

Assessment of Acute Toxicity (LC₅₀) of the Insecticide Voliam Targo in the Freshwater Fish *Channa gachua*

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Abstract

The increasing use of pesticides in modern agriculture poses a significant threat to aquatic biodiversity. The present study evaluates the acute and sublethal toxicity of the insecticide Voliam Targo a combination of chlorantraniliprole and abamectin in the freshwater fish *Channa gachua*, a species of ecological and nutritional importance. The LC₅₀ values decreased progressively with increasing exposure duration, indicating enhanced toxicity over time: **0.51 ppm at 24 hours, 0.38 ppm at 48 hours, 0.27 ppm at 72 hours, and 0.10 ppm at 96 hours**. Sublethal toxicity was assessed at one-tenth of the 96-hour LC₅₀ (0.01 ml/L) over a 28-day exposure period. These findings demonstrate that Voliam Targo exerts both lethal and sublethal effects on *Channa gachua*, emphasizing the ecological risks associated with its agricultural runoff and the necessity of implementing stricter pesticide regulation near aquatic systems.

Keywords: LC₅₀, Voliam Targo, *Channa gachua*, acute toxicity, sublethal exposure.

Introduction

Biodiversity, encompassing the diverse forms of life on Earth, is fundamental for maintaining ecosystem services that support human health, food security, and economic stability. However, the rapid decline of biodiversity across terrestrial, marine, and freshwater ecosystems has emerged as a global concern, threatening both ecological balance and sustainable development. Freshwater habitats, although accounting for only about 0.01% of the planet's total water and covering 0.8% of the Earth's surface, sustain nearly 6% of all known species, representing over 100,000 forms of life (Dudgeon et al., 2006). This immense biodiversity of inland waters provides economic, cultural, aesthetic, scientific, and educational value (Dudgeon 2019).

Water is indispensable for life and ecosystem integrity, forming the foundation for sustainable and inclusive growth. However, freshwater quality and availability are under increasing stress due to climate change, biodiversity loss, and widespread pollution. These pressures undermine ecosystem functions and pose significant risks to human health, livelihoods, and economic productivity (OECD,

2012). The need for clean and safe water is therefore crucial for public health and environmental sustainability (APHA, 2017).

Water pollution originates from multiple anthropogenic sources, with industrial discharges, domestic sewage, agricultural runoff, and excessive use of agrochemicals being major contributors. Among these, pesticides and herbicides play a dominant role in contaminating aquatic ecosystems, adversely affecting aquatic flora and fauna (Shefali et al., 2021, Chaudhry and Malik, 2017).

India, being a predominantly agrarian nation, has nearly 70% of its population engaged in agriculture. Although the average pesticide use is relatively low (0.31 kg/ha in 2017) compared to countries like China and Japan, the environmental residues remain a major concern. This arises mainly from unregulated and indiscriminate pesticide use, inadequate awareness of safe application practices, weak enforcement of regulatory frameworks, and increasing pest resistance (Nayak Pragati and Solanki Hitesh, 2021). Insecticides constitute the largest share of total pesticide usage in India, significantly contributing to environmental degradation and threatening freshwater ecosystems (OECD, 2021).

High levels of agricultural pollutants in aquatic systems also pose health risks to humans when these contaminants enter sources of drinking and bathing water. Pesticides, herbicides, and fungicides infiltrate aquatic environments through multiple routes such as leaching, runoff, subsurface drainage, and aerial drift and are subsequently absorbed by aquatic organisms. Fish are particularly vulnerable, as they can ingest contaminated food, absorb toxins through the skin, or take up pollutants across gill membranes (Abera et al., 2024, Pimentel et al., 2005).

Fish biodiversity serves as a sensitive indicator of the ecological health of aquatic systems (Okwuosa et al., 2019, Chovanec et al., 2003). Pollutants such as xenobiotics exert both immunotoxic and physiological effects, contributing to diseases and population declines (Wester, Vethaak, and van Muiswinkel, 1994). Acute exposure to toxicants often results in mortality, whereas sublethal exposure can alter fish behavior, metabolism, hematology, enzyme activity, reproduction, cause oxidative stress and overall health (Anil Kumar, 2025) Moezzi et al., 2025, Sagar et al., 2024, Camargo and Martinez, 2007).

Acute toxicity refers to the harmful effects that occur after a single or short-term exposure, typically up to 96 hours in aquatic organisms, and is expressed as LC₅₀ the concentration that causes 50% mortality among test organisms. In contrast, sublethal toxicity involves non-fatal physiological or biochemical disruptions resulting from prolonged exposure to lower concentrations of toxicants (OECD, 2019).

Voliam Targo® is a suspension concentrate formulation containing Abamectin (1.70% w/w) and Chlorantraniliprole (4.30% w/w), designed to control a broad spectrum of agricultural pests (**Syngenta, 2025**). Chlorantraniliprole acts by binding to ryanodine receptors, leading to uncontrolled calcium release, paralysis, and death of insects, while Abamectin interferes with nerve transmission and inhibits feeding activity.

Despite the extensive use of Voliam Targo, a combination formulation of chlorantraniliprole and abamectin, there is a marked lack of acute toxicity data on non-target freshwater fishes, particularly on locally important species such as *Channa gachua*. This gap limits accurate ecological risk assessment in agricultural regions where pesticide runoff frequently contaminates freshwater bodies. Therefore, the present study aims to determine the 24, 48, 72, and 96-hour LC₅₀ values of Voliam Targo in *C. gachua*, providing essential baseline data for environmental monitoring and pesticide regulation.

Material and Method

Test Pesticide-

The pesticide utilized in this study was Voliam Targo, produced by Syngenta, which comprises Abamectin (1.70% w/w) and Chlorantraniliprole (4.30% w/w) as its active components. This test chemical was of analytical grade and was handled in accordance with the guidelines set forth by the OECD and APHA to ensure the reliability of the experimental results.

Collection of Test Fish –

Adult specimens of the freshwater fish *Channa gachua*, from local fishermen in villages along the Girna River in Malegaon Tehsil, Maharashtra, India. Collection sites included Patane Shivar, Aghar, Dabhadi, and Rokdoba Wasti. This species was chosen for its significant commercial value, ease of access, abundance in the region, and adaptability to laboratory environments.

Acclimatization Procedure

Upon arrival at the laboratory, the fish underwent a treatment with a 0.5% potassium permanganate (KMnO₄) solution for two minutes to eliminate any potential dermal infections, following the protocol established by **Pandey et al. (2003)**. The specimens were then acclimatized for 20 days for maintained to minimize handling stress and ensure physiological stabilization of the fish before experimentation in aerated glass aquaria under a natural light cycle (12 hours light and 12 hours dark), with continuous monitoring of temperature, pH, and dissolved oxygen levels, in line with OECD recommendations. Daily feeding consisted of earthworms and locally sourced fish feed, with any uneaten food removed after 30 minutes to maintain water quality. Water was refreshed every 24 hours with dechlorinated freshwater, and any diseased or deceased individuals were promptly removed to

prevent water quality deterioration, as advised by **Schreck and Brouha (1975)**. Feeding was halted 24 hours before the commencement of exposure experiments.

Water Parameter with quality analysis

Water quality assessment forms a fundamental basis for interpreting ecotoxicological data, as the physicochemical characteristics of the aquatic environment strongly influence the bioavailability and toxicity of pollutants to aquatic organisms. Critical parameters such as temperature, pH, dissolved oxygen (DO), total dissolved solids (TDS), alkalinity, hardness, and conductivity play vital roles in regulating fish physiology and determining the degradation rates of pesticides. To ensure the comparability and reliability of ecotoxicological data across studies, adherence to standardized monitoring protocols is essential. The Indian Council of Agricultural Research (ICAR 2022) guidelines recommend maintaining optimal water quality conditions specific to the test species, thereby minimizing confounding environmental stressors. Continuous monitoring throughout the exposure period is vital for ensuring data consistency and experimental validity.

The analytical procedures outlined by the American Public Health Association (APHA, 2017) remain the benchmark for the accurate measurement of physicochemical parameters, ensuring reproducibility in aquatic toxicology experiments. Similarly, the OECD (2019) guidelines emphasize maintaining stable water quality conditions during static-renewal or flow-through bioassays to prevent variability in exposure concentrations. The United States Environmental Protection Agency (USEPA, 2002) also underscores the necessity of strict compliance with water quality standards for credible toxicity testing, particularly when dealing with sensitive aquatic species. Wetzel (2001) highlighted the ecological significance of water chemistry, noting that variations in DO, pH, and ionic composition can significantly alter metabolic and behavioral responses in fish. Likewise, the American Water Works Association (AWWA) and Water Environment Federation (WEF, 2017) standards stress the importance of accurate water analysis in both environmental monitoring and laboratory testing, as deviations from optimal conditions may obscure the true toxicological responses of test organisms.

In this study, water quality was meticulously maintained within the recommended parameters, ensuring that the biological responses observed were directly linked to the toxicant rather than influenced by environmental variability.

Water Parameter	Measured Value	Unit
pH	7.4	–
Temperature	26.6	°C
Dissolved Oxygen	7.8	mg/L

Free Carbon Dioxide	0.62	mg/L
Hardness	150	mg/L as CaCO ₃
Conductivity	350	μS/cm
Total Dissolved Solids (TDS)	280	mg/L
Salinity	0.1	Ppt
Total alkalinity	88.76	mg/L
Turbidity	3.5	NTU

The recommended range values have been compiled from various authoritative sources, including guidelines from ICAR, APHA (2017), OECD (2019), USEPA (2002), Wetzel (2001), and AWWA/WEF (1998).

(**Abbreviations:** °C = degrees Celsius; mg/L = milligrams per liter; mg/L as CaCO₃ = milligrams per liter as calcium carbonate; μS/cm = microsiemens per centimeter; ppt = parts per thousand; NTU = nephelometric turbidity units.)

Experiment Design-

The acute toxicity study was performed following the **OECD Guidelines for the Testing of Chemicals**, specifically **Test No. 203: Fish, Acute Toxicity Test** (OECD, 2019). Acute toxicity (LC₅₀) values were determined at 24, 48, 72, and 96 hours using probit analysis as described by **Finney (1971)**. Compliance with these internationally recognized standards ensured the reliability, reproducibility, and scientific validity of the results.

The experimental fish were divided into **two groups**:

1. **Control group** – maintained in dechlorinated water without exposure to the test chemical.
2. **Treatment group** – The treatment group was exposed to the LC₅₀ concentrations of Voliam Targo for 24, 48, 72, and 96 hours, as determined from the preliminary range-finding trials.

Each group consisted of **ten healthy fish**. To maintain a constant exposure level, the test medium was **renewed every 24 hours** (semi-static system). Reish, D. J. and Oshida, P. S. (1987), Sprague, J. B. (1970).

Mortality was carefully recorded at 24, 48, 72, and 96 hours. Any moribund or dead fish were promptly removed to prevent deterioration of water quality. Throughout the exposure period, water-quality parameters including temperature, pH, dissolved oxygen (DO), and total hardness were measured daily using the standard procedures described by the American Public Health Association (APHA, 2017). Such control of environmental variables ensured that mortality responses reflected true toxicant

effects rather than external stressors. All parameters were maintained within the acceptable limits recommended by OECD to ensure uniform test conditions and data reliability.

Toxicity Assessment

The freshwater snakehead fish, *Channa gachua*, with an average total length of 15.2 ± 0.78 cm and body weight ranging from 53.2 ± 1.06 g, was used as the test organism for the acute toxicity bioassay.

Acute toxicity tests were conducted in transparent glass aquaria containing 10 liters of dechlorinated tap water, maintained under controlled laboratory conditions. Test concentrations were prepared from the commercial formulation Voliam Targo®, which contains Chlorantraniliprole (4.30% w/w) and Abamectin (1.70% w/w) as active ingredients. The nominal concentrations selected for the acute toxicity exposure were 0.05, 0.10, 0.20, 0.30, 0.40, 0.50, 0.60, and 0.70 ppm, based on range-finding trials. The percentage mortality of *Channa gachua* exposed to different concentrations of Voliam Targo over 24, 48, 72, and 96 hours was recorded and is presented in Tables 1–4. A clear dose-dependent increase in mortality was observed, demonstrating a direct correlation between pesticide concentration and mortality rate. The control group exhibited no mortality, and all control fish remained active and healthy throughout the exposure period, confirming the absence of extraneous stressors.

Throughout the exposure period, fish were not provided with feed, in accordance with the recommendations of Ward and Parrish (1982) and Reish and Oshida (1987), to prevent metabolic interference and maintain stable water quality. Mortality data (%) obtained at different exposure durations were used for Probit analysis in order to determine LC₅₀ values with 95% confidence intervals.

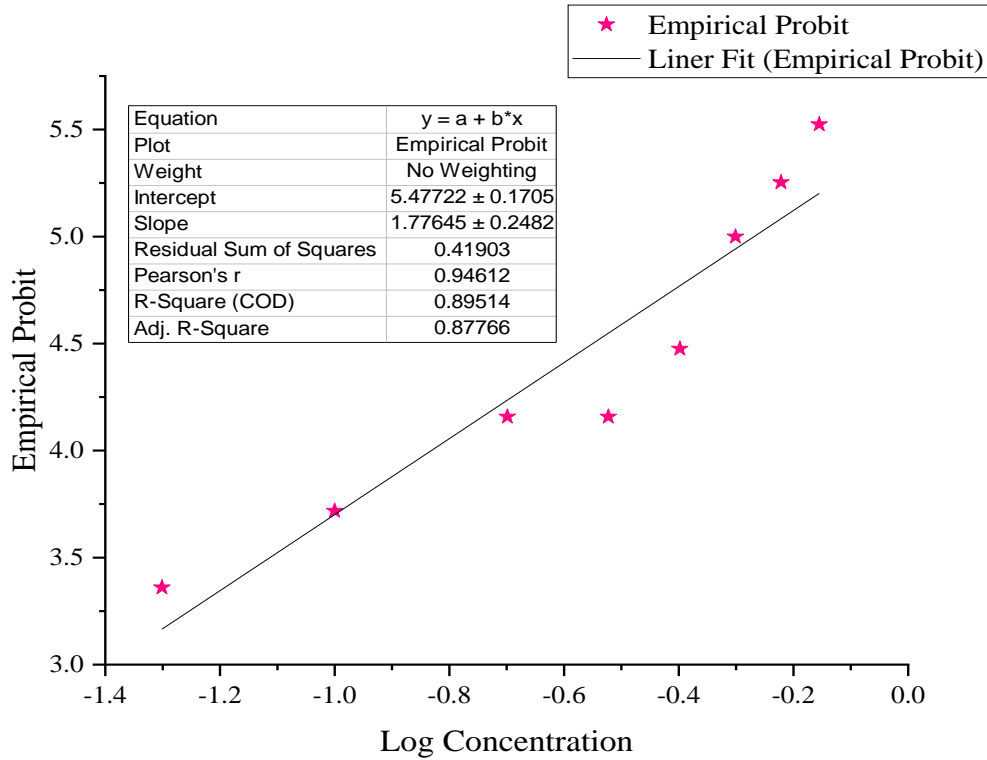
Result

Table 1. Mortality of *Channa gachua* exposed to different concentrations of Voliam Targo at 24 hrs.

Concentration / dose (PPM)	Total No. Exposed	No. Killed	Percent Mortality	Corr Mortality	Log dose/concen	Emp Probit
0.05	10	0	0.00	0.0	-1.30	3.36
0.10	10	1	10.00	10.0	-1.00	3.72
0.20	10	2	20.00	20.0	-0.70	4.16
0.30	10	2	20.00	20.0	-0.52	4.16
0.40	10	3	30.00	30.0	-0.40	4.48
0.50	10	5	50.00	50.0	-0.30	5.00
0.60	10	6	60.00	60.0	-0.22	5.25
0.70	10	7	70.00	70.0	-0.15	5.52

slope(b)	1.776453503
intercept(a)	5.477218898

Exposure period	Regression equation: $y' = (\bar{y} - b \bar{x}) + bx$	LC 50 value in ppm	Variance	Chi square	F. L up to 95% confidence	
					Lower Limit (M1)	Upper Limit (M2)
24 hrs	4.629179061	0.51	0.006392811	1.662729774	0.354543812	0.667967627



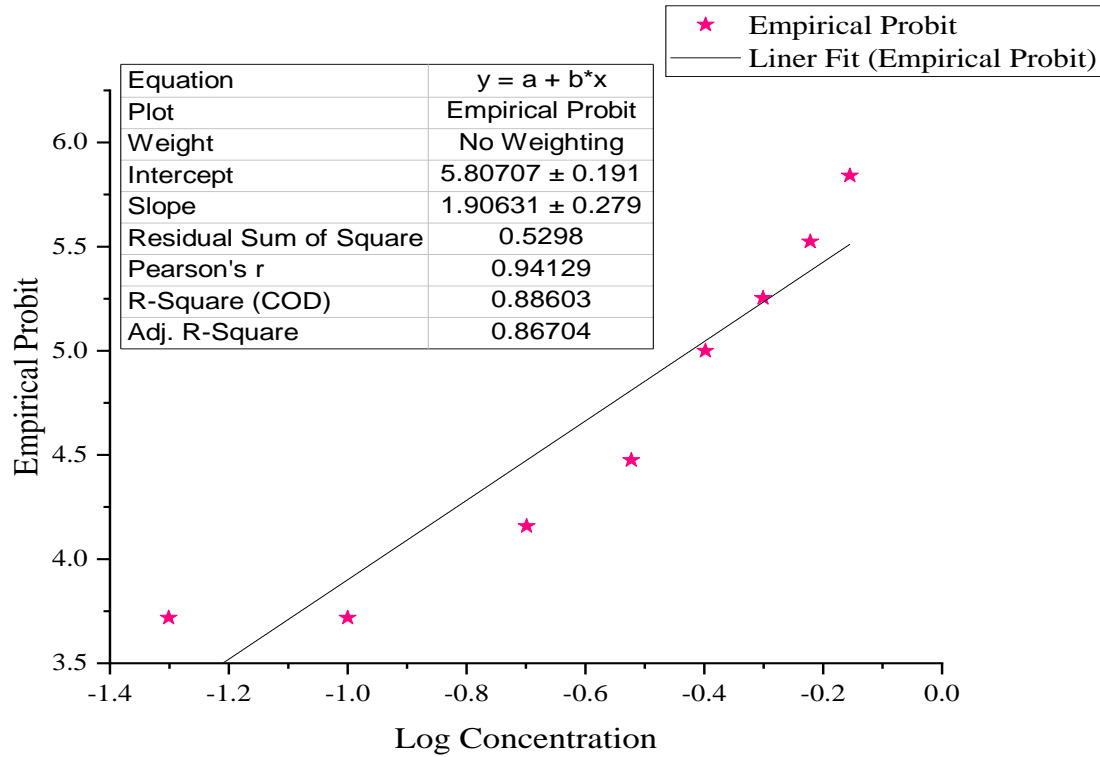
Graph No. 1- Relationship Between Probit Mortality and Log Concentration of the Pesticide Voliam Targo in the Freshwater Fish *Channa gachua* After 24 hrs. Exposure

Table 2. Mortality of *Channa gachua* exposed to different concentrations of Voliam Targo at 48 hrs.

Concentration / dose (PPM)	Total No. Exposed Fish	No. Killed Fish	Percent Mortality	Correct Mortality	Log dose/concentration (x)	Emp Probit
0.05	10	1	10.00	10.0	-1.30	3.72
0.10	10	1	10.00	10.0	-1.00	3.72
0.20	10	2	20.00	20.0	-0.70	4.16
0.30	10	3	30.00	30.0	-0.52	4.48
0.40	10	5	50.00	50.0	-0.40	5.00
0.50	10	6	60.00	60.0	-0.30	5.25
0.60	10	7	70.00	70.0	-0.22	5.52
0.70	10	8	80.00	80.0	-0.15	5.84

slope(b)	1.906305133
intercept(a)	5.807072216

Exposure period	Regression equation: $y' = (\bar{y} - b \bar{x}) + bx$	LC 50 value in ppm	Variance	Chi square	F. L up to 95% confidence	
					Lower Limit (M1)	Upper Limit (M2)
48 hrs	4.866764029	0.38	0.0058169	2.4512144	0.2312754	0.5302492



Graph No. 2- Relationship Between Probit Mortality and Log Concentration of the Pesticide Voliam Targo in the Freshwater Fish *Channa gachua* After 48 hrs. Exposure

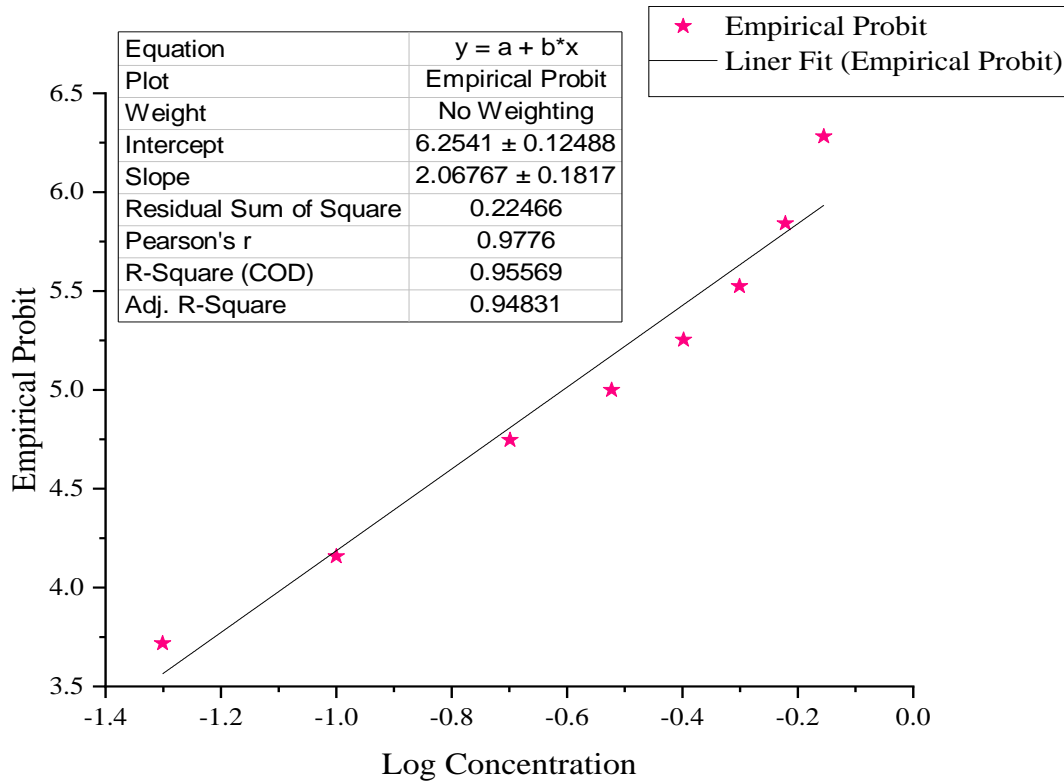
Table 3. Mortality of *Channa gachua* exposed to different concentrations of Voliam Targo at 72 hrs.

Concentration / dose (PPM)	Total No. Exposed Fish	No. Killed Fish	Percent Mortality	Corr Mortality	Log dose/concen (x)	Emp Probit
0.05	10	1	10.00	10.0	-1.30	3.72
0.10	10	2	20.00	20.0	-1.00	4.16
0.20	10	4	40.00	40.0	-0.70	4.75
0.30	10	5	50.00	50.0	-0.52	5.00
0.40	10	6	60.00	60.0	-0.40	5.25
0.50	10	7	70.00	70.0	-0.30	5.52
0.60	10	8	80.00	80.0	-0.22	5.84
0.70	10	10	100.00	90.0	-0.15	6.28

slope(b)	2.067669492
intercept(a)	6.25409804

Exposure period	Regression equation: $y' = (\bar{y} - b \bar{x}) + bx$	LC 50 value in ppm	Variance	Chi square	F. L up to 95% confidence	
					Lower Limit (M1)	Upper Limit (M2)
48 hrs	4.866764029	0.38	0.0058169	2.4512144	0.2312754	0.5302492

72 hrs	5.135489791	0.26	0.00585132	0.91164057	0.10632259	0.40617882
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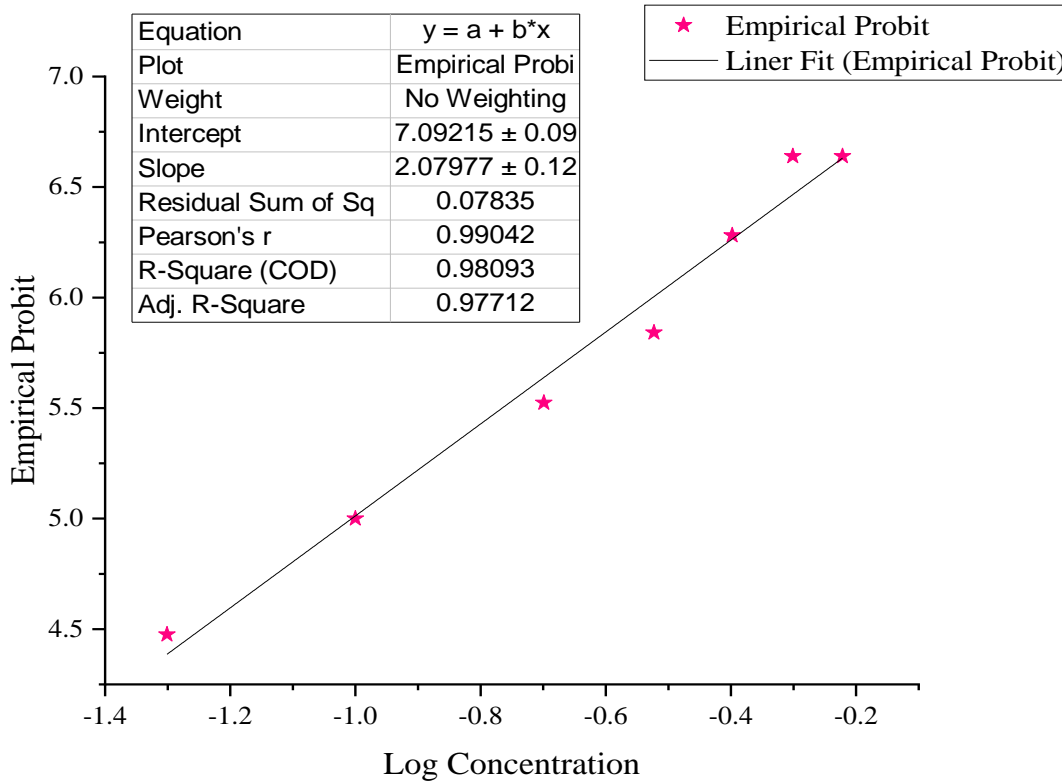
Graph No. 3- Relationship Between Probit Mortality and Log Concentration of the Pesticide Voliam Targo in the Freshwater Fish *Channa gachua* After 72 hrs. Exposure

Table 4. Mortality of *Channa gachua* exposed to different concentrations of Voliam Targo at 96 hrs.

Concentration / dose (PPM)	Total No. Exposed Fish	No. Killed Fish	Percent Mortality	Corr Mortality	Log dose/concen (x)	Emp Probit
0.05	10	3	30.00	30.0	-1.30	4.48
0.10	10	5	50.00	50.0	-1.00	5.00
0.20	10	7	70.00	70.0	-0.70	5.52
0.30	10	8	80.00	80.0	-0.52	5.84
0.40	10	9	90.00	90.0	-0.40	6.28
0.50	10	10	100.00	100.0	-0.30	6.64
0.60	10	10	100.00	100.0	-0.22	6.64

slope(b)	2.079770955
intercept(a)	7.092149399

Exposure period	Regression equation: $y' = (\bar{y} - b \bar{x}) + bx$	LC value in ppm	50	Variance	Chi square	F. L up to 95% confidence	
						Lower Limit (M1)	Upper Limit (M2)



Graph No. 4- Relationship Between Probit Mortality and Log Concentration of the Pesticide Voliam Targo in the Freshwater Fish *Channa gachua* After 96 hrs. Exposure

The assessment of Voliam Targo’s toxicity on *Channa gachua* involved exposing the fish to a series of concentrations ranging from 0.05 to 0.70 ppm for exposure durations of 24, 48, 72, and 96 hours. The results demonstrated a distinct increase in mortality rates corresponding to both the concentration of the pesticide and the duration of exposure. After 24 hours, mortality rates ranged from 10% at the lowest concentration (LC₅₀) to 70% at 0.70 ppm. The calculated median lethal concentration (LC₅₀) was 0.51 ppm with a 95% confidence interval (CI) of 0.35–0.66 ppm, indicating a moderate level of acute toxicity. The probit regression analysis yielded a slope of 1.77 and an intercept of 5.47, with variance and chi-square (χ^2) values of 0.0063 and 1.66, respectively, confirming the robustness of the model used. At 48 hours, mortality increased further, ranging from 10% at both 0.05 and 0.10 ppm to 80% at 0.70 ppm. The LC₅₀ value decreased to 0.38 ppm (95% CI: 0.23–0.53 ppm),

suggesting enhanced toxicity with prolonged exposure. The corresponding regression parameters included a slope of 1.90 and an intercept of 5.80, with variance = 0.0058 and chi-square (x^2) = 2.45.

By the 72-hour exposure period, mortality intensified markedly, ranging from 10% at 0.05 ppm to 100% at 0.70 ppm. The LC_{50} further decreased to 0.26 ppm (95% CI: 0.10–0.40 ppm). The regression model parameters were slope = 2.06 and intercept = 6.25, with variance = 0.0050 and chi-square (x^2) = 0.91, validating the statistical fit of the probit curve.

Finally, after 96 hours of exposure, mortality increased significantly, even at the lowest concentration (0.05 ppm), where 30% of the fish were affected. Complete mortality (100%) occurred at 0.60 ppm. The lowest LC_{50} value was recorded at 0.10 ppm, with a 95% CI ranging from –0.10 to 0.29 ppm, highlighting the pronounced cumulative toxic effects over time. The regression parameters included a slope of 2.07, intercept of 7.09, variance = 0.01024573, and chi-square (x^2) = 1.26

Collectively, these findings clearly demonstrate the dose-dependent and time-dependent toxicity of Voliam Targo in *Channa gachua*. The progressive decline in LC_{50} values with increasing exposure duration underscores the compounding impact of prolonged chemical exposure and emphasizes the ecological risk associated with persistent pesticide contamination in aquatic environments.

Graphical Representation and Interpretation

The log concentration–probit mortality relationship for *Channa gachua* exposed to Voliam Targo at 24, 48, 72, and 96 hours revealed a distinct linear trend (Graphs 1–4). Each regression line exhibited a positive correlation between pesticide concentration and mortality, confirming conformity of the data to Finney's (1971) probit model. The slope values ranged from 1.77 to 2.07, indicating a moderately steep concentration–response curve across all exposure periods. A gradual increase in slope from 24 to 96 hours suggests that the mortality rate accelerated with time and that the fish population became increasingly sensitive to the toxicant due to cumulative physiological stress and bioaccumulation effects. The coefficients of determination (R^2) were exceptionally high, ranging from 0.886 to 0.981, which confirms the statistical reliability and precision of the regression analysis. These high R^2 values indicate that approximately 89 to 98 % of the variation in mortality was explained by the regression model, validating the robustness of the data and the accuracy of the LC_{50} estimations. Overall, the linearity of the regression plots and the strong correlation coefficients demonstrate that mortality in *Channa gachua* followed a predictable, concentration-dependent pattern, emphasizing the time-dependent enhancement of Voliam Targo toxicity which show in Table No.5

Table No.5- Regression equations and coefficient of determination (R²) for *Channa gachua* exposed to Voliam Targo at different exposure periods.”

Exposure period	Regression equation	R ²	Statistical interpretation
24 hrs.	$y = 1.7765x + 5.4772$	0.8951	The regression line shows a good linear correlation between log concentration and probit mortality. The slope indicates a moderate rate of increase in mortality with concentration, while an R ² of 0.8951 confirms that about 89.5 % of the variation in mortality is explained by the model- representing a statistically reliable fit.
48 hrs.	$y = 1.9063x + 5.8071$	0.8860	The regression analysis demonstrates a strong and consistent linear relationship. The slightly higher slope compared to 24 h indicates an enhanced toxic effect with extended exposure. The R ² value (0.8860) suggests that 88.6 % of observed variation is well-explained by the regression, ensuring statistical soundness.
72 hrs.	$y = 2.0677x + 6.2541$	0.9557	The highest slope (2.0677) reflects the steepest concentration–response curve, indicating increased sensitivity of <i>Channa gachua</i> after prolonged exposure. An R ² value of 0.9557 implies that 95.6 % of the variation in mortality is explained by the model, validating excellent linearity and precision of the probit fit.
96 hrs.	$y = 2.0798x + 7.0921$	0.9809	The regression equation shows a nearly perfect linear relationship (R ² = 0.9809), meaning 98 % of the mortality variation is predicted by the model. The slope indicates strong concentration dependency, and the fit confirms exceptional accuracy of the probit analysis at 96 hrs. its representing maximum toxic intensity and model reliability.

Discussion

Overview

The present investigation clearly demonstrated that Voliam Targo a pesticide formulation containing chlorantraniliprole and abamectin exhibited pronounced dose- and time-dependent acute toxicity in *Channa gachua*. The LC₅₀ values progressively declined from 0.51 ppm (95% CI: 0.35–0.66 ppm) at 24 hours to 0.38 ppm (0.23–0.53 ppm) at 48 hours, 0.26 ppm (0.10–0.40 ppm) at 72 hours, and 0.10 ppm (–0.10–0.29 ppm) at 96 hours. This consistent decline indicates cumulative physiological stress and increased susceptibility of fish to Voliam Targo with prolonged exposure. A comparable trend was reported by (Vats et al., 2010) for Cypermethrin (Synthetic Pyrethroids) pesticides, where chronic physiological stress intensified toxicity in freshwater fish over time.

Time and Dose-Dependent Toxicity

The gradual decrease in LC₅₀ values with increasing exposure duration suggests that bioaccumulation of pesticide residues, metabolic exhaustion, and impaired detoxification mechanisms amplify toxicity during prolonged exposure (James P. Meador 2021). Mortality in the present study rose from 0 % at 0.05 ppm to 70 % at 0.70 ppm within 24 hours, reaching 100 % at higher concentrations during extended exposure.

Similar dose–mortality patterns were observed in *Channa punctatus* exposed to organophosphate pesticides (Verma et al., 2020) and in *Labeo rohita* subjected to pyrethroids (Gupta and Roberts, 2019). Similar time- and dose-dependent toxicity trends have been reported on *Tilapia guineensis*; *Clarias gariepinus* (Cat Fish) (Ogeleka et al., 2016; Kingsley et al., 2019). Its toxicity is comparable to or greater than that of other commercial formulations such as Confidor (imidacloprid) and Bavistin (carbendazim), which exhibited LC₅₀ values of 0.9 ppm and 0.10 ppm respectively in *Channa gachua* (Waghmare et al., 2022).

Mechanism of Action and Physiological Implications

Voliam Targo contains two active components with distinct biochemical targets. Chlorantraniliprole acts on ryanodine receptors, triggering uncontrolled calcium release from the sarcoplasmic reticulum of muscle cells, leading to hyper-contraction and paralysis (Cordova et al., 2006). Although these receptors are less abundant in fish, cross-reactivity at elevated concentrations disrupts calcium signaling, impairing neuromuscular coordination and cardiac rhythm.

Abamectin, a macrocyclic lactone, potentiates GABA-mediated chloride influx, causing neuro-paralysis in arthropods (Lasota and Dybas, 1991). In fish, similar interference in neurotransmission

results in erratic swimming, hyperactivity, surface gulping, and loss of equilibrium responses also observed in the present study. These findings concur with neuro-behavioral alterations described by Ram and Mishra (2015) in pesticide-exposed freshwater species. Both compounds possess moderate environmental persistence (USEPA, 2002), which may enhance their bioavailability and cumulative toxic impact under continuous exposure.

Probit Analysis and Model Reliability

In the present investigation, the probit regression slopes ranged between 1.73 and 2.07, while the coefficients of determination (R^2) varied from 0.886 to 0.981, indicating strong linearity between log concentration and probit mortality. These high R^2 values confirm that approximately 89–98 % of mortality variation was explained by the model, validating the precision of LC_{50} estimation. Comparable slope ranges have been reported in acute toxicity assays on teleost fishes (Gupta and Roberts, 2019; Verma et al., 2020).

The marginal variation in slope across time may reflect differences in physiological tolerance among individuals, consistent with Rand and Petrocelli (1985), who stated that slope flattening often signifies increased biological heterogeneity under extended chemical stress.

Comparative Toxicity and Previous Findings

Several earlier investigations have confirmed the sensitivity of air-breathing fish such as *Channa gachua* and *Channa punctatus* to diverse chemical stressors. Koul et al. (2006) reported acute toxicity and biochemical alterations following monocrotophos exposure in *C. gachua*. The LC_{50} values obtained in the present study reveal that Voliam Targo exerts comparable or higher toxicity than most conventional insecticides, particularly during extended exposure, emphasizing its potential ecological hazard to freshwater species.

Behavioural and Physiological Indicators of Stress

Exposure to Voliam Targo induced noticeable behavioral deviations in *Channa gachua*, including erratic swimming activity, increased opercular movement, hyperactivity, and loss of equilibrium clear indicators of neuro-toxic stress. Such responses are consistent with observations of (Ankita V. Bhamare, A. K. Sonawane and Resham Bhalla, 2025) also the insecticide Chlorantraniliprole exhibits marked toxicity to freshwater fish species such as *Channa punctatus*, altering behavior and respiratory functions (Chandra et al., 2023 and Bantu and Vakita, 2013). Similarly, exposure to cadmium chloride and carbaryl has been found to affect fish morphology, behavior, and survival (Singh et al., 2018), while mercury has been shown to exert higher toxicity than cadmium in *C. punctatus* (Sulaiman et al., 2024). These findings highlight the importance of standardized toxicity testing for different chemical agents and species.

Observable behavioral symptoms including erratic movement, hyperactivity, air gulping, and loss of equilibrium are well-established indicators of neurotoxic stress in fish (Ram and Mishra, 2015; Velisek et al., 2006). These responses typically precede mortality and are associated with disruption of ion regulation and neurotransmitter balance. Persistent behavioral impairment can further impact feeding efficiency, predator avoidance, and reproductive success, thereby compounding ecological consequences in natural habitats.

Ecological and Public Health Implications

Channa gachua is an ecologically important predatory freshwater species and also holds nutritional and economic significance for rural and tribal communities across South Asia (Talwar and Jhingran, 1991). The acute toxicity responses observed in the present laboratory study indicate that exposure to the tested pesticide formulation under controlled conditions may pose potential ecological and socio-economic concerns if similar exposures occur in natural aquatic environments. Agricultural runoff carrying pesticide residues has been reported to reach freshwater habitats, where it may adversely affect non-target aquatic organisms, leading to population declines and reduced availability of fish protein for dependent human populations. In addition, the persistence of pesticide residues and their possible bioaccumulation and biomagnification across trophic levels may increase the risk of human exposure through the consumption of contaminated fish, as reported in earlier studies (Nwani et al., 2013). Although sublethal effects were not examined in the present investigation, earlier studies have shown that prolonged exposure to low concentrations of pesticide residues may result in physiological disturbances such as impaired growth, reproductive dysfunction, and suppressed immune responses in fish (Mishra et al., 2018). Such alterations, if sustained, could contribute to long-term ecological imbalance in freshwater ecosystems.

Recommendations for Environmental Safety

The findings of the present study highlight the importance of responsible pesticide use and environmental management practices, particularly in agricultural areas located near freshwater systems. Preventive measures such as the maintenance of buffer zones around water bodies, adoption of Integrated Pest Management (IPM) strategies to reduce chemical dependency, and routine monitoring of pesticide residues in aquatic environments have been widely recommended to minimize ecological risk (APHA, 2017). In addition, farmer awareness and training programs may play a crucial role in promoting judicious pesticide application and reducing unintended impacts on non-target organisms. Given that the present work focused exclusively on acute toxicity, further investigations examining

sublethal hematological, biochemical, and histopathological responses in *Channa gachua* under controlled laboratory conditions would be valuable. Such studies would enhance understanding of potential chronic exposure effects and contribute to the development of environmentally sustainable pest management practices.

Conclusion

This study establishes that the pesticide formulation Voliam Targo, comprising chlorantraniliprole and abamectin, induces pronounced acute toxicity in the freshwater fish *Channa gachua* under controlled laboratory conditions. The consistent decline in LC₅₀ values from 0.51 ppm (24 h) to 0.10 ppm (96 hrs.) clearly demonstrates a time- and concentration-dependent increase in mortality, reflecting progressive physiological stress and disruption of essential metabolic functions during exposure.

The observed sensitivity of *Channa gachua* to Voliam Targo underscores the vulnerability of non-target freshwater organisms to pesticide contamination, particularly under conditions of improper application or agricultural runoff into aquatic systems. While the results are based on standardized laboratory bioassays, they provide valuable toxicological benchmarks for environmental risk assessment and highlight the importance of species-specific responses in pesticide safety evaluation.

These findings emphasize the necessity of cautious pesticide usage and strict adherence to recommended application guidelines, especially in agro-ecosystems adjacent to freshwater habitats. The implementation of Integrated Pest Management (IPM), maintenance of buffer zones, and routine monitoring of pesticide residues in aquatic environments are essential measures to mitigate unintended ecological impacts.

Further research addressing chronic, sublethal, and tissue-level effects under environmentally relevant exposure conditions is warranted to strengthen the ecological risk profile of Voliam Targo. Overall, the present study reinforces the need to balance agricultural pest control with long-term conservation of freshwater biodiversity through responsible pesticide management and sustained environmental surveillance.

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