

Study of embryonic development and evolution stages of Yellow fin sea bream (*Acanthopagrus latus*).Gholamhossein Mohammadi¹, Mojgan Khodadadi²¹South of Iran Aquaculture Research center, gmohammady@gmail.com²Department of fisheries, Islamic Azad University, Ahvaz Branch, Ahvaz, Iran

Abstract: In present survey development and evolution stages of Yellow fin sea bream (*Acanthopagrus latus*) fetus in two averages temperature 21 and 24 centigrade degree were studied. The brooders were collected from fishery grounds of south of Persian Gulf. Average diameter of lipid globule was 185 ± 0.005 micron and eggs diameter after fertilization were 740.33 ± 0.012 . Length of newly birth larvae was 1679.34 microns. Incubation period in 24 °C was 26 hours and 15 minutes. During the incubation 2-cells stage was observed at 20 minutes. Embryonic evolution stages were distinguished in these study as follows: 1: Two-Cells Blastomere, 2 four -Cells Blastomere, 3: eight -Cells Blastomere, 4: sixteen - Cells Blastomere, 5: Thirty two -Cells Blastomere, 6: Morula, 7: High Blastula Stage 8: Flat Blastula Stage, 9: Starting of Gastrulation, 10: Gastrulation, 11: Neurula, 12: Observation Embryo Profile, 13: Closing of blastopore, 14: The Formation of Somite, 15: The Appearance of Heart, 16: The Formation of Optic Cup, 17: Increasing of Pigmentation, 18: Hatching (10%), 19: Hatching (100%).

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Key words: Yellow fin sea bream, *Acanthopagrus latus*, fetus, evolution, Incubation, Persian Gulf

1. Introduction

The porgies (Percoidei: Sparidae) are primarily coastal fishes with about 110 species and 33 genera. Smith (1938) and Smith and Smith (1986) placed the genera of Sparidae in four subfamilies (Boopsinae, Denticinae, Pagellinae, and Sparinae) based primarily on dentition.

The Sparidae is a predominantly marine family, which is found in the Indian, Pacific and Atlantic oceans (Nelson, 1994) and contains many species of commercial and/or recreational importance and some that are used for aquaculture (e.g. Foscarini, 1988; Kailola *et al.*, 1993; Ingram *et al.*, 2002).

The yellow fin sea bream, *Acanthopagrus latus* (Hottuyn), is one of the important species commercially exploited in Kuwait. In view of its economic importance, Popularity with consumers and increased rate of exploitation (Al-Kanaan, 1999).

A. latus is a protandrous hermaphrodite (Kinoshita, 1939; Abol-Munaf &

Umeda, 1994; Abu-Hakima, 1984; Abou-Seedo *et al.* in press) although nearly Half (51.7%) of the individuals with in a generation is dioecious, out of which 22.9% are females (Abou-Seedo *et al.* 2003).

Various studies have been conducted on the properties and the development of sea bream eggs. It was reported that survival rate predominantly depended on egg size and eggs obtained by hormonal treatment were relatively smaller in size (Boulineau, 1974; Nash and Kuo, 1975). Feeding of the broodstock under favourable conditions has a major impact on the quality of egg (Bromage and Roberts, 1995). It was also reported that an increase of 2-3% in

salinity during incubation would be risky (Freddi, 1985).

The main factor affecting the rate and the quality of the embryonic development is temperature. Going beyond optimal limits during incubation leads to the deterioration of the cellular symmetry and the breaking of the oil globule; it also causes mass mortality and consequently a drop in the rate of larvae production during gastrulation (Jennings and Pawson, 1991). In this study, cleavage and embryonic phase of 18.5°C, which is accepted as the optimal limit of temperature for incubation, have been investigated.

2. Materials and methods

Yellow fin sea bream brood stock, 3 females and 3 males, were selected from wild breeders and stocked in two 10 m³ tank with a seawater flow of 1.5 m³ per h. Frozen cuttlefish (*Sepia officinalis*) and small trash fish food source, were provided daily. No hormonal treatment was applied to the breeders and spawning occurred spontaneously. Eggs were incubated in 30-l dark covered incubators at an initial density of 50eggs/l, with a gentle flow of 22 ± 0.2 °C seawater. Oxygen was 6.5 – 7.8 mg/l, salinity was 42 ppt, and pH was approximately 8.2. Photographs were taken from samples using stria microscope. Diameter of the egg and oil globule, fetus diameter, fetus length, eye size, larve length, larve yolk sack length was measured using Bicocom@Visolob soft wear. To determine the common embryonic developments, 100 eggs were taken from each incubator and were determined every 5 min until the blastomere (32 cells)

stage and then at 45 minutes intervals. Samples fixed in formaline 1%.

Results:

Yellow fin sea bream fertilized eggs are buoyant, transparent, and typical of sparid fish. Fertilized eggs were had a blastomer cell and in this stage circular blastomer was observed in animal polar.

During the incubation, the salinity of natural seawater was 36.3-37.4‰. The oxygen levels ranged between 6.5 and 7.8 mg/l. Fertilized eggs ranged in

diameter from 735.9 to 787.2 μm with a mean of $740.33 \pm 0.0112 \mu\text{m}$ eggs containing a single unpigmented oil globule were positively buoyant (Figure 1-1A). Oil average globules $0.185 \pm 0.005 \mu\text{m}$ to diameter. Incubation period in 24 °C was 26 hours and 15 minutes.

During the incubation 2-cells stage was observed at 20 minutes.

Photographs of all stages are presented in Figures (1 and 2 gropes).

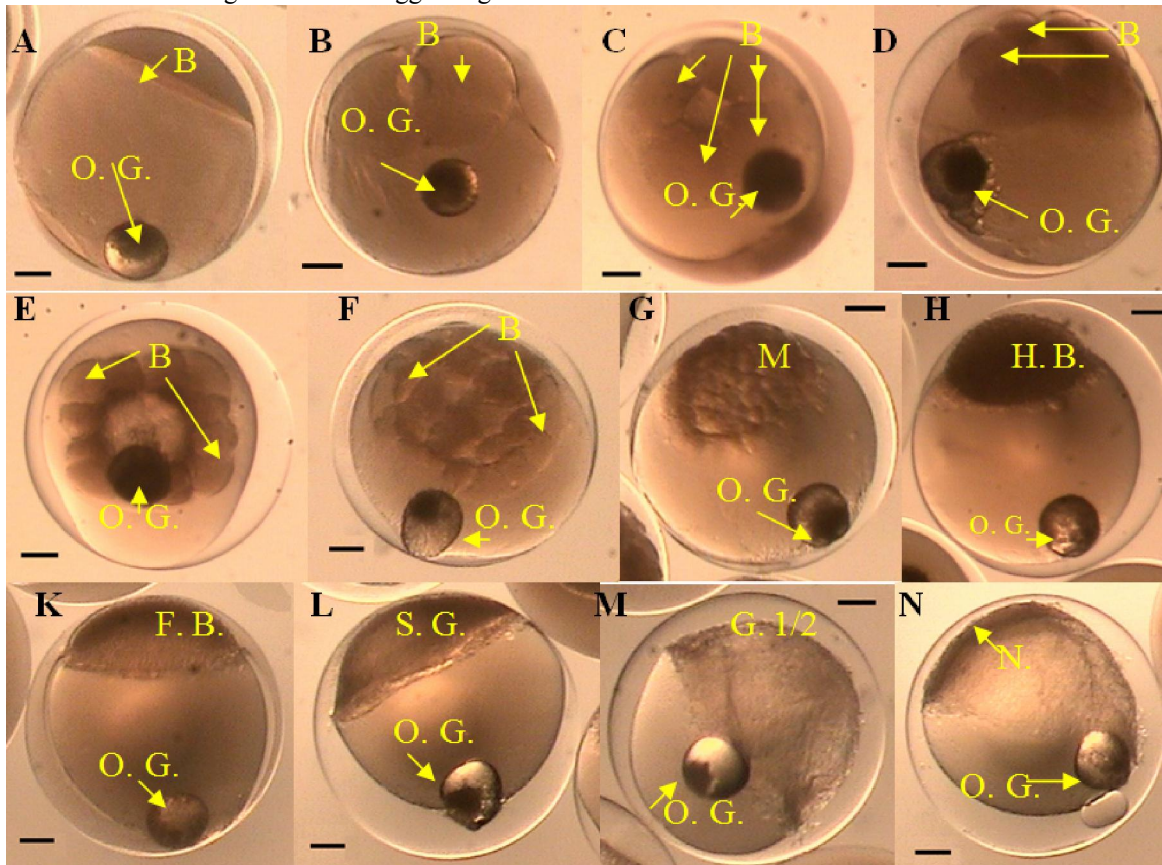


Fig.1 (A to N): Yellow fin sea bream embryonic stages, left to right A: fertilized egg; B: 2-cell blastomere; C: 4-cell blastomere; D: 8-cell blastomere; E: 16-cell blastomere; F: 32-cell blastomere; G: Morulla; H: Early Blastula; K: Late Blastula; L: Early gastrula; M: Gastrulation 1/2; N: Neurula. (Abbreviations, B: Blastomer, O.G.: Oil Globule, M: Morulla, H. B.: High Blastula, F. B.: Flat Blastula, S. G.: Starting of Gasterulation, G1/2: Gasterulation 1/2, N: Notochord)

We distinguished 19 developmental stages for preserved snapper eggs from spawning to hatching as follow.

This is the first stage after fertilization (Figure: 1-A) and zygote have a blastomere cell. The blastomere is completely circular in animal pole.

2-cell Blastomeres: by the time the first cell division stage appeared to be (Figure: 1-B) this step will create a furrow in the cell and spread toward the edges of the Blastodisk and this time the egg is a two-

cell blastomeres. Blastomeres at this stage are almost spherical and transparent.

4-cell blastomeres: the eggs contain four similar cell blastomeres. In this stage, the blastomeres lost their spherical shape and form to be drawn (Figure: 1-C).

8-cell blastomeres: In these stage the eggs contain eight cells care in two rows of are arranged together in a foursome. The blastomeres at this stage are similar to each other (Figure: 1-D).

16- cell blastomeres: The egg is a sixteen-cell blastomeres at the four rows of four have been together wand. Sixteen-cell stage blastomeres have different outward forms (Figure: 1-E).

32- cell blastomeres: The eggs is a thirty-two-cell blastomeres. Unlike in the earlier stages, blastomeres are not regularly together (Figure: 1-f).

Morulla: solid mass of blastomeres resulting from a number of cleavages of a zygote, or fertilized egg. Its name derives from its resemblance to a mulberry (Latin: *morum*). A morula is usually produced in those species the eggs of which contain little yolk and, consequently, undergo complete cleavage (Figure: 1-G).

Early Blastula Stage: In eggs of yellow fin sea bream, blastula stage, blastomeres form a cell layer

that is transparent and colorless in the animal pole and the yolk to form a hollow sphere or hemisphere can be observed (Figure: 1-H).

Late Blastula Stage: At this stage of embryonic development of yellow fin sea bream, height and thickness of the reduced cell mass and its size will increase. At this stage, a layer of embryonic (Epiboly) in animal cells is formed and develops as a pod and the blastopore grow (Figure: 1-K). Unlike the previous steps in this phase, cell division (consequently fetal growth layer) is more uniform. Mitotic divisions because of the inconsistency phenomenon that does not happen in a short timeframe (Kimmel *et al.*, 1995).

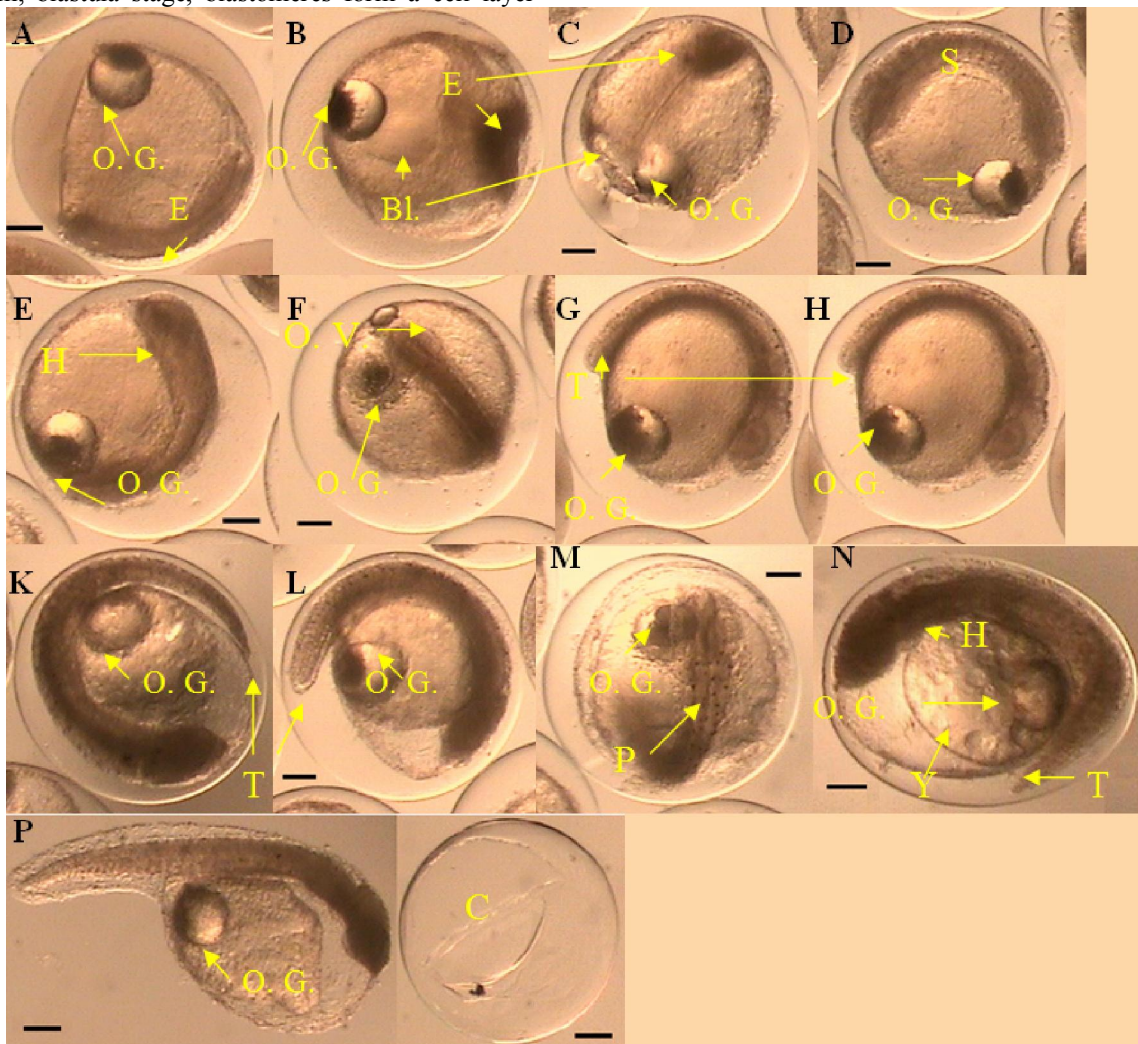


Fig. 2 (A to P): Yellow fin sea bream embryonic stages, left to right A: Observation Embryo Profile; B & C: Closing of blastopore; D: The Formation of Somite; E: The Appearance of Heart; F: The Formation of Optic Cup; G & H, K, L: Tail bud period; M: Increasing of Pigmentation; N: Start of dezhatching; P: Hatching (Abbreviations, Bl: Blastopore, O.G.: Oil Globule, S: Somite, H.: Heart, O. V.: Optic Vesicle, S. T.: Tail, P: Pigment, Y: Yolk sack, C: Corion)

Early gastrula: Blastulation when finished. The egg stage is Gastrulation (Figure: 1-L). The embryo at this stage is called embryonic shield (Kimmel *et al.*, 1995).

Gastrulation 1/2: The growth of the embryo in the egg yolks and half the fish to form a hemisphere around the yolk is visible (Figure: 1-M). In these stage emboly layer has two layers.

Neurula: End of gastrulation and organ regeneration

(Organogenesis) begins in the egg. At this time the nerve cord (Neural tube) will be back in the ectoderm (Figure: 1-N).

Observation Embryo Profile: *A. latus* embryos at this point in the profile appear. Notochord is formed in this step (Figure: 2-A). Embryos at this stage is completely immobile and critical activities are performed as before (Nutrition of the yolk and breathing is performed on all surfaces). The head is thicker than the other end of the embryo.

Closing of blastopore stage:

Blastopore a roughly circular area in the egg yolk is teleost before the end of the embryo does not cover any Epiboly, so close blastopore, as well as all the steps epiboly. At this stage the embryo is fully developed, so that the sheath surrounding the yolk into the hole to block blastopore (Figure: 2-B, C).

The Formation of Somite stage: Somitogenesis occurs at this stage (Figure: 2-D). During embryonic development the somites that create organs such as the spine, machine shed, and other digestive tract is the body.

The appearance of heart stage: The heart appears. Heart is positioned just below the head and the throat (Figure: 2-E).

The Formation of Optic Cup stage: The eye vesicle appears to be two clear oval bubble. The tail bud at the end of the embryo starts to grow (Figure: 2-F, G, H, K, L).

Increase of Pigmentation stage: The sensible way than before, increasing the density of pigment was observed. The eyes may also develop, as the eyes of fetus has circular lens (Figure: 2-M).

Start of hatching (10%) stage: The whole fish embryos and larvae can become. Fetal movements and pressure into the corion eggs out of a perfectly spherical and are roughly oval or ovoid (Figure: 2-N).

Hatching (100%) stage: At this stage, all eggs hatched and the corion is left.

Oral and anal newborn larvae is closed, So they do not have the ability to get foreign food and their only source of food and energy supplies is the yolk sac and oil globule (Figure: 2-P).

In our study, egg and oil globule diameters (Table 1) were lightly smaller to another sparidae species were mesard, for example: those obtained by artificial fecundity of red porgy captured in Argentine shelf and described by Ciechowski & Weiss (1973) and Kentouri *et al.* (1992), the egg size from Mediterranean Sea red porgy, obtained by natural spawning. The range of egg sizes could change according to the period of observations within the spawning season and the different considered areas (Laura *et al.*, 2003).

Table 1: Components eggs and oil globule diameter in different studies

Species	Egg diameter (μm)	Oil globule diameter (μm)	Author
<i>Pagrus pagrus</i>	810-880	176-194	Ciechowski & Weiss (1973)
	980-1025	250 \pm 7	Kentouri <i>et al.</i> (1992)
	890-930	180-200	Machinandiarena <i>et al.</i> (2003)
<i>Pagrus major</i>	660-1030	250	Fukuhara (1985)
	940-1050	-	Estévez (1991)
	900-1100	-	Pillay (1995)
<i>Sparus aurata</i>	1000-1030	220-250	Méndez (1994)
	1.001 \pm 0.005	0.217 \pm 0.001	Okan Kamacı <i>et al.</i> , 2005
<i>Archosargus rhomboidalis</i>	800-940	800-940	Houde & Potthoff (1976)
<i>Acanthopagrus latus</i>	740.33 \pm 0.012	185 \pm 0.005	This study

Also less than average diameters of Gilthead Sea Bream eggs were determined Okan Kamacı *et al.* in 2005, approximately 1.001 \pm 0.005 mm and 0.217 \pm 0.001 mm, respectively. Egg and oil globule diameters measured in their study were relatively similar with the other author's results (Uçal, 1983; Glamuzina *et al.*, 1988; Camus and Koutsikopoulos,

1984). It might be thought that natural conditions (temperature, food, geographic location) and broodstock management (salinity, temperature, nutrition) in captivity could be effected broodstock and egg quality (Okan *et al.*, 2005).

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