

## Article

# Euryhaline Aliens Invading Italian Inland Waters: The Case of the Atlantic Blue Crab *Callinectes sapidus* Rathbun, 1896

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**Abstract:** Alien invasive species represent a major threat in Italian freshwaters. Despite national and European regulations aimed at the control and prevention of new invasive species, the number of allochthonous species is increasing and so is the rate of spread. One of the most widespread invasive animals in coastal areas of the Mediterranean Sea is the blue crab *Callinectes sapidus*, a native of the Western Atlantic from Nova Scotia to Argentina. The species is euryhaline, and besides coastal waters, it is also commonly recorded in upper estuaries and lower reaches of rivers. Considering its fast adaptability and invasiveness, the main purpose of this work is to assess whether its biological plasticity and invasive capability may represent, in addition to marine and coastal habitats, also a potential threat for inland waters. Samples were collected in seven sites distributed between Southern and Central Italy. The new records are herein presented, starting from the original introduction by ballast waters and the actual westward migration in inland waters of Latium and Tuscany. In addition, the threat to biodiversity is also discussed, considering the blue crab aggressiveness toward both vertebrates and invertebrates, which may limit the distribution of native freshwater fish and macrofauna.

**Keywords:** Decapoda; Brachyura; non-native species; aquatic threat; freshwater invasion



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## 1. Introduction

In the last decades, the introduction of non-indigenous species (NIS) in European waters has increased remarkably, both in marine and freshwater ecosystems [1]. There are currently more than 14,000 alien species recorded in Europe (EASIN Catalogue [2], <https://easin.jrc.ec.europa.eu/>, accessed on 30 March 2022), with more than half originating from outside EU territories [2]. Their numbers are rapidly increasing, and in some cases so is their rate of spread [3]. The establishment success of NIS depends on the ecological characteristics of the invaded habitat as well as on the ecological plasticity of the species (with particular reference to reproductive capacity, response to environmental stress, trophic flexibility, and behavioral aspects) [4,5]. Once introduced, NIS have the opportunity to establish self-sustaining populations, acting as sources from which to spread widely, and they become invasive by inflicting environmental and economic harm (invasive alien species, IAS hereafter [6]).

This issue is particularly important in Italian rivers, where invasion by NIS has been listed among one of five main causes threatening freshwater biodiversity [7]. Interestingly, major attention has recently been dedicated to euryhaline taxa that can reach lotic habitats, migrating from transitional waters, due to their ecological and biological characteristics [8]. Climate changes are enhancing this migratory pattern due to persistent upstream shift of the salt wedge.

One of the most widespread invasive species in coastal areas of the Mediterranean Sea is the blue crab *Callinectes sapidus* Rathbun, 1896, belonging to the Portunidae family, a native of the Western Atlantic from Nova Scotia (Canada) to Argentina [9].

*Callinectes sapidus* has recently spread in Mediterranean coasts [10–12] following its introduction by ballast waters [9]. A comprehensive database on its distribution in its native and non-native ranges has recently been published [13], which verified 458 records in the Mediterranean Sea, stating the species can now be considered as “virtually ubiquitous” [13]. Indeed, despite the blue crab occurring mainly in the northern and eastern Mediterranean, the species is also expanding southward along the African coasts [13]. Therefore, the species is rapidly expanding in European waters and is currently considered invasive, as it represents a real threat to biodiversity at the local level as a predator and competitor of other benthic species [14,15]. *Callinectes sapidus* may play a major structuring and functional role in benthic systems, being a keystone consumer and feeding on fish, invertebrates, detritus, macrophytes, and algae [16]. It is therefore clear how the blue crab inserts itself on several levels of the trophic network and has a direct and indirect impact on benthic communities, both through direct predation and through the bioturbation of the sediment [17–19].

It is noteworthy to mention that the species is euryhaline, and besides coastal waters, it is also commonly recorded in upper estuaries, lower reaches of rivers, and even under fully freshwater conditions in coastal lakes [13,20]. The occurrence of the species in freshwaters is not exceptional and is related to its biological cycle. Specifically, mating occurs in the upper estuaries, after which females migrate to areas having higher salinities for spawning. After hatching, juveniles gradually migrate into shallower, less-saline upper estuaries and rivers, where they grow and mature until one to two years of age, before returning to both lower estuaries and sea coastal habitats [21].

Considering this scenario, the main purposes of this study are to (i) update the data available on the records of *Callinectes sapidus* in Italian inland waters and (ii) discuss whether its biological plasticity and invasive capability may represent, besides marine and coastal habitats, also a potential threat for inland waters.

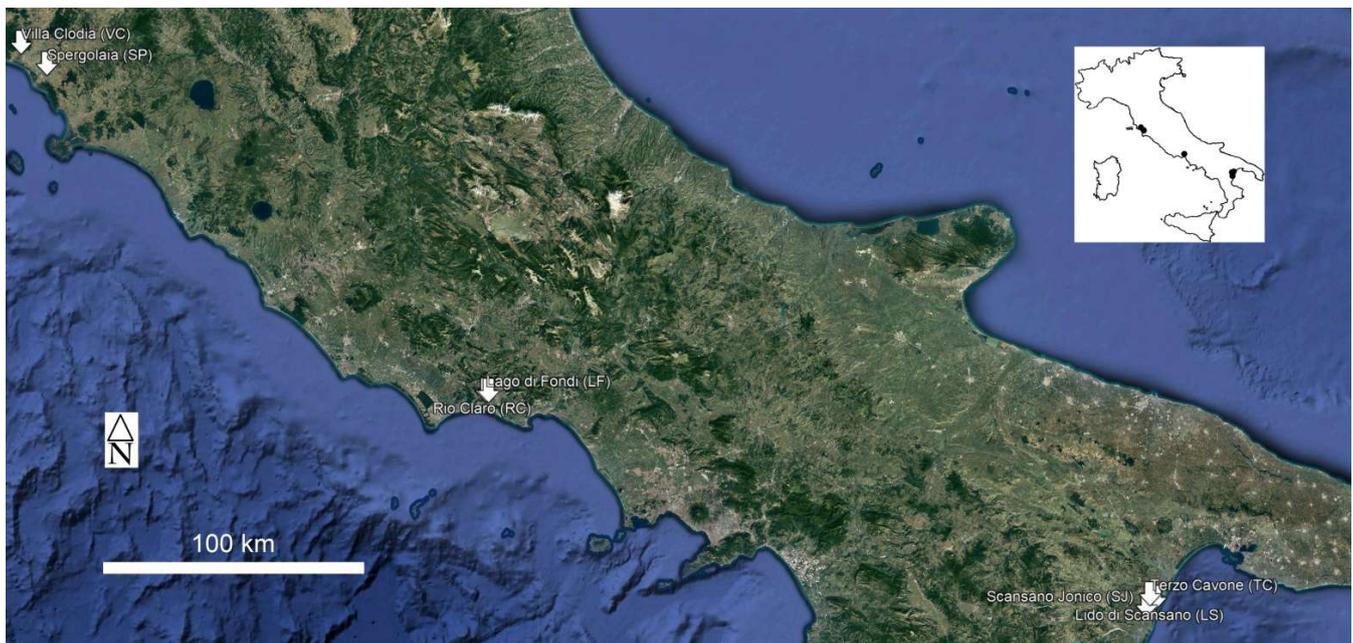
## 2. Materials and Methods

In this study we report our findings on specimens of the blue crab in Italian inland waters during other local environmental monitoring sessions, namely monitoring programs of the fish fauna and benthic macroinvertebrates for conservation purposes. In particular, samplings were carried out to define major environmental and ecological threats affecting the population dynamics of threatened fish species. Samplings were carried out between February 2020 and November 2021.

Seven different sites were investigated in three regions (Table 1 and Figure 1): Villa Clodia (VC—5.5 km away from the sea) and Spergolaia (SP—9.2 km) in Tuscany; Lago di Fondi (LF—3.7 km) and Rio Claro (RC—4 km) in Latium; Terzo Cavone (TC—7.5 km), Lido di Scansano (LS—2.5 km), and Scansano Jonico (SJ—4 km) in Basilicata. Crabs were collected using fyke nets with the following characteristics: 14 m in length, 5 rigid rings on each side (bilateral chambers) with a diameter decreasing from 55 to 30 cm, and mesh size descending from 1 to 0.5 cm in the terminal part of the net. The nets were placed between 0.5 to 1.5 m in depth for approximately 12 h overnight.

**Table 1.** Sites (from north to south) of the blue crab occurrence within the study area: distance from the sea is also reported.

| Region     | Province | Site             | Acronym | N            | E            | Distance from the Sea | Degree of Preservation                               |
|------------|----------|------------------|---------|--------------|--------------|-----------------------|--|
| Tuscany    | Grosseto | Villa Clodia     | VC      | 42°47'03.91" | 10°56'26.22" | 5.5 km                | Natural Reserve (Site of Community Importance)       |
|            |          | Spergolaia       | SP      | 42°41'41.20" | 11°04'54.50" | 9.2 km                | Maremma Natural Park                                 |
| Latium     | Latina   | Lago di Fondi    | LF      | 41°18'45.66" | 13°21'47.05" | 3.7 km                | Regional Natural Park (Site of Community Importance) |
|            |          | Rio Claro        | RC      | 41°18'34.84" | 13°22'39.90" | 4 km                  |  |
| Basilicata | Matera   | Terzo Cavone     | TC      | 40°17'44.80" | 16.43'54.08" | 7.5 km                | Nature 2000 network                                  |
|            |          | Lido di Scansano | LS      | 40°16'44.87" | 16.45'37.38" | 2.5 km                | Nature 2000 network                                  |
|            |          | Scansano Jonico  | SJ      | 45°15'07.49" | 16.42'31.24" | 4 Km                  | Nature 2000 network                                  |



**Figure 1.** Locations of sampling sites and the blue crab occurrence in Italian freshwaters to date.

We recorded sex (on the basis of abdomen shape), weight (by using a field balance), and carapace width (by a vernier caliper) of each captured individual, after having georeferenced each site (by an outdoor handheld GPS system) and having surveyed temperature, pH, conductivity, and salinity (by immersion probes). A checklist of sympatric fish species and benthic macrofauna was also recorded where specifically sampled/observed.

In addition, we used the information provided in a recent database of *C. sapidus* records in native and invaded ranges [13] to contextualize our findings and to emphasize the ability of the species to colonize freshwater environments. To this end, the coordinates of non-marine occurrences were extracted from the database and used to calculate their distance from the coastline (resolution = 10 m) using the R package rgeos (gDistance function).

### 3. Results

In total, we captured six males of *C. sapidus*, one per site, with a carapace width ranging from 14.6 to 24.1 mm and a weight from 134 to 253 g (Table 2). All individuals were

observed in coastal inland waters showing a salinity within values of typical freshwaters (Table 3). In the case of the Spergolaia sampling station, many more samples could have been caught due to a consistent demographic density of the species distributed over a wide area. However, a numeric evaluation of the population was not assessed, since it was not a specific focus of the project.

**Table 2.** Carapace width (cm) and weight (g) of the blue crab males observed in each sampled site (from northern to southern). Site acronyms (in alphabetical order): LF = Lago di Fondi; LS = Lido di Scansano; RC = Rio Claro; SJ = Scansano Jonico; SP = Spergolaia; TC = Terzo Cavone; VC = Villa Clodia.

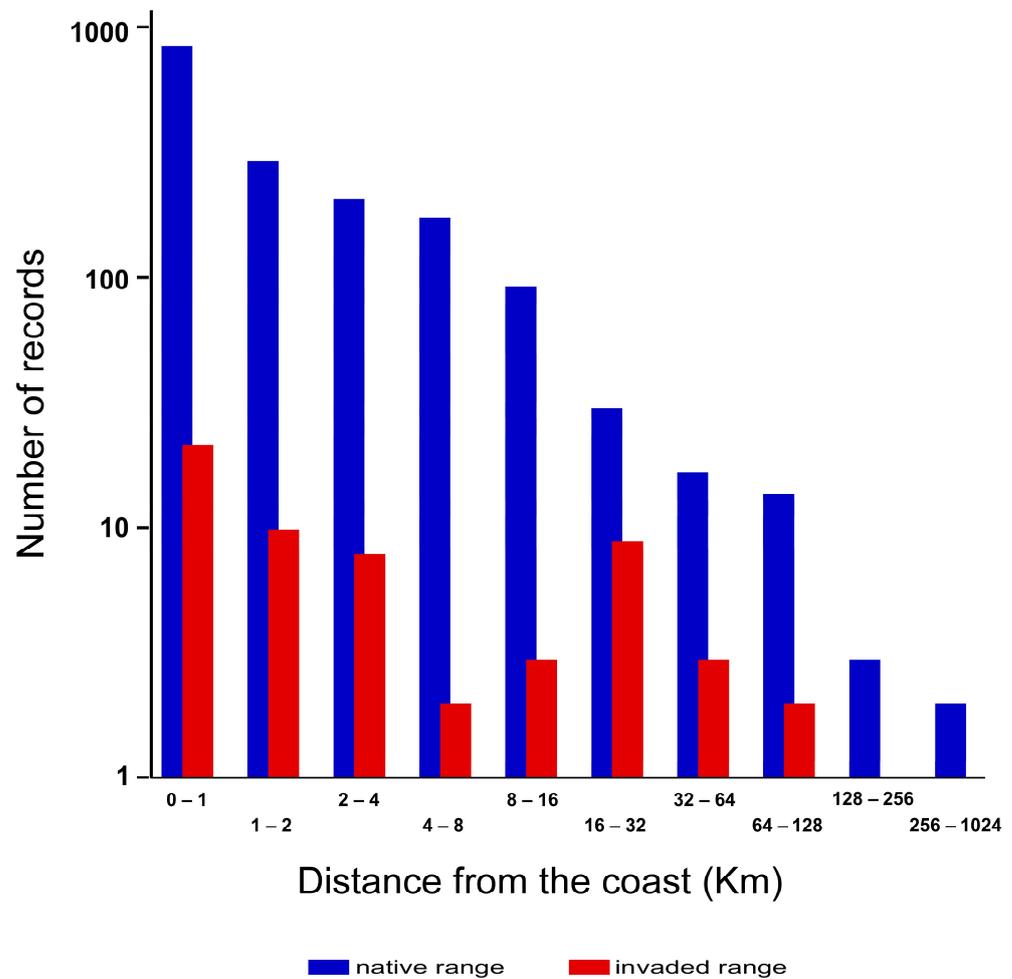
| Region     | Province | Site | Carapace Width (cm) | Weight (g) |
|------------|----------|------|---------------------|------------|
| Tuscany    | Grosseto | VC   | 15.0                | 160        |
|            |          | SP   | 14.7                | 140        |
| Latium     | Latina   | LF   | 14.6                | 134        |
|            |          | RC   | 21.4                | 208        |
| Basilicata | Matera   | TC   | 24.1                | 253        |
|            |          | LS   | 18.4                | 184        |
|            |          | SJ   | 22.8                | 199        |

**Table 3.** Environmental and ecological descriptors surveyed during the blue crab findings in each sampled site. Site acronyms (in alphabetical order): LF = Lago di Fondi; LS = Lido di Scansano; RC = Rio Claro; SJ = Scansano Jonico; SP = Spergolaia; TC = Terzo Cavone; VC = Villa Clodia. NA = Not available. \* Freshwater life stage.

| Region     | Province | Site | Temperature °C | pH  | Conductivity (µScm) | Salinity (‰) | Freshwater Fish Fauna/Benthic Macrofauna (Where Available)   |
|------------|----------|------|----------------|-----|---------------------|--------------|--|
| Tuscany    | Grosseto | VC   | 12.5           | 8.8 | 530                 | 3.8          | <i>Anguilla anguilla</i> *, <i>Carassius carassius</i> , <i>Cyprinus carpio</i> , <i>Scardinius erythrophthalmus</i>   |
|            |          | SP   | ND             | ND  | ND                  | ND           | <i>Alburnus alburnus</i> , <i>Alosa fallax</i> *, <i>Barbus graellsii</i> , <i>Carassius carassius</i> , <i>Cyprinus carpio</i> , <i>Ictalurus punctatus</i> , <i>Squalius squalus</i> |
| Latium     | Latina   | LF   | 16.4           | 8.0 | 1060                | 7.1          |  |
|            |          | RC   | 18.8           | 8.2 | 884                 | 6.3          | <i>Anodonta cygnea</i>   |
| Basilicata | Matera   | TC   | 15.7           | 7.9 | 701                 | 5.0          | NA   |
|            |          | LS   | 17.6           | 8.1 | 654                 | 4.7          | <i>Anodonta cygnea</i>   |
|            |          | SJ   | 13.3           | 8.4 | 589                 | 4.2          | NA   |

Environmental descriptors (Table 3) witnessed low salinity concentrations, with the latter being mostly referable to freshwater affected by saltwater encroachment. Despite measured values of 3.8‰ in station Villa Clodia and Spergolaia (salinity unknown), all sympatric fish species were strict freshwater taxa, such as carps, rudds, spanish barbels, chubs, channel catfish, bleaks, and crucian carps, together with diadromous eels, shads, mullets, sand smelts, Mediterranean tooth carps, and seabasses; in other sites, such as Rio Claro and Lido di Scansano, the sampled blue crab specimens were observed in sympatry with freshwater mussels. Scientific names of freshwater species are reported in Table 3.

The analysis performed on non-marine occurrences from [13] clearly confirmed the high euryhaline adaptability of *C. sapidus* (Figure 2). In total, 1820 records were extracted; in both native (1761 records) and invaded ranges (59 records), the species was reported to occur in inland waters at distances >64 km from the coastline (>250 km in native areas). Noticeably, the distance data were calculated “as the crow flies” without considering the hydrographic network; thus, they necessarily provide a conservative estimation of the actual ability of the blue crab to colonize freshwater environments.



**Figure 2.** Occurrence of *Callinectes sapidus* under brackish and freshwater conditions in native and invaded habitats. The histogram refers to non-marine occurrences provided in [13]. For the sake of clarity, distances are binned over a Log2 scale, while the number of records in each distance bin is reported using a Log10 scale.

#### 4. Discussion

Although crabs mainly inhabit marine waters, some living clades have colonized freshwater habitats, experiencing the life cycle therein [22]. In this study, we described the occurrence of an euryhaline species, *Callinectes sapidus*, within freshwater habitats. The invasion of the coastal marine environments of the Mediterranean Sea by *C. sapidus* is mainly due to its high ecological tolerance and trophic flexibility, its swimming ability, and the typical traits of its life cycle, such as high fecundity [23].

In light of our results, it is difficult to predict the degree of damage that might arise from the spread in freshwater habitats along the Tyrrhenian coast. Therefore, the tolerance of *C. sapidus* to inland waters in the secondary range of distribution should be further investigated as a fundamental management activity for the conservation of aquatic habitats, as well as to have a comprehensive assessment of the actual ecology of the species in invaded habitats.

Its impact on the local fauna and flora in the monitored freshwater environments is not yet evident, but considering the damage that *C. sapidus* can cause in the different areas where it was introduced, the possibility that this species may affect fragile ecosystems such as those of inland waters is concrete. Indeed, freshwater habitats in Europe are already threatened by NIS, with particular reference to the impact on native fish fauna, which is vulnerable to the introduction and spread of freshwater alien species [24]; the occurrence

of additional invaders coming from marine and coastal waters, such as the euryhaline *C. sapidus*, represents an additional risk for their conservation.

The new records of blue crab presented in this work can be discussed, considering the current distribution of the species along the Italian coasts. Records of *C. sapidus* in freshwaters of the Basilicata region herein collected occurred along the Ionian coast, in accordance with recent data on species distribution, showing that the blue crab in Italian coasts is more abundant in the Adriatic/Ionian area [15,25,26], with a “hotspot” of records from the Apulia region, especially around the Gargano Peninsula and in the Gulf of Taranto [25]. It is noteworthy that in the eastern Ionian Sea, the number of records increased significantly in recent years [10].

Although the occurrence of *C. sapidus* in the Tyrrhenian basin is lower, the species is currently expanding towards the western Mediterranean Sea [11–13,15,25,27,28], with confirmed records also in the Ligurian Sea [29]. Moreover, recent data obtained by online questionnaires for recreational fishermen confirmed the species occurrence in the Central and Southern sectors of the Tyrrhenian Sea, such as the Latium region, Naples, Tyrrhenian Calabria, and Sicily [25].

Records herein describing the freshwater habitats of Latium and Tuscany may be also related to the occurrence of two important harbors of the Central Tyrrhenian Sea: Civitavecchia (Latium) and Livorno (Tuscany). Ballast waters are acknowledged as the primal vector of blue crab introduction [9]; therefore, ports are particularly important as hot spots of introduction [23]. Even if traditional port surveys may fail to detect the blue crab [30], the species may also occur and can be detected with different, complementary methods, such as LEK (Local Ecological Knowledge), as previously observed in the Adriatic Sea [30].

Within the Mediterranean basin, the blue crab has been recently reported in several freshwater habitats, including the Ichkeul Lake in Tunisia [28], wetlands of the Moulouya mouth and Oued Kert mouth (Morocco) [31], the Tirso and Fenosu rivers in Western Sardinia [27], the mouth of the Basento River (Italy) [32], and Lake Volvi in Northern Greece [33].

As already stated by [10], the occurrence of *C. sapidus* in freshwaters itself is not unusual (Figure 2): blue crab can exploit different salinity regimes for its life cycle, and low-salinity environments play a role in mating and nursery habitats [21]. Recent data collected in its native range in North America showed that blue crabs were distributed along a salinity gradient, depending on their sex and ontogenetic life stage [21,34]. Upriver oligohaline waters were preferentially inhabited by male and juvenile crabs, whereas meso- and polyhaline portions of the rivers were selected mainly by adult females [34].

This pattern of distribution may explain why in our study only adult males were collected in portions of running waters characterized by freshwater salinity levels. The same results were also obtained by [27], who collected two male specimens in freshwater habitats of Sardinia, and by [31] in Morocco, where adult males were predominantly caught at river mouths. Unfortunately, in the records obtained by [28] in Tunisia, the gender of collected specimens was not reported.

Although for invaded ranges the majority of data regards marine and estuarine environments (see [13]), the blue crab may also represent a dangerous threat to biodiversity for freshwater habitats (Figure 2). Due to its wide trophic niche, the blue crab can predate both vertebrates (i.e., fish fauna) and invertebrate species (i.e., bivalves) of conservation and commercial interest and also affect native riparian and aquatic vegetation. In our work, the blue crab posed a major concern for its sympatric recurrence with the European eel population of the Diaccia Botrona (see Supplementary Material displaying a video of eel predation by a blue crab inside a fishing cage). On the other hand, samples from Spergolaia were collected in one of the last remaining spawning areas of the Mediterranean shad *Alosa fallax*. It should be noted that the two native species are protected under the Regulation 1100/2007/EU and the Directive 43/1992/CEE, respectively. Similarly,

predation on freshwater bivalves and herpetofauna, besides threatened fish species, has recently been described in the Iberian Peninsula [8,35,36].

Moreover, many freshwater ecosystems are nowadays suffering from the colonization and spread of multiple co-occurrent alien species [37]. In the specific case of euryhaline invaders, the co-occurrence of such benthic decapods as *C. sapidus* and *Procambarus clarkii* (Girard, 1852) at a specific local scale can enhance the impact on local biodiversity. Indeed, although they are following two different routes, *C. sapidus* (from marine to inland waters) and *P. clarkii* (from inland to marine waters) are deeply altering coastal waterbodies, occurring more and more frequently in both brackish and freshwater habitats [38].

This is why it is necessary to continue monitoring activities on the spread of *C. sapidus* both along marine coastal habitats and in inland waters. Taking into account the present state of the art in Central Italy, more data on *C. sapidus* in freshwaters are needed, considering the richness of endemic species in this part of the country.

Involving local communities and tourists in the monitoring of “unusual” records is fundamental for the early detection of the blue crab [30]. In fact, recent approaches based on citizen science and LEK (Local Ecological Knowledge) may be particularly useful to update data on distribution and abundance of the species in freshwater habitats, as successfully demonstrated for marine and estuarine habitats [25,26,29,30,39]. The cultural dissemination is fundamental, considering that although it is possible to propose the blue crab exploitation as a fishery resource to contain its demographic expansion, it must be remembered that *C. sapidus* may bioaccumulate contaminants, such as organic compounds, metals and metalloids, or plastics [40]. Like other macrofauna species, it is particularly exposed to contaminants occurring in sediments. This scenario could therefore represent a risk to the health of consumers.

## 5. Conclusions

Although mainly descriptive and limited to a low number of specimens, this work adds further knowledge on the important topic of highly invasive species in the Mediterranean area, with reference to euryhaline species. Most literature data on blue crab *C. sapidus* in Europe and in the Mediterranean basin is concentrated on records, distribution, and ecology of this invasive species in marine and estuarine waters.

However, the blue crab can easily move from river mouths up to freshwater habitats, where its impact can be valuable, although poorly investigated up to now, particularly on endemic species. Therefore, efforts should be made to monitor freshwater ecosystems to eventually detect blue crab occurrence and to better understand its population dynamics and possible impacts on local freshwater biodiversity.

**Supplementary Materials:** The following is available online at <https://www.mdpi.com/article/10.3390/app12094666/s1>, Video: Blue crab grabbing European eel.

**Author Contributions:** Conceptualization, M.S., S.C., G.M., F.N.M.; methodology, A.V., P.M.R.; formal analysis, M.S., A.V., P.M.R.; investigation, M.S., A.V., P.M.R., F.N.M.; resources, M.S., A.V., P.M.R., F.N.M.; data curation, M.S., F.N.M.; writing—original draft preparation, M.S., S.C., G.M., F.N.M.; writing—review and editing, all authors; visualization, M.S., S.C., G.M.; supervision, F.N.M.; project administration, F.N.M. All authors have read and agreed to the published version of the manuscript.

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