

Determination of Lc50 Values of Heavy Metals in Fish

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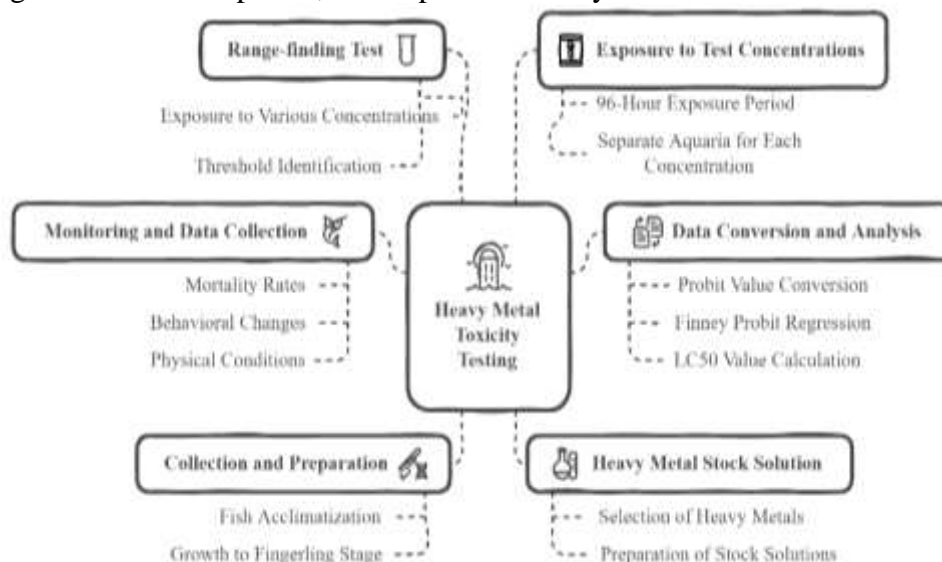
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SUMMARY

This review investigates the acute toxicity of heavy metals to aquatic organisms using the LC50 method, which determines the lethal concentration at which 50% of the exposed fish population dies. The research involves the collection and acclimatization of fish samples, preparation of heavy metal stock solutions, and the implementation of a range-finding test to select appropriate concentrations for the main experiment. Over a 96-hour exposure period, fish are monitored for mortality, behavioural changes, and physical conditions at different concentrations. Mortality data is converted into probit values, and Finney's Probit Regression is employed to calculate the LC50 value. The study highlights the significance of LC50 determination in assessing environmental risks associated with heavy metal contamination. By providing crucial insights into the toxic effects of pollutants, this method supports the development of effective pollution control and environmental management strategies.

INTRODUCTION

In ecotoxicology, tests mostly assessed how different concentrations of hazardous components responded to the morphologies of the experimental organisms over a specific period of time. The median lethal concentration (LC50) of the concentration-response relationship is used to determine the toxicant's potency (Ghosh and Saha, 2022). The LC50 and EC50 represent the concentration of a substance that causes mortality (LC50) or a specific effect (EC50) in 50% of an exposed population. Acute fish toxicity tests can be used to determine whether a xenobiotic is harmful to aquatic ecosystems and the food chain. The oxidative processes of biological macromolecules are catalysed by metals, and oxidative tissue damage can result from their toxicity. The lack of comprehensive research on toxicity caused by environmental contaminants is a significant challenge in experimental ecotoxicology. Animals' reactions to environmental contamination can be both beneficial and detrimental at different structural levels, including cells, tissues, and organs. The type of pollutant, its concentration, the rate of exposure, and the susceptibility of the organism all affect these reactions (Ali *et al.*, 2016). LC50 testing is widely used in regulatory frameworks, such as those outlined by organizations like the Organisation for Economic Co-operation and Development (OECD) and the U.S. Environmental Protection Agency (EPA). It is applied in assessing the toxicity of industrial effluents, agricultural pesticides, heavy metals, pharmaceuticals, and other pollutants. These tests are especially significant in areas experiencing rapid industrialization or agricultural development, where pollutants may enter water bodies and threaten aquatic life.



What does Lethal Concentration (LC₅₀) Means?

LC₅₀ (Lethal Concentration 50%) refers to the concentration of a chemical in the experimental water at which 50% of the test population is either unable to function or succumbs to mortality when exposed under controlled testing conditions for a specified duration (APHA, 1960).

Methods involved in heavy metal toxicity tests (LC₅₀)

Collection and Preparation of fish samples

The study begins by collecting fish sample from selected coastal sites. These fish are then reared in controlled laboratory conditions for one month, ensuring they adapt to the new environment. During this time, the fish grow to a fingerling stage, weighing between 8–15 grams and measuring about 6-7cm, which is ideal for toxicity testing. Proper acclimatization during this period is crucial to reduce stress and ensure the reliability of the test results.

Preparation of heavy metal stock solution

The heavy metals with the highest concentrations from residue analysis will be selected for toxicity testing, and precise stock solutions will be prepared to create the required test concentrations for the experiment.

Range-finding test

A preliminary range-finding test exposes fish to a wide range of concentrations to identify toxicity thresholds. Based on the results, low, medium, and high concentrations are selected for the main experiment (OECD, 2000).

Exposure to test concentrations

The selected test concentrations are introduced into separate aquaria, each representing a unique heavy metal concentration. Once the fish reach the desired size, they will be exposed to different tests concentrations of heavy metals in three separate tanks, each containing a different concentration. The exposure lasts for 96 hours, during which the fish are monitored to observe their response to the heavy metals.

Monitoring and Data collection

Throughout the 96-hour exposure period, data on mortality rates, behavioural changes (e.g., swimming patterns and feeding), and physical conditions (e.g., discoloration or lesions) are recorded at intervals of 24, 48, 72, and 96 hours (Sadeghi and Peery, 2018). This step helps in identifying the concentration at which 50% of the fish population dies. The observed mortality percentages at each concentration are critical for calculating the LC₅₀ value.

Conversion of data to Probit values

The mortality data is converted into probit values using statistical tables or software. Probit transformation linearizes the sigmoid dose-response curve, making it suitable for regression analysis. This step simplifies the relationship between concentration and mortality, allowing for the use of linear regression methods (OECD, 1992).

Finney Probit Regression Analysis

Using Finney's Probit Regression method, a regression equation is derived. The logarithms of the heavy metal concentrations are plotted on the x-axis, and the corresponding probit values of mortality are plotted on the y-axis (Finney, 1971).

Determination of LC₅₀ value

A straight line is fitted to the data, and the equation is used to calculate the LC₅₀ value. This value represents the concentration at which 50% of the fish population is expected to die and corresponds to a probit value of 5.0.

CONCLUSION

In summary, the LC₅₀ determination of heavy metals is a vital process for assessing their acute toxicity in aquatic organisms. By identifying the concentration at which 50% mortality occurs, this method provides

crucial insights into the environmental risks posed by heavy metal contamination. The use of standardized methodologies, such as Finney's Probit Regression and additional validation techniques, ensures accurate and reliable results, supporting effective environmental management and pollution control strategies.

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