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Growth Performance and Brood Stock Management of Small Indigenous Fish Mola Carplet, *Amblypharyngodon mola* (Hamilton, 1822)

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ABSTRACT

Spontaneous spawning of wild caught and domesticated mola carplet, Amblypharyngodon mola was attempted in controlled condition using an aquatic plant hydrilla, Hydrilla verticillata as a modified habitat. Growth performance of hatched larvae revealed that the larval growth (weight gain %) was significantly higher ($p < 0.05$) with a mixed diet (63.76 ± 0.52%) than natural diet (40.42 ± 0.26%) or artificial diet (13.74 ± 0.39%) whereas juvenile mola grew better on the artificial diet. Significantly higher maturity rate (> 80 %) of mola was observed in tanks/ ponds having hydrilla as an additional habitat than the tanks /ponds having no hydrilla. Similarly, higher fecundity of mola (>900) was recorded in tanks having hydrilla. Better spawning performance was also noted using hydrilla based habitat. The findings can be useful in artificial propagation and diversification of small indigenous fish species in captivity.

Key words: Habitat manipulation, Hydrilla, Larval recovery, Spawning, Small indigenous fish

INTRODUCTION

Fish are not only a good source of animal protein but also provide other beneficial micronutrients such as calcium, iron, zinc, phosphorous, selenium, fluorine, iodine, and polyunsaturated fatty acids (Pal *et al.,* 2018; Thilsted *et al.,* 2014). The protein requirement of the global population is partially met from aquaculture. The global annual aquaculture production in the year 2016 was 80 million tonnes that comprised of nearly 598 species out of which 369 are finfish (FAO, 2018). This production is mostly contributed by medium to large size fish. It is pertinent to mention that spawning and culture of small indigenous fish species have received limited attention over the years though they are richer in nutrients and vitamins than other cultured table fish. One of the small fish that requires attention is Amblypharyngodon mola (Hamilton, 1822), commonly referred to mola carplet of Cyprinidae family (Menon, 1999; Saha *et al.,* 2009). It is a small freshwater fish found in the rivers, lakes, reservoirs, slow-moving streams, floodplains, inundated fields, ditches and ponds in the Indian sub-continent. Mola is preferred for its taste (Roos *et al.,* 1999; Saha *et al.,* 2009) and is rich in nutrients: every 100 g of the mola contains 2680 retinol activity equivalent units of vitamin A, 5.7 mg of iron, 776 mg of calcium and 3.2 mg of zinc. Mola contains 20 times more calcium and 80 times more vitamin A than carps such as the silver carp (Hypophthalmichthys molitrix) and rohu (*Labeo rohita*) (Thilsted *et al.,* 2014).

Despite being highly nutritious and particularly suitable for farming and diversification in pond ecosystems, its artificial propagation and culture is hampered by insufficient information and research. The present study aimed at production of mola seed by habitat manipulated spontaneous spawning, evaluation of growth performance and brood stock management to promote farming of mola.

MATERIALS AND METHODS

Wild mola stock was collected using a dragnet in the morning hours from river Mahanadi at Banki in Cuttack district, Odisha, India with the help from local fisherfolk. A total of 30,000 mola conditioned and packed in 10 L capacity plastic bags (50 numbers in each bag) with one part of oxygen and two parts of water (vol/vol) and transported to the Carp Breeding Unit, ICAR-Central Institute of Freshwater Aquaculture, Bhubaneswar which is 70 km away from the collection site. After arrival, all the live mola were treated with 5% potassium permanganate

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for 30 to 60 seconds and stocked in cement tanks of 7500 L capacity with 24-h aeration. However, due to its delicate nature only 40% could survive despite the best care. Spontaneous spawning was simulated in two types of habitats viz. earthen pond and cement tank in duplicate with or without addition of submerged weed. One set of ponds/tanks were added with hydrilla (Hydrilla verticillata) to the tune of 20% of their water spread area and other set of tanks/ponds were devoid of hydrilla that served as control. Hydrilla was used in this study due to the fact that large amount of hydrilla could be seen in natural habitat from where mola was collected.

Healthy brood males (5.2 ± 1.1 cm length, weight 1.3 ± 0.56 g) and females (5.8 ± 1.2 cm length, weight 1.9 ± 0.59 g) mola were identified by their sexual dimorphism and kept together in equal proportions in cement tanks (7500 L; 3.0 m × 2.5 m × 1.0 m deep) and also in 0.01 ha earthen ponds at a stocking rate of 130/m³. To maintain the concentration of dissolved oxygen and to simulate natural environment, 30% of water was removed from the tank and replaced with freshwater every day. Two aerators were used in the tank but not in the pond. Spawning tanks were kept under natural photoperiod (12L:14D). Brood fish were given a commercially formulated powdered feed (protein 37.3%, crude fat 6.7%, crude fibre 3.8%, dry matter 94.7%, total ash 7.0%) at 3% of the body weight twice a day and live plankton (crude protein 43.8%, crude fibre 2.7%, dry matter 8.9%) collected from the wild twice a week. The proximate composition of the plankton and the artificial diet was determined using standard methods (AOAC, 2005).Various parameters such as relative fecundity, hatchlings/larval recovery (%) were calculated as follows:

To measure the growth of mola larvae during the initial days, 20 individuals of average length 16.2 ± 0.5 mm and average weight 38 ± 0.1 mg were reared in 150 L glass tanks for almost 10 weeks and fed three times a day with plankton and artificial diet at 5% of their body weight/day. Water quality was maintained by exchanging water to the tune of nearly 75% during one-week period and continuous aeration throughout the experiment. Feeding was discontinued 24 hours before sampling to avoid any larval excrement. In every 15 days, samples of the larvae were collected using a hand net and their length was measured with a scale and weight with an electronic weighing balance (Afcoset ER-200A, India). To find out a suitable diet for better growth and survival of mola fry, each time hatchery produced 20 numbers of active mola fry (15.5 ± 0 mm long, weighing 37.5 ± 0 mg) were introduced into each 75L tank. Three treatments were tested in duplicate, namely (1) natural diet (plankton feed) (crude protein 43.8%, crude fibre 2.7%, dry matter 8.9%), (2) artificial diet (Formulated powdered feed) (protein 37.3%, crude fat 6.7%, crude fibre 3.8%, dry matter 94.7%, total ash 7.0%) and (3) mixed diet (plankton feed + formulated powdered feed). The feed was given three times a day at 5% of the body weight of fish. All the tanks were aerated round the clock throughout the experiment for 45 days. Water exchange was done twice a week and uneaten feed and excreta were removed before feeding once in day. At the end of the experiment, 15 larvae were collected from each unit and their length was measured to the nearest millimetre and weight to the nearest milligram. Similarly, 6-month-old juveniles (42.4 ± 0.6 mm long, weighing 701.7 ± 2.9 mg) were also studied for 30 days.

Instantaneous growth rate (mg/day)

$$= \frac{[\text{Ln}\{\text{final weight (mg)}\} - \text{Ln}\{\text{initial weight (mg)}\}]}{\text{duration of rearing period (days)}}$$

Instantaneous growth rate (mg/day)

$$= \frac{[\text{Ln}\{\text{final weight (mg)}\} - \text{Ln}\{\text{initial weight (mg)}\}]}{\text{duration of rearing period (days)}} \times 100$$

$$\text{Condition factor} = \frac{\text{weight}}{\text{Length}} \text{x100}$$

RESULTS AND DISCUSSION

Significantly higher ($p < 0.05$) relative fecundity of mola was recorded in tanks and ponds with addition of hydrilla (937 and 906) than the tanks and ponds without hydrilla. Maturity rate of females were higher (85%) in cement tanks with hydrilla than controls (48%) without hydrilla. In earthen ponds too, the same was higher (80%) with the addition of hydrilla than its absence (60%). Similar trend was also noticed in larval recovery (yolk sac absorbed larvae) rate in cement tanks. (Table 1).

Table 1. Manipulation of habitat for spontaneous spawning of mola with or without addition of an aquatic weed, hydrilla (Hydrilla verticillata)

Parameter	Cement tank		Pond	
	With hydrilla	Without hydrilla	With hydrilla	Without hydrilla
Total number of broodfish stocked (male and female)	700 ± 0^a	700 ± 0^a	1000 ± 0^a	1000 ± 0^a
Average wt. of female	1.9 ± 0.12	1.9 ± 0.22	1.9 ± 0.21	1.9 ± 0.25
Relative fecundity	937 ± 1.22^b	145 ± 0.12^a	906 ± 1.12^b	123 ± 0.15^a
Maturity rate (%) (female)	85 ± 2.22^b	48 ± 0.12^a	80 ± 2.10^b	60 ± 1.22^a
Larval recovery rate (%)	85 ± 1.52	80 ± 1.22	-	-
	700 ± 0^a	700 ± 0^a	1000 ± 0^a	1000 ± 0^a

Different superscript in same row indicates the significant differences (P<0.05).

The fry showed good growth: between 5 dph and 80 dph, the average length increased from 16.2 ± 0.5 mm to 29 ± 0.4 mm and average weight, from 38 ± 0.1 mg to 248 ± 1.6 mg. Significant differences were seen in four parameters related to weight: final weight, weight gain, daily weight gain, per cent weight gain and specific growth rate (Table 2).

Table 2. Growth and survival of mola fry under different diets

Parameters	Natural diet (Plankton feed)	Artificial diet (Formulated powdered feed)	Mixed diet (Plankton feed + Formulated powdered feed)
Initial length (mm)	15.49 ± 0	15.49 ± 0	15.49 ± 0
Initial weight (mg)	37.5 ± 0	37.5 ± 0	37.5 ± 0
Final length (mm)	18.5 ± 0.43	17.47 ± 1.2	19.3 ± 0.23
Final weight (mg)	52.66 ± 0.1^b	42.65 ± 0.15^a	61.41 ± 0.2^c
Weight gain (mg)	15.16 ± 0.1^b	5.15 ± 0.15^a	23.91 ± 0.2^c
Daily weight gain (mg)	0.34 ± 0^b	0.11 ± 0^a	0.53 ± 0^c
Weight gain percent (%)	40.42 ± 0.26^b	13.74 ± 0.39^a	63.76 ± 0.52^c
Instantaneous growth rate (mg/day)	0.008 ± 0.00001^b	0.003 ± 0.00001^a	0.011 ± 0.00001^c
Specific growth rate (%/day)	0.75 ± 0.004^b	0.29 ± 0.01^a	1.1 ± 0.01^c
Condition factor	0.83 ± 0.06	0.82 ± 0.17	0.855 ± 0.03
CV for length (%)	17.09 ± 1.37	15.35 ± 1.37	14.68 ± 0.74
CV for weight (%)	33.15 ± 4.31	27.03 ± 7	22.87 ± 4.57
Yield (mg L-1)	14.48 ± 0.47^b	9.95 ± 0.68^a	16.89 ± 0.46^b
Survival (%)	82.5 ± 2.5	70 ± 5	82.5 ± 2.5
Mortality rate (%)	17.5 ± 2.5	30 ± 5	17.5 ± 2.5
No of larval output	17 ± 0.5	14 ± 1	17 ± 0.5

Different superscript in the same row indicates the significant differences (P<0.05).

Mixed diet proved significantly ($p < 0.05$) superior in terms of per cent weight gain, followed by the natural diet and the wholly artificial diet. Similarly, specific growth rate was also significantly higher in fish fed with the mixed diet than that with the other two diets. No significant differences ($p > 0.05$) in terms of survival, mortality and the number of larval outputs were noticed among all the three treated diets. The juvenile mola gained weight and grew longer as days passed in all the treatments. The growth parameters were significantly influenced by the diet (Table 3).

Table 3. Growth performance and survival of mola juvenile (6-month-old) under different diets

Parameters	Natural diet (Plankton feed)	Artificial diet (Formulated powdered feed)	Mixed diet (Plankton feed + Formulated powdered feed)
Initial length (mm)	42.36 ± 0.57	42.36 ± 0.57	42.36 ± 0.57
Initial weight (mg)	701.67 ± 2.89	701.67 ± 2.89	701.67 ± 2.89
Final length (mm)	44 ± 0.1^{a}	43.87 ± 0.07^{a}	45.93 ± 0.07^{b}
Final weight (mg)	817.32 ± 1.31^{a}	922.32 ± 2^{c}	869.25 ± 0.99^{b}
Weight gain (mg)	115.66 ± 1.31^{a}	220.65 ± 2^{c}	167.59 ± 0.99^{b}
Daily weight gain (mg)	3.86 ± 0.04^{a}	7.36 ± 0.07^{c}	5.59 ± 0.03^{b}
Weight gain percent (%)	16.48 ± 0.19^{a}	31.45 ± 0.29^{c}	23.88 ± 0.14^{b}
Instantaneous growth rate (mg/day)	0.005 ± 0.0001^{a}	0.009 ± 0.0001^{c}	0.007 ± 0.0001^{b}
Specific growth rate (%/day)	0.51 ± 0.01^{a}	0.91 ± 0.01^{c}	0.71 ± 0.001^{b}
Condition factor	0.96 ± 0.005^{b}	1.09 ± 0.007^{c}	0.897 ± 0.003^{a}
CV for length (%)	9.7 ± 1.06	11.45 ± 0.6	6.77 ± 2.65
CV for weight (%)	45.11 ± 0.13^{b}	33.81 ± 5.57^{ab}	22.57 ± 6.49^{a}
Yield (mg L-1)	272.44 ± 0.44^{a}	307.44 ± 0.67^{c}	289.75 ± 0.33^{b}
Survival rate (%)	100	100	100

Different superscript in the same row indicate the significant differences ($P<0.05$).

After 30 days of culture, the weight of mola was significantly higher with the artificial diet followed by mixed diet and natural diet. Other growth parameters such as weight gain, per cent weight gain, daily weight gain and specific growth rate were noticed to be higher in artificial diet than other two (artificial and mixed) diets. Juvenile mola reared on the artificial diet showed higher values of the condition factor than those reared on the other two diets. However, the survival rate of fry was not affected by the diets used.

Aquatic macrophytes provides a conducive spawning environment besides being a source of food and recycle of nutrients and maintenance of water quality (Cowx and Welcomme, 1998). In the present study, hydrilla (an aquatic macrophyte) was introduced into the experimental tanks/ponds to create a modified habitat for mola. This resulted in substantial higher relative fecundity (> 900) and larval recovery (85%) in mola brood reared in habitat modified tanks. This may be attributed to the habitat modified environment that provided a favourable environment for spawning in mola. Moreover, the biological nature of these fish is to find a safe place to spawn and also to avoid predators and sometimes cannibalism. Habitats with moderate amounts of aquatic vegetation provide an optimal environment for reproduction, breeding and raising fish nurseries and also protect the fish from predators (Gettys *et al.,* 2014; Petr, 2000). In some fish, spawning occurs only on ideal spawning grounds (Ganapati *et al.,* 1951). Fish like Cyprinus carpio, Colisa sota also require aquatic vegetation such as hydrilla sp., najas, eichornia, commelina sp. etc. for their spawning (Adamek and Pardo, 2015; Mitra *et al.,* 2006). Artificial spawning substrates are also used to enhance spawning and growth of fish. A comparative account of habitat manipulations used for spawning and larval survival of different fish species is presented in table 5.

We obtained the best growth (23.9 ± 0.2 mg) in mola fry using mixed diet than natural diet (15.2 ± 0.1 mg) or artificial diet (5.2 ± 0.2 mg) given alone. Nearly a four-fold higher weight gain was recorded in mola fry using mixed diet than the artificial diet which is attributed to the fact that it provided the required micronutrients and the protein during the developmental stage. When natural diet was provided alone, the growth rate was slower than the mixed one. This may be due to the protein insufficiency. The pattern of other growth parameters viz. daily weight gain, per cent weight gain and specific growth rate were similar. The superiority of the mixed diet may be due to the fact that it provides all the essential amino acids. It is reported that higher dietary protein level could meet the requirements of body protein synthesis in early stages and supports fast growth of larvae (Bengtson *et al.,* Leger and Sorgeloos, 1991). Trout larvae (Oncorhynchus mykiss), for example, grew better when higher amount of protein was provided (Akbary *et al.,* 2010). Similar findings in the larvae of Rhamdia quelen showed a higher survival rate when given a diet that contained brine shrimp (Artemia nauplii) (Salhi and Bessonart, 2012). In common carp, C. carpio, growth was better when fed with a mixed diet than artificial dry food and live zooplankton (Szlaminska and Przybyl, 1986). It is also reported that Labeo parvus showed better survival, weight gain and specific growth rate when fed with a mixed diet (Montchowui *et al.,* 2012).

Natural live food is considered ideal for freshwater larviculture, especially in the initial days (Awaiss *et al.,*1992; Wang *et al.,* 2005). Here, our observations also suggest that just-hatched larvae (yolk sac absorbed larvae) need live plankton because it is easily digested. We provided plankton and artificial diet as a supplement because enzymes in live food help digestion in first-feeding larvae (Lavens and Sorgeloos, 1996; Verreth *et al.,* 1993). Moreover, the formulated diet given to mola larvae helped them to adapt faster to the artificial diet. Feeding only the artificial diet resulted in significantly greater weight gain (220.6 ± 2 mg) after 30 days than with the other two diets, probably because the juveniles adapted more easily to the artificial diet, as evident from the condition factor: the condition factor is an indicator of fish health including growth and feeding intensity (Froese, 2006) was greater than 1 in the artificial but less than 1 in the other two diets. The factor is also used in evaluating the efficiency of feeding regimes, that is to assess how efficiently a given food is used (Weatherlley, 1972). The higher growth and conversation efficiency obtained with the artificial diet could also be due to higher intake of food (given that the consistency of the artificial diet was better than that of the other two diets leading to greater cohesion of particles), greater palatability and higher protein content (Quintana *et al.,* 2008). The mola juveniles grew to nearly 1 g after the experimental period of 30 days, which is a marketable size and can be used for domestic consumption.

CONCLUSION

Taking into account consumer preference and nutritional value, mola deserves to be grown in large scale. The present study provides essential benchmark information on artificial propagation of mola in captivity. By manipulating their habitat, mola can be made to spawn twice in a year. The potentiality of habitat manipulated spontaneous spawning of mola in captive condition and higher survivability of the hatchery produced individuals made it easier for mola breeders to take up this in a commercial scale. Formulated artificial diet may be recommended for the growth of mola juveniles whereas mixed diet was most suitable for fry rearing. This study opens new vistas for sustainable aquaculture production and species diversification.

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