

Seasonal abundance and trophic position of the Atlantic blue crab *Callinectes sapidus* Rathbun 1896 in a Mediterranean coastal habitat

Leonardo Carrozzo · Luigi Potenza ·
Pasquale Carlino · Maria Letizia Costantini ·
Loreto Rossi · Giorgio Mancinelli

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Abstract The blue crab *Callinectes sapidus*, native to the western coasts of the Atlantic Ocean, has been introduced in Mediterranean waters where it is currently considered an invasive species. Here, we verified the occurrence of an established population of *C. sapidus* in the Torre Colimena basin, a Mediterranean habitat located in SE Italy, and provided a first assessment of its functional role in a Mediterranean coastal ecosystem. Crab traps were used to estimate the species abundance at a seasonal frequency; in addition, its trophic position was estimated in summer using nitrogen stable isotopes. Estimations were performed using both a generally adopted value of 3.4 ‰ for the nitrogen trophic level fractionation factor $\Delta^{15}\text{N}$, and species-specific estimations obtained from the literature. *C. sapidus* was sampled throughout the year, reaching peak abundances in summer. This result, taken together with the observed patterns of variation in the average body size and sex ratio of captured specimens, provided evidence of a fully established population in the Torre Colimena basin. The estimated trophic position of the crab was consistent with the literature information on western Atlantic populations, and resulted, irrespective of the fractionation factor used, significantly higher than those characterizing an autochthonous brachyuran (*Pachygrapsus marmoratus*) and a fish predator (*Sparus aurata*). The present study

provided novel information on the occurrence and potential functional impact of this non-indigenous species in Mediterranean coastal habitats, highlighting current knowledge lacunae and identifying future research lines on marine alien species.

Keywords Invasive species · *Callinectes sapidus* · Trophic level · Stable isotopes · Ecological impact

1 Introduction

In the Mediterranean Sea, alien species introduction is accelerating at an unprecedented rate, causing remarkable effects on species composition and biodiversity of native marine communities, and ultimately triggering ecosystem-wide changes such as trophic cascades and productivity regime shifts (Boudouresque et al. 2005; Galil 2009; Zenetos 2010; Coll et al. 2010). The example provided by crustaceans is emblematic of this threat: between 2002 and 2008, the CIESM Atlas of Exotic Species listed 70 crustacean species introduced in the Mediterranean; in 2011, an updated review provided by Galil (2011) listed 106 species. To date, a number of new alien crustacean species have been recorded (e.g., *Paracaprella pusilla* Mayer, 1890; Ros and Guerra-Garcia 2012; *Scalpellum scalpellum* Linnaeus, 1767; Topaloğlu and Gönülal 2012).

Among the species listed in Galil (2011), the blue crab *Callinectes sapidus* Rathbun, 1896 (Brachyura: Portunidae) was actually ubiquitous, occurring along the coasts of 11 out of the 12 Mediterranean countries considered. The species is native of the western coasts of the Atlantic Ocean, from New England to Uruguay and was probably introduced in Europe by ballast waters (Galil 2000). The

L. Carrozzo · L. Potenza · G. Mancinelli (✉)
Department of Biological and Environmental Sciences and
Technologies, University of Salento, Centro Ecotekne,
sp Lecce-Monteroni, 73100 Lecce, Italy
e-mail: giorgio.mancinelli@unisalento.it

P. Carlino · M. L. Costantini · L. Rossi
Department of Environmental Biology, Sapienza University of
Rome, via dei Sardi 70, 00185 Rome, Italy

species was originally described along the Atlantic French coast (Bouvier 1901), while in the Mediterranean Sea it was first reported in Italy (Giordani Soika 1951) and in Israel (Holthuis and Gottlieb 1955). To date, the blue crab is considered an invasive alien species (Zenetos et al. 2005; IAS hereafter), even though the majority of the reports on the occurrence of the species in Mediterranean and, in general, European waters have been based on numerically limited, episodic catches (Dulčić et al. 2010; Nehring 2011 and literature cited; but see Tureli Bilen et al. 2011).

In the present study, we analyzed the seasonal occurrence of *Callinectes sapidus* in the Torre Colimena coastal basin (SE Italy) over a whole year, allowing an assessment of the patterns of abundance, size, and sex ratio characterizing this IAS in a Mediterranean coastal habitat. In addition, we calculated the blue crab trophic position and compared it with those of two autochthonous brachyuran and fish species. The crucial functional role of the blue crab within native coastal ecosystems has long been acknowledged (e.g., Baird and Ulanowicz 1989; Dittel et al. 2000); conversely, in invaded Mediterranean habitats it is to date virtually unexplored. Here, *C. sapidus* trophic position was estimated using nitrogen stable isotopes, a method that has been proven successful for freshwater and, more recently, marine food webs, providing useful information on mechanisms regulating the establishment success and ecological impact of invasive species (Vander Zanden et al. 1999; Gido and Franssen 2007; Layman and Allgeier 2011; Muñoz et al. 2011).

In both terrestrial and aquatic food webs, feeding relationships among species from different trophic levels are scrutinized assuming that a stepwise enrichment occurs in the heavier isotope ^{15}N of nitrogen with each trophic level, and that this enrichment is constant irrespective of the animal's biology and feeding behavior (i.e., 3.4 ‰: Minagawa and Wada 1984; Post 2002). Yet, the magnitude of this per trophic-step isotope fractionation (generally indicated as $\Delta^{15}\text{N}$) has been recognized to be influenced by a number of factors including taxonomy, body size, excretory mechanisms and feeding rates (Mill et al. 2007 and literature cited), and the generalized, unquestioned use of a $\Delta^{15}\text{N}$ of 3.4 ‰ has been criticized (McCutchan et al. 2003; Vanderklift and Ponsard 2003b; Caut et al. 2009; Bond and Diamond 2011; see also Mancinelli 2012a). Here, the trophic position of the blue crab was calculated and compared to those characterizing a native brachyuran (i.e., *Pachygrapsus marmoratus*) and predatory fish (*Sparus aurata*) using both a generally adopted value of 3.4 ‰ for the nitrogen trophic level fractionation $\Delta^{15}\text{N}$, and species-specific estimations obtained from the literature.

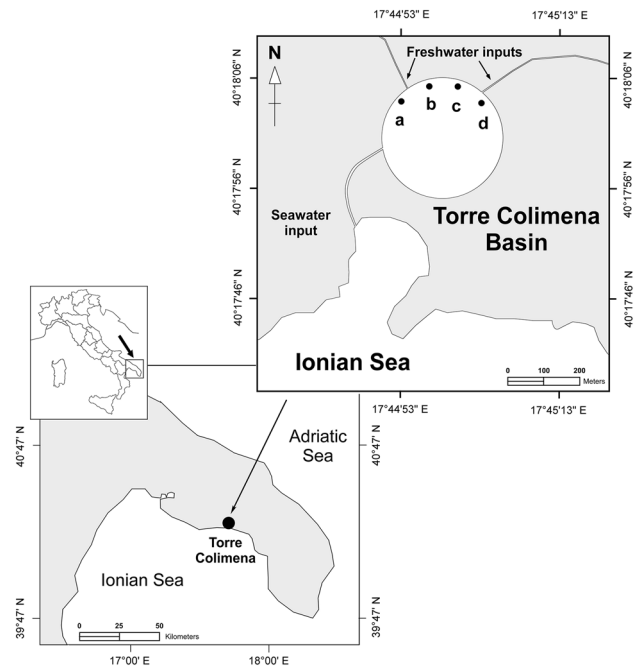


Fig. 1 Map of Torre Colimena coastal basin; sampling stations used during the study period are indicated by black circles and consecutive letters

2 Materials and methods

2.1 Study site

The study was carried out in the Torre Colimena basin, located along the Ionian coasts of the Salento peninsula ($40^{\circ}17'59''\text{N}$; $17^{\circ}44'57''\text{E}$). The basin is a shallow (1.2 m average depth) water body with a surface area of 0.1 km² and about 0.35 km in diameter, created during reclamation activities carried out in the area in the '30 s, when several retrodunal astatic ponds and swamps characterizing the Ionian coasts were regulated (see Mancinelli et al. 2013a, b; Longo and Mancinelli 2014 for additional information on the basin). It receives freshwater inputs from two drainage ditches, while a channel connects the basin with the Ionian Sea (Fig. 1). The seagrass *Cymodocea nodosa* (Ucria) Ascherson characterizes the soft bottom of the basin, while other seagrasses of the genera *Zostera* and *Ruppia* and macroalgae of the genera *Caulerpa* and *Chondria* are locally abundant. The benthic macrofauna is a brackish-water assemblage typical of other lagoons and coastal habitats of the area (e.g., Mancinelli and Rossi 2001; Mancinelli et al. 2005, 2007, 2009; Alemanno et al. 2007a, b; Potenza and Mancinelli 2010; Mancinelli 2010, 2012b; Vignes et al. 2012). Among primary consumers, amphipods, isopods and gastropods dominate the epifaunal assemblage associated to seagrasses and macroalgal beds. Autochthonous brachyurans include *Pachygrapsus*

marmoratus Fabricius and *Carcinus mediterraneus* Czer-niavsky, while representatives of the genera *Sparus*, *Liza* and *Gobius* are the most common fish predators reported in the basin (Mancinelli et al. 2013a).

2.2 Sampling procedures and laboratory analyses

Crabs were sampled with modified crab traps (60 × 60 × 60 cm) of the type described by Sturdivant and Clark (2011). In brief, each trap was constructed of vinyl-coated 2 × 2 cm mesh wire with an upper and lower chamber. A 15 cm wide entrance was located on each of the four sides of the lower chamber, and a conical bait well was situated in the centre. The two chambers were separated by a wire-mesh panel, raised in the middle to form an inverted V. There were two openings along the apex of the V leading into the upper chamber. In December 2011, four sampling stations were located along the northern banks of the basin at a distance of ~100 m from each other. One crab trap was located at each station at a depth of 70–100 cm and baited with fish carcasses. Traps were deployed between 6.00 and 7.00 p.m. and retrieved the day after between 9.00 and 10.00 a.m. Crabs were harvested and transferred to the laboratory in refrigerated containers. Sampling operations were repeated adopting an identical procedure in spring (March), summer (June), and autumn (September).

At each sampling occasion, before trap retrieval bottom water temperature and salinity were measured in triplicate with a YSI 556 meter (YSI Inc., Yellow Springs, OH). Collected crabs were identified, enumerated and sexed based on the shape of the abdomen apron. Consequently, for each specimen, the carapace width (CW) was measured as the distance (in mm) between the two outermost lateral spines and total wet weight (WW) was determined to the nearest 0.01 g.

2.3 Stable isotope analysis and trophic position estimation

Primary consumers were collected from each of the four sampling stations 3 weeks before crab trap deployment in June 2012 using a hand net and by inspecting submerged rocks or litter accumulations. The species included in the analysis (see Sect. 3) were chosen as they dominate the grazer and filter-feeder assemblage (e.g., Longo and Mancinelli 2014; Mancinelli, unpublished data) and are likely to constitute a primary component of the diet of benthivorous vertebrate and invertebrate species. The time lag between resources and crabs collection—i.e., 21 days—was decided to take into consideration short-term temporal variations in resource isotopic signatures and crab tissue-specific turnover rates of nitrogen isotopes (e.g.,

Suring and Wing 2009). With the only exception of the crab *Pachygrapsus marmoratus*, of which three individuals were captured by hand-held nets at station b (Fig. 1), at least four specimens per taxon per station were collected. In addition, three living specimens of gilthead sea bream *Sparus aurata* (142 ± 34 mm total length, mean ± SD) were captured by local fishermen in the basin by fyke nets and analyzed. The gilthead sea bream is known to prey actively on bivalves, crabs and other benthic invertebrates (Pita et al. 2002), and is largely dominant over other predatory fish species in lagoons and other coastal habitats of the Salento peninsula during summer months (e.g., Lumare et al. 2009).

In the laboratory, invertebrates were inspected for contaminants; shells were removed from both gastropod and mollusk samples. Portions of the right claw muscle were removed with a scalpel from four male blue crabs chosen at random among those captured in traps and from the three *P. marmoratus* specimens. A portion of dorsal muscle tissue was removed from each fish specimen. All samples were stored in falcon tubes and freeze-dried for 24 h. Subsequently, samples were ground to a fine powder using a ball mill and analyzed using an Elementar Vario Micro-Cube elemental analyzer (Elementar Analysensysteme GmbH, Germany) coupled with an IsoPrime100 isotope ratio mass spectrometer (Isoprime Ltd., Cheadle Hulme, UK).

The isotopic signature of each sample was expressed in delta notation (‰ deviation from atmospheric nitrogen used as a standard material) $\delta^{15}\text{N} = [(R_{\text{Sample}}/R_{\text{Standard}}) - 1] \times 1,000$, where $R = {}^{15}\text{N}/{}^{14}\text{N}$. The standard deviation of $\delta^{15}\text{N}$ replicate analyses was 0.23 ‰.

The trophic position (TP hereafter) of *Callinectes sapidus*, *Pachygrapsus marmoratus* and *Sparus aurata* was calculated as

$$\text{TP} = \left[\frac{\delta^{15}\text{N}_{\text{Cons}} - \delta^{15}\text{N}_{\text{Base}}}{\Delta^{15}\text{N}} \right] + \lambda$$

The formula is a generalization of that presented in Jepsen and Winemiller (2002), where $\delta^{15}\text{N}_{\text{Cons}}$ is the nitrogen isotopic signature of the consumer species under analysis, $\Delta^{15}\text{N}$ is the trophic level fractionation of $\delta^{15}\text{N}$, while $\delta^{15}\text{N}_{\text{Base}}$ and λ are the nitrogen isotopic signature and the trophic level of the baseline indicator, respectively. $\delta^{15}\text{N}_{\text{Base}}$ and the associated standard deviation SD_{Base} was calculated using the mean isotopic signature of primary consumers taxa (i.e., dominant invertebrate grazers-detritivores and filter feeders; Mancinelli et al. 2013c); accordingly, a $\lambda = 2$ was adopted.

To consider the effect of inter-specific variability in the nitrogen signatures of baseline taxa (see Sect. 3), a parametric bootstrap procedure was used (Vander Zanden and

Table 1 Fractionation factors ($\Delta^{15}\text{N}$) obtained in the literature search for *Callinectes sapidus* and *Sparus aurata* feeding on different resources under controlled laboratory conditions. Data on *Portunus pelagicus* are also reported. They were averaged with those specific of *C. sapidus* to estimate the grand mean $\Delta^{15}\text{N}$ value for brachyuran crustaceans used to calculate *Pachygrapsus marmoratus* trophic position (see Sect. 2 section)

Taxon	References	Food	$\Delta^{15}\text{N}$
<i>Callinectes sapidus</i>	Fantle et al. 1999	Meiofauna	3.2
		<i>Spartina detritus</i>	2.2
		<i>Uca pugnax</i>	0.85
		<i>Littorina littorea</i>	0.7
		Zooplankton	0.1
	Dittel et al. 2000	<i>Artemia</i> sp.	1.5
		<i>Littoraria irrorata</i>	0.8
		<i>Uca pugnax</i>	0.9
		Zooplankton	0.1
<i>Portunus pelagicus</i>	Møller et al. 2008	Commercial pellets	1.6
<i>Sparus aurata</i>	Serrano et al. 2008 ^a	Fish feed	2.7
			2.6
			2.9
			3.6
			4.4
			4.1
	Martin-Perez et al. 2013	Fish feed type 1	4.1
		Fish feed type 2	4
		Fish feed type 3	3.8
		Fish feed type 4	3.7
		Fish feed type 5	3.5
		Fish feed type 6	3.6

^a The different $\Delta^{15}\text{N}$ values refer to different sample collections

Fetzer 2007; Mancinelli et al. 2013c). In brief, for each crab or fish specimen, a value was randomly drawn from a distribution of mean $\delta^{15}\text{N}_{\text{Base}}$ and standard deviation SD_{Base} , and used to calculate a TP value. This procedure was repeated 9,999 times, and the generated data were averaged.

TP estimations were performed twice; the first adopting for all the species under analysis a mean trophic fractionation $\Delta^{15}\text{N}$ of 3.4 ‰ derived from the literature data on non-herbivorous aquatic consumers (Post 2002), the second using a species-specific $\Delta^{15}\text{N}$ value of $1.15 \text{ ‰} \pm 1.01$ (mean ± 1 SD, $n = 9$), calculated as the average of controlled laboratory measurements performed by Fantle et al. (1999) and Dittel et al. (2000) on *Callinectes sapidus* reported in Table 1, and more consistent with other literature syntheses presenting lower $\Delta^{15}\text{N}$ estimates (e.g., Vanderklift and Ponsard 2003a; see also Mancinelli 2012a). For *Pachygrapsus marmoratus*, no species-specific $\Delta^{15}\text{N}$ data are available; noticeably, consumer-diet $\Delta^{15}\text{N}$ values for brachyurans have been reported only for

Portunus pelagicus (Møller et al. 2008). The estimation for this species resulted within the range of the blue crab fractionation factors (Table 1); thus, for *P. marmoratus* a species-specific $\Delta^{15}\text{N}$ identical to *C. sapidus* was assumed. For *Sparus aurata*, a species-specific $\Delta^{15}\text{N}$ value of $3.54 \text{ ‰} \pm 0.58$ (mean ± 1 SD, $n = 11$; Table 1), calculated as the average of controlled feeding trials performed by Serrano et al. (2008) and Martin-Perez et al. (2013) was used.

2.4 Statistical analysis

Values in the test are expressed as mean ± 1 SE if not otherwise specified. One-way ANOVAs were used to assess the effect of seasonality on the abundance and mean weight of *Callinectes sapidus*. One-way repeated measures ANOVA was used to compare the trophic positions of *C. sapidus*, *P. marmoratus* and *S. aurata* calculated using different $\Delta^{15}\text{N}$. For ANOVA analyses, data were tested for conformity to assumptions of variance homogeneity (Cochran's C test) and transformed if required. When significant effects were detected, post hoc multiple comparisons of means were performed using the Tukey HSD test.

3 Results

During the study period, the mean water temperature measured over the four sampling stations increased progressively from $13.5 \pm 0.6 \text{ °C}$ in winter to 31.1 ± 0.5 and $29.7 \pm 0.7 \text{ °C}$ in summer and autumn, respectively (Fig. 2a). Salinity followed a similar pattern, passing from $19 \pm 2.3 \text{ ‰}$ in winter, indicating a considerable influence of freshwater inputs, to $32.4 \pm 1.3 \text{ ‰}$ in summer, close to values observed for nearby marine waters ($36.5 \pm 0.4 \text{ ‰}$, Mancinelli, unpublished data).

In total, 32 specimens of *Callinectes sapidus* were collected; no specimens belonging to other crab species were captured. The majority of the captured specimens were males (27) with an overall sex ratio of 0.18:1 (females:males). However, sex ratio progressively decreased during the year from 1 to 0 (Fig. 2b). The carapace width of sampled specimens ranged between 60 and 158 mm ($\text{CW}_{\text{♂}} = 125.6 \pm 4.2 \text{ mm}$), and the wet weight between 16.1 and 335 g ($\text{W}_{\text{♂}} = 162.3 \pm 13.9 \text{ g}$). The species was sampled throughout the study period, reaching minimum and maximum abundances in winter and summer, respectively (Fig. 2b; one-way ANOVA, $F_{3,12} = 6.46$, $P = 0.007$; HSD test: summer > autumn > spring = winter). The average weight of specimens varied significantly, with minimum values observed in spring (Fig. 2b; one-way ANOVA, $F_{3,28} = 5.28$, $P = 0.005$; HSD test:

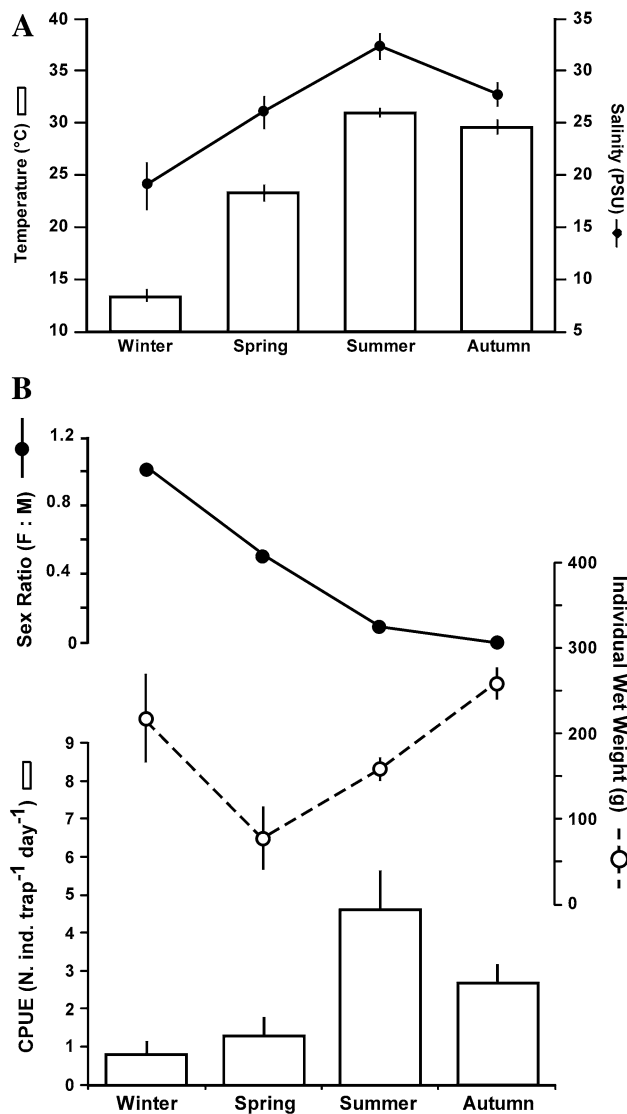


Fig. 2 **a** Bottom water temperature and salinity measured in the Torre Colimena basin over the study period. Values are means calculated over the four sampling stations. Bars = 1 SE; **b** Seasonal patterns of variation in the abundance (expressed as CPUE, catch per unit effort: N . individuals Trap⁻¹ Day⁻¹), mean individual size (wet weight, in g) and sex ratio (female:male) of *Callinectes sapidus* in the Torre Colimena basin during the study period

autumn = winter > summer > spring), followed by an increase up to values in autumn comparable with those observed in winter.

Five taxa of invertebrate primary consumers were sampled and analyzed, including crustacean and gastropod grazers (i.e., *Gammarus aequicauda*, *Sphaeroma serratum*, and *Cerithium* sp.) and sessile crustacean and mollusk filter feeders (*Balanus* sp. and *Mytilus* sp.). The grand mean $\delta^{15}\text{N}$ of primary consumers was 7.29 ± 0.89 (mean \pm SD), and significant inter-specific differences were observed (Table 2; one-way ANOVA, factor “species”, $F_{4,18} = 4.82$, $P = 0.01$).

Table 2 Stable nitrogen signatures $\delta^{15}\text{N}$ (mean, SD in brackets) of primary consumers used to calculate the baseline indicator $\delta^{15}\text{N}_{\text{Base}}$ and of *Callinectes sapidus*, *Pachygrapsus marmoratus* and *Sparus aurata*. The mean trophic positions (SD in brackets) calculated for these species using general (i.e., 3.4 ‰) and species-specific (i.e., 1.15 ‰ for both *C. sapidus* and *P. marmoratus*, and 3.54 ‰ for *S. aurata*; see text for further details) fractionation factors $\Delta^{15}\text{N}$ (TP_{3,4} and TP_{SS}, respectively) are also reported. Means and standard deviation calculated on four replicates for all species, with the exception of *P. marmoratus* and *S. aurata* ($n = 3$)

Taxon	$\delta^{15}\text{N}$	TP _{3,4}	TP _{SS}
<i>Sphaeroma serratum</i>	6.48 (0.04)		
<i>Gammarus aequicauda</i>	8.39 (1.04)		
<i>Cerithium</i> sp.	7.91 (0.62)		
<i>Mytilus</i> sp.	6.33 (0.94)		
<i>Balanus</i> sp.	7.36 (0.19)		
<i>Callinectes sapidus</i>	10.64 (0.28)	2.98 (0.08)	4.22 (0.19)
<i>Pachygrapsus marmoratus</i>	7.67 (0.39)	2.12 (0.12)	2.27 (0.26)
<i>Sparus aurata</i>	8.72 (0.28)	2.62 (0.14)	2.41 (0.09)

Decreasing the enrichment factor $\Delta^{15}\text{N}$ from 3.4 to 1.5 ‰ affected differently the three compared species (One-way repeated measures ANOVA, interaction factor $F_{2,7} = 56.2$, $P < 0.001$). For *Callinectes sapidus*, the trophic position calculated using a 3.4 ‰ $\Delta^{15}\text{N}$ value was significantly higher than that estimated using a 1.5 ‰ value (Table 2; HSD test, $P < 0.01$) while for *Pachygrapsus marmoratus* the effect was negligible ($P = 0.51$). Similarly, no significant effects related to the fractionation factor were observed for *Sparus aurata* (HSD test, $P = 0.67$).

However, independently from the $\Delta^{15}\text{N}$ value used, the trophic position of *C. sapidus* resulted always higher than those calculated for the other two species (HSD tests, *Callinectes* > *Sparus* > *Pachygrapsus*).

4 Discussion

In the present study, the total records, the occurrence of juveniles in spring and, more significantly, the regular capture during the whole sampling period, together constitute evidence of a well-established population of *Callinectes sapidus* in the Torre Colimena basin. There have been other claims of established populations in the Adriatic and Ionian Seas; however, reports have been generally limited in the temporal extent and the total number of specimens analyzed (Dulčić et al. 2011 and literature cited); constant and numerically important captures have been reported only for the eastern Mediterranean Sea (e.g., Tureli Bilen et al. 2011). This study provides original information about the seasonal patterns of variation characterizing the abundance, size and sex ratio of the species

in a Mediterranean coastal habitat. Although caution should be used when interpreting these results, given the relatively low number of total captures and the known limitations of traps in providing reliable quantitative estimations of crab population parameters (e.g., Sturdivant and Clark 2011), the observed seasonal patterns in abundance, size and sex ratio were highly consistent with biological information on the species in its native habitats. Specifically, the occurrence of small-sized specimens in spring and the low female : male ratio in summer and autumn confirmed the tendency of juveniles to migrate in brackish habitats at the end of winter months and of females to move to more saline waters for spawning (Miller and Smith 2003; Hines 2003; Kennedy and Cronin 2007).

In this study, the most apparent result was that, independently from the enrichment factor used, the trophic position of the blue crab resulted significantly higher than those characterizing the two autochthonous species included in the analysis, i.e., the omnivore crab *Pachygrapsus marmoratus* (Cannicci et al. 2002) and, more importantly, the benthivorous predator *Sparus aurata* (Pita et al. 2002). Our results suggest that *Callinectes sapidus* may occupy a predatory position in the Torre Colimena basin food chain, exerting a potentially high ecological impact on the coastal habitat. Noticeably, irrespectively of the fractionation factor, the estimated trophic position of the blue crab was higher than that calculated for *Sparus aurata*, whose adult stages are known to feed actively, among other preys, on crabs (e.g., Pita et al. 2002). This result may suggest that *C. sapidus* feeds on the same preys of the gilthead sea bream (e.g., native crab species) and, in addition, preys on live predatory fish species (including *S. aurata*). Alternatively, given the limited size of the analyzed breams (i.e., 142 ± 34 mm total length), it is likely that their feeding habits still relied mainly on small-sized benthic and planktonic invertebrates such as polychaetes, mysids and amphipods (Russo et al. 2007). This may have ultimately reflected on a lower $\delta^{15}\text{N}$ compared to the isotopic signature of the baseline species and, in turn, on a lower trophic position. Further isotopic studies including a wider—in both taxonomic and size span terms—representative of the predatory fish assemblage will provide an advanced resolution of the trophic relationships linking the blue crab to other autochthonous components of the benthic food web. In addition, it must be considered that the species is known to have generalist trophic habits including, besides predation, also cannibalism and scavenging (Kennedy and Cronin 2007); thus, the high trophic position observed here may actually reflect a diet based on conspecifics or fish remains. Alternatively, the nitrogen isotopic signature of *C. sapidus* may be ascribed to the consumption of prey outside the basin, as the species in its native habitats is characterized by a high mobility towards adjacent marine

areas (e.g., Carr et al. 2004). However, carbon isotope analyses suggest that this possibility is unlikely (Mancinelli et al. 2013a). Gut content examinations will help in clarifying the trophic ecology of the blue crab in invaded habitats, and whether any sex-related functional difference occurs between females and males, the former seasonally migrating to nearby marine habits for spawning. Additionally, long-term analyses are required to extend the resolution of *C. sapidus* trophic relationships throughout the year. Noticeably, blue crab populations from temperate Atlantic habitats are characterized by a period of winter quiescence during which specimens become inactive (Millikin and Williams 1984; Kennedy and Cronin 2007). In the Torre Colimena basin, the species was sampled in every season, suggesting that it may have adapted to the different abiotic conditions extending its activity throughout the year, ultimately making the species potentially more influential on the structure and dynamics of the benthic system.

Depending on the enrichment factor adopted, the trophic position of *C. sapidus* varied considerably. Specifically, the TP value calculated using an enrichment factor of 3.4 ‰ to 2.99 ± 0.08 —was consistent with other estimations provided in the literature, ranging from 2.8 to 3.3 (Moncreiff and Sullivan 2001; Rodriguez-Graña et al. 2008; Winemiller et al. 2007). Using a species-specific enrichment factor of 1.15 ‰, the calculated TP value increased to 4.22. The estimation was nonetheless consistent with the literature data: Abreu et al. (2006) and Rooker et al. (2006) indicated for the blue crab a trophic position of 4.35 and 3.4 in a Brazilian lagoon and in an open sea environment, respectively. These results, even though discordant, cannot be in principle considered biased or implausible, and lend further support to the current concern with trophic enrichment factors and their influence on the outcome of food web studies based on stable isotope analysis (Vanderklift and Ponsard 2003a; Mancinelli 2012a). Further laboratory investigations focusing on species-specific variations in isotopic metabolism are required for overcoming ambiguities determined by the adoption of generalized enrichment factors (Mancinelli 2012a; Mancinelli et al. 2013c).

In summary, we presented novel information on the seasonal occurrence of *Callinectes sapidus* in a Mediterranean coastal habitat, and, by estimating its trophic position, provided a first estimation of the potential ecological impact of the species within invaded coastal food webs. An estimation of the impact of introduced species on native communities has long been considered crucial in any policy of management and conservation of the ecological quality of aquatic habitats (Elliott 2003; Olenin et al. 2007); however, the impact of alien crustaceans, and, in general of IAS, on Mediterranean marine ecosystems has to date been poorly investigated (Galil 2011). The present

study represents a preliminary contribution to fill this lacuna.

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