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Contribution to the biology of Zagros tooth-carp, *Aphanius vladykovi* (Cyprinodontidae) in central Iran

Yazdan Keivany & Nasrollah M. Soofiani
Department of Fisheries, Faculty of Natural Resources, Isfahan University of Technology, 84156 Isfahan, Iran (e-mail: keivany@cc.iut.ac.ir)

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Synopsis

We studied some aspects of the biology of the Zagros tooth-carp, *Aphanius vladykovi*, an endemic and poorly known species from Chahar-Mahall-va-Bakhtiari province in central Iran, by regular monthly collections and direct observation in aquaria over a full year. We collected individuals from Modar-Dokhtar spring of Gandoman region; we preserved some in formalin and transferred some alive to aquaria. The stomach contents consisted mostly of freshwater crustaceans, suggesting a carnivorous habit. The eggs had an average diameter of 1 mm and the average absolute and relative fecundity was 415 and 110, respectively. The gonadosomatic ratio and ovary condition suggested that the reproductive season of the species was between late March and June with a peak in early April. The species is euryhaline and eurythermal and prefers neutral to basic waters. It is usually found in well-oxygenated waters, but is tolerant of hypoxia as well.

Introduction

The Cyprinodontidae are represented in Iran by six named species (Scheel 1990, Wildekamp 1993, Coad 1995, Coad 1996, Keivany & Soofiani 2002). The Zagros pupfish, *Aphanius vladykovi*, is an endemic species found in a restricted area of the central Zagros Mountains of Iran and is an attractive species for keeping in aquaria. However, its biology is unknown and there is little information on this species beyond the original description (Coad 1988) and some brief biological observations (Coad & Keivany 2000).

This species is a close relative of *A. sophiae*, but characterized by a higher lateral line scale counts and by its color pattern: males have darker coloration on the dorsal fin and a lighter one on the anal fin. Females lack the typical, large, lozenge-shaped spot at the base of the caudal fin seen in *A. sophiae* (Coad & Keivany 2000). The main objective of this study was to provide more data on some aspects of the biology of this species for conservation purposes and possibly, introduction to aquarium hobbyists.

Material and methods

Study area and collecting methods

The main collection site was Modar-Dokhtar spring, near the town of Gandoman (31°52′12″ N, 51°08′29″ E) in the province of Chahar-Mahall-va-Bakhtiari (Figure 1). We made regular monthly collections (twice in April) from this site. We caught the individuals with a small seine net (1 mm mesh size, 5 m length, and 1.5 m height) and each time we preserved 20 of them (10 female and 10 male) in 5% formalin (a total number of 260 specimens) and kept others alive in polyethylene bags and later transferred them to aquaria for further investigations. We carried out

Figure 1. The known distribution and collection sites of the Zagros toothcarp. The arrow shows the Modar-Dokhtar spring. This species is recorded from Behesht-Abad River for the first time in the present study.

measurement of chemical and physical properties of water at each sampling date using Ciba Corning EC/pH/TDS meter, WTW oxygen meter model OXI196, and an ordinary thermometer.

Biometry, feeding and reproduction

After measuring the length and mass of all the preserved specimens (260), we opened their stomachs to examine the contents. We calculated the hepatosomatic index (HSI), monthly to determine the feeding status of the fish throughout the year by

$$\text{HSI} = (\text{wet weight of liver}/\text{total wet weight of fish}) \times 100$$

We calculated the gastrosomatic index which is widely used to estimate the feeding intensity of fishes (Desai 1970), monthly by

$$\text{GSI} = (\text{wet weight of gut}/\text{total wet weight of fish}) \times 100$$

and the relative length of gut (RLG) monthly following Biswas (1993) by

$$\text{RLG} = (\text{length of gut}/\text{standard length}) \times 100$$

We examined the ovaries and calculated the absolute fecundity (total number of ova), relative fecundity (number of ova per unit of body weight), and total working fecundity (the free and ripe ova) by method of Bagenal (1967). We calculated the gonadosomatic ratio (GSR) (Nikolsky 1963) or maturity index, an indirect method for estimating spawning period (Hopkins 1979), for each month, but twice in April, by

$$\text{GSR} = (\text{wet weight of ovaries}/\text{total wet weight of fish}) \times 100$$

We kept live specimens in a 40 l aquarium and fed them aquarium fish flakes and live food. We studied the reproductive behavior by direct observation.

Results

Measurements

Maximum size recorded was 76 mm in total length and 7.10 g in mass for females, and 58 mm in total length and 3.70 g in mass for males. The general relationship between weight and length of captured fish, representing a sample taken from the wild at different times of the year, took the following logarithmic form:

$$\text{Log}(W) = -3.86 + 2.58\ \text{Log}(\text{SL})$$

$$(\text{df} = 1, 258;\ F = 2262;\ p < 0.0001)$$

Feeding

The main recognisable food item found in guts of all the specimens from Modar-Dokhtar spring was the freshwater crustacean, *Gammarus* sp., which is abundant in the spring pool. However, in one specimen caught in the river, we could recognize some algae, *Cyclops* sp., snail eggs, and even fish larvae. The gastrosomatic index ranged from 1.8 to 7.3 and the lowest value occurred in late April (Figure 2). The HSI ranged from 1.3 to 2.2 and we recorded the lowest value in late April, 2 weeks after the peak of reproduction time and the highest value occurred in late November (Figure 2). The relative length of gut was between 0.4, in larger specimens, and 1.1 in smaller ones, suggesting a more omnivorous diet in juveniles.

Spawning and reproduction

The absolute fecundity ranged from 220 to 650 (415 ± 169). The relative fecundity was 45–155 (110 ± 25) per gram of body mass. However, the working fecundity was much lower (absolute = 110, relative = 73). Size of the ripe eggs was 0.8–1.2 mm (1 ± 0.1). Based on our observations, eggs are adhesive and are attached to aquatic plants in small patches (varying from 3 to 30). The GSR was used as an indicator of fish spawning time. This ratio ranged between 2.7 and 12.4, with the highest value in early April (Figure 2). After that, the index decreased to 6.6 in late April. This suggests that the peak reproduction time is in early April.

Mature fish in aquaria performed courtship behavior including color changes in males (darker dorsal fin, brighter stripes and lighter yellow body pigmentation), chasing the female by the male and pushing it to a corner, pectoral fin flipping, and shivering, but never led to spawning in aquaria, with one exception, where some batches of 3–10 eggs were found. These eggs never hatched. Nevertheless, on 8 May 2002, we extracted 31 ova and fertilized them with sperm of a freshly caught mature male from Modar-Dokhtar spring by pressing their bellies. The ova were fertilized in a petri dish and then kept in a small jar. The incubation temperature was 21–22°C; at this temperature, it took them 9–13 days to hatch

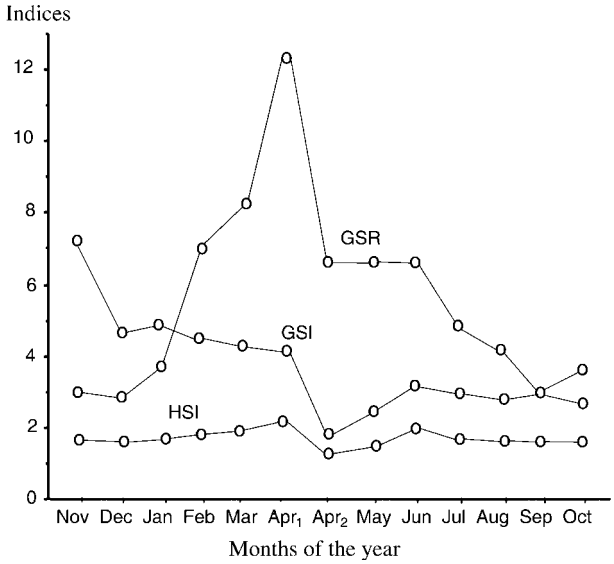

Figure 2. Variation of hepatosomatic index (HSI), gastrosomatic index (GSI), and gonadosomatic ratio (GSR) in Zagros toothcarp from November 2001 to October 2002 in Modar-Dokhtar spring (Apr_1 = early April, Apr_2 = late April).

(190–270°-h). After 3 days, some pigment appeared on the eggs. After 4 days, their eyes could be observed, and after 6 days, the embryo was almost fully shaped, occupying the whole egg space. After 11 days, amongst the 22 live larvae, four had still not hatched, four hatched, but still had their yolk sac, and 14 had absorbed their yolk sac. After 3 months, the young reached the size of 11–27.2 mm (18.5 ± 4.8) in standard length, suggesting individual differences in growth rate.

Habitat

Aphanius vladykovi is commonly found in fresh water with a conductivity of 240–280 μS (in aquaria they survived salinities up to 30 ppt). Temperature is almost stable in the spring (12.9–13.5°C), but in rivers it was found at a temperature as high as 19°C (in aquaria they survived temperatures up to 30°C), with pH 6.9–8.5, and TDS 125–138 mg l^{-1}. The habitat bottom is generally muddy, interspersed with pebbles. The water is clear in the Modar-Dokhtar spring and running slowly, but cloudy in the river. *Myriophyllum* sp. and *Potamogeton* sp. are the dominant aquatic plants in their habitat which are used as spawning substrate.

Discussion

The study of the life history of the fish shows that the maximum age is 2^{+} in both females and males. The seasonal growth period starts in June and continues until December and females reach larger sizes than males. The smaller size of the male could perhaps be due to the cessation of male growth at maturity and higher male mortality rates as reported for a population of *A. fasciatus* (Penaz & Zaki 1985). In general, as shown by the length-weight equation, there is allometric growth (Ricker 1979). The biology of this tooth-carp as in some related species (e.g., Vargas & De Sostoa 1997, Oltra & Todoli 2000, Caiola et al. 2001), is characterized by slow growth, low reproductive effort, and relatively late maturity.

Based on the jaw and gut morphology, RLG value, and studies on related species (Krupp & Schneider 1989, Huber 1996), this fish is expected to be carnivorous and surface feeder, however, in the Modar-Dokhtar spring where freshwater crustaceans are the dominant invertebrate, it mostly consumes this item which lives on or near the bottom. Nevertheless, omnivorous and herbivorous habits are also reported in some other *Aphanius* species (Huber 1996, Al-Daham et al. 1977, Krupp & Schneider 1989). According to our findings, this fish is a partial spawner that releases small batches of eggs between March and June. The release of eggs is greatest in early April as found in *A. fasciatus* (Leonardos & Sinis 1998). Such a lengthy breeding season is a kind of adaptation of short – lived small fishes to environmental conditions (Miller 1979). As the fish approach the peak of spawning, the intensity of feeding declines, followed by a decrease in HSI, suggesting the mobilization of the liver reserves toward the metabolic requirements of the fish during this period.

The distribution of the species was thought to be limited to waters neighbouring the town of Boldaji. This limited distribution could result in significant loss or even extinction if habitats are disturbed or destroyed, but we also found some populations in other water bodies such as the Gandoman wetland, the Chaghakhur wetland, Taqanak Bridge, 10 km southwest of ShahreKord (32°12′32″ N, 50°49′29″ E), Zanian stream (32°22′05″ N, 50°47′24″ E), and Behesht-Abad River in the province (Figure 1). These areas are small plains lying at altitudes above 2000 metres. Rivers in this area drain into Karun River which eventually discharges into the Persian Gulf. At the present, it seems that the species is in a good condition and can survive local adverse events. However, it should be noted that in the type locality (a large pool 3 km west of Boldaji) a mass mortality of this species was observed (for unknown reasons) in late March or early April (Coad & Keivany 2000).

The species is euryhaline and eurythermal and mostly found in neutral to basic waters. Although tolerant to hypoxia, it is usually found in well-oxygenated waters. Obvious sex determination, beautiful coloration, small size, high tolerance, and easy adaptation to aquarium conditions, make them a very suitable species for keeping in aquaria. However, we found it very susceptible to *Trichodiniasis* and *Ichthyophthiriasis*.

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