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The African Striped Grunt, *Parapristipoma octolineatum* (Valenciennes, 1833), in the Mediterranean Sea: The Third Record with Biological and Ecological Notes, and Identification Key for Haemulidae Recorded in the Mediterranean

Francesco Tiralongo ^{1,2,3}, Anna Maria Pappalardo ¹, Sara Ignoto ^{1,2}, Bianca Maria Lombardo ¹, Venera Ferrito ¹, Aitor Campos Sosa ⁴ and Andrea Spinelli ^{4,*}

- Department of Biological, Geological and Environmental Science, University of Catania, 95124 Catania, Italy; francesco.tiralongo@unict.it (F.T.); pappalam@unict.it (A.M.P.); sara.ignoto@phd.unict.it (S.I.); bm.lombardo@unict.it (B.M.L.); vferrito@unict.it (V.F.)
- ² Ente Fauna Marina Mediterranea, Scientific Organization for Research and Conservation of Marine Biodiversity, 96012 Avola, Italy
- Institute for Biological Resources and Marine Biotechnologies, National Research Council, 60125 Ancona, Italy
- ⁴ Research Department, Fundación Oceanogràfic de la Comunitat Valenciana, Oceanogràfic, Ciudad de las Artes y las Ciencias, 46013 Valencia, Spain; conservacion@oceanografic.org
- * Correspondence: aspinelli@oceanografic.org

Abstract: The Mediterranean Sea biodiversity is undergoing a rapid reshaping due to different factors, many of which are directly related to human activities (e.g., pollution, habitat destruction, overfishing and introduction of non-indigenous species). In this context, climate changes, and in particular water warming, are the main factors that favor the arrival and subsequent spread of thermophilic species, regardless of their area of origin (e.g., Red Sea, Atlantic Ocean). In this research, we report the third and eastern-most well-documented record of the thermophilic species Parapristipoma octolineatum (Valenciennes, 1833) in the Mediterranean Sea, providing new morphological, genetic, ecological and biological data on this rare species in the basin. The integrative taxonomical approach was robust and unambiguously identified the species as P. octolineatum. The specimen was a large-sized female in maturation. The first high-quality photographic illustrations of otoliths were provided. Recent direct observations of fishermen in the area suggested a species abundance increase. However, data from the Mediterranean Sea remain scarce and further efforts are needed to better understand the abundance and distribution of this species. Modified keys for a rapid color pattern-based determination of all Haemulidae species recorded in the Mediterranean Sea to date are also provided. Finally, the potential further expansion under the future prediction of climate changes of this Atlantic origin fish in the Mediterranean Sea is discussed.

Keywords: citizen science; global change; Haemulidae; thermophilic species; water warming



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1. Introduction

The Mediterranean bioregion represents one of the main biodiversity hotspots in the world, with a high percentage of endemic species and a marine biodiversity estimated at about 17,000 species [1,2]. On the other hand, the Mediterranean Sea is also a critical area from a climatic perspective, being particularly susceptible to climate changes [3], a phenomenon that affects biological and ecological processes, favoring the establishment and spread of thermophilic species from different origin areas [4]. In this regard, we can call into question the phenomena of "meridionalization" and "tropicalization". The first one refers to Mediterranean native fishes that expand northwards, invading new areas. The second phenomenon instead refers to exotic fishes invading and colonizing the Mediterranean basin [5]. The "blue runner", *Caranx crysos* (Mitchill, 1815), the "false scad", *Caranx rhonchus*

Geoffroy Saint-Hilaire, 1817 and the "parrotfish", Sparisoma cretense (Linnaeus, 1758) are relevant examples of Mediterranean native species involved in the process of "meridionalization" [6-8]. On the other hand, alien fishes such as the "bluespotted cornetfish", Fistularia commersonii Rüppell, 1838, the "silver-cheeked toadfish", Lagocephalus sceleratus (Gmelin, 1789) and the "devil firefish", Pterois miles (Bennett, 1828) are remarkable cases of exotic invasive species involved in the phenomenon of "tropicalization" [6,9]. However, Bianchi et al. (2018) [10] disagreed with this division proposed by Boero et al. (2008) [11]. Indeed, they proposed to consider the arrival and/or expansion of thermophilic species in the Mediterranean Sea, regardless of their origin area, as driven by the same climatic, hydrological and ecological factors, including in the "tropicalization" phenomenon all these species and reported no differences with the "meridionalization" phenomenon. In other words, they consider all thermophilic expanding species within the "tropicalization" phenomenon. Some of these thermophilic species have become invasive and can have devasting impacts on the invaded areas/ecosystems; others may impact fishing activity, although not always negatively; and a small number of them can have negative, and sometimes deadly, effects on human health [12].

Most members of Haemulidae (represented by a total of 137 valid species) are marine demersal fishes native to tropical and subtropical areas [13,14]. In the Mediterranean Sea, the family Haemulidae Gill, 1885 includes six species and four genera, namely Orthopristis chrysoptera (Linnaeus, 1766), Parapristipoma octolineatum (Valenciennes, 1833), Plectorhinchus gaterinus (Forsskål, 1775), Plectorhinchus mediterraneus (Guichenot, 1850), Pomadasys incisus (Bowdich, 1825) and Pomadasys stridens (Forsskål, 1775) [15]. Among these, those belonging to the genus *Pomadasys* Lacepède, 1802 have received particular attention from the scientific community due to their wider distribution in the Mediterranean basin [16,17]. In fact, P. stridens (striped piggy) is native to the Indo-Pacific region and entered the Mediterranean Sea through the Suez Canal. This species is therefore considered a Lessepsian immigrant and colonized the eastern part of the basin up to Greece, with a single reported record from Italy in 1968 [18,19]. Pomadasys incisus (bastard grunt), on the other hand, is a thermophilic species of Atlantic origin, widely recorded in the Mediterranean Sea [17-20]. Between the two species of the genus *Plectorhinchus* Lacepède, 1801, only *P. mediterraneus* is a native fish, widespread in the eastern Atlantic, from Portugal to Angola, and in the western Mediterranean, along the northwestern African coast and in the Adriatic Sea [21–23]. On the other hand, P. gaterinus, native to the western Indian Ocean, is known in the Mediterranean Sea through a single unusual record, as a juvenile specimen was found in the mantle cavity of the European squid *Loligo vulgaris* Lamarck, 1798 [24]. *Orthopristis chrysoptera* is also known from a single record in the central Mediterranean Sea. This alien fish, native to the western Atlantic, was recorded in the harbor of Siracusa (Sicily, South-East of Italy, Ionian Sea) in 2020, where it was most probably introduced through ballast water [15].

Parapristipoma octolineatum, commonly known as the African striped grunt, is a fish of Atlantic origin distributed in the eastern Atlantic, from Angola northward to Spain and France (Bay of Biscay), including Cape Verde, Madeira and Canary Islands, and in the western Mediterranean Sea [21,25–31]. In the western Mediterranean Sea, Bauchot (1987) [32] reported the presence of the species along the coasts of Morocco, Algeria and western Spain; recent records come from Spain (off Salobreña) and Algeria (off Béni Saf) [31]. It inhabits shallow coastal waters to a depth of about 60 m on rocky or sandy bottoms, where it can be found as isolated individual or gregarious (schooling individuals). It is a medium-sized fish and its maximum size rarely exceeds 50 cm in total length (TL), while common sizes are between 20 and 30 cm TL [32,33]. Small specimens seem to prefer shallower waters than adults [21,34]. The biology and ecology of this species are still poorly known. We only have fragmented data, meaning that the data are oviparous, and adults mainly feed on crustaceans and mollusks [21]. In the Mediterranean Sea and in its whole distribution range, only a small number of studies have been conducted on *P. octolineatum*, and they mostly concern a single specimen and distributional data.

The aim of this research is to provide an updated review on the Mediterranean distribution of *P. octolineatum*, with a new record from Spain (eastern-most record for the Mediterranean Sea, considering recent literature). We also provide new biological, morphological and meristic data from a Mediterranean specimen, and, for the first time, its genetic identification. Moreover, we also provide a new modified key for rapid and non-destructive identification of Haemulidae species recorded in the Mediterranean basin to base. The presence of this species in the Mediterranean Sea is finally discussed and related to the possible expansion linked to climate change.

2. Materials and Methods

2.1. Data Collection

On 5 December 2022, a large specimen of *Parapristipoma octolineatum* was caught with a trammel net at a depth of 20 m off Teulada-Moraira (Mediterranean Spain) (38.67011 N, 0.13271 E; Figure 1) on mixed bottoms (sand and rocks). A picture of the specimen was sent to one of the authors (A.S.) by a skipper of the fishing boat, questioning the identity of the species. He asked the fishermen to keep the specimen, which was handed over to the researchers immediately after landing. Furthermore, fishermen of the area said that this was not the first time they caught this "strange-looking" fish, but in last few years, they have noticed an increase in catches of the species (2–3 per year in the last 5 years). The specimen was immediately collected at a landing point by experts and taken to the laboratory of Fundación Oceanogràfic, Research Department (Valencia, Spain) for analysis, which was performed on the fresh specimen (Figure 2A). After analysis, the fish was stored frozen at the laboratory.

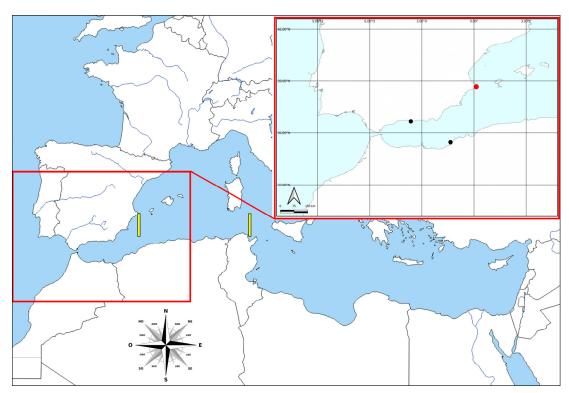


Figure 1. Updated distributional map of *Parapristipoma octolineatum* in the Mediterranean Sea. Record of the current research is indicated with a red circle (insert). Published records indicated with black circles (see Table 1 for details). Yellow lines (main map) indicate the limits of the historical Mediterranean distribution of the species (from west).

Table 1. Main morphometric and meristic data on the collected specimen of <i>Parapristipoma octolineatum</i>

Morphometric and Meristic Data					
	cm				
Total length (TL)	35.0				
Standard length (SL)	29.5				
Fork length (FL)	33.5				
Head length (HL)	9.2				
Pre-anal length (PAL)	18.0				
Weight	g 542				
Dorsal fin	XIII + 14				
Anal fin	III + 7				
Pectoral fin	12				
Pelvic fin	I + 5				

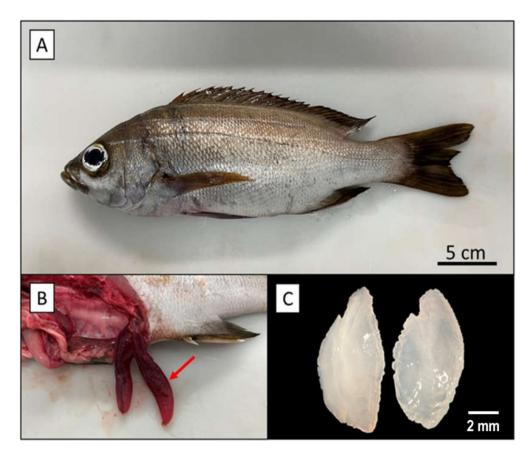


Figure 2. Adult female specimen of *P. octolineatum* collected in Spain (off Teulada-Moraira) on 5 December 2022; (**A**): general lateral view of the fish, (**B**): gonads in evidence, (**C**): otoliths (sagittae), outer and inner face. Red arrow in (**B**) indicates the gonads.

2.2. Laboratory Analysis

All the main morphometric and meristic characters were recorded in the freshly caught specimen. Histological analysis on the gonads was performed in order to identify the sex and maturation stage. The stomach content was analyzed. Otoliths (sagittae) were extracted and photographed. The fish was easily identified following the general morphological and color pattern description reported in [34], and meristic provided by Bañón et al. (2019) [30].

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2.3. Molecular Identification via DNA Barcoding

Total genomic DNA was extracted from muscle tissue stored in 95% ethanol using the DNeasy blood and tissue kit (Qiagen) following the manufacturer's instructions. A COI barcode sequence was obtained using the primers VF2_t1- and FishR2_t1 [35] and the PCR amplification conditions described in Pappalardo et al. (2015) [36]. The amplicon was checked through agarose gel electrophoresis and purified with the QIAquick PCR purification kit (Qiagen). Subsequently, sequencing was bidirectionally performed by Eurofins Genomics (https://eurofinsgenomics.eu (accessed on 20 July 2023)). The obtained sequence was carefully checked and deposited in the GenBank public database (http:// www.ncbi.nlm.nih.gov/genbank/ (accessed on 20 July 2023)) with the following Accession number: OR050541. The individual taxonomic identity was verified by comparison of the COI sequence within the GenBank database using the Blast search tool (https://blast.ncbi. nlm.nih.gov/Blast.cgi (accessed on 20 July 2023)). In addition, a Maximum Likelihood (ML) tree was built using a HKY + G model calculated in MEGAX [37] and included in the dataset COI barcode sequences (at least 650 bp long) of P. octolineatum and P. trilineatum retrieved from GenBank (MH980024, JF952805, JF952807, JF952808, EF607481, EF607482, HQ676782, JX042222, JX042223,). A COI sequence of Plectorhinchus macrolepis was used as the outgroup (HQ676788).

2.4. Preparation of a Rapid Identification Key

Being the species of the family Haemulidae reported in the Mediterranean Sea, and properly identifiable without the utilization of destructive techniques and laboratory analysis, we deem it useful to provide a rapid identification key. To achieve this, we used simple, constant and easily verifiable characters, mainly based on color pattern differences between adult specimens of the treated species. This key will be useful for rapid identification in the field (specimens caught or observed in their environment) and from photographic evidence. The dichotomous identification key was constructed following both the most relevant published manuscripts/manuals on the treated species and authors' experience of them [15,17,21–24,28,31–34,38]. Due to the unusual story of the record of *P. gaterinus* described above (see Corsini-Foka and Sarlis, 2016 [24]), it was not included in the key, as the species most likely never swam in Mediterranean waters to date.

3. Results

3.1. Biological, Morphological and Genetic Characterization of the Analyzed Specimen

The specimen caught by fishermen in Mediterranean Spanish waters was an adult female (stage II—maturing virgin and recovering spent: ovaries increasing in size, orange in color; external blood vessels started to develop) and showed a total length (TL) of 350 mm and a weight of 542 g (Figure 2A,B). The meristic formula was D = XIII + 14; A = III + 7. All the main morphometric and meristic characters are reported in Table 1. The stomach was empty. The body was laterally compressed and relatively stout; the snout was pointed with a terminal and slightly upturned mouth. The caudal peduncle was rather robust. The sides and ventral part of the body were silvery-white; the dorsal area had three clearly visible narrow longitudinal white stripes on a light brown background color. A fourth stripe of the same color extended below the third one (on the sides) and was poorly visible because it was on a light (whitish) background color. Caudal, anal, pelvic and dorsal fins were predominantly blackish in color; pectoral fins were yellow-brownish. The lateral line was highly visible and slightly curved following the dorsal profile. Relatively big eyes with a silvery and black iris and black pupil were noted. A mouth of moderate size with small teeth was also noted. Otoliths (sagittae) morphology is shown in Figure 2C. Recent Mediterranean records of the species are summarized in Table 2.

Date	Country	Location	Coordinates	Depth	Size	References
4 October 2008	Spain	Off Salobreña	36.7230 N 3.5808 W	1 m	n.a.	Tiralongo and Azzurro; In: Tsagarakis et al., 2021 [31]
21 October 2020	Algeria	Off Béni Saf	35.3014 N 1.4250 W	15 m	24.8 cm	Al Mabruk et al.; In: Tsagarakis et al., 2021 [31]
5 December 2022	Spain	Off Teulada- Moraira	38.6701 N 0.1327 W	20 m	35.0 cm	Present study

Table 2. Recent records with related main data on collected/observed specimens of *P. octolineatum* in the Mediterranean Sea; n.a. = not available.

A 681bp-long region of the COI gene was sequenced from the specimen identified based on morphological characters. The COI sequence was functional without stop codons and NUMTs. The Blast search against the GenBank database revealed an identity percentage of 99.85% with a 100% query coverage corresponding to a COI barcode of *Parapristipoma octolineatum* (GenBank Accession Number MH980024). This result was confirmed by the ML tree (Figure 3).

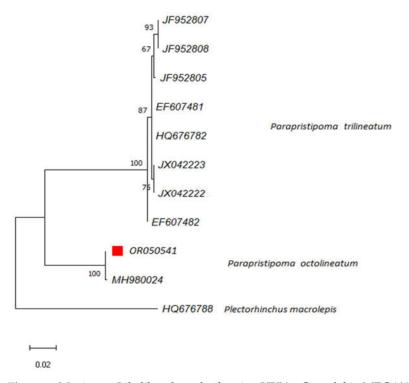


Figure 3. Maximum Likelihood tree built using HKY + G model in MEGAX showing the clustering of the *P. octolineatum* COI sequences obtained in this study (\blacksquare).

In summary, the morphological and meristic data agreed with those reported in the literature [30], and also molecular techniques confirmed the identity of the species as *P. octolineatum*.

3.2. Identification Key for Rapid Identification of Haemulidae Species Recorded in the Mediterranean Sea

- Body with highly visible and regular longitudinal bars (white or yellow-brown) extending on the sides from the head/anterior part of the body to the caudal peduncle

3.3. Remarks on Identification Key

Orthopristis chrysoptera: juveniles with two regular dark brown stripes extending longitudinally along the sides (dorsal area). Between these two dark stripes, an additional longitudinal and narrow bright yellow stripe is present [39].

Plectorhinchus mediterraneus: juveniles with whitish-greyish bodies with two regular dark brown (or blackish) stripes extending longitudinally along the sides (dorsal and midlateral area). The dorsal one is curved following the dorsal profile, while the midlateral one is straight and runs from the head to the caudal peduncle. The superior part of the eye is edged with golden yellow [39].

Common sizes (total length in cm) [39] in ascending order are as follows: *P. stridens* (15 cm); *P. incisus* (25 cm); *O. chrysoptera* and *P. octolineatum* (30 cm); *P. mediterraneus* (65 cm).

4. Discussion

The species shift range in relation to the water warming of the Mediterranean Sea is increasing the spatial overlap between endemic/native and exotic fishes, representing a threat to biodiversity and to the balance of the marine ecosystem. Several studies have now proved an acceleration in biological invasion processes with the introduction of thermophilic species from the Atlantic and the Red Sea in relation to the increasing water temperature of the Mediterranean basin [40,41]. Hence, this biotic homogenization through the gradual semi-replacement of endemic/native species by non-indigenous ones is critical, especially in a semi-enclosed basin with a high diversity of endemic species, such as in the Mediterranean. The Mediterranean Sea is becoming warmer and the impact on its biota by southern species looks set to increase in the coming years. In that scenario, a serious concern about the reversed balance between exotic and endemic species richness looks realistic. Furthermore, some of these species can also represent a problem for the economy related to fishing activities; however, on the other hand, some of them could represent new economic resources for fishery. Finally, some of these species, being venomous, also represent a threat to human health.

The molecular identification via DNA barcoding agreed with the morphological and color pattern-based identification of the specimen belonging to *Parapristipoma octolineatum*. The identification key here provided was developed to make the process of identification of Haemulidae species recorded in the Mediterranean to date a rapid, simple and non-destructive method based on color pattern analysis. This can be particularly useful for in situ observation or during fishery surveys, during inspections at fish markets, or in subsequent analyses and identification from photos on fish fauna [42,43].

The present study provides new distributional, morphological and biological data on *P. octolineatum* in the Mediterranean Sea, with a new well-documented record from Spain (eastern-most record for the Mediterranean Sea, considering recent literature), supporting the presence of a well-established population in the western-most part of the basin. Indeed, in addition to the few records present in literature, local fishermen said that they

sporadically catch this species in recent times. It is therefore reasonable to think that the abundance of the species is increasing. Moreover, the caught specimen was a large female in maturation, almost ready to reproduce and release its eggs in all likelihood, in Spanish Mediterranean waters. We also provided high-quality photographic illustrations of otoliths (sagittae), which helped us to identify the species from the fish that remained during the stomach content analysis of its predators, as these structures are species-specific [44]. The African striped grunt (P. octolineatum) has such a color pattern that it would hardly go unnoticed, which is why we think that the lack of reports in past years is not due to the fact that it went unnoticed by fishermen, but due to its scarce presence and low abundance in the area. Within the current global warming scenario, the establishment and increase in the abundance of this species in the western Mediterranean Sea could be the beginning of its rapid expansion towards the east, in a similar way to that followed by the bastard grunt (P. incisus) [17]. Indeed, this latter species (another member of the family Haemulidae), recorded for the first time in the Mediterranean Sea in 1840 [45], has currently expanded its presence to the entire basin, becoming abundant in some areas [9–31]. The reasons for its success in colonizing the Mediterranean basin are to be found in its ecological characteristics, such as its fast growth and relatively short life cycle [46,47], as well as its preference for warm waters [48]. In a similar way, the "newcomer" object of our study (P. octolineatum) could continue its invasion eastward in the Mediterranean following two different dispersal pathways: along the northern African coast (Algeria and Tunisia) and along the northern coasts of Spain and France, in opposition to the main Mediterranean currents [38]. On the other hand, the Lessepsian immigrant Pomadasys stridens followed an opposite direction in colonizing the Mediterranean basin. Indeed, this other haemulid is expanding further and further west in the basin [49,50], although established populations still remain confined to the eastern part only [51]. The Mediterranean is acting as a catchment area for thermophilic species of whatever origin they are. In this context, the basin can be also considered as a large open-air laboratory to study biological invasion processes and interaction between native/endemic species and non-indigenous ones, but also between exotic species of different origins (e.g., Atlantic and Red Sea). The colonization of the Mediterranean Sea by the family Haemulidae can be an excellent example to study and deepen our knowledge on the overlapping of species' niches. On the other hand, the two most common species (*P. incisus* and P. stridens) already have a partially overlapping distribution, although interactions and competition dynamics between these two fish species need to be adequately studied. The future further expansion of *P. octolineatum* will make this scenario even more complex, hence the need to better study and understand biological and ecological aspects of this species, which still remain unknown known, and to trace its eastward expansion into the Mediterranean basin.

The first documented records of *P. octolineatum* in the basin have only recently been published by Tiralongo and Azzurro (2021) from Spain and Al Mabruk et al. (2021) from Algeria (see Tsagarakis et al., 2021 [31]). These records date back to 2008 and 2020, respectively. In the last 15 years, no other records of *P. octolineatum* have been reported in the Mediterranean Sea. Furthermore, previous data on the Mediterranean presence and abundance of the species are only of a general nature, and regard its general distribution, with no specific records [32]. Hence, to the best of our knowledge, no other records of the species are available for the Mediterranean Sea. Furthermore, the species is absent from the fish checklist provided by Báez et al. 2019 from eastern Spain [29], while in Algeria, the species is considered very rare [26]. Also considering the Atlantic distribution of the species, the situation is similar, with the northern-most and the only well-documented record reported by Casamajor [28] in 2013. In this oceanic sector, the historical distribution of the species is confined to the eastern part, from the coast of Angola northward to Spain, including Cape Verde, Madeira and Canary Islands [21,27,28]. However, in the latter two areas, the species is reported as rare [27,52]. In light of the overall Atlantic–Mediterranean distribution of the species analyzed by us, it is clear that our knowledge of this species is still very limited, not only from a biological and ecological point of view, but also from a distributional one.

Indeed, more detailed and well-documented data on these fish data are available only from recent times and were obtained thanks to the collaboration of "citizen scientists". Indeed, the northern-most record from the Atlantic Ocean (Bay of Biscay, France) reached scientists thanks to a scuba diver. The Mediterranean records of the species were obtained thanks to the collaboration of an amateur fisher and a scuba diver [31], and of a professional fisherman (present record, third most well-documented in the Mediterranean Sea), hence the need to strengthen the collaboration with the different "sea users" because our knowledge on most marine species is very limited, even with species that we take for granted. Despite the quite wide distribution reported for *P. octolineatum* and its "attention-catching" color pattern, we realized that our knowledge on this species is still very limited.

The published distribution of *P. octolineatum* in the Mediterranean Sea still remains patchy and limited to the western sector of the basin; furthermore, biological and ecological aspects of the species are not well-known and further research is needed to improve our knowledge on this thermophilic fish. In this context, the monitoring of the species through citizen science is of fundamental importance to track its further expansion in the Mediterranean Sea; indeed, citizen science has proven to be particularly effective in the detection (sometimes at an early stage of colonization) of "strange looking" species [6,38,46–48,53], especially for those species with unusual color patterns that can be easily identified even by non-experts, as in the case of *P. octolineatum*. Moreover, as in the present study, professional fishermen in particular play a key role in providing researchers with other specimens to study and further data on its distribution and habits.

5. Conclusions

The Mediterranean Sea is rapidly changing due to different human activities, but the warming of marine waters is one of the main factors contributing to reshaping its biodiversity and ecological processes [1]. The basin is connected to the west with the Atlantic Ocean through the natural opening of the Strait of Gibraltar, and to the east with the Red Sea and the entire Indo-Pacific area through the artificial opening of the Suez Canal. This allows thermophilic species of any origin (including native) to spread through the whole basin, with Atlantic species that spread eastward, Lessepsian immigrants that spread westward, and native southern species that spread northward (from southern to northern part of the Mediterranean Sea) [7,40]. In other words, the Mediterranean Sea is acting as a catchment basin for southern thermophilic species. Some of these species are invasive and successfully colonized the whole basin [4] or have become more common than endemic species in some areas [54]. The rapid spread of some non-indigenous and thermophilic species in general is changing the biodiversity of the Mediterranean, with consequences on ecosystems, fishery and human health [55], hence the urgent need to improve our knowledge of the processes underlying biological invasions.

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