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**Michele Gristina, Frine Cardone, Andrea Desiderato, Serena Mucciolo, Tamara Lazic & Giuseppe Corriero**

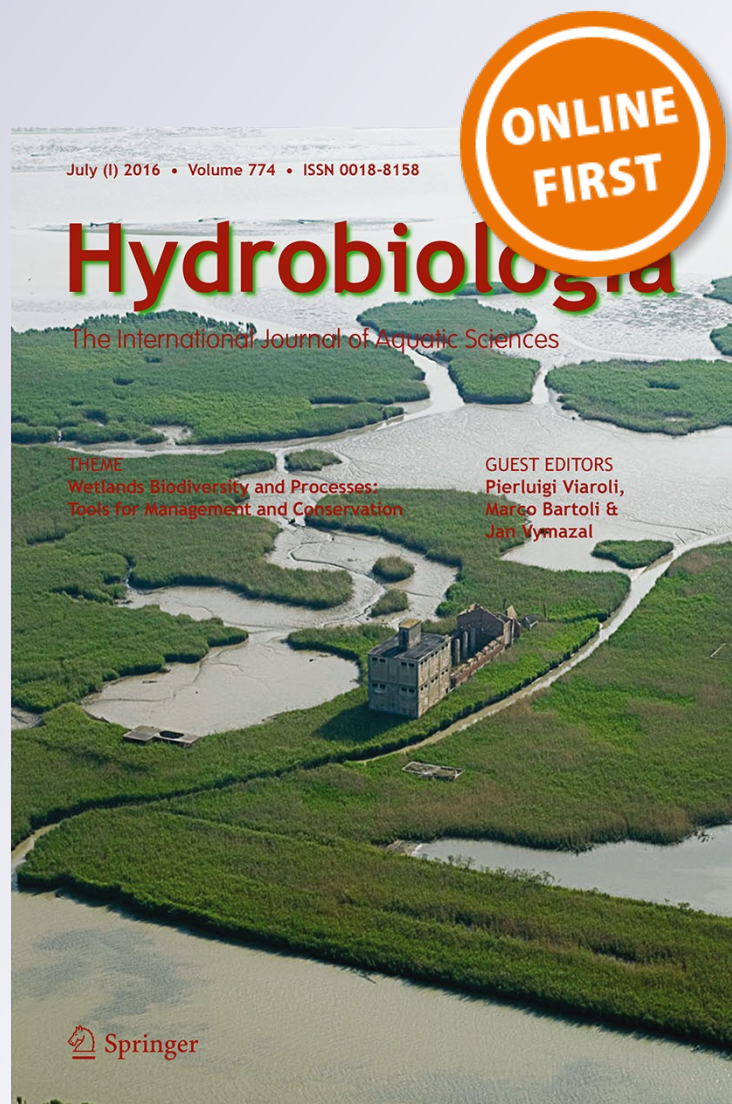
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# Habitat use in juvenile and adult life stages of the sedentary fish *Hippocampus guttulatus*

Michele Gristina · Frine Cardone · Andrea Desiderato · Serena Mucciolo · Tamara Lazic · Giuseppe Corriero

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**Abstract** Understanding of the spatial distribution and habitat use in different stages of a life cycle represents the essential aspect of threatened species management and conservation. In the present paper, the spatial and temporal patterns of habitat use in juvenile and adult life stages of the long-snouted seahorse *Hippocampus guttulatus* in the Mar Piccolo of Taranto (Apulia—Italy) (40°28'N, 17°16'W) were examined. From October 2012 to January 2014, monthly visual censuses were conducted in six coastal habitats of a focal area (Buffoluto site). Of a total of 317 individuals of the long-snouted seahorse that were sighted, 148 were juveniles, 82 adult females and 87 adult males. Our results showed significant differences in the habitat use between adult and juvenile life stages of *H. guttulatus*. Adult individuals were mainly concentrated within *Cladophora prolifera* beds and the wood poles of a mussel farm at 2–4 m of depth, while juvenile individuals were mainly present in shallower water (0.4–1 m) on artificial hard substrates

covered by a brown algae turf. Assessing and describing the habitat use within different life stages of *H. guttulatus* represent a useful tool to support species conservation by protecting the habitats it uses.

**Keywords** *Hippocampus* · Underwater visual census · Habitat use · Nursery area · Threatened species · Conservation

## Introduction

Identification and conservation of essential habitats are important for sustainable fisheries and conservation purposes (Rosenberg et al., 2000; Wells & Rooker, 2004). In the last two decades, fisheries scientists have made increasing effort to identify the distribution and habitat needs of species throughout their life cycle (Tuckey & Dehaven, 2006) giving valuable information regarding the physical attributes and biological significance of such habitats. As a consequence, the nursery and spawning areas of the main target species have been actively investigated to increase knowledge about the biology of individual species and to develop management plans to protect critical habitats (Garofalo et al., 2011).

Understanding how well marine animals can cope with different habitat features is of great significance for their conservation, and it is the crucial trait for sedentary animals that have developed life history

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strategies to feed, grow, and reproduce while minimizing the need for locomotion (Caldwell, 2012; Caldwell & Vincent, 2012).

Sedentary marine animals usually inhabit shallow coastal areas, where anthropogenic disturbances tend to be most frequent and severe, representing the main threat to their survival. Because life history strategies of sedentary animals are in association with high habitat specialization, these animals are particularly vulnerable both to habitat loss/degradation and to exploitation (Reynolds et al., 2005).

The identification of habitats in which sedentary organisms can complete the distinct stages of their life cycle referring primarily to the vulnerable juvenile stage has become a priority in management and conservation actions.

Among sedentary fishes, seahorses (genus *Hippocampus* Rafinesque, 1810) appear as a useful model. They avoid predation by using camouflage, they are ambush predators, and most species are monogamous within one or several breeding seasons (Foster & Vincent, 2004; Harasti et al., 2012). Due to their strict association with preferential habitat and peculiar life strategies (sparse distribution, low mobility, small home ranges, low fecundity, lengthy parental care, and mate fidelity) they become extremely vulnerable to the anthropogenic impacts that affect coastal areas. For this reason, seahorses are listed on the World Conservation Union's Red List of Threatened Species (Vincent & Hall, 1996; IUCN, 2013) and in 2002 were added to the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES, 2001).

Like most of its congeneric, also the long-snouted seahorse *Hippocampus guttulatus* Cuvier, 1829 is a cryptobenthic fish species which, in adult life stage, exhibits high site fidelity, tends to stay within small home ranges and has relatively limited swimming ability, although it has been reported that in the presence of natural or anthropogenic disturbances is capable of longer distance movement in search of more suitable environmental conditions (Curtis & Vincent, 2005, 2006; Caldwell & Vincent, 2013). Long-snouted seahorses (in British also called the Spiny Seahorses) inhabit shallow inshore waters of the Mediterranean Sea, the Black Sea and the north-eastern Atlantic Ocean from the North African coast (Lourie et al., 2004) to the coast of Shetland Isles, out into the North Sea and all around the United Kingdom

(Garrick-Maidment et al., 2014), usually associated with complex habitats with greater seagrass density, vegetation cover and sessile invertebrates that can provide holdfasts and abundant food (Curtis & Vincent, 2005; Caldwell & Vincent, 2012). However, due to its preferences to shallow areas of seagrass beds that are prone to both natural and human-induced disturbances, *H. guttulatus* is highly sensitive to the habitat loss and destruction. Especially in the Mediterranean, coastal aquatic phanerogams (*Posidonia oceanica* (L.) Delile, 1813, *Cymodocea nodosa* (Ucria) Ascherson, 1870 and *Zostera marina* Linnaeus, 1753) are experiencing a severe decline with significant effects on biodiversity, food webs and associated ecological processes (Calizza et al., 2013; Bostrom et al., 2014; Tuya et al., 2014).

Since little data are available across most of its geographic range, *H. guttulatus* cannot be reliably classified regarding global risk according to the IUCN categories, and is listed, at present, as data deficient (DD). Several ecological and biological aspects of the long-snouted seahorse adult population are well known (Curtis & Vincent, 2006; Garrick-Maidment, 2011; Caldwell & Vincent, 2012; Gristina et al., 2015) though information about recruits and/or young of the year (yoy) are limited and scanty. In fact, due to their cryptic, camouflaged appearance and variability in the timing and magnitude of recruitment events (Doherty & Williams, 1988), sightings of juveniles are uncommon and rare. Several authors have suggested that the rarity of juvenile findings may be due to the inherent limitations in the sampling approach (e.g. visual census) (Woodall, 2009) and to the ontogenetic habitat shift (different habitats from adults) (Boiseeau, 1967).

This study focused on the population of *H. guttulatus* in the focal area (Buffoluto station) of the Mar Piccolo of Taranto, which is a semi-enclosed marine system located in south-eastern Italy. Across the whole Mar Piccolo, the use of towed fishing gears is hindered by the presence of extensive mussel farming systems, which should make fishing pressure negligible. The *H. guttulatus* population in this area was described in the last years (Gristina et al., 2015), while the persistent nursery area has been identified and monitored in the Buffoluto focal area (Gristina, 2015).

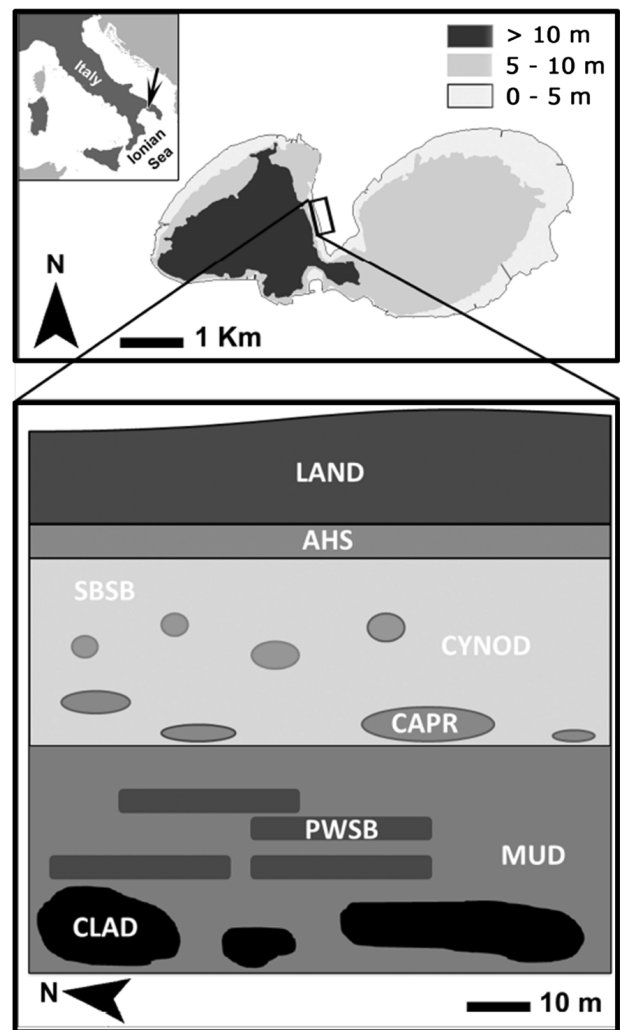
For *H. guttulatus*, such as for many other fishes in which the period of the highest mortality occurs during the early life stages (Houde, 2008), it is important to

describe the features of habitats used by juveniles aiming better prediction of the human pressure's effects. Moreover, identification and description of the habitats used by juveniles represent an essential step for determination of habitats that act as a nursery area, contributing to the adult populations sustaining (Beck et al., 2001). The central goal of this paper was individuation and description of the habitats mainly used by juveniles of *H. guttulatus* in the focal area of the Mar Piccolo. Besides, the comparison of habitat use in adult and juvenile individuals, both at spatial and temporal scale was conducted.

### Study area

The Mar Piccolo (40°28'N, 17°16'W) is located at the northern end of the Gulf of Taranto (Ionian Sea), with a total surface area of about 20.63 km<sup>2</sup> (Fig. 1). It is characterized by low water turnover and water movement (Capasso et al., 1989). Water temperature varies from 8.4°C (December) to 26.8°C (August) (Caroppo & Cardellicchio, 1995). Salinity values are around 36 psu (Caroppo & Cardellicchio, 1995). Dissolved oxygen shows extensive seasonal variability: from May to October, oxygen deficit in the deeper layers of Mar Piccolo (from about 7–10 m of depth) can reach marked levels, and may be responsible for dystrophic events (Alabiso et al., 1997; Gaino et al., 2010). *Cymodocea nodosa* and unattached macroalgae are covering the dominant soft bottoms habitats (Cecere & Petrocelli, 2009).

Where vegetation is missing, a large number of benthic filter feeders (sabellids, molluscs, ascidians and ceriantharia) colonize the soft bottoms. Hard substrates are very abundant and consist of stones and anthropogenic debris, such as ropes and material discarded by mussel farmers and fishermen. On hard substrates, the macrozoobenthic community is characterized by the wealthy assemblages of filter feeders (mainly poriferans, hydrozoans, polychaetes, bryozoans, bivalves, crinoids and ascidians) forming pluristratified assemblages. The sea bottom of the Buffoluto focal area exhibits the same edaphic and biocenotic characteristics described for the rest of the Mar Piccolo (Gristina et al., 2015). Moreover, our focal area is characterized by a continuous stone wall along the coastline (Fig. 1). This artificial structure supports an algal turf mainly constituted by perennial



**Fig. 1** Study area and schematic representation of the sampled habitats. *AHS* Artificial Hard Substrates; *SBSB* Soft Bottoms with Scattered Bioconcretions; *CYNOD* *Cymodocea nodosa*; *CAPR* *Caulerpa prolifera*; *PWSB* Poles With Soft Bottom; *CLAD* *Cladophora prolifera*. Mud habitat was not sampled in this work

*Cystoseira* C. Agardh, 1820 spp. and other frondose algae (i.e. *Corallina elongata* J. Ellis & Solander, 1786, *Dictyopteris* J. V. Lamouroux, 1809 spp.).

### Underwater visual census

In the summer 2012, preliminary surveys were carried out by SCUBA divers with the scope of habitat features mapping in the focal area of interest. A total of six principal habitats (Table 1) were identified, measured with a metric rope, mapped and considered for further investigation (Fig. 1). In these habitats,

**Table 1** Descriptions of sampled habitat types and depth range

Habitat type	Code	Description	Surface (m <sup>2</sup> )	Depth range (m)
Artificial hard substrates	AHS	Concrete wall, stones and debris of human origin on the bottom with abundant algae ( <i>Cystoseira</i> sp., <i>Dictyota dichotoma</i> , <i>Corallina elongata</i> ) and filter feeders (large sabellids, both colonial and solitary ascidians, demosponges, bryozoans and hydrozoans)	5,504	0.4–0.7
Soft bottoms with scattered bioconcretions	SBSB	Near the coastline, soft bottoms are mixed with a large amount of organogenous concretions (bivalve and gastropods shells), small stones and artificial hard substrates that are mainly colonized by sabellids and solitary ascidians	4,419	1.6–2.2
<i>Cymodocea nodosa</i>	CYNOD	Patches more or less dense interspersed with soft bottom. Few benthonic organism are present (sabellid polychaetes and ascidians). Abundant pipefishes	574	0.8–1.4
<i>Caulerpa prolifera</i>	CAPR	<i>C. prolifera</i> algal beds represent a very monotonous habitat, with a few associated zoo benthic species (some gastropods, large opisthobranchs, the sea anemone <i>Aiptasia</i> sp.), mainly distributed in the upper layer of the phytocoenosis	959	1.0–1.5
Poles with soft bottom	PWSB	Wood pools for mussel farming are colonized by a rich sessile filter feeder fauna. This benthic assemblage is distributed in a pluristratified pattern. The basal layer is mainly colonized by bivalves ( <i>Mytilus galloprovincialis</i> and <i>Ostrea edulis</i> ), demosponges ( <i>Tethya</i> spp., <i>Hymeniacidon perlevis</i> , <i>Oceanapia isodyctiformis</i> , <i>Suberites carnosus</i> ) and ascidians ( <i>Styela</i> spp.). Since the usual occurrence of large amounts of hard artificial substrates (ropes, chains, bricks) and benthic organisms (mussel shells, bryozoa) also the soft bottom displaced around each pool has been sampled	420	3.3–4.2
<i>Cladophora prolifera</i>	CLAD	Large beds interspersed with soft bottom. Sabellid polychaetes, solitary and colonial ascidians are scattered on the substrate. The Ceriantharia <i>Pachycerianthus solitarius</i> may be locally abundant	7,471	3.3–3.9

underwater visual censuses (Samoilys, 1997) were performed monthly, from October 2012 to January 2014 during the morning/afternoon periods. Monthly data were classified according to the four seasonal levels (see experimental design). For each habitat, three randomly selected 2 m × 10 m linear transects were monitored ( $n = 9$  for each season).

For each seahorse sighted, habitat and depth have been recorded in situ to obtain further biometric information. All specimens encountered were gently placed in a semi-rigid plastic bag and then photographed with a ruler positioned as close as possible. Although this method slightly increases the specimens manipulation compared to the method proposed by Lourie (2003), it allows to align the seahorse with the ruler and thus to obtain more precise measurements. Seahorses were photographed frontally and laterally to identify reproductive stage and increase the reliability

of gender determination from photos. After the procedure, animals were released onto the same holdfast from which they were collected. Later, photos were analysed to determine sex and standard length using the tpsDig2 1.11 program. Seahorse standard length was measured as a linear distance from the snout to the middle of operculum, and then down the body axis to the end of a tail, following the measurement protocol suggested by Lourie (2003).

In our study *H. guttulatus* specimens were considered juveniles at  $96.0 \pm 8.0$  mm (SL) (Curtis, 2004). The used cut-off roughly corresponds to the first two size/age categories proposed by Correia (2015) in a more detailed classification.

The undertaken research in this project was accomplished with the accordance of the permit 00210337/18.09.2012 issued by the Italian Ministry of the Environment and Protection of Land and Sea, II

Division. During the study, as the stress due to seahorse handling was minimized and as sampling procedures were carried out in situ without damaging, sacrificing or removing any specimen from the water, approval from the Italian Institutional Animal Care and Use Committee “Organismo preposto al benessere degli animali” (O.P.B.A.) (Article 26 of the Legislative Decree No 26/2014 of the Italian Republic) was not required.

#### Experimental designs and statistical analyses

A three-way distance-based permutational ANOVA (Anderson, 2001) was performed to test potential differences in the patterns of distribution between juvenile and adult life stages. This analysis, in which the F-statistics are calculated, but *P* values are obtained by permutation, was performed rather than traditional univariate ANOVA as it avoids any assumptions about either the nature of the distribution of the original variables or the homogeneity of variances (Anderson, 2001; Anderson & Ter Braak, 2003). A three-way design was conducted, in which ‘Season’ was treated as a fixed factor with four levels (Autumn, Winter, Spring and Summer), ‘Habitat’ as a fixed and orthogonal factor with six levels (CYNOD, AHS, CAPR, CLAD, SBSB, PWSB) (Table 1), and ‘Life Stage’ as a fixed and orthogonal factor with two levels (Adults and Juveniles). Nine replicates of density counts were collected for each combination of the factors. All analyses were based on Euclidean distances in the original raw data, with all *P* values obtained using 9999 permutations of the appropriate exchangeable units (Anderson & Ter Braak, 2003). Significant terms in the full model were examined individually using appropriate a posteriori pairwise comparisons, also conducted by permutations (Anderson, 2001). The software applications PRIMER 6.1.10 and PERMANOVA+ b20 (<http://www.primers-e.com>) were used to perform all procedures.

#### Results

In the six habitats surveyed, a total of 317 sightings of the long-snouted seahorse were reported. Of a total number of animals sighted during the surveys, 148 were juveniles (20.6% males; 23.8% females; 2.72% undetermined), 82 adult females (25.5%) and 87 adult

males (27.2%). The *H. guttulatus* sex ratio (females/total adults) was substantially unbiased (48.5%). Height in juvenile specimens ranged from 2.8 to 9.6 cm (mean =  $6.76 \pm 1.05$  cm), while in adults height ranged from 9.7 to 13.5 cm (mean =  $10.2 \pm 0.83$  cm). The mean density of long-snouted seahorses during the sampling period ranged from 0.031 ind.  $m^{-2}$  ( $\pm 0.0063$  SE) in January 2014 to 0.081 ind.  $m^{-2}$  ( $\pm 0.0108$  SE) in September 2013.

In spring (May, June) and autumn (September) months, the density values were highest, while summer (July, August) and winter (February, March) months showed the lowest densities (Fig. 2). Total abundance of juveniles and adults revealed wide fluctuation during the study period (Fig. 2). Adults had the main peak in September 2013, while juveniles showed two peaks, one in December 2012 and the other one in June 2013. Males with a fully developed brooding pouch (Lourie, 2003) peaked in June but sighted from April to September.

*Hippocampus guttulatus* was unequally distributed across the habitats monitored. Population density of this species is shown to be lower within natural substrates such as *Cymodocea nodosa* (CYNOD) ( $0.027$  ind.  $m^{-2} \pm 0.007$  SE), *Caulerpa prolifera* (Forsskål) J.V.Lamouroux, 1809 (CAPR) ( $0.017$  ind.  $m^{-2} \pm 0.01$  s.e), *Cladophora prolifera* (Roth) Kützing, 1843 (CLAD) ( $0.059$  ind.  $m^{-2} \pm 0.009$  SE) and in Soft Bottoms with Scattered Bioconcretions (SBSB) ( $0.032$  ind.  $m^{-2} \pm 0.074$  SE), than in Artificial Hard Substrates (AHS) ( $0.18$  ind.  $m^{-2} \pm 0.016$  SE) and Poles With Soft Bottom (PWSB) ( $0.09$  ind.  $m^{-2} \pm 0.02$  SE) (Fig. 3). Adult individuals resulted widely distributed in all six habitats monitored. However, very high density values were recorded in PWSB ( $0.069$  ind.  $m^{-2} \pm 0.018$  SE), CLAD ( $0.054$  ind.  $m^{-2} \pm 0.009$  SE) and AHS ( $0.049$  ind.  $m^{-2} \pm 0.012$  SE) (Fig. 4). Juveniles, by contrast, were mainly concentrated in the AHS habitat ( $0.132$  ind.  $m^{-2} \pm 0.018$  SE), being rare in other habitats.

Density distribution patterns of *H. guttulatus* juvenile and adult life stages were significantly different in the interaction term Life Stage  $\times$  Habitat (LixHA) (3-way PERMANOVA; pseudoF<sub>5,420</sub> = 22.585, *P* = 0.0001). For the former interaction, pairwise *t* tests revealed that density of adults was significantly higher in CLAD, PWSB and SBWC, while juveniles were significantly more abundant in AHS habitat (Fig. 4; Tables 2, 3).



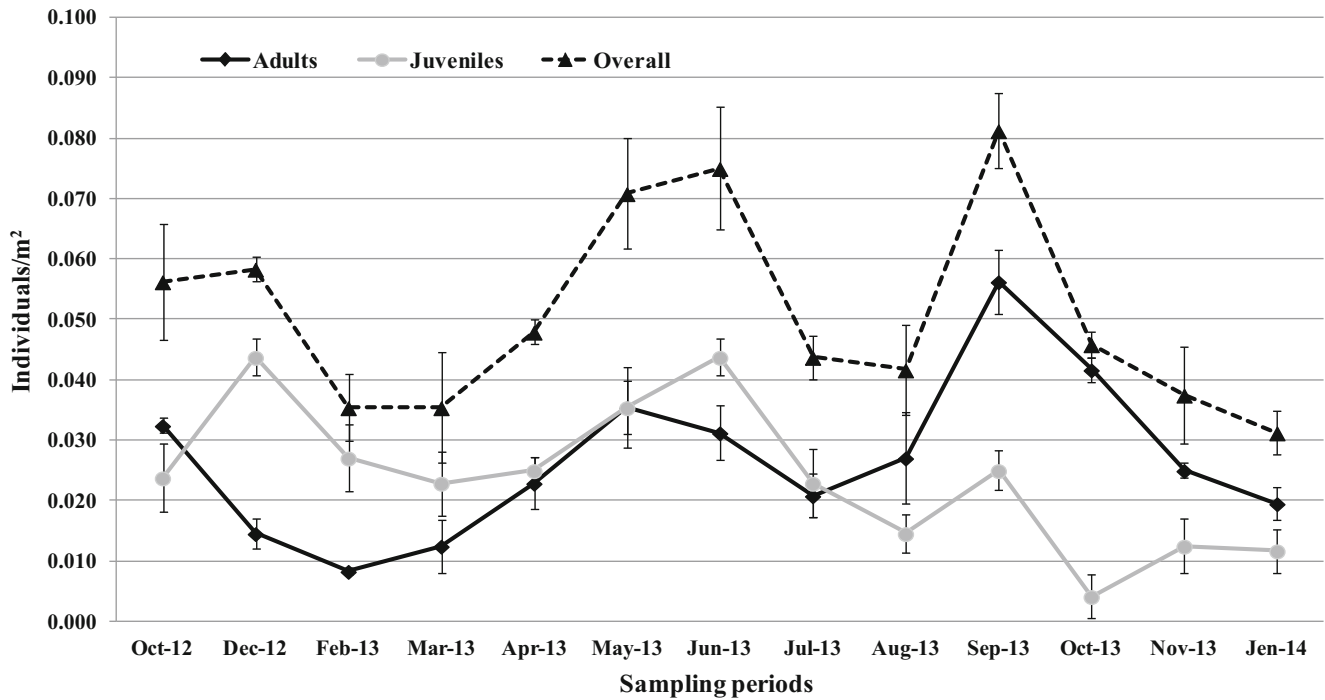


Fig. 2 Density of juvenile, adult and overall long-snouted seahorse specimens during the study period

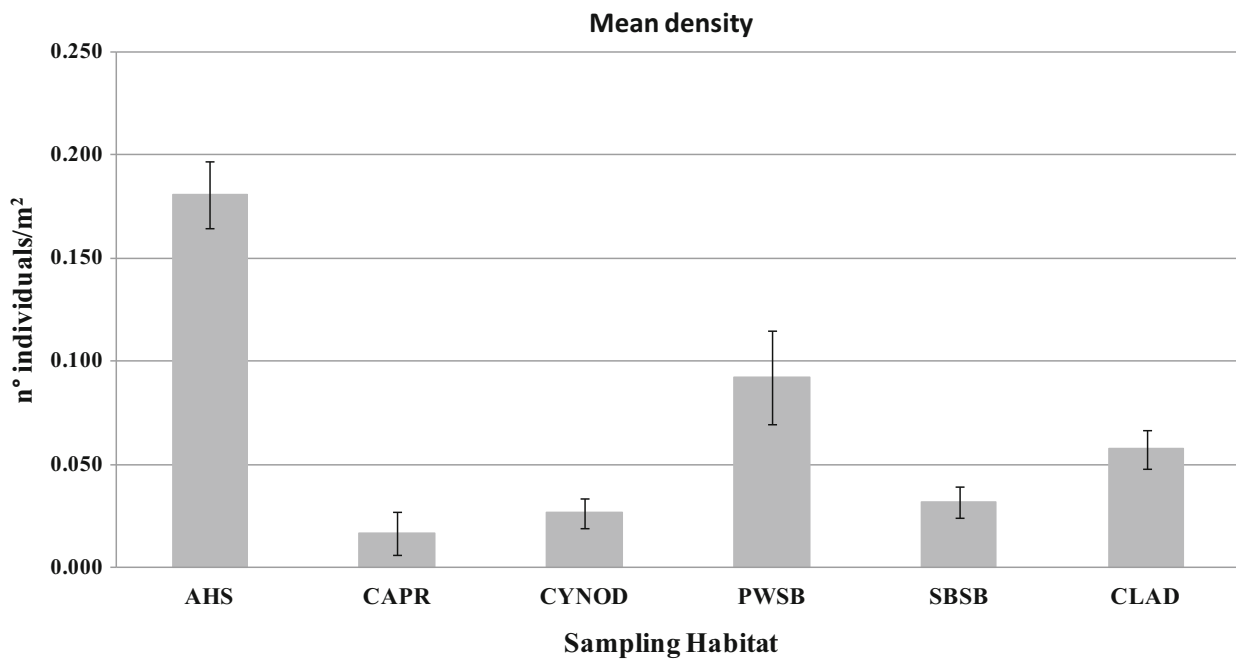
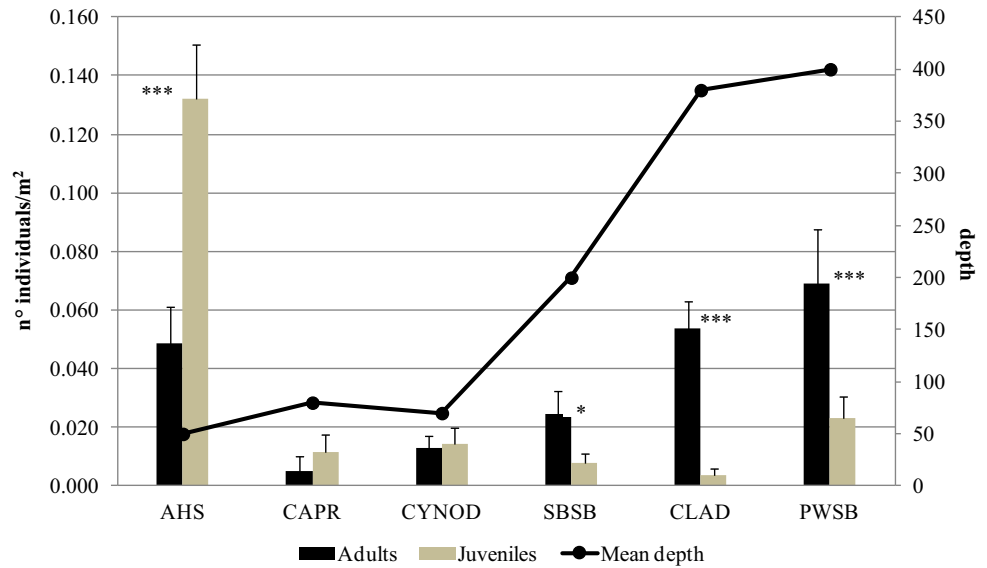


Fig. 3 Mean density of *H. guttulatus* in the sampled habitats

As previously reported (see also Fig. 2), the overall density of *H. guttulatus* differed significantly between habitats and seasons (3-way PERMANOVA; pseudoF<sub>5,420</sub> = 1.8094, *P* = 0.0333) independently from life stages. However, the pairwise *t* test revealed

significant seasonal differences only in those habitats least used by juvenile and adult stages (e.g. CYNOD, CAPR, SBWC), while in the habitats mainly used by seahorses, any difference in the overall density was not found (Table 4).

**Fig. 4** Mean density of juveniles and adults of *H. guttulatus* in the sampled habitat. \*  $\leq 0.05$ ; \*\*\*  $\leq 0.001$



**Table 2** Three-way distance-based permutational ANOVA on untransformed *H. guttulatus* density

Source	df	SS	MS	Pseudo-F	P (perm)	Unique perms
Season (SE)	3	9.1318	3.0439	3.5783	0.0143	9,894
Habitat (HA)	5	150.39	30.077	35.358	0.0001	9,920
Life stage (LI)	1	0.45	0.45	0.529	0.4668	9,330
SExHA	15	23.088	1.5392	1.8094	<b>0.0333</b>	9,923
SExLI	3	5.8269	1.9423	2.2833	0.0804	9,941
HAxLI	5	96.063	19.213	22.585	<b>0.0001</b>	9,948
SExHAxLI	15	19.752	1.3168	1.548	0.0834	9,912
Residuals	420	357.28	0.85066			
Total	467	656.28				

Bold values indicate  $P < 0.05$

**Table 3** Pairwise *t* tests within each level of factor ‘Habitat’ for the levels of factor ‘Life Stage’

Groups	<i>t</i>	P (perm)	perms
CLAD Adult/Juvenile	<b>5.966</b>	<b>0.0001</b>	<b>4,213</b>
PWSB Adult/Juvenile	<b>3.704</b>	<b>0.0001</b>	<b>6,837</b>
AHS Adult/Juvenile	<b>4.923</b>	<b>0.0001</b>	<b>8,359</b>
CYNOD Adult/Juvenile	0.446	0.6695	1,678
CAUL Adult/Juvenile	1.032	0.3304	1,042
SBWC Adult/Juvenile	<b>2.333</b>	<b>0.0227</b>	<b>1,947</b>

Significant interaction HAxLI; significant pairwise comparisons are in bold

## Discussion

Adult and juvenile life stages of many marine species of both invertebrates and vertebrates exhibit

ontogenetic changes in the habitat use during their life cycle (Childress & Herrnkind, 2001; Gristina et al., 2013). These changes are usually triggered by alterations in animal’s needs (i.e. feeding, predator avoidance) (Dahlgren & Eggleston, 2000) and different perceptions of habitat features between juvenile and adult specimens of the same species (Grol et al., 2011).

Information about the *H. guttulatus* distribution along the Italian coast is very fragmented. It has been found that large and persistent populations of this species are mainly concentrated in the confined lagoon systems, such as Taranto, Venice, Chioggia and, perhaps, in some coastal lakes of Sardinia. As already observed in the exposed coastlines of other regions (Curtis & Vincent, 2005; Garrick-Maidment, 2012), also in Italian open coastal waters, the occurrence of *H. guttulatus* is occasional or at most limited to a small

**Table 4** Pairwise *t* tests within each level of factor 'Habitat' in the significant interaction SExHA

Groups	CLAD	PWSB	AHS	CYNOD	CAUL	SBWC
Fall, winter	0.1967	0.3465	0.6283	0.5285	0.5966	<b>0.0068</b>
Fall, spring	0.5625	0.8614	0.1939	<b>0.0352</b>	0.0912	<b>0.0318</b>
Fall, summer	0.1812	0.0810	0.3881	0.7223	0.3335	0.1857
Winter, spring	0.1339	0.2741	0.0609	0.1214	<b>0.0452</b>	1.0000
Winter, summer	1.0000	0.0630	0.0988	0.2429	1.0000	0.1281
Spring, summer	0.0960	0.1154	0.5515	<b>0.0058</b>	<b>0.0194</b>	0.3242

Only P(permutation) values are reported; significant pairwise comparisons are in bold

group of individuals (Gristina, 2015). This distribution pattern can be explained by the combination of several factors: significant regression of several aquatic phanerogams along the Italian coast (Bostrom et al., 2014; Tuya et al., 2014; Telesca et al., 2015), dramatic impact of commercial towed gears on the coastal biocenosis (Ardizzone et al., 2000; De Biasi, 2004), decreased holdfasts availability (Correia et al., 2015) and by *H. guttulatus* migration into deeper water at the beginning of stormy and cold winter (Garrick-Maidment et al., 2014). In spite of this general pattern, in the Mar Piccolo of Taranto, where the presence of a large mussel farm prevents any impact from towed fishing gears, a large population was estimated (Gristina et al., 2015).

The present study aims addition of new data about the habitats used by different life stages (juveniles/adults) of the long-snouted seahorse population in the focal area of the Mar Piccolo of Taranto (Southern Italy). The *H. guttulatus* specimens at this site showed the preferential use of Artificial Hard Substrates (AHS), Poles With Soft Bottom (PWSB) and *Cladophora prolifera* beds (CLAD) (Fig. 2) habitats. Throughout the 12 months of monitoring, population of the long-snouted seahorse demonstrated a stable and persistent use of habitat. In fact, when subjected to the seasonal pairwise comparison (Table 4), the density of *H. guttulatus* in AHS, PWSB and CLAD habitats showed no differences during the period of monitoring.

Although persistent observations of juveniles appear poorly documented in the literature, the present study recorded a high number of juvenile specimens (i.e. 148). The high water transparency and low average depth in the Buffoluto study site allowed us to locate carefully and describe broad size spectrum of

the *H. guttulatus* specimens within the population. Results of this study revealed that juveniles were recruiting to the benthic habitat mainly during the summer and autumn months. Juveniles with a minimum of 2.5 cm of a total length were sighted recruiting to the sea bottom, grasping with their prehensile tails to mainly vegetated habitats. The main concentration of juveniles was in the shallow water habitats and they showed the preferential use of AHS habitat to anchor and hide. This habitat is characterized by the presence of algal turf, primarily constituted by perennial *Cystoseira* spp., found associated with other frondose algae (i.e. *Corallina elongata*, *Dictyopteris* spp.), and it apparently results as appropriate substrate for juvenile specimens, optimizing their camouflage efficiency and reducing the risk of predator encounter.

The importance of a complex habitat which allows hiding from predators by camouflage was widely described for other species of genera *Hippocampus*. It was found that the density of *Hippocampus capensis* Boulenger, 1900 was positively related to the presence of *Zostera capensis* Setchell, 1933 (Whitfield, 1995), while the density of the Australian *Hippocampus whitei* Bleeker, 1855 was associated with *Posidonia König*, 1805 sp. (Middleton et al., 1984) and to the soft coral *Dendronephthya australis* Kükenthal, 1905 (Harasti et al., 2014). Moreover, the importance of holdfast availability in natural seahorse population sustaining was highlighted in some paper both for artificial (Hellyer et al., 2011; Correia et al., 2015) and natural holdfasts (Perante et al., 2002). Although well documented for *Hippocampus* spp. adults, the information about the habitat use in juveniles is rare and occasional, and only recently, Harasti et al. (2004) reported that *H. whitei* juveniles prefer complex

habitat represented by the gorgonian *Euplexaura* Verrill, 1869 sp.

The high algal cover on Artificial Hard Substrates (AHS) appears to encourage the settlement of seahorse juveniles. This habitat seems to act as a “sink area” in which newborns concentrate from April to September, within the period when pregnant males can be sighted and when the tally of the frondose brown algae reach maximum development (Sales & Ballesteros, 2012). After the pelagic phase, when new recruits reach the sea bottom, they will find a highly favourable habitat where they can safely stay until they reach the adult or pre-adult stage. Once reached, individuals will move towards the deeper habitats where they will use distinct microhabitats to optimize feeding strategies and reproductive success (Morgan & Vincent, 2007).

Adult individuals (both males and females) preferentially use the Poles With Soft Bottom (PWSB) and *Cladophora prolifera* beds (CLAD) (Fig. 4). PWSB is a complex habitat that provides many suitable microhabitats used for camouflaging and it is a good source of amphipods and copepods, regarded as the main prey of seahorses (Kitsos et al., 2008; Kuitert, 2009). Unlike PWSB, the CLAD habitat, due to its rapid turnover, can be considered as a habitat of low complexity, which gives rise to a rich detritus food chain by supporting an abundant population of crustacean detritivores. As observed for other bed-forming algae, also *Cladophora prolifera* beds are mostly exploited by amphipods (Prato & Biandolino, 2003), where the high production of algal detritus leads to rapid population and somatic growth of deposit-feeding invertebrates (Levinton & McCartney, 1991).

As for *H. guttulatus*, similar ontogenetic changes in the habitat preferences between juvenile and adult life stages have been described for *Hippocampus comes* Cantor, 1849 (macroalgal and coral habitats for juveniles and adults, respectively) (Morgan & Vincent, 2007) and for *H. whitei* (gorgonians for juveniles, and both sponges and soft corals for adults) (Harasti et al., 2014). For the long-snouted seahorse, although a vast amount of scientific production has been done, no specific works explored the spatial distribution of different life stages. The lack of available information about the habitat use in juveniles of this species can be explained by the low number of juveniles sighted during random Underwater Visual Census surveys (Curtis & Vincent, 2005).

Our results are giving an insight into the habitat used by the *H. guttulatus* juveniles and adults, showing the important differences in the habitat use between these two life-cycle stages. As already suggested by Caldwell (2012), this species is showing a great flexibility and adaptability to cope with the habitat loss/degradation.

In the last 20 years, marine environment in the Mar Piccolo of Taranto appears strongly influenced and threatened by different anthropic pressures. Nevertheless, *H. guttulatus* is able to fit all habitats available, forming a large and temporally persistent population. The juveniles are capable of recruiting to the habitats of human origin (AHS), to stay there and to use it until they reach the adult or pre-adult phase. The juveniles are not subjected to the impact of mechanical gears, due to the mussel farm presence, and they seem able to take advantage of the rich epifauna “pabulum” supported by the high trophic load in the study area.

Although in a context of environmental degradation, this paper allows the nursery ground identification of the long-snouted seahorse population in the Mar Piccolo of Taranto, giving a fundamental contribution to the development of spatial conservation planning. To support its conservation, assessment and description of the habitat use in *H. guttulatus* life stages represent a useful tool.

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