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Studies on the *Pistia stratiotes* Reduction of Acute Toxicity of Malathion on Growth and Protein Content in Fresh Water Fish *Catla catla* Fingerlings

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Abstract: The primary objective of this research was to examine the effect of malathion exposure on growth and protein content in *Catla catla* fingerlings (3.87±0.10 g). Malathion's 96 h LC_{50} value for fingerlings of *C. catla* was 10.0 µl/l. There was no mortality below the concentration of 9.5 µl/l. During 24, 48, 72, and 96 h, the growth and biochemical composition of fish of control group were significantly greater than those of the malathion-treated groups. The conclusion is that both low and high amounts of malathion are very harmful and affects vital organs of fish. Experiment was performed to examine the growth and protein content of fingerlings of *Catla catla* exposed to malathion for eighty days. During this experiment, the fish were fed Water Lettuce, *Pistia stratiotes* young leaf meal at various concentrations as a control diet (0% *P. stratiotes*) and experimental diets containing 10%, 20%, 30%, 40% and 50% of *P. stratiotes* in place of fish meal as a protein source and designated as P0, P10, P20, P30, P40, and P50, respectively. There was a statistically significant difference ($P < 0.01$) between growth and protein content. The findings also shown that malathion-exposed fingerlings of *C. catla* fed meals containing up to 30% *P. stratiotes* meal performed the best among experimental diets. Although fish meal is irreplaceable, aquatic weed meal may be added up to the optimal quantity to make cost-effective feed for *C. catla.*

Keywords: Malathion, *Catla catla*, *Pistia stratiotes*, Development, Protein, LC_{50}

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Introduction

Any undesired alteration in the natural quality of the environment caused by physical, chemical, or biological forces is referred to as pollution. In aquatic ecosystems, water pollution is often generated by a variety of human sources, industrial facilities, and agrochemicals. It has serious environmental issues now. Water quality is one of the most critical factors in aquaculture. However, these particles and industrial discharges may be successfully transported by air, wind, rivers, and floods into water reservoirs, altering their physiochemical characteristics and

increasing their toxicity. It causes harm to aquatic organisms, particularly fish. Many toxicants, such as herbicides, pesticides, heavy metals, and polychlorinated biphenyls, are actively ingested and accumulated by many kinds of fish in water bodies. The accumulation of some pesticides causes growth, biochemical, physiological, and morphological alterations in freshwater animals. (Ramamurthy *et al*., 1987).

The presence of harmful substances in aquatic ecosystems and organisms is reduced by the use of approved chemical compounds. The chemical compound reduces harmful metabolites in ponds located in aquatic environments (Chien, 1992; Briggs and Smith, 1996, Bharathi *et al*., 2019). Substituting fish meal with a diverse blend of plant protein may limit the exposure of fish to anti-nutritional factors and enhance growth performance (Borgeson, 2006) In contrast, green plants have been recognised for a long time as the cheapest and most plentiful possible source of protein due to their capacity to manufacture amino acids from a broad variety of almost endless and easily accessible raw materials (Fasuyi and Aletor, 2005). The current study was conducted to determine the effects of *Pistia stratiotes* leaf meal on the reduction of malathion toxicity in fish and the enhancement of development and protein content in the freshwater fish *C. catla* fingerlings.

Materials and Methods

Experimental diet:

For the experimental supplemental feed, young *P. stratiotes* leaves and selected components were applied. Fresh colonies of *P. stratiotes* were obtained from a pond in the district of Tuticorin, Tamil Nadu, India, and cleaned properly to eliminate dirt. Then, they were dried at room temperature for one week. After that, the samples were ground into powder. Six dry diets designated as P0, P10, P20, P30, P40, and P50, were produced in which fish meal was substituted with *P. stratiotes* at 0%, 10%, 20%, 30%, 40%, and 50%, respectively.

Feed processing:

In accordance with Hardy (1980), a 35% protein diet was created for experimental purposes. Diets for testing were composed of fishmeal, crushed oil cake, rice bran, cod liver oil, vitamins and minerals. The powdered and dry components were first combined to form a homogeneous mixture. The mixture was then combined with the appropriate amount of dried *P. stratiotes* leaf meal (0%, 10%, 20%, 30%, 40%, and 50%) along with an aliquot of heated water and pressure-cooked for 15 to 20 min. After that, mild cooking pellets (2 mm) were made using a manually powered pelletizer and dried in the sun. After dehydration, feeds were kept separately for experimental usage.

Experimental animal:

C. catla fingerlings were chosen for the current study. It was chosen because to its strong growth potential, great customer preference, and widespread availability. It is the most significant freshwater species cultivated in India and neighbouring countries. Given its significance in the cultural system, India has also prioritised its genetic enhancement via selective breeding. The experimental animal *C. catla* was obtained from the Sabari fish farm in Tirunelveli, Tamil Nadu, India. Then they were brought straight to laboratory conditions. A month was spent acclimating these fish to the laboratory environment. During acclimation, the animals were given pellets containing 20% protein.

Experimental Design:

For the experiment fingerlings of *C. catla* (3.87± 0.10 g) were selected from the acclimation tank and fasted for 24 h prior to the start of the study. The fish were examined for pathogenic infections. The fish were separated into seven groups of twenty individuals each, and the experiment lasted eighty days. Group 1 was the control and was raised in fresh water and given a control diet. The second through seventh groups of test animals were subjected to 0.9 µl/l (sublethal dose of malathion). The second malathion-exposed

group was fed a control diet, while the third, fourth, fifth, sixth, and seventh groups were given 10, 20, 30, 40, and 50 per cent *P. stratiotes* leaf diets, respectively. For simplicity, the trial groups 1, 2, 3, 4, 5, 6, and 7 will be denoted as C, C+M, M+P1, M+P2, M+P3, M+P4, and M+P5, correspondingly. Each group was represented by triplicates. All tests were carried out in epoxy-coated cement tanks (0.6 × 0.6 m; 110 L capacity) holding 100 L of water. The test media were altered in two ways once. The hydro-biological parameters were temperature 29.4±0.5℃, dissolved oxygen 4.28 ±0.15 ml O_2l^{-1}, pH 7.8±0.3, salinity 0.30±0.07 ppt, and water hardness 73±3.30 mg $CaCO_3$ l^{-1}.

During the experimental period, test animals were given ad libitum with experimental meals twice a day for 1 h at 07.00 and 18.00. Unconsumed feed was eliminated after one h of feeding. Every 20 days, feeding, growth, and protein contents were assessed.

Statistical analysis:

All data were reported as the mean± standard deviation. All group data were subjected to an analysis of variance (ANOVA) using Microsoft Excel 2007 and a significance threshold of $P<0.01$ was established.

Results

The mean body weight and length of C.catla subjected to malathion and fed *P. stratiotes* increased. The feeding and growth indices of malathion-exposed fish fed *P. stratiotes* were increased (Table 1). On day 80, the weight increased, feed intake, and feeding rate of *C. catla* treated with C+M vs M+P3 were considerably improved (Table 1). In contrast, the conversion rate, cross conversion efficiency, and feed conversion ratio did not vary substantially ($P>0.05$). However, there were statistically significant ($P<0.01$) differences between M+P2 and M+P3 treatments. However, the cross conversion ratio did not vary (Table 1). In comparison to previous *P. stratiotes* diets, *C. catla* fingerlings fed 30% *P. stratiotes* exhibited a higher rate of feed consumption and growth characteristics when exposed to malathion. The two-way ANOVA test revealed that the growth and feed utilisation characteristics of *C. catla* are significantly ($P<0.01$) affected by the concentration of *P. stratiotes*. The tissue protein content improved considerably ($P <0.01$) in fish exposed to malathion and fed *P. stratiotes*. Among the *P. stratiotes* diets, malathion-exposed *C. catla* fingerlings given 30% *P. stratiotes* diet (M+P30) showed a greater increase in tissue protein than any other diet, except the control diet (Table 2). A substantial ($P<0.01$) positive connection was found between the tissue protein concentrations of malathion-exposed fish fed *P.stratiotes* (Fig. 1). It demonstrated that the *P. stratiotes* diet decreased the toxicity of malathion in *C. catla* fish hence, increasing their protein content. In addition, a two-way ANOVA test revealed that protein content was substantially ($P<0.01$) associated with *P. stratiotes* diet levels. The highest amount of protein was found in muscle, followed by liver. In malathion-exposed *C. catla* fed a 30% *P. stratiotes* diet, the muscle and liver protein regression equations were $Y=0.045+23.36$ and $Y=0.032+4.05$, respectively. Compared to control fish, protein content was decreased five-fold in the liver and thrice in the muscle of the studied tissues. A substantial ($P<0.01$) and positive connection was found between protein (of both muscle and liver) and *P. stratiotes* diet concentrations (Fig. 1).

Discussion

In the current investigation, *C. catla* fingerlings were exposed to sublethal levels of malathion and fed various concentrations of *P. stratiotes*. In aquatic environments, pesticides operate as metabolic depressants and exert pressure on physiologically active chemicals. Mathers *et al*. (1985) discovered that when a large proportion of dietary energy was used for metabolism, less energy was available for development in *Micropterus salmodius* that had been exposed to toxicants. Numerous researchers have explored the effects of various pesticides and heavy metals

Table 1: Effect of *Pistia stratiotes* diets on growth and feeding parameters in malathion exposed carp, *Catla catla* as a function of time. Each value is the mean ±SD of three estimations. Rates are exposed as mg g^{-1} live fish day^{-1} and efficiencies as %.

Exposure period(days)	Control (C)	C + M	M + P1	M + P2	M + P3	M + P4	M + P5
Whole Fish Weight (g wet weight)							
20	58.45±0.04	57.45±0.05	58.56±0.04	58.45±0.08	58.45±0.08	58.48±0.09	57.44±0.04
40	61.34±0.03	50.82 ±0.52	51.78±0.49	53.95±0.10	54.58±0.37	51.89±0.56	50.04±0.07
60	65.55±0.32	39.47±0.58	54.93±0.13	57.92±0.28	61.02±0.18	54.91±0.25	52.11±0.11
80	72.19±0.15	37.20±0.19	58.83±0.16	63.30±0.40	67.53±0.45	58.64±0.20	54.39±0.26
Weight Gain (g wet weight)							
20	2.89±0.03	-9.63±0.10	-5.78±0.12	-4.66±0.11	-2.9±0.06	-5.59±0.22	-7.4±0.72
40	5.21±0.13	-9.35±0.21	3.15±0.12	4.97±0.17	6.47±0.31	3.02±0.03	2.07±0.07
60	6.64±0.15	-12.27±0.18	3.90±0.73	5.38±0.40	6.5±0.08	3.73±0.09	2.28±0.05
80	8.10±0.09	-11.74± 0.1	3.94±0.12	7.16±0.03	10.98±0.47	5.80±0.18	3.29±0.27
Feed Intake (g dry matter)							
20	30.66±1.69	19±0.81	24±1.63	26.66±1.25	27.80±0.17	24.66±0.47	21.33±1.25
40	35.33±2.05	15.66±1.25	26.33±2.62	30±2.62	31.33±2.05	27.66±1.69	23±2.44
60	39±0.82	12±3.26	28±4.54	33.66±2.62	37.33±2.05	32.38±2.66	25.94±1.93
80	45.11±1.69	10.65±1.41	31.23±2.78	35.85±2.30	43.32±1.96	36.76±1.31	27.10±1.17
Feeding Rate (mg g-1 live fish day-1)							
20	28.89±0.93	17.79±0.52	20.04±0.66	22.86±0.20	23.84±0.27	22.09±0.34	18.83±0.48
40	30.93±0.40	15.37±0.45	21.90±0.38	25.19±0.74	27.65±0.49	24.22±0.66	20.68±0.64
60	34.93±0.74	10.53±1.17	23.27±1.42	25.48±0.96	32.21±0.64	26.07±1.35	21.54±0.82
80	36.79±1.21	7.69± 0.43	26.08±1.63	29.39±2.28	35.30±0.86	30.23±1.65	24.1±1.63
Conversion Rate (mg g-1 live fish day-1)							
20	2.53±0.08	-8.31±067	-5.17±0.16	-4.19±0.25	-2.53±0.34	-4.66±0.29	-6.46±0.03
40	4.51±0.31	-8.23±0.15	2.69±0.18	4.33±0.22	5.55±0.30	2.67±0.18	1.92±0.08
60	5.61±0.32	-10.6±0.15	3.50±0.13	4.70±0.06	5.61±0.22	4.17±0.05	2.03±0.03
80	7.12±0.66	-10.32± 0.06	3.54±0.09	6.46±0.20	9.59±0.40	5.09±0.04	2.81±0.18
Cross Conversion Efficiency (%)							
20	8.55±0.47	-48.14±0.80	-27.15±2.13	-17.39±0.80	-10.78±1.01	-22.54±1.21	-34.84±1.22
40	14.65±0.50	-54.33±1.69	12.52±0.44	17.53±0.42	20.58±0.49	11.53±0.28	10±0.82
60	17.33±0.26	-101.31±0.88	14.65±0.26	17.53±0.33	18.41±0.25	12.19±0.13	9.23±0.24
80	18.75±0.22	-130.22±0.74	13.47±0.29	21.25±0.22	26.53±0.18	16.67±0.24	11.84±0.08
Feed Conversion Ratio							
20	11.23±0.15	-2.14±0.05	-3.40±0.28	-5.71±0.18	-9.37±0.14	-4.57±0.08	-2.79±0.16
40	6.75±0.16	-1.90±0.08	7.99±0.05	5.82±0.13	9.02±0.09	9.02±0.09	11.52±0.32
60	5.88±0.14	-1.02±0.06	6.90±0.12	5.53±0.30	8.55±0.10	5.66±0.10	10.86±0.22
80	5.40±0.16	-0.86± 0.07	7.65±0.18	4.74±0.10	6.24±0.23	3.88±0.11	8.71±0.19

Student 't'test : 80 days

Parameters	C+M Vs M+P3	M+P2 Vs M+P3
Weight gain	t =7.6; P < 0.01	t = 31.83; P < 0.01
Feed intake	t = 19.33; P < 0.01	t = 3.50; P < 0.01
Conversion rate	t = 41.20; P < 0.01	t = 3.43; P < 0.01
Cross conversion efficiency	t = -195.64; P < 0.01	t = 5.79; P > 0.05
Feed conversion ratio	t = -37.75; P > 0.05	t = 8.6; P < 0.01

Table 2: Effect of *Pistia stratiotes* diets on protein content (mg g^{-1} wet tissue) in malathion exposed carp, *Catla catla* as a function of time. Each value is the mean±SD of three estimations.

Exposure period(days)	Control (C)	C + M	M + P1	M + P2	M + P3	M + P4	M + P5
Muscle							
0	20.45±1.0	20.45±1.0	20.45±1.0	20.45±1.0	20.45±1.0	20.45±1.0	20.45±1.0
20	22.10±1.17	18.78±1.30	29.35±1.17	19.05±0.95	29.98±1.03	20.78±1.07	19.03±1.28
40	26.73±1.25	13.32±2.35	27.39±0.27	22.19±0.78	22.92±1.95	23.15±2.30	20.60±1.36
60	28.79±1.25	10.48±1.38	19.25±1.22	24.65±1.40	25.02±1.10	26.42±139	22.10±1.19
80	35.42±0.95	7.31± 2.34	10.58±0.09	26.01±2.35	27.43±0.09	32.63±1.53	24.02±1.19
Liver							
0	4.93±0.15	4.93±0.15	4.93±0.15	4.93±0.15	4.93±0.15	4.93±0.15	4.93±0.15
20	5.40±0.19	3.03±0.15	3.93±0.27	4.02±0.09	4.14±0.49	4.09±0.20	3.85±0.17
40	6.88±0.25	2.95±0.32	3.99±0.17	4.09±0.31	4.60±0.52	4.28±0.05	3.95±0.18
60	8.16±0.15	2.10±0.51	4.68±0.45	4.86±0.32	5.69±0.25	5.18±0.38	4.18±0.52
80	10.05±0.27	1.85±0.13	6.02±0.52	6.85±0.09	7.42±0.23	6.19±0.28	5.56±0.35

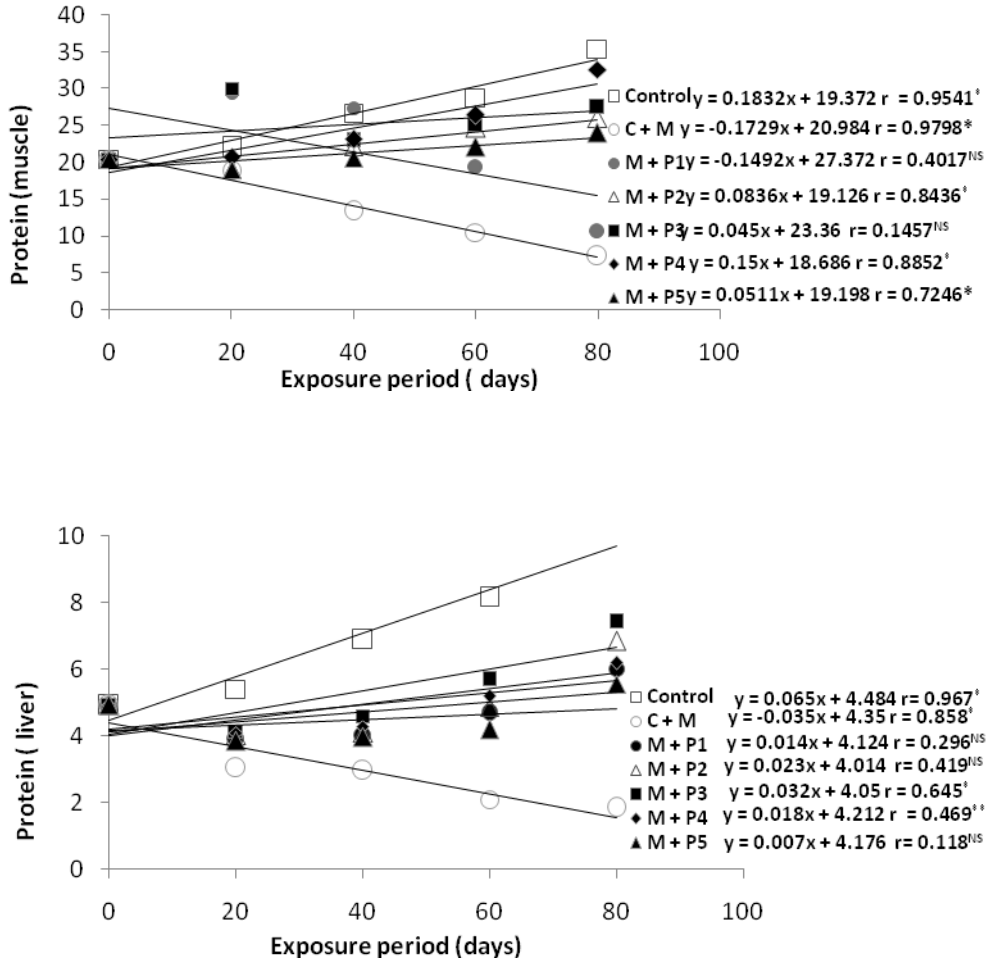

Fig.1: Regression lines for protein (muscle and liver) in malathion exposed *Catla catla* fingerlings fed diets with different levels of *Pistia stratiotes* diets as a function of time.

on the development and biochemical components of a variety of aquatic creatures. James *et al*. (2009) discovered that a 6% Spirulina supplemented meal decreased copper absorption in tissues and increased copper excretion through fish faeces hence, enhancing food use in *Cirrhinus mrigala*. Similarly, Sunisa *et al.* (2012) discovered that a diet containing *Moringa oleifora* decreased the lead poisoning in *Puntius altus*, which improved the deteriorated kidney, liver, and gills of lead-exposed fish.

In outdoor tests, the *P. stratiotes* plant absorbs cadmium and chrome. Specifically, the rise in Cd content in plant tissues occurred in the roots. Cd is absorbed by plant roots quicker than it is transported to the plant's leaves (Maine and Duarte, 2001). Similar to *P. stratiotes*, *Scrirpus tabernaemontam*, and *Colocasia esculenta*, aquatic plants were assessed for their ability to remove mercury from water by Skinner (2007). Biochemical parameters improved in malathion-exposed *Catla catla* given *P.stratiotes*-supplemented diets, according to the current research. Based on the location of action, the biochemical parameters of the metabolic rate either rise or decrease. According to Magar and Afsar Shaikh (2012), proteins are the building blocks of the animal's body and are the most essential and biochemical components to increase blood glucose and energy source during periods of stress. This study found that the addition of *P. stratiotes* to fish diets increased feed utilisation and biochemical markers in fish exposed to malathion.

Conclusion

From the observations of this study, it was possible to infer that aquatic weed (*P. stratiotes*) may be supplemented to *C. catla* up to 30% of the diet, as well as growth and protein alterations in both muscle and liver in response to malathion pesticides. In the current investigation, alterations in the biochemical contents of *C.catla* subjected to varying concentrations of malathion were detected. The protein content of several tissues of fish subjected to malathion was dramatically reduced. Changes in biochemical markers, such as protein levels, are crucial indicators of the sensitivity of organ systems to environmental contaminants, since they affect their function. According to the findings of the current experiment, fish feed made from aquatic vegetation is less expensive than fish meal. Consequently, replacing it with fish meal in fish diets showed tremendous promise and might lower the price of fish feed. Although the results of the experiment indicated that fish meal cannot be completely replaced with plant material, partial replacement with *P. stratiotes* meal is optimal for the development of *C. catla*.

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