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# Determination of $LC_{50}$ of Lead Nitrate for a fish, *Labeo rohita* (Hamilton-Buchanan)

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## Abstract

*The contamination of water by heavy metals is a worldwide problem which is increasing day by day due to the anthropogenic activities. These heavy metals poses a serious health risk on human population and aquatic organisms. One such heavy metal is Lead. This paper emphasises on the determination of 96hr $LC_{50}$ value of Lead Nitrate for the fish, Labeo rohita. The acute toxicity test was performed according to the standard methods in APHA and the value was calculated by probit analysis. The fish specimens were acclimatized in the laboratory conditions for 15 days. The stock solution of Lead Nitrate was prepared and the fish fingerlings were treated with various concentrations ranging from 1 mg/l to 50 mg/l for 96 hours. The results showed that the median lethal concentration ($LC_{50}$) of Lead Nitrate for the fish, Labeo rohita is 34.20 mg/l. The susceptibility of Labeo rohita to the lethal effect of Lead Nitrate were dependent on duration as well as on concentration. The mortality of the fishes is directly proportional to the concentration. The use of Lead should be discouraged to protect valuable biodiversity.*

**Keywords**: Lead Nitrate, Acute Toxicity, *Labeo rohita*, 96hr $LC_{50}$.

## Introduction

Water pollution has become a global problem as various pollutants like heavy metals and toxic chemicals are discharged without prior treatment into the water bodies most commonly in developing countries. These heavy metals due to their properties like long half life period, bioaccumulation, biomagnification in the food chain and non-biodegradability are hazardous to the aquatic organisms and their consumers which on being exposed to these heavy metals can suffer from immense health problems and risk of life.

Fishes have direct economic importance and are quite sensitive to the wide array of pollutants discharged in the aquatic ecosystems. Fishes are widely used to assess water quality of aquatic ecosystems because they serve as pollution bioindicators[1-2]. Fish may concentrate large quantities of toxic metals from polluted aquatic environments[3]. The heavy metal concentration in the body of fish depends upon feeding habits, trophic status, food availability, physico-chemical properties of water, metabolic rate of animal and toxicity of heavy metals[4-6].

Lead is not necessary for biological functions. It is discharged into different natural aquatic ecosystems from various industries like petroleum, chemical, dye and mining industries. It poses serious threats from the toxicity point of view and cause mass mortality of various aquatic organisms[7-9]. Lead is known to cause neurological, haematological, gastrointestinal, reproductive, circulatory, immunological, histopathological and histochemical changes[10-12]. The bioaccumulation of the highly toxic heavy metal has been observed in various tissues of the fishes like scales, bones, gills, kidneys and liver[13].

The toxic effects of various heavy metals may hinder the physiological and metabolic functions, rate of growth, reproductive efficacy and ultimately causes mortality in fishes[14]. Toxicity tests have been performed on fishes to evaluate the effect of toxicants on various aquatic organisms under laboratory conditions. To assess susceptibility and survival potential of the test organisms 96 hr $LC_{50}$ tests of some particular toxicants have been conducted. The fingerling stage of fish is more reliable to conduct toxicity test of various waterborne toxicants[15,16].

Lead as a heavy metal is known to cause detrimental effects on aquatic organisms. *Labeo rohita* is a commercial fish and widely preferred as edible fish in india. It is very important to evaluate edible organisms like *Labeo rohita* from toxicity point of view as health of human being is directly associated with it. During the present course of investigation acute toxicity tests of Lead Nitrate to determine 96 hr $LC_{50}$ have been conducted on *Labeo rohita*, a fish having high nutritional value as well as a it serves as a good pollution indicator.

## Material and Methods

The experimental fish, *Labeo rohita* with an average weight of $10\pm2$ g, length of $10\pm1$ cm and of less than one year of age were collected from Nanoki fish farm located at Nanoki village of District Patiala, Punjab. They were acclimatized to laboratory conditions in dechlorinated tap water for 15 days in a plastic

tank of 1000 litre capacity equipped with filters and aerators. The heaters were also used during winters to protect the fingerlings from extreme cold. The water was changed after every 24 hours and fishes were given bath for 2-3 minutes in 0.1% $KMnO_4$ solution for the prevention of any disease. The fingerlings of the fish, *Labeo rohita* were fed with commercial artificial feed equal to $1/10^{th}$ of their body weight. Unconsumed feed and faecal wastes were siphoned daily with a rubber hose and the water replenished regularly. Feed was not provided to the test fishes 24 hrs before the commencement of the experiments. The water used for the test was made free of chlorine by exposing it to air for 24 hrs. The dead fish was immediately removed from the test tanks.

Toxicity tests have been performed in accordance with the standard methods given in APHA[17]. These were carried out in plastic tanks of 25 litre capacity. The stock solution of Lead Nitrate was freshly prepared which was renewed after every 24 hours. The fishes were exposed to different concentrations (1 mg/l to 50 mg/l) of $Pb(NO_3)_2$ for 96 hours. On the basis of fish mortality in each tank performing static bioassay test, 96hr $LC_{50}$ of Lead Nitrate calculated by Probit analysis was 34.20 mg/l[18].

## Results and Discussion

During the present investigation the 96hr $LC_{50}$ of Lead Nitrate for the fish, *Labeo rohita* was found to be 34.20 mg/l. The relation between the percentage mortality and the concentration of Lead Nitrate have been drawn (table-1). Figure-1. Shows the regression line between the probit kill of *Labeo rohita* and log concentration of Lead Nitrate.

**Table-1**
**Relation between concentration of Lead Nitrate and the percentage mortality of the fish**

| Dose mg/l | Log conc. | mortality | % mortality | Probit kill |
|---|---|---|---|---|
| 14 | 1.146 | 2 | 20 | 4.16 |
| 25 | 1.397 | 3 | 30 | 4.48 |
| 27 | 1.431 | 4 | 40 | 4.75 |
| 34.20 | 1.527 | 5 | 50 | 5.00 |
| 35 | 1.54 | 6 | 60 | 5.25 |
| 36 | 1.55 | 7 | 70 | 5.52 |
| 60 | 1.778 | 8 | 80 | 5.84 |
| 65 | 1.812 | 9 | 90 | 6.28 |
| 85 | 1.929 | 10 | 100 | - |

Regression line Equation: y = 0.30 + 3.19x

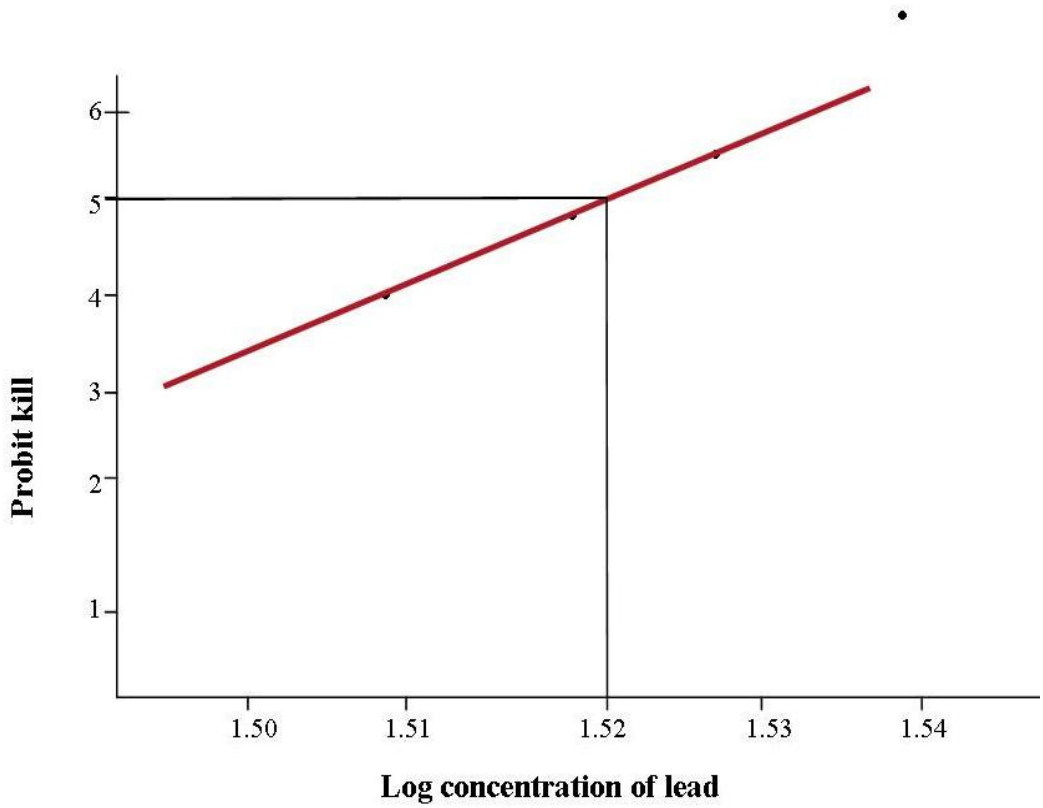


**Figure-1**
**Regression line between the probit kill of *Labeo rohita* and log concentration of Lead Nitrate**

Earlier the $LC_{50}$ values for the fish, *Labeo rohita* was found to be as 27.2 mg/l and 32.70 ± 2.23 mg/l when treated with Lead by different workers[19,20]. Similar results were also observed with other metals and different fish species as for example $LC_{50}$ value of $CuSO_4.5H_20$ for *Labeo rohita* was found to be 0.56mg/l and 3.15 mg/l and 96hr $LC_{50}$ of Lead Nitrate for *Clarias batrachus* was found to be 378 mg/l and for *Cyprinus carpio* was 2.624 mg/l by different workers[21-24]. There are differences in the value of $LC_{50}$ found in the same fish species for same heavy metal. Sometimes it is observed that some fishes are very sensitive towards the toxicity caused by one heavy metal and shows less sensitivity towards another equally toxic heavy metal at the same concentration[25]. In the same way some toxicants which can cause detrimental effects to some organisms even at low concentrations may be less or more toxic to some other organisms at higher or same concentration[26]. This is attributed to the fact that several factors including differences in the test species, age, feeding habit, sex, composition of toxicant and also the experimental conditions under which the tests are performed[19,27].

The high level of total dissolved solids in the water resultantly causes hardness. This high hardness play to reduce the availability of Lead Nitrate to the fish[28-31]. The variation in different physico-chemical parameters like low level of dissolved oxygen causes hypoxia, pH causes acidification and temperature causes hindrance to various physiological process and render fishes prone to intoxication but high hardness gives vice-versa effects to the toxicants[32]. The susceptibility of *Labeo rohita* to the toxic effect of Lead Nitrate is directly proportional to the concentration and duration of dose. If the dose increases the rate of mortality will also increase hence, confirms the observations made in *Oreochromis niloticus*, *Cyprinus carpio*, *Labeo rohita*, and *Poecilia reticulate* by the effect of different heavy metals[33-36].

## Conclusion

It is concluded that some organisms become sensitive to high concentrations of some heavy metal in the aquatic ecosystem and that causes deleterious effects on them. It helps us to determine the permissible limit of a toxicant in an ecosystem. Acute toxicity test reveals about the health of given aquatic ecosystem and eventually help us to propose policies to protect the aquatic ecosystem. It helps us to evaluate the environmental damage resulting from the pollutants and the establishment of water quality criteria to protect aquatic life.

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## References

**1.** Lopes P.A., Pinheiro T., Santos M.C., Mathias M., Collares-Pereira M.J. and Viegas-Crespo A.M., Response of antioxidant enzymes in freshwater fish populations (*Leuciscus alburnoides* complex) to inorganic pollutant exposure, *Sci. Tot. Environ*., **280**, 153-163 **(2001)**

**2.** Williams N.D. and Holdway D.A., Effect of pulse-exposed cadmium and zinc on embryo hatchability, larval development and survival of Australian crimson spotted rainbow fish (*Melanotaenia fluviatilis*), *Environ. Toxicol*., **15**, 165-173 **(2000)**

**3.** Olaifa F.E., Olaifa A.K., Adelaja A.A. and Owolabi A.G., Heavy metal contamination of *Clarias gariepinus* from a lake and fish farm in Ibadan, *Nig. Afr. J. Biomed. Res*., **7,** 145-148 **(2004)**

**4.** Chen C.Y. and Folt C.L., Bioaccumulation and diminution of arsenic and lead in a freshwater food web, *Environ. Sci. & Technol*., **34**, 3878-3884 **(2000)**

**5.** Canli M. and Atli G., The relationship between heavy metal (Cd, Cr, Fe, Pb, Zn) levels and the size of six Mediterranean fish species, *Environ. Pollut*., **121**, 129-136 **(2003)**

**6.** Yilmaz A., Turan C., and Toker T., Uptake and distribution of hexavalent Cr in tissues (gill, skin and muscle) of a freshwater fish, tilapia, *Oreochromis aureus*, *J. Environ. Chem. Ecotoxicol*., **2**, 28-33 **(2010)**

**7.** Sorensen E.M., Metal poisoning in fish. CRC Press, Boca Raton, USA., 243 **(1991)**

**8.** Heath A.G., Water pollution and fish physiology. CRC Press. Boca Raton, Florida, **(1995)**

**9.** Ciftci-Soyedemir N., Cicik B., Erdem C. and Ay O., Effects of lead concentrations on sera parameters and hematocrit levels in *Anguilla anguilla* L., 1758, *J. Fish. Sci. Com*., **2(4)**, 616-622 **(2008)**

**10.** Reglero M.M., Taggart M.A., Monsalve-Gonzalez I. and Mateo, R. Heavy metal exposure in large game from a lead mining area: effects on oxidative stress and fatty acid composition in liver, *Environ. Pollut*., **157**, 1388-1 **(2009)**

**11.** Abdallah G.M., El-Shayed S.M. and Abo-Salem O.M., Effect of lead toxicity on coenzyme Q levels in rat tissues, *Food. Chem. Toxicol*., **48**, 1753-1756 **(2010)**

**12.** Rout P.C. and Niak B.N., Quantitative precipitation tests for anti avidin during experimental plumbism in *Clarias batrachus*, Linn. Asian resonance, **2(3)**, **(2013)**

**13.** Dallas H.F. and Day J.A., The effect of water quality variables on riverine ecosystem. A review: Water Research Commission Report No., 351-360 **(1993)**

**14.** Woodward D.F., Brumbaug W.G., Delonay A.J. and Smith C., Effects on rainbow trout of metals contaminants diet of

benthic invertebrates from the Clark Fork river, Moutana, *Trans. American fish. Soc*., **23**, 51-62 **(1994)**

**15.** Mckim J.M., Evaluation of tests with early life stages of fish for predicting long term toxicity, *Can. J. Fish. Aquat. Sci*., **34**, 1148-1154 **(1977)**

**16.** Phillips T.A., Wu J., Summerfelt R.C. and Atchinson G.J., Acute toxicity and cholinesterase inhibition in larval and early juvenile walleye exposed to chloropyrifos, *Environ. Toxicol. Chem*., **21**, 1469-1474 **(2002)**

**17.** APHA, Standard methods for examination of water and wastewater, 22nd Edition. American Public Health Association. Washington D.C., **(2012)**

**18.** Finney D.J, Probit analysis. Cambridge University Press, London/Newyork, 20-47 **(1971)**

**19.** Latif A., Ali M., Sayyed A.H., Iqbal F., Usman K., Rauf M. and Kaoser R., Effect of copper sulphate and lead nitrate, administered alone or in combination, on the histology of liver and kidney of *Labeo rohita*, *Pak. J. Zool*., **45(4)**, 913-920 **(2013)**

**20.** Abdullah S., Javed M. and Javid A., Studies on acute toxicity of metals to the fish (*Labeo rohita*), *Int. J. Agri. Biol*., **9**, 333-337 **(2007)**

**21.** Javid A., Javed M. and Abdullah S., Nickel bioaccumulation in the bodies of *Catla catla*, *Labeo rohita* and *Cirrhinus mrigala* during 96hr $LC_{50}$ exposures. *Int. J. Agri. Bio*., **9**, 139-142 **(2007)**

**22.** Adhikari S., Effect of calcium and magnesium hardness on acute copper toxicity to Indian major carp, *Labeo rohita* (Hamilton) and catfish, *Channa punctatus* (Bloch), *Aquacul. Res*., **34**, 975-980 **(2003)**

**23.** Shamshun N., Chandan K., Kavita M. and Mamta K., Lead induced alteration in blood profile of air breathing catfish, *Clarias batrachus* Linn. *The Bioscan*:, **2, (2010)**

**24.** Hamed N. and Esmail G., Determination of $LC_{50}$ of lead nitrate and copper sulphate in common carp (*Cyprinus carpio*), *Amer. Eura. J. Toxicol. Sci.,* **4(2)**, 60-63 **(2012)**

**25.** Das K.K. and Banerjee S.K., Cadmium toxicity in fishes, *Hydrobiol*., **75**, 117-121 **(1980)**

**26.** Shah S.L. and Altindu A., Effects of heavy metal accumulation on the 96hr $LC_{50}$ values in tench, *Tinca tinca* L., 1758. *Turk. J. vet. Anim. Sci*., **29**, 139-144 **(2005)**

**27.** Witeska M., Jezierska B. and Chaber J., The influence of cadmium on common carp embryos and larvae, *Aquaculture*, **129**, 129-132 **(1993)**

**28.** Demayo A., Taylor M.C., Taylor K.W. and Hodson P.V., Toxic effects of lead and lead compounds on human health, aquatic life, wildlife plants and livestock, *Critical reviews in environmental control*., **12(4)**, 257-305 **(1981)**

**29.** Hodson P.V., Whittle D.M., Wong P.T.S., Borgmann U., Thomas R.L., Chau Y.K., Nriagu J.O. and Hallet DJ., Lead contamination of the great lakes and its potential effects on aquatic biota. *In:* Nriagu J.O. and Simmons M.S. (eds), *Toxic contaminants in the great lakes*, John Wiley and Sons, Indianapolis **(1984)**

**30.** Nussey G., Vuren V.J.H.J. and Prez H.H., Bioaccumulation of chromium, manganese, nickel and lead in the tissues of the moggel, (*Labeo umbratus*) from Witbank Dam. Mpumalanga, *Water S.A.,* **26(2)**, 264-284 **(2000)**

**31.** Martinez C.B.R., Nagae M.Y., Zaia C.T.B.V and Zaia D.A.M., Acute morphological and physiological effects of lead in the neotropical fish, *Prochilodus lineatus*, *Braz. J. Biol*., **64**, 797-807 **(2004)**

**32.** Witeska M. and Jezierska B., The effect of environmental factors on metal toxicity to fish, *Fresenius. Environ. Bull*., **12**, 824-829 **(2003)**

**33.** Mulley D., Kamble G.M. and Bhilave M., Effect of heavy metals on nucleic acids in *Cyprinus carpio*, *J. Environ. Bio*., **21**, 367-370 **(2000)**

**34.** Yilmaz M., Gul A. and Karakose E., Investigation of acute toxicity and the effect of cadmium chloride metal salt on behaviour of the guppy (*Poecilia reticulate*), *Chemospher*, **56**, 375-380 **(2004)**

**35.** Dardenne F., Nobels I., De C. and Blust, R., Dose response relationships and statistical performance of a battery of bacterial gene profiling assays, *Apps. Microbiol. Biotechnol*., **75**, 223-234 **(2007)**

**36.** Singh A., Jain D. and Kumar P., Determination of $LC_{50}$ of cadmium chloride in *Heteropneustes fossilis*, *GERF Bull. of Biosci*., **1**, 21-24 **(2010)**