**Acute toxicity of Cadmium on *Labeo rohita*: Understanding Behavioral Patterns and Dose-Dependent Effects**

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**Abstract:**

This comprehensive study delves into the intricate realm of acute toxic effects incited by cadmium on *Labeo rohita*, a highly pertinent freshwater fish species. Rigorous toxicity tests were conducted, employing a static method extending over varying durations: 24, 48, 72, and 96 hours, aiming to ascertain the lethal concentration (LC50) values. Astonishingly, the LC50 values obtained were found to be 0.309, 0.247, 0.207, and 0.160 ppm, presenting empirical evidence of the escalating menace cadmium poses over time to aquatic ecosystems. Evidencing the severity of the impact, the exposed fish displayed a myriad of dose-dependent behavioral anomalies, encompassing erratic swimming, loss of balance, surfacing, and convulsions. These pronounced modifications parallel previous research that has documented similar trends. Augmenting the credibility of these results, a meticulous analysis of the existing literature was performed, supplemented by the utilization of the aptly named Statistical Package for the Social Sciences (SPSS) software, enabling the determination of regression values and 95% confidence limits for the LC50 values during each experimental interval. Such quantified insights synthesize a comprehensive understanding of the detrimental effects of cadmium, propelling the urgency to adequately monitor and address its environmental contamination, while preserving the delicate balance of aquatic ecosystems.

**Keywords:** Cadmium, Acute toxic effects, *Labeo rohita*, LC50 values, Aquatic ecosystems

1. **INTRODUCTION:**

Cadmium, a heavy metal pollutant, poses a grave threat to aquatic ecosystems due to its widespread presence resulting from numerous industrial activities (Biswas et al., 2018, Rahman and Singh, 2019, Fatima et al., 2019,). Understanding the acute toxicity of cadmium on freshwater fish species is crucial for assessing its impact on aquatic organisms and ecosystems (Malik et al., 2020, Vilas boas et al., 2020, Gu et al., 2021). *Labeo rohita*, a commonly studied fish species, serves as an ideal indicator of environmental contamination due to its high sensitivity to toxic substances (Pal et al., 2016, Tabrez et al., 2022, Kumar et al., 2022). This study aims to unravel the acute toxic effects of cadmium on *Labeo rohita* and critically examine the dose-dependent behavioral patterns exhibited by the fish.

1. **MATERIALS AND METHODS:**

**Test Organism:**

A healthy population of *Labeo rohita* was obtained from a local freshwater fish hatchery (Devuni kadapa). Prior to experimentation, the fish (7-8cm) were acclimated for one week in laboratory conditions, ensuring temperature (25±2°C) and dissolved oxygen levels (6–8mg/L) consistent with their natural habitat.

**Chemical Preparation:**

Cadmium chloride (CdCl2), of analytical grade, was procured from a reputable chemical supplier. Stock solutions of different concentrations (1000 ppm) were prepared using deionized water. Serial dilutions were made to achieve the required test concentrations. Toxicity Tests: Static toxicity tests were conducted, adhering to the standard procedure recommended by the OECD guidelines for testing chemicals (OECD, 2012). The experimental setup comprised of four test groups exposed to different concentrations of cadmium (0.1 ppm, 0.2 ppm, 0.3 ppm, 0.4 ppm) for durations of 24, 48, 72, and 96 hours, respectively. A control group, maintained in deionized water, was also included for comparison.

**Behavioral Observations:**

Throughout the exposure periods, continuous observation was carried out to detect any behavioral changes exhibited by the fish. Key observations encompassed swimming patterns, loss of balance, surfacing, and convulsions.

**Data Analysis:**

The obtained data was subjected to probit analysis to calculate the lethal concentration

(LC50) values. Percentage mortality observed in each test group was incorporated into the analysis (Finney, 1981). Regression values and 95% confidence limits for the LC50 values were determined using the Statistical Package for the Social Sciences (SPSS) software, enhancing the reliability and precision of the results.

**III. RESULTS AND DISCUSSION:**

The acute toxicity of cadmium on *Labeo rohita* was assessed through static toxicity tests conducted over different durations: 24, 48, 72, and 96 hours. The obtained LC50 values (lethal concentration at which 50% of the fish population is expected to die) were found to be 0.309 ppm, 0.247 ppm, 0.207 ppm, and 0.160 ppm, respectively. These values provide significant empirical evidence of the increasing menace cadmium poses over time to aquatic ecosystems.

The fish exposed (1/10th of 96h LC50) to cadmium exhibited varying degrees of behavioral anomalies that were dose-dependent, including erratic swimming, loss of balance, surfacing, and convulsions. These findings align with previous studies that have reported similar dose-dependent effects of cadmium on fish behavior (Durmaz et al, 2006, Biswas et al., 2018, Pal et al., 2016; Gade et al, 2021). The alterations in behavior observed in *Labeo rohita* serve as clear indicators of the toxic impact of cadmium on this species.

Comparing the results obtained in this study with earlier works sheds light on the consistent trends in the toxic effects of cadmium on fish. Biswas et al. (2018) documented cadmium-induced oxidative stress in *Labeo rohita*, supporting the notion that cadmium leads to significant physiological disturbances in fish (Malarvizhi et al., 2017, Junejo et al., 2019). Furthermore, Pal et al. (2016) conducted an acute toxicity test on *Labeo rohita* using various heavy metals, including cadmium, and found similar behavioral changes in fish exposed to these substances (Mohanty et al., 2013, Robey et al., 2017, Chakraborty and Chakraborty, 2021, Singh & Sexena, 2020). Such consistency in the observed effects suggests a robust pattern of cadmium toxicity in *Labeo rohita* and strengthens the validity and generalizability of the present findings

The regression analysis for the LC50 values and the calculation of 95% confidence limits provide essential statistical insights into the toxicity of cadmium on *Labeo rohita* during each experimental interval. These values contribute to a more comprehensive understanding of the relationship between cadmium concentration and fish mortality.

These results underscore the urgent need for effective monitoring and regulation of cadmium contamination in aquatic environments to protect the delicate balance of ecosystems. The hazardous effects of cadmium on *Labeo rohita* serve as an indication of its potential impact on other fish species and highlight the importance of implementing measures to mitigate its environmental presence.

**Fig.1.**

Logit (P) and Probit graph

-

1.2166

-

0.2087

0.202

1.0554

2.4087

y = 0.8515x

-

2.1063

R² = 0.9698

-1.5

-1

-0.5

0

0.5

1

1.5

2

2.5

3

0

1

2

3

4

5

6

Logit(P)

Log (C%)

10

30

50

70

80

84

3.71

4.47

56

5

5.5244

5.8416

y = 0.5295x + 3.3234

R² = 0.9797

0

1

2

3

4

5

6

7

0

10

20

30

40

50

60

70

80

90

0.677

0.585

0.508

0.443

0.387

Probit mortality

Per cent mortality

Log Concentration

Per cent mortality

Probit mortality

Linear (Probit mortality)

**Fig.2.** per cent mortality and Probit mortality at 24 hour

20

40

50

70

80

4.1584

4.7467

5

5.5244

5.8416

y = 0.4144x + 3.811

R² = 0.9864

0

1

2

3

4

5

6

7

0

10

20

30

40

50

60

70

80

90

0.3665

0.3979

0.4317

0.4685

0.5086

**Probit mortality**

**Per cent mortality**

**Log Concentration**

Per cent mortality

Probit mortality

Linear (Probit mortality)

**Fig.3.** Per cent mortality and Probit mortality at 96 hour

**Table.1.** Static 24 h per cent mortality and probit mortality of the fish, *Labeo rohita* exposed to cadmium.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| S.NO. | DOSE IN  PPM | PERCENT MORTALITY | PROBIT VALUE  (Y) | LOG(100\* DOSE) =X | X\*X | Y\*Y | XY |
| 1 | 0.21 | 10% | 3.7184 | 1.322219295 | 1.748263863 | 13.82649856 | 4.916540226 |
| 2 | 0.26 | 30% | 4.4756 | 1.414973348 | 2.002149575 | 20.03099536 | 6.332854716 |
| 3 | 0.31 | 50% | 5 | 1.491361694 | 2.224159702 | 25 | 7.456808469 |
| 4 | 0.36 | 70% | 5.5244 | 1.556302501 | 2.422077474 | 30.51899536 | 8.597637535 |
| 5 | 0.41 | 80% | 5.8416 | 1.612783857 | 2.601071768 | 34.12429056 | 9.421238177 |
|  |  |  | 24.56 | 7.397640694 | 10.99772238 | 123.5007798 | 36.72507912 |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  | X | 1.479528139 |  |  |  |  |
|  |  | Y | 4.912 |  |  |  |  |
|  |  | SXX | 0.052704815 |  |  |  |  |
|  |  | SYY | 2.86205984 |  |  |  |  |
|  |  | SXY | 0.387868034 |  |  |  |  |
|  |  | SLOPE B | 7.359252299 |  |  |  |  |
|  |  | VARIANCE B | 1.897359835 |  |  |  |  |
|  |  | VARIANCE A | 4.253327345 |  |  |  |  |
|  |  | M | 1.491485875 |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  | Log (100) | -2 |  |  |  |  |
|  |  | Anti log | -0.509 |  |  |  |  |
|  |  | LC50 Value | 0.309 |  |  |  |  |

**Table.2.** Static 48 h per cent mortality and probit mortality of the fish, *Labeo rohita* exposed to cadmium.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| S.NO. | DOSE IN  PPM | MORTALITY | PROBIT VALUE | LOG(100\* DOSE) =X | X\*X | Y\*Y | XY |
| 1 | 0.16 | 20% | 4.1584 | 1.204119983 | 1.449904933 | 17.29229056 | 5.007212536 |
| 2 | 0.21 | 30% | 4.4756 | 1.322219295 | 1.748263863 | 20.03099536 | 5.917724676 |
| 3 | 0.25 | 50% | 5 | 1.397940009 | 1.954236268 | 25 | 6.989700043 |
| 4 | 0.31 | 70% | 5.5244 | 1.491361694 | 2.224159702 | 30.51899536 | 8.238878541 |
| 5 | 0.36 | 80% | 5.8416 | 1.556302501 | 2.422077474 | 34.12429056 | 9.091296688 |
|  |  |  | 25 | 6.971943481 | 9.79864224 | 126.9665718 | 35.24481248 |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  | X | 1.394388696 |  |  |  |  |
|  |  | Y | 5 |  |  |  |  |
|  |  | SXX | 0.07704306 |  |  |  |  |
|  |  | SYY | 1.96657184 |  |  |  |  |
|  |  | SXY | 0.385095081 |  |  |  |  |
|  |  | SLOPE B | 4.998439587 |  |  |  |  |
|  |  | VARIANCE B | 1.297975443 |  |  |  |  |
|  |  | VARIANCE A | 2.6236794 |  |  |  |  |
|  |  | M | 1.394388696 |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  | Log (100) | -2 |  |  |  |  |
|  |  | Anti log | -0.606 |  |  |  |  |
|  |  | LC50 Value | 0.247 |  |  |  |  |

**Table.3.** Static 72 h per cent mortality and probit mortality of the fish, *Labeo rohita* exposed to cadmium.

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| S.NO. | DOSE IN PPM | MORTALITY | PROBIT VALUE | LOG(100 \*DOSE) =X | X\*X | Y\*Y | XY |
| 1 | 0.11 | 10% | 3.7184 | 1.041392685 | 1.084498725 | 13.82649856 | 3.87231456 |
| 2 | 0.16 | 20% | 4.1584 | 1.204119983 | 1.449904933 | 17.29229056 | 5.007212536 |
| 3 | 0.2 | 50% | 5 | 1.301029996 | 1.69267905 | 25 | 6.505149978 |
| 4 | 0.26 | 60% | 5.2533 | 1.414973348 | 2.002149575 | 27.59716089 | 7.433279489 |
| 5 | 0.31 | 90% | 6.2816 | 1.491361694 | 2.224159702 | 39.45849856 | 9.368137616 |
|  |  |  | 24.4117 | 6.452877705 | 8.453391984 | 123.1744486 | 32.18609418 |
|  |  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  | X | 1.290575541 |  |  |  |  |
|  |  | Y | 4.88234 |  |  |  |  |
|  |  | SXX | 0.125465848 |  |  |  |  |
|  |  | SYY | 3.988229192 |  |  |  |  |
|  |  | SXY | 0.680951244 |  |  |  |  |
|  |  | SLOPE B | 5.42738325 |  |  |  |  |
|  |  | VARIANCE B | 0.79702964 |  |  |  |  |
|  |  | VARIANCE A | 1.427520794 |  |  |  |  |
|  |  | M | 1.312254497 |  |  |  |  |
|  |  |  |  |  |  |  |  |
|  |  | Log (100) | -2 |  |  |  |  |
|  |  | Anti log | -0.684 |  |  |  |  |
|  |  | LC50 Value | 0.207 |  |  |  |  |

**Table.4.** Static 96 h per cent mortality and probit mortality of the fish, *Labeo rohita* exposed to cadmium.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| DOSE IN PPM | MORTALITY | PROBIT VALUE | LOG(100\* DOSE) =X | X\*X | Y\*Y | XY |
| 0.6 | 20% | 4.1584 | 1.77815125 | 3.161821869 | 17.29229056 | 7.39426416 |
| 0.11 | 40% | 4.7467 | 1.041392685 | 1.084498725 | 22.53116089 | 4.943178659 |
| 0.16 | 50% | 5 | 1.204119983 | 1.449904933 | 25 | 6.020599913 |
| 0.21 | 70% | 5.5244 | 1.322219295 | 1.748263863 | 30.51899536 | 7.304468272 |
| 0.26 | 80% | 5.8416 | 1.414973348 | 2.002149575 | 34.12429056 | 8.26570831 |
|  |  | 25.2711 | 6.760856561 | 9.446638965 | 129.4667374 | 33.92821931 |
|  |  |  |  |  |  |  |
|  |  |  |  |  |  |  |
|  | X | 1.352171312 |  |  |  |  |
|  | Y | 5.05422 |  |  |  |  |
|  | SXX | 0.304802678 |  |  |  |  |
|  | SYY | 1.741038328 |  |  |  |  |
|  | SXY | -0.242637134 |  |  |  |  |
|  | SLOPE B | -0.796046596 |  |  |  |  |
|  | VARIANCE B | 0.328081107 |  |  |  |  |
|  | VARIANCE A | 0.699852754 |  |  |  |  |
|  | M | 1.420282903 |  |  |  |  |
|  |  |  |  |  |  |  |
|  | Log (100) | -2 |  |  |  |  |
|  | Anti log | -0.58 |  |  |  |  |
|  | LC50 Value | 0.16 |  |  |  |  |

1. **RECOVERY STUDY**

In addition to investigating the acute toxicity of cadmium on *Labeo rohita*, a recovery study was conducted to assess the fish's ability to recover from the toxic effects after exposure to cadmium. The recovery period lasted for 14 days, during which the fish were kept in a clean and uncontaminated environment.

The results of the recovery study revealed that *Labeo rohita* exhibited a considerable capacity for recovery from the toxic effects of cadmium. As the fish were removed from the contaminated environment and placed in clean water, their behavioral patterns gradually returned to normal. The previously observed anomalies such as erratic swimming, loss of balance, surfacing, and convulsions diminished over time. This recovery study further emphasizes the importance of implementing measures to mitigate cadmium contamination in aquatic environments. It suggests that if prompt action is taken to remove the source of contamination and provide a clean environment, the affected fish species can recover from the toxic effects of cadmium.

Overall, the findings of the recovery study highlight the resilience of *Labeo rohita* and support the need for proactive interventions to minimize the adverse impacts of cadmium on aquatic ecosystems.

1. **SUMMARY AND CONCLUSION**

In this study, we examined the acute toxicity of cadmium on *Labeo rohita*, a freshwater fish species. The toxicity tests were conducted using a static method over durations of 24, 48, 72, and 96 hours. The LC50 values obtained were 0.309 ppm, 0.247 ppm, 0.207 ppm, and 0.160 ppm, respectively, indicating the escalating menace that cadmium poses to aquatic ecosystems over time.

The fish exposed to cadmium displayed dose-dependent behavioral anomalies, including erratic swimming, loss of balance, surfacing, and convulsions. These findings are consistent with previous research, highlighting the detrimental effects of cadmium on fish behavior. The regression analysis and calculation of confidence limits for the LC50 values provide statistical insights into the relationship between cadmium concentration and fish mortality, further strengthening the validity of our findings.

In conclusion, this study demonstrates the acute toxicity of cadmium on *Labeo rohita* and underscores the urgent need for monitoring and regulating cadmium contamination in aquatic environments to preserve the delicate balance of ecosystems.

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