Reidentification of leuciscid fish (Cypriniformes) reported as a host of *Argulus coregoni* (Crustacea: Branchiura: Argulidae) from a stream, central Japan, with a new host record for the argulid

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Abstract

Both fat minnow *Rhynchocypris lagowskii steindachneri* (Sauvage, 1883) (Leuciscidae) and dark chub *Nipponocypris temminckii* (Temminck and Schlegel, 1846) (Xenocyprididae) were recently reported as hosts of *Argulus coregoni* Thorell, 1864 from the Kushiko River, a tributary of the Hasu River, in Fukui Prefecture, central Japan. However, more recently, because the identification of the former fish species was found doubtful, we reexamined three individuals of leuciscid reported as "fat minnow" and identified them as a morphologically similar congeneric species, upstream fat minnow *Rhynchocypris oxycephala jouyi* (Jordan and Snyder, 1901) (Leuciscidae). Furthermore, to confirm the occurrence of upstream fat minnow in the Kushiko River, we collected fishes in this river in October 2024 and identified specimens of leuciscid as the fish species. As a result of these works, we have concluded that the leuciscid recently reported from the Kushiko River was upstream fat minnow and the record of "fat minnow" was incorrect. Alternatively, upstream fat minnow becomes a correct host name and represents a new host record for *A. coregoni* because this parasite has never been found from the fish species occurring in the Far East. A specimen of *A. coregoni* was collected from a dark chub during the sampling in October 2024 and is reported as its third record from this xenocypridid fish.

Introduction

In parasitology, host identification is important to understand various aspects of the biology of parasites, such as their ecology, host-parasite relationships, and coevolution with hosts. Nonetheless, since parasitologists are usually not experts of the taxonomy of hosts, they sometimes encounter difficulties in host identification, especially when they examine morphological similar, closely related host species.

In our recent paper, *Argulus coregoni* Thorell, 1864 was reported to infect two species of cypriniform fishes in the Kushiko River, a tributary of the Hasu River, Fukui Prefecture, central Japan, and they are fat minnow *Rhynchocypris lagowskii steindachneri* (Sauvage, 1883) (Leuciscidae) and dark chub *Nipponocypris temminckii* (Temminck and Schlegel, 1846) (Xenocyprididae) (Nagasawa and Nishimura, 2025). However, after this paper was accepted for publication in mid-October 2024, we doubted our own identification of leuciscid reported as "fat minnow" because a congeneric species, upstream fat minnow *Rhynchocypris oxycephala jouyi* (Jordan and Snyder, 1901) (Leuciscidae), has been reported to also occur in the Hasu River and its tributaries (Kato, 1998) and this

Nagasawa, K., H. Nakano and H. Nishimura. 2025. Reidentification of leuciscid fish (Cypriniformes) reported as a host of *Argulus coregoni* (Crustacea: Branchiura: Argulidae) from a stream, central Japan, with a new host record for the argulid. *Nature of Kagoshima* 51: 257–261.

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Received: 4 March 2025; published online: 8 March 2025; https://journal.kagoshima-nature.org/archives/NK_051/051-055.pdf

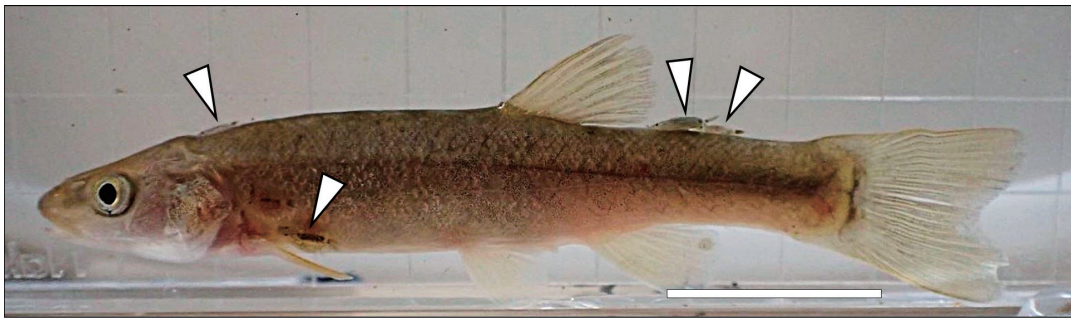

Fig. 1. Live individual of leuciscid (76 mm SL) infected with *Argulus coregoni* from the Kushiko River, a tributary of the Hasu River, Fukui Prefecture, central Japan. This leuciscid was reported as "fat minnow *Rhynchocypris lagowskii steindachneri*" by Nagasawa and Nishimura (2025), but the ethanol-preserved specimens of this and two other individuals of leuciscid collected on 22 August and 15 September 2022, respectively, are identified in the present study as upstream fat minnow *R. oxycephala jouyi* based on their number of transverse scales above the lateral line. Arrowheads indicate individuals of *A. coregoni* attaching on the host fish. Scale bar: 20 mm.

species is morphologically similar to fat minnow (Nakamura, 1969; Hirai and Hikido, 1976; Itai, 1977, 1978; Fujita and Hosoya, 2003; Hosoya, 2013; Hata et al., 2018). When we wrote and revised our paper, we were not aware of Kato's (1998) paper and, based on the gross morphology of the leuciscid collected, reported it as "fat minnow" without discussing the possibility of the occurrence of upstream fat minnow in the Kushiko River. In the present study, to resolve uncertainty in such fish identification, we reexamined three individuals of leuciscid reported as "fat minnow" and identified them as upstream fat minnow. Moreover, we collected fishes again at the site reported by Nagasawa and Nishimura (2025) in the Kushiko River in late October 2024 and identified specimens of leuciscid as upstream fat minnow. Based on these facts, we have concluded that the recently reported leuciscid was not "fat minnow" but upstream fat minnow and the latter species serves as a host for *A. coregoni* along with dark chub in the Kushiko River. Moreover, upstream fat minnow here represents a new host record for *A. coregoni* because there is no previous record of this parasite from the fish species occurring in the Far East. In this paper, we also report on the occurrence of *A. coregoni* on fishes collected in the Kushiko River in late October 2024.

Materials and Methods

Identification of leuciscid in the genus *Rhynchocypris*. For identification of leuciscid from the Kushiko River, two samples were used: one sample is three ethanol-preserved specimens [62–76 mm standard length (SL)] reported as "fat minnow" in our recent study (Nagasawa and Nishimura, 2015), and another sample is seven similarly preserved specimens (48–71 mm SL) that were newly collected on 26 October 2024 at the site reported by the above authors (see the paragraph below). The former sample was found in the fish collection of the third author (HN) of this paper, and one and two fish were collected on 22 August and 15 September 2022, respectively. The fish sampled on the former date was found infected with four individuals of *A. coregoni* (Fig. 1).

All specimens (n = 10) of both samples were examined at the Aquaparasitology Laboratory in Shizuoka Prefecture, where the number of transverse scales above the lateral line (NTS) was recorded for each of individuals larger than 45 mm SL. The NTS has been reported as the most reliable morphological character to differentiate upstream fat minnow from fat minnow collected in central Honshu, Japan (Itai, 1977, 1978; Fujita and Hosoya, 2003; Hata et al., 2018) and it is possible to precisely count the NTS in fish that are larger than 45 mm SL (Hata et al., 2018).

Fish collection and parasite examination. To confirm the occurrence of fat minnow and/or upstream fat minnow in the Kushiko River, fishes were collected with a scoop net in the lower reaches of the river (35º31'48"N, 135º54'30"E) at Fujii in Wakasa, Fukui Prefecture on 26 October 2024. The collection site (Fig. 2) was the same as that reported by Nagasawa and Nishimura (2025). Within the day of collection, fishes were transported alive to the laboratory of the Kyoto Marine High School in Miyazu, Kyoto Prefecture, where they were individually identified, measured to the SL (to the nearest 1 mm), examined for ectoparasites, and fixed in 70% ethanol. An argulid was collected from the body surface of a dark chub

Fig. 2. Collection site in the lower reaches of the Kushiko River, a tributary of the Hasu River, Fukui Prefecture, central Japan. The photograph was taken on the day of collection (26 October 2024).

and fixed in 70% ethanol. As stated above, leuciscid specimens were later examined at the Aquaparasitology Laboratory. Detailed information on the collection site and its environmental conditions is found in Nagasawa and Nishimura (2025).

Identification of specimen of argulid and its occurrence on fishes from the Kushiko River. The argulid specimen was examined at the Aquaparasitology Laboratory and identified with an Olympus SZX10 stereo microscope and Olympus BX51 phase-contrast compound microscope. The specimen was cleared in lactophenol and observed using the wooden slide procedure (Humes and Gooding, 1964; Benz and Otting, 1996). The coxae of the first and second legs were illustrated with the aid of a drawing tube attached to the compound microscope. After the specimen was identified, its sex and total length (TL, to the nearest 0.1 mm, from the anterior tip of the carapace to the posterior tip of the abdomen) were recorded.

Specimen deposition in museums. The fishes collected on 26 October 2024 in the Kushiko River consisted of four species: dark chub, upstream fat minnow, pond loach *Misgurnus anguillicaudatus* (Cantor, 1842) (Cypriniformes: Cobitidae), and yoshinobori goby *Rhinogobius* sp. (Gobiiformes: Gobiidae). The specimens of these fishes and three specimens of upstream fat minnow collected in August and September 2022 have been deposited at the Lake Biwa Museum (LBM) in Otsu, Shiga Prefecture [catalogue number 1210061298, dark chub (n = 36); 1210061299, upstream fat minnow (n =12); 1210061300, pond loach (n =2), all collected on 26 October 2024; 1210061303–1210061305, upstream fat minnow (n = 3) collected on 22 August and 15 September 2022] and the Wakayama Prefectural Museum of Natural History (WMNH) in Kainan, Wakayama Prefecture [WMNH-PIS 14184–14186, yoshinobori goby (n = 3) collected on 26 October 2024]. The scientific names of fishes mentioned in this paper follow Motomura (2025), and the order and family, to which each fish species belongs, is based on Froese and Pauly (2024).

A voucher specimen of *A. coregoni* collected on 26 October 2024 has been deposited in the Crustacea collection of the National Museum of Nature and Science, Tsukuba, Ibaraki Prefecture (NSMT-Cr 32870).

Results and Discussion

Identification of leuciscid from the Kushiko River. The NTS in three and seven specimens of leuciscid collected in the recent (Nagasawa and Nishimura, 2025) and present studies ranges from 14–15 and 13–17, respectively.

The NTS is known to differ between upstream fat minnow and fat minnow collected in central Honshu, Japan. Itai (1977) reported that upstream fat minnow has 14–19 scales, whereas fat minnow possesses 20–25 scales. Similar data have been shown by Fujita and Hosoya (2003), Hosoya (2013), and Hata et al. (2018): for example, 12–19 scales in upstream fat minnow but 20–24 scales in fat minnow (Hata et al., 2018). Fujita (2024) mentions that the former species has 18 or less scales but the latter species does 19 or more scales. The overall NTS (13–17, n = 10) counted here corresponds to that reported from upstream fat minnow, which demonstrates that the leuciscid examined in our recent and present studies is not “fat minnow” but can be identified as upstream fat minnow. In other words, the later fish species serves as a host for *A. coregoni* along with dark chub in the Kushiko River.

According to Fujita (2015, 2024), upstream fat minnow has a wide geographical distribution in the Far East with its records from central and western

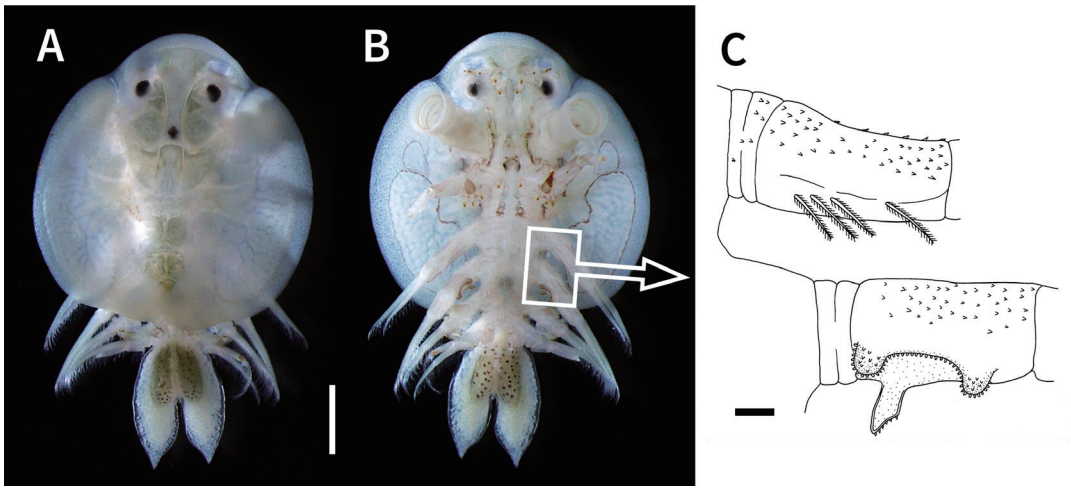

Fig. 3. *Argulus coregoni*, adult male (7.2 mm TL), NSMT-Cr 32870, collected from the body surface of a dark chub *Nipponocypris temminckii* on 26 October 2024 in the Kushiko River, a tributary of the Hasu River, in Fukui Prefecture, central Japan. A, habitus, dorsal view; B, habitus, ventral view; C, coxae of first and second legs, ventral view. The specimen was fixed in 70% ethanol on the day of collection and photographed on 3 February 2025. Scale bars: A, B, 1 mm; C, 0.1 mm.

Japan, the Korean Peninsula, the Amur River system (Russia), and the Min River system (Fujian Province, China). Since there is no record of *A. coregoni* from upstream fat minnow in these regions (Nagasawa et al., 2024, tables 1–2, 4), this fish species is regarded here as a new host of the parasite.

Identification of argulid and its occurrence on fishes from the Kushiko River. The argulid specimen collected on 26 October 2024 is an adult male (7.2 mm TL, Fig. 3A, B) and is characterized by sharply pointed abdominal lobes; four plumose setae near the ventroposterior margin of each coxa of the first legs; and two protrusions adorned with small spines and a digitiform projection on the ventro- and dorsoposterior margins of each coxa of the second legs (Fig. 3C). Moreover, 60 and 61 supporting rods are each present in marginal membranes of the first maxillae. This number of supporting rods per first maxilla is fewer than that (65–70) reported recently from the adult male of *A. coregoni* from the Kushiko River (Nagasawa and Nishimura, 2025), but the above morphological characters are identical to those of *A. coregoni* from the river and other localities in Japan (e.g., Tokioka, 1936; Hoshina, 1950; Nagasawa et al., 2024; Nagasawa and Nishimura, 2025). Thus, the argulid specimen collected in this study is identified as *A. coregoni*.

The fish sample collected on 26 October 2024 consisted of four species, i.e., dark chub, upstream fat minnow, pond loach, and yoshinobori goby. A single individual (82 mm SL) of 36 dark chub [51–121 (mean, 70) mm SL, n = 36] was found infected with *A. coregoni* on its body surface. There was no infection of the parasite on three other fish species, including 12, 2, and 3 individuals of upstream fat minnow [30–71 (54) mm SL], pond loach [87–88 (88) mm SL], and yoshinobori goby [41–46 (43) mm]. Prevalence of infection on dark chub was as low as 2.8% and the intensity of infection was one. The present collection of *A. coregoni* represents its third record from dark chub in Japan and worldwide: the previous records from this fish species were from the Takase River, Wakayama Prefecture (Nagasawa et al., 2024) and the Kushiko River (Nagasawa and Nishimura, 2025), both of which lie in central Japan.

Future study. In this study, upstream fat minnow was the only leuciscid collected at the study site in the Kushiko River, a tributary of the Hasu River. However, according to Kato (1998), 23 fish species including upstream fat minnow and fat minnow are found in the Hasu River and its tributaries. He also has suggested that these two species have different habitats in the rivers of Fukui Prefecture: upstream fat minnow occurs in the upper reaches, whereas fat minnow is found from the mid-reaches to a lower region of the upper reaches. Thus, as has been similarly suggested before by Nagasawa and Nishimura (2025), it is important to examine the occurrence of *A. coregoni* on these closely related leuciscid species in the main stream and various tributaries of the Hasu River for clarifying host and environmental factors contributing to the distribution of the parasite in the river.

Acknowledgments

We thank Dr. Seigo Kawase (LBM, Otsu) and Dr. Kentaro Hirashima (WMNH, Kainan) for their support with museum deposition of fish specimens. Dr. K. Hirashima also identified our specimens of yoshinobori goby and measured them.

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