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FECUNDITY AND INDUCTION OF SPAWNING IN SPINY GUCHI, *MACROGNATHUS PANCALUS* (BLOCH & SCHNEIDER, 1822) IN BANGLADESH

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Abstract: The fecundity of *Macrognathus pancalus* was measured by Gravimetric method and fecundity was found to range from 866.93±117.65 to 2074.25±88.79 from 100 samples having a total length from 47.90±1.27 to 83.19±1.71 mm, body weight from 5.71±0.37 to 30.74±1.62 g and gonad weight from 3.56±0.44 to 5.06±0.48 g. A screening test of different doses of common hormonal substance pituitary gland (PG) from 5.0 mg to 185.0 mg/kg body weight and 5.0 mg to 60.0 mg/kg body weight were tested for different length of time to find out the accurate dose which responses the ovulation and spermiation of *M. pancalus*. A double dose of PG extracts (2.0 mg and 180.0±1.01 mg/kg body weight) was the best effective dose in ovulation and a single dose of PG (60.0±1.05 mg/kg body weight) was most effectual in spermiation.

mvi-ms‡¶c: ¯x ¸wP evBg (*Macrognathus pancalusn*) gv‡Qi wWg Drcv`b ¶gZv Gravimetric c×wZ‡Z wbi¡cb Kiv n‡q‡Q Ges ¯x gv†Qi Mo ^`N¨ 47.90±1.27 - 83.19±1.71 wg.wg., kvixwiK Mo IRb 5.71±0.37 - 30.74±1.62 Mvg Ges Rbb As‡Mi wW¤vbi Mo IRb 3.56±0.44 - 5.06±0.48 Mvg m¤úb 100wU gv‡Qi wWg Drcv`b ¶gZv 866.93±117.65 †_‡K 2074.25±188.79 cvIqv hvq| *M. pancalus* gv‡Qi KwÎg cRb‡b cwicK gv‡Qi wcUBUwi MwŠn n‡Z †Mvbv‡WvUwcb ni‡gvb wbhvm ^Zwi K‡i cwZ‡KwR ¯x gv‡Qi IR‡bi Rb¨ 5.0 wg.Mv. †_‡K 185.0 wg.Mv. Ges cwZ‡KwR ci"I gv‡Qi IR‡bi Rb¨ 5.0 wg.Mv. †_‡K 60.0 wg.Mv. wewfb mg‡qi mv‡_ mvgÄm¨ †i‡L Bb‡RKkvb c‡qvM K‡i ^Re D‡ËRbv mwói gva¨‡g gv‡Qi wWg Qvov I ïµvbi wbhv‡mi cKZ wcUBUwi MwŠn wbhv‡mi gvÎv wbi¡cb Kiv nh| m‡evcwi, *M. pancalus* gv‡Qi wWg Qvovi †¶‡Î me‡P‡q djcm gvÎv n‡jv wcUBUix MwŠni †Mvbv‡WvUwcb ni‡gvb wbhvm cwZ †KwR gv‡Qi IR‡bi Rb¨ 2.0 wg.Mv. I 180.0±1.01 wg.Mv. Ges ïµv‡bv wbhv‡mi †¶‡Î mdj cKZ gvÎv n‡jv wcUBUix MwŠni †Mvbv‡WvUwcb ni‡gvb wbhvm cwZ †KwR gv‡Qi IR‡bi Rb¨ 60.0±1.05 wg.Mv.|

Key wards: Ovary, Linear relationship, Correlation coefficient, PG, Ovulation, spermiation, fertilization and hatchling.

INTRODUCTION

World aquaculture has entered into a new and challenging phase in the last few decades. Continued demands on animal protein supply focused major attention on fisheries research and innovation of new methods for increased production. A development of new technology particularly induction of spawning offers a good opportunity to develop the culture stock of fish. Breeders in Bangladesh have significantly raised productivity for a many commercially important species of carps, catfish and tilapia, but the successful breeding and cultivation of spiny eel like Guchi, *Macrognathus pancalus* would be a tremendous boost to high-value aquaculture.

The knowledge of fish fecundity has much relevance in fish population studies, commercial potentials, and culture and management practices. Fecundity is usually studied in fishes to establish relationship with length, weight and age (Ibrahim 1957), to provide relative index of density dependent factors affecting the population size for reporting the various stocks and races of population (Baxter 1959) and also to develop numerical relationship between egg-production and recruitment (Beverton and Holt 1957).

The spiny eel, Guchi, *Macrognathus pancalus* is one of the important fresh water fishes of the South-east-Asia. The species is distributed to Bangldesh, India, Pakistan (Talwar and Jhingran 1991) and Nepal (Froese and Pauly 2006). It is commonly found in open water (rivers, canals and beels), boro-paddy fields and swamps of Bangladesh and total length is recorded 135 mm ((Rahman 1989). This species is very much popular for its excellent taste and high market value. This species plays an important role in meeting the nutritional requirements of the people of Bangladesh. This fish was abundantly available in our open water system but due to over exploitation and various ecological changes in its natural habitat; this species is gradually declining. The entire demand for this species is meeting through collection from the wild. Indiscriminate destructive practices have caused havoc to aquatic bio-diversity. Recent studies suggest that world wide 20% of all fresh water species are extinct, endangered or vulnerable (Moyle and Leidy 1992). International Union of Conservation of Nature (IUCN), Bangladesh (1998) enlisted *M. pancalus* as not threatened spiny eel in Bangladesh. But due to rough and unplanned water management policy for irrigation, over exploitation, illegal practice of capture fisheries and various ecological changes in its natural habitat, this species is threatened now. Considering the importance of this species in nutritional, economic and biodiversity point of view, its conservation and propagation are considered through fisheries regulation and a development of appropriate artificial propagation technique of *M. pancalus.* This technology will be helpful to prevent the fish from being extinct and this tasty fish will be available for the rural people of Bangladesh. Nevertheless, comprehensive information on the induction of *M. pancalus* is not available to provide necessary imputes to this study. There is a study on the age and growth (Nabi and Hossain 1990) and induction of spawning of *Macrognathus aral* (Chakraborty 2008). However, a few studies that are related to secretory cells in the gills of fresh water spiny eel (Kapoor 1957, Maheshwari 1971) cephalic sensory canals and a case of abnormality in the testes are available. For large scale production of fish seed, comprehensive information on dose optimization and efficacy of different ovulatory agent is required. Therefore, the present study aims to find out the

effectiveness of pituitary gland extracts (PG) on the reproductive response of *M. pancalus* under hatchery condition for commercial seed production.

The present study aims to find out the fecundity of Guchi, *M. pancalus* to establish relationship with length, weight and age, and dose optimization and effectiveness of pituitary gland extracts (PG) on the reproductive response of *M. pancalus* under hatchery condition for commercial seed production to meet up the nutritional requirements.

MATERIAL AND METHODS

The experiment was carried out from October 2006 to September 2007 at the hatchery of fish seed farm of Babul Fish Seed Farm, Ishorgonj, Mymensingh, Bangladesh. The brood fishes were collected from the open water body (beel) of Ishorgonj, Mymensingh district and released into the ponds of the farm. A balanced diet of fish meal (70%), mustard oilcake (20%), flour (9%) and Vitamin E (1%) was supplied twice a day at the rate of (6-8)% of the body weight (Chakraborty 2008). Proximate composition of the feeds was analyzed according to AOAC (1995) method, nitrogen free extract (NFE) by subtraction (Castell and Tiews 1980). Proximate composition (% dry matter) of the supplementary feeds (crude protein, crude lipid, crude fiber, ash and nitrogen-free extract) of experimental feeds was 33.09, 7.45, 11.05, 18.24 and 30.17%, respectively. Brooders were fed twice a day on an average daily. The ponds were also fertilized weekly intervals with only cowdung (494 kg/ha). Water temperature was measured daily with Celsius thermometer.-

During the study period, 100 samples were collected and brought to measure the total length and body weight of individual fish. Then the ovary of each fish was taken out carefully and preserved in 10% buffered formalin with labeled vials for subsequent analysis. The weight of the ovary was measured carefully with the help of a sensitive portable electronic balance (Model HL 400 EX). Gravimetric method was used to estimate the fecundity of *M. pancalus* following Phillips (1969). The fecundity was estimated by counting the number of mature ova from known weights of sub sample collected from the anterior, posterior and middle portions of both the ovaries and calculating the total number of mature ova in the ovary following Grimes and Huntsman (1980). In this method, the ovaries were dissected out by a pair of scissors. The external connective tissues were removed from the surface of each pair of ovaries. The moisture of the ovaries was removed with the help of a piece of blotting paper. The mean number of eggs in 20 mg was determined and then multiplied by the total weight of the ovary, which gave the total number of eggs.

Induced breeding was carried out in brooders showing an advanced stage of maturity: females with well-developed ovarian follicles (i.e. mode diameter ≥1.0 mm) and males discharging milt. Ovarian follicles were sampled in females with a flexible catheter. The diameter of about 40 fresh follicles was measured in each sample using a dissecting microscope (×12.5) equipped with an ocular scale. Small follicles were always numerous, measurements included follicle with diameter >0.2 mm. The diameter of the ova ranged from 0.2 to 1.3 mm. The pick reproduction period occurred largely between May and July. The mature female and male brood fishes of *M. pancalus* for induction were collected from the rearing pond with cast net, bamboo trap and fish aggregating device (FAD), and then anesthetized in a solution of 0.2-0.3 ml/L phenoxy-2-ethanol. They were placed in different breeding aquaria for about 12 hrs for conditioning prior to injection of PG extracts. The experiment contained a series of trials where the numbers of male and female *M. pancalus* were 256 and 148 (including three different treatments) with an average body weight of 18.06 ± 2.42 g and 30.74 ± 1.62 g, respectively. The experiment had three replications. The selected fish were placed in an aerated aquarium. But for confirmation test, the female and male (1:2) were placed in the aerated aquarium. Permanent water exchange was provided through supplying process of over headed tank. Water temperature ranged from 26°C to 30°C during the experimental period.

Dry pituitary gland (PG) extracts (M/S Quddus; 1 g PG ample) were collected from a Local market and the required amount of PG extracts was homogenized in a tissue homogenizer with small amount of distilled water. Homogenized glands were poured in a centrifuge tube and diluted with distilled water to obtain the desired concentration of stock solution. The prepared solution was stored in a refrigerator and required dose of PG/kg body weight for screening test was injected to both male and female spawners. The identified doses of male received one injection containing at the rate of 60.0 mg PG/kg body weight and female received two injections containing at the rate of 2.0 mg and 180.0 mg PG/kg body weight. During fish handling for hormone injection and stripping period to collect milt and eggs the male and female were anesthetized with phenoxy-2-ethanol solution. Spawning took place usually within 8.0-10.0 hrs and milt was collected from the male onto 8.5 cm sterilized petridishes. One cc of milt was mixed with Hanks Balanced Salt solution in the ratio of 1: 4.

After successful determination of the ovulation doses of *M. pancalus* from the screening test, a final test was designed to find out the ovulation and hatching period of the fish. After successfully ovulation, the eggs from female fishes of *M. pancalus* were collected into a bowel and eggs were fertilized with the milt of *M. pancalus.* The eggs and milts were mixed thoroughly with a soft and clean

feather. A few drops of water were added to the mixture on the tray and was shaken gently to ensure effective fertilization. Fertilized eggs were washed several times with water. Trials A, B, C and D were conducted on (5-45) mg, (46-95) mg, (96-145) mg and (146-185) mg/kg body weight to find out accurate doses of ovulation. Trials A, B and C tested were (5-20) mg, (21-40) mg and (41-60) mg/kg body weight of the fix to detect accurate doses of spermiation. Male received a single PG injection in order to enhance milt production. After screening test, a confirmation test (Treatment T_1, T_2 and T_3) was designed to identify best combination of doses of spermiation and ovulation, spawning duration, fertilization and hatching rate of *M. pancalus*. To promote fertilization a special solution named Fertilization solution (carbamide, 4g salt and 3g urea/L of water) was added to the fertilized eggs and the mixture was stired continuously for 8-10 minutes. Due to adhesive and stickiness characteristics of the eggs of *M. pancalus* tannin solution (5 g of tannin/10 L water) was added and mixed very quickly. Then the eggs were washed several times with fresh water because tannin solution was very toxic to the eggs. The fertilized swollen eggs (approximate 1000) were transferred into Pepsi bottle jars (2 L capacity each) connecting with water flow system. The flow of water in the jar was regulated and the rate of flow was maintained at 400-500 ml/minute during incubation period. Numbers of live eggs in each group were determined within two to three hrs of fertilization. Eighty to eighty two hours after hatching, the yolk sac totally disappeared. At that time the fine egg yolk emulsion was prepared and fed at the rate of 300% egg yolk per day to meet up the dietary requirement of the hatchlings. During incubation, dead embryos were removed to prevent fungal growth. Number of live eggs was determined within two to three hrs of fertilization. The fertilization rate (%), hatching rate (%) and survival rate (%) upto yolk sac were estimated. The larvae were counted and stocked in a nursery pond.

Statistical package MICROSTAT and EXCEL were used to determine correlation co-efficient (R^2) between total length and fecundity, body weight and fecundity, and gonadal weight and fecundity following the methods of Zar (1984). The data collected on fertilization rate, hatching rate, survival upto yolk sac and production were subject to statistical analysis ANOVA and DMRT.

RESULTS AND DISCUSSION

The fecundity was estimated from 100 randomly collected fish samples ranging in total length from 87.29 ± 1.90 to 123.58 ± 1.38 mm, body weight from 5.71 ± 0.37 to 30.74 ± 1.62 g and ovary weight from 3.56 ± 0.44 to 5.06 ± 0.48 g.

The fecundity was found to vary from 866.93±117.65 to 2074.25 ± 88.79 (Table 1).

Table 1. Body length and weight, ovary weight and fecundity of different sizes of *Macrognathus pancalus*

Group size length (mm)	No. of fish examined	Total length (mm) (Mean ± SD)	Body weight (g) (Mean ± SD)	Ovary weight (g) (Mean ± SD)	Fecundity (Mean ± SD)
85-90	14	87.29±1.90	5.71±0.37	2.39±0.41	866.93±117.65
91-95	13	92.85±1.46	10.62±1.93	3.22±0.52	1066.92±66.12
96-100	13	98.23±1.66	15.66±1.76	4.15±0.64	1033.33±130.29
100-105	12	102.92±1.62	18.06±2.42	4.44±0.45	1163.62±61.81
106-110	12	108.25±1.48	23.81±2.33	4.65±0.42	1476.17±71.71
111-115	12	113.17±1.70	26.87±1.86	4.96±0.38	16.64.42±50.54
116-120	12	118.08±1.51	28.58±1.55	5.10±0.33	1870.75±107.60
121-125	12	123.58±1.38	30.74±1.62	5.63±0.48	2074.25±88.79

Relationship between fecundity and total length of fish: The relationship between fecundity and total length (mm) is shown in Fig. 1. The relationship between them was found to be polynomial of second order of body weight and is expressed as $Y=370.72–938X+0.2683X^2$ ($R^2=0.9473$, fecundity was highly correlated ($p< 0.01$) with the total length of fish (Fig. 1).

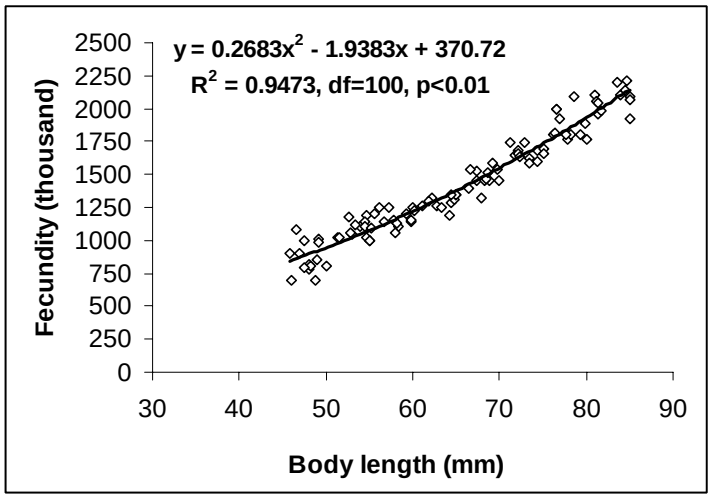

Fig. 1. Correlation coefficient (R^2) between fecundity and body length of *Macrognathus pancalus*.

Relationship between fecundity and body weight of fish: The relationship between fecundity and total body weight of *M. pacalus* is shown in Fig. 2. The regression equation of fecundity on total body weight was Y=580.96+42.368X showing a curvilinear relationship between fecundity and body weight. It was

also found that fecundity was high correlation with the body weight of fish (R_2 = 0.8759, p<0.01).

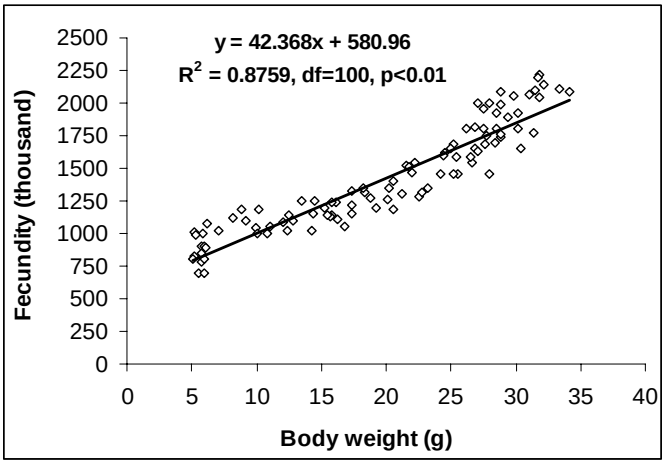

Fig. 2. Correlation coefficient (R^2) between fecundity and body weight of *Macrognathus pancalus*.

Relationship between fecundity and ovary weight of fish: Fig. 3 shows the relationship between fecundity and gonadal weight of *M. pancalus*. A highly significant (*P*<0.01) linear relationship was found to exist between fecundity and gonadal weight. The fecundity was highly correlated (R2 = 0.6606, P<0.01) with the ovary weight of fish (Fig. 3).

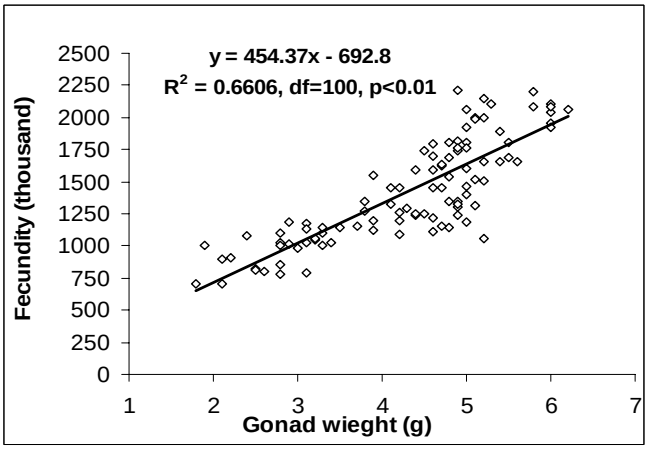

Fig. 3. Correlation coefficient (R^2) between fecundity and ovary weight of *Macrognathus pancalus*.

The data presented in the Tables 2-3 show detailed relationship between different hormonal doses and reproductive response of the breeder *M. pancalus*. During screening test, ovulation was successfully responded to the 2nd doses of

injection in trail D bodyweight, but not in trials A and B (Table 2). Trial C (96.0-145.0) mg/kg bodyweight had shown a little success in the induction of spawning but response of spawning percentage of individual was 4.0% to 57.0% and ovulation was responded 2 to 35%, respectively. Hormone trail D at (146-155) mg/kg, (156-165) mg/kg and (166-175) mg/kg bodyweight was found to be the better effective for ovulation in *M. pancalus* and response of spawning of individual was 75, 87 and 100%, and ovulation was responded 60, 89 and 90%, respectively (Table 2). Prolongation of hormone trail at (176-185) mg/kg bodyweight was the best effective for ovulation and response of spawning individual was 100%, and percentage of ovulation was responded 100%. Only matured ovarian follicles (i.e. diameter ≥ 1.2 mm) were undergone ovulation. Diameter of the dry ova ranged from 1.1 to 1.4 mm and the mean weight of single ovum was 2.1± 0.2 mg.

Table 2. Response of different doses of Pituitary gland (PG) on the screening test of induction of spawning in female *Macrognathus pancalus*.

Month /year	Trial	Doses of 1st inject -ion PG (mg/kg)	Interval period (hr)	Doses of 2nd injection PG (mg/kg)	Observed ovulation period (hr)	n	% of response	Ovulation (%)
May-July 2007								
	A	2	6	<5	12	2	0	0
		2	6	6-15	12	20	0	0
		2	6	16-25	12	20	0	0
		2	6	26-35	12	20	0	0
		2	6	36-45	12	20	0	0
	B	2	6	46-55	12	20	0	0
		2	6	56-65	12	20	0	0
		2	6	66-75	12	20	0	0
		2	6	76-85	12	20	0	0
		2	6	86-95	12	20	0	0
	C	2	6	96-105	12	20	4	2
		2	6	106-115	12	20	15	5
		2	6	116-125	11-12	20	30	10
		2	6	126-135	11-12	20	41	20
		2	6	136-145	10-11	20	57	35
	D	2	6	146-155	10-11	20	75	60
		2	6	156-165	9-11	20	87	89
		2	6	166-175	9-10	20	100	90
		2	6	176-185	8-10	20	100	100
				Total		238		

Figures with different superscripts in the same column varied significantly ($P<0.05$)
n = Total number of female used in the experiment

During screening test of male, amount of PG extract required to promote spermiation was found to be 5-60 mg PG/kg body weight administered at the time of application of second injection to the female. It was recorded that the male fish did not respond to spermiation from (5-25) mg/kg body weight (Table 3). Trial B, the doses of hormone at (26-40) mg/kg bodyweight had shown a partial success in spermiation but response of spawning percentage of individual was 13% to 35% and spermiation was responded 1% to 32%, respectively. Trial C, hormone treatment at (41-45) mg/kg and (46-50) mg/kg bodyweight was effective for the spermiation of *M. pancalus* and response of spawning percentage of individual was 55.0% and 86% and spermiation was responded 52% and 69%, respectively. Hormone treatment at (51-55) mg/kg was better effective for spermiation and response of individual number was 100%, but spermiation was responded 87% only. Prolongation of hormone treatment at (56-60) mg/kg bodyweight was recorded to be the best effective for spermiation and response of individual number was 100% and percentage of spermiation was responded 100%.

Table 3. Response of different doses of Pituitary gland (PG) on the screening test of induction of spermiation in male *Macrognathus pancalus*. Number of replications was two.

Period	Trial	Doses of 1st injection PG (mg/kg)	Interval period (hr)	Doses of 2nd injection (mg/kg)	Observed ovulation period (hr)	n	% of response No.	Spermiation (%)
May-July 2007								
	A	-	6	<5.0	12	2	0	0
		-	6	6-10	12	10	0	0
		-	6	11-15	12	10	0	0
		-	6	16-20	12	10	0	0
	B	-	6	21-25	.12	10	0	0
		-	6	26-30	12	10	13.0	1
		-	6	31-35	12	10	25.0	18
		-	6	36-40	10-11	10	35.0	32
	C	-	6	41-45	10-11	10	55.0	52
		-	6	46-50	9-11	10	86.0	69
		-	6	51-55	9-10	10	100.0	87
		-	6	56-60	8-10	10	100.0	100
				Total		112		

Figures with different superscripts in the same column varied significantly (P<0.05)
n = Total number of male used in the experiment.

Pertinent data regarding the time of injection and best combination doses of spermiation and ovulation, fertilization rate, time of hatching, and hatching rate of a confirmation test (Treatment T_1, T_2 and T_3) are furnished in the Fig.1. The administration of the PG extract at 2.0 mg dose and 177.0 ± 1.04 mg/kg body

weight showed fertilization, hatching and survival rate at 62.0±2.22%, 58.0±1.3% and 50.0±1.1%, respectively. Best spawning occurred under dual hormonal regime at the PG dose of 2.0 mg and 180.0±1.0 mg/kg body weight in the case of female and highest fertilization, hatching and survival rate were recorded at 92.0±1.02%, 89.0±2.05% and 81.0±1.24%, respectively. Second highest spawning was recorded at the PG dose of 2.0 mg and 183.0±1.1 mg/kg body weight in the case of female and fertilization, hatching and survival rate were recorded at 88.0±1.25%, 81.0±1.10% and 68.0±1.06%, respectively. In treatment T_3, an increasing tendency of doses, decreasing tendency of fertility and hatching rate were found. In the case of male, the amount of the PG required to promote spermiation was found to be 60.0±1.05 mg/kg body weight administered at the time of second injection to the female. After administering the 2nd injection, the male and the female fishes moved together in anti-clockwise direction and the female was held by the male, later bending its body, rubbing, knocking and nudging her. Their bodies were twisted round each other and firmed with the fins. They were started to nudge themselves by snout in the mouth and ventral region of the female up to ovulation time.

Ovulation occurred after 8.0 to 10.0 hrs of the 2nd injection and hatchlings came out after 50.0 to 52.0 hrs of fertilization. Best fertilization, hatching and survival rate were found to be at 92.30 ± 2.11%, 89.50 ± 2.05% and 81.0 ± 1.50%, respectively in treatment T_2 (Fig. 4). Thus, the treatment T_2 (2.0 mg and 180.0 ± 1.01 mg/kg body weight) was the best dose of pituitary extracts for the induced breeding of *M. pancalus*. The administration of higher amount of PG extract at 2.0 mg and 183.0±1.02 mg/kg body weight in the treatment C resulted in reduced success. The fertilization, hatching and survival rate were found to be at 88.15 ± 2.02%, 81.11 ± 1.17% and 68.2 ± 1.88%, respectively in treatment T_3. The lowest fertility, hatching and survival rate (62.22 ± 1.88%, 58.22 ± 2.11% and 50.40 ± 2.11%) were recorded in treatment T_1, where the lower doses of pituitary gland extracts were applied.

Statistical analysis using ANOVA followed by DMRT revealed that there was a significant difference (p>0.05) in the effectiveness of different hormone doses from 177.0±1.04 to 183.0±1.1 mg/kg bodyweight and percentage of individual number and ovulation of *M. pancalus*. Significant variation was observed in fertilization and hatching rate of *M. pancalus* eggs following administration of pituitary extract. Significant differences (p<0.05) were observed in their effects on the rates of hatching of embryos and survival of larvae. A little number of offspring died and settled on the bottom of the hatching jar. They were found to be deformed, curved and shortened.

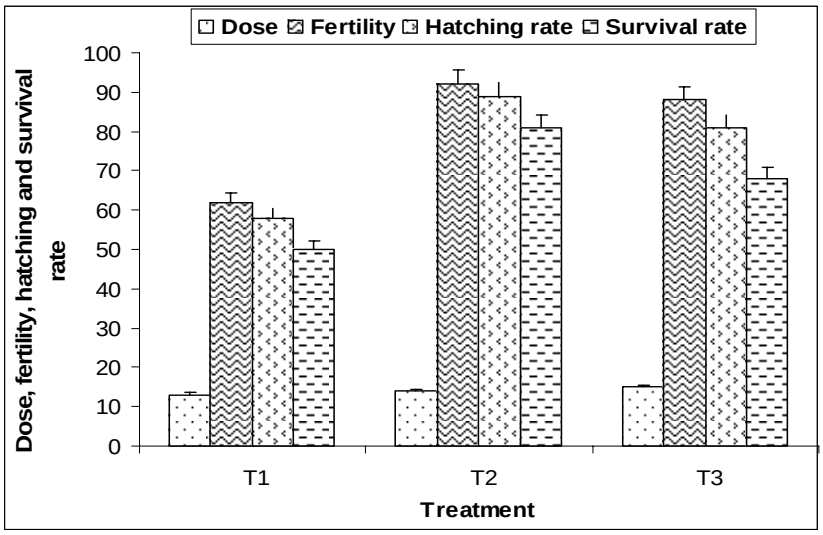

Fig.4. Dose, fertility, hatching and survival rate of *Macrognathus pancalus* at double doses of pituitary gland (PG). Bars represent standard deviation. Significant differences between the successive steps of treatments (P<0.05).

M. pancalus attained sexual maturity at the end of the first year of life which is very much similar to the study of *Macrognathus aral* (Nabi and Hossain 1990). Females of *M. pancalus* were larger in size (123.58±1.38 mm, 30.74 ± 1.62 g) than males (91.11 mm, 10.02 ± 1.44 g) (Swarup *et al.* 1972). They also reported similar size difference in the species with females of 10.6-17.8 cm and males of 7.2-14 cm length in the population. The breeding period of a fish will be for a definite duration if the mature ova are sharply separated from the stock of immature ova (Hickling and Rutenberg 1936).

From the present observation, it is clear that fecundity is directly proportional to total length and weight which is in conformity with the findings of Simpson (1951) and Suresh *et al.* (2006). The relationship between fecundity and weight of *M. pancalus* was found to be curvilinear. A similar result was found by Yuen (1955) and Gupta (1968). Gupta (1968) also observed a linear relationship between fecundity and ovary weight which was similar to this study. But the relationship between fecundity and length of *M. pancalus* was found to be cubic which is very much similar to the study of Sinha (1975). Present study also indicates that this species belonging to the same size group had varying in number of eggs in their ovaries. Lagler *et al.* (1967) reported that the number of eggs produced by an individual female was dependent on various factors like size, age and condition of the species. In this study, it was also observed in some cases that the fecundity of some larger fishes was much less than that of some smaller fish. This type of variation was also reported by

different workers (Doha and Hye 1970, and Ahmed *et al.* 1979). During the study period, the smallest and largest ovaries were measured 31 mm and 40 mm, respectively and weight recorded 2.39 ± 0.41 and 2.39 ± 0.41 g, respectively. The suitable for spawning induction in the female *M. pancalus* was found to be correlated with ovarian follicle size for full ovulation. This criterion for maturity was also observed in catfishes, such as *Pseudoplatystystoma fasciatum* (Kossowski 1996) and *Heterobranchus longifilis* (Legendre 1986).

Dose specificity of PG extract as observed in the present study is in conformity with the findings in *Macrognathus aral* (Chakraborty 2008). He found better spawning performances with pituitary extracts.

In the present study, ovulation occurred after 8.0 to10.0 hours of the 2nd hormonal injection and hatchlings came out after 50.0 to 55.0 hours of fertilization. The successful induction of spawning in *M. pancalus* indicated that the spawners might have received hormone treatment at optimal breeding conditions which is very similar to the findings of Khan and Mukhapadhyay (1975), who pointed out that the success of entire operation of induced breeding depends largely on the proper selection of brood fishes, which has proved very true in the present experiment. Accomplishment of successful spawning depends on the selection of suitable recipient fish at the proper stage of ovarian development and creation of congenial spawning conditions (Nash and Shehadesh 1980). This is successfully carried out artificial breeding, fertilization and hatching of *M. pancalus* by using pituitary gland extracts hormone which is very much similar to the investigation in *Anguilla japonica* (Yamamoto and Yamauchi 1974), *Anguilia anguilla* (Boetius and Boetius 1980) and *Macrognathus aral* (Chakraborty 2008). But for easy and successful breeding for higher output, it is essential to use specific fertilization and tannin solution to remove the adhesive and sticky characteristics of the fertilized eggs of *M. pancalus* (Horvath *et al.* 1992 and Chakraborty 2008). The breeding behavior of *M. pancalus* was very much similar to *Macrognathus aral* (Chakraborty 2008).

The number of eggs produced by an individual female *M. pancatus* was dependent on various factors like size, age and condition of the species Lagler *et al.* (1967). In some cases, the fecundity of some larger fishes was much less than that of some smaller fish (Ahmed *et al.* 1979). Rearing brood stock in ponds at a low density and adequate quality feeding seem to provide suitable conditions of spawning. No hormonal treatment was applied for an advanced stage of follicle development.

CONCLUSION

It is evident from the findings of the present study that PG extracts is the effective in the induction of spawning and the hatchery operators may use pituitary gland extracts (PG) as a source of reproductive hormone for induced spawning of the spiny eel, *M. pancalus.*

SUMMERY

The fecundity and artificial propagation study of this threatened species is essential to save this species in our open water system. Reference to the few published work have been reported in the study. The result has been discussed under Result and Discussion of the chapter. The important findings of the fecundity and artificial propagation have been shown under conclusion. The relationship between body length and fecundity was found to be polynomial and the relationship between fecundity and total weight; fecundity and gonad weight was curvilinear. The regression co-efficient were highly significant ($p<0.01$). A common hormonal substance pituitary gland (PG) extracts was tested to evaluate the efficacy on ovulation, fertility and hatching rate of *M. pancalus* under controlled conditions. A double dose of PG extracts was the best effective dose in ovulation and a single dose of PG was most effectual in spermiation. Highest fertility, hatching and survival rate of offspring was recorded in the treatment T_2.

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