

# Occurrence of a Skin Parasite *Argulus coregoni* (Branchiura: Argulidae) on Salmonids in Mountain Streams, Central Japan, with Discussion on Its Longitudinal Distribution and Host Utilization in Rivers

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*Argulus coregoni* Thorell, 1864 was collected from white-spotted char, *Salvelinus leucomaenis* (Pallas, 1814), red-spotted masu salmon, *Oncorhynchus masou ishikawae* Jordan and McGregor, 1925, masu salmon, *O. m. masou* (Brevoort, 1856), and a hybrid between white-spotted char and masu salmon in mountain streams, Gifu Prefecture, central Japan. The host's body surface under and near the pectoral fins was the most common infection site for *A. coregoni*. The infected white-spotted char were caught at high elevations (461–873 m) in the headwater streams. The infected individuals of the two masu salmon subspecies were mostly caught in the upper river reaches, but the elevations where they were caught were lower (237–733 m and 660–707 m, respectively) than those of the white-spotted char. Since *A. coregoni* is also known as a parasite of ayu, *Plecoglossus altivelis altivelis* (Temminck and Schlegel, 1846), in the middle and lower river reaches in the prefecture, this parasite utilizes different fish species as its hosts along a river course: white-spotted char and the two masu salmon subspecies serve as the hosts, respectively, in the headwater and the middle to lower sections of the upper reaches, but in the mid- and lower river reaches, ayu is the important host.

**Key Words:** freshwater fish parasite, white-spotted char, red-spotted masu salmon, masu salmon, ayu.

## Introduction

The argulid branchiuran *Argulus coregoni* Thorell, 1864 is a skin parasite of freshwater fishes in northern Europe and eastern Asia (Neethling and Avenant-Oldewage 2016). In Japan, the species infects wild fishes belonging to five families; Salmonidae, Plecoglossidae (both Salmoniformes), Cyprinidae (Cypriniformes), Amblycipitidae (Siluriformes), and Odontobutidae (Gobiiformes) (e.g., Tokioka 1936; Yamaguti 1937; Nagasawa and Kawai 2008; Nagasawa 2009, 2011; Nagasawa et al. 2014; Nagasawa and Ishikawa 2015; Nagasawa and Taniguchi 2021) but is also found on farmed salmonid and plecoglossid fishes (Hoshina 1950; Nagasawa and Yuasa 2020). Its larval development, host associations, impacts on host fishes, and treatment have all been studied (Inoue et al. 1980; Shimura and Egusa 1980; Shimura 1981, 1983; Shimura et al. 1983a, b; Shimura and Inoue 1984; Katahira et al. 2021).

*Argulus coregoni* is known as a parasite of ayu, *Plecoglossus altivelis altivelis* (Temminck and Schlegel, 1846), and white-spotted char, *Salvelinus leucomaenis* (Pallas, 1814) in rivers and of salmonids reared at a fisheries research insti-

tute in Gifu Prefecture (Nagasawa et al. 2018, 2020a, b, 2021; Nagasawa and Morikawa 2019a). This prefecture lies in a mountainous inland region of central Japan and is home to many freshwater fish populations in numerous rivers and streams (Mukai 2017). During an ecological study of freshwater fishes in the prefecture, *A. coregoni* was collected from the body surface of salmonids in mountain streams (Fig. 1). Due to the low population size of the salmonids and difficulty in sampling them, limited information is available on the biology of *A. coregoni* infecting stream-dwelling salmonids, especially those occurring at high elevations. This paper reports on the distribution and habitats of salmonids infected with *A. coregoni* and its occurrence on those salmonids from mountain streams in Gifu Prefecture. The paper also discusses the longitudinal distribution and host utilization of *A. coregoni* in rivers, because this parasite infects ayu in the middle and lower reaches of rivers in the prefecture as well (see above literature).

## Materials and Methods

The specimens of *A. coregoni* were carefully removed by

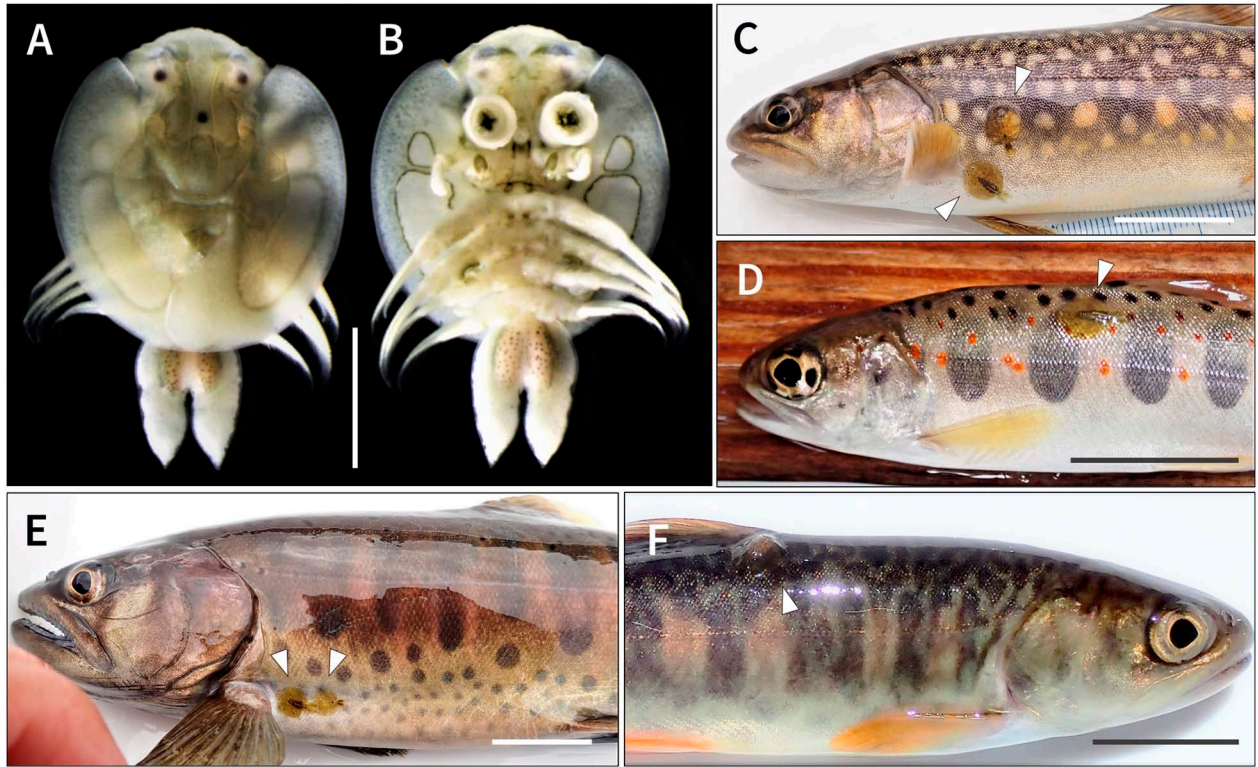


Fig. 1. *Argulus coregoni*, male, NSMT-Cr 30777, from a white-spotted char, *Salvelinus leucomaenis*, from the Gamada River, Gifu Prefecture, ethanol-preserved specimen, A, Dorsal view; B, ventral view; C, two females infecting a white-spotted char (180 mm FL) near the left pectoral fin (from the Maze River); D, one female infecting a red-spotted masu salmon, *Oncorhynchus masou ishikawae* (103 mm FL), near the base of the dorsal fin (from a tributary of the Hida River); E, one female (left) and one male (right) infecting a masu salmon, *O. m. masou* (257 mm FL), near the left pectoral fin (from the Itoshiro River); F, one female infecting a hybrid between white-spotted char and masu salmon (165 mm FL) near the dorsal fin (from the Itoshiro River). Arrowheads indicate individuals of *A. coregoni*. See Fig. 2 for the locations of the rivers. Scale bars: A, B, 2 mm; C–F, 20 mm.

hand from the body surface of live salmonids caught by electrofishing from July to October 2019 in seven rivers belonging to the five larger river systems in Gifu Prefecture (Fig. 2; Table 1). No data on prevalence of infection were taken. Soon after capture, infected fish were individually identified to species, anesthetized with FA100 (containing mainly eugenol), measured for fork length (FL) to the nearest mm, and then photographed for recording the attachment site of each *A. coregoni* specimen. One fish with an irregular body color pattern (Fig. 1F) was identified as a hybrid between a white-spotted char and a masu salmon, *Oncorhynchus masou masou* (Brevoort, 1856) (see Kato 1977; Kishi and Tokuhara 2012; Mukai et al. 2015). The specimens of *A. coregoni* were fixed and preserved in 99% ethanol. Their identification was later confirmed at the Aquaparasitology Laboratory, Shizuoka, based on Tokioka (1936), Yamaguti (1937), and Nagasawa and Taniguchi (2021). The specimens of *A. coregoni* were examined for their sex and total length (TL) measured to the nearest 0.1 mm. Voucher specimens of *A. coregoni* have been deposited in the Crustacea (Cr) collection of the National Science Museum of Nature and Science, Tsukuba, Ibaraki Prefecture [NSMT-Cr 30777 (Fig. 1A, B), one male from white-spotted char from the Gamada River; NSMT-Cr 30778, two males and one female from red-spotted masu salmon from the Maze River; NSMT-Cr 30779, one male and one female from masu salm-

on from the Itoshiro River]. Data on the elevation (m) at each collection site were obtained from a GSI map (<https://maps.gsi.go.jp/>) provided by the Geospatial Information Authority of Japan.

Previously, two subspecies of white-spotted char [*Salvelinus leucomaenis pluvius* (Hilgendorf, 1876) and *S. l. japonicus* Oshima, 1961] were identified from Gifu Prefecture (e.g., Kawanabe 1989; Hosoya 2015). However, a recent molecular work has not supported the recognition of these subspecies in white-spotted char (Yamamoto et al. 2004), thus, *S. leucomaenis* is used in this paper. The common name of *S. leucomaenis* follows Dunham et al. (2008), and those of two subspecies of *O. masou* (*O. m. masou* and *O. m. ishikawae* Jordan and McGregor, 1925) are based on Kishi et al. (2016).

## Results

### Distribution and habitats of infected salmonids.

*Argulus coregoni* was found infecting 20 individual salmonids, which were identified as white-spotted char (*S. leucomaenis*) (n=11), red-spotted masu salmon (*O. m. ishikawae*) (n=5), masu salmon (*O. m. masou*) (n=3), and a hybrid between white-spotted char and masu salmon (n=1) (Fig. 1; Table 1). The infected white-spotted char were caught at seven sites in six rivers at high elevations (461–

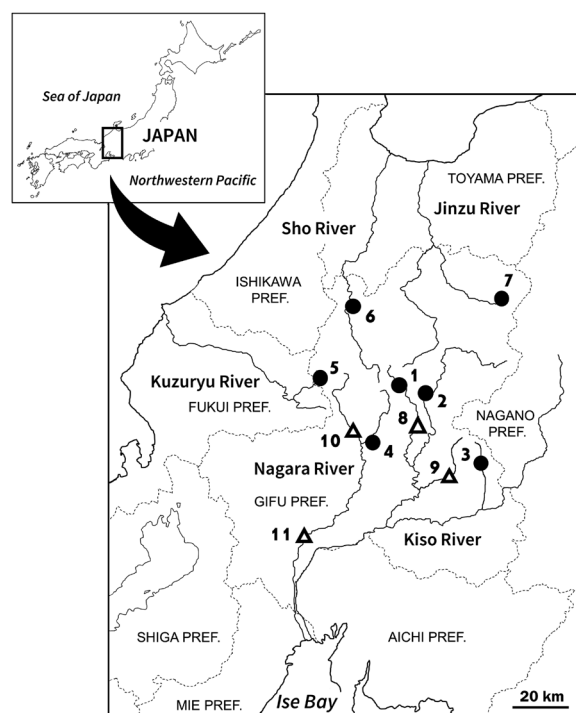


Fig. 2. Map showing the collection localities of salmonids infected with *Argulus coregoni* (closed circles 1–7) in rivers of Gifu Prefecture, central Japan. The collection localities of ayu, *Plecoglossus altivelis altivelis*, infected with *A. coregoni*, are also shown (open triangles 8–11). 1, Upper reaches of the Maze River; 2, tributary of the Hida River; 3, tributary of the Tsukechi River; 4, tributary of the Yoshida River; 5, the Itoshiro River; 6, tributary of the Sho River; 7, the Gamada River; 8, middle reaches of the Maze River; 9, middle reaches of the Shira River; 10, middle reaches of the Nagara River; 11, lower reaches of the Nagara River.

873 m) (Table 1). The six rivers belong to the five large river systems: three (Kuzuryu, Sho, and Jinzu) and two (Kiso and Nagara) of these river systems flow into the Sea of Japan and the Northwestern Pacific through Ise Bay, respectively (Fig. 2). The infected red-spotted masu salmon were caught at four sites in three rivers (Table 1), which are major tributaries to the Kiso and Nagara river systems (Fig. 2). The elevations at which the red-spotted masu salmon were caught (237–733 m) were lower than those of the white-spotted char catch. The infected masu salmon were caught at two sites in the Itoshiro River at 660–707 m elevation, and the infected hybrid was also caught at a site in the same river at 715 m elevation (Table 1). The Itoshiro River is an upstream tributary in the Kuzuryu River system (Fig. 2).

All infected salmonids were collected from mountain streams, including the main streams of the rivers (Fig. 3A, E, H) and small tributaries flowing into the main streams (Fig. 3B–D, F, G). These sites showed a wide range of conditions in space, width, water volume, and gradient, but the water was constantly clear, the water depth was usually less than 1 m, and the substratum consisted of a mixture of gravel and cobble, often with boulders.

**Occurrence on hosts, attachment sites, and body size.** In total, 32 specimens of *A. coregoni* were collected from 20 infected salmonids [103–257 (mean=174) mm FL]: one

specimen per host was most common (60%), followed by two (25%) and three (10%) specimens. Only a single fish (white-spotted char, 195 mm FL, from the Tsukechi River) harbored four specimens of *A. coregoni* (Table 1).

The lateral body surface under and near the pectoral fins of salmonids was the most common attachment site for *A. coregoni*, followed by the lateral body surface below the dorsal fin and the dorsal body surface of the anterior trunk (Figs 1, 4). Only a few specimens of *A. coregoni* were found on the posterior body surface of the host.

The 32 specimens of *A. coregoni* included 14 males (44%) and 18 females (56%). Excluding a single small male specimen (3.1 mm TL) collected in early September 2019, the other 13 males were stouter and bigger [5.3–9.6 (mean=7.5) mm TL]. The females were composed of 15 ovigerous specimens [7.5–11.0 (mean=8.7) mm TL] and three smaller, non-ovigerous specimens [6.0–7.0 (mean=6.5) mm TL]. These non-ovigerous females were collected from early to late September 2019, but the ovigerous females were collected during the whole survey period (from late July to mid-October 2019).

## Discussion

Following a recently published record of *A. coregoni* from a white-spotted char in the Maze River (Nagasawa et al. 2021), this paper represents the second record of the species from salmonids in the mountainous region of Gifu Prefecture and reveals that it also parasitizes red-spotted masu salmon, masu salmon, and the hybrid between white-spotted char and masu salmon. The infected whitespotted char were caught in the streams in all five river systems investigated. In contrast to this, as the two masu salmon subspecies have separated natural distribution ranges in central Japan (Kawanabe 1989; Kato 1991), the infected individuals of masu salmon and red-spotted masu salmon occurred in the river systems emptying into the Sea of Japan and the Northwestern Pacific, respectively (Table 1).

Among stream-dwelling salmonids, white-spotted char occurs in the uppermost reaches of rivers in central Japan (e.g., Kato 1992; Kitano and Kubota 1999; Kishi and Tokuhara 2012; Kishi et al. 2016). During this study, one infected white-spotted char (145 mm FL) was caught in a tributary of the Sho River at 873 m elevation, which represents the second highest elevation record of the natural distribution of *A. coregoni* in Japan. The highest elevation collection record (1,075 m) of the species was taken from a white-spotted char (reported as *S. l. japonicus*) in a mountain stream, Nagano Prefecture, east of Gifu Prefecture (Nagasawa and Kawai 2015). *Argulus coregoni* was also reported from brown trout, *Salmo trutta* Linnaeus, 1758, in a high elevation lake (1,269 m), central Japan, but it had not been naturally distributed in the lake but was introduced there (Nagasawa 2009; Nagasawa and Kawai 2015). Based on these collection records, *A. coregoni* has been regarded as one of the fish parasites found in high elevation mountain streams of Japan (Nagasawa and Kawai 2015).

Table 1. *Argulus coregoni* collected from salmonids in mountain streams of Gifu Prefecture, central Japan.

River system	River	Collection locality			Host		Fork length (mm)	<i>A. coregoni</i> collected (total length, mm)	Collection date
		Site	Elevation (m)	Location	Number in Fig. 2	Species			
Kiso River	Maze River	Main stream	733	Oppara, Takayama	1	<i>Salvelinus leucomaenis</i>	162	One female (9.5)	1 October 2019
							180	One male (8.3), one female (9.0)	2 October 2019
	Hida River	Inflowing tributary-1	519	Shimi, Gero	2	<i>Oncorhynchus masou ishikawae</i>	128	One male (8.0)	23 July 2019
							255	Two males (7.7, 7.0), one female (7.5)	1 October 2019
							159	Two females (8.2, 6.4)	24 September 2019
Nagara River	Yoshida River	Inflowing tributary	237	Asahi, Gujo	4	<i>Oncorhynchus masou ishikawae</i>	147	One male (7.0)	13 September 2019
							152	One female (8.0)	
							212	One female (8.0)	
							103	One female (7.9)	24 September 2019
							195	One male (9.2), three females (11.0, 9.8, 6.0)	10 September 2019
Kuzuryu River	Itoshiro River	Main stream-1	707	Itoshiro, Gujo	5	<i>Oncorhynchus masou masou</i>	200	One male (7.6)	13 September 2019
							117	One male (7.0), one female (9.0)	30 September 2019
	Inflowing tributary	715	Itoshiro, Gujo	5	5	<i>Salvelinus leucomaenis</i>	112	One female (7.0)	
							197	Two females (10.0, 9.0)	17 October 2019
							257	Two males (7.5, 6.9), one female (8.2)	
Sho River	Gamada River	Main stream	836	Kansaka, Takayama	7	<i>Salvelinus leucomaenis</i>	204	One male (8.8)	7 September 2019
							152	One male (9.6)	
Sho River	Inflowing tributary	873	Kurodani, Takayama	6	<i>Salvelinus leucomaenis</i>	165	One female (10.0)		
						145	Two males (5.3, 3.1)	4 September 2019	
Jinzu River	Gamada River	Main stream	836	Kansaka, Takayama	7	<i>Salvelinus leucomaenis</i>	240	One male (6.0)	8 August 2019

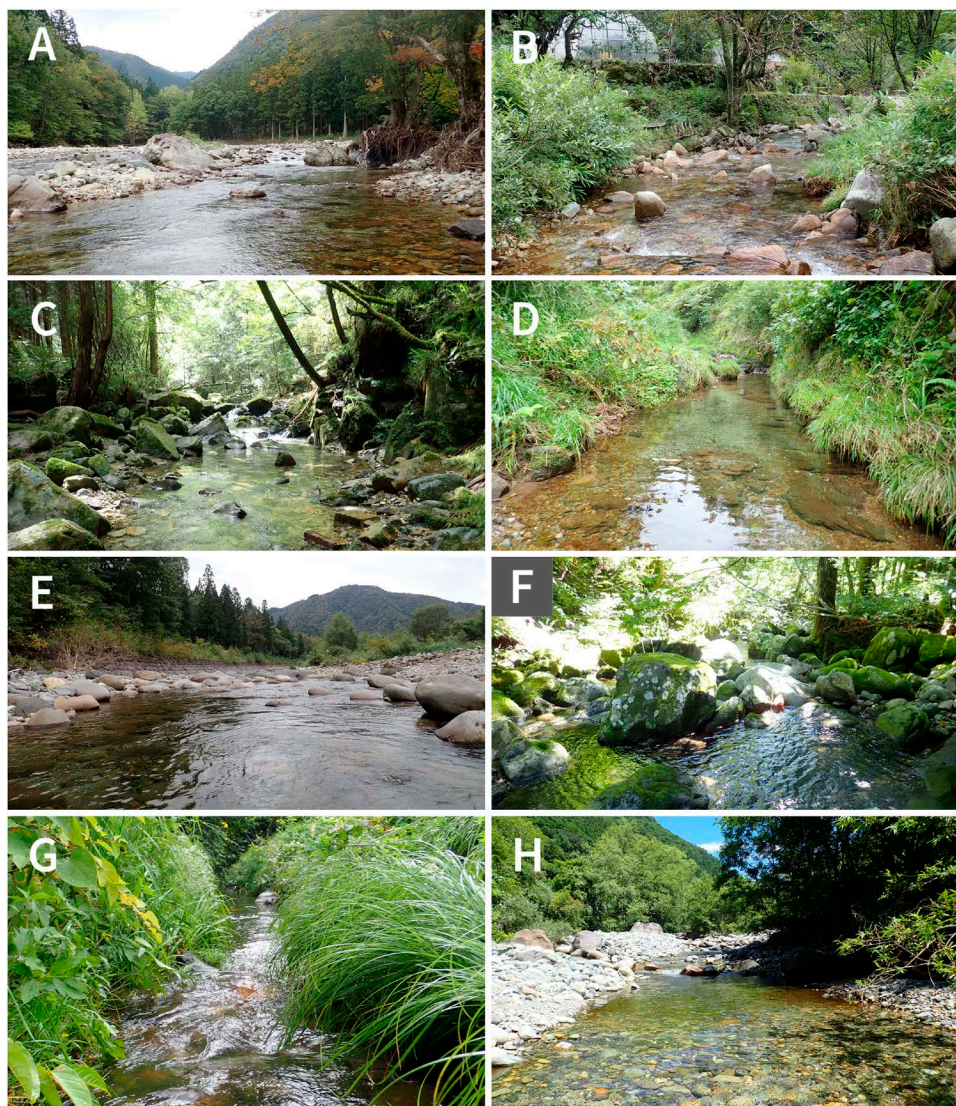


Fig. 3. Mountain streams where the salmonids infected with *Argulus coregoni* were caught in Gifu Prefecture, central Japan. A, Main stream of the upper Maze River (locality 1 in Fig. 2); B, tributary of the Hida River (locality 2); C, tributary of the Tsukechi River (locality 3); D, tributary of the Yoshida River (locality 4); E, main stream of the Itoshiro River (locality 5); F, tributary of the Itoshiro River (locality 5); G, tributary of the Sho River (locality 6); H, main stream of the Gamada River (locality 7).

Both red-spotted masu salmon and white-spotted char often co-occur in the upper reaches of rivers in central Japan, but the former is found in slightly lower streams than the latter (Kitano and Kubota 1999, 2007; Kishi and Tokuhara 2012). Similar distribution differences are also known for masu salmon and white-spotted char in the upper reaches in central Japan (Miyasaka et al. 2003). The highest elevations where the infected masu salmon and red-spotted masu salmon were caught in this study are, respectively, 707 and 733 m, both of which are lower than that (873 m) of the white-spotted char catch (Table 1). One extraordinarily collection was an infected red-spotted masu salmon was at 237 m elevation in the Yoshida River (location 4 in Fig. 2; Table 1), but this fish occurred in a small tributary flowing into the river (Fig. 3D). This tributary had clean water and gravel and cobble substratum conditions which were similar to those in the upper reaches. White-spotted char and the two masu salmon subspecies are predominant

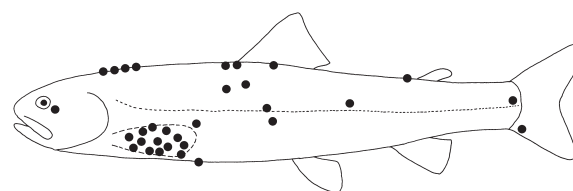


Fig. 4. Distribution of 31 specimens of *Argulus coregoni* (closed circles) on the host's body surface. A total of 32 specimens of *A. coregoni* were collected, but the attachment site for one individual was not recorded.

in the upper reaches of rivers in Gifu Prefecture (Kishi and Tokuhara 2012; Kishi et al. 2016), and these salmonids most probably serve as the major hosts of *A. coregoni* there. Further, based on the observed difference in their distribution within the upper reaches, it is likely that white-spotted char and the two masu salmon subspecies are important hosts in

Table 2. Previous records of *Argulus coregoni* collected from ayu, *Plecoglossus altivelis altivelis*, in rivers of Gifu Prefecture, central Japan.

River system	Collection locality			Location	Number in Fig. 2	Reference
	River	Reaches	Elevation (m)			
Kiso River	Maze River	Middle reaches	557	Maze-Nakagiri, Gero	8	Nagasawa et al. (2018)
	Shira River	Middle reaches	329	Kando, Higashi-Shirakawa	9	Nagasawa et al. (2018)
Nagara River	Nagara River	Middle reaches	430	Nagataki, Gujo	10	Nagasawa et al. (2020a)
			367	Shirotori, Gujo	10	Nagasawa et al. (2020a)
			269	Tokunaga, Gujo	10	Nagasawa et al. (2018); Nagasawa and Morikawa (2019a)
		Lower reaches	65	Sodai (reported incorrectly as "Soda"), Mino	11	Nagasawa et al. (2018)

the headwater and the middle to lower sections of the upper reaches, respectively.

There are six records of *A. coregoni* collected from ayu (*P. a. altivelis*) in the Kiso and Nagara river systems (Nagasawa et al. 2018, 2020a; Nagasawa and Morikawa 2019a) (Fig. 2; Table 2): the infected ayu were caught in the middle reaches at 269–557 m elevation and in the lower reaches at 65 m elevation. As stated in the Introduction, *A. coregoni* is not a strictly host-specific parasite, and many species of freshwater fishes are present in such reaches: for example, 36 fish species have been recorded from the lower reaches of the Nagara River (Goto and Goto 1971). Thus, freshwater fishes other than ayu possibly serve as the hosts of *A. coregoni* in the middle and lower reaches of rivers, but the presence of the six collection records of this parasite from ayu indicates that this fish is one of the important hosts in these reaches. Recently, Nagasawa and Morikawa (2019b) have suggested the importance of ayu as the host of *A. coregoni* in the middle reaches of rivers in Mie Prefecture, central Japan.

Based on the above discussion on the distribution and host utilization of *A. coregoni* in rivers, it can be summarized that 1) the species occurs from the upper through the middle to the lower reaches; 2) it utilizes different fish species as its hosts along a river course; 3) white-spotted char and the two masu salmon subspecies (masu salmon and red-spotted masu salmon) serve as the hosts, respectively, in the headwater and the middle to lower sections of the upper reaches; and 4) ayu is the important host in the middle to lower reaches.

The number of specimens of *A. coregoni* found on a single host was low, ranging from one to four, and, most commonly (60%), only one specimen was found per host. The smallest specimen was a male of 3.1 mm TL (collected in early September 2019) and is herein regarded as a larva at the eighth or ninth stage (see Shimura 1981). In our survey, we recognized the infected salmonids by naked eye, and there is the possibility that we overlooked smaller larvae at earlier stages in the field. In other words, the highest number of individuals of *A. coregoni* per host may be more than four.

The most common attachment site of *A. coregoni* on the examined salmonids was the body surface under and near the pectoral fins (Fig. 4). In this study, all of the infected salmonids were caught in mountain streams (Fig. 3), and it is inferred that *A. coregoni* utilizes that site not to detach

from the host's skin under the fast-flow stream conditions. A similar suggestion was made by Shimura (1983) regarding the distribution of the same parasite on masu salmon reared at a governmental fish hatchery in Tokyo. He stated that, as masu salmon are a swift swimmer, *A. coregoni* selects the sites sheltered from water flow.

The specimens of *A. coregoni* collected during the survey period (from late July to mid-October 2019) consisted of 14 males and 18 females. Based on their TL range (male, 3.1–9.6 mm; female, 6.0–11.0 mm) and the information on the hatching season and growth of the species at the fish hatchery in Tokyo (Shimura 1983), the specimens collected in this study are inferred to have hatched during the spring to summer of 2019 from the overwintered eggs that were deposited in the early to late fall of 2018. The presence of the above mentioned male larva (3.1 mm TL) in early September 2019 implies that this specimen hatched in July or August 2019. Further, ovigerous females of *A. coregoni* were collected throughout the whole survey period, which indicates that eggs are deposited at least from late July to mid-October in the mountain streams.

During this study, we collected *A. coregoni* from salmonids without recording prevalence of infection data, and the infected salmonids were caught at 10 sites in the seven rivers in Gifu Prefecture (Table 1). In this prefecture, due to intensive studies on the ecology of freshwater fishes, sufficient information has been accumulated on the distribution and abundance of salmonids in high elevation mountain streams (Kishi and Tokuhara 2012; Kishi et al. 2016). It is thus desirable to examine salmonids from many streams in various regions in order to clarify the host utilization, prevalence of infection, and life cycle of *A. coregoni* occurring in those mountain streams.

White-spotted char is distributed in Far East Asia, ranging from around the Kamchatka Peninsula, Russia, to central Honshu, Japan (Dunham et al. 2008). Gifu Prefecture is located near the southern limit of the distribution of the species, and its range and habitats in central Japan including the prefecture have been predicted to be limited by global warming (Takegawa et al. 2017), which suggests that *A. coregoni* will lose their local host populations. The geographical distribution of masu salmon is similar to that of white-spotted char, and red-spotted masu salmon occurs only in central and western Japan (Kato 1991). Gifu Prefec-

ture also lies near the southern limit of the distribution of these two masu salmon subspecies, especially masu salmon (Kawanabe 1989), and they may also be negatively affected by global warming. It is, therefore, important to study and record the present state of the distribution of the salmonids and *A. coregoni* in high elevation mountain streams for a future evaluation of the impact of global warming on the populations of these animals in central Japan.

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