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# Effect of dietary vitamin C on the growth and survival rate of walking catfish *Clarias batrachus* (Linnaeus)

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**Abstract.** The aim of the study was to evaluate the effect of dietary vitamin C in 3 levels ($T_2$-500, $T_3$-1000, $T_4$-1500 mg vit C) per kg feed on the growth and survival rate of walking catfish (*Clarias batrachus*) in aquaria system. *Clarias batrachus* fingerling having an average weight of 6.11± 0.47 g were reared in aquaria for a period of 11 weeks fed on diets containing 35% protein. The highest survival rate was found in $T_4$ and lowest in $T_1$. Best growth performance was found in $T_4$ followed by $T_3$, $T_2$ and $T_1$ (control). Best on the findings of the present study it is recommended to supplement 1500 mg vitamin C/kg feed containing diet for *C. batrachus*.
**Key words:** Walking catfish, vitamin C, growth, survival rate, *C. batrachus*

## Introduction

In freshwater aquaculture, major culturable fish species of Bangladesh are Indian major carps, Chinese carps, common carps and tilapia. Now-a-days attention has been given to the culture of air breathing like shing (*Heteropneustes fossilis*), walking catfish (*Clarias batrachus*) and climbing perch (*Anabas testudineus*). Total production of catfishes has crossed 68000 metric tons (DoF 2018). Among these fishes, walking catfish is one of the popular indigenous air breathing catfish which could be cultured in shallow and small ponds by facing various adverse conditions (Sidthimunka 1971).

Vitamin C (AA-Ascorbic acid) is essential for normal physiological functions in animals including fish (Wilson and Poe 1973, Lim and Lovell 1978) and has been found to be one of nutrients correlating with fish immunity (Anbarasu and Chandran 2001). Like other teleost fishes *C. batrachus* lacks the capability of biosynthesis of vitamin C due to absence of an essential enzyme gulono-lactone oxidase (Fracalossi *et al*. 2001) that is responsible for the synthesis of vitamin C. Therefore, an exogenous source of AA is required in fish diets. For fish, optimization of the amounts of vitamin C in formulated feeds is important because either low or high levels of vitamin may lead to poor growth. Therefore the present study was designed to determine the effect of different levels of vitamin C on the growth preformances and survival rate of *C. batrachus* with a view to find an optimal dose.

## Materials and Methods

***Experimental site*:** The research was undertaken at the Wet Laboratory, Faculty of Fisheries, Bangladesh Agriculture University (BAU), Mymensingh. Stored ground water was used for

rearing the fish. Aeration was provided from a 1.0- HP compressor to all the experimental aquaria.

***Experimental design*:** The experiment was conducted in 12 glass aquaria (50L capacity) comprising four treatments: Treatment 1 ($T_1$), Treatment 2 ($T_2$), Treatment 3 ($T_3$), and Treatment 4 ($T_4$)] each having three replications. The diets for T1, T2. T3 and T4 contained 0 mg, 500 mg, 1000 mg and 1500 mg vitamin C respectively. *C. batrachus* fingerlings having an initial weight of $6.11 \pm 0.47$ g were randomly distributed at a rate of 15 fish per aquarium. An adequate level of dissolved oxygen, in each aquarium was maintained through artificial aeration during the experimental period. All the aquaria were kept on 1m high wooden table to facilitate better observation and accessibility. Partial change of water from each aquarium was done daily during the removal of uneaten feed and faces.

***Sample collection and acclimatization*:** Fingerlings of *C. batrachus* were collected from Al-Helal Fish Seed Hatchery adjacent to Bangladesh Agriculture University. Transportation of fry was done in oxygen bag to avoid stress and injury. During the period of acclimation, adequate oxygen supply was maintained through artificial aeration and fish were fed formulated pelleted feed.

***Feed ingredients collection and feed formulation*:** The proximate composition of the commonly available ingredients and their inclusion levels in the experimental diets are presented in Table I and the proximate compositions of the formulated diet are presented in Table II. Four graded levels of vitamin C at 0, 500, 1000, and 1500 mg/kg diets were included in the basal diet (Vitamin C was supplemented separately to the basal diet at the expense of wheat flour). The ingredients were weighed, ground and thoroughly mixed. A dough was prepared and pelleted with meat a mincer through a 0.5 mm diameter. After pelleting, feed were air dried and put in an air-tight plastic container and stored at -20°C until fed.

***Proximate composition analysis*:** Proximate composition of prepared feeds and individual ingredients were analyzed in the Fish Nutrition Laboratory, Department of Aquaculture, BAU following Association of Officials Analytical Chemists (AOAC, 2000) method.

**Table I. Proximate composition (dry basis) of the feed ingredients and their inclusion levels in the formulated experimental diets**

| Feed ingredients | Proximate composition | | | | | | % Inclusion |
|---|---|---|---|---|---|---|---|
| | Crude Protein (%) | Crude Lipid (%) | Moisture (%) | Ash (%) | Crude Fibre (%) | NFE | |
| Fish meal | 58.74 | 10.60 | 8.51 | 16.60 | 2.80 | 2.75 | 40 |
| Rice bran | 13.61 | 11.40 | 12.01 | 13.60 | 6.80 | 42.58 | 25 |
| Wheat bran | 16.13 | 5.60 | 13.14 | 13.40 | 6.95 | 44.78 | 25 |
| Soybean oil | 32.34 | 4.60 | 13.48 | 8.40 | 6.88 | 34.3 | 4 |
| Molasses | - | - | - | - | - | - | 5 |
| Vitamin Mineral Premix | - | - | - | - | - | - | 1 |

**Table II. Proximate composition analysis of formulated feed (dry basis)**

| Diet | Moisture (%) | Crude Lipid (%) | Crude Protein (%) | Ash (%) | Crude Fibre (%) | NFE |
|---|---|---|---|---|---|---|
| T1 diet | 14.23 | 6.88 | 35.17 | 12.73 | 5.80 | 25.19 |
| T2 diet | 14.48 | 7.40 | 35.35 | 12.73 | 5.35 | 24.69 |
| T3 diet | 14.45 | 7.68 | 35.37 | 12.66 | 5.46 | 24.38 |
| T4 diet | 14.19 | 7.80 | 35.95 | 13.69 | 5.65 | 22.72 |

***Sampling Procedure*:** The experiment was conducted for 11 weeks (from 11 February to 29 April 2017) having three replications for each treatment. Feeding was done twice daily @ 5%/BW at 9.00 am and 5.00 pm throughout the study period. Initial and final weight of fish in each aquarium was recorded. Fish were bulk weighed at every seven days interval to keep record of fish weight. Fish were caught by using a fine meshed scoop net and excess water then removed from fish body gently by using a blotting paper before weighing by the digital balance. After weighing the fingerlings were released in the aquarium.

***Statistical analysis*:** The collected data were subjected to a one way analysis of variance (ANOVA) by Microsoft Excel and XL-Stat (version 2013). The level of significance was set at $p<0.05$ to see whether the influence of different treatments on these parameters were significant or not. The means of different treatment were compared by Duncan Multiple Range Test (Duncan, 1955) to test the significance of variation between the treatment means.

## Results

***Weight gain (g)* and *specific growth rate (%/day)-SGR:*** The weekly increment in weight is shown in Fig. 1. The mean final weight of *C. batrachus* in different treatments varied from 8.31g to 10.23g and the mean weight gain of different treatments ranged from 2.20g to 4.12g. The final weight and mean weight gain (g) in treatment $T_4$ was significantly higher ($p<0.05$) than those of $T_3$, $T_2$, and $T_1$, respectively (Table III). The specific growth rate (%/day) ranged from 0.40% to 0.67% /day. The highest SGR (0.67%/day) was found in treatment $T_4$ followed by $T_3$, $T_2$, $T_1$, respectively. There was significance variation of SGR between $T_1$ and $T_4$, $T_2$ and $T_4$ ($p<0.05$) but no significance variation between $T_3$ and $T_2$; $T_3$ and $T_4$ (Table III).

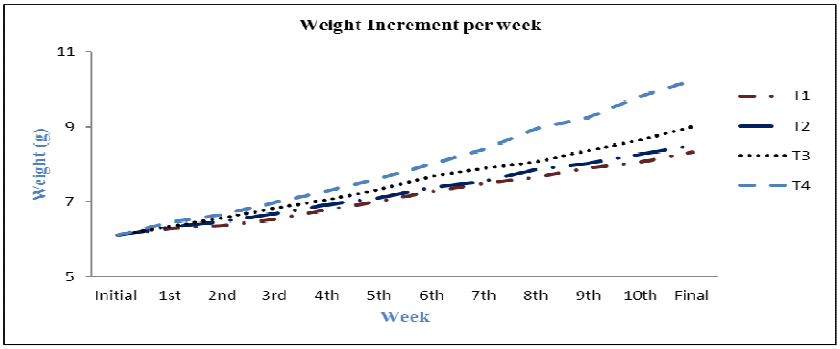


**Fig. 1.** Mean weight increment (g) of *C. batrachus* in different treatments during the 11 week experimental period.

***Food conversion ratio (FCR):*** Mean food conversion ratio in different treatments ranged from 1.46 to 2.22.The highest FCR was obtained in treatment T1 followed by $T_4$, $T_3$, and $T_2$, respectively. No significant ($p>0.05$) variation was observed in mean FCR among the four treatments (Table III).

***Food conversion efficiency (FCE) and Protein efficiency ratio (PER)*:** Mean food conversion efficiency in different treatments ranged from 0.452 to 0.684. The highest FCE was obtained in treatment $T_4$ followed by $T_3$, $T_2$, and T1, respectively. There was no significant ($P\geq0.05$) variation in mean FCR among the four treatments. Mean protein efficiency ratio in different treatments varied from 0.236 to 0.285.The highest PER was found in treatment $T_4$ followed by $T_3$, $T_2$, and T1, respectively. There was significance ($p<0.05$) difference between T1 and $T_4$; $T_2$ and $T_4$ but no significance variation between $T_3$ and $T_2$; $T_3$ and $T_4$; $T_3$ and T1; T1 and $T_2$ (Table III).

***Survival rate (%)*:** The mean survival rate (%) of *C. batrachus* under different treatments ranged from 77.50 to 95.00%. Significant differences ($p<0.05$) in survival rates were found between the T1, and $T_4$; $T_2$ and $T_4$; $T_2$ and $T_3$; $T_3$ and $T_4$ but not between T1 and $T_2$ (Table III).

**Table III. The effect of different treatments on growth performance, feed utilization and survival of walking catfish (*C. batrachus*) reared in aquarium (Mean ± SE) during the study period**

| Variable | Treatment 1 | Treatment 2 | Treatment 3 | Treatment 4 |
|---|---|---|---|---|
| Initial weight (g) | 6.11 ± 0.47 | 6.11 ± 0.47 | 6.11 ± 0.47 | 6.11 ± 0.47 |
| Final weight (g) | 8.31 ± 0.21$^c$ | 8.52 ± 0.14$^c$ | 9.01 ± 0.01$^b$ | 10.23 ± 0.32$^a$ |
| Weight gain (g) | 2.20 ± 0.21$^c$ | 2.41 ± 0.14$^c$ | 2.90 ± 0.01$^b$ | 4.12 ± 0.23$^a$ |
| SGR (%/day) | 0.40±0.03$^c$ | 0.43±0.02$^c$ | 0.50±0.00$^b$ | 0.67±0.03$^a$ |
| FCR | 2.22±0.13 | 1.80±0.08 | 1.80±0.03 | 1.46±0.05 |
| FCE | 0.45±0.02 | 0.56±0.26 | 0.54±0.01 | 0.68±0.02 |
| Survival rate (%) | 77.50±1.77$^c$ | 77.50±0.00$^c$ | 85.00±7.07$^b$ | 95.00±7.07$^a$ |
| PER | 0.236±0.005$^c$ | 0.241±0.002$^c$ | 0.254±0.001$^b$ | 0.285±0.009$^a$ |

The values in the same row having similar letter (s) do not differ significantly otherwise differ significantly ($p<0.05$) as per Duncan Multiple Range Test (DMRT).

## Discussion

In this experiment effects of dietary vitamin C on the growth and survival rate of catfish (*C. batrachus*) in aquarium were investigated. Compared with other treatments, growth performance of magur were significantly ($p<0.05$) better in treatments 4 which were provided with higher levels of vitamin C (1500 mg/kg) feed. Tewary and Patra (2008) assessed the effect of dietary vitamin C on the growth performance rohu fingerlings at a concentration of 500, 1000 and 1500 mg/kg diet for 60 days and recommended that vitamin C concentration of up to 1000 mg/kg diet would be the most appropriate concentration for the better growth of rohu fingerlings. In our study, the maximum mean weight gain was 4.12±0.32g in treatment 4, whereas, the minimum mean weight gain was 2.20±0.22g in T1. Nsonga et al. (2009) showed that maximum mean final weight in juvenile tilapia (*Oreochromis karongae*) was 13.79±0.28 g where vitamin C at a dose of 60 mg/kg feed was supplied and minimum mean final weight was 4.78±0.12 g where no vitamin C was provided. Peng *et al.* (2013) conducted an experiment to evaluate the effect of

high dose vitamin C (100mg, 450mg and 800mg) on the growth, tissue ascorbic acid concentrations and physiological response to transportation stress in juvenile silver pomfret (*Pampus argenteus*) and did not found significant variations. Misra *et al.* (2007) evaluated the effects of different dietary levels of vitamin C on the immunity, growth and survival of Indian major carp *Labeo rohita* through a 11 week feeding trial feeding on diets containing 0 mg/kg, 500 mg/kg, 1000 mg/kg, and 1500 mg/kg vitamin C. Results showed that supplementation of vitamin C at a dose of 1500 mg/kg feed had significant positive effects on the food conversion ratio (FCR), specific growth rate (SGR), average daily gain. They suggested that Diet 4 containing 1500 mg/kg vitamin C feed would be more effective for better growth of *C. batrachus*.

In the present study, the mean feed conversion ratio (FCR) in T1 (control) was 2.2± 0.13, T2 was 1.80 0.08, T3 was 1.80 0.03 and T4 was 1.4±0.053. The lowest FCR value was recorded in $T_4$. The mean food conversion efficiency (FCE) $T_1$, $T_2$, $T_3$ and $T_4$ was 0.452± 0.026, 0.556±0.026, 0.536±0.009 and 0.68 ± 0.025, respectively. The higher FCE value was recorded in $T_4$. Tewary and Patra (2008) reported that the food conversion ratio (6.10 ± 0.18) was found the highest in the control (without vitamin C) while the lowest food conversion ratio (3.43 ± 0.19) was measured when the diet was supplemented with 1000 mg vitamin C per kg feed. The FCR values in the present study were lower than the above studies. This might be due to good quality and high energy value of the feed ingredients used in this experiment, quality of the fry and overall good condition during the experimental period.

In the present study protein efficiency ratio in T1 (control) was 0.236±0.05, in T2 was 0.241±0.002, in T3 was 0.254±0.001 and in T4 was 0.285±0.009. The lowest protein efficiency ratio was observed in T1 and the highest in T4. Gbadamosi *et al.* (2006) found the highest protein efficiency ratio in African catfish *C. gariepinus* which was 1.08± 0.02 for the diet supplemented with vitamin C at a dose of 200 mg/kg feed and lowest protein efficiency ratio was 0.55±0.01 for control diet which was not supplemented with vitamin C. The PER in the present study were lower than (0.55 to 1.08) reported by Gbadamosi *et al.* (2006).

In the present study, specific growth rate (SGR) varied from 0.40±0.03 to 0.67±0.04. SGR value was higher in T4 which contained the highest vitamin C level (1500 mg/kg feed) and lower in treatment 1 (control) which was not supplemented with vitamin C. Alam et al. (2009) found the highest value of SGR for *H. fossilis* fed with diet supplemented with vitamin C at a dose of 1200 mg/kg feed (1.75±0.10% per day) and lowest in control (without vitamin C), 0.81 ±0.016% per day. The SGR in the present study were lower than (0.81 to 1.75) reported by Alam *et al.* (2009).

In the present study survival rate varied from 77.50±1.77 to 95.00±7.07. Survival rate was higher in T4 and lower in T1. Ashraf and Rauf (2008) observed 70±6.0% survival rate when the diet was supplemented with vitamin C at a dose of 300 mg/kg feed and lowest in control 30 ± 6.0% where no vitamin C was supplemented. In the present study, survival rate was higher than the above study, which might be due to the use of aerator in the experimental aquaria during the experimental period and regular exchange of water. The better water quality might have positively contributed to the higher survival (%) of the fish.

It can be concluded that best growth performance and survival rate of *C. batrachus* can be obtained by supplementing 1500 mg/kg feed with a diet having 35% protein and 7% lipid.

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