December, 2017
Received in revised form 8th January, 2018
Accepted 4th February, 2018
Published online 28th March, 2018

Key Words:

Spirulina platensis, Zinc, phosphofructokinase, Labeo rohita, Clarias batrachus , Channa punctatus.

ABSTRACT

The enzymes are the mediators of the metabolism and the interference of heavy metals with osmo-regulatory mechanisms may influence the osmotic pressure, cardiac function and nerve conduction sequentially. The presence of *Spirulina platensis* might have protected the protein part of the enzyme despite the fish species were exposed to sub-lethal levels of zinc *in Labeo rohita, Clarias batrachus & Channa punctatus* reflects that the aquatic autotrophs have soothing impact hence the fish species are able to survive under ability of' adaptation for survival.

In the present study the influence of *Spirulina platensis* on sub - lethal concentrations of zinc on *phosphofructokinase* in cerebrum, diencephalon, cerebellum and medulla oblongata in *Labeo rohita* (Ham.), *Clarias batrachus* (Linn.) and *Channa punctatus* (Bloch) under chronic studies (15,30 & 45 days).

INTRODUCTION

Heavy metal induces oxidative damage in different organs by increasing per-oxidation of membrane chemistry and altering the antioxidant system of the cells/tissues *[Bashiru & Rosemary, 2007; Kumar & Kalonia, 2007]*. Interaction of metal ions with the cell organelles causes injury to cellular components. Heavy metal intoxication further depletes glutathione & protein bound sulfa-hydryl groups resulting into the production of reactive oxygen species like hydrogen peroxides, superoxide ions & hydroxyl radicals. These reactive oxygen species induce elevated visceral per -oxidation *[Bano et al., 2007; Gini et.al., 2006; Gupta & Flora,2006* and *Kaur & Bansal ,2008]*.

The biological compounds with anti-oxidant properties protect cells & tissues against deleterious effects of reactive oxygen species & other free radicals. Protective compounds from biological source with anti - per oxidative and antioxidant properties play a vital role in protecting the visceral organs against heavy metal toxicity*[Bert, et. al., 2009; Nichat, 2016 a, b]*. Newtechnologies are innovated to conserve energy and save expenditure. Involving aquatic autotrophs to combat aquatic contamination is attaining momentum *[Kushwaha et.al. , 2001& 2004; Nichat, et. al. 2014 & Dosi et.al. 2007]*.

In the present investigation the researcher want to study the influence of *Spirulina platensis* on sub - lethal concentrations of zinc on *phosphofructokinase* in cerebrum, diencephalon, cerebellum and medulla oblongata in *Labeo rohita* (Ham.), *Clarias batrachus* (Linn.) and *Channa punctatus* (Bloch) under chronic studies (15,30 & 45 days).

MATERIAL and METHODS

Alive, healthy, mature, disease-free & active *Labeo rohita* (Ham.), *Clarias batrachus*(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 for a period of seven days and were subjected for various exposures and investigations.

Method of microbe Feeding: *Spirulina platensis* was given daily for *Labeo rohita in* flour as pellets and for *Clarias batrachus* and *Channa punctatus* the *Spirulina platensis* mixed with 35% animal flesh made pellets and these three fish species were kept under observation for thirty minutes.

Determination of safety, Sub-lethal and lethal concentration: Safety, sub-lethal concentrations of zinc metal was determined on *Labeo rohita,Clarias batrachus* and *Channa punctatus* by the *Probit Analysis Method [Finney,1971]*. Higher

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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, Clarias batrachus* 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, Clarias batrachus* and *Channa punctatus* were exposed to sub-lethal concentration of zinc metal the detail were described somewhere else *[Shaffi & Kakaria2006]*.

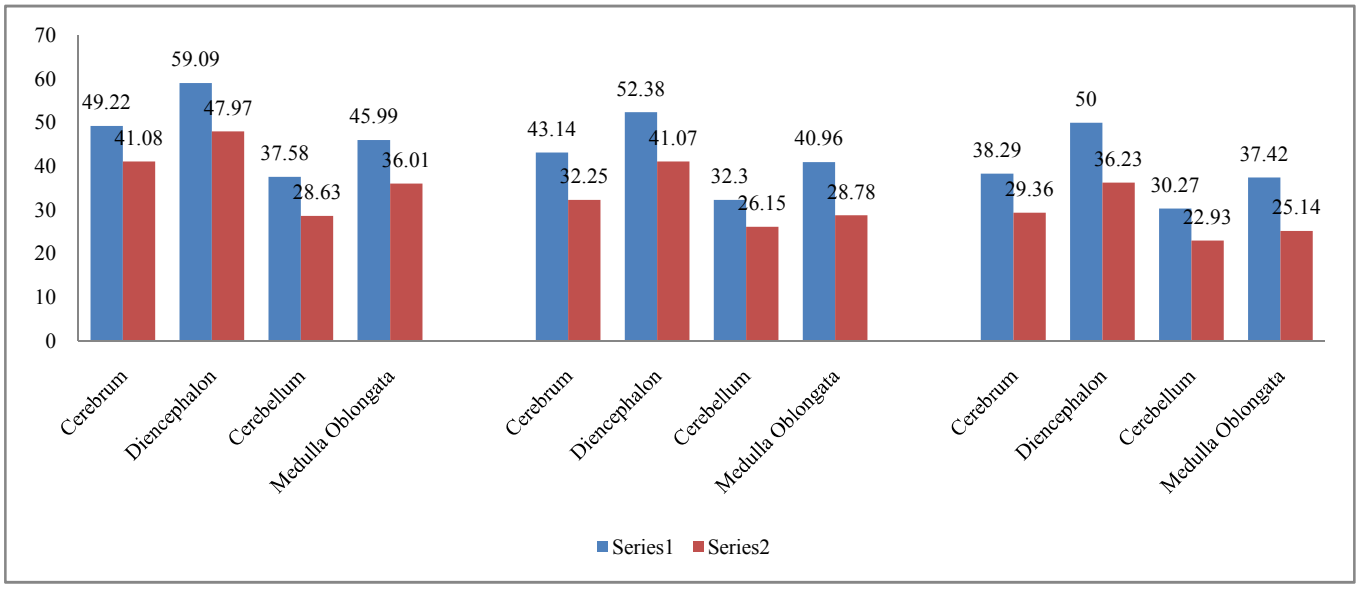
Preparation of tissue extract: The termination of the experiment preparation of tissue extract and enzyme assays were described elsewhere *[Colowick & Kaplon, 1975 , Shaffi & Habbibulla,1977]*.

Table No 1 Impact of *Spirulina platensis* on Zinc Metal (sub-lethal)caused toxicity in three fresh water Teleosts *Phospho fructokunase*- Chronic Studies

Regions of the brain	Control	Duration of Sub-Lethal Concentration Exposure			% Of Fall/ Rise	Duration of Sub-Lethal Concentration Exposure With *Spirulina platensis*			% Of Fall/ Rise
		15 DAYS	30 DAYS	45 DAYS		15 DAYS	30 DAYS	45 DAYS	
(a) *Labeo rohita* (HAM)									
Cerebrum	0.258 ±.062	0.184 ±.026	0.158 c ±.029	0.131 c ±.021	49.22	0.194 ±.032	0.186 ±.026	0.152 c ±.031	41.08
Diencephalon	0.198 ±.039	0.172 ±.019	0.116 ±.016	0.081 c ±.014	59.09	0.178 ±.021	0.122 ±.018	0.103 c ±.019	47.97
Cerebellum	0.149 ±.024	0.138 ±.021	0.109 ±.018	0.093 ±.014	37.58	0.123 ±.019	0.112 ±.028	0.105 ±.021	28.63
Medulla Oblongata	0.211 ±.026	0.192 ±.041	0.168 ±.026	0.114 c ±.021	45.99	0.174 ±.024	0.155 ±.032	0.135 ±.026	36.01
(b) *Clarias batrachus* (LINN.)									
Cerebrum	0.248 ±.036	0.206 ±.029	0.179 ±.032	0.141 c ±.021	43.14	0.202 ±.021	0.189 ±.029	0.168 c ±.019	32.25
Diencephalon	0.168 ±.021	0.148 ±.019	0.110 ±.014	0.080 c ±.013	52.38	0.142 ±.019	0.122 ±.018	0.099 ±.014	41.07
Cerebellum	0.130 ±.016	0.121 ±.016	0.112 ±.012	0.088 ±.012	32.30	0.122 ±.021	0.112 ±.012	0.096 ±.021	26.15
Medulla Oblongata	0.108 ±.024	0.164 ±.018	0.139 ±.021	0.117 ±.016	40.96	0.162 ±.032	0.153 ±.014	0.141 ±.022	28.78
(c) *Channa punctatus* (BLOCH)									
Cerebrum	0.235 ±.032	0.194 ±.022	0.159 ±.026	0.145 c ±.021	38.29	0.204 ±.014	0.182 ±.024	0.166 ±.021	29.36
Diencephalon	0.138 ±.018	0.102 ±.016	0.079 ±.010	0.069 ±.010	50.00	0.129 ±.019	0.105 ±.014	0.088 ±.013	36.23
Cerebellum	0.109 ±.019	0.092 ±.012	0.084 ±.012	0.076 ±.009	30.27	0.101 ±.016	0.091 ±.012	0.084 ±.011	22.93
Medulla Oblongata	0.171 ±.024	0.151 ±.021	0.128 ±.016	0.107 ±.019	37.42	0.154 ±.021	0.136 ±.016	0.128 ±.012	25.14

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

Fall in *phosphofructokinase*



Series 1 –Zinc treated, **Series 2** –Zinc & *Spirulina platensis* treated

Diagram 1 Variation of *phosphofructokinase* in defferent brain region of *Labeo rohita, Clarias batrachus* and *Channa punctatus*

Statistical analysis: The experiments with acute and chronic studies were repeated at least seven times separately the data for analysis of variance.

RESULTS

It is clear that *phosphofructokinase* in different brain regions (cerebrum, diencephalons, cerebellum & medulla oblongata) of *Labeo rohita*(sub-lethal concentration of Zn- 0.72 mg/ltr.), *Clarias batrachus* (sub-lethal concentration of Zn- 2.75mg/ltr.), and *Channa punctatus* (sub-lethal concentration of Zn- 2.90mg/ltr.,) exposed to sub-lethal zinc concentration in presence of *Spirulina platensis* underwent marked variations at 15, 30 & 45 days. Please see Table-01 and chart diagrams - 01.

The variations in *phosphofructokinase* was maximum (Fall) at 30 days in diencephalons than at 15 & 45 days in comparison to cerebrum, medulla oblongata & cerebellum in *Labeo rohita* . The fall in *phosphofructokinase* was highest in diencephalons at 30 days than at 15 & 45 days accompanied by cerebrum, medulla oblongata & & cerebellum in *Clarias batrachus* exposed to sub- lethal levels of zinc metal in presence of *Spirulina platensis* under chronic studies.

In *Channa punctatus* the *phosphofructokinase* fall was optimum in diencephalons at 30 days exposed to sub-lethal level of zinc in presence of *Spirulina platensis* in comparison to 15&45 days than in cerebrum, medulla oblongata & cerebellum.

Among the fish species *Labeo rohita* was most affected than *Clarias batrachus* & *Channa punctatus* exposed to sub- lethal concentrations of zinc metal in presence of *Spirulina platensis*.

DISCUSSION

The exposure to sub-lethal levels of zinc might have secreted significant secretion of humoral & cell mediated responses in exposed animals *like Labeo rohita, Clarias batrachus & Channa punctatus*. If the immunology becomes weak the organism will face difficulty to overcome the toxicant poisoning and subsequently the toxicant impact will affect physiology of the organism and the variations in enzymes of brain regions may be understood on these lines and the presence of these aquatic autotroph might have reduced the toxic impact and the less fall in enzymes may be due to the presence of the above said autotroph which have the ability to accumulate the toxicant and the less fall in the enzymes may be understood as the detoxification property of the aquatic autotrophs *[Mench,et.al.,2009; Page,et.al., 2009*; *Schroder, et.al., 2009 and Nichat, et. al., 2015]*.

CONCLUSION

Novel methods to clean up the heavy metal polluted water bodies by replicate transitional physical & chemical methods of environmental cleanup through phyto-remediation.

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How to cite this article:

Avinash R.Nichat.2018, Impact of Spirulina Platensis on long-Term Zinc Metal Toxicity on Labeo Rohita (Ham.), Clarias Batrachus (linn.) And channa punctatus (bloch.). *Int J Recent Sci Res.* 9(3), pp. 25101-25104.
DOI: http://dx.doi.org/10.24327/ijrsr.2018.0903.1792

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