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Analysis of the Gut Contents of the Internal Exotic Fish Species *Opsariichthys uncirostris uncirostris* in the Futatsugawa River, Kyushu Island, Japan

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Opsariichthys uncirostris uncirostris was introduced from Lake Biwa to several freshwater areas in Kyushu Island, and the gut contents of this fish were observed in order to evaluate the effect of internal invasion on native aquatic organisms. The fish specimens were captured in the Futatsugawa river, north-western Kyushu Island, Japan, between February 2007 and January 2008, and their gut contents were examined under a stereomicroscope. The gut contents of this exotic species comprised various prey animals, mainly fishes in most of the seasons. The empty gut ratio was approximately 80% in several seasons and 100% during February and March. This exotic species showed high selectivity for *Zacco* spp., *Pseudorasbora parva*, *Tanakia* spp., and *Rhodeus* spp., which had large populations at the study sites. In contrast, this exotic species showed no selectivity for *Carassius* spp. that also had a large population. *Acheilognathus* spp. that had a small population at this study site showed high selectivity as prey by this exotic species. *A. tabira nakamurae*, a native species, is listed as threatened in the Japanese Red Data Book. Native species with a small population and high prey selectivity were estimated to be the most negatively effected species by the internal invasion of the study site by this exotic species.

INTRODUCTION

The effect of the invasion of nonnative fishes on native fishes is a serious problem worldwide. Exotic species are introduced into different environments for aquaculture (Casal, 2006), eliminating mosquito larva (Hirose *et al.*, 1997; Tashiro *et al.*, 2007), game fishing (Hosoya, 2005), or from ships' ballast water (Sapota and Skora, 2005). These invaders have undesirable effects on native fish species: the population of the native species decreases because they are driven to seek alternative habitats, and their hatching success is reduced due to predation, depletion of food resources, or other pressures resulting from the invader's presence (Azuma, 1992; Yodo and Kimura, 1998; Maezono and Miyashita, 2003; Townsend, 2003; Corkum *et al.*, 2004; Katano *et al.*, 2005; Madeira *et al.*, 2005). In Japan, most studies have evaluated the effects of invasions by exotic fishes such as *Micropterus salmoides*, *Lepomis macrochirus*, and *Gambusia affinis* (Yodo and Kimura, 1998; Maezono and Miyashita, 2003; Tashiro *et al.*, 2007) on native fish species and have recommended methods for the extermination of the nonnative fishes (Katano *et al.*, 2005). Today, the rearing, movement, and release of nonnative fishes in the environments of native fishes are regulated by legislation.

Japan is facing other challenges due to such invasions. Exotic fishes have been introduced into different environments from internal regions for aquaculture and game fishing; these fishes commingle with the juveniles of the ayu *Plecoglossus altivelis altivelis*, which have been released into rivers (Katano and Mori, 2005). However, invasions by fishes from internal regions have not been as well investigated as those by fishes from foreign countries. Evaluation of the effects of internal invasion on native species is also an urgent need.

Opsariichthys uncirostris uncirostris introduced from Lake Biwa is known to distribute in the creeks around Ariake Bay in northwestern Kyushu Island, Japan (Onikura *et al.*, 2008). In Lake Biwa, this species feeds on small fishes such as *P. altivelis altivelis*, bitterlings, and gobies (Tanaka, 1964). This exotic species needs to be investigated in detail as a species that poses an environmental threat. We investigated the gut contents of this exotic species, which has been introduced from Lake Biwa, in order to evaluate the effect of internal invasion on native aquatic organisms.

MATERIALS AND METHODS

Specimen collection

O. uncirostris uncirostris individuals were collected from the Futatsugawa river (32°10' N, 130°26' E) and an irrigation ditch around this river in northwestern Kyushu Island, Japan. Samplings were conducted every month or twice a month from February 2006 to January 2007. The individuals were sampled 50 times using casting nets (mesh size: approximately 12 mm).

Observation

The individuals that were collected were used as

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specimens. In total, 490 (70–249 mm in body length) specimens were collected. These specimens were fixed in 10% buffered formalin, their guts were removed, and the gut contents were observed under a Nikon stereomicroscope.

Empty gut ratio and food selective index

Chiou *et al.* (2005) calculated the empty stomach ratio in order to analyze the feeding activities of Taiwan mauxia shrimp. Since *O. uncistrostris uncistrostris* has no stomach, we estimated the seasonal changes in its feeding activities by calculating the empty gut ratio (*EGR*). The *EGR* was calculated by using the following equation:

$$EGR = 100 \times N_{empty} / N_{total}$$

N_{empty} : number of individuals with an empty gut
 N_{total} : total number of specimens

Prey selectivity was also investigated in the context of seasonal changes by calculating the food selective index (E_i) of Yoshihara and Fukuda (1998). The number of individuals of each species captured as bycatch in 50 casts was counted, and this number was used as the stock biomass for each species at the sampling site (p_i). We also used the number as the population of each species at the site. In addition, the number of individuals of each species observed in the guts of the exotic species was counted and considered as the predation intake (r_i). The E_i was calculated using the following equation:

$$E_i = (r_i - p_i) / (r_i + p_i)$$

The E_i values are positive when the gut contents contain prey preferred by this exotic species, and they are negative when the prey preferred by this species decreases in the gut.

The E_i values and fish population data were established for each prey fish species every 2 months in order

to evaluate the impact of this exotic species on the native species. Log transformations were conducted on the fish population data, and a cluster analysis was performed by the Ward method by using these data.

RESULTS AND DISCUSSION

Food composition

Figure 1 shows the seasonal variation in prey–animal composition in the gut contents of *O. uncistrostris uncistrostris*. During April and May, the gut contents mainly comprised insects (38.5%) and Malacostraca (23.1%). This exotic species fed mainly on fishes (96.3%) during June and July, and the *Rhinogobius* sp. OR constituted 40.7% in the gut contents. During August and September, the fish species were mainly observed, and 58.3% of the gut contents comprised Cyprinidae individuals. Cyprinidae individuals were also preyed on between October and January. However, during February and March, all individuals of this exotic species had empty guts.

The analysis of the gut contents of *O. uncistrostris uncistrostris* revealed various prey animals, mainly fishes, in almost all seasons. Among the prey fishes, *Rhinogobius* spp. were the main prey in summer, while Cyprinidae individuals were the chief prey after summer. These characteristics of the feeding habit of *O. uncistrostris uncistrostris* corresponded with that of the native inhabitants of Lake Biwa (Tanaka, 1964). However, in the study sites in the Futatsugawa river, this species showed no feeding during winter. However, in Lake Biwa the juveniles and young fish of *P. altivelis altivelis* are often preyed on by this exotic species in winter (Tanaka, 1964). Although individuals of *P. altivelis altivelis* inhabiting Lake Biwa survive winter, those inhabiting the rivers in Kyushu Island survive winter by migration to coastal regions (Nishida, 2005). The ecological differences between *P. altivelis altivelis* from the 2 areas might be related to the difference in the feeding habits of *O. uncistrostris uncistrostris* between these 2 areas.

Seasonal changes of empty gut rate and food selectivity

Figure 2 shows the seasonal changes in the *EGR* of *O. uncistrostris uncistrostris* captured from the Futatsugawa river, as determined in this study, and the *EGR* of the individuals inhabiting Lake Biwa, as reported by Tanaka (1964). In the Futatsugawa river, the *EGR* was calculated as approximately 80% between April and January, and the values were 100% during February and March. In Lake Biwa, the values were less than 40% all year long; this was significantly lower than those in the Futatsugawa river (Mann–Whitney *U* test; $p < 0.01$). The *EGR* in the Futatsugawa river may be higher than those in Lake Biwa due to insufficient prey in the Futatsugawa river, which is a transmigration area of *O. uncistrostris uncistrostris*.

Figure 3 shows the E_i of *O. uncistrostris uncistrostris* and the population (stock biomass) of each prey animal.

Fig. 1. Seasonal changes in the composition of the gut contents of *O. uncistrostris uncistrostris* captured from the Futatsugawa river, Kyushu Island, Japan.

Fig. 2. Seasonal changes in the empty gut ratio (EGR, %) of *O. uncirostris uncirostris* captured from the Futatsugawa river, Kyushu Island, Japan and the EGR of the individuals inhabiting Lake Biwa, as reported previously by Tanaka (1964).

There were large populations of *Zacco* spp., *Carassius* spp., *Pseudorasbora parva*, *Tanakia* spp., and *Rhodeus* spp. at our study site. The E_i values of this exotic species were positive between August and January for *Zacco* spp.; during June and July, for *P. parva*; during April and May, for *Tanakia* spp.; and during August and September, for *Rhodeus* spp. In contrast, the E_i for *Carassius* spp., which had a large population, was negative, suggesting that this exotic species does not prefer this native fish as prey. In case of native fishes with small populations, the E_i was positive during October and November for *Abbottina rivularis* and *Biwia zezera*; during December and January, for *Acheilognathus* spp.; from June to July and from December to January, for *Rhinogobius* sp. OR; and during April and May, for *Odontobutis obscura*. These native fishes with small populations were estimated to be preyed on selectively by this exotic species.

Cluster analysis was performed using the E_i values and fish population data, and the prey fishes were classi-

Fig. 3. Food selective index (E_i) of *O. uncirostris uncirostris* for the population of each prey species in the Futatsugawa river, Kyushu Island, Japan. The dots, bars, and parentheses indicate the E_i values, populations, and the averages of the populations, respectively. The acronyms used for a prey animal species or genus are as follows: Ou1, *O. uncirostris uncirostris*; Zsp, *Zacco* spp.; Csp, *Carassius* spp.; Ar, *Abbottina rivularis*; Bz, *Biwia zezera*; Ph, *Pungtungia herzi*; Pp, *Pseudorasbora parva*; Tsp, *Tanakia* spp.; Asp, *Acheilognathus* spp.; Rsp, *Rhodeus* spp.; ROR, *Rhinogobius* sp. OR; Oo, *Odontobutis obscura*; O-C, other Cypriniformes; O-P, other Perciformes; O-O, other fishes.

fied into 6 groups: no population and no selectivity (Group 1), low population and no selectivity (Group 2), low population and high selectivity (Group 3), moderate population and no selectivity (Group 4), high population and moderate selectivity (Group 5), and high population and no selectivity (Group 6) (Fig. 4). This classification suggested that this exotic species had the largest impact in each season on the native species in Group 3. *O. obscura*, *Acheilognathus* spp., and *Rhinogobius* sp. OR were estimated to be at high risk from April to May; December to January; and June to July, and from October to January, respectively.

Predation of threatened fish species

Among the native species that were present in the gut of this exotic species, *Rhodeus ocellatus kurumeus* (Fig. 5), *Tanakia limbata*, and *T. lanceolata* are ranked as threatened species in the Japanese Red Data Book (Ministry of Environment, 2003). Although the E_i values for these native species were positive in several

Fig. 5. *Rhodeus ocellatus kurumeus* found in the gut of *O. uncicrostris uncicrostris* captured in the Futatsugawa river, Kyushu Island, Japan.

seasons, their average population was more than 50 individuals/50 casts (Fig. 3), suggesting that these threatened native species were not largely effected by the feeding behavior of *O. uncicrostris uncicrostris*. The E_i for *A. rivularis*, another endangered native species, was also positive from October to November (Fig. 3). Although the average annual of the population in this species was small (approximately 9.2), it increased from October to January. Therefore, this native species was estimated to be preyed upon by *O. uncicrostris uncicrostris* when its population size was large but not when its population size was small. Therefore, by performing the cluster analysis (Fig. 4), we classified this native species into Group 5 (high population and moderate selectivity) from October to November, and this result was confirmed in this study.

In this field sampling, 28 individuals of *Acheilognathus tabira nakamurae*, which are classified as *Acheilognathus* spp., were present in the gut contents; this species is also a threatened native species (Ministry of Environment, 2003). The population of *Acheilognathus* spp. indicates one-seventh of the population of the exotic species (*Acheilognathus* spp./ *O. uncicrostris uncicrostris*, 4.5/29.1) and had positive E_i values in winter (Fig. 3). By using cluster analysis, individuals belonging to this genus were classified into Group 3 (low population and high selectivity) (Fig. 4). These results suggest that *A. tabira nakamurae* is probably the most negatively effected species due to internal invasion of the study site by the exotic species *O. uncicrostris uncicrostris*.

Further ecological research involving parameters such as age, growth, and reproduction of this exotic species is needed in order to control internal invasion by this species. In addition, for some time in the future, we must monitor the changes in the populations of both the exotic and native threatened species at sites where both species are sympatric.

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Fig. 4. Similarity in the relationship between the population and the prey selectivity in each prey fish in each season. The acronyms used for a prey animal species or genus are the same as those used in Figure 3. The parentheses indicate the following seasons: AM, April to May; JJ, June to July; AS, August to September; ON, October to November; DJ, December to January. In each group, the average values of E_i and population are indicated in this figure.

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