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Embryonic Development of Barbel (*Barbus barbus*)

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Key words: *Barbus barbus*, barbel, embryo, development

Abstract

Barbel (*Barbus barbus* L.) is rarely bred and reared in hatcheries, and data on the early development of this species are scarce. Thus, the aim of the study was to describe its embryonic development in detail. Eggs and sperm were obtained from artificially stimulated spawning. Fertilized eggs were incubated in ten 2000-ml aquaria filled with aerated dechlorinated tap water and maintained at a constant 18°C, the optimal temperature for embryonic development of barbel. The eggs swelled to a maximum of 18% during the first hour after fertilization. There were eight distinct stages of embryonic development: two blastomeres, eight blastomeres, small-celled blastula, embryo body formation, body segmentation, formation of brain and eye germs, change of yolk sac shape, and first movement of the embryo. Survival during development was over 81% and during hatching 74%. Of the newly hatched larvae, 88% were normal, 7% were dead, and only 5% had morphological abnormalities, the most common of which were yolk sac malformations, spinal cord curvatures, and heart edema.

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Introduction

Barbel (*Barbus barbus*) is a member of the Cyprinidae family. The genus includes over one thousand species and sub-species which are widely distributed throughout Europe, central Asia, and Africa. Barbels are usually found in gravel and rocky-bottomed, fast-flowing waters with a high dissolved oxygen content. Barbels have two pairs of barbs, a longer pair points forwards and slightly downwards and is positioned on the side of the mouth, and a smaller pair is located under the chin. The fish received its name from the Latin *barba*, meaning beard, a reference to the hair-like feelers (also known as barbels) growing around the mouth.

Barbel is rarely bred and reared in hatcheries and data on the early development of this species are scarce. Morphological characteristics of the early development stages, including embryo, larvae, and first juvenile stage, have been described under experimental conditions (Krupka, 1988), as have selected aspects of incubation and hatching (Korwin-Kossakowski et al., 1998). The egg development of shabbout (*Barbus grypus*) was described as a preliminary step towards assessing its aquaculture potential (Sahinoz et al., 2007). The larval development of barbel was examined by measuring their wet and dry weight, whole body mineral contents, skeletal and gill development (Calta, 1998).

The aim of present study was to describe in detail the embryonic development of barbel under controlled rearing conditions.

Materials and Methods

Eggs and sperm were obtained from artificially stimulated spawning in the Inland Fisheries Institute in Zabieniec on March 6, 2008, and transported in a cold box (5°C) to the laboratory of the Department of Animal Physiology in Siedlce. The eggs from three females and milt from three males were mixed with a small amount of water for 3 min and fertilization took place two hours after spawning. Fertilized eggs were incubated in 2000-ml aquaria (four replicates of 80 embryos per aquarium) filled with aerated dechlorinated tap water. A thermostat, placed in the water, maintained a constant temperature of 18°C.

The diameters of 25 whole eggs and their yolks were measured hourly under a stereoscope (12 x 1.6 magnification) during the first four hours after fertilization. The percent increase of the egg diameter (swelling) was calculated as: $x = (c - d) \times 100/d$, where x is the swelling (in %), c is the egg diameter, and d is the yolk diameter.

The embryos were observed daily to evaluate the rate of development. Dead embryos were counted and removed at four embryonic stages (metamere formation, formation of eye and brain germs, body movement, before hatching) to calculate embryo survival. Newly hatched larvae were counted and inspected. The hatching rate was calculated as the percent of eggs that hatched, assuming the initial number of incubated eggs was 100%.

Hatched larvae were observed to evaluate their quality. The larvae were divided into three groups: normal (live, motile, without visible anomalies), deformed (live, moving erroneously, showing body malformations), and dead (immobile, opaque, whitish). The proportion of each group among the entire pool of hatched larvae was calculated. Embryos and larvae were observed using a computer MultiScan image analysis system and a stereoscopic microscope that allowed photographs to be taken. Results concerning egg swelling and embryonic survival were subjected to Student's *t* test to evaluate the significance of differences ($p<0.05$).

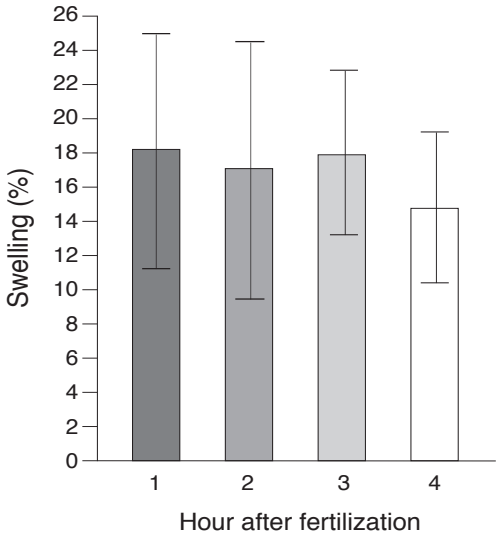

Fig. 1. Swelling of barbel eggs.

Results

The barbel eggs swelled a maximum of 18% within the first hour after fertilization (Fig.1). Cleavage started two hours after fertilization, then the blastodisc divided into the first two blastomeres (Fig. 2). Eggs reached the 8-blastomere stage three hours after fertilization. As a result of continual cell division, the small-cell blastula developed ten hours after fertilization. The blastoderm covered almost half the egg 29 h after fertilization, and the embryo body formed. Some somites were discernible 51.5 h after fertilization. The germs of eyes and brain appeared 53.5 h post fertilization. The body became elongated due to the rapid growth of the tail and a change of the yolk sac shape (it differentiated into an enlarged anterior section and a narrow posterior section) 73 h post fertilization. The first movement of the embryos was observed 110 h after fertilization. The first individuals hatched 116 h after fertilization and the last ones 141 h after fertilization.

Embryo survival during development was over 81%. During hatching it dropped to 74%, but the difference was statistically insignificant (Fig. 3). Among the newly hatched larvae, 88% were normal, 7% were dead, and 5% had morphological abnormalities. The most common abnormalities were yolk sac malformations, spinal cord curvatures, and heart edema.

Discussion

In the present study, the phytophilous barbel eggs reached the maximum swelling (18%) one hour after fertilization. The increase in egg diameter in other phytophilous fish such as the common carp (*Cyprinus carpio*) reached about 40% (Witeska et al., 1995; Jezierska and Slominska, 1997; Calta, 2001; Jezierska et al., 2001). Pelagic eggs of grass carp (*Ctenopharyngodon idella*) swelled by 200-250% within 5-6 h from fertilization (Jezierska et al., 2001).

In the present study, embryonic stages were based on the description of barbel development by Krupka (1988) and *Barbus grypel* by Sahinoz (2007). They are similar to those described for other *Cyprinidae* species, even those not closely related to barbel. The same stages of gastrulation (formation of 2, 4-8 blastomeres, and blastula) and organogenesis (beginning of embryo formation, formation of eyes) were observed in common carp (Witeska et al., 1995), *Candidia barbatu*s (Sado and Kimura, 2002), and *Barilius canarensi*s (Sado and Kimura, 2005).

The high embryo survival (81%) resulted in a high hatching (74%) rate. Similar values were obtained in studies carried out under hatchery conditions for common carp, where the hatching rate was 79-80% (Witeska et al., 1995; Calta, 2001) and 98% (Lugowska and Jezierska, 2000). The proportion of normal fry (88%) with a small amount of dead and deformed hatch is similar to other *Cyprinda*e fish incubated under optimal laboratory conditions. The share of normal larvae was 75-92% in common carp (Lugowska and Jezierska, 2000) and 45-73% in grass carp 45-73% with only 9-14% deformed (Lugowska et al., 2002) .

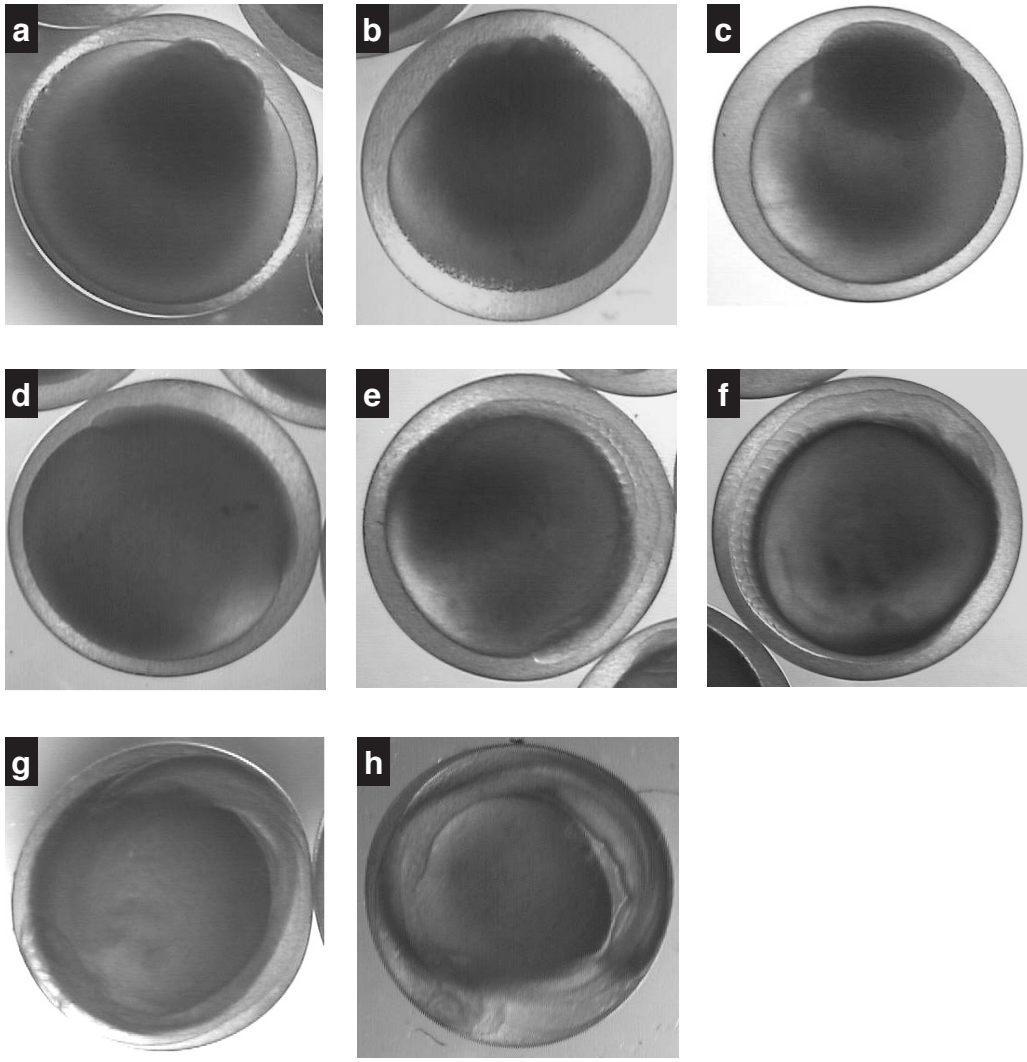

Fig. 2. Stages of barbel embryonic development: (a) two blastomeres, (b) eight blastomeres, (c) small-cell blastula, (d) formation of embryo body, (e) formation of metameres, (f) eye germs and brain formation, (g) body elongation, (h) body movement.

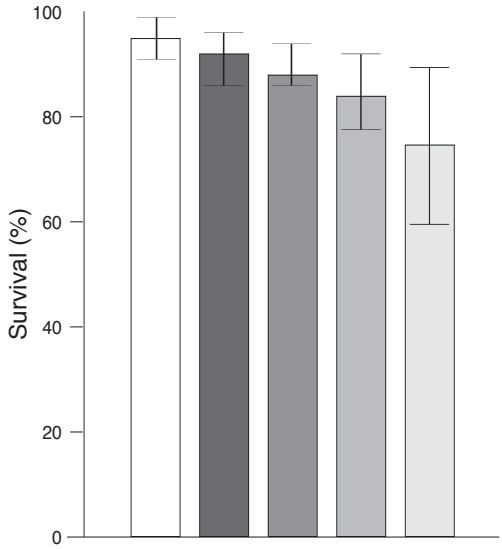

Fig. 3. Survival of barbel embryos in the metamere, developed eyes and brain, body movement, prehatching, and post hatch stages.

Acknowledgements

The experiments comply with current Polish law (Certificate of Permission from the III Local Ethical Committee No. 25/2007).

References

Calta M., 1998. Early development of barbel (*Barbus barbus* L.) larvae. *Turk. J. Zool.*, 22: 17-22.

Calta M., 2001. Effects of aqueous cadmium on embryos and larvae of mirror carp. *Indian J. Anim. Sci.*, 71:885-888.

Jezierska B. and I. Slominska, 1997. The effect of copper on common carp (*Cyprinus carpio* L.) during embryonic and postembryonic development. *Pol. Arch. Hydrobiol.*, 44: 261-272.

Jezierska B., Witeska M. and K. Lugowska, 2001. The effect of temperature on swelling of unfertilized eggs of common carp and grass carp. *Folia Univ. Agric. Stetin.* 218, *Piscaria*, 28:21-28.

Korwin-Kossakowski K., Wolnicki J., Kaminski R. and L. Myszkowski, 1998. Incubation of *Barbus barbus* (L.) eggs - rare in Polish hatcheries. *Kom. Ryb.*, 5:1-3 (in Polish).

Krupka I., 1988. Early development of the barbel. *Prace Ust. Rybar. Hydrobiol.*, 6:115-138.

Lugowska K. and B. Jezierska, 2000. Effect of copper and lead on common carp embryos and larvae at two temperatures. *Folia Univ. Agfic. Stetin.*, 205 *Piscaria*, 26:29-38.

Lugowska K., Jezierska B., Witeska M. and P. Sarnowski, 2002. Deformations of newly hatched grass carp larvae. *Acta Sci. Pol., Piscaria*, 1(1):15-22.

Sado T. and S. Kimura, 2002. Developmental morphology of the cyprind fish, *Candidia barbatus*. *Ichthyol. Res.*, 49:350-354.

Sado T. and S. Kimura, 2005. Developmental morphology of the Indian Cyprinidae fish *Barilius canarensis*. *Ichthyol. Res.*, 52:360-363.

Sahinoz E., Dogu Z. and F. Aral, 2007. Embryonic and pre-larval development of shabbout (*Barbus grypus* H.). *Isr. J. Aquac. - Bamidgeh*, 59(4):235-238.

Witeska M., Jezierska B. and J. Chaber, 1995. The influence of cadmium on common carp embryos and larvae. *Aquaculture*, 129:129-132.