

A fine scale analysis of heavily invaded Italian freshwater fish assemblages

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Abstract

Inland waters are highly vulnerable to the introduction and spread of non-native species, due to heavy human use of aquatic ecosystems and the natural linkages among streams and lakes. This is particularly noticeable in freshwater fish communities. To better evaluate how these communities are affected by non-native species introductions, we conducted a fine scale analysis of the changes in Italian freshwater fish assemblages after species introduction. For this analysis, we collected information on fish species present in 44 basins.

The present Italian freshwater fish fauna is composed of 48 native and 41 established introduced species, while a further 15 introduced species were reported but not yet considered naturalized. The changes of the fish assemblages mostly took place in the last two centuries and have increased recently, with nearly 60% of the species introduced in the last three decades. The number of species introduced per basin ranged from 0 to 35 (mean 10.85 ± 7.77 species/basin), and in ten basins the number of species introduced is now equal to the number of native species or even higher. In the past, introduced species mainly originated from America, but in last decades, an increase of

introductions from other parts of Europe and Asia was recorded. Our results show that basins already rich in native species tend to get even richer as a consequence of the establishment of introduced species. This confirms the trend toward a biotic homogenization of ecosystems even at a local scale, due to an increase in the human-mediated spread of generalist species.

Key words: biological invasions, Italy, non-native species, Pisces, species introductions.

Introduction

The introduction of non-native species changes ecosystem composition and functioning, altering the relationship between species and energy flow and, consequently, affecting human well-being (Ehrenfeld 2010; Strayer 2012). Compared to terrestrial systems, inland waters are highly vulnerable to the introduction and spread of new species. This is related to heavy human use of aquatic ecosystems, the natural linkages among streams and lakes, the effects of water flow regulation and the linked dispersal capability of aquatic species (Leppäkoski et al. 2002; Gherardi 2007; Gherardi et al. 2008). The extent of species introduction is particularly impressive in freshwater fish communities. Leprieur et al. (2008) identified six major worldwide invasion hotspots where non-native fish species represent more than a quarter of the total number of species. The impact of these changes is yet to be well understood. According to the IUCN Red List of Threatened Species (Vié et al. 2009), introduced species are major threats to freshwater fishes in South Africa, coming second after water pollution, with southern Europe as one of the invasion hotspots identified by Leprieur et al. (2008).

In general, the major drivers of fish species introductions (revised by Tricarico 2012) have been: 1) angling (through increasing wild stocks for professional or sport fishing); 2) biocontrol (species released as control agents of other species); 3) commercial (introductions for fish farming and related purposes); 4) filling a vacant niche (introductions to fill a perceived ecological void in the fish community, although the concept of a vacant niche is erroneous, given that the niche is a property of the species and not a space to fill); 5) ornamental (aquarist or other ornamental purposes); and 6) research (introductions for experimental purposes). By far, the primary motivation for introduction was the desire to have new species for fishing, followed by ornamental purposes (Elvira 2001; Copp et al. 2005; Tricarico 2012).

In recent years, a certain number of papers have been published on changes in freshwater fish assemblages after species introduction at the country level (e.g. Elvira 1995; Rahel 2000; Musil et al. 2010; Rabitsch et al. 2013). However, these studies focused on the inventory and subsequent analysis of main basins. Considering that most of the introductions are human-mediated and intentional and that fish species can spread along river systems, changes in fish communities are likely to have reached even the secondary reservoirs. Therefore, a comprehensive analysis is necessary to understand better the effects of fish introductions and, consequently, to develop appropriate mitigation strategies.

Italy is an excellent case study to assess the situation of alien fishes, as its freshwaters have been heavily invaded for several years (Gherardi et al. 2008; Nocita & Zerunian 2007). Here, we present a fine scale analysis of the changes in the Italian freshwater fish assemblages following species introduction. For the purposes of the present study, the Italian territory was divided into 44 basins, for which information on native and introduced species was collected. This allowed an evaluation of species richness and the importance of species introductions at a fine spatial scale. Our aims were to 1) assess the native species richness in all the Italian watersheds, 2) compile a list of the basins more invaded and of the species more widely introduced, 3) evaluate how freshwater fish communities have changed after species introduction, and 4) identify spatial and temporal trends in native species assemblages and the intensity of introductions.

Material and Methods

We divided the Italian territory into 44 groups of drainage basins (ISTAT 2006), i.e.: Po, Fissero-Tartaro-Canalbianco, Adige, Brenta-Bacchiglione, Piave, Livenza, Tagliamento, Isonzo, Lemene, Friuli Venezia Giulia, Internazionale, Veneto, Liguria, Emilia-Romagna, Conca-Marecchia in the North, Reno, Magra-Vara, Arno, Toscana, Fiora, Tevere, Marche, Lazio, Abruzzo, Sangro, Terno in the Centre, and Ofanto, Liri-Garigliano, Volturno, Sele, Campania, Fortore, Molise, Trigno, Saccione, Puglia, Noce, Basilicata, Sinni, Bradano, Lao, Calabria in the South, and the two main islands of Sicilia and Sardegna (Table 1; see also map in Figure S1).

We measured the effects of introductions and translocations on the freshwater fish assemblages considering changes in α and β diversity. In our case, α -diversity is the mean species richness at the basin level and β -diversity is the turnover in species occurrence across basins. We built matrices of fish species presence/absence at the basin level, composed of 1) native species as proxies of the original assemblages, 2) introduced species, 3) native and introduced species, 4) native, introduced, and translocated species (i.e. Italian native species moved within the country into a basin different

from its native one). The last two matrices describe the present assemblages with or without translocated species.

To evaluate α -diversity for each basin, we retrieved data on the presence of native, translocated and introduced fish species. For three basins (i.e. Lao, Puglia, Internazionale), we could not collect any information on the presence of fish species, and therefore they were not considered in the analyses. We also collected information on the area of origin for 41 introduced fish species and their introduction dates, here considered as the date of first detection of established species in Italy (N = 39). We considered only introduced or translocated species for which there was evidence of naturalization (i.e. presence with breeding populations) and, therefore, species with only occasional reports were not included in the database. Some cases of single and sporadic occurrences are cited separately. The database has been compiled gathering the information available so far from about a hundred scientific articles, papers from grey literature and unpublished data of one of the authors. The collected information was processed after validation and implementation (Table S3). In particular, most information was obtained from local reports called “Carta ittica” (i.e. Fish map), where the list of fish species for the basins of a certain area is drafted in order to manage the ichthyofauna for conservation and angling purposes.

For some fish species complexes, it was not possible to evaluate if a native component of the complex is still present and whether introduced species of the same complex or even hybrids are now present. Therefore, we decided to use a conservative approach, considering as native in every basin whenever present the complexes for *Salvelinus alpinus*, *Thymallus thymallus*, *Gasterosteus aculeatus*, *Gobio gobio* and *Esox lucius* (Bianco & Delmastro 2011; Bianco 2014; Meraner & Gandolfi 2012). On the contrary, the *Salmo trutta* complex was considered introduced everywhere because, even if the native species was present in Italy before the 19th century, due to continuous restocking with alien taxa for decades, it is no longer possible to distinguish morphologically native from introduced trout. For example, the presence of a native lineage of trout was reported from the XVI century in the Fiora Basin in Tuscany (Piccolomini 1584). We acknowledge that these complexes include species introduced in at least part of their range, and therefore the list of species introduced in Italian basins is probably longer than what is presented here.

For nomenclature, we used the book by Kottelat & Frejof (2007). However, for very similar species (e.g. species belonging to the *Salmo* complex), the name used in the aforementioned local reports did not correspond to the real taxon to which the species belongs. Thus, the information was integrated with scientific papers on genetics and systematics of that species (e.g., Meraner & Gandolfi 2012; Bianco 2014). In some cases, the introduction of alien species was rapidly progressing and an integration of data was needed (Piazzini et al. 2010; Puzzi et al. 2010).

To evaluate the level of alteration of fish assemblages and allow comparisons with other countries, a zoogeographic integrity coefficient (ZIC) was calculated as the ratio of the native to the total species present in a basin (Bianco 1990; Elvira 1995). This index ranges from 1, indicating a basin in pristine condition, to 0 (at least theoretically) for completely altered water bodies hosting only introduced species.

We measured the β -diversity among every pair of basins using Jaccard's index of similarity (Jaccard 1912). The index equals zero when two assemblages share no species and equals unity when they have identical species composition. Changes in β -diversity were assessed by comparing the native historical fish assemblages and the present assemblages including introduced and translocated species. An increase in the index indicates that assemblages become more similar from historical to current periods with a tendency toward homogenization. On the contrary, a decrease in the index refers to assemblages that become more dissimilar with time due to introductions.

To better evaluate the spatial trend of species introduction, we used explorative analyses with the aim to identify homogenous (i.e. similar) groups of basins based on presence-absence of introduced species. Since classical cluster analysis also returns groups composed of only one or two basins (outgroups), whereas we were more interested in identifying larger spatial aggregations of basins, we used a K-means clustering procedure. This method allows separating a set of data into a user-specified number of clusters that are as distinct as possible. Computationally, the program starts with K random clusters and then moves objects (basins) between those clusters with the goal of minimizing variability within clusters and maximizing variability between clusters. We started specifying basins to be separated into two clusters, then continued with three, four clusters, and so on, until one or more clusters were composed only of a single basin, considering then the previous aggregation.

When building the regression curves, the models that fitted the data were assessed by comparing the fit of the more complicated equations with that of a simpler equation for a straight line, using the significance and the level of R^2 as guidance. Correlations were evaluated with the Spearman's rank test. All statistical analyses were performed using SPSS version 22.0.1. (IBM Corp. Released 2013), except Jaccard indexes computed with EstimateS (Colwell 2013).

Results

The freshwater fish fauna native to Italy is composed of 48 species (Table S1), although for five species complexes, no distinction between native and introduced populations was possible. Species

richness per basin ranged from 1 to 42 (mean: 15.88 ± 10.60 species/basin: Table 1). Twenty native species have been translocated into other basins outside their native range (Table S1). Five species have been translocated into 11-18 basins (*Barbus plebejus* 18, *Scardinius hesperidicus* 16, *Barbus caninus* 13, *Perca fluviatilis* 13, *Alburnus arborella* 12), all the others into 1-8 basins (overall mean 6.30 ± 5.51 basins/species). Each basin received 0-13 new species (mean 3.07 ± 3.17 , Table 1).

We collected information on 445 introductions relating to 41 non-native species (Table S2). The number of species introduced per basin ranged from 0 to 35 (mean 10.85 ± 7.77 species/basin, Table 1), and in 10 basins the number of species introduced was equal to the number of native species or even higher (100-166.7%). Adding the species translocated to those introduced from outside Italy, the basins with an equal or higher number of non-native species increased to 19 (46.3% of the total).

The number of native and introduced species per basin was correlated ($R^2 = 0.68$, slope 0.83, $P < 0.001$; Fig. 1a), while there was no correlation between native and translocated species numbers.

Fifteen species were introduced into more than ten basins and ten species in more than 20, with a mean of 10.85 ± 11.00 basins/species (Table 2). Three species are now present in most of the basins: *Salmo trutta* 39, *Cyprinus carpio* 34, *Oncorhynchus mykiss* 31. The number of basins where single species were introduced followed a pattern similar to the number of basins where single native species were present (Fig. 1b, $r_s = 0.99$, $P < 0.001$).

The mean Jaccard index considering all possible pairs of basins with historical assemblages (i.e. only native species) was 0.31 ± 0.18 . Present assemblages were more similar in respect to the original state, with an increase in the mean Jaccard index to 0.32 ± 0.16 , including introduced species, and to 0.35 ± 0.16 , adding translocated species. Changes in similarities and dissimilarities through time for every pair of basins are reported in Fig. 2.

The K-means procedure resulted in three clusters composed of 6, 7 and 27 basins, respectively. Continuing the analysis with four or five clusters, we obtained one or two clusters with a single basin respectively. The three clusters were composed of basins close together in north Italy plus the Tevere River, the basins in south-central and south of the country and the basins in the center-north and north-east of the country (Fig. 3).

Among the fifteen more invasive species, five are listed in the IUCN list of the 100 of the World's Worst Alien Species (Lowe et al. 2000) and one in the 100 of Europe's Worst Invasive Alien Species (Vilà et al. 2009) (Table 2). According to Genovesi et al. (2015a), four of these species have an impact on native species considered threatened in Europe (IUCN Red List: CR, EN, VU).

The zoogeographic integrity coefficient was high in some basins with few species (Saccione 1.00, Campania 0.82), but also in some rivers with complex and slightly altered communities (e.g.

Tagliamento and Lemene 0.74). However, mean ZIc was low, at 0.53 ± 0.15 , and in 21 out of 41 basins (51.2%), the index ranged between 0.40-0.60 (Fig. 4a).

Fish species introductions in Italian freshwaters sharply increased in recent times (Fig. 4b). Before 1800, only two species were introduced: *Cyprinus carpio* and *Carassius auratus*. In the last two centuries and until 1980, 0.50-1.50 species have been introduced every ten years (mean 0.84 ± 0.42 /ten years). The rate of introduction species increased to 6.76 species/ten years from 1980 to 2014 (exponential curve $R^2 = 0.65$, $F_{1,6} = 11.25$, $P < 0.05$). The cumulative number of introductions is explained by a cubic curve (Fig. 4d, $R^2 = 0.96$, $F_{3,5} = 41.20$, $P < 0.001$).

Regarding the area of origin, about one third of the species originated from America (31.7%), another third from other parts of Europe (34.1%) and one third from Asia (14.6%), the Palearctic (12.2%) and Africa (7.3%). Dividing the introductions into three periods (up to 1900, 1901-1980, 1981-2014), there was a decrease of introductions from America and an increase from other parts of Europe and Asia (Fig. 4c) over time.

Discussion

For the first time, we present a fine scale analysis of the composition of Italian freshwater fish assemblages composed of native, translocated and introduced species. Today, Italian basins have been invaded by at least 41 established introduced fish species, almost equalling the number of native species. Another five species complexes are composed of groups of species difficult to interpret as native or introduced in Italian basins. Applying a conservative approach, these species were considered only as native. However, these complexes include species probably introduced in some basins, and a clarification of their status could lengthen the list of species introduced in Italy. Furthermore, in the reviewed literature and on the basis of personal communications with other researchers, at least another 15 introduced species are reported as single records or acclimatized in at least one Italian basin without evidence of their naturalization (e.g. *Channa micropeltes*, Piazzini et al. 2014; *Neogobius melanostomus*, Busatto et al., 2016; *Clarias gariepinus*, *Poecilia reticulata*, *Xiphophorus helleri*, *Luciobarbus graellsii*, Nocita et al., 2014; some other Poeciliidae, Nonnis Marzano pers. comm.). It is, however, likely that at least some of them will become naturalized, also facilitated by the ongoing climate change, further increasing the list of established species.

Although there is a long history of species introductions around the world, only two fish species were reported to have been introduced into Italy before the 19th century. The alteration in the Italian freshwater fish assemblages mostly happened in the last two centuries and has recently been

increasing, with nearly 60% of the species introduced in the last three decades. This evidence is particularly worrying because it highlights a trend with a steady increase in the number of introductions in recent years, as already shown for other European countries (e.g. Elvira & Almodóvar 2001; Keller et al. 2009; Rabitsch et al. 2013).

At present, in ten basins the number of species introduced is equal to the number of native species or even higher. This number nearly doubles when we also consider species that were translocated into other basins. A few species were introduced in most of the basins and many others in nearly half of them, indicating a human interest to introduce a pool of species that are now ubiquitous in the Italian basins as well as in many other parts of the world where they have been widely introduced (García-Berthou et al. 2005; Marr et al. 2013). The alteration of the fish assemblages is further accentuated by the translocation of Italian species from one basin to another outside their native ranges. This phenomenon was usually limited to a few species per basin, but, in some cases, we found many more. As a result, in nearly half of the basins, the number of species was twice as high compared to the native situation.

Considering the composition of the introduced community, the basins were aggregated into three groups generally composed of basins close together, indicating similar patterns of species introduction in neighbouring areas. However, and quite surprisingly, the communities of introduced species in basins in north-east Italy were more similar to basins in central Italy than to the rest of the northern country. This could be due to historical reasons: in central Italy local aquaculture facilities produced stocking materials for local needs, especially for salmonids, but, when this production was not sufficient, trout were provided by aquaculture facilities located in north-east Italy (Sommani 1969). Similarly, in Rome (Tevere basin) there was an important “ichthyogenic center” that frequently used the facilities located in north Italy (Po basin) as sources for stocking materials, especially in the 1950s and 1960s (Sommani 1969). These movements of animals between north and central Italy could help to explain the observed patterns of introduced species dissimilarity between basins. In southern basins, hydrographical characteristics and climate together with a less marked tradition of angling and aquaculture production allowed the maintenance of native structure in the fish communities more than in north and central Italy.

Introduced and translocated species together increased the mean similarities between basins by 4%. However, translocated species had a greater homogenization effect (+3%) in respect to introduced ones (+1%). This is in agreement with evaluations at a European scale (Leprieur et al. 2007), where translocated species increased similarities while those introduced contrasted this process, decreasing similarities. Clearly, native species already present in some basins and translocated into others increase the similarities between them, while the introduction of new species could have contrasting

effects. Species introduced in a few basins differentiate them from the others, while the introduction of common species in many basins leads to a higher homogenization (Olden & Poff 2004; Toussaint et al. 2016).

Humans have provided a variety of pathways by which aquatic species can circumvent historical, ecological and geographical barriers that contributed to the establishment of unique regional fauna assemblages (Rahel 2000). Among the species most widely introduced in Italian rivers, twelve are listed among those most introduced in Mediterranean-climate regions (Marr et al. 2013) and six are included among the ten freshwater fishes most frequently introduced worldwide (García-Berthou et al. 2005). This confirms the trend toward a biotic homogenization of ecosystems, due to an increase in the human-mediated spread of generalist species, while specialized species are exposed to the risk of extinction as they are more sensitive to global changes (McKinney & Lockwood 1999; Clavel et al. 2010).

The number of native and introduced species per basin and the number of basins where single native or introduced species were present were highly correlated. This indicates that the distribution patterns of native and introduced species are currently quite similar. This positive relationship seems to confirm several studies that have falsified Elton's (1958) original hypothesis of "biotic resistance" (e.g. Gido & Brown, 1999; Jeschke & Strayer, 2005; Pino et al., 2005; Leprieur et al., 2007). According to Elton (1958), species-rich communities resist biotic invasions better than species-poor communities do, as the higher number of biotic interactions in species-rich communities excludes or restricts the recruitment or persistence of newcomers. Our results, on the contrary, show that basins already rich in native species tend to get even richer as a consequence of the establishment of aliens. There are other examples in freshwater ecosystems of the "rich get richer" effect (Stohlgren et al., 2003). Californian watersheds, subject to a high rate of invasions by fishes, are today characterised by a large community of both native and alien species (Marchetti et al., 2004). Similarly, the Mississippi River basin, with the world's richest endemic assemblage of freshwater mussels, has been invaded by the zebra mussel in extremely high densities (Ricciardi et al., 1998). On the contrary, a single invader, such as the Nile perch in Lake Victoria, has greatly devastated diverse endemic fish communities (Goldschmidt et al., 1993). Our results might be explained by the wide habitat heterogeneity as indicated by the high richness in native species. This ecological heterogeneity might in turn provide more opportunities of success to alien species (Eriksson et al., 2006), partly in accordance with the "biotic acceptance" hypothesis. Alternatively, however, this phenomenon might be a by-product of other factors, such as latitude. In fact, alien

species richness follows a latitudinal gradient in Italy, along with the extent of artificial areas and the density of roads, fish farms and fishing shops (Gherardi et al., 2008).

Overall, human introductions have produced an assembly of alien fishes in Italian freshwater that mimics the native community in number of species and their distribution in basins. In just two centuries, humans have built through introduced species what took evolution thousands of years with native species. This reshuffling was done by bringing species from all over the world into Italy. In the past, most of the species were imported from North America, but there is a progressive increase of species from Eastern Europe and Asia. A similar pattern was already reported for Germany and Austria (Rabitsch et al. 2013), being probably common in Europe with Asian species often used in aquaculture and Eastern European species most introduced after the fall of the Berlin Wall (Britton & Gozlan 2013). The effect of this reshuffling is the loss of the typical Italian fish communities with their evolutionary and biogeographic uniqueness.

The magnitude of the impacts of introduced species on the native Italian fish fauna is still largely unknown. The European catfish *Silurus glanis*, for example, is one of the most remarkable invasive species in freshwater habitats of Southern Europe. Introduced into North Italy in 1957 (Manfredi, 1957), it is considered responsible for the fish fauna decline in terms of biodiversity and community composition, due to its wide piscivorous diet and large size (adults of 100 kg are quite common in the Po River). This is in accordance with the international literature, suggesting that catfish may be responsible for a reduction in biodiversity (Copp et al., 2009). Unfortunately, for Italy, in-depth scientific studies on the impacts caused by this species are lacking, and information is only anecdotal. The topmouth gudgeon *Pseudorasbora parva*, introduced into Italy in 1990 (Sala & Spampinato, 1991), shows highly invasive characteristics, such as plasticity in habitat exploitation, as it occupies diverse types of waterbodies and microhabitats, although more frequently, the species is associated with submerged vegetation where it occurs in high densities. The species shows a wide range of food preferences and the capacity to change its diet in the introduced range (Gozlan et al. 2010), resulting in a large overlap of the food spectrum with other species.

A recent study conducted on the brook trout *Salvelinus fontinalis* in Italian alpine lakes showed how adults are responsible for selective predation on the large crustacean zooplankton, driving the impact of introduced fish throughout the entire zooplankton community (Tiberti et al., 2014).

In such a situation, avoiding new introductions is of pivotal importance. However, in the EU Regulation 708/2007 on the use of alien species in aquaculture, most of these impacting species are included in Appendix IV, a white list of species whose introduction for aquaculture purposes is allowed.

The new EU Regulation 1143/2014 on invasive alien species establishes a framework for tackling invasive species at the European level, with the aim of protecting biodiversity, ecosystem services and human well-being (Genovesi et al., 2015b). However, only two fishes, *Perccottus glenii* and *Pseudorasbora parva*, are included in the proposed first list of invasive alien species of Union concern that will be banned from Europe and for which management actions should be undertaken. For the other species, at present, just two recommendations on a European code of conduct exist, one on pets and invasive alien species (154/2011) and another on recreational fishing and invasive alien species (170/2014).

In conclusion, our study highlights the importance of alien fishes in Italy, a process that is completely reshuffling and changing the communities, and the depth of the gap between scientific research on their impacts and management actions. Indeed, according to EU Regulation 1143/2014, national lists of invasive species of regional concern to be managed could be implemented. This, however, requires scientific evidence of impacts as well as a risk assessment for every species candidate to be restricted and managed. Nevertheless, as well emphasised by Genovesi et al. (2015b), even if fully implemented, the legislative framework cannot alone address the increasing threat of biological invasions. The best way to cut back on unauthorized introductions (e.g. through the release of unused baits as well as ornamental species) and to increase awareness of the risks posed by invasive alien fishes, is indeed public education that should be intensively promoted together with the adoption of the European codes on pets and recreational fishing at the national level.

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Table 1. Numbers of native, introduced and translocated fish species present with established populations in the Italian basins. ZIc = zoogeographic integrity coefficient. See Figure S1 for a map.

	Native	Introduced	Translocated	Total	Introduced/ Native (%)	ZIc
Po	42	35	0	77	83.3	0.55
Adige	35	22	1	58	62.9	0.60
Piave	32	16	1	49	50.0	0.65
Tagliamento	31	11	0	42	35.5	0.74
Livenza	31	17	0	48	54.8	0.65
Isonzo	26	13	1	40	50.0	0.65
Brenta-Bacchiglione	36	24	1	61	66.7	0.59
Arno	15	21	9	45	140.0	0.33
Tevere	20	25	11	56	125.0	0.36
Liri-Garigliano	15	11	7	33	73.3	0.45
Volturno	11	8	5	24	72.7	0.46
Lemene	25	9	0	34	36.0	0.74
Fissero-Tartaro-Canalbianco	25	20	0	45	80.0	0.56
Magra-Vara	12	2	4	18	16.7	0.67
Reno	23	14	2	39	60.9	0.59
Conca-Marecchia	13	7	0	20	53.8	0.65
Fiora	11	11	5	27	100.0	0.41
Tronto	6	3	3	12	50.0	0.50
Sangro	7	2	3	12	28.6	0.58
Trigno	6	4	2	12	66.7	0.50
Saccione	2	0	0	2	0.0	1.00
Fortore	3	3	1	7	100.0	0.43
Ofanto	7	9	3	19	128.6	0.37
SELE	10	4	2	16	40.0	0.63
Bradano	6	6	4	16	100.0	0.38
Noce	3	5	4	12	166.7	0.25
Sinni	8	5	6	19	62.5	0.42
Friuli Venezia Giulia	19	10	0	29	52.6	0.66

Veneto	31	20	0	51	64.5	0.61
Liguria	7	4	4	15	57.1	0.47
Emilia Romagna	25	17	4	46	68.0	0.54
Toscana	18	21	13	52	116.7	0.35
Marche	19	12	4	35	63.2	0.54
Lazio	13	11	6	30	84.6	0.43
Abruzzo	12	9	7	28	75.0	0.43
Molise	7	7	4	18	100.0	0.39
campania	9	2	0	11	22.2	0.82
Basilicata	6	7	5	18	116.7	0.33
Calabria	8	5	4	17	62.5	0.47
Sicilia	8	6	0	14	75.0	0.57
Sardegna	8	7	0	15	87.5	0.53

Table 2. List of the most invasive established species introduced in Italian basins and their impacts on native species.

Species	Number of basins invaded	100 of the worst global (GISD) ¹	100 of the worst Europe (DAISIE) ²	Threatened species affected in Europe ³
<i>Salmo trutta</i>	39	x		
<i>Cyprinus carpio</i>	34	x		
<i>Oncorhynchus mykiss</i>	31	x		
<i>Carassius carassius</i>	29			
<i>Lepomis gibbosus</i>	28			
<i>Micropterus salmoides</i>	28	x		2
<i>Carassius auratus</i>	26			
<i>Ameiurus melas</i>	24			
<i>Gambusia affinis/holbrooki</i>	24	x*		3
<i>Pseudorasbora parva</i>	20		x	
<i>Rhodeus sericeus</i>	15			
<i>Silurus glanis</i>	14			2
<i>Ictalurus punctatus</i>	12			
<i>Sander lucioperca</i>	11			3
<i>Rutilus rutilus</i>	11			

¹ Present in the IUCN list of “100 of the World's Worst Invasive Alien Species”

² Present in the DAISIE “100 of the Europe's Worst Invasive Alien Species”

³ Number of threatened native species (IUCN Red List CR, EN, VU) that are affected in Europe (from Genovesi et al. 2015a)

* In GISD, *Gambusia affinis* is reported; however, *G. affinis* is closely related to the eastern mosquito fish (*Gambusia kolbrooki*) which was formerly classed as a sub-species. Their morphology, behaviour and impacts are almost identical.

Figure 1. A: Correlation between the original native species richness of each Italian basin and the number of introduced species (filled circles, black regression line) or translocated species (open circles, the regression is not significant). B: Number of Italian basins where single native or introduced species are present.

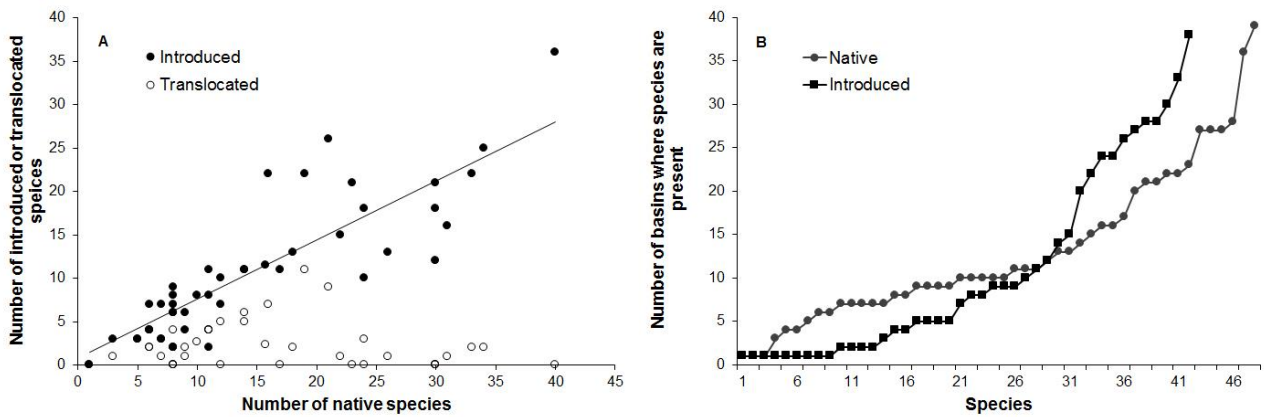


Figure 2. Pairwise comparison of present and past similarities between fish assemblages of basins using the Jaccard index. Past historical assemblages consider only native species, while present assemblages are evaluated including native and introduced species or adding also translocated species. Values over the diagonal line indicated basins that are more similar in the present than in the past, while values below the diagonal line indicated basins that were more similar in the past than in present.

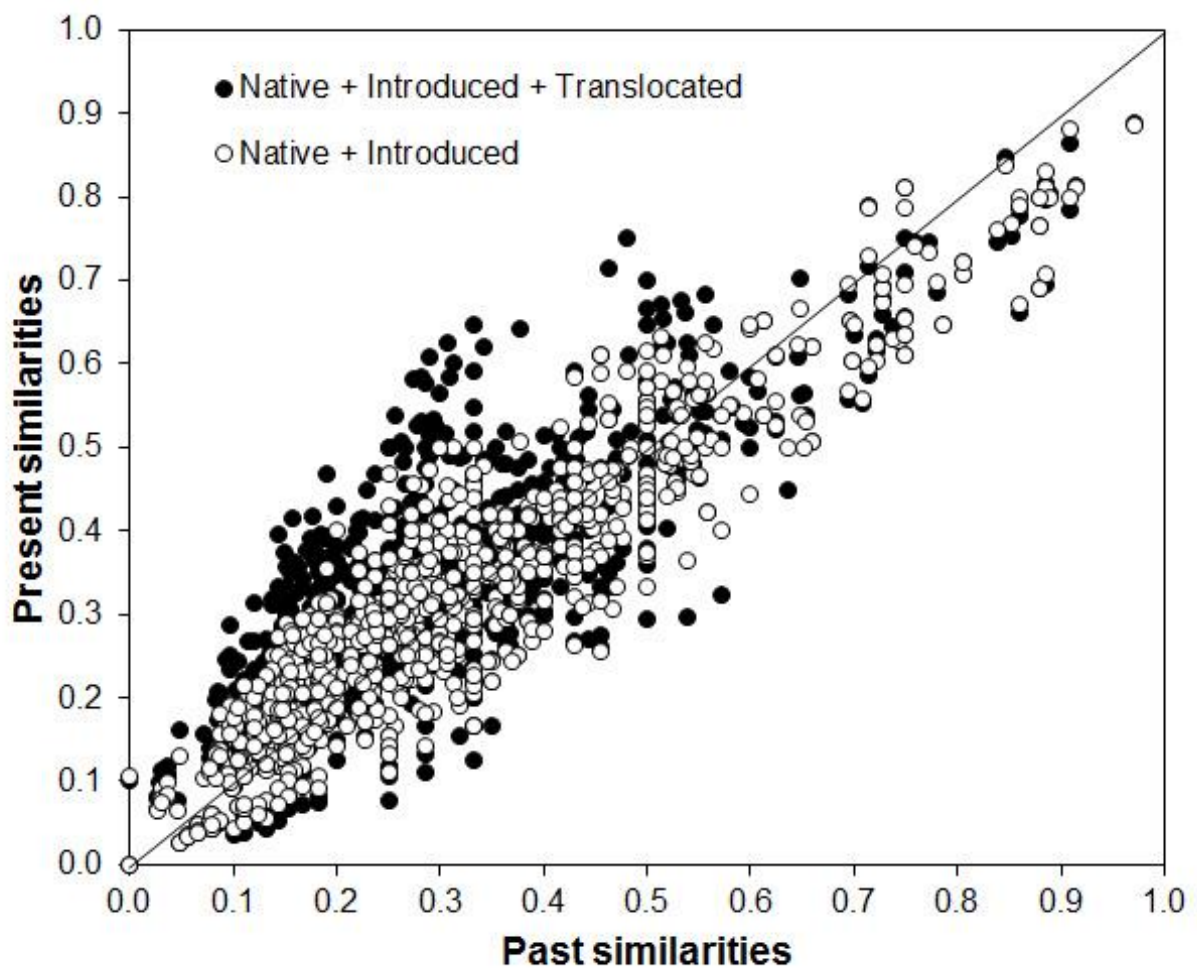


Figure 3. Three clusters of basins identified with the K-means clustering procedure considering only introduced species.

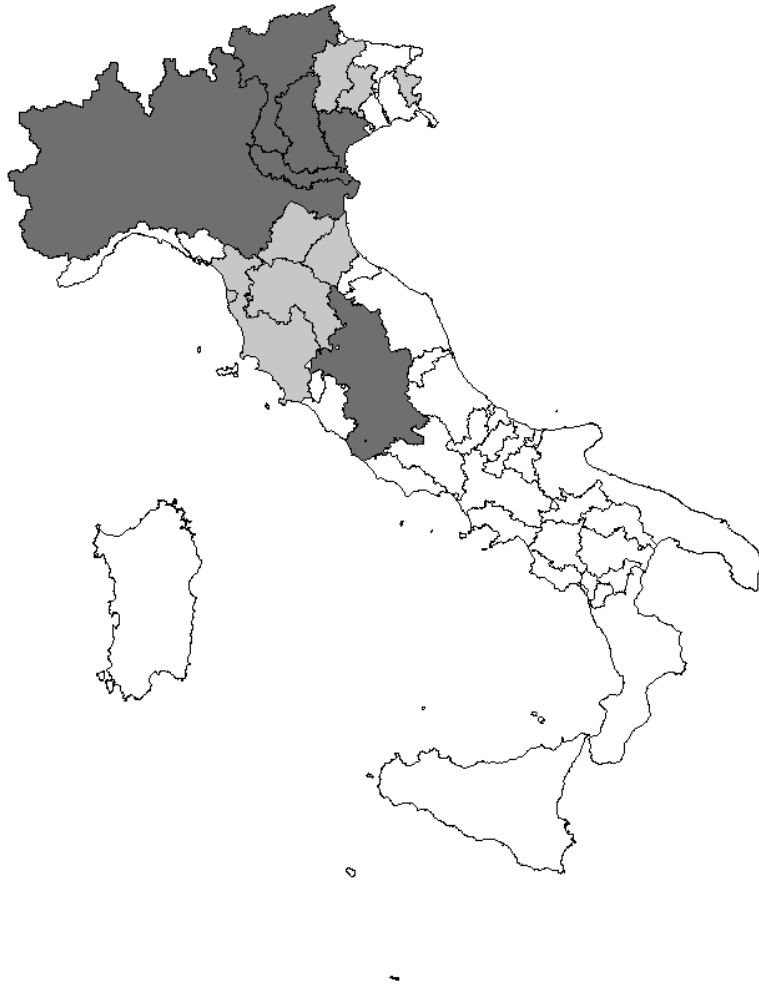


Figure 4. A: Number of basins with similar zoogeographic integrity coefficients (ZIC). B: Mean number of alien fish species introduced in Italy. Bars represent the mean number of species introduced in ten years for a given period. C: Areas of origin of fish species introduced in Italy. D: Cumulative number and trend (dashed line) of alien fish species introduced in Italy. Species without information about the year of first record were excluded.

