Ichthyol Res (2013) 60:396–400
DOI 10.1007/s10228-013-0361-z

SHORT REPORT

# Polymorphic spawning-nest types in a local population of the fluvial sculpin, *Cottus nozawae* (Teleostei: Cottidae)

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Received: 27 February 2013 / Revised: 14 June 2013 / Accepted: 17 June 2013 / Published online: 1 August 2013


**Abstract** The nesting behavior of a fluvial sculpin *Cottus nozawae* population was investigated in a stream–reservoir system on the Sainai River, northern Honshu Island, Japan in 2003, 2004 and 2006. Except for the usual rock nest-spawning type (RSN), two new types of spawning nest were found: hole nest spawning (HNS) and crevice nest spawning (CNS). The HNS type was characterized by males digging one or more nest holes in the wall of the banks of the stream and reservoir, or on the sandy silt bottom of the reservoir. The CNS type involved the use of a crevice between large boulders or crevice of crag for their nests. The HNS type was dominant in the reservoir section, whereas the RNS type was dominant in the rapid riffles of stream sections. The HNS and CNS types appeared to be rarely or first observed in the *Cottus* species. These new nest-spawning types might have occurred in relation to the artificial environment changes in the river by dam construction.

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**Keywords** Nesting behavior · Hole nest spawning · Crevice nest spawning · Stream–reservoir system · *Cottus*

## Introduction

The quality of reproductive resources such as nest sites can be a constraint on an animal's reproductive success. A male defending a spawning nest may attract few mates and eventually lose his opportunity to mate. Such males are expected to move to a better nesting site if one is available (Wolf et al. 1997). According to this expectation, nesting males are more likely to move to an unexplored area to construct better nests when the current nest sites are restricted to a narrow area.

The genus *Cottus* consists of approximately 60 species (Scott and Crossman 1973; Lee et al. 1980; Goto 1990). Its reproductive style involves the typical nest spawning, with the male caring for the developing eggs and the larvae in some cases (Morris 1954; Goto 1982; Burr and Warren 1988; Takeshita et al. 1999). The river bullhead *Cottus gobio* is a typical rock nest spawner whose females deposit the eggs on the surface of rocks in rivers, usually during April. Thereafter, the nesting male takes care of the eggs by fanning and cleaning them, and defends the nest against intruders until hatching of the larvae (Morris 1954).

The fluvial sculpin *Cottus nozawae*, an endemic species to the northern part of the Japanese Archipelago, has been reported as a rock nest spawner that usually spawns in early spring (Goto 1975, 1982; Goto et al. 2001). Adult fish of *C. nozawae* preferentially inhabit slow riffles in the middle and upper courses of the rivers in Hokkaido Island, using moderately embedded cobble-sized rocks as spawning sites (Goto 1975, 1982). The polygenous mating system of *C. nozawae* is fundamentally similar to other *Cottus* species

(Brown 1982; Goto 1987, 1993; Marconato and Bisazza 1988; Takeshita et al. 1999).

Recently, we found the hole nest-spawning (hereafter, referred to as HNS) and crevice nest-spawning (CNS) types together with the usual rock nest-spawning type (RNS) in a population of *C. nozawae* in the Omono River system, the northern part of Honshu Island, Japan. This study examined the characteristics and frequency of occurrence of the three nest-spawning types, their association with the size of nesting males and physical environments of the nest sites. We discuss how the nest-type polymorphism has occurred, in relation to the artificial environmental changes by dam construction.

## Materials and methods

*Study sites.* The field research was conducted in the stream–reservoir system (39°32'14"N, 140°39'55"E) of the Sainai River (approximately 22 km long and with an area of 84 km$^2$), a secondary tributary of the Omono River basin of Akita Prefecture, Tohoku District, Japan, in 2003, 2004 and 2006 (Fig. 1a, b). The reservoir–stream system, which was formed by dam construction in 1993, is located approximately 12 km upstream from the confluence of the Sainai River with the Tama River, which is the first tributary of the Omono River (Fig. 1b, c). It is approximately 700 m long between dams (3–8 m in height) and consists

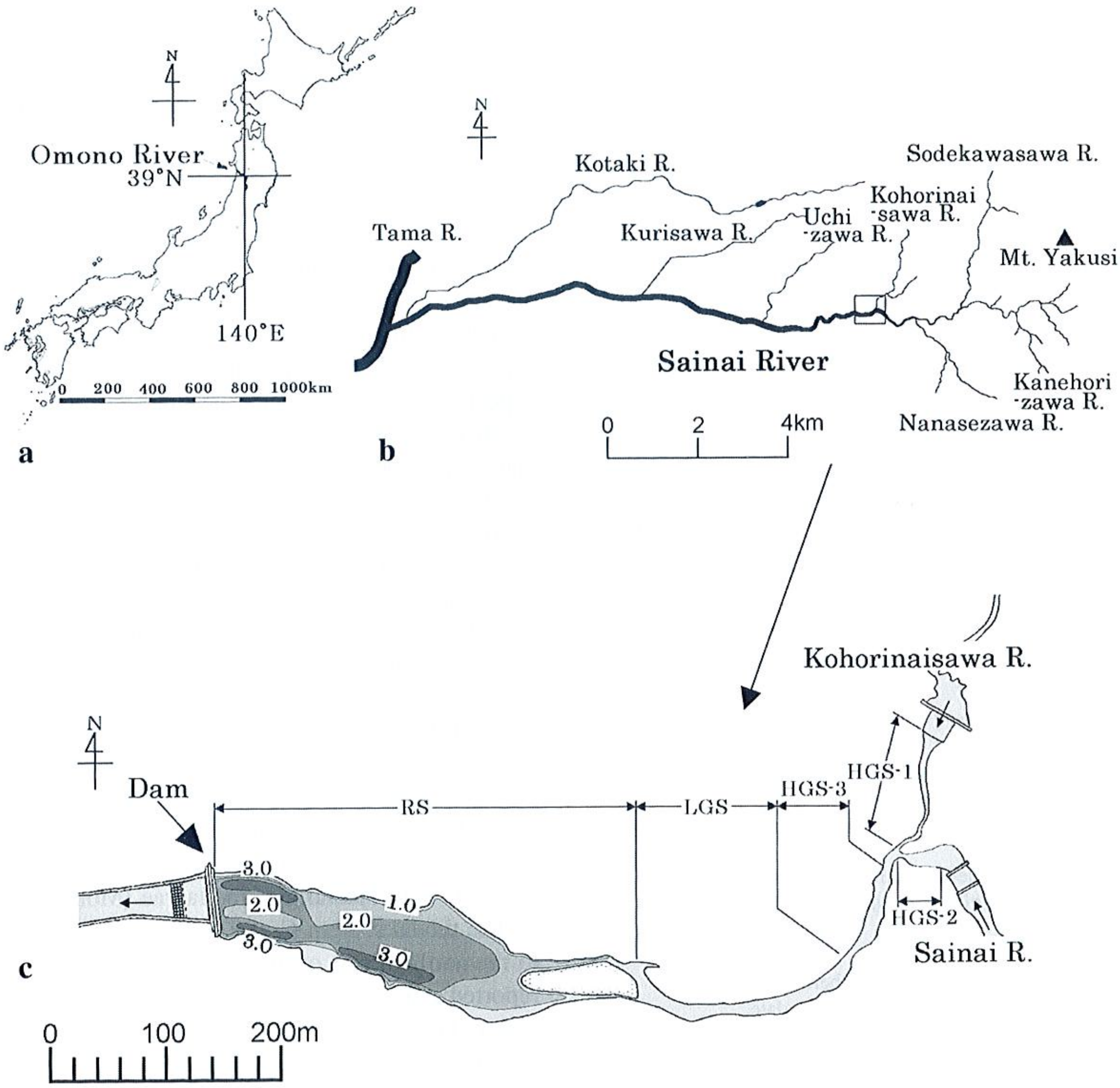


**Fig. 1** Location of **a** the Omono River basin, **b** maps of the observation sites in the Sainai River and **c** a stream–reservoir system in the Sainai River. *Numbers* in Fig. 1c indicate the water depth (m). *RS* reservoir, *LGS* low-gradient stream section, *HGS-1, 2, 3* high-gradient stream sections in which only HGS-3 was surveyed for nest spawning types of *Cottus nozawae*

**Table 1** Geopysical characteristics of sections surveyed in the stream–reservoir of the Sainai River in 2006

| Geophysical characteristics/section surveyed | HGS-3 | LGS | RS |
|---|---|---|---|
| Area in sections surveyed ($m^2$) | 1,200 | 2,200 | 200,000 |
| Length in sections surveyed (m) | 120 | 230 | 350 |
| Width in sections surveyed (m) | 5–15 | 13–15 | 20–30 |
| Depth in sections surveyed (m) | 0.1–0.5 | 0.1–1.5 | 3–6 |
| Current velocity in sections surveyed (m/sec) | 0.5–1.4 | 0.3–0.8 | 0.05–0.8 |

of relatively high-gradient (6–10 %) upstream sections (hereafter, referred to as HGS), a relatively low-gradient (2–5 %) downstream section (LGS) and a small reservoir section (RS) (Fig. 1c).

The HGS-3 is composed of fast riffles and characterized by the substratum that consists of gravel, cobble and boulder (Fig. 1c; Table 1). The HGS-1 and 2, which locate in the most upper part of the reservoir–stream system, were not used as our survey sections because they were found to support only fluvial *Cottus pollux* by our preliminary surveys. The LGS consists of riffle–pool reaches with slow riffles just upstream from the reservoir, and is characterized by the substratum that is composed of sand, gravel, cobble, boulder and fragments of bedrock fallen from the banks (Fig. 1c; Table 1). On the left side of the stream at the stream–reservoir boundary is a large spring-fed pool adjacent to an eroded bank. The RS consists of almost still water area, and the substrate is mainly composed of sandy silt, although gravel and debris of bedrock are scattered on the bank side. The banks of the reservoir consist of rocks and sandy silt (Fig. 1; Table 1).

*Nest site and spawning style.* Field surveys for finding the spawning nests of *C. nozawae* were conducted for 3–5 days starting from 15 April to 10 June 2003, 19 April to 20 June 2004 and 28 April to 29 June 2006. In both 2003 and 2004, the surveys were restricted to the HGS and LGS, whereas in 2006 the surveys were conducted in the whole reservoir–stream system including HGS, RGS and RS. During these surveys, the nest sites and nesting behaviors of parental males were examined by snorkeling in the shallow ($\leq$5 m) stream area and by scuba diving in deep (>5 m) water areas. Almost all nests and parental males were photographed. In narrow spaces, such as between rocks, a hand mirror was used to observe the nests and parental males.

The nests of *C. nozawae* were defined as sites in which both deposited egg clusters and parental males were observed. For successive observations of the developing eggs and behavior of parental male in the nest, the nest site was marked with a float that was tied to a fishing line and a weight at its end. Parental males in nests were collected using a small hand net, and their standard lengths were measured to the nearest 0.1 mm. After the measurements, the males were carefully released at the entrance of their nests.

Nest characteristics, including diameter of nest entrance in long and short axes, and depth of the nest hole were measured to the nearest 1 mm by using a divider or steel measure, immediately before the parental male swam away from the nest and just after removing the larvae from the nest.

For statistical analysis, the Mann-Whitney *U* test was used for pairwise comparison of the nest sizes among nest types and that of the nesting male size among nest types. Significant *P* values (0.05) were corrected by using the Bonferroni method. The Fisher's exact tests were used to examine differences in the frequency of occurrence of spawning-nest types among the research sections.

## Results and discussion

**Polymorphic spawning-nest types.** Spawning nests of *Cottus nozawae* were found at 100 sites during the three years of survey. Fifteen and nineteen nests were found in 2003 and 2004, in which observation was conducted only in the stream sections (HGS-3 and LGS). In 2006, a total of 66 nests were observed from the stream and reservoir sections (HGS-3, LGS and RS).

Based on the male nesting behaviors and nest characteristics such as site and materials, the spawning nests were classified into three types: HNS, CNS and RNS. The HNS type was characterized by the nesting male digging one or more horizontal holes in the wall of banks of the stream and reservoir, or vertical holes on the sandy silt bottom of the reservoir (Fig. 2a). The nesting male's behaviors for digging nest holes comprised following three behavioral elements: digging a hole on the wall composed of sandy silt using the mouth, carrying the sandy silt to the outside of a nest hole by the mouth, and extending the nest hole by fanning of the caudal fin. The CNS type was characterized by the nesting male using a crevice between large boulders or crevice of crag located on the bank of the stream and reservoir using a gap or hole in the woody debris for the nest sites (Fig. 2b). The RNS type was characterized by the nesting male using a rock or a few rocks on the streambed as its nest site (Fig. 2c).

This is the first record of HNS and CNS types in *C. nozawae*; only the RNS type has been previously observed in this species (Goto 1975, 1982). Morris (1954) extensively examined the reproductive behavior using nest stones and fanning activity of the bullhead *C. gobio* under experimental conditions in an aquarium and showed that its nest corresponds to the RNS type. Since Morris (1954), the RNS type has been reported in many *Cottus* species

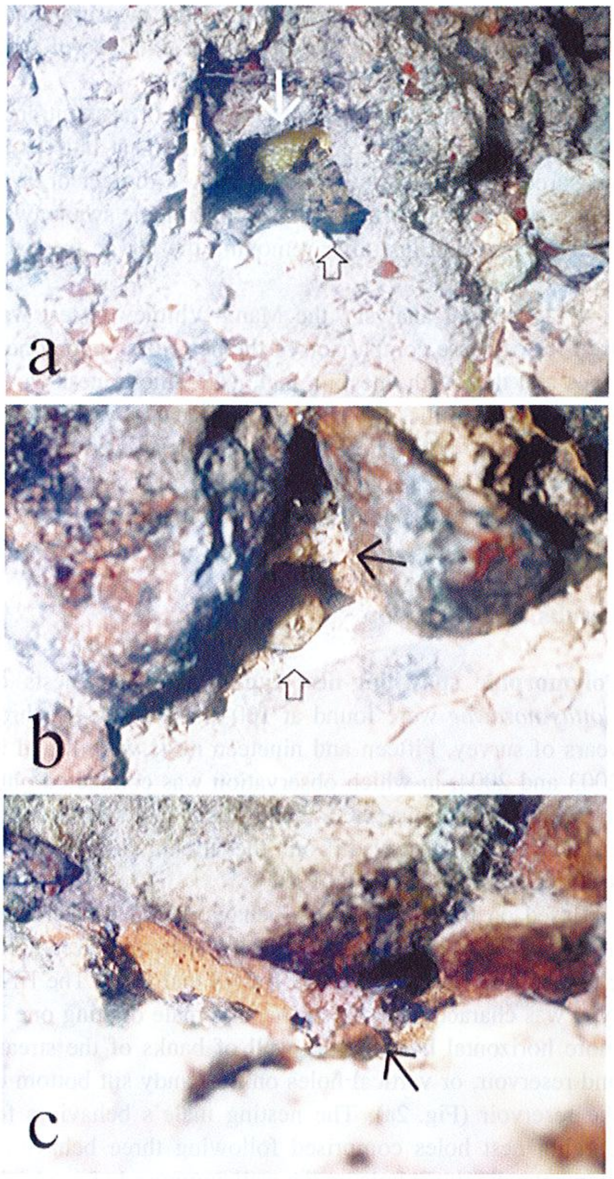

**Fig. 2** Photographs of three spawning-nest types taken in the reservoir–stream system of the Sainai River. **a** A typical HNS type of which a parental male [80 mm in standard length (SL)] guards the eggs at the entrance of a hole located immediately above the stream bed, **b** a typical CNS type of which a parental male (67 mm SL) guards the eggs at a crevice between large rocks located on the reservoir bed and **c** a typical RNS type of which a parental male (93 mm SL) guards the eggs beneath another rock in the rapid riffle area. *Black* and *white arrowheads* indicate the nest, *white arrow* shows a parental male, and *black arrow* indicates the deposited eggs

distributed in different geographic regions (Savage 1963; Goto 1987; Takeshita et al. 1999; Bateman and Li 2001), and the CNS type has been reported only in *Cottus asper* and *Cottus cognatus* from North America as far as we know (Krejsa 1967; Scott and Crossman 1973). Therefore, this is the first report of a member of the genus *Cottus* using the HNS type of digging an entire hole used as a spawning nest, although similar male digging behavior was observed in several benthic species of the cottoids in Lake Baikal (Sideleva 2003).

**Size of spawning nests and nesting males.** With regard to the mean long and short diameter of the entrance and the mean depth of the nests (Table 2), no differences were found among the three nest types at 5 % significance level (Mann-Whitney *U* tests after Bonferroni correction, $P > 0.0167$). A positive correlation was found between the mean entrance size and depth of nests only in the RNS type ($r^2 = 0.295$, $P < 0.01$), although no correlation was detected for the HNS and CNS types.

No significant differences were found in the mean standard length of nesting males for all the pair comparisons of the three spawning-nest types (Mann-Whitney *U* tests after Bonferroni correction, $P > 0.36$ for all pairwise comparison; Table 2). A significant correlation was found between the depth of nests and the standard length of nesting males in CNS type ($r^2 = 0.826$, $P = 0.032$, $n = 5$). Furthermore, significant correlations were found both between nesting male size and size of nest entrance and between male size and depth of nests in the HNS type ($r^2 = 0.356$, $P = 0.015$, and $r^2 = 0.252$, $P = 0.048$).

These results indicate that there are marked differences in the materials of nests among the three spawning-nest types, but no difference in nest size among them. In addition, larger nesting males likely use larger nests in the CNS and HNS types, although the result for the CNS type may not be reliable for the small sample size. Because the males dig the nest hole by themselves in the HNS type, it seems possible that the nest size was adjusted to the males' size. By contract, males in the CNS and RNS types seem to utilize naturally existing holes as their nests.

**Comparison of frequency of occurrence of spawning-nest types among the sections.** The frequency of occurrence of spawning-nest types among three sections (HGC-3, LGC and RS) in 2003, 2004 and 2006 is shown in Fig. 3. No significant difference in the nest type frequency in LGS (only the section surveyed in all the three years) was found among the three years (Fisher's exact test, $P = 0.50$). The frequency of occurrence of spawning-nest types (HNS, CNS and RNS) in 2006 or all the three years was significantly different between LGS and RNS (extended Fisher's exact test, both $P < 0.01$). No conclusion was obtained for HGS-3 because of the small number of nests observed ($n = 2$ in total). These results suggest that RNS type is dominant in the rapid riffles (found only in LGS and HGS-3) where males can utilize rocks as the nest materials, whereas HNS type is dominant in the reservoir where males are able to utilize the walls and bottom, which are constructed by sandy silt, as the nest materials, and CNS type is dominant in the slow riffles where males can utilize both rocks and sandy silt as the nest materials.

**Table 2** Diameter of nest entrance, depth of nests and standard length of parental males of three nest types of *Cottus nozawae* in 2006

| Item measured | Hole nest type | Crevice nest type | Rock nest type |
|---|---|---|---|
| Long diameter of nest entrance (mm) | 109 ± 41 ($n$ = 54) | 94 ± 36 ($n$ = 11) | 102 ± 31 ($n$ = 35) |
| Short diameter of nest entrance (mm) | 53 ± 16 ($n$ = 54) | 54 ± 20 ($n$ = 11) | 49 ± 17 ($n$ = 35) |
| Depth of nests (mm) | 76 ± 40 ($n$ = 54) | 82 ± 41 ($n$ = 11) | 84 ± 26 ($n$ = 35) |
| Standard length of parental males (mm) | 84.3 ± 13.0 ($n$ = 16) | 78.2 ± 12.1 ($n$ = 5) | 81.6 ± 11.0 ($n$ = 10) |

Data are shown as mean ± SD

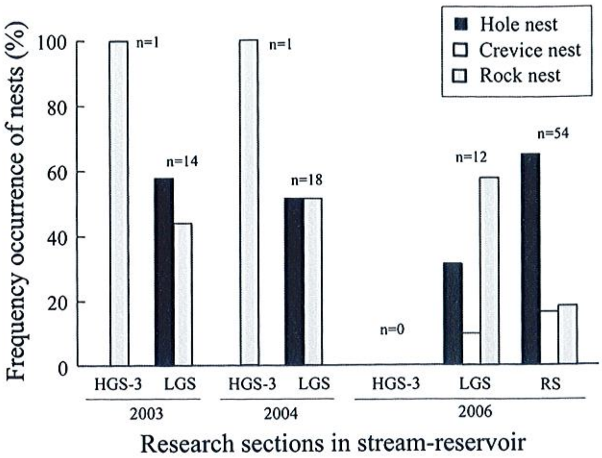


**Fig. 3** Comparison of frequency of occurrence of three spawning-nest types among three sections (HGS-3, LGS, and RS) in 2003, 2004 and 2006

Despite much intensive research effort, we could not find the HNS and CNS nest types in 8 other tributaries of the Omono River basin (Kumagai, Sugiyama and Goto, unpubl. data), and more than 25 rivers in the Hokkaido Island of which two rivers, the Hekiriji and the Saru, include dam reservoirs; the RNS type was the only observed spawning nests in the riffles of the rivers with gravel bottom (Goto 1975, 1982). These results suggest that male nesting behavior in both the HNS and CNS types might have been modified from that of the original RNS type, in relation to the artificial environmental changes accompanying the alteration from the stream to the stream–reservoir systems after the dam construction in 1993. Further studies on the biological advantage (e.g., fitness) of the nesting males using the HNS, RNS and CNS types are needed.

**Acknowledgments** We wish to express our gratitude to R. Yokoyama, T. Kusakari, Y. Sasaki, S. Kimura and M. Sato for their devoted help in the field survey and sample collections.

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