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ISSN 0792 - 156X

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PUBLISHER:
Israeli Journal of Aquaculture - BAMIGDEH -
Kibbutz Ein Hamifratz, Mobile Post 25210,
ISRAEL

Phone: + 972 52 3965809

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Short Communication

Embryonic and Pre-Larval Development of Shabbout (*Barbus grypus* H.)

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(Received 1.4.07, Accepted 4.7.07)

Key words: *Barbus grypus*, shabbout, artificial insemination, embryo development

Abstract

As a preliminary step towards assessing its aquaculture potential, egg development and artificial breeding in shabbout (*Barbus grypus* Heckel 1843) was studied. Artificial insemination was achieved by mixing eggs and a sperm-testes extract. The fertilization rate was 60%. Diameters of fertilized eggs ranged 2.58-2.70 mm. The perivitelline space formed 20 min after insemination and contained no oil droplets. A blastodisc split two blastomeres of nearly equal size 80 min after insemination and first cleavage occurred 4-4.5 h after insemination. The gastrula stage was completed after 12.5 h and the embryonic body formed after 14 h. The first somites were observed at 28 h and the first heartbeat at 72 h. The first hatched larva appeared at 84 h and all eggs hatched within 92 h. The mouth opening occurred after 188 h. Head pigmentation was nearly complete together with the formation of the tail fin by 480 h, after which the tail fin gained its homocercal formation and the pigmentation spread throughout the body.

Introduction

The Talmud, a massive Jewish work completed in Persia approximately 1500 years ago, contains references to a fish named shibuta. There have been various attempts to identify it over the last several centuries (Zivotofsky, 2006). One possibility is the new aquaculture candidate, *Barbus grypus* (Heckel 1843). The

Barbus genus of Cyprinidae is widely distributed in eastern Asia, eastern Europe, and Africa. It is commonly called barb or shabbout, also spelled shabbout or shabut.

Barbus grypus is a vagile species that prefers rivers but is also found in estuaries. It is commercially fished and can reach nearly

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two meters and over 50 kg (Coad, 1996). Its growth, sexual maturity characteristics, and reproduction biology have been studied by Al-Hakim et al. (1981), Khalaf et al. (1984), Epler et al. (2001), Pyka et al. (2001) and Szypula (2001). Spawning generally occurs from May to mid June (Geldiay and Balik, 1988). The spawned eggs are scattered above aquatic plants and cling to the vegetation (Geldiay and Balik, 1988; Epler et al., 2001).

Like elsewhere, the aquaculture industry in Turkey is continually evaluating new candidates and systems to diversify its production as effectively as possible. Mastering of the reproduction cycle, including larval stages, is of great importance and basic for the adaptation of a new species to aquaculture (Kamler, 2005). Hence, the present study was carried out to determine the embryonic and pre-larval development of captive shabbout.

Materials and Methods

The study was conducted in 2005 at the Department of Fisheries of Harran University Bozova Vocational School with *B. grypus* caught with gill nets (80 x 80 mm) in Ataturk Dam Lake. Three mature female and three mature males (2.0-2.5 kg, 65-69 cm) were randomly selected and stocked in small tanks in natural lake water conditions (23.0°C, 8.3-9.1 mg/l oxygen, pH 8.5, salinity 0.2%). Scales were removed from the lateral line and dorsal fin to determine age (Lagler, 1966).

Artificial insemination was achieved by mixing 200 eggs from the females with sperm from the males. Lake water was added to cover the egg surfaces and one-third of the water was replaced every 2 min several times. Egg stickiness was neutralized according to Al Hazzaa and Hussein (2003) and the eggs were placed in a Zuger glass for incubation. Water circulated by aeration.

The embryonic development of the fertilized eggs was examined under a microscope and unfertilized eggs were removed daily. Examination of egg stage and measurements were performed with an ocular micrometer at x10 magnification and photos were taken with a stereo-microscope (Nikon SMZ 2 T stereo).

Results

The age of the fish ranged 6-7 years. The fertilization rate was 60%. Diameters of fertilized eggs ranged 2.58-2.70 mm. Eggs with a chorion were spherical, transparent, and without an oil globule. Egg development is shown in Fig. 1. The gastrula stage was completed 12.5 h, the embryonic body formed 14 h, the first somites were observed 28 h, and the eyed egg stage was first observed 48 h after insemination. Newly hatched free larvae were slightly pigmented. Absorption of the yolk sac occurred approximately 166 h after insemination. The mouth opening occurred after 188 h. Head pigmentation was nearly complete together with the formation of the tail fin after 480 h.

Discussion

Egg size is a key feature in the early history of fish. It may be expressed as egg diameter, egg volume, wet weight, dry weight, energy content per egg, or content of a key substance such as carbon, nitrogen, or protein (Kamler, 2005). The length and diameter of the fertilized shabbout eggs were similar to those of the himri barbel (*Barbus luteus* Heckel; Al Hazzaa and Hussein, 2006).

The embryonic stages were typical of most cyprinids. However, in bouni (*Barbus sharpeyi*), two and four blastomere stages occurred 20 and 30 min after fertilization, the multi cellular (morula) stage was observed 9.5 h after fertilization, and the first embryonic movements and heartbeat were observed after 65 h of incubation (Pyka, 2001). In our study, hatching occurred 84-92 h after insemination while Pyka (2001) reported that it began after 96, 81, and 72 h of incubation in shabbout, bouni, and gattan (*Barbus xanthopterus*), respectively. Morphological and physiological characters of fish larvae vary considerably during development (Blaxter, 1986). Alami-Durante (2000) found that the quantity and quality of yolk reserves of embryos are not identical, causing differences in the endogenous potential of embryos for growth.

The newly hatched free larvae were pigmented and the yolk sac was absorbed approximately 166 h after insemination, similar to findings of Pyka (2001). However, in the

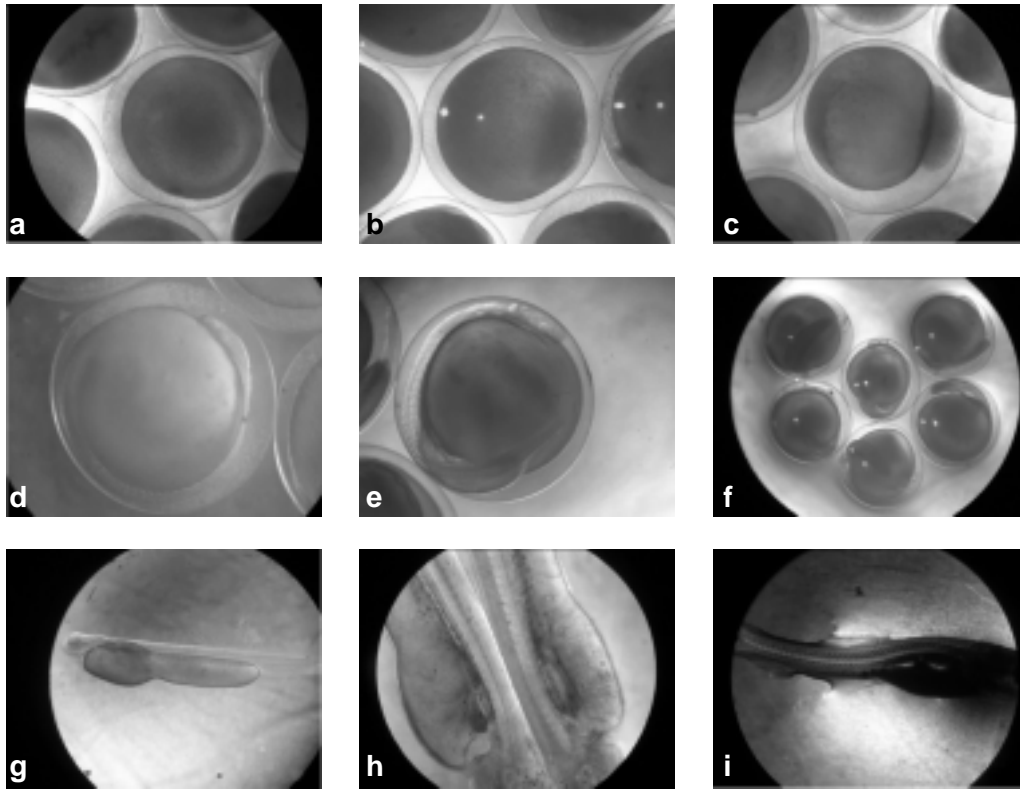


Fig. 1. Development of fertilized eggs in *Barbus grypus*: (a) formation of perivitelline space, 20 min after insemination, (b) 2-cell stage, 80 min after insemination, (c) morula stage (blastomeres), 4-4.5h after insemination, (d) head region started to be shaped, 32 h after insemination, (e) eye formation (otic vesicle), 34 h after insemination, (f) first heartbeat and embryonic movement, 72 h after insemination, (g) newly hatched larvae, 84-92 h after insemination, (h) air bladder and body pigmentation, 172 h after insemination, and (i) intestinal formation, 720 h after insemination.

himri barbel, the yolk sac was absorbed earlier (Al-Hazza and Hussein, 2006), possibly due to an insufficient availability of good quality food. Absorption in shabbout was first observed 188 h after insemination, as reported for other marine and freshwater species (Blaxter, 1969; Economou et al., 1991).

In conclusion, artificial fertilization and embryonic development of shabbout were achieved but further investigation and development are required before this technique can be used in aquaculture.

Acknowledgements

We gratefully acknowledge the valuable comments of Prof. Necmettin Tekin, Prof. Selcuk Secer, and Assoc. Prof. Hasan Atar of the Faculty of Fisheries at Ankara University in Ankara. We gratefully acknowledge the valuable comments of Assoc. Prof. Hasan Musa Sari and Asst. Prof. Tulin Coker of the Faculty of Fisheries at Ege University in Izmir. Also, we thank the Harran University Bozova Vocational School Department of Fisheries Directorate in Sanliurfa, Turkey, for equipment and chemical support.

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