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Effect of L-tryptophan on Growth and Survival of Pabda Fry, *Ompok bimaculatus* (Bloch, 1794)

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Abstract

Availability of quality seeds of Pabda, *Ompok bimaculatus* (Bloch, 1794) remains a constraint for development of aquaculture of this species. Poor fry survival, attributed to several factors, is a major bottleneck for the culture of Pabda. A study was conducted for 30 days to evaluate the effect of tryptophan solution on growth and survival of Pabda, *O. bimaculatus* fry. Fifty numbers of fry (0.113±0.001 g; 2.43±0.01 cm) were stocked into aerated aquariums following a completely randomized design consisting of four treatments with three replicates each. Four different doses of L-tryptophan *viz.,* 0 ppm (control), 2 ppm (T1), 4 ppm (T2) and 6 ppm (T3) were prepared and tested. Fishes were fed twice with chopped tubifex to satiation. The results showed that groups fed with tryptophan had significantly lower cannibalism and higher survival rate than control group. However, the growth performance of tryptophan groups were significantly lower than the control group. The maximum survival (51.33±0.67%) and minimum cannibalism (22.00±1.15%) coupled with high growth (0.38±0.01 g) was recorded in group treated at 2 ppm tryptophan. The study concludes that a dose of 2 ppm tryptophan will reduce cannibalism without compromising the growth of pabda fry during rearing.

Keywords: Pabda, *Ompok bimaculatus*, L-tryptophan, growth, survival

Introduction

In India, diversification of aquaculture with endemic catfishes is widely gaining importance due to lucrative return on investment. Pabda or Indian butter catfish, *O. bimaculatus* is an indigenous catfish (Family: Siluridae) that has gained importance due to its superior taste, excellent nutritional profile, soft bony structure, rich lipo-protein and market value (Banik et al., 2012; Biswas et al., 2018). Although successful induced breeding of the fish has been established and reported by several researchers (Chakrabarti et al., 2012; Biswas et al., 2018; Rawat et al., 2018), there is an urgent need to resolve the issue of low survival during fry rearing that occurs due to heavy cannibalism, inadequate food, improper feeding regime, stocking density, and rearing conditions (Banik et al., 2012; Rawat et al., 2018). In case of rearing of predatory fish, intra-cohort aggression and cannibalism has a major impact on survival (Loadman et al., 1986; Katavic et al., 1989). During the initial stage of rearing, type I cannibalism (biting the abdomen or tail) dominates as the size heterogeneity among the larvae is low (Baras & Jobling, 2002). However, as the size heterogeneity increases, type II cannibalism occurs, which is characterised by complete prey ingestion (Hecht & Pienaar, 1993). The approaches made in reducing cannibalism during seed rearing of *O. bimaculatus* involves extrinsic factors such as reduced stocking density, routine size-grading and provision of shelters (Debnath et al., 2013; Biswas et al., 2018). But, all these approaches are labour intensive and also may cause additional stress to the fishes. The use of anti-aggressives through water might be an alternative strategy to improve survival during rearing of fry.

Tryptophan is an essential amino acid for fish (Wilson, 1989), which is a precursor of serotonin (5-HT) and is widely used to reduce aggressive behaviour (Winberg et al., 2001; Hseu et al., 2003; Höglund et al., 2005), stress-releasing effects (Johnston et al. 1990) and feeding performance (Pedro et al., 1998; Ortega et al., 2005). The dietary supplementation of tryptophan has been reported to

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increase the levels of brain serotonergic activity which suppresses cannibalism in fish species (Johnston et al. 1990; Pedro et al. 1998; Winberg et al., 2001; Król & Zakêœ, 2016). Studies on fish species such as rainbow trout (Winberg et al., 2001), grouper (Hseu et al. 2003), Atlantic cod (Höglund et al., 2005), pikeperch (Król & Zakêœ, 2016) and Asian seabass (Kumar et al., 2017) have shown that supplementation of tryptophan significantly reduces the cannibalistic behaviour. It is also reported that the high levels of brain serotonin reduces the food intake in fish (Pedro et al., 1998; Hseu et al., 2003), thereby resulting in depressed fish growth. In this backdrop, the present work has been undertaken to find the effect of L-tryptophan solution on growth and survival during fry rearing of Pabda, *O. bimaculatus*.

Materials and Methods

The study was conducted for 30 days at the College of Fisheries, Central Agricultural University, Tripura, India aiming to evaluate four different doses of tryptophan on the rearing performance of *O. bimaculatus*. All the preparation needed for induced breeding of Pabda was done in the wet laboratory. After successful breeding (Biswas et al., 2018), 2 days old larvae were obtained and fed with natural food like planktons, chopped tubifex up to 12 day post hatch (dph). Subsequently, the resulting seed has been collected and stocked for the experiment. Twelve aquariums (30.0x15.0x15.0 cm), filled with 20 l of water were used for the experiment. Each aquarium was stocked with fifty Pabda seeds (0.113±0.001 g; 2.43±0.01 cm) following a completely randomized design (CRD) consisting of four treatments with three replicates each. Four different doses of tryptophan solution were tested for the study namely control (0 ppm), T1 (2 ppm), T2 (4 ppm) and T3 (6 ppm) daily after the exchange of water routinely followed by feeding. Fishes were kept under continuous aeration and fed with chopped tubifex twice a day upto satiation. The daily water exchange amounted to 50% of the total unit volume with chlorine-free water. The water quality parameters such as temperature, pH, dissolved oxygen (DO), free carbon dioxide (CO_2), carbonate hardness, ammonia-N and nitrate–N were recorded every week following standard method (APHA, 1998). The fish were fed twice at 05.00 and 18.00 h under normal light regime and sampling of fish was done every 15 days.

A sub-group of 20 randomly selected fish from each treatment were individually measured to the nearest 0.1 mg. The growth response and survival were assessed by using the following formulae:

Length increment (cm) = Final mean length – initial mean length

Body Weight gain (BWG; %) =100[(final mean weight – initial mean weight)/ initial mean weight]

Specific growth rate (SGR; % day^{-1}) = 100[{ln (final mean weight) – ln (initial mean weight)}/ experimental periods]

Mean daily weight gain (%) = 100[(total final weight – total initial weight)/days of experiment]

Fulton's condition factor (K) = (final live weight/final length^3)

Total biomass (g) = final number of fish × mean final weight

Performance index (PI) = Survival rate (%) [(Final mean body weight –Initial mean body weight)/ days of experiment]

Survival (%) = 100 [(Number of surviving fish/ Total number of larvae stocked)]

Cannibalism (%) = 100 × {Initial number of fish - (Final number of fish + Number of dead fish registered)}/Initial number of fish

Data were analyzed using statistical package SPSS, in which data were subjected to one-way ANOVA and Duncan's multiple range tests to determine the significant differences between the means. Comparisons were made at the 5% probability level.

Results and Discussion

The range of different water quality parameters in different treatments are summarized in Table 1. All the parameters were within the tolerable limits as reported for pabda fry rearing (Chakrabarti et al., 2012; Rawat et al., 2018). Such values of the parameters indicated prevalence of suitable growing environment for the species. Growth in terms of final length, length increment, final weight, body weight gain and specific growth rate of *O. bimaculatus* fry under different treatments are shown in Table 2. The final weight (g), body weight gain (%), mean daily weight gain and specific growth rate (SGR; %day^{-1}) was found significantly ($p<0.05$) higher in control group followed by T1 treatment compared to other tryptophan groups. The final length and length increment of fish was recorded significantly higher in control group. Such lower growth performance among the tryptophan treatments is probably attributed to the tryptophan induced reduction in increased brain 5-HT concentration which in turn significantly reduces feed intake and hence growth over the control group (Pedro et al., 1998; Hseu et al., 2003; Ortega et al., 2005; Król & Zakêœ, 2016). Peng & Peter (1997) observed that 5-HT can directly act on somatotrophs in the pituitary gland and inhibit growth hormone secretion in goldfish. Growth hormones play a key role in stimulating the growth as well as the food intake in fish (Peng & Peter, 1997; Lin et al., 2000). Several other studies also reported that the tryptophan decreased the growth in fish such as grouper, *Epinephelus coioides* (Hseu et al., 2003); European seabass *Dicentrarchus labrax* (Papoutsoglou et al., 2005); goldfish *Carassius auratus* (Pedro et al., 1998). It was also observed from the study that tryptophan dose beyond 2 ppm showed significantly lower growth rate. Young (1996) and Hseu et al. (2003) reported that much higher levels of TRP in diet reduced the growth rate in juvenile grouper, which is relevant to the present findings. This could be due to the higher tryptophan intake from water that increases the brain serotonergic activity and subsequent decrease in the appetite.

Production parameters such as survival, biomass, condition factor and performance index are depicted in Table 3. The tryptophan treated groups showed significantly higher survival compared to control group. However, no significant differences in survival were found among the tryptophan groups. Moreover, significantly higher cannibalism (%) was observed in control group (46.67±1.33%) compared to tryptophan groups (Fig. 1). Chakrabarti et al. (2007) and Debnath et al. (2013) reported the carnivorous and cannibalistic mode in *O. bimaculatus* during early seedling stage which can be attributed to intrinsic genetic effects that result in heterogeneous growth. Cannibalism contributes a lot to mortality during hatchery rearing, even where

Table 1. Physico-chemical parameters of water in different experimental groups

Parameters	Control	T1	T2	T3
Dissolved oxygen (mg l^{-1})	7.16- 8.24	7.21-8.34	7.12-8.59	7.14-8.23
Temperature (°C)	26.6-29.0	26.5-29.6	26.2-29.2	26.6-28.9
Water pH	7.27-7.92	7.07-7.84	7.45-7.96	7.34-7.87
Carbon dioxide (mg l^{-1})	1.1-2.4	1.3-2.5	1.1-2.3	1.1-2.5
Total alkalinity (mg l^{-1})	39-49	42-50	40-48	39-49
Hardness (mg l^{-1})	35-39	34-41	35-41	34-39
Ammonia (mg l^{-1})	0.02-0.05	0.01-0.04	0.02-0.05	0.01-0.03

Table 2. Growth Parameters in different experimental groups

Parameters	Control	T1	T2	T3	P-value
Initial stock (nos.)	50	50	50	50	-
Final length (cm)	3.49±0.05^{c}	3.45±0.02^{bc}	3.39±0.01^{ab}	3.34±0.01^{a}	0.032
Final weight (g)	0.41±0.01^{c}	0.38±0.01^{b}	0.35±0.01^{a}	0.33±0.02^{a}	0.000
Body weight gain (%)	259.37±7.94^{c}	233.56±4.56^{b}	211.33±7.23^{ab}	195.06±9.23^{a}	0.001
Length increment (cm)	1.06±0.05^{b}	1.01±0.02^{ab}	0.96±0.02^{ab}	0.91±0.02^{a}	0.039
Specific growth rate (% day^{-1})	6.37±0.03^{b}	6.29±0.02^{ab}	6.23±0.04^{a}	6.17±0.04^{a}	0.022
Mean daily weight gain (%)	0.98±0.03^{c}	0.88±0.01^{b}	0.79±0.02^{a}	0.73±0.03^{a}	0.001

*Overall mean value having different superscript in the same row shows significance difference ($p<0.05$) (mean ± S.E.); Superscript with same alphabets denotes no significance difference between the treatments.

Table 3. Yield parameters in different experimental groups

Parameter	Control	T1	T2	T3	P-value
Survival (%)	32.00±1.15^{a}	51.33±0.67^{b}	53.33±1.76^{b}	50.67±1.76^{b}	0.000
Total biomass (g)	6.49±0.14^{a}	9.70±0.15^{c}	9.34±0.25^{c}	8.43±0.13^{b}	0.000
Condition factor (K)	1.42±0.03^{c}	1.30±0.02^{b}	1.19±0.02^{a}	1.11±0.03^{a}	0.000
Performance index	0.31±0.005^{a}	0.45±0.008^{d}	0.42±0.010^{c}	0.37±0.008^{b}	0.000

*Overall mean value having different superscript in the same row shows significance difference (p<0.05) (mean ± S.E.); a, b, c, d denotes significance differences between different treatments.

conditions appear to be ideal (Parameswaran et al., 1970; Rawat et al., 2018) and it is usually more serious when food availability is limited. The decreased intensity of cannibalism in tryptophan treated groups is due to the calming effect on the aggression of pabda fry. The capability of tryptophan to represses aggression could be due to the increased activity of 5-HT in the brain and in turn, may suppress the aggressive behavior of dominants as confirmed in several studies on cultured fish species (Winberg et al., 2001; Hseu et al., 2003; Höglund et al., 2005; Król & Zakêœ, 2016; Kumar et al., 2017). Moreover, mortality caused due to cannibalism (mainly type I cannibalism) was more prone in control group rather than the tryptophan groups.

The total biomass of fish was significantly high in T1 and T2 groups, while lower was in control group. Performance index was significantly higher in T1 group. This might be due to more individual growth as well as survival in case of T1 group as evidenced by the high level of survival percentage. Similar studies have been reported in *Oncorhynchus mykiss* (Winberg et al., 2001), Atlantic cod (Höglund et al., 2005) and *Brycon amazonicus* (Wolkers et al., 2012).

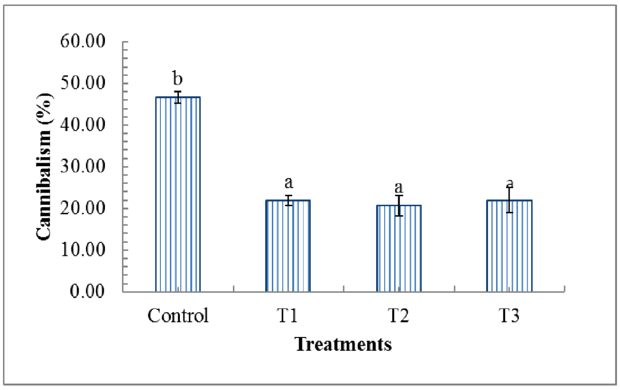

Fig. 1. Effects of tryptophan on cannibalism (%) of *O. bimaculatus* fry. Mean values are data of three replicates. Same letters on bars signify no significant differences at (p>0.05) level

In summary, it is found that the growth of pabda fry is higher in the control treatment, however, survival was significantly lower. Therefore, additions of tryptophan could be a promising aquaculture management strategy for carnivorous fish like *O. bimaculatus* for better survival. Moreover, for hatchery production of many of the fishes, survivability of the fry is more important than growth as economic returns through the sale of the larger amount of fry is high, so L-tryptophan at 2 ppm dose in solution can be recommended to end users as it gives better survival without compromising the growth of pabda fry than other tryptophan groups. The current study is laboratory-based, so further studies are needed to clarify the effect of tryptophan solution on feed intake and feeding behaviour in *O. bimaculatus* fry in commercial conditions.

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