

## Assessing the potential impact of nonnative fish on native fish by relative condition

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**Abstract:** Nonnative species (NNS) pose one of the most common threats to the conservation of biodiversity. Given the difficulty in measuring and documenting the specific impacts of introduced species on native species, indices of condition may provide an easy and inexpensive alternative. The aim of this study was to understand the impact of NNS on some common and endemic freshwater fishes, examining their relative conditions in the absence (allopatry) and presence (sympatry) of nonnative fish species from 3 different regions in the western part of Turkey. Data from 19 sites, a total of 1672 fish of 11 cyprinid species, were used for calculation of condition indices. Relative condition was significantly higher in allopatry than sympatry for *Capoeta sieboldii*, *Gobio gobio*, *Ladigesocypris ghigii*, *Leucaspius delineatus*, and *Phoxinus phoxinus*; however, it was higher in sympatry only for *Rhodeus amarus*, whereas *Barbus tauricus*, *Cyprinus carpio*, *Rutilus rutilus*, *Squalius fellowesii*, and *S. pursakensis* showed no significant differences in relative condition between allopatry and sympatry. Results suggest that there was a possible impact of NNS on native species and that relative condition would provide an easy and useful alternative to get a preliminary evaluation of the impact of nonnative fish species as long as sampling follows proper methodology.

**Key words:** Nonindigenous species, growth, endemic species, conservation, *Carassius gibelio*

### 1. Introduction

Among the many factors cited as common threats to the conservation of biodiversity, invasion of introduced nonnative species (NNS) is one of the most important causes leading to range reductions, population declines, and the extinction of freshwater fish around the world (Darwall et al., 2008). The spread of invasive species has also been associated with important social and economic impacts (Pimentel et al., 2000) and this is an ongoing phenomenon in many European countries (Gozlan et al., 2010; Aydın et al., 2011).

The freshwater fish fauna of the Mediterranean basin includes a high number of endemic freshwater fish species, most of which are threatened (Myers et al., 2000). Running waters in Mediterranean-climate regions are reported to be the most invaded systems around the world (Marr et al., 2010). In this respect, Turkey has an important role in Mediterranean-climate countries with 73 endemic species, 27% of which are listed as Critically Endangered and 32% as Endangered (Hermoso and Clavero, 2011).

Although the impacts of NNS on native fish communities in Turkey have become evident through recent studies (Gaygusuz et al., 2007; Aydın et al., 2011; Tarkan et al., 2012b), these studies have been restricted either to one highly invasive species (gibel carp, *Carassius gibelio*: Aydın et al., 2011) or a single catchment area (Ömerli Reservoir: Tarkan et al., 2012b). Furthermore, these studies mostly focused on life history traits of NNS (e.g., Balık et al., 2004; İzci, 2004; Tarkan et al., 2012a), probably due to difficulties in measuring and documenting the specific role of the introduced species on native species (e.g., Copp et al., 2010).

However, determining fish health by indices of condition may provide an easy and inexpensive way to understand the impact of NNS, as length and weight of threatened fish species can be measured while sacrificing them in minimum numbers (e.g., Fechhelm et al., 1995). One of these condition indices is relative condition (Le Cren, 1951), which has commonly been used for comparison of health of different populations of the

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same species with and without intraspecific interactions (Cucherousset et al., 2009; Tarkan et al., 2010). It would be a very useful tool as long as the populations are sampled at the same time of the year and in the same geographic region, hence under similar environmental conditions (Knaepkens et al., 2002). Similar size ranges and maturity stages of compared species should also be used (Copp et al., 2010). In this study, to assess the impact of NNS we used the relative condition of some common and endemic freshwater fish species from 3 different regions in the western part of Turkey, and we compared their conditions in the absence (allopatry) and presence (sympatry) of nonnative fish species.

## 2. Materials and methods

### 2.1. Study sites

Three different regions located in western Anatolia, Turkey (Muğla, İzmit, and Eskişehir), were visited for fish sampling. In total, 19 water bodies including mainly reservoirs and streams were used for the analyses. Study sites in the Muğla region included Mediterranean-type streams, which usually dry up during the summer months. These were 7 similar-sized streams ranging in length from 3.1 km (Akarca) to 32 km (Sarıçay). In the Eskişehir region, a big river and 2 relatively smaller streams were sampled. The Sakarya River is the third longest river in Turkey (810 km); its basin is divided into 3 parts under the names of Upper, Middle, and Lower. However, samples were only collected from the Upper basin in the present study. Porsuk and Emet are relatively smaller streams that are under threat by pollution caused by human disturbance. The İzmit region has many water reservoirs established mainly for the aim of irrigation and drinking water supply and one small stream (Çınarlı Stream). From these reservoirs, the Yuvacık Reservoir is the largest, which provides drinking and water usage for the İzmit area (approximately 1.5 million urban population). The other 7 reservoirs are close to each other and range from 0.17 to 1.6 km<sup>2</sup> with similar water depths (approximately 10 m). All water bodies sampled for each region were similar in trophic level, climate conditions, and environmental factors (Table 1).

Fish samples were collected by electrofishing in November 2010 from Eskişehir region and in May 2010 from the İzmit and the Muğla regions. Immediately after capture, fish were killed with an overdose of 2-phenoxyethanol, immersed in a slurry of iced water, and chilled to freezing. For threatened fish species, individuals sacrificed were kept to a statistical minimum. In the laboratory, specimens were defrosted and each was measured to the nearest millimeter for total length (TL) and to the nearest 0.1 g for total weight (W).

For each species, the length–weight relationship was calculated using a growth model of the type  $W = aTL^b$ ,

where  $a$  and  $b$  are the parameters of the equation using all fish data collected during the study as per Ricker (1975). Accounting for variation in body condition due to size differences, Le Cren's (1951) relative body condition was calculated for each species as per Copp (2003):  $LK = w/w'$ , where  $w$  is the observed and  $w'$  is the expected body weight as estimated from the length–weight relationship using the entire data set. According to Le Cren's (1951) relative body condition, values of  $>1.0$  indicate that the individual is in better condition than an average individual of the same TL range, whereas  $LK$  values of  $<1.0$  indicate that the individual is in worse condition than an average individual of the same length. As this index requires that populations are sampled at the same time of the year, region, and similar stage of lifespan (i.e. Knaepkens et al., 2002), all analyses and comparisons were done accordingly.

For nutrient analyses (nitrate [NO<sub>3</sub>], nitrite [NO<sub>2</sub>], total phosphorus [TP], orthophosphate [o-PO<sub>4</sub>], and sulfate [SO<sub>4</sub>]), water samples were taken from all study sites and kept cool in the dark until they were brought to the laboratory. All analyses were done according to standard methods (APHA et al., 1985). Several physical water parameters such as temperature, dissolved oxygen, conductivity, and pH were measured in situ using a multiparameter probe (YSI 650 MDS). To determine chlorophyll-*a* content, water samples were filtered and extracted through ethanol. After the centrifugation, absorbance was measured before and after acidification in a spectrophotometer and then calculated (Ryther and Yentsch, 1957). All analyzed environmental factors were compared between study sites that contained allopatric and sympatric populations separately for the 3 studied regions.

Relationships between length and weight of fish species were tested using nonlinear (power curve) regressions. Differences in relative condition values between allopatry and sympatry were assessed using Student's t-test while differences in environmental factors among the study sites were tested with one-way analysis of variance (ANOVA). When significant differences among the study sites were detected ( $P < 0.05$  was accepted as the level of significance), Tukey's honestly significant difference (HSD) test was used to determine which study sites and environmental factors were different.

### 3. Results

There were no significant differences in environmental factors measured among the study sites (ANOVA,  $P > 0.05$ ), except for o-PO<sub>4</sub> in the Muğla region having significantly higher values in study sites that have sympatric populations (ANOVA, Tukey's HSD test,  $P < 0.05$ ) (Table 1). Fish species were sampled from 19 water bodies, 11 of which contained NNS (Table 2). The most sampled region

**Table 1.** Environmental factors of the study sites from İzmit, Eskişehir, and Muğla regions. All comparisons within the regions were insignificant at  $P = 0.05$  except for orthophosphate in the Muğla region. T: Temperature ( $^{\circ}\text{C}$ ), C: Conductivity (mS/cm),  $\text{O}_2$ : Dissolved oxygen (mg/L), A: Altitude (m),  $\text{PO}_4$ : Phosphorus ( $\mu\text{g/L}$ ),  $\text{NO}_2$ : Nitrite ( $\mu\text{g/L}$ ),  $\text{NO}_3$ : Nitrate ( $\mu\text{g/L}$ ), TP: Total phosphorus ( $\mu\text{g/L}$ ),  $\text{SO}_4$ : Sulfate (mg/L), Chl-*a*: Chlorophyll-*a* (mg/L).

	T ( $^{\circ}\text{C}$ )	C (mS/cm)	pH	$\text{O}_2$ (mg/L)	A (m)	$\text{PO}_4$ ( $\mu\text{g/L}$ )	$\text{NO}_3$ ( $\mu\text{g/L}$ )	$\text{NO}_2$ ( $\mu\text{g/L}$ )	TP ( $\mu\text{g/L}$ )	$\text{SO}_4$ ( $\mu\text{g/L}$ )	Chl- <i>a</i> (mg/L)
İZMİT REGION											
Tahtalı Reservoir	21.1	355	8.3	8.40	253	2.50	0.06	0.10	5.86	17.31	2.66
Denizli Reservoir	21.2	361	8.3	8.47	246	7.38	0.03	0.10	8.31	15.83	0.89
Sevindikli Reservoir	20.0	262	8.4	8.83	280	3.97	0.06	0.10	13.74	17.44	0.00
Sipahiler Reservoir	21.3	346	8.2	8.39	275	2.82	0.01	0.10	5.19	17.16	1.33
Çınarlı Stream	13.3	450	8.4	9.51	208	3.23	0.05	0.10	12.78	9.30	0.00
Çayırköy Reservoir	22.6	381	8.2	12.00	64	3.81	0.11	0.09	4.30	18.72	1.77
Bayraktar Reservoir	21.7	391	8.4	8.58	94	7.24	0.04	0.10	16.89	21.57	2.66
Kirazoğlu Reservoir	20.8	409	8.5	8.65	105	4.95	0.16	0.10	8.31	19.40	0.89
Yuvacık Reservoir	18.5	219	8.7	8.60	190	17.85	0.07	0.10	20.46	16.96	0.00
ESKİŞEHİR REGION											
Porsuk Stream	11.3	435	7.5	8.70	800	1.62	0.02	0.03	-	30.70	-
Emet Stream	11.2	712	7.5	9.60	918	35.00	0.01	0.02	-	7.65	1.23
Sakarya River	11.1	976	8.1	8.20	874	2.75	0.01	0.01	-	218	11.50
MUĞLA REGION											
Akarca Stream	13.7	380	8.3	5.80	667	25.36	0.01	0.04	-	-	0.00
Tahliye Stream	12.7	504	8.4	6.92	115	12.20	0.01	0.01	-	-	0.00
Balıkli Stream	17.0	629	8.5	4.93	110	11.40	0.01	0.01	-	-	0.90
Sarıöz Stream	16.6	649	8.1	6.14	119	7.82	0.01	0.09	-	-	0.00
Gelibolu Stream	14.2	378	7.7	8.00	25	5.18	0.01	0.09	-	-	0.99
Tersakan Stream	13.9	997	8.4	7.80	16	4.12	0.01	0.09	-	-	0.91
Sarıçay Stream	11.7	337	8.3	8.42	25	41.74	0.01	0.03	-	-	10.12

was İzmit with 9 water bodies, followed by Muğla with 7, and Eskişehir with 3. A total of 1672 specimens of 11 fish species (4 of which are endemic) were used for calculation of the condition indices (Table 2). All of the length-weight relationships were significant ( $P < 0.001$ ). There were 823 individuals in allopatric populations as opposed to 849 in sympatric populations (Table 3). The mean relative condition (*LK*) value for allopatric populations ( $1.0400 \pm 0.06$ ) was significantly higher than in sympatric populations ( $0.9899 \pm 0.03$ ) (*t*-test,  $P = 0.007$ ). *LK* ranged from 0.9255 (*P. phoxinus* – sympatry) to 1.1251 (*L. ghigii*

– allopatry) (Table 3). Considering each species separately, *LK* was significantly higher ( $P < 0.05$ ) in allopatry than sympatry for 5 species (*C. sieboldii*, *G. gobio*, *L. ghigii*, *L. delineaatus*, *P. phoxinus*) but it was higher in sympatry only for *R. amarus*. Four species (*B. tauricus*, *C. carpio*, *R. rutilus*, *S. fellowesii*) showed no significant differences in *LK* between allopatry and sympatry ( $P > 0.05$ ) although they had higher *LK*s in allopatry (Table 2). Conversely, *LK* was higher in sympatry than allopatry for *S. porsakensis*; however, this difference was also not statistically significant ( $P > 0.05$ ) (Table 3).

**Table 2.** Latitude, longitude, altitude, native, and coexisting nonnative fish species of study sites in western part of Turkey.

Site name	Region	Latitude	Longitude	Native species	Nonnative species
Tahtalı Reservoir	İzmit	40°54'25"N	29°53'42"E	<i>Cyprinus carpio</i> , <i>Leucaspius delineatus</i> , <i>Squalius pursakensis</i>	<i>Carassius gibelio</i>
Denizli Reservoir	İzmit	40°53'59"N	29°33'57"E	<i>Rhodeus amarus</i> , <i>Squalius pursakensis</i>	<i>Carassius gibelio</i>
Sevindikli Reservoir	İzmit	40°53'14"N	29°45'40"E	<i>Cyprinus carpio</i>	N/A
Sipahiler Reservoir	İzmit	40°52'21"N	29°47'30"E	<i>Cyprinus carpio</i> , <i>Squalius pursakensis</i>	<i>Carassius gibelio</i>
Çınarlı Stream	İzmit	40°48'42"N	29°51'56"E	<i>Phoxinus phoxinus</i>	N/A
Çayırköy Reservoir	İzmit	40°48'30"N	29°59'10"E	<i>Gobio gobio</i> , <i>Leucaspius delineatus</i> , <i>Rutilus rutilus</i> , <i>Squalius pursakensis</i> , <i>Rhodeus amarus</i>	<i>Lepomis gibbosus</i>
Bayraktar Reservoir	İzmit	40°47'56"N	30°05'33"E	<i>Gobio gobio</i> , <i>Rhodeus amarus</i> <i>Rutilus rutilus</i> , <i>Squalius pursakensis</i>	<i>Carassius gibelio</i> , <i>Lepomis gibbosus</i> , <i>Pseudorasbora parva</i>
Kirazoğlu Reservoir	İzmit	40°45'46"N	30°06'44"E	<i>Rutilus rutilus</i>	<i>Carassius gibelio</i> , <i>Pseudorasbora parva</i>
Yuvacık Reservoir	İzmit	40°40'12"N	29°58'00"E	<i>Gobio gobio</i> , <i>Phoxinus phoxinus</i> , <i>Squalius pursakensis</i>	<i>Carassius gibelio</i>
Porsuk Stream	Eskişehir	39°38'43"N	30°17'27"E	<i>Capoeta sieboldii</i>	<i>Carassius gibelio</i>
Emet Stream	Eskişehir	39°25'44"N	29°07'13"E	<i>Barbus tauricus</i> , <i>Capoeta sieboldii</i>	N/A
Sakarya River	Eskişehir	39°21'27"N	31°03'05"E	<i>Barbus tauricus</i>	<i>Carassius gibelio</i> , <i>Oreochromis niloticus</i> , <i>Clarias gariepinus</i>
Akarca Stream	Muğla	37°07'38"N	28°23'20"E	<i>Ladigesocypris ghigii</i> , <i>Squalius fellowesii</i>	<i>Carassius gibelio</i>
Tahliye Stream	Muğla	37°00'48"N	28°30'34"E	<i>Ladigesocypris ghigii</i>	N/A
Balıkli Stream	Muğla	37°00'26"N	28°32'55"E	<i>Ladigesocypris ghigii</i> , <i>Squalius fellowesii</i>	N/A
Sarıöz Stream	Muğla	37°00'25"N	28°30'43"E	<i>Squalius fellowesii</i>	N/A
Gelibolu Stream	Muğla	36°57'08"N	28°17'11"E	<i>Squalius fellowesii</i>	N/A
Tersakan Stream	Muğla	36°48'25"N	28°52'50"E	<i>Ladigesocypris ghigii</i> , <i>Squalius fellowesii</i>	N/A
Sarıçay Stream	Muğla	38°19'43"N	27°42'46"E	<i>Squalius fellowesii</i>	<i>Carassius gibelio</i> , <i>Lepomis gibbosus</i> , <i>Pseudorasbora parva</i>

**Table 3.** Total length (TL, mm) range, number of specimens, parameters (*a* and *b*) of length–weight relationships, and relative conditions of native species from 3 different regions with (sympatry) and without (allopatry) nonnative species. Results of t-tests are also shown as P-values. Bold font shows significant differences between relative conditions at 95% confidence limits.

Species	TL (min–max)	Number of specimens		Parameters of length–weight relationships		Relative condition		P
		Allopatry	Sympatry	<i>a</i>	<i>b</i>	Allopatry	Sympatry	
<i>Barbus tauricus</i>	92–400	48	17	0.0068	3.0885	1.0129 (0.02)	0.9887 (0.02)	0.282
<i>Capoeta sieboldii</i> †	115–439	83	26	0.0060	3.1704	1.1147 (0.01)	0.9638 (0.01)	<b>0.022</b>
<i>Cyprinus carpio</i>	28–317	39	32	0.0177	2.9581	1.0094 (0.01)	0.9983 (0.02)	0.295
<i>Gobio gobio</i>	38–128	89	107	0.0096	3.0891	1.0521 (0.01)	0.9757 (0.01)	<b>&lt;0.001</b>
<i>Ladigesocypris ghigii</i> †	39–74	17	34	0.0105	3.0706	1.1251 (0.03)	1.0155 (0.03)	<b>0.001</b>
<i>Leucaspius delineatus</i>	30–55	41	140	0.0114	2.8946	1.0602 (0.03)	0.9973 (0.01)	<b>0.001</b>
<i>Phoxinus phoxinus</i>	20–90	274	180	0.0100	3.1321	1.0690 (0.01)	0.9255 (0.01)	<b>&lt;0.001</b>
<i>Rhodeus amarus</i>	31–90	35	67	0.0111	3.1521	0.9316 (0.02)	1.0293 (0.01)	<b>0.001</b>
<i>Rutilus rutilus</i>	28–130	15	46	0.0109	2.9925	1.0519 (0.03)	0.9964 (0.02)	0.095
<i>Squalius fellowesii</i> †	34–365	98	23	0.0081	3.1351	1.0104 (0.01)	0.9933 (0.01)	0.181
<i>Squalius pursakensis</i> †	36–224	84	177	0.0092	3.0992	0.9990 (0.01)	1.0049 (0.01)	0.356

† indicates endemic species for the region.

#### 4. Discussion

Nonnative freshwater fish introductions have been demonstrated to cause decline in native freshwater fish species (Byers et al., 2002; Tarkan et al., 2012b) and in some cases even extinction (Lorenzoni et al., 2006). Similar reports have increasingly been noted from Turkish waters in recent years (Fricke et al., 2007; Gaygusuz et al., 2007; Aydın et al., 2011; Tarkan et al., 2012b). In the present study, impacts of NNS are implied by differences in relative condition values between allopatric (native fish species living without NNS) and sympatric populations (native fish species living with NNS) (Table 3).

It is well known that growth in fishes is a crucial mechanism affecting their reproduction and survival (Mangan et al., 2005). The introduction of NNS, in this respect, would generate a considerable decline in native fishes by worsening their individual growth and condition through interspecific competition (Bohlin et al., 1994; Latini and Petrere, 2004), and this may provide for a simple and easy way to assess the impact of NNS. Indeed, interspecific competition, especially in cases of food shortage, can have a substantial adverse effect for fish

species (Resetarits, 1997) and this can be tested better with closely related species that use similar resources (Werner, 1986), as is the case in the present study. Initial evaluation of feeding behavior between native and nonnative fishes from the İzmit region revealed that there were significant food overlaps, especially between gibel carp and native species (unpublished data). Similarly, it has been indicated recently that growth of crucian carp, *Carassius carassius*, in a small lake was negatively affected by high densities of coexisting fish species (Tarkan et al., 2011) and this would be more prominent for an indigenous fish species when competing with a nonnative fish species (e.g., Cucherousset and Olden, 2011). A long-term study on this matter clearly demonstrated competitive exclusion of a native fish (*Coregonus lavaretus*) from its original niche by its invading congener nonnative fish species *C. albula* (Bohn et al., 2008).

The comparison of relative condition of fish species in the present study suggested a possible impact of NNS on native species (Table 3). In contrast to other examined species, only *R. amarus* had higher condition values in sympatry than allopatry. This might be due to unique

biological features of this species with its unusual mode of reproduction and habitat preference (i.e. obligate parasite of unionid mussels) (Reichard et al., 2010). Although Blackwell et al. (2000) emphasized that the relationship between relative weight and density in fish would only be possible in lentic environments, this was not the case in the present study, as in both lentic and lotic environments (reservoirs – İzmit; streams and rivers – Muğla and Eskişehir) significant relationships were detected. Indeed, our results revealed that the relative conditions of 3 stream-dwelling fish (*P. phoxinus*, *L. ghigii*, *S. fellowesii*) were also lower in sympatry (Table 3). This may suggest that NNS occupy recipient environments completely covering all parts of the habitats where native species exist. It seems likely, as the most common NNS was gibel carp in the present study and it was found in 10 of 11 sympatric populations being mostly or only NNS (Table 3). Since this fish species is very robust, it can live under adverse environmental conditions (Liasko et al., 2011; Tarkan et al., 2012a) and easily adapt to a wide range of different habitat types (Aydın et al., 2011). Therefore, this species has become one of the dominant fish in most of the freshwater systems of Turkey, facilitated mainly by human-aided dispersal (Önsoy et al., 2011). Undeniably, the negative impact of gibel carp through reproductive interference has recently been recognized and the decline of native cyprinid fish populations has been associated with the introduction of this species in many parts of its invaded areas (Vetemaa et al., 2005; Smartt, 2007), including Turkish waters (Aydın et al., 2011; Tarkan et al., 2012b). Lower conditions in native species living with gibel carp may reflect the negative effect of gibel carp by a high interspecific competition in sympatry. Another NNS in the present study, pumpkinseed *Lepomis gibbosus* as a predatory species, and topmouth gudgeon *Pseudorasbora parva*, North African catfish *Clarias gariepinus*, and Nile tilapia *Oreochromis niloticus* as powerful competitors for native species, might have an influence on native species, but given that their abundance and presence were not so high in the study sites, these species would have not contributed to the decline of native species' condition, at least not as much as gibel carp.

Although this study revealed a consistent pattern for greater condition in allopatric populations, differences in condition values between allopatric and sympatric populations could not be attributed directly to introduced NNS, as these differences may be related to biotic and abiotic variations among the sites. To compare condition factors of all studied populations, environmental factors of examined study sites regardless of the presence of

nonnative competitors should be identical, or differences should be taken into account in analyses, because native fishes considered in the present study may be influenced by environmental factors, such as food availability, temperature, or presence of predators (e.g., Önsoy et al., 2011). However, these factors are not likely to have had a major influence on our results, as all study sites examined in the same region are geographically close and have similar environmental characteristics in terms of trophic level and climate regime. Comparison of large numbers of environmental factors including chlorophyll-*a* production also clearly indicated that all examined water bodies had very similar environmental characters (Table 1). The only detected difference was significantly higher orthophosphate concentrations in sympatric populations of the Muğla region (Table 1), which is often considered an indicator of productivity and a stimulating factor for growth of fishes (e.g., Motzkin et al., 1982). However, conditions of native fish species in sympatric populations of this region were also lower than those in allopatric populations (Table 3), which strengthens the suggested negative impact of nonnative on native fish in the present study. Finally, our data did not confirm the presence of any predatory fish in all water bodies sampled, which could prey on both native and nonnative fishes (Table 2). Furthermore, we paid attention to having similar phases of life and size ranges for compared species, since different maturity stages can cause a bias in comparisons (Table 3).

In conclusion, it can be expected that reduced condition levels of native fish species could be due to coinhabitation with NNS. Recent detailed studies on the effect of NNS on native fauna and ecosystems have highlighted difficulties of assessing the impact of NNS (Copp et al., 2010). On the basis of our results, however, relative condition would provide a useful way to obtain preliminary results for evaluating the impact of NNS as long as sampling follows proper methodology (i.e. Knaepkens et al., 2002; Copp, 2003). However, to understand the real-world impact of NNS, more comprehensive studies should be performed with large and long-term datasets of both native and nonnative species.

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