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Early larval development stages of nest building dwarf gourami *Trichogaster lalius* (Hamilton, 1822) in laboratory condition

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Abstract
The dwarf gourami, *Trichogaster lalius* is a high valued ornamental fish remarkable for its nest building behavior. The species, mainly the male is having high domestic as well as foreign demands to the aquarium traders due to its sparkling, translucent blue colour with red or dark orange stripes in the body. The present study was conducted to know about the early larval development of dwarf gourami in control condition. The hatchlings become free swimming by 3 to 4 day of hatching. Yolk size was totally absorbed by the 4th day and they were fed with laboratory prepared green water containing micro algae, mostly chlorella up to 10th days. From 10th day onwards the hatchlings were fed on laboratory prepared pellet feed and *Tubifex* worm. For successful conservation of the species, mass propagation as well as larval development technique should be disseminated among the ornamental fish breeders which will generate employment opportunities to the unemployed rural youths.

Keywords: Early larval stages, *Trichgaster lalius*, control condition

1. Introduction
Gouramis commonly known as lybyrinth fishes belong to the suborder *Anabantoidei*. The dwarf Gourami, *Trichogaster lalius* is a peaceful freshwater fish which have both ornamental as well as food values. In West-Bengal, the drawf gourami is locally known by ‘Kholisa or LalKholse’ distributed in Bangladesh, India and Pakistan. It is a hardy fish and can breathe air from the surface with aid of an accessory air-breathing organ, the labyrinth (Sutradhar *et al.* 2016) [26].

The fish is commonly inhabited in freshwater pools, ditches, ponds, wetlands and marshes as well as rivers and lakes with vegetation (Hayakawa and Kobayashi 2010a) [13].

The sexually mature male is sparkling; translucent brightly blue coloured with red or dark orange stripes in the body and the female is less colorful but has an attractive silvery body. Its bright lucrative color and the ease with which the dwarf gourami adapts to captivity make it a popular aquarium fish (Hayakawa and Kobayashi 2010b) [14].

The trade of indigenous ornamental fishes is highly unorganized and mainly based on natural collection. The fishes are collected by local fishermen and marketed by traders who actually control the activities. Besides these, indiscrimination exploitation from natural sources leads to extinction of some of the rare varieties of some indigenous ornamental fishes and declination in number of others (Tarali and Deka, 2013) [27].

The stock assessment of these species is immediately required and species identification particularly in larval and juvenile stages is important. Hence, accumulation of early life-history information is essential, including morphological descriptions of indigenous species (Ruber *et al.* 2006) [25].

The descriptions of early life-stage morphology should provide significant new information for rearing of this species. The present study was carried out to observe the early larval development stages of *Trichogaster lalius* in control condition. The detail morphology of larvae and juveniles are described in from a series of laboratory-reared specimens.

2. Materials and Methods
2.1 Site of experiment
The present study was carried out in the laboratory of department of Fisheries Resource Management, Faculty of Fishery Sciences, West-Bengal University of Animal and Fishery Sciences, Chakgaria, Kolkata.

2.2 Natural breeding

Ten set of experiment were conducted by maintaining male and female (1:1 ratio) in the breeding tank (30×30×30 cm) filled with tap water and sufficient aeration to facilitate nest building by male. All the aquariums were decorated with floating weed Hydrilla (*Hydrilla verticulata*) along with thermocol plate to provide them natural breeding environment as well as to make their own bubble nest.

2.3 Study of larval development

The larval samples were collected from the larval rearing tank a day interval to study their development such as morphology and their behavior. The larval development was observed up to ten days and photograph was taken whenever necessary. At least five larvae were collected and immediately put into 70% ethanol for further study. During these whole study period water quality parameters like water temperature, pH, Dissolved Oxygen, Alkalinity, Hardness was monitored using the standard methodology (APHA, 2010) [4]**.**

3. Results and Discussion

3.1 Water quality parameters of rearing tank

The water quality parameters during the study period are depicted in Table 1.

Table 1: Water quality parameters recorded during larval stages of *Trichogaster lalius*

Parameters	Months		Mean Value (±SD)
	May	June	
Water temperature (°C)	29.03±0.03	30.01±0.06	29.52±0.04
pH	7.22±0.21	7.23±0.24	7.25 ± 0.22
DO (mg l^{-1})	6.02±0.11	6.18±0.14	6.10 ± 0.12
Total alkalinity (mg l^{-1})	137.51±5.95	137.10±6.21	137.30±5.90
Hardness (mg l^{-1})	465.66±24.05	466.12±25.01	465.89±24.53

The water quality parameters during the study period were found to be optimum for larval development, except hardness. In the present study, the mean temperature was found 29.52±0.04 which was found to be suitable for larval rearing. Marilyn (1993) [21] reported that the optimum temperature of 28-30 ºC support the normal growth of fishes in India which was similar to our present study.

Rinna *et al.* (2014) [24] reported the same observation in case of *Trichogaster trichopterus.* Kumar (1993) [19] has reported that the water temperature ranged between 20-30º C is ideal for aquaculture practices in India. The mean pH value 7.25 ± 0.22 was very much effective for the larval rearing which is supported by Rinna *et al.* (2014) [24] for *Trichogaster trichopterus.* Kumar (1993) [19] reported that water quality management in aquaculture the preferable pH was 6.8- 8.6 and slightly alkaline pH is preferable in India. The above reason could be a factor for larval survival. Goodwin (2001) [12] has suggested that *Carassius auratus* preferred pH of water is 6.0-8.0 and pH 7.0 for widow tetra (*Gymnocorymbus ternetzi*). Lee and Harney (1999) [20] found that much lower level of pH can reduce the survival rate of fish.

In this study, mean dissolved oxygen level was observed to be 6.10±0.12 mg l^{-1}. The present findings coincide with the report of Gautam and Gautam (2005) [11] where feed influence the quality of water, and also he reported that the dissolved oxygen level varied depending on the polluted nature of water. Regarding the dissolved oxygen concentration in water, Bindu *et al.* (2014) [9] noticed that the survival rate of larvae was maximum at oxygen level from 3-9 mg l^{-1}. Molokwu and Okpokwasili (2002) [22] recommended that the ideal range of total alkalinity should be 60-300 mg l^{-1} which was within the recommended limit. They also reported that higher larval growth and survival in *Rhamdia quelen* (*Siluriformes, Pimelodidae*) was obtained at 30 and 70 mg l^{-1} $CaCo_3$ for best hatching rate, high viability and maximum larval survival. But in the present study hardness was observed little higher (461-471 mg l^{-1}), which reduced the survival rate of the larvae.

3.2 Larval development

Yolk size was totally absorbed by the 4th day and they were fed with laboratory prepared green water containing micro algae, mostly chlorella up to 10th days. From 10th day onwards the hatchlings were fed on laboratory prepared pellet feed and *Tubifex* worm. The different characters of larval developmental stages were elaborated in Table 2, from the newly born larvae to 10th day old fry of *Trichogaster lalius.*

Table 2: Larval development stages of *Trichogaster lalius* in control condition

Days after hatching	Morphological description of the larvae
Newly hatched larvae	Larvae will remain motionless in the surface of water with black eyes. Mouth and gills were developing slowly but pulsing of heart is seen on the anterior side of the body. Lateral line was starting to develop and pigmentation extended throughout the yolk sac.
1 day old larvae	Yolk size reduced due to absorption of nutrients. Mouth just started moving, oil granules were visible, chromatophores were well grown throughout the body (Fig. 1, a).
2 day old larvae	Larvae size increased, yolk sac reduced and three lobed patterns of yolk, mouth and nostril were well developed (Fig. 1, b).
3 day old larvae	Eyes are pigmented, larvae able to settle at the bottom, size of oil globule reduced, pectoral movement started, larvae started to eat from exterior particularly planktons, mouth become stronger (Fig. 1, c).
4 day old larvae	Melanophores are started to appear on jaws. Folding of pectoral fin was seen in larvae.Yolk totally absorbed and larvae started to feed external feed (Fig. 1, d).
5 day old larvae	Larvae size further increased, chromatophores are aggregated on the body surface, caudal fin started to develop, dorsal and anal fin with fin rays, well develop in both the jaws (Fig. 1, e).
6 day old larvae	Myomeres was seen, no yolk found, jaws became stronger, intestine is well developed. Caudal fin rays and pectoral fin buds appear, pigmentation founds over the body (Fig. 1, f).
7 day old larvae	The size of yolk sac is reduced and pushed upwards. The notochord becomes segmented and the upward extension of the hinder end is more pronounced giving the caudal region a heterocercal condition. The rudiments of the caudal rays are formed. The neural and haemal arches can also been seen (Fig. 1, g).
8 day old larvae	With the increasing development of musculature and bones, especially in the head, the larvae were becoming thick and opaque. The chromatophores become numerous and prominent all over the body. The air bladder is seen

	developing as an evagination from the dorsal aspect of anterior part of the gut (Fig. 1, h).
9 day old larvae	The media fin-folds were gradually diminishing in size. The caudal fin is well defined assuming a homocercal condition and the caudal rays are formed (Fig. 1, i).
10 day old larvae	Larvae were gradually growing in bulk. The yolk sac region now presents the normal contour of the body. The heart become thick and occupies the normal position in the thorax. The dorsal and ventral fins were now distinct from the caudal and have developed rays. The larvae finally developed to fry (Fig. 1, j).

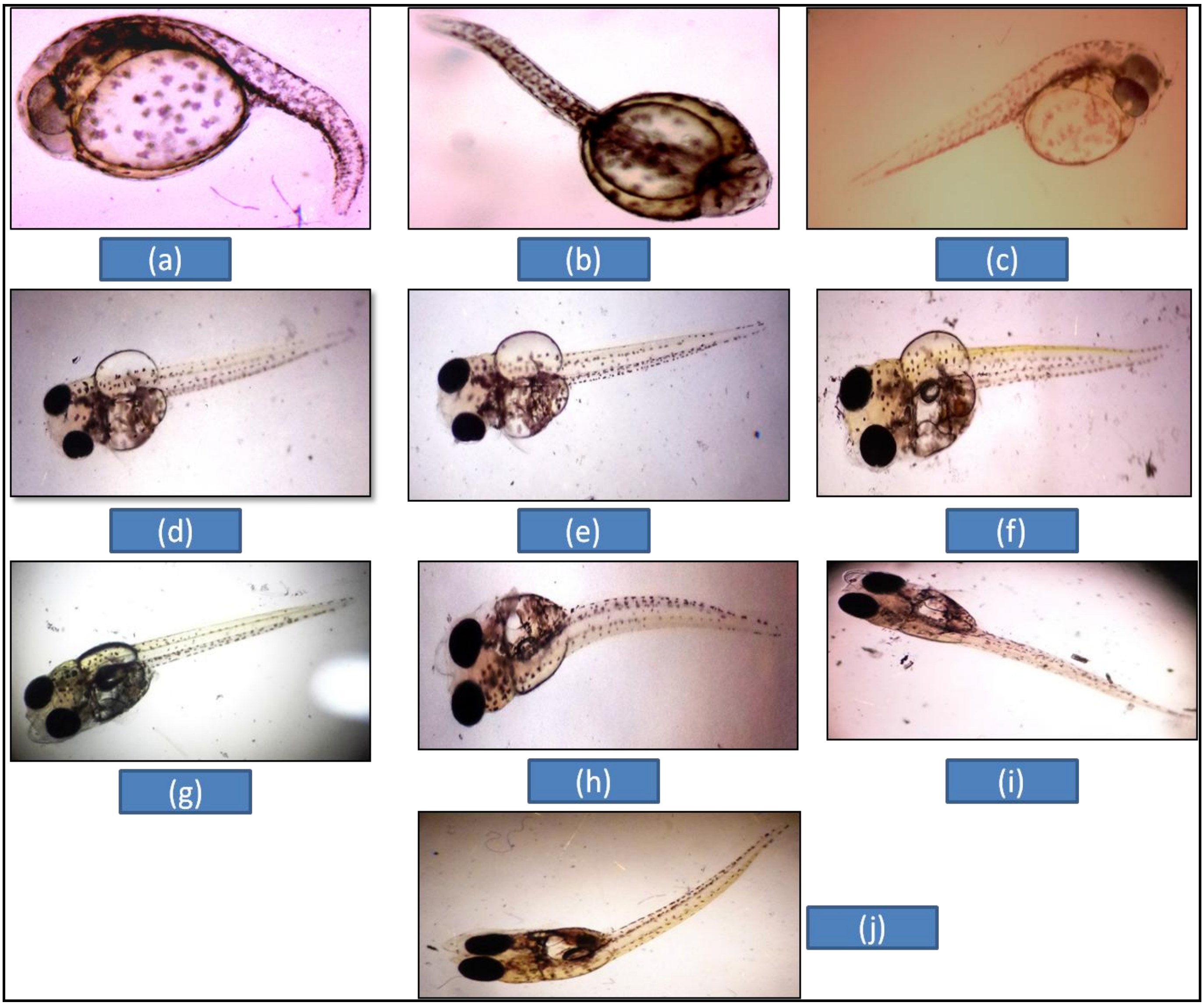

Fig 1: Larval development stages of *T. lalius* (a) 1st day old hatchling,(b) 2nd day old hatchling, (c) 3rd day old hatchling, (d) 4th day old hatchling, (e) 5th day old hatchling, (f) 6th day old hatchling,(g)7th day old hatchling, (h) 8th day old hatchling, (i) 9th day old hatchling, (j) 10th day old hatchling.

The yolk sac of newly hatched larvae of *Trichogaster lalius* totally absorbed at 4th day of hatching which was similar to several related work for different fishes. Amornsakun *et al.* (1997) [3] reported that the yolk absorption of larval green catfish *Mystus nemurus* was complete at 3 days after hatching. Amornsakun (1999c) [2] reported that the yolk absorption of larval red-tail catfish, *Mystus wyckioides* was complete at 4.3 days after hatching and at 3.4 days for larval sand goby, *Oxyeleotrism armoratus*, after hatching (Amornsakun *et al.* 2002) [1]. Houde *et al.* (1976) [16] also reported the yolk absorption of larval white mullet *Mugil curema* was complete at 3.5 days after hatching. The yolk of milkfish *Chanos chanos*, was completely absorbed in about 2.5 dayold larvae (Chaudhuri *et al.* 1978) [10]. The yolk absorption of larval freshwater catfish, *Clarias sp.* was completed 3-4 days after hatching (Tarnchalanukit *et al.* 1982) [28].

The larvae of rabbitfish, *Siganus guttatus*, have rapid development of the eye, mouth and alimentary tract during the yolk-sac stage which makes it possible for the larvae to feed before the yolk is completely absorbed (Bagarinao, 1986) [5]. Morphological investigations of the jaw and the digestive tract showed that larval cod, *Gadus morhua*, is able to absorb ingested food well before exhaustion of the yolk sac (Kjorsvik *et al.* 1991) [18].

In the present study the larval development was same as described by Hayakawa and Kobayashi (2012) [15] in the dwarf Gourami *Colisa lalia;* they reported that on day 2, the yolk-sac occupied most part of the body cavity. The operculum was clearly observed. On day 3, the yolk sac still occupied most of the body cavity. The mouth was confirmed to open and move by this day, but larvae did not consume exogenous food. Eyes began to show pigmentation.

Melanophore series were observed on the lateral side and along the dorsal and ventral finfold. On day 5, myomeres began to be clearly observed. The yolk sac still occupied a large part of the body cavity but larvae began to take food. Larvae actively swam by moving fin fold pectoral fins, and larvae remaining in the aquarium left the nest and moved to bottom of the aquarium. Relatively large stellate melanophores were observed on the head and ventral regions, and small stellate melanophores were also observed on the posterior region of trunk.

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Several authors were also studied different morphological larval development stages with respective fish species, Yurembum (2015) [29] in *Trichogaster labiosa*; Morioka *et al.* (2010) [23] in *Trcichogaster pectoralis*; Hayakawa and Kobayashi (2012) [15] in *Colisa lalia*; Bhimachar *et al.* (1944) [8] in *Osphronemus gourami*; Barman *et al.* (2013) [6] in *Colisa fasciatus.* The present finding was resembles with the earlier studies.

4. Conclusion

The success of this study will be useful for the ornamental fish breeders, aquarium keepers and further study on genetic characterization and the survivability of the larvae and fry for persistence of colour by feeding of different natural flora will conserve this species in natural environment.

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