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A Review on Embryonic Development of Inland Fishes of Bangladesh

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Abstract

The early developmental pattern of inland fishes of Bangladesh are not well studied though it has a great importance in fisheries and aquaculture sector. The embryonic study provides interesting information on further growth and health of the fish and considered as an essential component for optimization of fish seed production by natural and induced breeding. Therefore, the current review work has been undertaken to provide a detail information on embryonic development of important inland fishes of Bangladesh. Information was collected from published scientific papers, un-published Masters and PhD dissertations from universities, popular articles and other published and grey literature. Diameters of unfertilized egg of the reviewed fish species were found to be 0.5 to 1.3 mm and fertilized egg were 0.49 to 1.6 mm. Shapes of the egg were also variable from species to species. There is little information available on egg activation and egg micropyle of fish species of Bangladesh. The fertilization rate of different fishes ranged from 40.1% to 93.9%. There are different stages of early development in different species and time needs to complete the stages also vary. The timing of post hatching development by metamorphosis was found to vary based on the fish species from several days to weeks. Different factors like temperature, photoperiod, DO, seasonality and presence of chemicals in water were found to affect the early development of fish. The review included eighteen inland fishes and unearthed useful insights of their embryonic development and influence of different factors. As we expect, the outcome of the study would provide a baseline and would be very useful in conducting further research on the embryology of indigenous fishes of Bangladesh.

Keyword: Fish embryo, ontogenic development, early life stage, hatching, larvae

1. Introduction

Bangladesh is a land with massive potential water bodies with a wide diversity of fishes. During several decades' fishes of Bangladesh has declining due to various natural causes. Different man-made activities are also influencing the process. IUCN (2015) reported that 64 fish species are threatened which comprises 9 Critically Endangered, 30 Endangered and 25 Vulnerable fish species. Therefore, the conservation of this fishes is considerably important. The knowledge on early developmental pattern is essential for establishment of proper conservation measure. However, there is a very little knowledge available about the early embryonic and larval development of fishes of Bangladesh. Embryonic development is a complex process in which cellular differentiation and proliferation occur simultaneously at different rate (Hall, 2003). Changes in the pattern of the entire structure of an organ or of specific organ in relation to the environment are decisive for evaluating the developmental pattern of a species. Information on early life history is an essential requirement for optimization of mass seed production, culture and management of fishes. Embryonic development and larval development providing remarkable information in itself are imperative and consequential to the successful rearing of larvae for large scale seed production. Therefore, it is indispensable to conduct study to characterize different embryonic and larval stages of fish. In addition, embryonic developmental stages of fish life are also used in various investigational studies; especially in aquaculture as well as toxicological studies (Rahman *et al.,* 2009).

1.1 Fish Embryo

Embryo is the earliest developing stages of fish from the time when the fertilized egg starts to divide, while it is continued within the egg until hatching. The embryo goes through several complex stages before hatching. Embryo is the result of fusion of male and female gamete and the first stage of life in fish as well as other animals (Langeland & Kimmel, 1997).

1.2 Gametogenesis

The process of gamete formation in the sexually reproducing animals is gametogenesis. The male gamete is known as spermatozoon or sperm, and the female gamete is known as ovum or egg. Fish produce gametes directly through meiosis in organs called gonads (testis in males and ovaries in females). Gametogenesis are of two types; spermatogenesis and oogenesis (Andrade *et al.,* 2001).

1.2.1 Spermatogenesis

Spermatogenesis involves two distinct process known as formation of the spermatids and Spermiogenesis (Sharma *et al.,* 2018). The primordial germinal cells called spermatogonia undergoes repeated mitotic divisions to maintain a supply of cells for the production of sperm. After meiosis primary spermatocytes form secondary spermatocytes; after the second meiotic division, they are spermatids (Avidor-Reiss *et al.,* 2015). The metamorphosis or differentiation of the spermatids into the sperm is called spermiogenesis. Spermiogenesis is characterized by changes in the nucleus, acrosome formation and Centrioles (Avidor-Reiss *et al.,* 2015).

1.2.2 Oogenesis

The primordial germinal cells divide repeatedly to form the oogonia which multiply by the mitotic divisions and form the primary oocytes and pass through the growth phase. In the primary oocyte, large amount of fats and proteins become accumulated in the form of yolk and due to its heavy weight, it is concentrated towards the lower portion of the egg (forming vegetal pole). After this process cytoplasm of egg divides unequally forming three polar body and one egg (Lubzens *et al.,* 2010).

1.3 Different Embryonic Developmental Stages of Fishes

1.3.1 Fertilized Eggs

Prior to fertilization, the egg is in a quiescent state, arrested in metaphase of the second meiotic division. Upon binding of a sperm, the egg rapidly undergoes a number of metabolic and physical changes. The yolk is usually translucent and yellowish in color; the oil droplets are unchanged (Kinsey *et al.,* 2007). Aerobic respiration increases in the egg (Nakano, 1953). Enzyme systems become activated. In most animals, a burst of protein synthesis begins and the nucleus undergoes the second division of meiosis after fertilization.

1.3.2 Cleavage

The zygote experiences a quick cell cycles with no significant growth, producing a cluster of cells within few minutes of fertilization is called cleavage (Sperber, 1995). Cleavage is basically occurring in the blastodisc region of the animal pole of the eggs which further converts into embryo (Fukazawa *et al.,* 2010). After several successive cell division, the egg forms a thick layer of cell known as germ ring and is made up of superficial layer, ectoderm, endoderm mesoderm (Lee *et al.,* 2004). A dorsal-ventral axis forms at that time that may be referred as pre-notochord from which a neural plate is formed. Cells of the neural plate fold to form the neural groove and the surrounding neural folds which fuse, forming a hollow neural tube (Forgacs & Newman, 2005).

1.3.3 Morula, Blastula and Gastrula

A series of cleavage of zygote forms a solid ball of cell is called morula which occurs within few hours of fertilization. Soon after development of the 8-cell or 16-cell embryo (depending on the species), the blastomeres begin to form mulberry-shaped mass of cells called a morula. This change in shape of the embryo is called compaction (Forgacs & Newman, 2005). This compaction leads to form a hollow sphere called blastula, surrounding by a hollow blastocoel (Forgacs & Newman, 2005). The blastula enhances gastrula formation in which embryo form germ layers (Gilbert, 2010). Gastrula is a dramatic rearrangement of the cells of the blastula. Initially blastoderm cells move outwardly to intercalate with the more superficial cells which leads to the formation of gastrula (Warga & Kimmel, 1990). At this stage the yolk syncytial layer starts expansion around the yolk cell (Trinkaus, 1984). By the end of gastrulation, embryonic cells have rearranged into three layers' ectoderm, mesoderm and endoderm.

1.4 Importance of Embryonic Developmental Study of Fishes of Bangladesh

Different organs of fish develop in different embryonic stage. Knowledge about the timing of different organ development of fish can be extended by embryonic study. Knowledge on cleavage pattern of Bangladeshi fish can be achieved by embryonic study. Embryonic study can be helpful to optimize survival and growth rate of fish larvae in our country. Embryonic development besides, providing interesting information are imperative and substantial to the successful rearing of larvae for large scale seed production of any species and thus important for hatchery operators.

1.5 Factors Affecting Embryonic Development

A wide-ranging literature is present on the problem of the effect employed by some environmental factors upon the embryonic stage in fishes. These factors are like temperature, salinity, light, and some mechanical factors. Among this factors temperature is the most prominent factor as it is well known, the rate of the embryonic development of a given species is directly related to the temperature. Salinity is another important factor that affect the embryonic development of fishes. Every fish species has a tolerance limit of salinity beyond which in cannot survive. Light is not always controls the development of fish embryo. However, it has been shown in case of some species e.g. some salmonids, light usually has a negative impact on the early development of embryo (Eisler, 1957). Rather than these factors, some mechanical factors like pressure, shock etc. can sometime influence the embryonic development of fishes (Ciechomski, 1964).

1.6 Justification and Objectives of the Study

The embryonic and larval stages are considered very sensitive indicators of environmental disturbances (Marimuthu and Haniffa 2007). They are also indispensable in the study on ontogeny and phylogeny of their families (Legendre and Teugels 1991; Verreth *et al.,* 1992). In addition, such studies on the embryonic development of any cultivable species can be useful in directing the husbandry efforts of fish farmer. In Bangladesh the embryonic study of fish has been not yet gained so much attention though it has a huge potential in the sector of fish and fisheries. The current review work is therefore has been undertaken to provide a details knowledge about the embryonic development of fishes of Bangladesh, its current status and future research importance on embryonic development of fishes of Bangladesh.

2. Materials and Methods

2.1 Literature Collection and Review

Literature were collected from different journals, published paper, Magazines related to fish and fisheries. Unpublished research from masters and PhD research were also considered. Personal communication with the experts on the field of fish embryology were done for collection of some information. The fish species under this review are given in Table 1.

Table 1. Fish species under review.

Local name	Scientific name	Family	Author
Rui	*Labeo rohita*	Cyprinidae	Das *et al.,* 2006
Catla	*Gibelion catla*	Cyprinidae	Tumbahangfe *et al.,* 2014
Mrigel	*Cirrhinus cirrhosus*	Cyprinidae	Chakraborty and Murty, 1972
Desi Sarputi	*Puntius sarana*	Cyprinidae	Chakraborty *et al.,* 2007
Bata	*Labeo bata*	Cyprinidae	Hossain *et al,* 2007; Miah *et al.,* 2009
Silver berb	*Barbodes gonionotus*	Cyprinidae	Basak *et al.,* 2014
Tara Baim	*Macrognathus aculeatus*	Mastacembelidae	Farid *et al.,* 2008
Guchi baim	*Mastacembelus pancalus*	Mastacembelidae	Hasan, M.R. *et al.,* 2016; Rahman *et al.,* 2009
Shol	*Channa striatus*	Channidae	Roy *et al,* 2016; Marimuthu & Haniffa, 2007
Taki	*Channa punctatus*	Channidae	Banerji, 1975
Tengra	*Mystus cavasius*	Bagridae	Rahman *et al.,* 2004
Rita	*Rita rita*	Bagridae	Molla *et al.,* 2008; Mollah *et al.,* 2011
Pabda	*Ompok pabo*	Siluridae	Purkayastha *et al.,* 2012; Sarma *et al.,* 2012
Kani Pabda	*Ompok bimaculatus*	Siluridae	Raizada *et al.,* 2013
Pangas	*Pangasius pangasius*	Pangasiidae	Khan & Mollah, 2004; Ferosekhan *et al.,* 2015
Local Koi	*Anabas testudineus*	Anabantidae	Karim *et al.,* (2012)
Gutum	*Lepidocephalicthys guntea*	Cobitidae	Sayeed *et al,* 2009
Meni, Bheda	*Nandus nandus*	Nandidae	Pal, *et al.,* 2003; Das *et al.,* 2002
Shing	*Heteropneustes fossilis*	Heteropneustidae	Puvaneswari *et al.,* 2009; Nesa *et al.,* 2017

2.2 Data Analyses, Tabular and Graphical Representation

All collected data were subjected in computer software MS Excel v2016 for analysis and graphical representation.

3. Results and Discussion

3.1 Fertilization

The egg and spermatozoa are the main component for fertilization. The eggs and spermatozoa of many fish have an extremely short functional life after spawning. Marimuthu & Haniffa, (2007) showed in case of *Channa striatus* fertilized egg were free floating, spherical, non-adhesive translucent and yellow in color. In case of *Rita rita*, Mollah *et al.,* (2011) reported the same result but it was demersal and brownish in color. They reported a redish spot in the fertilized egg which indicates the blastodisc. Same blastodisc has been reported in case of *Macrognathus aculeatus*, *Labeo bata*, *Anabas testudineus*, *Nandus nandus*, *Heteropneustes fossilis, Mystus cavasius,* by Farid *et al.,* (2008); Miah *et al.,* (2009); Karim *et al.,* (2012); Das *et al.,* (2002); Nesa *et al.,* (2017); Rahman *et al.,* (2004). The diameter of fertilized and unfertilized eggs is varying in species to species. Fertilization rate, hatching rate, diameter of fertilized and unfertilized egg of different fish species are given in the Table 2.

The fertilization rate of different fish species is different. Fertilization rate are also season dependent in case of many species. Fertilization rate also differs depending on the natural or striping.

Table 2. Fertilization rate, hatching rate and egg diameter of different inland fishes of Bangladesh

Species name	Highest fertilization rate (%)	Highest hatching rate (%)	Egg diameter		Author
			Unfertilized (mm)	Fertilized (mm)	
Catla Catla	93.9	90.98	-	4.5	Tumbahangfe *et al.,* 2014
Labeo bata	92.33	88.33	0.7±0.01	0.8±0.01	Hossain *et al,* 2007; Miah *et al.,* 2009
Mystus cavasius	-	-	-	0. 49-0.51	Rahman *et al.,* 2004
Rita rita	71.66±7.64	48.33±7.64	1.0 to 1.3	1.3 - 1.6	Molla *et al.,* 2008; Mollah *et al.,* 2011
Mastacembelus pancalus	75.23± 1.13	55.12± 1.07	0.50±0.00	0.70±0.02	Hasan, *et al.,* 2016; Rahman *et al.,* 2009
Macrognathus aculeatus	-	-	0.7±0.11	0.8±0.11	Farid *et al.,* 2008
Ompok pabo	75.5	60.5	0.99-1.1	1.0-1.3	Purkayastha *et al.,* 2012; Sarma *et al.,* 2012
Ompok bimaculatus	75–90	80–90	-	-	Raizada *et al.,* 2013
Heteropneustes fossilis	62.33±4.51	41.33±5.69	1-1.1	1.3 -1.4	Nesa *et al.,* 2017
Anabas testudineus	-	-	0.6±0.01	0.7±0.0	Karim *et al.,* (2012)
Lepidocephalicthys guntea	78.60±3.21	65.49±5.23	-	-	Sayeed *et al,* 2009
Nandus nandus	92±5%	90±2	0.6	0.8	Pal, *et al.,* 2003; Das *et al.,* 2002
Pangasius pangasius	40.1±3.1	65.0±2.3	1.09-1.28	1.2-1.45	Khan & Mollah, 2004; Ferosekhan *et al.,* 2015
Channa striatus	59.5±2.50	67.5±2.50	-	1.20 - 1.40	Roy *et al,* 2016; Marimuthu & Haniffa, 2007

3.1.1 Egg Activation

The event of egg activation is thought to be the result from the introduction of protein or other component from the sperm plasma to the egg. The existence of some factors which are capable of triggering egg for activation has been reported by Coward *et al.,* (2003). The majority of the evidence to date indicates that the critical sperm component is a phospholipase isoform (phospholipase zeta) (Cox *et al.,* 2002; Saunders *et al.,* 2002). One of the exceptions of this was found in the study by Lee *et al.* (1999), which described the apparently normal activation

of zebrafish egg in the absence of sperm. This may be defined as parthenogenic activation egg. In general, some morphological changes occur during fertilization while the egg is being activated. According to Kinsey *et al.* (2007), some morphological changes occur in the fish egg at fertilization like progressive disintegration of cortical alveoli, reduction in volume, transformation of the chorion, expulsion of second polar body, bipolar differentiation. Kusa (1953) says that the first observable change after fertilization involves the outline of the alveolus becoming indistinct. The alveolus then disappears. Till date no researches were recorded on egg activation of fishes of Bangladesh. Details study is needed for the proper knowledge about the activation of fish egg of Bangladesh.

3.1.2 Micropyle

There is no significant record of micropyle study of Bangladeshi fish. At the internal aperture of the micropylar canal a site of sperm attachment was found on the egg membrane (Kudo, 1982), recognized as a gentle cytoplasmic swelling, bearing 19-27 cytoplasmic finger-like projections (Kudo, 1982).

3.2 Embryonic Developmental Stages and Timing of Development in Different Fish Species

The timing of development is species specific and also stage specific, that means different developmental stage needs different time for each species. In Bangladesh, the developmental timing for all species is not well known. However, in case of some species, research shows some good findings. A complete chart on the timing of development is shown in the Table 3.

Table 3. Timing of early developmental stages of inland fishes of Bangladesh

Stages/Species	*Labeo bata* (Miah *et al.,* 2009)	*Ompok pabo* (Sarma *et al.,* 2012)	*Heteropneustes Fossilis* (Nesa *et al.,* 2017)	*Channa striatus* (Marimuthu and Haniffa 2007)	*Macrognathus aculeatus* (Farid *et al.* 2008).	*Anabas testudineus* (Karim *et al.,* 2012)	*Barbodes gonionotus* (Basak *et al.,* 2014)
Fertilized egg	00 min	00 min	00 min	00 min	00 hrs	00 hrs	00 hrs
Two cells	45 min	36 min	20.60 min	15-20 min	-	1.20 hrs	0.35hrs
Four cells	55 min	46 min	39.8 min	-	-	1.50 hrs	0.50 hrs
Eight cells	80 min	60 min	71.67 min	-	-	2.20 hrs	1.00 hrs
Sixteen cells	-	1.08 hrs	-	30-50 min	-	-	1.20-1.40 hrs
Thirty-two cells	-	-	-	1.00-1.20 min		-	2.00 hrs
Morula	45-55 min	2.06 hrs	2.20 hrs	1.3-2.0 hrs	5.00hrs	5.10hrs	3.00hrs
Blastula	4.30 hrs	3.30 hrs	4.15 hrs	5.00-6.00 hrs	9.00 hrs	-	5.00 hrs
Gastrulation	8.30-12hrs	-	6.35 hrs	8.00-9.00 hrs	15.10hrs	-	6.15-6.25 hrs
Germinal ring formed	-	-	5.1 hrs	-	-	8.20 hrs	-
14 somites	-	-		-	-	32.30 hrs	-
16-18 somites	-	-		-	--	37.30 hrs	-
Yolk plug stage	16 hrs	5.0 0hrs	8.25 hrs	-	-	-	--
Organogenesis	16-18hrs	14.00hrs		-	-	-	-
Just before hatching	18-20 hrs	-	22.5 hrs	-	36-40 hrs	75.20 hrs	-

Fully active embryo	-	17.0-18.0 hrs	-		-	-	-
Hatchling competed	20-21 hrs	20 hrs	39.5 hrs	23.30-24.0 hrs	40 hrs	80.30 hrs	13.40-14.00 hrs

3.3 Organogenesis and Hatching Temperature

In organogenesis stage different organs of the fish are formed. In case of Bangladeshi fish, organogenesis is not well studied except only a limited number of species. In an experiment conducted by Farid *et al*. (2008) reported that organogenesis in case of Tara baim starts almost 11 hours after fertilization. Both tail and head ends were clearly differentiated and the beating heart was visible in this stage. Gills and pectoral fins start to appear. Auditory and optic vesicle develops. These aspects of organogenesis were similar to *L. rohita* ((Khan, 1943) and *C. mrigala* (Chakraborty and Murty, 1972). In another study in case of *Labeo bata* conducted by Miah *et al.* (2009) concluded that, at the organogenesis stage, appearance of heart rudiment pectoral fin buds and gill rudiment occurs. Notochord becomes visible, auditory and optic vessels developed in 16-18 hours after fertilization. Das *et al*. (2002) on the other hand, reported that organogenesis occurs 13 hours after fertilization. At this stage both tail and head-end were clearly visible and heart beat starts in case of *Nandus nandus.* Out of this species discussed, there are many commercially important fish species which organogenesis are not well defined in the previous research. However, the pattern of organogenesis is found more or less similar in the species of the country. It may be due to similarity in environmental condition, food supply and geographical location.

Optimum incubation temperature for different fish species is different found. A list of optimum incubation temperature reported for different fish species of Bangladesh is presented in Table 4.

Table 4. Optimum incubation temperature of different fish species of Bangladesh

Name of the species	Scientific name	Temperature	Author
Bata	*Labeo bata*	27-31°C	Miah *et al.,* 2009
Sharputi	*Barbonymus gonionotus*	26.6-27.5 °C	Chakraborty *et al.,* 2007
Mrigal	*Cirrhinus cirrhosus*	25-30 °C	Chakrabatry & Murty., 1972
Taki	*Channa punctatus*	26-30 ºC	Ramanathan *et al.,* 1985
Local Koi	*Anabas testudineus*	27-29 ºC	Karim *et al.,* 2012
Shing	*Heteropneustes fossilis*	29 ºC	Puvaneswari *et al.,* 2009
Pabda	*Ompok pabo*	29.3 ºC	Sarma *et al.,* 2012
Rita	*Rita rira*	27-29 °C	Mollah *et al.,* 2011

3.4 Post Hatching Development of the Hatched Larvae

Post hatching development means the development of the larvae from the hatching till metamorphosis. The pattern of post hatching development of different species is different. Timing of development is also dependent on some environmental factors, like temperature of the water, available DO of the water, salinity etc. A typical chart on the development timing of different fish species reported for Bangladesh is given in the Table 5.

Table 5. Timing of post hatching development of different fishes of Bangladesh

Species name	Time after hatching					Author
	Twelve hours	Twenty-four hour	Thirty-six hour	Forty-eight hour	Seventy-two hour	
Labeo bata	Larvae size 3.0±0.05 mm. Chromatophores seen in the eye. Ventral embryonic fin fold more prominent.	Larvae length 4.4±0.01 mm. Operculum & Myomeres visible. Prominent pectoral and pelvic fins fold.	Eye size increased with pigmentation. Pectoral fin more prominent. Brain lobe visible mouth cleft formed. Larvae reached to	Larvae length 5.9±0.02 mm. Yolk sac convex interiorly, air bladder distinct elliptical. Large black chromatophores on head, prominent gills.	Larvae silver-blackish and transparent, 6.5±0.02 mm in size. Myomere visible. Larvae swim and feed actively.	Harun *et al*., 2009

	Pectoral fin bud appeared.		5.5±0.05 mm in size.			
Puntius sarana	Larvae increased 3.2±0.05 mm in size. Pectoral fin bud appeared. Melanophore bands prominent at the posterior end of the body, also appeared above the eye and around the yolk sac.	Larva length 5.0 ± 0.01 mm. Colour changed to silvery-yellow. Myomeres visible and mouth cleft formed.	Larve color whitish-black. Pectoral and pelvic fin buds found. Length of the larva 5.2 ± 0.04 mm.	Air bladder distinct. A few black chromatophores found in the area posterior to the auditory and large black chromatophores observed on head. Larvae size 5.7 ± 0.02 mm.	Larval length 5.8 ± 0.01 mm. Eyes fully pigmented and pectoral fin bud more pronounced.	Chakraborty *et al.,* 2007
Channa striatus	Length 4.2 mm, Caudal fin begins to separate and pectoral fin buds. Swim bladder formed and heart positioned in front of the yolk.		Average length 5.1 mm. Pectoral fin round shaped, mouth formed, the lower jaw less developed, vent formed. Rudimentary gill opening and pits differentiate. Thick band of melanophore observed.	Average length 5.4 mm. Pectoral fins paddle shaped, mouth formed with well-developed jaw. Vent and gill rudiments clearly visible. Larvae move at water surface and feeding exogenously.		Marimuthu and Haniffa, 2007
Rita rita.	Larvae length 2.2 mm. Yolk sac partially reduced. Alimentary canal tube like. Eye spot with a dark pigmented area, barbels found.	Larvae length 2.5 mm. Pectoral fin buds seen. Pigmentation gradually extended all over the body, blood circulation system fully developed	Length 2.8 mm. Distinct heart visible, functioned actively and reddish blood seen around the heart. Dark pigmented eyes and upper and lower jaws visible. Gills covered by the operculum. Mouth and anal pore found with small opening.	Larvae length 3.0 mm. Pectoral fin folds became distinct and the rudimentary rays developed in the caudal fin. Mouth well-formed and barbels became elongated. Alimentary tract straight and distinct and a small pouch like stomach formed. Larvae swimming smoothly and feeding exogenously at end of this stage.		Mollah *et al.,* 2011
Macrognathus aculeatus	Larvae length 2.8±0.05 mm. Pectoral fin bud appeared. Large number of melanophores appeared above the eye and around the yolk sac.	Pectoral and pelvic fin bud appeared. Air bladder visible. Anus became distinct. Larvae increased to 4.2±0.01 mm.	Length of the larva 5.2±0.04 mm. Colour whitish-black. Eyes became whitish black.	Distinct air bladder seen. Few black chromatophores found on the caudal fin. Large black chromatophores observed on head. Larvae increased to 5.7±0.02 mm in size	Larvae length 6.0±0.09 mm and the silver-blackish and transparent in colour. Eyes fully pigmented. Dorsal and ventral fin folds persistent. Larva swims actively.	Farid *et al*., 2008
Anabas testudineus		Pectoral fins paddle shaped and the movements of fins were		Larva 2.7 mm in length. Yolk sac reduced to half. Brain	Larvae length 3.5mm and mouth gap quite large. Head broadened	Karim *et al.*, 2012

	marked. Mouth elliptical in shape and inferior in position. Gill appeared in the form of comb and the supply of yolk diminished gradually.		formed completely and continuous heart beat visible. Mouth turned in terminal position.	than body and became round in shape. Larva started feeding at the end of this period.	
Heteropneustes fossilis	Larvae length 4.0±0.2 mm, reduced yolk sac seen. Eyes dark pigmented and prominent. Pectoral fin buds seen, jaws formed and the alimentary tract distinct. Heart visible and the blood circulatory system fully functional. Barbells appeared in the form of tiny knobs.	Length of larvae 4.3±0.3 mm. Pectoral fin oval shaped with a membranous flap. Mouth formed. Rudimentary gill openings and olfactory pits differentiated. The yolk reserve further diminished.	Larvae length 4.6±0.2 mm. Barbells became elongated and prominent around the mouth. Anal aperture and opercula well-formed and distinct. Blood circulation observed in the heart, tail and opercula region.	Larvae length 5.0±0.2 mm, brownish in color. Mouth and anus became fully functional. Head prominent and four pairs of barbells noticed. Pectoral fins vascularized and the caudal fin had 5 rudimentary rays. The yolk material completely absorbed and the larvae exhibited vigorous movements	Nesa *et al.*, 2017

3.5 Factors That Influence Embryonic Development

There are many factors that influences embryonic development of fish has been identified by different scientist. These factors may be categories into some category. These are internal factors like endocrine regulation of the fish or may be other external physico-chemical factors.

3.5.1. Endocrine Regulation

Study on endocrine regulation of the developing embryo is not well studied in Bangladeshi fishes except Shing and Koi. In case of Shing, Nesa et al., (2007) reported that a dorso-ventral unpaired fin, and some melanophores appeared on the head region, ventral side of the notochord and dorsal side of the body, probably by influence of some endocrine glands. Karim (2012) also reported some endocrine regulatory development of the Koi embryo, but didn't mentioned any special type of name of the gland. This two research clearly indicates that the development of early stages of the fishes are somehow regulated by the endocrine gland. But there is a lack of clear information that which gland is responsible for development of which organ. More research on this aspect is necessary in case of fish species of Bangladesh.

3.5.2 Physico-Chemical Factors

Embryonic stage is very crucial stage of a fish life. In this stage certain factors plays important role in the development of the embryo. There are many external factors that are responsible for regulation of the embryonic development of fish. These factors may be physical or may be chemical also. Temperature is the most vital factor that determines the proper development of the embryo. It has been proved as one of the major factors that regulate the development of the early embryonic stages of fish. Legendre & Teugels, (1991) indicated that temperature have an influence on the development of the embryo. In a study in case of *Labeo bata* embryonic and larval development were reported optimum from 27 °C-31°C (Miah *et al.,* 2009). Similar influence of temperature was observed in case of Shol*,* koi, indigenous Magur, Shing, Taki, pabda, Pangas in discrete study conducted by Ramanathan *et al.,* 1985; Karim *et al.,* 2012; Singh & Vidyarthi, 1990; Benerji, 1975; Sarma *et al.,* 2012; Ferosekhan *et al.,* 2015, respectively. In a study conducted by Chattopadhyay and Chattoraj (2017), reported that gonadal development and spawning stops with the fall of temperature that happen with the approach of winter. This provides a straight indication that temperature along with photoperiod is the key controlling factor for maturation and spawning in fish. In case of spawning of fish such as carp and many other cyprinids, gonadal maturation begins in late winter or early spring. Therefore, it can easily be said that increasing temperature influence the maturation of gonad as well as influence the early developmental stages of fish which is also agreed by the result showed by Chakraborty *et al.,* (2007) in case of *Puntius sarana* where the author found significant

variation in the larval development in different temperature treatments. In case of Australian strain of *Lates calcarifer* the rate of embryonic development was positively correlated to the increase in incubation temperature and the thermal tolerance range for the Australian strain of *L. calcarifer* eggs was found to be 28–34 °C in a study conducted by Thepot and Jerry (2015). In case of Bangladeshi strain of *L. calcarifer* it may be more or less similar however it needs further research on Bangladeshi strain. In a study conducted by Das *et al.,* (2006) reported highest hatching rate and least time for attaining each ontogenic stage at 31 > 33 > 26 > 36 °C and were significantly different ($p < 0.05$) in case of *Labeo rohita*. The lowest hatching percentage and maximum time duration for attaining a given ontogenic stage for *L. rohita* were observed at 36 °C and also resulted in malformed embryos. Another important factor that affect the embryonic development of fishes is salinity. In case of *L. rohita*, Pillai *et al.,* (2003) reported that survivability limit of *L. rohita* embryo in waters up to 8 ppt salinity but best embryonic development was obtained at 0 to 2 ppt.

Deprived of temperature and salinity, there are some other factors that may have influence on embryonic development of the fishes in Bangladesh. This are sound, light, chemical compounds of water such as DO, pH, Alkalinity etc. (Rahman *et al.,* 2011). But those factors are not well studied yet. Study on this factors that may influence the embryonic development is very important for Bangladeshi fish.

4. Conclusion

One of the major goals of fisheries biology is to inspect a fish stock and in terms of fisheries biology it is vital to know the embryonic and larval development. As these type of studies are essential to define the spawning periods and areas, to determine the chronological variations of the spawning period, to predict the mature stock of fish, to predict the rate of death of fish at the end of spawning period and to inspect the relation of the growth with its environment. In Bangladesh we need more research on the developmental biology of fishes, especially native or indigenous fish needs more concentration on this aspect.

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