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Induced Breeding of Freshwater Goby, *Glossogobius giuris* (Hamilton, 1822) in the Captivity: A Preliminary Study

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ABSTRACT

Glossogobius giuris (freshwater goby) is widely distributed and captured species in Bangladesh. Recent times, the species is highly demanded and expensive fish item but not yet reported to culture though decreased in capture. Present study was the first trial to induce propagation of *G. giuris* in the captivity using different hormones. There were four treatments conducted such as (T₁) Natural breeding in aquarium without hormones, (T₂) Induce breeding using Ovapirim, (T₃) Induce using HCG and (T₄) Induce using PG. Each treatment had one more replication. In each aquarium, 4 pairs of broods (sex ratio 1:1) were stocked. The ovapirim, HCG and PG double dose were injected for female and single dose for male in intramuscular. The water parameters such as pH, temperature and DO measured daily which recorded as 7.49-8.66, 27.00-30.93°C and 3.80-4.47 mg L⁻¹ respectively. Among the four treatments, highest spawn success (75% pairs) showed in treatment T₄ followed to T₃ and T₁. Spawning occurred within 48 h of injection of 2nd dose and the spawning duration recorded up to 30 min. The eggs were started to hatch within 36 h of spawning which completed (highest 80%) within 72 h. The fecundity of the species recorded 8050-10070. The results showed that induce breeding of *G. giuris* is possible either with HCG or PG hormones. However, further studies are suggested to confirm the findings as well as optimization of the hormone doses.

Key words: *Glossogobius giuris*, induce breeding, breeding behavior, spawning

INTRODUCTION

Gobiidae is the second largest teleost family after Cyprinidae and contains about 212 genera and approximately 1875 species (Nelson, 1994). The goby fishes occurs in tropical and subtropical fresh, brackish and marine waters along the East coast of Africa, Southern Asia, South-East Asia with its associated Islands, as well as Northern Australia. This species is most often associated with estuarine habitats, although, it is also found in marine water and can be found many kilometers inland in freshwater streams.

The species *Glossogobius giuris* (locally known as 'Bele') is widely distributed in the freshwater and estuaries of Bangladesh, India, Pakistan, Myanmar and Far East (Doha, 1974). In Bangladesh it is commonly found in the river, estuaries, haors, baors, beels, ponds and swamps. The inland water areas of the country comprise 1.03 million ha of rivers, canals and estuaries, 114,161 ha of

natural depressions such as the 'beels' and 'haors', 161,943 ha of ponds and tanks, 5,488 ha of ox-bow lakes, 2.8 million ha of flood plains and 87,300 ha of brackish water aquafirms (Rahman, 1989).

In Bangladesh about 80% population is poor and they depend on small size fish for their daily supply of animal protein as they are available at reasonable price (Siddiqui, 1985). Fish is the main source of animal protein in the diet of the people of Bangladesh because 80% of the animal protein in our diet comes from fish alone. Ahmed *et al.* (1984) reported on the biochemical composition of gobi fishes and showed as high protein-low fat in the flesh of *G. giuris*. This is important species for capture fishery in our country (Costa *et al.*, 1999) but no culture practice introduce yet due to the lack of fry availability.

The meajority of the goby fishes, being small in size, do not constitute an important fishery anywhere in Bangladesh but *G. giuris* which attains about a foot in length, is notable, as it form a fishery of some magnitude in the southern part of the country. *G. giuris* is very important food fish, specially to the low middle class and poor people, because of being comparatively cheaper but sometimes very expensive to them (Islam, 2004). However, the high demand in the present market and higher market values raise question whether the bele fish is poor or middle class family food or higher class family food. Now a days, it became out of buying capacity for the middle class and poor people. Anyway, there is no doubt about the needed to meet the high demand and thus we have to switch from capture to culture practice of the species. So, research details on this species specially on culture aspect and hence the improve breeding technology to ensure available supply of fry is the demand of time. In addition, though the species not in the redlist by the IUCN (probably due to lack of information), the species not in such available in our capture composition from the earlier time.

There were some breeding experiemnt have done on the other goby fishes in some countries. For example, induced breeding and early development of the Marble Goby (Tan and Lam, 1973); care and breeding of Peacock goby (aquaticcommunity.org); breeding Bumblebee goby (aquaticcommunity.org); breeding spotted gudgeon (aquaticcommunity.org); breeding ground profile of food fish species (Sarkar and Banerjee, 2010).

However, there are some research on biological and ecological aspect of goby fishes have done in our country and abroad such as habitat distribution (Doha, 1974; Allen, 1991; Talwar and Jhingran, 1992; Rainboth, 1996), food and feeding habit (Alikunhi *et al.*, 1951; Marquez, 1960; Bhowmick, 1965; David and Rajagopal, 1975; Doha, 1974; Bhuiyan and Haque, 1984; Islam, 2002a) and reproductive biology (Hora, 1935; Alikunhi *et al.*, 1951; Marquez, 1960; Bhowmick, 1965; Breder and Rosen, 1966; Doha, 1974; Fatema *et al.*, 1997; Pankhurst, 1998; Kovacic, 2007; Hajji *et al.*, 2013).

Thus, the present study was the first ever attempt to develop breeding technique of the species. The study focused on the details breeding aspects such as natural breeding performance in crol system, induced breeding performance in aquarium by the using of different doses of Ovaprim, Pituitary Gland (PG) and Human Chorionic Gonadotropin (HCG) hormones.

MATERIALS AND METHODS

Study site: The brood stock was reared in the newly excavated pond complex in the campus of Jessore Science and Technology University, Jessore. Natural and induce breeding performance was conducted in the laboratory glass aquarium of the Department of Fisheries and Marine Bioscience of the same university.

Table 1: Breeding performance and survival (%) of different experimental design for the induced breeding of *Glossogobius giuris* using different hormone dose, stocking density and performance

Designed	Treatment T ₁	Treatment T ₂		Treatment T ₃		Treatment T ₄	
-----	-----	-----	-----	-----	-----	-----	-----
Natural	Natural	Female	Male	Female	Male	Female	Male
Ovaprim injection							
1st dose		0.2 mL kg ⁻¹	N/A				
2nd dose		0.2 mL kg ⁻¹	0.1 mL kg ⁻¹				
3rd dose		0.3 mL kg ⁻¹	0.2 mL kg ⁻¹				
HCG injection							
1st dose				400I U kg ⁻¹	N/A		
2nd dose				400I U kg ⁻¹	400I U kg ⁻¹		
PG injection							
1st dose						1.5 mg kg ⁻¹	N/A
2nd dose						2.0 mg kg ⁻¹	2.0 mg kg ⁻¹
Stocking density	4 pairs = 8	4	4	4	4	4	4
(No./Aquarium)							
Performance	Spawn 25%		No response		Spawn 50%		Spawn 75%
Survival (%)	87		75		87		100

Experimental design: The experiment was separated in to two distinguished ways; (i) Natural breeding in the crol system and (ii) Induced breeding. There were four pairs of brood goby stocked in the crol aquarium for the observation of natural breeding of the species which was treated as T₁. For the induced breeding; ovaprim, HCG and PG were injected which was treated as T₂, T₃ and T₄ respectively and stocked in different aquarium to observe the breeding performance (Table 1). The each treatment had one more replication.

Collection and rearing of brood fish: The brood fishes of *Glossogobius giuris* were collected from nearby Baor (Monirampur, Jessore) and species was identified according to the reference books; Inland Fishes (Talwar and Jhingran, 1992). The average length and weight of the species were 9.3 cm and was 7.5 g, respectively. Fishes were reared about one month to develop their gonad properly. Fishes were feed with pellet feed daily at the rate of 5% of body weight. Broods were transferred to the aquarium to induce when gonad developed properly. Before injection of hormones, broods were kept one week in the aquarium for the adjustment in the captive condition.

Preparation of aquarium and stocking of brood fish: There were eight aquariums for the four treatments with size of 3×2 feet each. Some sand-stone layer prepared at the bottom in each aquarium. Lime was applied at the rate of 10 g tank⁻¹ to treat water. The water hyacinth was placed in each tank to provide shelter. In addition, Bamboo holes (Fig. 2b inset) was used in each tank for the shelter and also assumed to hatching substrate of the species. Four pairs of brood fish were stocked in each aquarium tank.

Induced technique: The 1st dose of hormone (Table 1) was injected at the base of pectoral fin and the 2nd and 3rd dose (when necessary) on the base of the dorsal fin to avoid the injury. The injected species released directly to the experimental tank. At treatment T₁, tubifex were supplied up to from the very first day of stocking. In other three treatments (T₂, T₃ and T₄) there were no food provided in first day of injection but later tubifex were provided twice daily.

Fecundity estimation: The fecundity of the species counted in two different ways. The ovarian developed female were dissected and counted the fecundity as the following equation:

$$F = \frac{\text{Total ground weight} \times N}{\text{Weight of small portion of total ground}}$$

Here, N is No. of eggs.

In addition, as the fishes spawned on the glass wall, the numbers of egg were counted manually by the plotting of the spread eggs.

Water quality measurement: The water quality parameters such as pH, Temperature and DO measured daily. The water exchange daily at the rate of 30-40% of the reservoir. Siphoning also performed to remove dust from the bottom of the tank daily.

RESULTS

Water quality: Water parameters such as pH, temperature and DO were measured daily in different treatments which are shown in Fig. 1. The water pH, temperature and DO ranged were 7.49-8.66, 27.0-30.93°C and 3.80-4.47 mg L⁻¹, respectively in different treatment during the experiment. There were no significant differences ($p < 0.05$) in water parameters in different treatments. However, a sudden water temperature fallen for few degrees on 8th day due to may be heavy rainfall.

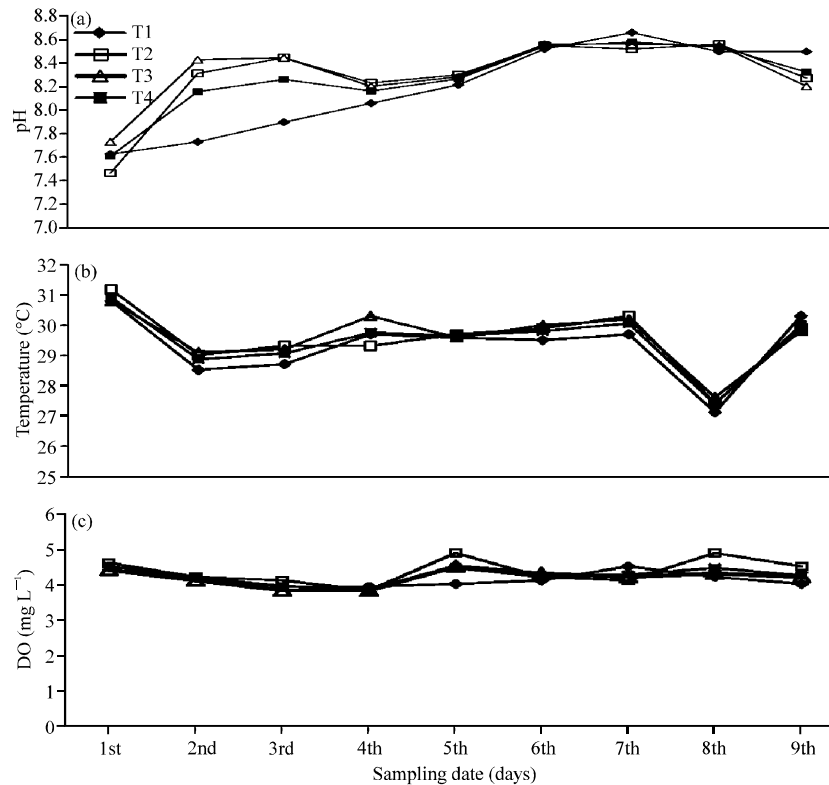


Fig. 1(a-c): Water quality parameters (a) pH, (b) Temperature (°C) and (c) Dissolved oxygen (mg L⁻¹) during induced breeding of *G. giuris* in different experimental aquariums

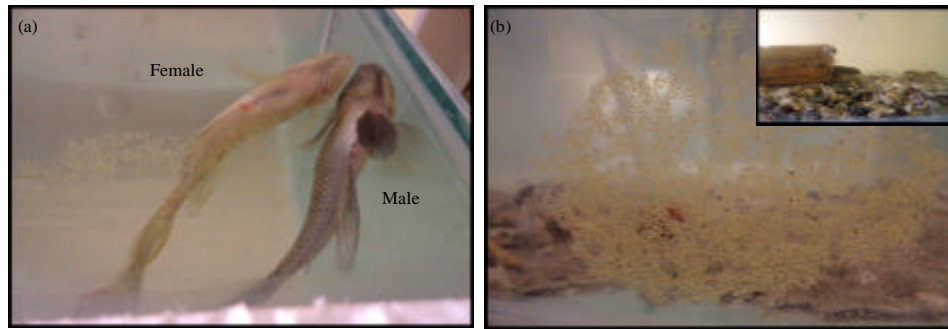


Fig. 2(a-b): Breeding behavior of *Glossogobius giuris*: (a) Female laying eggs on the aquarium glass wall and male agitating and guarding female during the induce breeding in the captive condition and (b) Spawn spread eggs on the glass wall of aquarium. Inset: Bamboo holes (shelter for the fishes)

Breeding behavior: The stock fishes were seems to make pair though it was not much clear about the exact partner at the beginning of pairing. They used to pass most of the time inside the bamboo shelter which was provided within the aquarium. However, when feed provided all of them actively took part in taking food. After the 2nd dose of injection (mostly for HCG and PG), the most pair noticed moving vigorously and showing crazy behave. After around 24 h of 2nd dose, the female selected a shadow wall side and moved around purposely. The male started to nip on female as well as guarding to protect from other fishes. After certain time, they come close together and move slowly (Fig. 2a). They also placed ventral to ventral as well as lateral to lateral with very affectionately. The female come on the wall side and spawn in a small batch on the wall. After each batch of spawn, it moved a bit for few seconds, come close to male and come back again and spawns another batch. At the same way, they complete spawn within about 30 min. After spawn, the pair showed parenting and guarding the eggs. However, for the safety of eggs, the broods were kept away from the aquarium.

Breeding performance: The breeding performances of four treatments are shown in Table 1. Among the four treatments, spawning performance showed of a single fish in T_1 within 10 days of stocking. However, within about 14 h of spawn, eggs were disappeared from the side wall of the aquarium. Later, about one mh cinuous culture period no spawn showed in natural treatment.

In treatment T_2 , induced with ovaprim did not showed any breeding performance even applied 3rd dose of injection and hence could not identified any specific reasons. In addition, highest (25%) broods died in this treatment. As no performance in both replications of ovaprim induced, it might be said that ovaprim is not suit for the species to influence the breeding.

Induced with HCG (T_3) were showed better breeding performance than T_1 and T_2 . There were 50% pair spawn within 48 h of injection of 2nd dose. However, the highest spawn (75%) and survivility (100%) showed in T_4 which treated with PG (Table 1). In case of PG hormone, the species also spawned within 48 h of injection of 2nd dose.

Spawning and hatching: The fishes were spawned on the side wall glass of the aquarium (Fig. 2b). Though there were shelter provided in each treatments and observed to take the shelter as nest for a pair but they did not spawned within the shelter. The eggs were started to hatch

Table 2: Hatching performance (mean %) of *Glossogobius giuris* in aquarium tank after induced with different hormones

Hours of spawned (h)	Hatch (%)
36	25
52	60
60	75
72	80

within 48 h which completed within 72 h (Table 2). It was noticed that there were about 80% spawned eggs hatched and remaining were lost. After the spawn, the brooders were kept away from the aquarium. Though there was no water flow or circulation, it was maintained aeration continuously around the egg mass.

Fecundity: Fecundity per spawn, based on number of eggs deposited on each substrate per collection as well as dissected ova count, varied between 8050 and 10070 eggs with an average 9050 ± 718 . Eggs were translucent, embedded in circular, jelly-like, spread as masses with an average area of 200-300 cm².

DISCUSSION

Water quality: The water parameters were comparable to other researchers who have done induced of other gobies in different regions. The marble goby, *Oxyeleotris marmorata* hatch within 2-5 days after fertilization at mean temperature $27 \pm 1^\circ\text{C}$ Tan and Lam (1973) and Ooi (2004) reported $27-32^\circ\text{C}$ temperature is suitable for the breeding of marble goby (*O. marmoratus*). Tavarutmaneegul and Lin (1988) reported temperature, pH and DO ranges are $26-30^\circ\text{C}$, 5.8-9 and $4.5-12 \text{ mg L}^{-1}$, respectively for the breeding of sand goby. In the natural system, it is found that food fishes are breed successfully where the temperature, pH and DO ranges are $19-30^\circ\text{C}$, 6-8.5 and $3-4.5 \text{ mg L}^{-1}$, respectively (Sarkar and Banerjee, 2010).

Breeding behavior: Like most of the fishes, *G. giuris* female showed larger in size than males. Doha (1974) observed sexual dimorphism in *G. giuris* after the species attaining a total length of 60 mm. In addition, different coloration was found in sexes of the species. In the present study, pelvic fins of the maturing and mature males were darker in color than those of the females (Fig. 2a) while vice versa reported by Doha (1974). However, the same black colored formed in males during mated in case of round goby (*Neogobius melanostomus*) whereas females are mottled, mimicking colors of bottom substrates (Wickett and Corkum, 1998).

In the present study, it was showed that the species formed a pair and might not promiscuous. On the other hand, round goby showed polygynous mating system in which many reproductive females deposit eggs in the nests of a single male (MacInnis and Corkum, 2000). The pair moved closely and they kept separate themselves than others in the captivity. They laid their eggs on the substrate (glass wall) rather than bamboo hole. Thus it may be said that freshwater goby like to spawn on the clean substrate. The same also reported for the sand goby, *Oxyeleotris marmorata* (Tavarutmaneegul and Lin, 1988).

The pair forming, guarding of male to female is more or less similar to other goby fishes round goby (Corkum *et al.*, 1998) desert goby (Symons *et al.*, 2011). In most gobiids have a reproductive strategy that is characterized by male parental care of eggs (maintaining oxygen levels by fanning eggs) egg or nest defense particularly aggressive behavior towards intruders or outsiders

(Corkum *et al.*, 1998). Most of the fishes showed stayed inside the bamboo holes or dark corner of the aquarium glass. This behavior also noticed by Natalie (2009) and Symons *et al.* (2011) in desert goby (*Chlamydogobius eremius*); by Iwata *et al.* (2001) in fresh water goby (*Micropercops swinhonis*) in the aquarium. After spawn, the pair showed parenting, fanning (such as desert goby, Natalie, 2009) and guarding the eggs. Indeed, during the spawning males of most gobiidi species guard nest and/or female (Hajji *et al.*, 2013).

In the treatment T₁, eggs were feed by the parents and/or other broods. It was not possible to notice whether there was any relationship of feeding eggs to sex. The eggs feeding behave also found in other gobies such as round goby (*Neogobius melanostomus*) those are poses a great threat by feeding of eggs of their native fishes (Steinhart *et al.*, 2004). However, cannibalism is a frequent occurrence in the freshwater goby *G. giuris* (Bhuiyan and Haque, 1984; Islam, 2002b) as well as many other freshwater fishes when the young ones are abundant such as *Anabas testudineus*, *Clariias batrachus*, *Notopterus chitala* and *Channa punctatus* etc. (Mookerjee, 1944; Javid, 1970). Though the pair showed guarding the eggs, however, it could be eaten by the other pair of goby. Alikunhi *et al.* (1951), Bhowmick (1965) and Doha (1974) reported the cannibalistic habit of *G. guiris* but they also did not show any relationship of cannibalism in relation to its size and sex. Environmental parameters i.e., day length, temperature, turbidity, availability and nature of food, inter-specific and intra-specific social relations may influence the nature and patterns of reproduction and development behavior in aquatic animals (Waterman, 1961).

Breeding performance: Among the four treatments, T4 i.e., induced by PG hormone showed the better survival (100%) and maximum spawning (75%), thus PG could be the best hormones for the induced breeding of the species. HCG could also be used for inducing of the species which reported to use by Tan and Lam (1973) to ovulate marble goby, *Oxyeleotris marmorata*. Though, ovaprim did not show stimulation to ovulate of the species, further study required if the hormone was bad in quality or really it not fit for the species. However, in case of fecundity and hatch of the eggs, there were no differences noticed among the different hormones injected.

There were about 80% hatched in all treatments within about 72 h of spwan and rest of them lost and attached with the glass wall. As the literatures are rare for the species, it may comparable to other goby species. In case of marble goby, *Oxyeleotris marmorata*, 90% hatching reported within 2-5 days after fertilization which was induced to ovulate using HCG (Tan and Lam, 1973). In sand goby, *Oxyeleotris marmorata*, 80% hatching within 2-4 days after fertilization (Tavarutmaneegul and Lin, 1988).

Fecundity: The findings fecundity (mean 9050±718) of *G. giuris* in the present study was a bit small in number compare to Bhowmick (1965) and Doha (1974) who were stated fecundity ranged 10,760-29,580. However, the variations of fecundity is very common in fishes and the number of eggs produced by an individual is depend on various factors, like size, age, types of species of the samples and their ecological habitats including food availability (Moyle and Cech, 2000; Lagler, 1956). In back goby (*Gobius niger*), fecundity range found 4280-14000 eggs fish⁻¹ with an average 8522±689 (Hajji *et al.*, 2013), in sand goby (*Oxyeleotris marmorata*) fecundity ranged 2000-30,000 eggs individual⁻¹ with an average 24,000 (Tavarutmaneegul and Lin, 1988) and in stripped goby (*G. vittatus*) it was 560-3045 with an average of 1426±89 eggs fish⁻¹ (Kovacic, 2007).

CONCLUSION

The induced breeding of *Glossogobius giuris* is possible in the captivity using hormones of PG and HCG. However, further study is suggested to confirmation of the findings as well as for the optimization of the hormone doses.

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