Effect of Melia Azedarach Extract on Some Selected Physiological Parameters of (Catla catla)

Introduction

Aquaculture is one of the important sectors contributing significantly in the Indian economy. Fish culturists are encouraged towards intensification of culture system to increase production and profit. In such practice of fish and shrimp farming, disease becomes major threats. Disease is one of the most important constraints of fish production both in culture system, as well as in wild condition [1]. Fish production is decreased due to the occurrence of disease caused by different pathogens in aquaculture. Viral diseases have posed significant problems in aquaculture for many years. In commercial aquaculture, antibiotics were used for prevention and control the diseases, and hormones were used for growth performance but the cost of antibiotics and hormones are expensive. Several studies have been carried out to find the new compounds from plant sources at cheap and best to prevent the disease causing organisms in aquaculture [2]. Aquaculture has been a tradition in several parts of Asia and according to FAO statistics, over 80% of fish produced by aquaculture come from Asia, where the production was 31.07 million metric tons valued at $ 38.855 billion [3].

Aeromonas hydrophila is a gram negative motile bacterium. The ulcerative disease is mostly caused by gram negative bacterium. A. hydrophila is pathogenic not only to fishes but also to amphibian, reptiles and mammals including man [4]. The Indian major carps, Catla catla, Labeo rohita and Cirrhinus mrigala are the most important commercial fishes in India with a maximum market demand and acceptability as food by the consumers due to their taste and flesh. They contribute about 67% of total freshwater fish production [5]. Melia azedarach is a well-known ethno medicinal tree used in Ayurveda, its use in the traditional folk medicine. Different parts of M. azedarach in traditional system of medicine. Hence, the present study has been carried out to find the new compounds from plant sources at cheap and best to prevent the disease causing organisms in aquaculture [2]. Aquaculture has been a tradition in several parts of Asia and according to FAO statistics, over 80% of fish produced by aquaculture come from Asia, where the production was 31.07 million metric tons valued at $ 38.855 billion [3].

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Materials and Methods

Alive and activity fishes (12± 1g) were collected from High-tech fish farm, Madurai, Tamil Nadu, India. The fishes were maintained in non-chlorinated at 20 day. The ground nut oil cake, fish meal and rice bran, tapioca, soybean, were mixed and sterilized. And then add a multivitamin tablet. The above mixed foods were added with different concentrations (1.0g, 1.5g and 2.0g) of plant extract prepared using shoklet apparatus. These extract Melia azedarach extract used for experimental fishes and without plant extract diet for control fish. The food was made into small pellets. 0.1 ml of 10 CFU/ml of Aeromonas hydrophila was injected intraperitoneally both for control and experimental. In every seven days following physiological studies such as,(Table 1).

Survival and mortality

The survival and mortality rate was calculated by dividing the number of fish died to the total number of fish.

\[
\text{Survival rate} = \frac{\text{Number of fish died}}{\text{Total number of fish}}
\]

\begin{table}[h]
\centering
\begin{tabular}{|c|c|c|c|c|c|c|c|}
\hline
\textbf{Dose (gm)} & \textbf{Days after treatment} & 0 & 7 & 14 & 21 & 28 & 35 & \textbf{Total mortality} \\
\hline
\textbf{Positive control} & 0 & 10 & 0 & 0 & 0 & 0 & 0 & 0 \\
\textbf{Normal fish} & 0 & 10 & 0 & 0 & 0 & 0 & 0 & 0 \\
\hline
\textbf{Negative control} & 1.0 & 10 & 0 & 0 & 0 & 0 & 0 & 0 \\
\textbf{(A. hydrophila)} & 1.5 & 10 & 0 & 0 & 0 & 0 & 0 & 0 \\
\hline
\textbf{Experimental fish} & 1.5 & 10 & 0 & 0 & 0 & 0 & 0 & 0 \\
\textbf{(A. hydrophila)} & + M. azedarach) & 2 & 10 & 0 & 20 & 20 & 20 & 30 \\
\hline
\end{tabular}
\end{table}
Antibody titer

This method is followed by this (Micheal 2002)

Phagocytic activity

The phagocytic activity assay was performed by the following modified method of Sahoo et al.(2000).

\[
\text{Number of WBC} \times \text{Dilution,} \\
\text{Number of WBCs/mm}^3 \quad \text{Area counted } \times \text{Depth of the fluid}
\]

Oxygen consumption in fish

The oxygen consumption of the fingerlings of the control and experimental fish was estimated by Winkler’s method.

Opercular movement

The fish is taken in a beaker containing water. The number of opercular movements for a minute was recorded with the help of a stop watch in the control and the experimental fish. The triplicate observation was recorded from each sample for the control and the experimental fishes.

Growth rate

The growth of the fish is defined as an increase in the body weight of the fish in definite intervals of time. The weight gain of Catla catla was calculated as

\[
\text{Weight gain} \% = \frac{\text{BWf} - \text{BWi}}{\text{BWi}} 
\]

The statistical significance between control and experimental groups were tested by ‘t’ test.

Results and Discussion

In this study the cumulative percentage of Mortality, Antibody titer, Phagocytic activity, oxygen consumption, Opercular movement and growth rate, were studied in disease induced Catla catla using different concentration of melia azedarach formulated died against Aeromonas hydrophila. In control groups showed 60% mortality 1g and 2g died fed groups was 20% and 30% mortality in experimental groups. Similar result were observed by [7], reported that Mikania cordata leaf powder significantly increased non-specific immunity and decreased mortality in C. catla experimentally infected with Aphanomyces invadans. The M. cordata leaf powder supplemented diet showed significantly (p<0.05) high disease resistance against A. invadans infection when compared with control group. The highest

| Dose (g) | Days after administration |
|---------|---------------------------|
| 0       | 0.62 ± 0.63               |
| 0.1     | 0.30 ± 0.46               |
| 0.2     | 0.18 ± 0.17               |
| 0.3     | 0.06 ± 0.46               |
| 0.4     | 0.17 ± 0.18               |
| 0.5     | 0.18 ± 0.17               |
| 0.6     | 0.17 ± 0.18               |
| 0.7     | 0.18 ± 0.17               |
| 0.8     | 0.19 ± 0.18               |
| 0.9     | 0.18 ± 0.17               |
| 1.0     | 0.17 ± 0.18               |
| 1.1     | 0.16 ± 0.17               |
| 1.2     | 0.15 ± 0.16               |
| 1.3     | 0.14 ± 0.15               |
| 1.4     | 0.13 ± 0.14               |
| 1.5     | 0.12 ± 0.13               |
| 1.6     | 0.11 ± 0.12               |
| 1.7     | 0.10 ± 0.11               |
| 1.8     | 0.09 ± 0.10               |
| 1.9     | 0.08 ± 0.09               |
| 2.0     | 0.07 ± 0.08               |

Each value (Mean±SD) represents the average of 3 replicates.* statistically significant, <0.05, ‘t’ test.

Table 2: Antibody titer (log, values) of C. catla fed with different concentrations of M. azedarach intraperitoneally injected with 0.1ml of 10^6 CFU/ml of A. hydrophila.

In the present study, the plant extracts administered experimental groups showed more phagocytic activity when compared with control. During the study period 1.5g (100g) of diet treated fishes showed maximum phagocytic when compared to other concentrations. Similar reports are also observed by [11]. They reported that phagocytic activity was significantly increased in fish fed with 2.0% Azadirachta indica formulated diet compared to the control, but not with 1.0% and 3.0%. They also suggested that 2.0% supplementation diet significantly influence the growth, haematology, and enhances the innate immune system in silver carp. Hypophthalmichthys molitrix against A. hydrophila [12], reported that Basella alba extract treated tilapia showed significantly higher (P<0.05) phagocytic activity compared to fish fed control diet.

The Oxygen consumption higher in 1.5g plant extract formulated diet treated fishes than control and other experimental groups. Similar reports are also observed by [13], reported that oxygen consumption of fishes in experimental group increased after the treatment of different concentrations (250,500,750mg/kg of food) of extract of Aloe vera were fed to common carp, Cyprinus carpio against Aeromonas hydrophila [14], observed that experimental group of 250 mg/kg showed more O2 consumption of fish compared with control. The experimental fish treated with 10 mg of turmeric powder showed increase in the O2 consumption of fish, when compared to 20mg and 30 mg of turmeric powder [15], reported that presence of toxicants in the medium could bind with globin fraction of haemoglobin of the fish and alter the physiological activity of the body. Thus the decrease oxygen consumption may be due to the damage caused to red blood cells. The concentration of red blood corpuscles can be increased to raise the oxygen carrying capacity of the blood per unit volume [16].

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In the present study was opercular movement was varied from both experimental and control fishes. The opercular movement in the control fish showed 60.00±0.57 and the plant extract formulated died found to be maximum number of opercular movement were observed in 1.5g plant extract formulated died found to be 65.00±1.00 in the initial day (0 day) and 86.00±0.57 (35 day) Similar result were observed by [17]. They reported that opercular movement of fishes in experimental group increased after the treatment of different concentrations (250,500,750mg/kg of food) of extract of Aloe vera also reported that opercular ventilation of the concentrations (250,500,750mg/kg of food) of extract of Aloe vera observed by [18], also reported that opercular ventilation of the catfish, (Pangasius hypophthalmus) was increased and treated with increasing concentrations of Azadirachta indica leaf extract [12]. Observed that increase in opercular movement, mucous secretion, erratic movement etc., were noticed in neem leaf extract exposed fish [19] and also noticed similar behavioural changes in Channa punctatus exposed to Nerium indicum leaf extract (Tables 3,4).

Table 3: Phagocytic activity of C. catla feed with different concentrations of M. azedarach intraperitoneally injected with 0.1 ml of 10^5 CFU / ml of A. hydrophila.

| Dose (g) | 0 | 7 | 14 | 21 | 28 | 35 |
|---------|---|---|----|----|----|----|
| Positive control (Normal fish) | 0 | 27.66±0.57 | 29.66±1.52 | 31.66±1.52 | 34.66±1.52 | 36.66±1.52 | 39.66±2.08 |
| Negative control (A. hydrophila) | 0 | 19.66±1.52 | 18.66±1.52 | 17.66±0.56 | 16.00±1.00 | 15.00±1.00 | 14.00±1.00 |
| Experimental fish (A. hydrophila + M. azedarach) | 1.0 | 33.00±2.60 | 36.00±1.00 | 38.00±1.00 | 39.00±1.00 | 42.33±1.52 | 44.33±1.52 |
| | 1.5 | 40.33±0.57 | 42.00±1.00 | 43.66±1.58 | 45.33±1.52 | 47.33±1.52 | 49.33±2.51 |
| | 2.0 | 34.66±0.57 | 35.00±2.00 | 36.00±1.00 | 37.66±1.55 | 39.00±1.00 | 40.00±1.00 |

Each value (Mean±SD) represents the average of 3 replicates.* statistically significant, <0.05, ‘t’ test.

Table 4: Physiological changes of C. catla feed with different concentration of Melia azedarach and intraperitoneally injected with 0.1 ml of 10^5 CFU / ml of Aeromonas hydrophila.

| Dose (g) | Oxygen consumption | Opercular movement | Growth rate |
|----------|-------------------|-------------------|-------------|
|          | 0 | 7 | 14 | 21 | 28 | 35 | 0 | 7 | 14 | 21 | 28 | 35 | 0 | 7 | 14 | 21 | 28 | 35 |
| Normal fish |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |
| Control (A. hydrophila treated fish) |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |
| Exp. fish (A. hydrophila + M. azedarach) |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |   |

Each value (Mean±SD) represents the average of 3 replicates.* statistically significant, <0.05, ‘t’ test.

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