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Captive breeding of Striped Spiny Eel, *Mastacembelus pancalus* (Hamilton, 1822) considering the various hormonal responses

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Abstract
The spawning performance of spiny eel, *M. pancalus* was investigated by using different hormones. The water quality parameters were within optimum ranges such as average temperature, DO and pH were 29.13 ºC, 4.19 mg/L and 8.32 respectively. Hormones were administered into females in two doses; a lower priming dose followed a 6 h interval by a higher resolving dose. In first dose, the female were injected ovaprim, HCG and PG at 0.25 ml/kg, 200 IU/kg and 30 mg/kg of body weight respectively. The second dose was applied to both male and female and female received double amount of hormone of the first dose. The male were administered a single dose at 0.5 ml/kg, 400 IU/kg and 60 mg/kg of ovaprim, HCG and PG respectively. It is indicated that the use of ovaprim was more effective in spawning (100%), fertilization (75%) and hatching (55%) of *M. pancalus* compared to the other stimulators.

Keywords: *Mastacembelus pancalus*, induced breeding, hormones, spawning

1. Introduction
The Striped spiny eel (*Mastacembelus pancalus*) is one of the common species of fishes of the family Mastacembelidae and important eel fish in Bangladesh with great demand as a good traditional table fish [1]. Locally the fish is called guchi, baim, turi or chirka. However, very commonly known as guchi baim. The species is distributed in India, Pakistan, Bangladesh [22] and Nepal [8]. Guchi baim was available in rivers, canals, beels and inundated fields in the past throughout Bangladesh but presently it is under threat due to tremendous loss of natural habitat [14]. According to IUCN [10] the fish is considered to be a critically endangered in the country. The fish is not yet incorporated in inland culture system. The main reasons behind this are insufficient information on the captive induced breeding, inadequateness of natural fry and absence of commercial hatchery production. Considering the importance of the species, adequate research in this field is required. Notable works have been done on striped spiny eel such as Talwar and Jhingran [22] have studied on the age and growth; Karim and Hossain [11], Vettath *et al.* [23], Saha *et al.* [17] and Pathak *et al.* [12] have studied on the reproductive biology; Alam *et al.* [3] and Rahman *et al.* [14] have studied on the induced breeding of *M. pancalus* with different doses of PG; Das and Kalita [6] have studied on the induced captive breeding of *Macrognathus aculeatus* with different doses of ovaprim. But so far no works have been done on the induced breeding of *M. pancalus* with different hormones. It is necessary to undertake proper study to identify and characterized the breeding protocol of the fish. The research aim was to establish an induced breeding protocol considering the various hormonal responses of *M. pancalus*.

2. Materials and Methods
The major part of the experiment was conducted at the Laboratory, Department of Fisheries and Marine Bioscience, Jessore University of Science and Technology (JUST), Bangladesh. The experiment was done from 10 March to 30 June 2014.

2.1 Brood fish collection and rearing
Samples of *M. pancalus* for induced breeding were collected from Jhapa boar, Jessore and were kept in the previously prepared experimental pond of the university. For successful

induce breeding, proper rearing and maintenance of brood stock is a pre-requisite. Monthly manuring and fertilization was done with cow dung, urea and TSP at 5 kg/decimal, 250 g/decimal and 100 g/decimal respectively to stimulate the growth of natural feed. Liming was also performed whenever necessary at 250 g/decimal. Besides this, the fishes were fed daily with a mixture of mustard oil cake, rice bran, wheat bran, and vitamin premix at the ratio of 20:49:30:1 by weight at 5% of total body weight of fish.

2.2 Brood fish selection
The ripe and healthy male and female brood fish were selected based on physical and visual examination of secondary sexual characteristics i.e., size, color, swollen abdomen and genital openings. Males were comparatively small in size, developed with dark greenish color on the top and whitish in beneath. While mature females were comparatively large in size, greenish color on the top and yellowish in beneath with soft and swollen abdomen. Mature males and females from the brood rearing ponds were collected and kept in the laboratory aquarium.

2.3 Aquarium preparation
The aquarium was rectangular in shape and size was 36 inches x 15 inches x 14 inches. The aquariums were supported with sand, gravel and bamboo slits, used as their hiding place. A lid was used to prevent the fish from dying outside the tank. Filter inlets and outlets were secured with mosquito nets. Besides this, water hyacinths was supplied to hold the sticky eggs.

2.4 Experimental design
Three treatments were designed and considered as T_1, T_2 and T_3 for Ovaprim, HCG and PG respectively. For each treatment both one pair of male as well as female were used. The hormones were applied at two different doses. The primer dose for female was 0.25 ml/kg, 200 IU/kg and 30 mg/kg of Ovaprim, HCG and PG respectively. The seconddose of female was doubled of the first dose after the interval of 6 h. The male only received the second dose of hormones (Table 1).

2.5 Measurement of water quality
The water quality of the experimented aquarium were maintained and recorded carefully. Water parameters such as dissolved oxygen (DO), pH and water temperature were measure daily. A pH meter (EZoDO; Model-7200; Made in Taiwan) was used for the measurement of pH and temperature; a DO meter (LTLutron; Model- YK-22 DO; Made in Taiwan) was used for the measurement of DO.

2.6 Hormone administration
The hormones were administrated near the base of pectoral and dorsal fin at an angle of 45° with the body. Then the fishes were caught very carefully from the spawning tank and a piece of clean, soft and wet cloth was used to wrap up the fish. The amount of hormone for each fish was determined before according to the body weight of the broods. After the administration of hormones the fishes were kept in separate aquarium which supported a continuous air flow and at the sex ratio of 1:1 for breeding.

Table 1: Doses of hormone applied to male and female brood of *M. pancalus.*

Treatment	Hormone	Sex	Doses		Time interval
			1st	2nd	
T_1	Ovaprim (ml/kg)	Female	0.25	0.5	6h
		Male	-	0.5	
T_2	HCG (IU/kg)	Female	200	400	
		Male	-	400	
T_3	PG (mg/kg)	Female	30	60	
		Male	-	60	

2.7 Determination of fertilization and hatching rate
The eggs were examined to determine the fertilization rate. The *M. pancalus* laid their sticky eggs on the root of water hyacinths. Approximately 60 eggs with roots were placed in plastic bowls with three replications of each having water flow from porous PVC pipe and outlet facility. The fertilized eggs were transparent and unfertilized one became whitish. The eggs were observed under a magnifying glass and fertilized eggs were counted with the help of a soft thin brush. The fertilization rate was determined by using the following formula:

$$\text{Fertilization rate} = \frac{\text{No. of fertilized eggs}}{\text{Total no. of eggs (fertilized and unfertilized)}} \times 100$$

Some fertilized eggs were then kept in specially prepared small PVC jar. Hatching started after 32-34h of fertilization. After completion of hatching, the hatchings were collected in another jar from the experimental jar and counted by visual observation. Hatching rate was determined by using the following formula:

$$\text{Hatching rate} = \frac{\text{No. of hatchlings}}{\text{Total no. of fertilized eggs}} \times 100$$

2.8 Statistical analysis
The results found in the study were subjected to statistical analysis, chi-square test that showed the significance (P<0.05) level of differences between the treatments. This statistical analysis was performed with the aid of the Microsoft Excel 2010.

3. Results
The results were varied significantly in variation with the different hormones in this experiment. Beside this, the variation also depend on the broods size and maturation, season of inducing, aquarium condition etc.

3.1 Maintenance of brood fishes
The brood fishes of *M. pancalus were* reared for almost three months in the experiment pond. Through the proper broodstock management, the broods were found to be healthy, fully mature and are ready to spawn. It was found that spawning period was from June to August.

3.2 Water quality
The average water quality parameters of the experimental aquarium are given in table 2.The physico-chemical condition such as temperature, dissolved oxygen and pH of water in experimental aquarium under different treatments of *M. pancalus* ranged between 27 to 29 ºC of temperature, 3.5 to 5 mg/L of DO and 7.2 to 8.5 of pH respectively with negligible variation. The mean values of water parameters were not

significantly (P<0.05) different among the treatments.

Table 2: Average water quality parameters of different treatment aquarium of *M. pancalus*

Parameter	Treatment		
	T_1	T_2	T_3
Temperature	29.02 ± 0.94[a]	29.16 ± 0.93[a]	29.21 ± 0.88[a]
P^H	8.19 ± 0.42[b]	8.34 ± 0.43[b]	8.42 ± 0.42[b]
DO	4.17 ± 0.27[c]	4.23 ± 0.37[c]	4.17 ± 0.25[c]

(Mean ± SE); values of the parameter in each column with different superscripts (a, b, c) differ significantly (P<0.05)

3.3 Breeding behavior and spawning

Spawning took place inside the breeding tank within 33 to 35 h after administration of second dose of hormone. Courtship began within 29-32h and took place for about 2-4 h. They swim in a tight circle (Fig.1) and the male chased the female with its caudal fin (Fig.1, inset).Later, the pair encircles around the roots of the water hyacinths for spawning. The female laid their sticky and adhesive eggs to the roots of the water hyacinths and immediately the male released sperm to fertilized them (Fig. 2). After fertilization both male and female were hide them into the plastic jar that were kept in the aquarium.

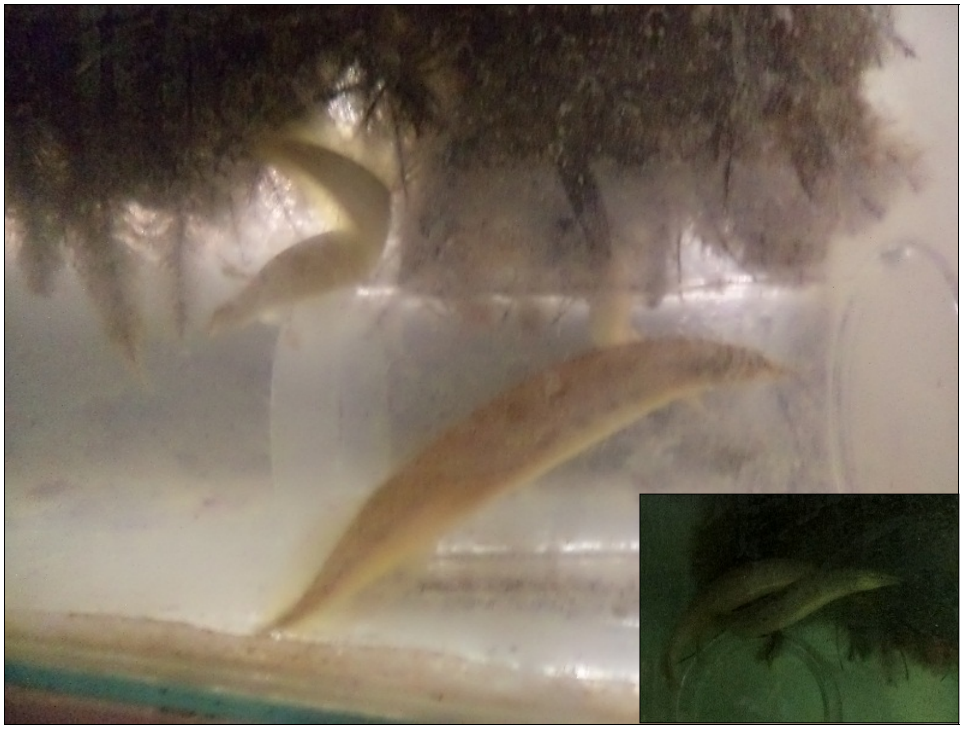

Fig 1. Courtship behavior: swim at circle and chased with caudal fin (inset) of *M. pancalus*

Fig 2. *M. pancalus* laying eggs at the root of the water hyacinths.

3.4 Breeding performance

The breeding performance varied with different hormones. The fertilization and hatching percentages were higher in T_1 rather than T_2 and T_3. The T_1 showed 100% spawning, 75% fertilization and 55% hatching performance (Table 3). This results was significant at 0.1% level (df=1, x^2=5.33, P>0.05). Hatching completed after 33-35 h from the fertilization period. Hatching rate of fertilized eggs was obtained at 55%, 46%, and 26% respectively in treatment T_1, T_2 and T_3.

Table 3: Responses of *M. pancalus* with different treatments.

Treatment	Stocking density (No./Aquaria)	Spawned performance (%)	Brood dead after spawn (%)	Hours of spawned	Fertilization rate (%)	Hours of hatched	Hatching rate (%)
T_1	4	100± 1.98[a]	0± 0.00[a]	31	75.23± 1.13[a]	32	55.12± 1.07[a]
T_2	4	100± 1.98[a]	25± 0.41[b]	33	54.87± 1.03[b]	33	46.49± 0.91[a]
T_3	4	50± 0.94[b]	0± 0.00[a]	35	35.4± 0.77[c]	34	26.39± 0.93[b]

(Mean ± SE); values of the parameter in each column with different superscripts (a, b, c) differ significantly (P<0.05).

4. Discussion

Proper care of broodstock is very important for assuring good production of eggs, fry and fingerlings [16]. The daily feeding of broodstock diets have direct effects on fecundity and egg size [7]. *M. pancalus* is a bottom feeder fish that feeds on debris matter near the bottom due to predominant occurrence of algae and detritus [4]. There was an effect of maturation of the brood on the ovulation, fertilization and hatching rate. Immature eggs were not fertilized and while fertilized sometimes produced deformed spores which did not hatch out. The experimental fishes were fed with supplementary feed containing mustard oil cake, rice bran, wheat bran, and vitamin premix at the ratio of 20:49:30:1 by weight at 5% of total body weight of fish. Alam *et al.* [3] had stated that fed with a mixture of mustard oil cake, rice bran, wheat bran, fish meal, vitamin premix and di-calcium phosphate at the ratio of 20:33:25:20:1:1 by weight at 4 - 5% of total body weight of fish per day for artificial propagation of *M. pancalus.*

Water quality parameters are an important factor for successful breeding. Fertilization and hatching rate depends largely on water quality [3]. Underground water was used during the experiment. The water was exchanged daily at 30% by siphoning so that excessive amount of iron, carbonate and other ions were not accumulated at the bottom of the tank and it provided a favorable breeding performance of the specimen. The water temperature range was 27ºC to 31ºC which was might be optimum for the breeding of *M. pancalus. M. pancalus* successfully breeds at 28.1 to 31.2 °C (see: Afroz *et al.*) [1] and at 27 to 31^{0}C (see: Alam *et al.*) [3].Besides this, Farid *et al.* [7] and Das and Kalita [6] successfully breeds *M. aculeatus* at 27 to 33ºC and at 28 to 30ºC respectively. This range of temperature is suitable for breeding of most indigenous small fishes (SIS) [2, 9, 19]. The average pH and DO was 7.2 to 8.5 and 3.5 to 5 mg/L respectively in this experiment. This is also supported by Afroz *et al.* [1] and Alam *et al.* [3]. Afroz *et al.* [1] successfully breeds *M. pancalus* at 7.13 to 8.69 of pH and 3.6 to 4.9 mg/l of DO; Alam *et al.* [3] successfully breeds the *M. pancalus* at 6.5 to 7.4 of pH and 6.5 to 7.5 mg/l of DO.

The inducing of ovaprim into *M. pancalus* showed 100% spawning performance, 75% fertilization rate and 55% hatching rate. Das and Kalita [6] have studied on *M. aculeatus* and injected 0.025 ml and 0.05 ml of ovaprim for individual female and male respectively. They found 100% spawning performance, 78% fertility and 67% hatchability. Sahoo *et al.* [18] have also studied on *M. aculeatus* using the same dose of ovaprim as Das and Kalita [7] and found 83% fertility and 76% hatchability.

There were 50% spawning, 35% fertilization and 26% hatching performances by using 60 mg PG/kg body weight of *M. pancalus.* Alam *et al.* [3] used 170 mg PG/kg body weight of *M. pancalus* and showed 77% ovulation rate, 91% fertilization rate and 80% hatching rate. Rahman *et al.* [14] have found 90% ovulation rate, 90% fertilization and 80% hatching rate of *M. pancalus* by using the same dose of PG. Farid *et al.* [7] have found 86% fertilization and 50% hatching rates of *M. aculeatus* by using 90 mg PG/kg body weight. Previous researchers were used PG into *M. pancalus* with very high doses, but in the study the PG dose was relatively lower. This could be a cause of the variation in results.

PG is usually use for breeding of small indigenous species (SIS) by many researchers with very high doses. Ovaprim and HCG is also common inducing hormone for SIS breeding. However, there were no experiment was conducted on *M. pancalus* with ovaprim. Ovaprim was used in *M. aculeatus* but no experiment yet using HCG. This variation of the treatments in spawning, fertilization and hatching may be arisen because they were treated with different hormones. Although some variations may arise due to experimental error. These differences in the results would also be due to environmental factors and bad quality of hormones, as it was collected from market. The water flow reduced in the small experimental jar because of double protection with wire mesh. This could lower the hatching rate.

5. Conclusion

The present study evident that *M. pancalus* responses with these three different hormones, but ovaprim successfully responses. So ovaprim could be an alternative hormone for the induced breeding of *M. pancalus.* The present study can through light for future researches on the improvement of induced breeding of *M. pancalus* concerning the optimization of ovaprim hormone application.

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7. Reference

1. Afroz A, Islam MS, Hasan MR, Hasnahena M, Tuly DM. Larval rearing of spiny eel, *Mastacembelus pancalus* in the captivity with emphasis on their development stages. Int. J. Fish. Aquat. Stu. 2014; 1(6):163-167.
2. Akhteruzzaman M, Kohinoor AHM, Shah MS. Observations on the induced breeding of *Puntius sarana* (Hamilton). Bangladesh J. Zool. 1992; 20(2):291-295.

3. Alam M, Alam MS, Alam MA, Islam MA, Miah MI. Dose optimization with PG hormone for induced breeding of *Mastacembelus pancalus*. Ecofri. Agric. J. 2009; 2(9):799-804.
4. Azadi MA, Naser A. Food and feeding habits of *Labeo bata* (Hamilton) from Kaptai reservoir. Chittagong University Studies, Part II: science 1996; 20(2):11-17.
5. Bromage N, Cumaranatunga R. Egg Production in Rainbow trout. In Recent Advances in Aquaculture, 1988; 4:63-138.
6. Das SK, Kalita N. Captive breeding of Peacock Eel, *Macrognathus aculeatus*. J. Assam Agric Uni. 2003; 8(3):35-36.
7. Farid SM, Miah MI, Habib MAB, Rahman MM. Effect of pituitary gland (PG) doses on artificial propagation of endangered Tarabaim, *Macrognathusaculeatus* (BLOCH). Progress of Agriculture 2008; 19(2):111-118.
8. Froese R, Pauly D. Fish Base, World-wide Web Electronic Publication. http://www.fishbase.org, 2015.
9. Islam Z, Chowdhury AQ. Induced spawning of major carp for commercial production of fry in the fish seed farm, Bangladesh J. Zool. 1976; 4(2):51-61.
10. IUCN. Red Book of threatened fishes of Bangladesh, 2000, 116.
11. Karim MA, Hossain A. Studies on the biology of spiny eel, *Mastacembelus pancalus* in artificial ponds. Part II. Sexual maturity and fecundity. Bangladesh J. Agric Sci. 1992; (1):15-18.
12. Pathak BC, Ali R, Serajuddin M. Comparative Analysis of Reproductive Traits in Barred Spiny Eel, *Macrognathu pancalus* (Hamilton, 1822) from Lotic and Lentic Ecosystems of Gangatic Basin, India. World J. F Mar Sci. 2012; 4(5):470-479.
13. Rahaman MM, Miah MI. Fecundity of guchi baim, *Mastacembelus pancalus*. J. Bangladesh Agric. Uni. 2007; 7(1):133-137.
14. Rahman MM, Miah MI, Hasan KR, Farid SM. Effects of pituitary gland doses on artificial propagation of guchibaim, *Mastacembelus pancalus* (Hamilton), J. Bangladesh Agric Uni. 2011; 9(2):305-310.
15. Rahman MM, Miah MI, Taher MA, Hasan MM. Embryonic and larval development of guchibaim, *Mastacembelus pancalus* (Hamilton). J. Bangladesh Agric Uni. 2009; 7(1):193-204.
16. Robert K, Piper G, Melweing IB, Orme LE, Mecraren JP, Fowler LG *et al.*, Fish Hatchery Management, 1982, 131-199.
17. Saha BK, Islam MR, Saha A, Hossain AM, Some aspect of reproduction of the indigenous spiny eel, *Mastacembelus pancalus* from netrakona, Bangladesh. Bangladesh J. Zool. 2006; 34(2):289-293.
18. Sahoo SK, Giri SS, Chandra S, Sahu AK. Larval rearing of the spiny eel, *Macrobnathus aculeatus* (Lacepede) under different rearing condition: a preliminary study. Indian J. Fish. 2009; 56(1):47-49.
19. Sardar AS. Artificial Breeding of Chinese and Indian major carps, training programmeon hatchery technology and aquaculture for Cambodian Fisheries Personnel (PADEK), 1997, 3-6.
20. Serajuddin M, Ali R. Food and feeding habits of striped spiny eel, *Macrognathus pancalus* (Hamilton), Indian J. Fish. 2005; 52(1):81-86.
21. Suresh VR, Biswas BK, Vinci GK, Mitra K, Mukherjee A. Biology and fishery of barred spiny eel, *Macrognathus pancalus* (Hamilton, 1822). J. Acta Ichthyol. Piscat. 2006; 36(1):31-37.
22. Talwar PK, Jhingran AG. Inland fishes of India and adjacent countries. Oxford and IBH Publishing Co. PVT. Ltd., New Delhi, India, 1991; 2:789-541.
23. Vettath RS, Benoy KB, Gobi KV, Krishna M, Mukherjee A. Biology and fishery of barred spiny eel, *Macrognathus pancalus* (Hamilton, 1822). J. Acta Ichthyol Piscat. 2006; 36(1):31-37.