Article

Abundance, Distribution, and Habitat Preference of Syngnathid Species in Sabaudia Lake (Tyrrhenian Sea)

Tamara Lazic [1], Cataldo Pierri [1,*], Giuseppe Corriero [1], Maria Flavia Gravina [2], Michele Gristina [3], Miriam Ravisato [1] and Armando Macali [4]

[1] Department of Biosciences, Biotechnologies and Environment, University of Bari, 70125 Bari, Italy; tamara.lazic@uniba.it (T.L.); giuseppe.corriero@uniba.it (G.C.); miriam.ravisato@uniba.it (M.R.)

[2] Department of Biology, University of Rome ''Tor Vergata'', 00133 Rome, Italy; maria.flavia.gravina@uniroma2.it

[3] Institute of Anthropic Impacts and Sustainability in Marine Environment (IAS), National Research Council, 90146 Palermo, Italy; michele.gristina@cnr.it

[4] Department of Ecological and Biological Sciences, Ichthyogenic Experimental Marine Centre (CISMAR), Tuscia University, 01016 Tarquinia, Italy; a.macali@unitus.it

*   Correspondence: cataldo.pierri@uniba.it

Abstract: Syngnathids are considered flagship species of estuarine and coastal environments. However, most of the Mediterranean species are still classified as data-deficient (DD) at a global level according to the IUCN Red List of Threatened Species. In central Italy, several areas host potentially suitable habitats for syngnathids but have not been previously reported in the literature; the estimation of population parameters and habitat partitioning at these sites may help to assess their conservation status. In this study, we investigated the abundance, distribution, and habitat preferences of sympatric populations of *Hippocampus hippocampus*, *H. guttulatus*, *Syngnathus abaster*, and *Nerophis ophidion* in Sabaudia Lake (Tyrrhenian Sea, Italy). While confirming the primary importance of a healthy coastal habitat, we retrieved hints about species ecology and habitat use. The species distribution in the study area highlights the role of habitat complexity in supporting local populations of these sensitive species.

Keywords: Syngnathidae; pipefishes; seahorses; Mediterranean; conservation

1. Introduction

Worldwide, species whose habitats overlap with human activities are considered to be under major conservation risk [1]. Environmental and anthropogenic threats, including climate changes, land-based pollution, and coastal eutrophication, usually have higher impacts in shallow coastal zones subjected to simultaneous acts of multiple pressures [2–4].

Among shallow coastal zones, coastal lakes and estuaries play an important role for many fish species, including syngnathids [5–8]. These charismatic fish have unique life cycle traits, such as a restricted home range, low mobility, and reduced reproductive output, which make them susceptible to direct and indirect threats; these include the degradation of habitats [9], incidental capture in fishing gear [10], and aquarium trade [11]. Moreover, the use of syngnathids (especially seahorses) in Traditional Chinese Medicine (TCM) is a widespread issue with a more recent emergence in the Mediterranean Sea [12]. Consequently, all syngnathids have been listed in Appendix II of CITES and the IUCN Red List of Threatened Species [13]; according to the latter, most syngnathids in the Mediterranean Sea are classified as data-deficient on a global level [13], indicating an urgent need for further research to fill the information gap and permit adequate conservation of these sensitive fishes. Monitoring of syngnathids, however, is considered difficult due to their cryptic nature and sedentary behaviour. Indeed, available data on diversity,

distribution, threats, and conservation status in the Mediterranean are mostly fragmented, and an updated overview is required to facilitate the identification of research and conservation priorities. Although syngnathids act as flagship species of the marine conservation effort as well as indicators of the health and diversity of coastal ecosystems [13], most of the research in the Mediterranean Sea has referred to seahorses on a local scale, e.g., [12,14–17], while studies on pipefishes are lacking and have been mostly limited to generic biodiversity projects and lifecycle studies [18–20]. Only a few studies have specifically focused on evaluating pipefish population dynamics, ecology, and spatial distribution [19,21], but most have referred to Turkey [22,23], Tunisia [24], and Spain [25], with only a few studies on the northern Italian Adriatic coast [19,21,26,27].

In this paper, we report, for the first time, the occurrence of a structured assemblage of four syngnathid species in Sabaudia Lake to determine their distribution and abundance while also investigating habitat association. The collected data will aid future population assessments at this site and provide valuable hints for future conservation efforts for these species. Estimating the conservation status of populations could benefit from knowledge about their distribution [8,28]. Effective conservation and management plans for syngnathid populations require improved and accurate information about their current spatial distribution and preferred habitats.

2. Materials and Methods

Sabaudia Lake (Figure 1), also known as Paola Lake (41°16′50″ N, 13°02′40″ E), is the largest of four coastal lakes within Circeo National Park (CNP) in Latium, Italy. The lake is designated as a Site of Community Importance (SCI IT6040013) and a Special Area of Conservation (SAC) under Habitat Directive 92/43/EEC. It has a long, narrow shape with a central section oriented in a northwest–southeast direction and five transversal branches that bring freshwater inputs from the surrounding landscape through small tributaries. Despite its protected status, the lake has faced various human impacts in recent decades, such as sewage discharge, fishing, tourism, boat traffic, and mariculture (including shellfish harvesting) [29,30]. The lake is connected to the sea via two tidal inlets, the Roman Channel (RC) on the southern side and the Caterattino Channel (CC) on the northern side, allowing for water exchange between the lake and the sea.

The lake mouth areas of both the primary and secondary channels are regularly influenced by lake and seawater flows according to the tidal phase, although the CC is frequently barred by sandy mounds that prevent the continuity of lake–sea exchanges.

As in most shallow Mediterranean coastal lagoons, hydrodynamic processes at this site are driven by the combined effects of the tide and wind, which promote horizontal advection and vertical turbulent diffusion [31–33]. The spatial distribution of macrobenthic communities varies seasonally according to the hierarchy of forcing environmental conditions [29]. The spatial distribution and biodiversity of molluscan assemblages mirror the levels of the tide- and wind-induced kinetic energy; indeed, maximum species diversity is observed where energy levels are high (RC inlets area with a dominance of coarse sediment and low depth), and diversity significantly declines in landward positions where there is a lower level of kinetic energy, associated with the dominance of fine sediments [29]. The decreasing trend of species richness from the inlet mouth to the inner lagoon zones agrees with the "confinement gradient" [29,34,35], suggesting that RC inlets, rather than CC inlets, can be considered zones of maximum spatial extension of marine influence.
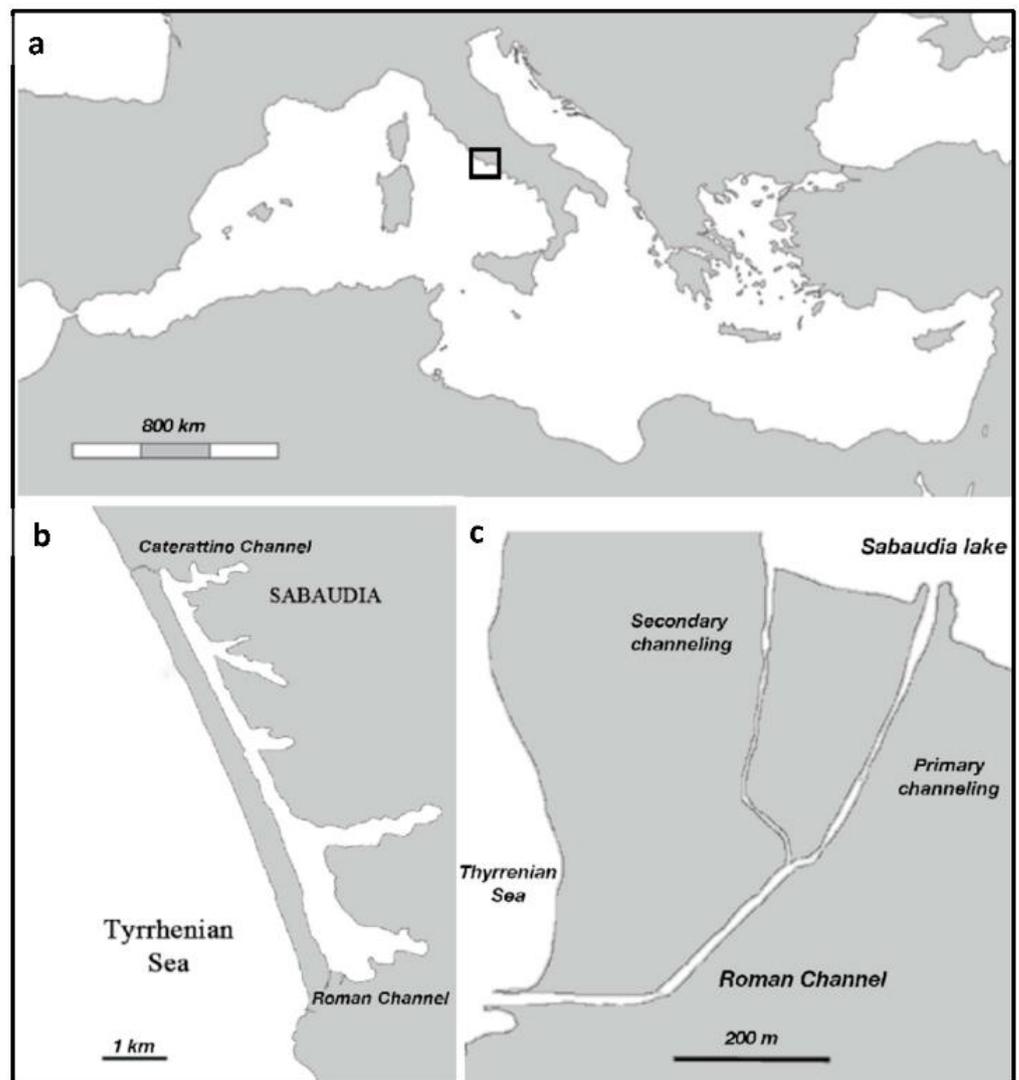
Figure 1. Study area. Sabaudia Lake, located in central Italy, Tyrrhenian Sea (**a**), is connected with the sea by two channels (**b**); details of Roman Channel and its primary and secondary channellings (**c**).

2.1. Sampling Stations and Data Collection

A preliminary survey to assess the presence of syngnathids was conducted across Sabaudia Lake, including areas of the Roman Channel and artificial walls around the sea mouth. A subsequent fieldwork study was carried out in March 2022 by four experienced operators while scuba diving and snorkelling. According to the high degree of ecosystem patchiness, 11 stations (Figure 2) were selected and monitored using a standard underwater census (UVC) [36] carried out on two standard linear 50 m transects. For each sighted syngnathid, the species, sex, total length, and sexual maturity stage according to [37] were recorded in situ. To retrieve additional biometric data, encountered specimens were photographed following Curtis et al. [38]. Information on salinity and temperature, as well as hydrodynamism and the influence of the sea on the ecological dynamics of the lagoon, were obtained from [29]. Ecological characterisation of the sampling stations, including descriptions of substrates and species, was performed following [29]. In cases of doubt regarding habitat-forming or dominant species identification, samples were taken, fixed in 96% ethanol, and then analysed in the laboratory.
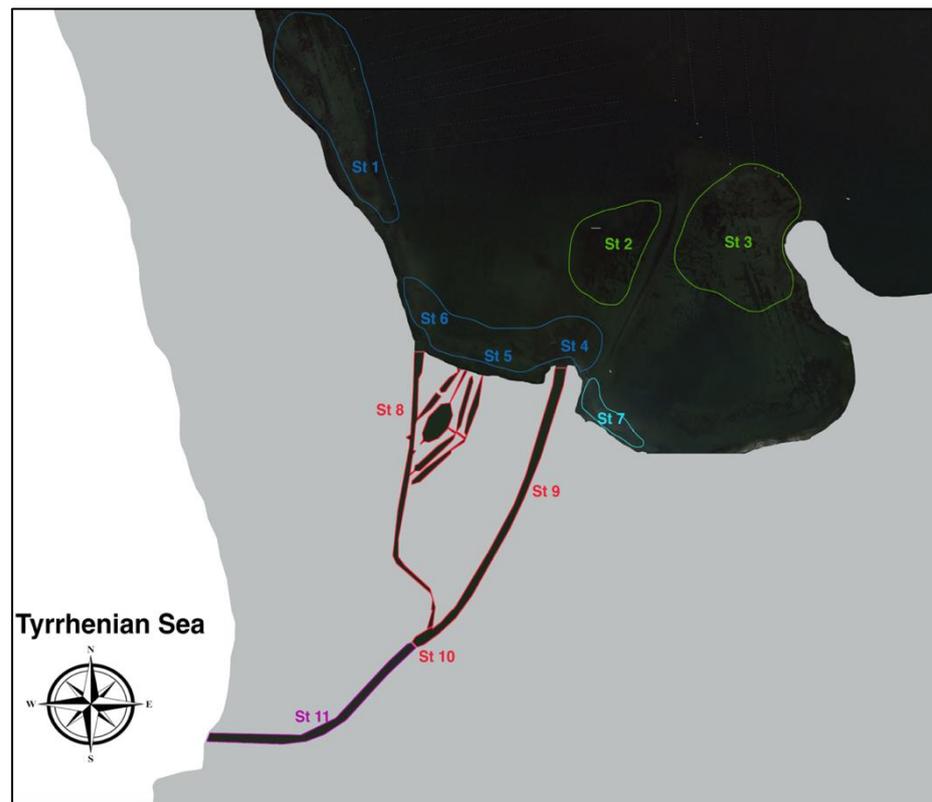
Figure 2. Aerial view of sampling stations (St 1–St 11) and distribution of different habitats. Green: muddy bottom with seagrass and algae; blue: *Cymodocea nodosa* meadows; light blue: dense colonial ascidians populations with low hydrodynamism; red: composed macro-invertebrate guilds with medium–high hydrodynamism; violet: predominantly marine habitat with high hydrodynamism.

2.2. Data Analysis

Differences in syngnathid assemblages composition at each station were assessed using an nMDS analysis of the abundance matrix based on square root transformed data. Five areas were identified in relation to their position with respect to communication channels with the sea, namely (A) the innermost parts of the area influenced by tidal currents (St 1–3), (B) inlet areas of different channellings (St 4–6), (C) an internal and protected area almost excluded from tidal influence (St 7), (D) a secondary channelling and its starting point (St 8 and 9), and (E) the Roman channel (St 10 and 11). To test the significance of the observed differences among stations, ANOVA was performed on the abundance matrix using the same experimental design. Data were analysed using the Bray-Curtis dissimilarity measure on square root transformed data with 9999 permutations of residuals under a reduced model. All analyses were performed using PRIMER 6.0 software.

3. Results

3.1. Habitat Characterisation

The average depth was approximately 1.5 m, with a maximum of 3.5 m at St 3 and St 10 and a minimum of 0.5 m at St 1, 4, and 5 and St 8–11 (Table 1). Habitat characterisation (Table 1) revealed the widespread presence of muddy bottoms often associated with fine vegetal deposits, consisting mainly of *Posidonia oceanica* rhizomes fibres, with *Chaetomorpha* sp. beds in the inner lake parts (Figure 2; St 1–3, 5, 7). *C. nodosa* meadows occurred sparsely in most areas near RC inlets in association with unconsolidated bottoms. The RC presented two well-defined and distinct aspects: shallow sandbars with high tidal currents at St 11 and sparse rocks and coarse shell-bearing sands associated with terrestrial and marine plant detritus, including tree branches, *P. oceanica* rhizomes, and detached thalli of *Codium* spp., at St 10. These sites were impacted by litter items mainly originating

from marine inputs; the same items were also observed in the primary and secondary channels (St 8, 9). As displayed by macrobenthic assemblage and seabed granulometry, St 3 and St 7 were the most influenced by lake conditions. Although St 3 was located approximately 600 m from the RC mouth (Figure 2), it was more affected by marine waters than St 7, as inferred from the presence of scattered *C. nodosa* meadows (Figure 3).

Figure 3. *Cymodocea nodosa* (**above**) and detrital bottom (**below**), with *Hippocampus hippocampus* and *H. guttulatus* recorded, respectively.

Table 1. Description of habitats, depth, and encountered syngnathids at the eleven investigated stations.

Station	Habitat	Depth	Syngnathid Species
St 1	Muddy–sandy seabed with patchy meadows of *C. nodosa* and *Chaetomorpha* spp. and abundant fine vegetal detritus. Dense population of *Cerithium vulgatum* and *Hexaplex trunculus*. The site is weakly affected by tidal streams, with hydrological features mainly referring to the lagoon ecosystem.	0.5–1.5 m	*Hippocampus hippocampus* *Nerophis ophidion* *Syngnathus abaster*
St 2	Muddy seabed with stable occurrence of *Chaetomorpha* spp. Patchy meadows of *C. nodosa* were also present. Structured macrobenthic community with dense populations of *H. trunculus*, *Haminoea japonica,* and *Gammarus insensibilis.* The site is not affected by tidal streams, with hydrological features mainly referring to the lagoon ecosystem.	1–3 m	*N. ophidion* *S. abaster*
St 3	Muddy seabed with stable occurrence of *Chaetomorpha* spp. Patchy meadows of *C. nodosa* were also present. Structured macrobenthic community with dense populations of *H. trunculus*, *H. japonica,* and *G. insensibilis.* The site is not affected by tidal streams, with hydrological features mainly referring to the lagoon ecosystem.	1–3.5 m	*N. ophidion* *S. abaster*
St 4	Muddy–sandy seabed with *C. nodosa* meadows extending to the whole channel mouth area with abundant fine vegetal detritus. Dense population of *Cerithium vulgatum* and *H. trunculus*. Strong tidal flow with hydrological features changing according to tidal phases.	0.5–1.5 m	*H. guttulatus* *H. hippocampus* *N. ophidion* *S. abaster*
St 5	Muddy–sandy seabed with patchy meadows of *C. nodosa and Chaetomorpha* sp. Dense populations of *C. vulgatum*. and *H. trunculus* be found, along with a more complex malacocenosis. The site is weakly affected by tidal streams, with hydrological features mainly referring to the lagoon ecosystem.	0.5–2 m	*H. hippocampus* *N. ophidion* *S. abaster*
St 6	Muddy–sandy seabed with patchy meadows of *C. nodosa* and abundant fine vegetal detritus. Dense populations of *C. vulgatum*. and *H. trunculus* were found. Moderate tidal flow with hydrological features changing according to tidal phases.	1–2.5 m	*H. guttulatus* *H. hippocampus* *N. ophidion* *S. abaster*
St 7	Muddy seabed with seasonal occurrence of algal coverage (*Ulva* spp., *Chaetomorpha* spp.). Poor seabed macrobenthic community. Dense population of *Ciona intestinalis, Styela plicata, Aiptasia mutabilis,* and *Ficopomatus enigmaticus* colonise artificial rocky substrates. The site is not	1–2 m	

	affected by tidal streams, with hydrological features mainly referring to the lagoon ecosystem.		
St 8	Sandy bottom with a thick coverage of organic matter mainly composed of *P. oceanica* leaves and rhizome fibres. Tidal flow is weaker than at Station 9.	0.5–1 m	*H. guttulatus* *H. hippocampus*
St 9	Sandy bottom with scattered accumulation of vegetal detritus, gravel, and compacted organic matter. Closer to the inner mouth, the channel is affected by a sparsely packed grassland of *C. nodosa*. Strong tidal flow with hydrological features changing according to tidal phases.	0.5–2 m	*H. guttulatus* *H. hippocampus*
St 10	Coarse shell-bearing sands with terrestrial and marine vegetal detritus, including tree branches, *P. oceanica* rhizomes, and detached thalli of *Codium* spp. High seabed slope, reaching 3.5 m in depth in the central part. Strong tidal flow velocity leaves large portions of rocks uncovered. Hydrological features change according to tidal phases.	0.5–3.5 m	
St 11	Coarse sandy bottom with scattered vegetal detritus, without areas of accumulation. Hard substrates colonised by dense populations of anthozoans (*Anemonia viridis*, *Paranemonia cinerea*). Strong tidal flow velocity. Hydrological features change according to tidal phases.	0.5–1.5 m	

St 7, located in a bend on the southern basin shore, was excluded from the direct influence of lake–sea water exchanges and presented a muddy seabed. The presence of *Chaetomorpha* spp. and *Ulva* spp. supported eutrophic conditions as a direct consequence of the weak hydrodynamics of the water column.

The values of salinity and temperature mostly referred to marine and lagoon environments according to the tidal phase, with T °C = 18.8 °C and salinity = 37.8 psu during high tide and T °C= 14.9 °C and salinity = 28.2 psu during low tide for all stations, except for St 3 and 7, where T °C= 14 ° C and salinity = 23 psu.

Hydrodynamism promoted by sea–lake tidal exchanges reached its maximum values at St 11, where RC had the lowest depth and broadness. Near St 10, this channel widened, and the current energy progressively decreased towards the lake.

3.2. Syngnathid Population

A total of 125 individuals belonging to four species (corresponding to 27% of the syngnathid species recorded for the Mediterranean) were observed: 11 (2F + 9M) *Hippocampus guttulatus*, 59 (20F + 39M) *H. hippocampus*, 19 (2F + 17M) *Nerophis ophidion*, and 37 (8F + 29M) *Syngnathus abaster*.

The sex ratio was in favour of males in all species. The total length (TL) of *H. guttulatus* and *H. hippocampus* ranged from 7 to 13.5 cm, while the lengths of *N. ophidion* and *S. abaster* varied between 11 and 15.5 cm and 9.5 and 13 cm, respectively (Figure 4).

N. ophidion had the widest distribution, being found in the innermost area (St 1, St 2, and St 3) in co-occurrence with *S. abaster* as well as near the two inlet areas of the primary and secondary channels (St 4, St 5, and St 6). At St 8 and St 9, only *H. hippocampus* and *H.*

guttulatus were found. No syngnathid species were recorded at St 7 (located in a bend of the basin), St 10, or St 11 (directly influenced by marine inputs) (Figure 5).

ANOVA revealed notable variations in the composition and relative abundance of communities at different stations. At the innermost stations specifically, only pipefishes were recorded, while the estuary contained both pipefishes and seahorses; in the channels, only seahorses were observed. These dissimilarities can be observed in the graphical representation of the populations (Figure 6) based on the same similarity matrix.
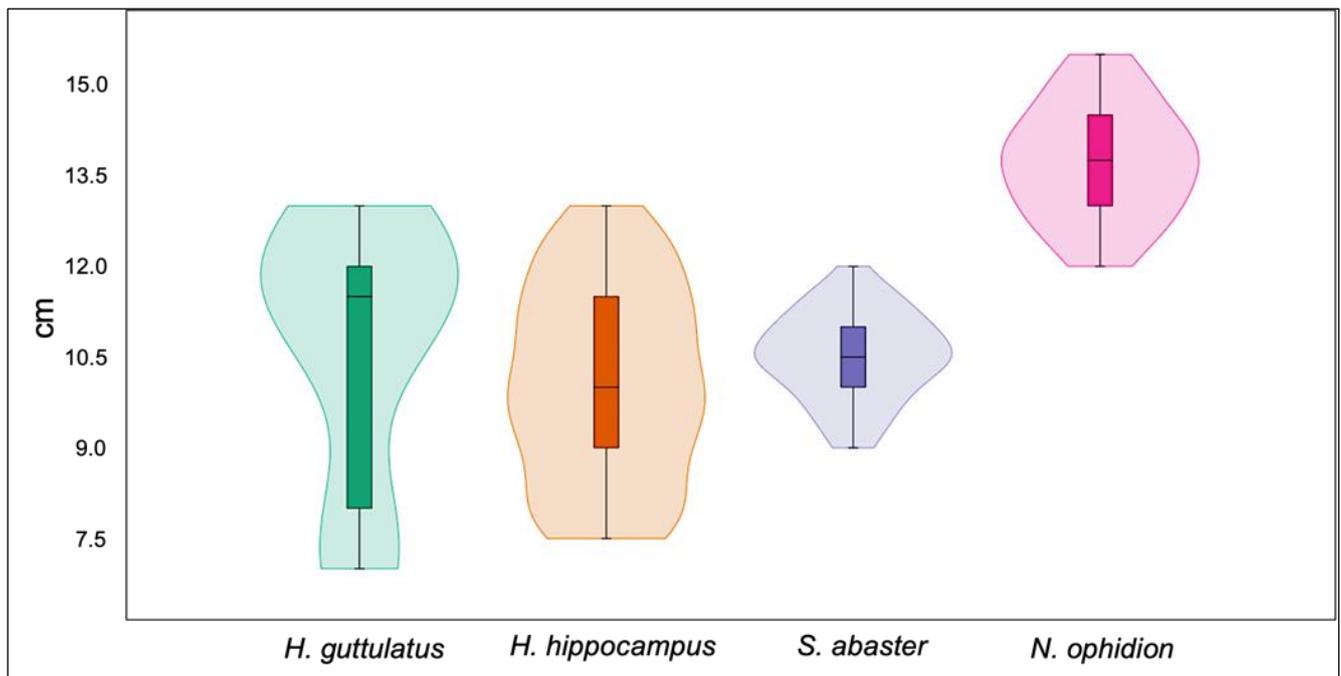

Figure 4. Size distribution of *Hippocampus guttulatus*, *H. hippocampus*, *Syngnathus abaster*, and *Nerophis ophidion*.
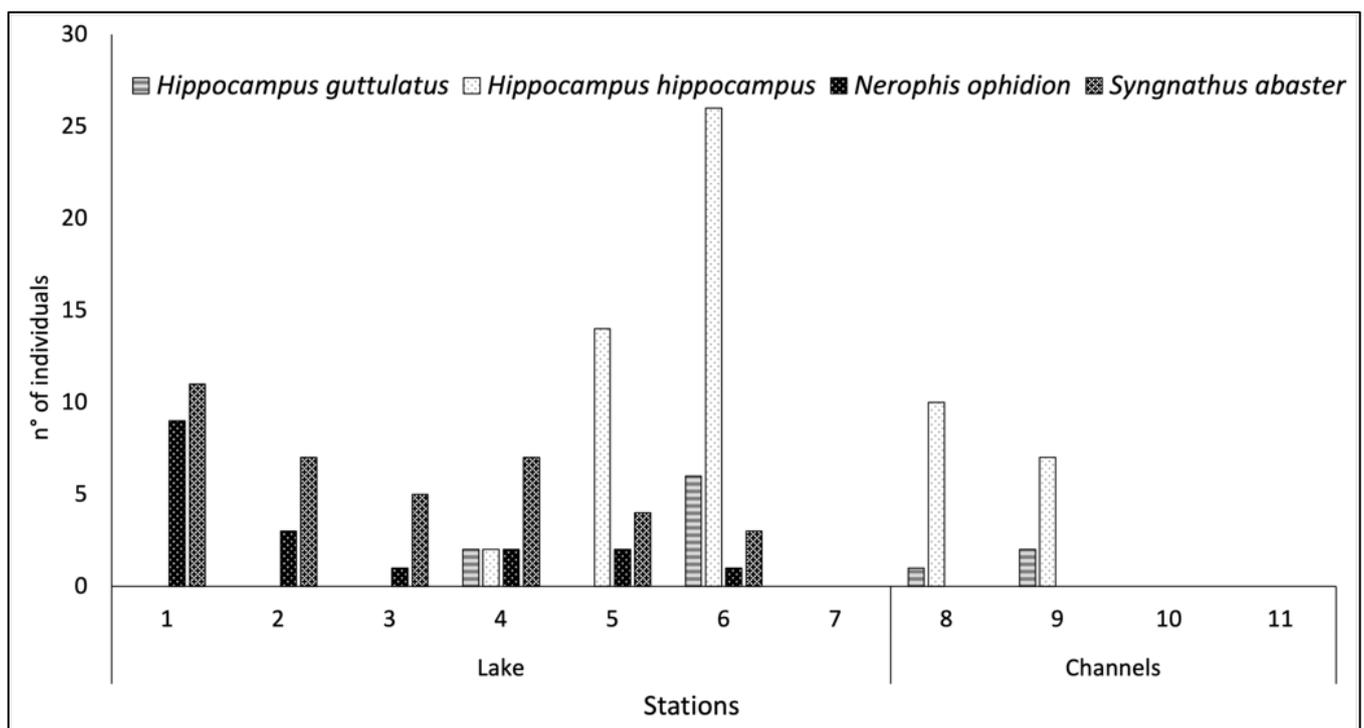

Figure 5. Number of syngnathid species at each sampling station within the lake and channels.
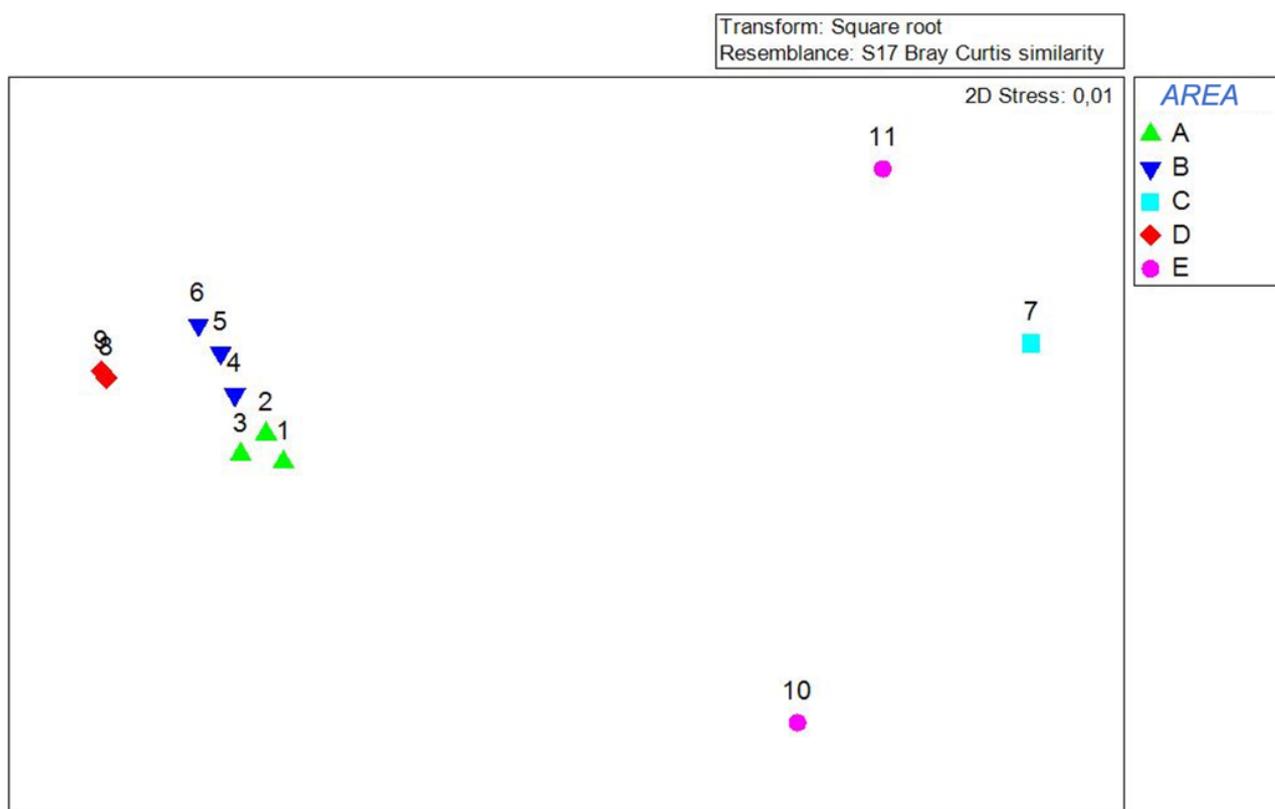
Figure 6. nMDS graphic representation of recorded syngnathids. Numbers indicate sampling stations; letters indicate the following habitats: A: lake habitat with muddy bottom with seagrass and algae; B: *C. nodosa* habitat near the channel's mouth; C: habitats with colonial ascidians and little or no hydrodynamism; D: habitat with macro-invertebrates with medium–high hydrodynamism; E: predominantly marine habitat with high hydrodynamism.

4. Discussion

This study provides the first evidence of the occurrence of *Hippocampus guttulatus*, *H. hippocampus*, *Syngnathus abaster*, and *Nerophis ophidion* in Sabaudia Lake. The number of individuals of four species was relatively high considering the extension of investigated habitats, and the observed scattered distribution is consistent with that observed in previous studies e.g., [6,16,39,40], suggesting that syngnathids display patchy distributions even on small spatial scales. It should be noted, however, that the numbers recorded in this work are conservative estimates of the species distributions in habitats and are certainly strongly influenced by the sampling method used. The need to standardise sampling efforts led to the choice of the underwater visual transects technique [36], which seems the most suitable for the census of seahorse populations, but it is probably less suitable for pipefishes; such a choice could have led to the underestimation of pipefish abundance, as these fishes are more difficult to observe in nature with respect to seahorses due to their morphology, preference towards vegetated habitats, and often small body dimensions. Indeed, the use of manual instruments (such as hand nets or beach seines) in the upper infralittoral plane may offer more realistic estimates of pipefish species and abundance but could heavily underestimate the number of observed seahorses, with values often reduced to zero (Pierri and Ravisato, unpublished data).

The coastal systems in which syngnathids usually occur are particularly vulnerable to anthropogenic factors [5,41,42]. All species are currently listed on the IUCN Red List of Threatened Species [13], and most of the species have been listed as data-deficient (eight out of ten in European waters; see Supplementary Material Table S1) with special concerns for seahorses, freshwater pipefishes, and estuarine species. Sensitivity to human-related

disturbances is enhanced by their unique life cycle traits, including low mobility, site fidelity, and lengthy parental care [8,14,16,17,43,44]. As highlighted by IUCN World Conservation Congress Resolution 95 (WCC-2020-Res-095), syngnathid populations should be further protected, as human activities together with climate changes are causing widespread degradation of these species' habitats on a worldwide level, while also underlining the need to enhance available knowledge and thus promote conservation actions. Globally, most of the current information is based on studies on a few populations that have been extensively researched, e.g., [17,20,45–52]. Most of this attention, however, has been dedicated to seahorses, and it was gained after severe declines in populations [7,15,51,53–56] whose causes are still not fully elucidated [12,15,51]. The presence of syngnathids represents a good indicator of the ecological status of habitats and could provide ideas for new models for validating the environmental quality of water bodies that have been historically subjected to anthropogenic pressures, such as confined areas. Indeed, the IUCN World Conservation Congress 2020 highlighted the fundamental role of syngnathids as sentinel organisms of structural environmental variations.

According to the available literature, it seems that the highest syngnathid abundances are reached in confined environments, including lagoons and estuaries [43,57,58]. Although it is still not clear which are the abiotic and biotic factors that ensure their presence, it is known that seahorses have a somewhat restricted optimum range for several environmental parameters [59,60], while pipefishes seem to have a wider tolerance to, for instance, salinity values [61–65]. The reliance of syngnathids on seagrass structural complexity as a form of shelter against hydrodynamic stress was recently suggested [65]; indeed, it seems that the deterioration of these habitats can have a significant effect on the energy expenditure of these fishes. Global decline, fragmentation, and loss of structural complexity in biogenic habitats significantly contribute to a reduction in hydrodynamically sheltered habitats, thus resulting in increased energy expenditure among associated fauna and possibly contributing to population declines [66]. The observed distribution of pipefishes in the study area highlights the role of habitat complexity in supporting local populations, in contrast to *Hippocampus* spp., which seem to have a greater tolerance to kinetic features. According to the results, seahorses were found at the most hydrodynamic stations (St 8, St 9) and seem to display a stronger dependency on holdfasts (vegetal detritus) [16,67] than habitat complexity. Different habitat complexities may also result in different benthic communities; thus, the role of trophic niche partitioning in describing species distributions cannot be excluded [45,68,69].

As for most Mediterranean lagoons, the morphology of Sabaudia Lake inlets is artificially modified, with the functionality of tidal channels depending upon maintenance and dredging to contrast their silting up. The missed management of artificial inlets due to the reduction in fishing efforts using inlet fish barriers has led to a reduction in the exchanges with the sea and a consequent reduction in marine migrants during the winter season. This has resulted in adverse effects on the water quality of the entire lagoon [30]; it has also promoted the massive intrusion of marine sediments and organic detritus in the inner mouths, boosting habitat fragmentation. The patchy distribution of *Cymodocea nodosa* meadows in the studied area suggests a possible effect of strong marine intrusion on its integrity.

The mean size of the recorded fishes in this study was within the range documented for other areas in the Mediterranean Sea [22–25,70]. According to the results, the sex ratio was slightly in favour of males; it was suggested that there may be a different ratio of mortality between males and females, as males may be more susceptible to predation when brooding [71], although this does not seem to be the case in Sabaudia.

H. hippocampus appeared to be the most common species, while *H. guttulatus* was relatively rare; such a species ratio is somewhat peculiar, as *H. hippocampus* is usually much less abundant when co-occurring [12,15,39,49]. However, such a ratio might be explained by the higher availability of low-complexity and muddy habitats, which seem to be preferred by *H. hippocampus* [12,72]. One of the recorded species, *N. ophidion*, was reported to

be common throughout its global range, but abundance estimates are scattered and rarely recorded in Italian waters [21,73]. Another pipefish species, *S. abaster*, is considered one of the most representative European syngnathids [74,75]. The available data on syngnathid populations in the Mediterranean Sea indicate spatial variability in distribution and abundance [21,75] and the apparent preference of these fishes for confined areas [5,7,39,42,70,76,77]. Such variability could be influenced by the extension and integrity of suitable habitats and the structure of epifauna assemblages [14,69,75,78] in correlation with the physical characteristics of the site [79]; however, their presence within virtually suitable confined areas cannot be considered a rule [16].

The present study is the first to describe the distribution and abundance of four syngnathids species at Sabaudia Lake and investigate their habitat preferences. The results and previous observations (Macali, unpublished data) indicate that these populations are temporarily stable. The conservation of syngnathids should be a priority for biological and ecological reasons and their intrinsic value [5,80]. Future research should aim at the year-round characterisation of syngnathids in Sabaudia Lake to assess eventual abundance changes, as observed in many other localities across their geographical range [19,68]. Furthermore, future research should also be oriented towards locating and describing hitherto unknown populations, which would be valuable for understanding large-scale distribution and ecology and could help preserve emblematic syngnathids.

Author Contributions: Conceptualisation, G.C. and A.M.; investigation and methodology, A.M., M.G., C.P., M.F.G. and T.L.; formal analysis and data curation, A.M. and C.P., writing—original draft preparation, A.M., C.P., T.L., M.R., M.F.G. and M.G.; writing—review and editing, A.M., C.P., T.L., M.R. and M.G. All authors have read and agreed to the published version of the manuscript.

Institutional Review Board Statement: Not applicable.

Data Availability Statement: The authors are available to share the data collected upon justified request to the corresponding author.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Kerr, J.T.; Currie, D.J. Effects of human activity on global extinction risk. *Conserv. Biol.* **1995**, *9*, 1528–1538. https://doi.org/10.1046/j.1523-1739.1995.09061528.x.
2. Lotze, H.K.; Lenihan, H.S.; Bourque, B.J.; Bradbury, R.H.; Cooke, R.G.; Kay, M.C.; Kidwell, S.M.; Kirby, M.X.; Peterson, C.H.; Jackson, J.B. Depletion, degradation, and recovery potential of estuaries and coastal seas. *Science* **2006**, *312*, 1806–1809. https://doi.org/10.1126/science.1128035.
3. Crain, C.M.; Kroeker, K.; Halpern, B.S. Interactive and cumulative effects of multiple human stressors in marine systems. *Ecol. Let.* **2008**, *11*, 1304–1315. https://doi.org/10.1111/j.1461-0248.2008.01253.x.
4. Crain, C.M.; Halpern, B.S.; Beck, M.W.; Kappel, C.V. Understanding and managing human threats to the coastal marine environment. *Ann. NY Acad. Sci.* **2009**, *1162*, 39–62. https://doi.org/10.1111/j.1749-6632.2009.04496.x.
5. Pierri, C.; Lazic, T.; Gristina, M.; Corriero, G.; Sinopoli, M. Large-scale distribution of the European seahorses (*Hippocampus* Rafinesque, 1810): A systematic review. *Biology* **2022**, *11*, 325. https://doi.org/10.3390/biology11020325.
6. Lim, A.; Chee, O.; Chong, V.; Wong, C.; Choo, C.K. Diversity, habitats and onservation Threats of Syngnathid (Syngnathidae) Fishes in Malaysia. *Trop. Zool.* **2011**, *24*, 193–222.
7. Vincent, A.C.J.; Foster, S.J.; Koldewey, H.J. Conservation and management of seahorses and other Syngnathidae. *J. Fish Biol.* **2011**, *78*, 1681–1724. https://doi.org/10.1111/j.1095-8649.2011.03003.x.

8. Claassens, L.; Hodgson, A.; Short, G.; Harasti, D. Diversity, distribution, ecology and conservation status of the family Syngnathidae in sub-Saharan Africa and adjacent islands. In *Oceanography and Marine Biology: An Annual Review*, 1st ed.; CRC Press: Boca Raton, FL, USA, 2022; Volume 60; pp. 169–242; ISBN 978-1-00-328860-2.

9. Olden, J.D.; Hogan, Z.S.; Zanden, M.J.V. Small fish, big fish, red fish, blue fish: Size-biased extinction risk of the world's freshwater and marine fishes. *Global Ecol. Biogeogr.* **2007**, *16*, 694–701. https://doi.org/10.1111/j.1466-8238.2007.00337.x.

10. Vincent, A.C.J. *The International Trade in Seahorses*; Species in Danger; Traffic International: Cambridge, UK, 1996; ISBN 9781858500980.

11. Salin, K.R.; Yohannan, T.M.; Mohanakumaran Nair, C. Fisheries and Trade of Seahorses, *Hippocampus* spp., in Southern India. *Fish. Manag.* **2005**, *12*, 269–273. https://doi.org/10.1111/j.1365-2400.2005.00450.x.

12. Correia, M.; Paulo, D.; Samara, E.; Koulouri, P.; Mentogiannis, V.; Dounas, C. Field studies of seahorse population density, structure and habitat use in a semi-closed north-eastern Mediterranean marine area (Stratoni, North Aegean Sea). *J. Fish Biol.* **2020**, *97*, 314–317. https://doi.org/10.1111/jfb.14371.

13. Iucn, Red List of Threatened Species. 2022. Available online: http://www.iucnredlist.org/ (accessed on 15 March 2023).

14. Curtis, J.M.R.; Vincent, A.C.J. Life history of an unusual marine fish: Survival, growth and movement patterns of *Hippocampus guttulatus* Cuvier 1829. *J. Fish Biol.* **2006**, *68*, 707–733. https://doi.org/10.1111/j.0022-1112.2006.00952.x.

15. Correia, M.; Caldwell, I.R.; Koldewey, H.J.; Andrade, J.P.; Palma, J. Seahorse (Hippocampinae) population fluctuations in the ria Formosa lagoon, south Portugal. *J. Fish Biol.* **2015**, *87*, 679–690. https://doi.org/10.1111/jfb.12748.

16. Lazic, T.; Pierri, C.; Gristina, M.; Carlucci, R.; Cardone, F.; Colangelo, P.; Desiderato, A.; Mercurio, M.; Bertrandino, M.S.; Longo, C.; et al. Distribution and habitat preferences of *Hippocampus* Species along the Apulian Coast. *Aquat. Conserv.* **2018**, *28*, 1317–1328. https://doi.org/10.1002/aqc.2949.

17. Pierri, C.; Lazic, T.; Corriero, G.; Cardone, F.; Onen Tarantini, S.; Desiderato, A.; Mirto, S.; Gristina, M. Site fidelity of *Hippocampus Guttulatus* Cuvier, 1829 at Mar Piccolo of Taranto (Southern Italy; Ionian Sea). *Environ. Biol. Fish.* **2020**, *103*, 1105–1118. https://doi.org/10.1007/s10641-020-01008-0.

18. Campolmi, M.; Franzoi, P.; Mazzola, A. Observations on pipefish (Syngnathidae) biology in the Stagnone Lagoon (West Sicily). *Publicaciones Espec. Inst. Esp. Oceanogr.* **1996**, *21*, 205–209.

19. Franzoi, P.; Maccagnani, R.; Rossi, R.; Ceccherelli, V. Life-Cycles and Feeding-Habits of *Syngnathus taenionotus* and *S. Abaster* (Pisces, Syngnathidae) in a Brackish Bay of the Po River Delta (Adriatic Sea). *Mar. Ecol. Prog. Ser.* **1993**, *97*, 71–81. https://doi.org/10.3354/meps097071.

20. Sanna, D.; Biagi, F.; Ben Alaya, H.; Maltagliati, F.; Addis, A.; Romero, A.; De Juan, J.; Jean-pierre, Q.; Castelli, A.; Franzoi, P.; et al. Mitochondrial DNA Variability of the Pipefish *Syngnathus Abaster*. *J. fish Biol.* **2013**, *82*, 856–876. https://doi.org/10.1111/jfb.12027.

21. Scapin, L.; Cavraro, F.; Malavasi, S.; Riccato, F.; Zucchetta, M.; Franzoi, P. Linking Pipefishes and Seahorses to Seagrass Meadows in the Venice Lagoon: Implications for Conservation. *Aquat. Conserv.* **2018**, *28*, 282–295. https://doi.org/10.1002/aqc.2860.

22. Gürkan, Ş. Investigations on the Ecomorphologic Characteristics of the Pipefish (Familia: Syngnathidae) Distributing in the Çamalti Lagoon (İzmir Bay). Ph.D. Thesis, Ege University, Bornova, İzmir, 2004; Volume 215.

23. Gürkan, S.; Taskavak, E. Length-Weigth Relationships for Syngnathid Fishes of the Aegean Sea, Turkey. *Belg. J. Zool.* **2007**, *137*, 227–242.

24. Amor, M.M.; Salem, M.; Reynaud, C.; Capapé, C. Length–Weight Relationships in Syngnathid Species from Tunisian Waters (Central Mediterranean). *Mar. Biodivers. Rec.* **2011**, *4*, e44. https://doi.org/10.1017/S1755267211000303.

25. Planas, M.; Piñeiro-Corbeira, C.; Bouza, C.; Castejon, I.; Vera, M.; Regueira, M.; Ochoa, V.; Bárbara, I.; Terrados, J.; Chamorro Valverde, A.; et al. A Multidisciplinary Approach to Identify Priority Areas for the Monitoring of a Vulnerable Family of Fishes in Spanish Marine National Parks. *BMC Evol. Biol.* **2021**, *21*, 77–92.

26. Franzoi, P.; Franco, A.; Torricelli, P. Fish Assemblage Diversity and Dynamics in the Venice Lagoon. *Rend. Fis. Acc. Lincei* **2010**, *21*, 269–281. https://doi.org/10.1007/s12210-010-0079-z.

27. Malavasi, S.; Franco, A.; Riccato, F.; Valerio, C.; Torricelli, P.; Franzoi, P. Habitat Selection and Spatial Segregation in Three Pipefish Species. *Estuar. Coast. Shelf S.* **2007**, *75*, 143–150. https://doi.org/10.1016/j.ecss.2007.02.022.

28. Hare, M.P.; Nunney, L.; Schwartz, M.K.; Ruzzante, D.E.; Burford, M.; Waples, R.S.; Ruegg, K.; Palstra, F. Understanding and Estimating Effective Population Size for Practical Application in Marine Species Management: Applying Effective Population Size Estimates to Marine Species Management. *Conserv. Biol.* **2011**, *25*, 438–449. https://doi.org/10.1111/j.1523-1739.2010.01637.x.

29. Macali, A.; Conde, A.; Smriglio, C.; Mariottini, P.; Crocetta, F. The Evolution of the Molluscan Biota of Sabaudia Lake: A Matter of Human History. *Sci. Mar.* **2013**, *77*, 649–662. https://doi.org/10.3989/scimar.03858.05M.

30. Zucchetta, M.; Capoccioni, F.; Franzoi, P.; Ciccotti, E.; Leone, C. Fish Response to Multiple Anthropogenic Stressors in Mediterranean Coastal Lagoons: A Comparative Study of the Role of Different Management Strategies. *Water* **2021**, *13*, 130. https://doi.org/10.3390/w13020130.

31. Millet, B.; Guelorget, O. Spatial and Seasonal Variability in the Relationships between Benthic Communities and Physical Environment in a Lagoon Ecosystem. *Mar. Ecol. Prog. Ser.* **1994**, *108*, 161–174. https://doi.org/10.3354/meps108161.

32. Perdicaro, R. Osservazioni sulle condizioni idrologiche del Lago di Sabaudia o di Paola nel biennio 1982-1983. *Quad. Ist.Idrobiol. Aquacolt. Brunelli* **1983**, *3*, 59–72.

33. Perdicaro, R. Condizioni chimico-fisiche del Lago di Sabaudia o di Paola dal 1933 al 1984. *Quad. Ist. Idrobiol. Aquacolt.Brunelli* **1984**, *4*, 71–139.

34. Guélorget, O.; Perthuisot, J.P. Indicateurs Biologiques et Diagnose Écologique Dans Le Domaine Paralique. *Bull. Ecol.* **1984**, *15*, 67–76.

35. Guélorget, O.; Perthuisot, J.P. Paralic Ecosystems Biological Organization and Functionning. *Vie et Milieu* **1992**, *42*, 215–251.

36. Samoilys, M. Underwater Visual Census Surveys. *Manual for Assessing Fish Stocks on Pacific Coral Reefs*; Queensland Department of Primary Industries: Brisbane, Training Series QE97009, 1997; pp. 16–29.

37. Gürkan, S.; Taylan, B. The first observations of oocyte diameters for the pipefish from the southern Black Sea. *J. Black Sea/Mediterr. Environ.* **2013**, *19*, 206–216.

38. Curtis, J.; Moreau, M.-A.; Marsden, D.; Bell, E.; Martin-Smith, K.; Samoilys, M.; Vincent, A. Underwater Visual Census for Seahorse Population Assessments. Project Seahorse, University of British Columbia, Vancouver 2004.

39. Curtis, J.; Vincent, A. Distribution of Sympatric Seahorse Species along a Gradient of Habitat Complexity in a Seagrass-Dominated Community. *Mar. Ecol. Prog. Ser.* **2005**, *291*, 81–91. https://doi.org/10.3354/meps291081.

40. Perera, N.; Dahanayaka, D.D.G.L.; Udagedara, S. Habitat Preference and Population Structure of Two Data Deficient Seahorse (Syngnathidae) Species. *OUSL Jnl* **2017**, *12*, 75. https://doi.org/10.4038/ouslj.v12i2.7408.

41. Fagan, W.F. Connectivity, fragmentation, and extinction risk in dendritic metapopulations. *Ecology* **2002**, *83*, 3243–3249. https://doi.org/10.1890/0012-9658(2002)083[3243:CFAERI]2.0.CO;2.

42. Hernández-Urcera, J.; Murillo, F.J.; Regueira, M.; Cabanellas-Reboredo, M.; Planas, M. Preferential Habitats Prediction in Syngnathids Using Species Distribution Models. *Mar. Environ. Res.* **2021**, *172*, 105488. https://doi.org/10.1016/j.marenvres.2021.105488.

43. Rosa, I.L.; Oliveira, T.P.R.; Castro, A.L.C.; Moraes, L.E.D.S.; Xavier, J.H.A.; Nottingham, M.C.; Dias, T.L.P.; Bruto-Costa, L.V.; Araújo, M.E.; Birolo, A.B.; et al. Population Characteristics, Space Use and Habitat Associations of the Seahorse *Hippocampus Reidi* (Teleostei: Syngnathidae). *Neotrop. Ichthyol.* **2007**, *5*, 405–414. https://doi.org/10.1590/S1679-62252007000300020.

44. Nester, G.M.; De Brauwer, M.; Koziol, A.; West, K.M.; DiBattista, J.D.; White, N.E.; Power, M.; Heydenrych, M.J.; Harvey, E.; Bunce, M. Development and Evaluation of Fish eDNA Metabarcoding Assays Facilitate the Detection of Cryptic Seahorse Taxa (Family: Syngnathidae). *Environ. DNA* **2020**, *2*, 614–626. https://doi.org/10.1002/edn3.93.

45. Vizzini, S.; Mazzola, A. The trophic structure of the pipefish community (Pisces: Syngnathidae) from a western Mediterranean seagrass meadow based on stable isotope analysis. *Estuaries* **2004**, *27*, 325–333. https://doi.org/10.1007/BF02803388.

46. Amor, M.M.B.; Salem, M.B.; Souissi, J.B.; Capape, C. On the occurrence of the deep-snouted pipefish *Syngnathus typhle* Linnaeus, 1758 (Osteichthyes: Syngnathidae) in Tunisian waters (Central Mediterranean). *Acta Adriat.* **2007**, *48*, 15–23.

47. Ben Alaya, H.; Sanna, D.; Casu, M.; Biagi, F.; Francalacci, P.; Carcupino, M.; Galzin, R.; Quignard, J.P.; Trabelsi, M. Analysis of meristic and mitochondrial DNA variation in *Syngnathus abaster* (Teleostea: Syngnathidae) from two western Mediterranean lagoons. *Biologia* **2011**, *66*, 1140–1147. https://doi.org/10.2478/s11756-011-0127-x.

48. Hablützel, P.I.; Wilson, A.B. Notes on the occurrence of *Syngnathus rostellatus* (Teleostei: Syngnathidae) in the Mediterranean. *Mar. Biodiv. Rec.* **2011**, *4*, E57. https://doi.org/10.1017/S1755267211000558.

49. Gristina, M.; Cardone, F.; Carlucci, R.; Castellano, L.; Passarelli, S.; Corriero, G. Abundance, distribution and habitat preference of *Hippocampus guttulatus* and *Hippocampus hippocampus* in a semi-enclosed central Mediterranean marine area. *Mar. Ecol.* **2015**, *36*, 57–66. https://doi.org/10.1111/maec.12116.

50. Moser, F.N.; Wilson, A.B. Reproductive Isolation Following Hybrid Speciation in Mediterranean Pipefish (Syngnathus Spp.). *Anim. Behav.* **2020**, *161*, 77–87. https://doi.org/10.1016/j.anbehav.2020.01.006.

51. Pierri, C.; Cardone, F.; Corriero, G.; Lazic, T.; Quattrocchi, F.; Alabiso, G.; Gristina, M. Density Decline in a Mediterranean Seahorse Population: Natural Fluctuations or New Emerging Threats? *Front. Mar. Sci.* **2021**, *8*, 692068. https://doi.org/10.3389/fmars.2021.692068.

52. Simal Rodríguez, A.; Grau, A.; Castro-Fernández, J.; Castejón, I.; Terrados, J.; Morales-Nin, B.; Arechavala-Lopez, P. Reproductive Biology of Pipefish *Syngnathus typhle* and *S. abaster* (Syngnathidae) from Western Mediterranean Sea. *J. Ichthyol.* **2021**, *61*, 608–615. https://doi.org/10.1134/S0032945221040147.

53. Martin-Smith, K.M.; Vincent, A.C.J. Exploitation and Trade of Australian Seahorses, Pipehorses, Sea Dragons and Pipefishes (Family Syngnathidae). *Oryx* **2006**, *40*, 141–151. https://doi.org/10.1017/S003060530600010X.

54. Rosa, I.; Sampaio, C. Collaborative Monitoring of the Ornamental Trade of Seahorses and Pipefishes (Teleostei: Syngnathidae) in Brazil: Bahia State as a Case Study. *Neotrop. Ichthyol.* **2006**, *4*, 247–252. https://doi.org/10.1590/S1679-62252006000200010.

55. Laksanawimol, P.; Petpiroon, S.; Damrongphol, P. Trade of Seahorses, Hippocampus Spp. (Actinopterygii: Syngnathiformes: Syngnathidae), on the East Coast of the Gulf of Thailand. *Acta Icth. Et Piscat.* **2013**, *43*, 229–235. https://doi.org/10.3750/AIP2013.43.3.07.

56. Harasti, D. Declining Seahorse Populations Linked to Loss of Essential Marine Habitats. *Mar. Ecol. Prog. Ser.* **2016**, *546*, 173–181. https://doi.org/10.3354/meps11619.

57. Masonjones, H.D.; Rose, E.; McRae, L.B.; Dixson, D.L. An Examination of the Population Dynamics of Syngnathid Fishes within Tampa Bay, Florida, USA. *Curr. Zool.* **2010**, *56*, 118–133. https://doi.org/10.1093/czoolo/56.1.118.

58. Claassens, L.; Booth, A.J.; Hodgson, A.N. An Endangered Seahorse Selectively Chooses an Artificial Structure. *Environ. Biol. Fish.* **2018**, *101*, 723–733. https://doi.org/10.1007/s10641-018-0732-4.

59. Salin, K.R.; Yohannan, T.; Nair, C. Captive Rearing of *Hippocampus kuda* and Issues of Seahorse Conservation in India. In Proceedings of the International Conference & Exposition on Marine Living Resources of India for Food and Medicine, Chennai, India, 27–29 February 2004.

60. Dody, S.; Manuputty, G.; Limmon, G. Habitat Characteristcs and Density of Spotted Seahorse *Hippocampus kuda* at Waters of Ternate Island and Surroundings, Maluku Utara, Indonesia. *IOP Conf. Ser. Earth Environ. Sci.* **2021**, *805*, 012006. https://doi.org/10.1088/1755-1315/805/1/012006.

61. Power, M.; Attrill, M.J. Long-Term Trends in the Estuarine Abundance of Nilsson's Pipefish (*Syngnathus rostellatus* Nilsson). *Estuar. Coast. Shelf S.* **2003**, *57*, 325–333. https://doi.org/10.1016/S0272-7714(02)00358-X.

62. Kuiter, R.H. *Seahorses and Their Relatives*; Aquatic Photographics: Seaford, UK, 2009; ISBN 9780977537211.

63. Berglund, A.; Sundin, J.; Rosenqvist, G. Baltic Pipefish Females Need Twice as Many Males as They Get. *Behav. Ecol.* **2017**, *28*, 827–832. https://doi.org/10.1093/beheco/arx046.

64. Whitfield, A.K.; Mkare, T.K.; Teske, P.R.; James, N.C.; Cowley, P.D. Life-Histories Explain the Conservation Status of Two Estuary-Associated Pipefishes. *Biol. Conserv.* **2017**, *212*, 256–264. https://doi.org/10.1016/j.biocon.2017.06.024.

65. Castejón-Silvo, I.; Terrados, J.; Nguyen, T.; Jutfelt, F.; Infantes, E. Increased Energy Expenditure Is an Indirect Effect of Habitat Structural Complexity Loss. *Funct. Ecol.* **2021**, *35*, 2316–2328. https://doi.org/10.1111/1365-2435.13876.

66. Pandolfi, J.M.; Bradbury, R.H.; Sala, E.; Hughes, T.P.; Bjorndal, K.A.; Cooke, R.G.; McArdle, D.; McClenachan, L.; Newman, M.J.H.; Paredes, G.; et al. Global Trajectories of the Long-Term Decline of Coral Reef Ecosystems. *Science* **2003**, *301*, 955–958. https://doi.org/10.1126/science.1085706.

67. Aylesworth, L.A.; Xavier, J.H.; Oliveira, T.P.R.; Tenorio, G.D.; Diniz, A.F.; Rosa, I.L. Regional-Scale Patterns of Habitat Preference for the Seahorse Hippocampus Reidi in the Tropical Estuarine Environment. *Aquat. Ecol.* **2015**, *49*, 499–512. https://doi.org/10.1007/s10452-015-9542-3.

68. Ape, F.; Corriero, G.; Mirto, S.; Pierri, C.; Lazic, T.; Gristina, M. Trophic flexibility and prey selection of the wild long-snouted seahorse *Hippocampus guttulatus* Cuvier, 1829 in three coastal habitats. *Estuar. Coast. Shelf S.* **2019**, *224*, 1–10. https://doi.org/10.1016/j.ecss.2019.04.034.

69. Lazic, T.; Fosso, B.; Bachir, B.; Corriero, G.; Gristina, M.; Marzano, M.; Pesole, G.; Santamaria, M.; Pierri, C. *Hippocampus guttulatus* Diet Based on DNA Metabarcoding. *Front. Mar. Sci.* **2023**, *10*, 1138279. https://doi.org/10.3389/fmars.2023.1138279.

70. Foster, S.A.; Vincent, A.C. Life history and ecology of seahorses: Implications for conservation and management. *J. Fish Biol*. **2004**, *65*, 1–61. https://doi.org/10.1111/j.0022-1112.2004.00429.x.

71. Joseph, E. A Study of the Systematics and Life History of the Gulf Pipefish, *Syngnathus scovelli* (Evermann and Kendall). Ph.D. Thesis, The Florida State University, Tallahassee, FL, USA, 1957.

72. Woodall, L.C.; Otero-Ferrer, F.; Correia, M.; Curtis, J.M.; Garrick-Maidment, N.; Shaw, P.W.; Koldewey, H.J. A synthesis of European seahorse taxonomy, population structure, and habitat use as a basis for assessment, monitoring and conservation. *Mar. Biol.* **2018**, *165*, 1–19. https://doi.org/10.1007/s00227-017-3274-y.

73. Frando, A.; Malavasi, S.; Zucchetta, M.; Franzoi, M.; Torricelli, P. Environmental influences on fish assemblage in the Venice Lagoon, Italy. *Chem Ecol.* **2006**, *22*, S105–S118. https://doi.org/10.1080/02757540600571836.

74. Pineiro-Corbeira, C.; Iglesias, L.; Nogueira, R.; Campos, S.; Jimenez, A.; Regueira, M.; Berreiro, R.; Planas, M. Structure and trophic niches in mobile epifauna assemblages associated with seaweeds and habitats of syngnathid fishes in Cíes archipelago (Atlantic Islands Marine National Park, Northwest Iberia). *Front. Mar. Sci.* **2021**, *8*, 773367. https://doi.org/10.3389/fmars.2021.773367.

75. Planas, M. Ecological traits and trophic plasticity in the greater pipefish *Syngnathus acus* in the NW Iberian Peninsula. *Biology* **2022**, *11*, 712. https://doi.org/10.3390/biology11050712.

76. Louisy, P. *Hippocampus Guttulatus, l'espèce Commune de l'étang de Thau, Hippo-Thau Bilan Scientifique 2005–2009*; CPIE: Bassin de Thau, France, 2011.

77. Faleiro, F.; Baptista, M.; Santos, C.; Aurélio, M.L.; Pimentel, M.; Pegado, M.R.; Paula, J.R.; Calado, R.; Rosa, R. Seahorses under a changing ocean: The impact of warming and acidification on the behaviour and physiology of a poor-swimming bony-armoured fish. *Conserv. Physiol.* **2015**, *3*, cov009. https://doi.org/10.1093/conphys/cov009.

78. Lazic, T.; Pierri, C.; Corriero, G.; Balech, B.; Cardone, F.; Deflorio, M.; Fosso, B.; Gissi, C.; Marzano, M.; Nonnis Marzano, F.; et al. Evaluating the efficiency of DNA metabarcoding to analyze the diet of *Hippocampus guttulatus* (Teleostea: Syngnathidae). *Life* **2021**, *11*, 998. https://doi.org/10.3390/life11100998.

79. Diaz-Ruiz, S.; Aguirre-Leon, A.; Perez-Soli, O. Distribution and abundance of *Syngnathus louisianae* and *Syngnathus scovelli* (Syngnathidae) in Tamiahua Lagoon, Gulf of Mexico. *Cienc. Mar.* **2000**, *26*, 125–143. https://doi.org/10.7773/cm.v26i1.567.

80. Lourie, S.A.; Pollom, R.A.; Foster, S.J. A global revision of the seahorses *Hippocampus* Rafinesque 1810 (Actinopterygii: Syngnathiformes): Taxonomy and biogeography with recommendations for further research. *Zootaxa* **2016**, *4146*, 1–66. https://doi.org/10.11646/zootaxa.4146.1.1.