Folia Zool. – 57(1–2): 139–146 (2008)

First report on the spawning behaviour of a golden spined loach, *Sabanejewia vallachica* (Teleostei: Cobitidae)

Jörg BOHLEN

Institute of Animal Physiology and Genetics, Academy of Sciences of the Czech Republic,v.v.i., Rumburská 89, 277 21 Liběchov, Czech Republic; e-mail: bohlen@iapg.cas.cz

Received 23 September 2006 ; Accepted 21 March 2008

A b s t r a c t .Very little is known about the spawning behaviour of loach fishes, despite the fact that reproduction is one of the most important aspects in the autecology of animals. Here the spawning behaviour of golden spined loaches of the genus *Sabanejewia* is described for the first time. In opposite to spined loaches of the genera *Cobitis* and *Misgurnus*, the spawning partners do some 'circle swimming' together prior to spawning and the male does not form a full ring around the female but catches the female with a 'V'-shaped body. This spawning behaviour explains why males of *Sabanejewia,* unlike nearly all other genera of Cobitidae, 1) are not significantly smaller than females, a condition that is required only to form a complete ring around the female, and 2) have swellings in the middle of the body instead of specialised rays of the pectoral fins like in most other genera. These swellings are in the part of the body that touches the female during the release of gametes, while the pectoral fins do not reach to female's body.

Key words: ecology, reproduction, freshwater fish, sexual dimorphism

Introduction

Morphological characters are most important in the identification and taxonomy of fishes, and the only known facts about many loaches. In Cobitidae, the most prominent morphologic feature is the sexual dimorphism, which has been used to define genera and subgenera (N a l b a n t 1963, 1994). However, understanding the function of a morphological structure is a stronghold for practical use in taxonomy and ecology.

In nearly all genera of Cobitidae (spined loaches), a remarkable sexual dimorphism is present (Table 1), including different size of the sexes in at least 11 of 19 genera, an enlarged pectoral fin in males in at least 12 genera, ossified structures on the rays of pectoral fins (at least eight genera) and fleshy swellings on the side of the body (four genera). In many cases, the sexual dimorphism is used to diagnose genera, but nothing is known about the function of the elements of sexual dimorphism. The sexual dimorphism in *Sabanejewia* is exceptional among Cobitidae, since this genus does not show a size difference between males and females and the males have neither enlarged rays of the pectoral fin nor any structure on the pectoral fin, as is present in nearly all other genera of Cobitidae. Instead, during breeding season the males develop a vertical swelling of the body side that reaches from the margin of the dorsal blotches to the margin of the belly (Fig. 1). Unlike the vertical swelling in *Sabanejewia,* the swelling in *Lepidocephalichthys irrorata* and all species of *Koreocobitis, Misgurnus* and *Paramisgurnus* is a small horizontal stripe located slightly above the midlateral on the body sides shortly behind the height of the dorsal fin base (T s u k a h a r a 1948).

Pronounced morphological specialisations always evoke questions as to the functional background of the involved structures. In sexual dimorphisms, the function has to be searched for in field of reproductive features, especially since the sexual dimorphisms in

Table 1. Sexual dimorphisms within the family Cobitidae. Character stages are described for males; females lack this character. A lamina circularis is an ossified broadened piece of the base of the second (in some species also the first) ray of pectoral fins. An X indicates the presence of the charackter, – the absence and ? unclear cases.

Genus	Smaller than females	Second pectoral fin ray prolonged and/or thickened	Last pectoral fin ray thickened	One or two lamina circularis on pectoral fin rays	Serration on second ray of pectoral fin	Horizontal swellings on body sides behind dorsal fin base	Vertical swellings on body sides before dorsal fin base	Pigmentation pattern different from females	Source
Acanthopsoides	X	X	-	-	-	-	-	-	Roberts 1989
Acantopsis	X	X	-	-	-	-	-	-	Roberts 1989
Cobitis	X	X	-	X[2]	-	-	-	X[2]	Vladykov 1935, Bohlen 2000
Enobarbus[1]	?	-	X	-	-	-	-	?	Menon 1992
Iksookimia	X	X	-	X	-	-	-	-	Nalbant 1993
Kichulchoia	X	-	-	-	-	-	-	-	Kim et al. 1997
Koreocobitis	X	X	-	X	-	X	-	-	Kim et al. 1997
Kottelatlimia	X	X	-	-	X	-	-	-	Roberts 1989
Lepidocephalichthys	X	-	X	-	-	X[3]	-	X[2]	Roberts 1989, Menon 1992
Lepidocephalus	?	X	-	-	-	-	-	-	Roberts 1989
Misgurnus	X	X	-	X[2]	-	X	-	-	Vasil'eva 2001
Neoeucirrhichthys[1]	?	X	?	?	?	?	?	-	Menon 1992
Niwaella	X	-	-	-	-	-	-	-	Suzuki 1966
Pangio	-	X[2]	-	-	-	-	-	-	Roberts 1989
Paralepidocephalus	?	?	?	?	?	?	?	?	---
Paramisgurnus	X	X	-	X	-	X	-	-	Vasil'eva 2001
Protocobitis	?	X	-	-	-	-	-	-	Yang et al. 1994
Sabanejewia	-	-	-	-	-	-	X	-	Vladykov 1935
Somileptes	?	-	-	-	-	-	-	-	Menon 1992

[1] genus known from a single specimen only; [2] not all species; [3] only a single species

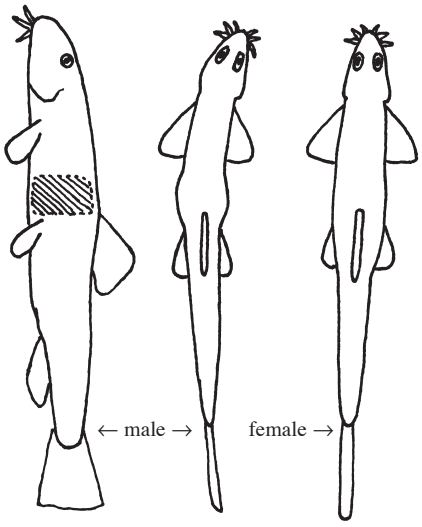

Fig. 1. Dorsal view of a pair of *Sabanejewia* during reproductive season, showing sexual dimorphism. The male bears a vertical swelling of the body sides at the level of the dorsal fin, but has no special structures on the pectoral fin rays and reaches a similar size than the female. The striped area indicates the position of the swelling in the horizontal view on the male.

Cobitidae are expressed only after fish have reached maturity and in some genera only during the reproductive season (swellings in *Misgurnus, Lepidocephalichthys, Paramisgurnus* and *Sabanejewia;* pigmentation patterns in some *Cobitis*). Unfortunately, nothing or very little is known about the reproduction of most genera of Cobitidae. Spawning behaviour has been described for *Cobitis* (B o h l e n 2000, L o d i & M a l a c a r n e 1990) and *Misgurnus* (K n a a c k 1960) as followed in both genera: the first visible behaviour is increased but undirected swimming activity, later one or more males follow the female and finally embrace the female in the way that a male lays around the female in a complete ring. In this position, gametes are released. The difference between *Cobitis* and *Misgurnus* lays in the choice of spawning site and egg size: while *Cobitis* spawns large non-sticky eggs inside clumps of very dense vegetation, *Misgurnus* spawns in open water and scatters the small, sticky eggs over the vegetation. Different egg surfaces were reported for different species of *Iksookimia*, leading P a r k & K i m (1996) to conclude that the species have different spawning biology. B o h l e n (2000) found the eggs of *Sabanejewia* as small and sticky and scattered across all kinds of substrate, and postulated that the spawning strategy of *Sabanejewia* can be expected to be different from that of *Cobitis*. However, due to the secretive nature of the loaches, spawning was never observed.

In the present paper, the spawning behaviour of *Sabanejewia vallachica* is described from direct observation. The spawning behaviour is taken to explain the functional background of the odd sexual dimorphism in *Sabanejewia*.

Material and Methods

Four pairs of *S. vallachica* were caught on 8 April 1998 in River Ialomitza at the village Crivina (type locality of *S. vallachica*) and kept in an aquarium 40x40x35cm under nearly natural conditions similar to those described in B o h l e n (1999). The aquarium was

equipped with a layer of fine sand, an air-driven sponge filter and floating plants (*Najas* sp.), a few plants in the ground (*Cryptocoryne affinis*), a clump of moss (*Vesicularia dubyana*) in one corner and some shelter in the form of plastic half-tubes. Fish were fed either live *Tubifex* worms or frozen chironomid larvae or food tablets for ornamental fish five times per week. On the morning of 27 June 2000 at about 8:00 a.m., unusual swimming activity of all loaches in the tank was observed. From 9:00 a.m., after the light of the aquarium switched on, the spawning of *S. vallachica* was observed and notes were taken while observing.

Results

Only one of the four females in the aquarium was spawning, the other three females did not show any other interest in the spawning event than feeding on the eggs. The fish were pigmented as usual, no change of colouration occurred during spawning as has been observed in some species of *Cobitis* (B o h l e n 2000). The spawning behaviour of *S. vallachica* can be divided into a number of elements:

S e a r c h – s w i m m i n g o f t h e f e m a l e (A i n F i g . 2)

Occasionally, the spawning female swam for one or two minutes slowly along the glass of the aquarium, the bottom, the filter and the plastic half-tubes, touching the substrate with the mouth. During this swimming, the female explored horizontal as well as vertical surfaces as well as interspaces between objects. Between the periods of swimming, the female rested on the bottom or joined the activities of the other fish (feeding on eggs or unspecific swimming).

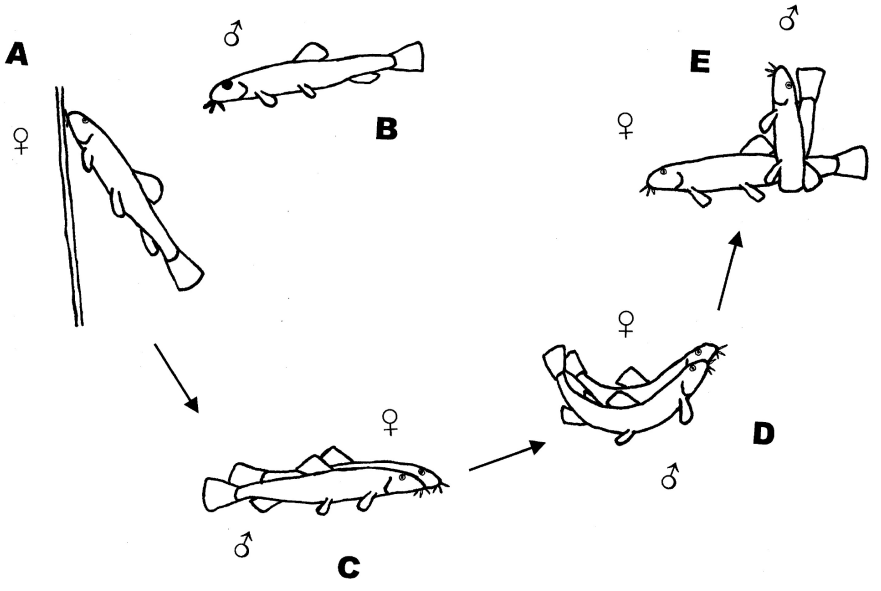

Fig. 2. Schematic drawing of the main behavioural elements during spawning of *S. vallachica*. A) Search-swimming female, the female swims slowly along structures and touches them with the mouth; B) Search-swimming male, the males swim around until they encounter another *Sabanejewia*; C) Approach by the male, the male places itself into parallel position with the female and presses with its head the operculum area of the female; D) Circling of pair, both partners rapidly swim in horizontal circles; E) Spawning, the males forms a 'V' with the female lying in the angle of the 'V'.

Search–swimming of the male (B in Fig. 2)

Also, all four males showed intense swimming activity along the structures in the aquarium with somewhat faster and more jagged movements than usual. Unlike the female, the males did not have contact with the surfaces during this swimming.

Approach by the male (C in Fig. 2)

Occasionally, when a male met another swimming specimen, independent of whether it was male or female, it approached it in a parallel position and pressed its head against the operculum region. Only swimming specimens (along the glass or over the ground) were approached, specimens resting on the ground were not. The approached specimens reacted in most cases either by pressing the approaching male away or avoiding the approaching male by a sudden change of swimming direction. The males did not focus on the spawning female with their approaching behaviour, but approached each specimen they met. Due to the increased swimming activity of the males, in most occasions the males approached other males, making it unlikely that the males can differentiate between males and females or that they are specifically attracted to the spawning female by specific signals such as behaviour, colour or smell. At this point, the behaviour of the males of *Sabanejewia* differs from that of *Cobitis*, since in *Cobitis* the males predominantly follow spawning females.

Circling of pair (D in Fig. 2)

When a male approached the spawning female during her search-swimming periods, the female reacted in four cases with avoidance of the male. However, in five cases, the female turned its head under the pressure from the male to the side opposite of the male and started to swim in narrow horizontal circles in the open water near to the substrate it had previously been investigating when the male approached. The males follow the female with the head still pressing the operculum region of the female. Since the female always started the circle by turning the head away from the male, the male was always on the outer side of the circling pair. On one occasion, a search-swimming male touched the side of the head of the spawning female by chance, and the female reacted by starting the circle-swimming, until it realised the lack of a male by its side. This observation shows that the press on the head is an intraspecific communication signal that prompts the female to start a ready-prepared behavioural element, in this case circle-swimming, but not that the pressure itself forces an otherwise straight swimming of the female into a circle. It further can be concluded that the male is following the female rather than leading it.

Spawning (E in Fig. 2)

The circle-swimming was rather rapid and lasted about two complete circles. Then the male slipped from the parallel position into a ‘V’ with head and caudal fin pointing upwards and the genital region of the female lying in the angle of the ‘V’. It appeared as if the male slipped its caudal peduncle under that of the female, but the movements were very fast and difficult to follow by eye. The male clamped the female tightly with its body sides and perhaps pressed the female gently, then the eggs were released. Afterwards, the fish separate and immediately start to search the ground under the spawning place for eggs. All specimens in the aquarium were intensely feeding on the eggs, including searching the surface of the filter,

the plastic half-tubes and the glasses, leaving not many eggs undetected. The whole process of circle-swimming and spawning took about two seconds. During each observed spawning event, about 30 to 40 eggs were released. Spawning took place at different places within the aquarium and at different distances from the filter current, indicating the water current is not important for the spawning fish. However, during the observation period with the aquarium lights switched off, spawning took place at the front glass in the lightest part of the aquarium, after the lights were switched on at the back glass in the darkest part of aquarium, indicating that there is an optimum light level for spawning. Spawning was repeated five times within one hour before an external disturbance stopped the spawning activity.

Discussion

The spawning behaviour of *Sabanejewia* differs in several important points from the spawning behaviour of the related genera *Cobitis* and *Misgurnus*. First, in both other genera, the males form a complete ring around the female, while in *Sabanejewia* the male forms a 'V' (Fig. 3). Furthermore, exploration of surfaces by the female or circle-swimming of pair were reported in neither *Cobitis* nor *Misgurnus*. Another difference regards the orientation of the two spawning partners to each other. During the approach, the male takes a parallel position to the female with its head touching the female at the operculum, while in *Cobitis* the male's head touches the belly of the female in the area of the ventral fins (B o h l e n 1999). In *Cobitis* and *Misgurnus*, the male changes from a parallel position to a ring by sliding its caudal peduncle across the caudal peduncle of the female, thus directing its dorsal fin towards the head of the female. In *Sabanejewia*, the male slips its caudal peduncle beneath the caudal peduncle of the female and thus ends up with its dorsal fin pointing towards the caudal fin of the female. However, due to difficulties in observation and the speed of the movements, it was not sure that the male in each spawning event was oriented in the same direction. A behaviour

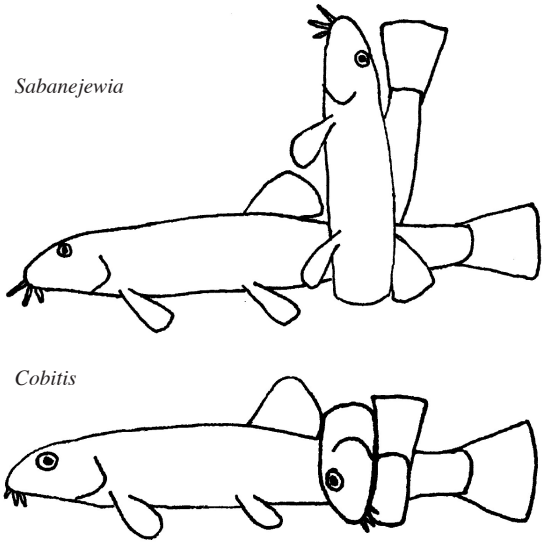

Fig. 3. Comparison of the 'V'-shaped spawning position in *Sabanejewia* with the complete ring which males of *Cobitis* form around their females during spawning. In *Cobitis*, the back of the male is directed towards the head of the female; in *Sabanejewia* the belly is directed towards the head of the female.

that does not directly belong to spawning is the intense egg cannibalism of *Sabanejewia*, in which even the spawning pair joined in immediately after the release of eggs. Such intense egg cannibalism has been observed neither in *Cobitis* nor *Misgurnus*. Similarities in spawning behaviour between *Sabanejewia* on the one hand and *Cobitis* and *Misgurnus* on the other regard the lack of interactions between males and between females.

Looking at the functional aspect of the described behavioural elements, the search-swimming of the female can be considered an investigation of the structures that surround the spawning place. At this moment, the female is able to make a choice of spawning environment by refusing spawning when the surroundings are not suited and to continue search-swimming until the surroundings are better suited. This possibility would represent a simple kind of parental care by choosing the environment the eggs and hatchlings have to face later. This hypothesis is supported by the observation that all spawning events took place at the aquarium glass, although also the filter, the bottom and the plastic half-tubes were explored during search swimming, indicating that some kind of choice is made. Unfortunately, no information about the spawning of *Sabanejewia* in nature is available. The search-swimming of the male represents a search for the female, although the way males are alarmed cannot be reconstructed from the present descriptions. The circle-swimming of the pair ensures that spawning takes place at a certain distance to the formerly investigated substrate.

Once the spawning behaviour is known, it is easy to understand the unusual sexual dimorphism in *Sabanejewia*. First, the males do not need to be smaller than the females because a smaller male is needed only for the formation of a tight ring around the female (due to the stiffness of a fish's body, overly large males could have problems bending the body enough to ensure a tight embrace of the female). However, in *Sabanejewia* this element of forming a ring is changed to forming a 'V', therefore there is no need to be smaller than the female. Second, the lack of any specialised structures on the pectoral fins of the males of *Sabanejewia* can also be explained by the new 'V'-shaped spawning position of the male. Since the pectoral fin is not in contact with the female, there is no need to have any specialised structures on them. This assumption leads to the conclusions that all Cobitidae with a specialised structure on the pectoral fin of males and males smaller than females can be expected to spawn in the way *Cobitis* does, however those genera or species without these elements of sexual dimorphism can be expected to spawn differently. It furthermore invokes the conclusion that specialised structures on the pectoral fins are important in intersex communication during embracement.

A c k n o w l e d g e m e n t s

I would like to thank T.T. N a l b a n t for help with obtaining the original specimens. This study was supported in part by grant 206/05/2556 of the Czech Science Foundation and of grant IAA600450508 of the Grant Agency of the Academy of Sciences of the Czech Republic. The Institute of Animal Physiology and Genetics receives continuous support from the Academy of Sciences of the Czech Republic (IRP lIAPG No. AV0Z50450515).

L I T E R A T U R E

Bohlen J. 1999: Reproduction of spined loach, *Cobitis taenia* (Cypriniformes; Cobitidae) under laboratory conditions. *J. Appl. Ichthyol. 15: 49–53.*

Bohlen J. 2000: Similarities and differences in the reproductive biology of loaches (*Cobitis* and *Sabanejewia*) under laboratory conditions. *Folia Zool. 49 (Suppl. 1): 179–186.*

Kim I.S., Park J.Y. & Nalbant T.T. 1997: Two new genera of loaches (Pisces: Cobitidae: Cobitinae) from Korea. *Trav. Mus. natl. Hist. nat. "Grigore Antipa" 34: 191–195.*

Knaack J. 1960: Beiträge zur Biologie und Parasitenfauna der mitteleuropäischen Cobitiden. Inauguraldissertation, *Mat.-Nat. Fak. Univ. Leipzig.*

Lodi E. & Malacarne G. 1990: Reproductive behaviour of the spined loach *Cobitis taenia* L. (Pisces, Cobitidae). *Ann. Sci. Nat. Zool. 13: 107–111.*

Menon A.G.K. 1992: Fauna of India. Cobitoidea: Cobitidae. *Zool. Survey of India, Calcutta.*

Nalbant T.T. 1963: A study of the genera of Botiinae and Cobitinae (Pisces, Ostariophysi, Cobitidae). *Trav. Mus. natl. Hist. nat. "Grigore Antipa" 4: 343–379.*

Nalbant T.T. 1993: Some problems in the systematics of the genus *Cobitis* and its relatives (Pisces, Ostariophysi, Cobitidae). *Rev. Roum. Biol. / Serie Biol. Anim. 38: 101–110.*

Nalbant T.T. 1994: Studies on loaches (Pisces, Ostariophysi, Cobitidae). I. An evaluation of the valid genera of Cobitinae. *Trav. Mus. natl. Hist. nat. "Grigore Antipa" 34: 375–380.*

Park J.Y. & Kim I.S. 1996: Adhesive membranes of egg in five cobitid species of *Iksookimia* (Pisces: Cobitidae). *Korean J. Zool. 39: 419–425.*

Roberts T.R. 1989: The freshwater fishes of western Borneo. *Mem. Calif. Acad. Sci. 14: 1–210.*

Suzuki R. 1966: Artificial spawning and early development of the loach, *Cobitis delicata* Niwa. *Bul. Freshw. Fish. Res. Lab., Tokyo 15: 175–188.*

Tsukahara H. 1948: The relation between the secondary sex-characters and the spawning habits in the loach, *Misgurnus anguillicaudatus* Cantor. *Seibutu 3: 64–69.*

Vasil'eva E.D. 2001: Loaches (genus *Misgurnus*, Cobitidae) of Russian Asia. I. The species composition in waters of Russia (with a description of a new species) and some nomenclature and taxonomic problems of related forms from adjacent countries. *J. Ichthyol. 41: 553–563.*

Vladykov V.D. 1935: Secondary sexual dimorphism in some Chinese cobitid fishes. *J. Morph. 57: 275–302.*

Yang J.X., Chen Y.R. & Lan J.H. 1994: *Protocobitis typhlops*, a new genus and species of cave loach from China (Cypriniformes: Cobitidae). *Ichthyol. Explor. Freshwaters 5: 91–96.*