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Research Article

# HEAVY METAL IMPACT OF NICKEL SULPHATE ON FISH *LABEO ROHITA*

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**ABSTRACT**

Ulterior effect of heavy metals present in water bodies on the aquatic life has been a subject of research since last few years. Cadmium, Cobalt, Copper, Lead, Mercury, Nickel and Zinc, are most harmful metabolic pollutants that cause such aquatic damage. The negative effects are further compounded by the reaction of these heavy metals with water soluble, non-biodegradable, strong oxidizing agents, polypeptides and proteins. Research carried out till-date has failed to precisely determine the mechanism of the toxic action of heavy metals on aquatic life, and no theory of heavy metals toxicity is completely convincing as of today. The present study deals with the effect of heavy metal, Nickel sulphate on laboratory acclimatized fish *Labeo rohita.* It has been observed that nickel salts affect the gills of the fish by covering them with a mucous layer and darkening of the colour. Nickel salts like Nickel sulphate is widely used in many industrial applications. Untreated effluents from these industries, laden with these salts gain unrestricted entry into various water bodies. Industrial growth in the last few decades has compounded this pollution problem leading to catasphoric conditions. Nickel is a wide spread element in nature, and is required in only particular minimum amount for the growth of all living organisms like plants, man and animals. Any ingestion above this minimum required quantity causes evident deleterious effects. Previous investigations have pointed towards the presence of amino acids like Proline, Aspartic acid, Glycine, Leucine in fish muscles by incorporating paper chromatography or column chromatography or circular chromatography techniques. These amino acids are further sub-classified into two types: free amino acids and bound amino acids. The current investigation has been focused on the free amino acids that are present in the fish muscles. Increased durations of exposure to the pollutant has led to a decrease in amino acids and resultant damage to the muscles. Amino acids, that were present in control: Proline, Aspartic acid, Glycine and Leucine, were found to be conspicuously missing in higher concentration of the pollutant.

**Key words:** Nickel Sulphate, Chromatography, Proline, Aspartic acid, Glycine and Leucine.

## INTRODUCTION

Pollution can be safely defined as an artificial variation in the equilibrium of the eco-system caused by the daily activities of man-kind. The agents that cause such pollution originates at one place, generally land, are then transported by aqueous medium to the water bodies causing damaging effect. So, pollution is a 'transport effect phenomenon'. Industrial growth coupled with human population explosion constitute to an increased demand on the natural resources, as well as an increased discharge of effluents causing pollution and global warming. Water bodies are polluted by the untreated discharge, both from municipal sewers and industrial effluents. Pulp and paper, textile, sugar and steel works, tanneries, distilleries, chemical and fertilizer plants are some of the major deleterious contributors in this field; leading to an extreme increase of 62.0 ppm chlorine content of the water bodies against a safe tolerant limit of 1.0 ppm.

Fish thriving in such contaminated bodies ingest the toxic products by their food and water for respiration [1,2]. This leads to the accumulation of heavy metals in different organs of the fish including the liver [3,4,5,6]. Though fish liver is seldom consumed, its qualitative

assessment gives better biomonitoring of metal pollution[7]. Continuous monitoring of the water bodies and the aquatic ecosystem for the ulterior effects of heavy metals is necessary for all the species that thrive on these water mass [8]. Human consumption of such affected fish leads to a number of temporary and permanent health problems in humans. Since fish are a natural and good source of protein and unsaturated omega-3 fatty acid for human, monitoring of their quality and health aspect is a subject of great importance [1,2,9,10,11,12,13,14,15,16]. A better example is the River Kaveri crossing three southern states of India. This study has been confined to the assessment of the effect of heavy metal pollution on the fish thriving in the delta of this river, with their different feeding habits and also to find the accumulation pattern in different organs viz., muscle tissue, liver and gill of fish from Anaikarai dam of River Kaveri. Heavy, dense, metallic elements which are toxic to human beings and animals and which tend to accumulate in the body are known as trace metals. These toxic metals even in trace amounts, such as Arsenic, Cadmium, Chromium, Cobalt, Iron, Lead, Manganese, Mercury, Nickel and Tin have an undesirable effect on the entire ecosystem.

## MATERIALS AND METHODS

### A. Processing of the fresh water fingerlings major carp Labeo rohita for study:

Healthy fresh water fingerlings major carp *Labeo rohita* of both sexes were collected from the local fish farms. The fingerlings were transported in polythene bag containing O2 saturated water and brought to the aquarium nearly 125 to 160 fingerlings of *Labeo rohita* and were kept as stock. Normal fresh water, commercial fish food was given twice a day. Dissolved Oxygen level and pH of water was maintained in the laboratory to acclimatize the fish for 15 to 20 days. Different concentration of Nickel was made by dissolving analytical grade Nickel sulphate in the fresh water. Amino acids were separated from the fish tissue with the help of centrifuge. For the qualitative and quantitative analysis of amino acids, ascending paper chromatographic techniques was used. 3 sets of different concn of Nickel sulphate (40, 50, 60 ppm) were prepared for treatment.

### Preparation of sample:

Extraction of free a. a. from the fish tissues: Weighed tissues (300 mg), were homogenized in glass homogenizer in 80 % alcohol and was left for 24 hours at 0 – 4 ºC. The homogenous mixture was centrifuged after 24 hours and alcoholic supernatant was filtered and evaporated over water bath at 80 ºC. The dried residue was extracted with 1 ml of 10 % aqueous ethanol. These samples were used for chromatography for the determination of free amino acids

**Solvent system:** *n*-Butanol: Glacial acetic acid: Distilled water - 4:1:5

**Locating reagent:** 0.2 % ninhydrin in acetone was used as location reagent.

**Procedure:** Paper chromatographic technique is used for the detection of amino acids present in the fish muscle. Chromatograms were obtained. The amino acid location appeared as pinkish violet spots. The spots were circled with a pencil. The individual spots were cut out from the chromatograms and coloured spots from paper pieces in test tubes were extracted with 0.42 % Sodium bicarbonate in 48 % ethanol and kept undisturbed overnight. In the morning the test tubes were shaken and after the filter paper pieces settled down the coloured solutions were decanted in colour Imation tubes. The colour intensity of known and unknown spots of chromatograms was determined with the help of photo colorimeter using green filter. Then optical densities of amino acids were measured with the help of photo colorimeter.

## RESULT AND DISCUSSION

The results denote that Nickel sulphate prominently affects the fish muscles as some amino acids are found to be reduced or even completely missing. This directly impairs the nutritive quality of the fish as they are of the main nutritive part of the fish containing high percentage of protein. Hence, the current study has been concentrated on presence of amino acids in the fish muscles including variation in its content,

The results obtained after performing the experiments are represented in the following table no. 01. It represents the results of all the four amino acids found to be present in fish *Labeo rohita*. The decreasing amount of the amino acid Glycine ,Aspartic acid, Proline and Leucine is shown in graph no.01, 02,03 and 04 respectively.

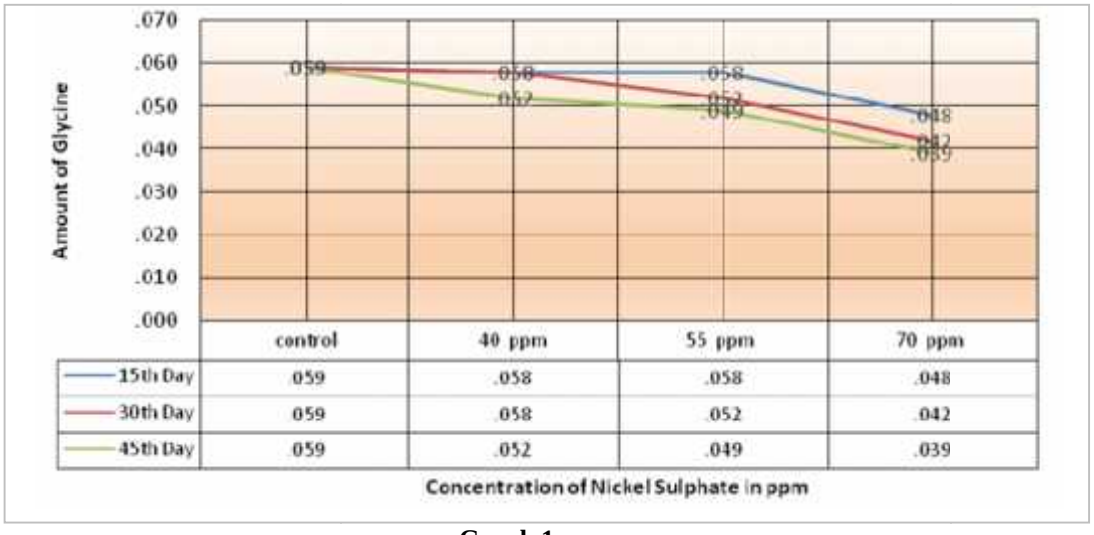
## CONCLUSION

Protein deficiency in a human body leads to a number of health related problems, including reduction of chondroblastic and osteoblastic activity. This leads to an acute situation where the normal growth and formation of the bones and the cartilage gets hampered, which finally results in irreparable deformities in the body of the growing children, and also in adults. Fish is the staple food of the people of South Gujarat region. Hence it is a matter of utmost importance to study and constantly monitor the various heavy metal pollutants in water in the Southern region of the state of Gujarat.

**Table 1**

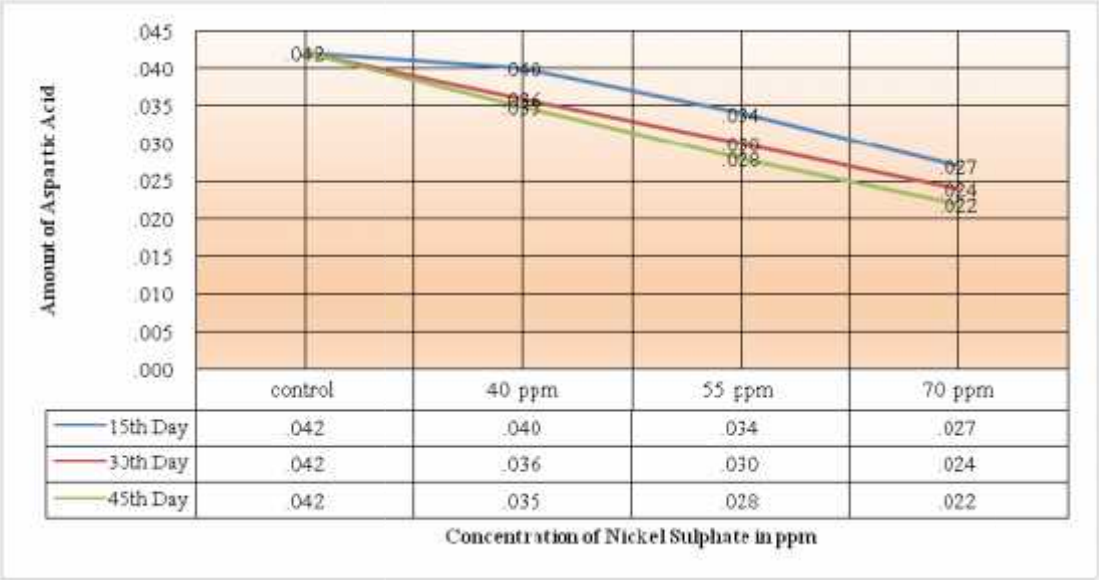
**Change in optical density of different amino acids after exposure to various concentrations of Nickel sulphate.**

| Amino acids | Nickel concentration | Exposure time ( Days ) | | |
|---|---|---|---|---|
| | | 15 | 30 | 45 |
| ( 01 ) | (02) | (03) | (04) | (05) |
| **Glycine** | Control | 0.059 | 0.059 | 0.059 |
| | 40 ppm | 0.058 | 0.058 | 0.052 |
| | 55 ppm | 0.058 | 0.052 | 0.049 |
| | 70 ppm | 0.048 | 0.042 | 0.039 |
| **Aspartic Acid** | Control | 0.042 | 0.042 | 0.042 |
| | 40 ppm | 0.040 | 0.036 | 0.035 |
| | 55 ppm | 0.034 | 0.030 | 0.028 |
| | 70 ppm | 0.027 | 0.024 | 0.022 |
| **Proline** | Control | 0.028 | 0.028 | 0.028 |
| | 40 ppm | 0.028 | 0.022 | 0.020 |
| | 55 ppm | 0.021 | 0.018 | 0.018 |
| | 70 ppm | 0.017 | 0.015 | 0.010 |
| **Leucine** | Control | 0.032 | 0.032 | 0.032 |
| | 40 ppm | 0.032 | 0.030 | 0.026 |
| | 55 ppm | 0.029 | 0.022 | 0.018 |
| | 70 ppm | 0.020 | 0.018 | 0.011 |

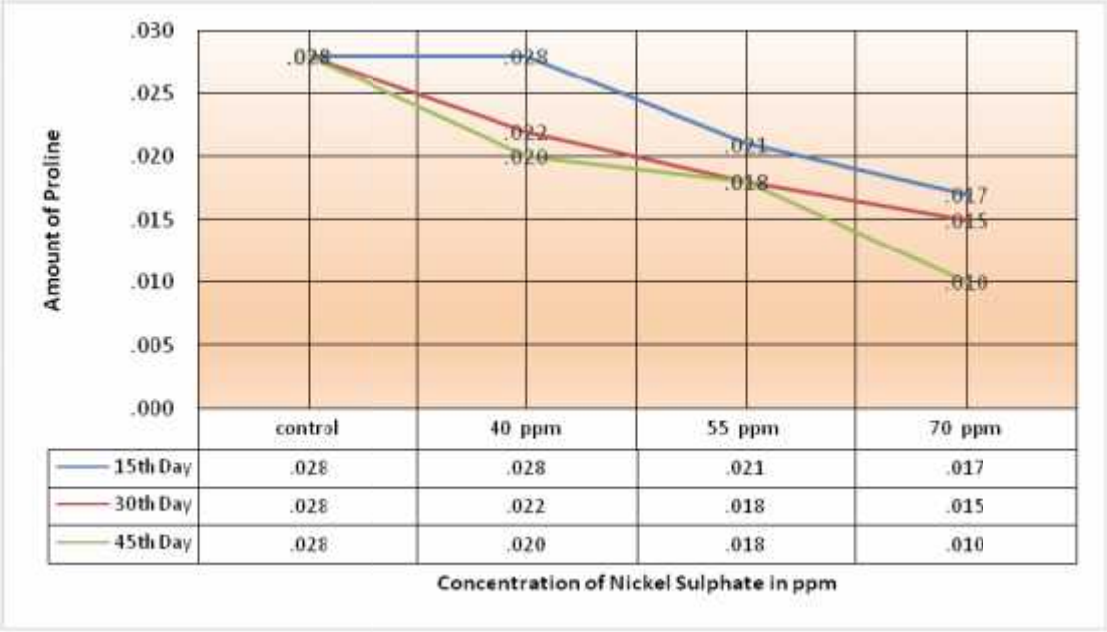



**Graph 1**

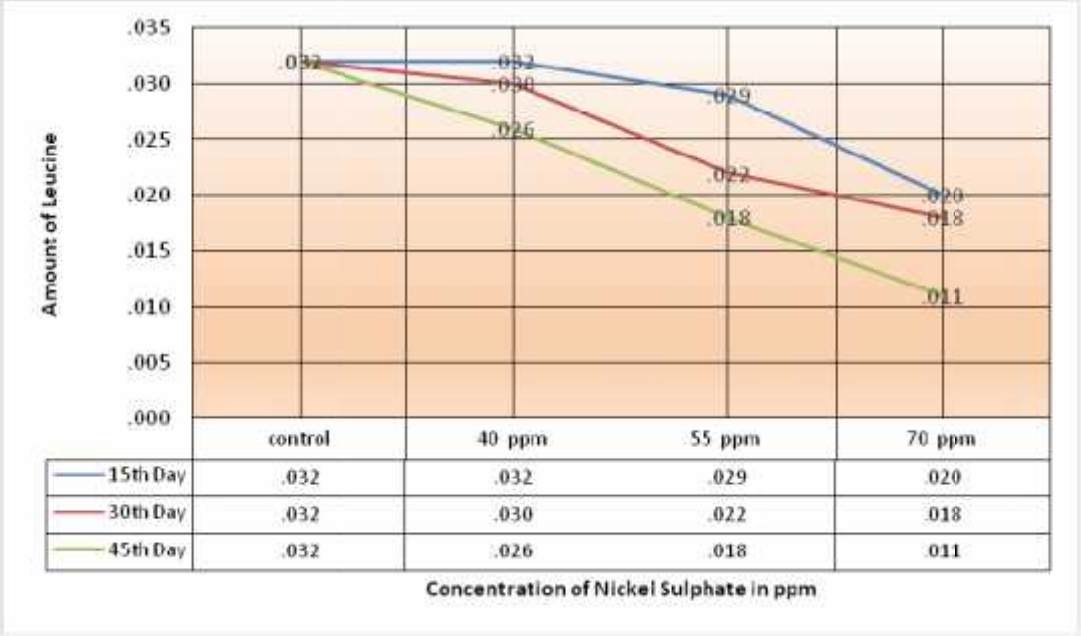
**Graph showing optical density change of Glycine after exposure to various concentration of Nickel Sulphate.**




**Graph 2**
**Graph showing optical density change of Aspartic Acid after exposure to various concentration of Nickel Sulphate.**




**Graph 3**
**Graph showing optical density change of Proline after exposure to various concentration of Nickel Sulphate.**




**Graph 4**
**Graph showing optical density change of Leucine after exposure to various concentration of Nickel Sulphate.**

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