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Contribution to the Special Issue: "Ocean Literacy across the Mediterranean Sea region"

Ocean literacy and scientific data acquisition through citizen science campaigns: a mixed approach in the Maltese Islands to collect information on *Pinna nobilis* and *Pinna rudis*

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Abstract

The genus *Pinna* includes two species in the Mediterranean Sea: *Pinna nobilis* and *Pinna rudis*. Both these species are under threat from multiple stressors. *Pinna nobilis*, in particular, has been exhibiting mass mortality events (MMEs) since 2016. The population and distribution of these species have never been comprehensively explored in the Maltese archipelago, and in this work, we collate information collected between 2006 and 2019 through a number of SCUBA underwater visual census monitoring programs. The logistical barriers surrounding SCUBA-based sampling techniques and the low-density distribution of these species constitute significant obstacles to an extensive conventional population assessment. Citizen science was thus also deployed in this study to supplement the data collected through SCUBA surveys: recreational SCUBA divers worked as citizen scientists, providing data on the distribution of these two endangered species from areas never explored before. This information can be used for assessing the conservation status of *P. nobilis* and *P. rudis* in Maltese waters, whilst contributing to the next generation of ocean-literate citizens.

Keywords: Pen Shell Mussel; Mass Mortality; Citizen Science; Ocean Literacy; SCUBA; population monitoring program.

Introduction

The Pinnidae family (Leach, 1819) is comprised of bivalve molluscs having a typically thin triangular shell with a pointed umbo anchored to the substratum through a byssus (Tebble, 1966). In the Mediterranean, two species of the genus *Pinna* can be found: *Pinna nobilis* (Linnaeus, 1758) and *Pinna rudis* (Linnaeus, 1758). *Pinna nobilis* and *P. rudis* are the largest bivalves in the Mediterranean (Zavodnik *et al*., 1991; Galinou-Mitsoudi *et al*., 2006), with the former being endemic to the Mediterranean. They are long-lived species, with *P. nobilis* known to exceed, in rare cases, a lifespan of 45 years (Rouanet *et al*., 2015), and *P. rudis* living up to 31 years (Colomer *et al*., 2016). *Pinna nobilis* is often associated with the occurrence of *Posidonia oceanica* meadows growing on soft bottoms (Vicente, 1990). The depth distribution of *Pinna* spp. individuals, ranging between 0.5 m and 60 m (Butler *et al*., 1993; Richardson *et al*., 1999; Garcia-March & Kersting, 2006; Katsanevakis, 2007; Coppa *et al*., 2013), appears correlated with size, with larger individuals frequently found in deeper waters which are less exposed to human disturbance (Addis *et al*., 2009; Guallart & Templado, 2012). *Pinna rudis* can occur in the same habitat as *P. nobilis*, but is more commonly found on rocky, hard substrata (García-March & Kersting, 2006). Since *P. rudis* is often confused with *P. nobilis*, little is known about its biology, ecology (Kersting & García-March, 2017) and its actual distribution in the Mediterranean (Gvozdenović *et al*., 2006).

Pinna nobilis and *P. rudis* provide several ecosystem functions and services. The shells act as a hard substrate on generally soft, less heterogeneous substrata, increasing the local biodiversity (Basso *et al*., 2015) by hosting different species assemblages (Çinar *et al*., 2021) including strictly-associated commensal species (Trigos & Vicente, 2019; Acarli *et al*., 2019). Moreover, they filter large volumes of water, retaining organic matter and decreasing water turbidity (Basso *et al*., 2015). They are used as bioindicators as they can bioaccumulate heavy metals (Rabaoui *et al*., 2014). Furthermore, they provide cultural services by being an attraction for SCUBAdivers and for the eco-tourist (Morrocco *et al*., 2018). Despite this, the two species are sensitive to a diverse array of pressures and impacts (Morrocco *et al*., 2018; Alomer *et al*., 2015).

In particular, *P. nobilis* is threatened by several anthropogenic pressures such as direct harvesting for decorative purposes, consumption and silk production from the byssus (Vicente & Moreteau, 1991; Islam & Tanaka, 2004; Rabaoui *et al*., 2010; Deidun, 2012; Basso *et al*., 2015), as well as by indirect threats such as habitat destruction, pollution, trawling, urbanization, boat anchoring and climate change (Centoducati *et al*., 2007; Rabaoui *et al*., 2010; Basso *et al*., 2015).

Since 2016, starting from the Alboran Sea in the western Mediterranean Sea, a still-ongoing Mass Mortality Event (MME), caused by the protozoan *Haplosporidium pinnae*, has been affecting populations of *Pinna nobilis* (Darriba, 2017; Vázquez-Luis *et al*., 2017; Catanese *et al*., 2018; Zotou *et al*., 2020). Recently, *H. pinnae* has been frequently recorded in association with other bacterial pathogens, making the MME scenario more complex than initially thought and far from being fully understood (Carella *et al*., 2020; Prado *et al*., 2020; Scarpa *et al*., 2020). In the Western Mediterranean, North Tyrrhenian Sea, Adriatic Sea and Aegean Sea, up to 100% mortality levels were recorded within monitored *P. nobilis* populations (Vázquez-Luis *et al*., 2017; Kersting *et al*., 2019; Acarli *et al*., 2020; Čižmek *et al*., 2020; Acarli et al., 2021; Betti *et al*., 2021; Katsanevakis *et al*., 2021), in contrast with *P. rudis* populations, which are not affected by *H. pinnae* (Catanese *et al*., 2018).

Pinna nobilis is protected by Annex IV of the EU Habitats Directive 92/43/EEC (EEC, 1992) and Annex II of the Barcelona Convention, and is included in the IUCN Red List as a Critically Endangered Species. *Pinna rudis* is considered a strictly-protected species in Annex II of the Bern Convention and as an endangered species in the Barcelona Convention. In a recent study by Vázquez-Luis *et al*. (2021), it emerged that natural hybridization occurs between *P. nobilis* and *P. rudis*. with these hybrids apparently being resistant to *H. pinnae*, similarly to *P. rudis*. This discovery highlights the importance of monitoring the current status of distribution of both species and of identifying areas of sympatry; indeed, despite the two species having different benthic habitat preferences (Vazquez-Luis *et al*., 2014; Kersting & Garcìa-March, 2017), they can coexist on the same substratum, making hybridization more probable.

In Maltese waters, no comprehensive studies concerning the distribution of these two species and the occurrence of the MME within local *P. nobilis* populations have been published to date. Moreover, these species are usually found in very low densities (García-March *et al*., 2007; Basso *et al*., 2015), making it easy to underestimate their real distribution and abundance. For this reason, in this study, two different monitoring techniques were deployed: systematic SCUBA diving visual census surveys as well as a citizen science questionnaire.

Citizen science is a non-conventional data acquisition tool, but it may also represent one of the most effective methods of monitoring and collecting data for these two ecologically important species. Indeed, many studies have demonstrated how citizen science can produce a high-quality dataset (Kosmala *et al*., 2016; McKinley *et al*., 2017; Krželj *et al*., 2020). Data acquisition is not the only contribution that citizen science programmes can make, given that the conduction of such programmes also promotes the implementation of ocean literacy. The training and engagement of citizen scientists, in fact, develops a population of final users with a better attitude towards scientific knowledge and an enhanced awareness on the nature of the science itself (Aristeidou & Herodotou, 2020). Citizen science is a participation and integration process involving both professional scientists and citizen scientists (Kelly *et al*., 2021), with citizen science being considered a stepping-stone in the nurturing of a new generation of Mediterranean-Sea-literate citizens (Mokos *et al*., 2020), in line with the goals of the United Nations Decade of Ocean Science for Sustainable Development 2021-2030.

This work is the first attempt at gathering information on *P. nobilis* and *P. rudis* distribution and density in Maltese nearshore waters, in order to investigate population dynamics, to highlight any trends in abundance between 2006 and 2019 and to formulate recommendations on future monitoring surveys for the two species by merging relevant data originating from different surveying methodologies. The objective of this paper is also to increase the level of involvement of SCUBA divers based in Malta in the conservation of the endangered fan pen shells, with the ultimate goal of increasing their related knowledge and thus become ocean-literate citizens.

Materials and Methods

Study area

For the systematic underwater visual census surveys, four sites located within different marine Sites of Community Importance (SCIs) (NATURA 2000 sites) were targeted (Fig. 1), at depths ranging between 5 m and 38 m. In 2006, the northern area of the MT0000101 Natura 2000 site (Site 1) was systematically explored through ten strategically selected stations. The exact location of these stations was chosen according to the following criteria: the occurrence of extensive *Posidonia oceanica* meadows and of previous knowledge, through preliminary surveys, on the presence of fan pen shells in the area. In 2012, over the January-February period, the same site 1 stations were explored once again so to assess changes with the data obtained in 2006. Three other census survey sites were added, located within the Natura 2000 sites MT0000104 (Site 2 – four survey stations), MT0000103 (Site 3 – ten survey stations) and MT0000105 (Site 4 – six survey stations). During July 2018, a new census survey was conducted across sites 1, 3 and 4 with five, four and three survey stations each, respectively, as part of Malta’s first national Marine Strategy Framework Directive (MSFD) monitoring programme. Site 2 was

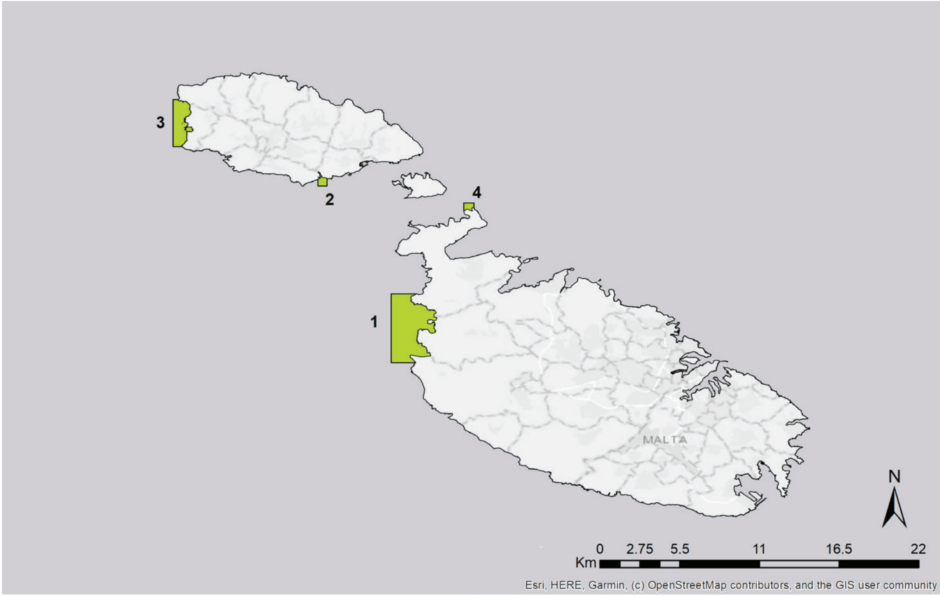

Fig. 1: Sites explored during the underwater visual census monitoring programme.

excluded from this monitoring program since no living specimens of pen shells were found in the 2012 monitoring program. The total benthic area explored during the various SCUBA surveys conducted over the 2006-2018 period was that of 14,100 m^2, including the total number of survey sites including the whole monitoring period relevant to each SCUBA sampling site for the 2006-2018 period (summarised in Table 1).

Sampling techniques

As part of the 2006 and 2012 monitoring surveys, a circular transect sampling method was adopted within each station in order to estimate the density of individuals of the two species.

Corresponding to each station's coordinates, a stainless-steel rod was inserted into the seabed, marking the centre of a circle. The edge of the circle was determined using a measuring tape with a pre-defined length (representing the extent of the circumference of the same circular area). This area was systematically searched for any live individual or empty shell fragments of the target species.

During the survey of 2006, at Site 1, all the live individuals of *P. nobilis* found were tagged with a float attached to their proximity in order to facilitate future location of the same site and to in order to find the exact position of the same individuals in case the tagged specimen was missing. The citizen science survey was conducted as part of the Interreg MED Programme (2014-2020) AMAre (Actions for Marine Protected Areas) project. This project worked on the development of shared geospatial tools for integrated management, with the project geoportal covering Marine Protected Areas (MPAs) participating in the project being publicly accessible at: https://amare.interreg-med.eu/toolbox/geoportal/. These tools will be used for achieving MPA-related objectives, including the establishment of best practices for monitoring present and future drivers of changes in biodiversity and ecosystem services within MPAs participating in the project.

56 diving centres across the Maltese archipelago were contacted. Different informative tools were designed and

Table 1. SCUBA surveys conducted over the 2006-2018 period, with details about the number of stations explored for each site and year in brackets.

Site ref.	Survey Area	Survey year/s (N. Survey stations)
Site 1	Northern reaches of the MT0000101 NATURA 2000 site ("il-Baħar bejn Rdum Majjiesa u Għar Lapsi"	2006 (10), 20112 (10), 2018 (5)
Site 2	The MT0000104 Natura 2000 site ("il-Baħar bejn il-Ponta tal-Hotba u Tal-Fessej in Gozo")	2012 (4)
Site 3	The MT0000103 NATURA 2000 site ("il-Baħar fl-inħawi tad-Dwejra in Gozo")	2012 (10), 2018 (4)
Site 4	Northern reaches of the MT0000105 NATURA 2000 site ("Żona fil-Baħar bejn il-Ponta ta' San Dimitri (Għawdex) u il-Qaliet") known as Daħlet ix-Xmajjar Embayment	2012 (6), 2018 (3)

distributed (e.g. flyers, online articles, explanatory videos) and face-to-face meetings with a number of SCUBA diving centres were organized. This procedure aimed to train SCUBA diving instructors and their students to recognize *P. nobilis* and *P. rudis* and to estimate the size of the shell in terms of height above the seabed. An online platform was developed and published as a user-friendly data submission portal (http://ioi.research.um.edu.mt/pinna-nobilis-survey/), through which the *ad hoc* questionnaire and sighting reports could be consulted and/or uploaded. Photo documentation was always required in order to validate the report, thus avoiding misidentification between the two species.

All underwater visual censuses and citizen science data collected were organised in a database, analysed and plotted through GIS (ArcMap GIS 10.8) by layering up the different sources of data and sampling methodologies of presence/absence of *P. nobilis* and *P. rudis*, generating distribution maps of the study areas for the *P. nobilis* population status between 2006 and 2019.

Results

A total of 79 live fan shells was recorded from all the census survey stations during the period of study (2006-2019), consisting of 60 *P. nobilis* and 19 *P. rudis* individuals.

Population dynamics between 2006 and 2012 at Site 1 (MT0000101)

A benthic area of 2597 m^2 at a depth between 6.4 m and 38 m (average 23.04 m) was surveyed in 2006 and in 2012. During the underwater visual census survey conducted in 2006, forty-eight live specimens of *P. nobilis* were recorded and tagged, with an average density of 1.8 individuals/100 m^2, reaching the highest density at Station 1 (5.1 individuals/100m^2). The mean size of the tagged individuals was that of 34.6 ± 11 cm. In 2012, none of the previously-tagged individuals were found, representing a mortality rate of 100% in five years. During the same year, only six live *P. nobilis* individuals were recorded, along with eighteen empty shells/fragments, resulting in an average live individual density for the same area of 0.2 individuals/100 m^2. The mean height of the live *P. nobilis* individuals was that of 25.6 cm ± 10.8 (Fig. 2).

Population assessment in 2012 for Sites 2, 3 and 4

An area of 5148 m^2 at a depth between 6 m and 35 m (average 13.7 m) was surveyed through an underwater visual census. At site 2, no living individuals were found, but only two empty *P. nobilis* shells. At site 3, across the ten survey stations, five individuals of *P. nobilis* were recorded, with a total density of 0.25 individuals/100 m^2 and a mean height of 18.3 cm ± 6.3, along with ten empty shells. At Site 4, across the six survey stations, two live individuals of *P. nobilis* and one live *P. rudis* were recorded (density = 0.2 individuals/100 m^2), with a mean live individual height of 15.5 cm ± 9.1. A total of eighteen empty shells/fragments were also found across these survey stations (Fig. 3).

Population assessment in 2018 for Sites 1, 3 and 4

An area of 3768 m^2 at a depth ranging between 5 m and 25 m (average 14.1 m) was surveyed through an underwater visual census. At sites 1 and 3, neither living *Pinna* spp. individuals nor empty shells were found. At Site 4, across the three stations, two live individuals of *P. rudis* were recorded (density = 0.2 individuals/100 m^2), with an average size of 11.85 cm, and empty shells of the species were also recorded (Fig. 3).

Citizen Science Survey (2017-2019)

Through the collaboration of volunteer SCUBA divers, thirty-seven presence/absence reports for *P. nobilis* and *P. rudis* were received, but only twenty-one of these were considered for this study, as only these were substantiated with photographic evidence which enabled discrimination between *P. nobilis* and *P. rudis*. Most of the records were submitted directly through social media platforms (e.g., Facebook) or by email. Most of the

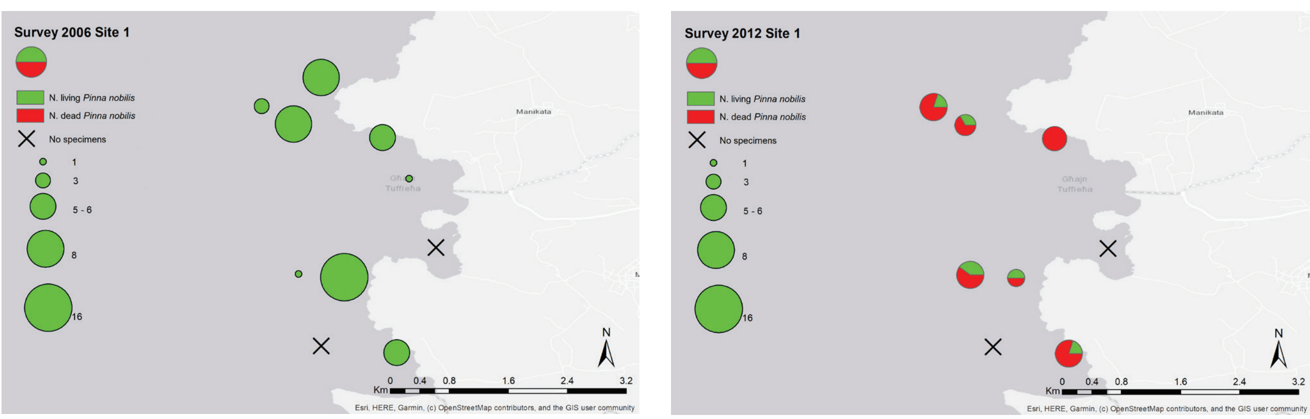

Fig. 2: Comparison of the results of the underwater visual census conducted in 2006 and in 2012 for Site 1. All the specimens recorded belong to *P. nobilis* (no *P. rudis* specimens were found at this site).

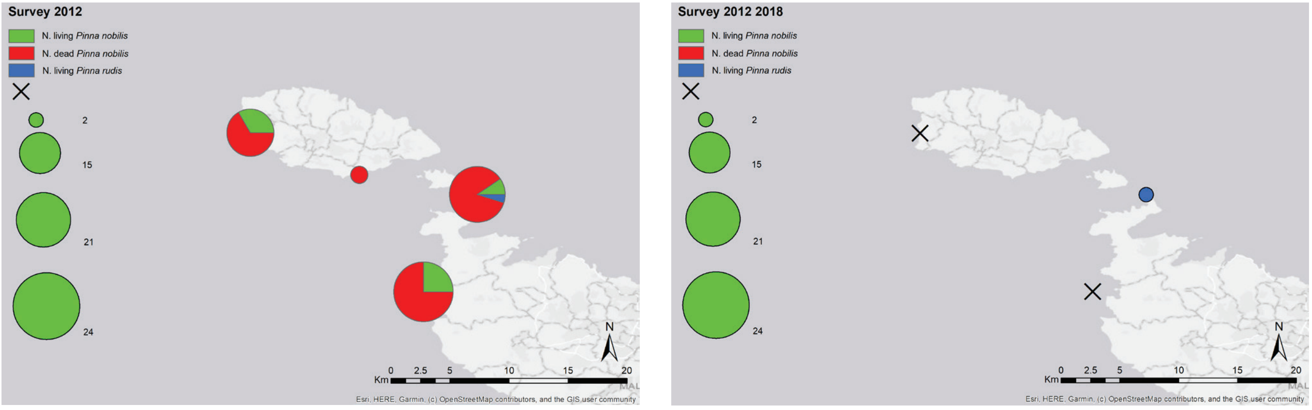

Fig. 3: Comparison of the results of the underwater visual censuses conducted in 2012 (Sites 1, 2, 3 and 4) and 2018 (Sites 1, 3 and 4).

sightings of living specimens referred to *P. rudis* (16), recorded at an average depth of 15.4 m (4–25 m), representing mostly young individuals (0-10 cm = 46.7%; 11-20 cm = 33.3%; 21-30 cm = 20-30%). No empty valves of this species were reported during this exercise. On 19-09-2019, a single live *P. nobilis* individual was recorded at a depth of 25 m (36.080736°N, 14.229153°E) and was estimated to have a height ranging between 20 and 30 cm (Fig. 4). Moreover, five empty shells/fragments of this species were found. All citizen science reports received are mapped in Figure 5.

Discussion

In testimony of the ongoing *P. nobilis* MME (IUCN, 2021; Carella *et al*., 2020; Cabanellas-Reboredo *et al*., 2019;), we here provide the first instance of this phenomenon recorded in Maltese waters, in line with other Mediterranean national observations (Katsanevakis *et al*., 2019; Acarli *et al*., 2020, 2021; Čižmek *et al*., 2020; Zotou *et al*., 2020). The average *P. nobilis* individual density registered during the first monitoring survey conducted in 2006 was that of 1.8 individuals/100 m^2 (max. 5.1 individuals/100 m^2). These density values have been compared with corresponding density values from Tunisia, given its proximity to Malta. *Pinna nobilis* density

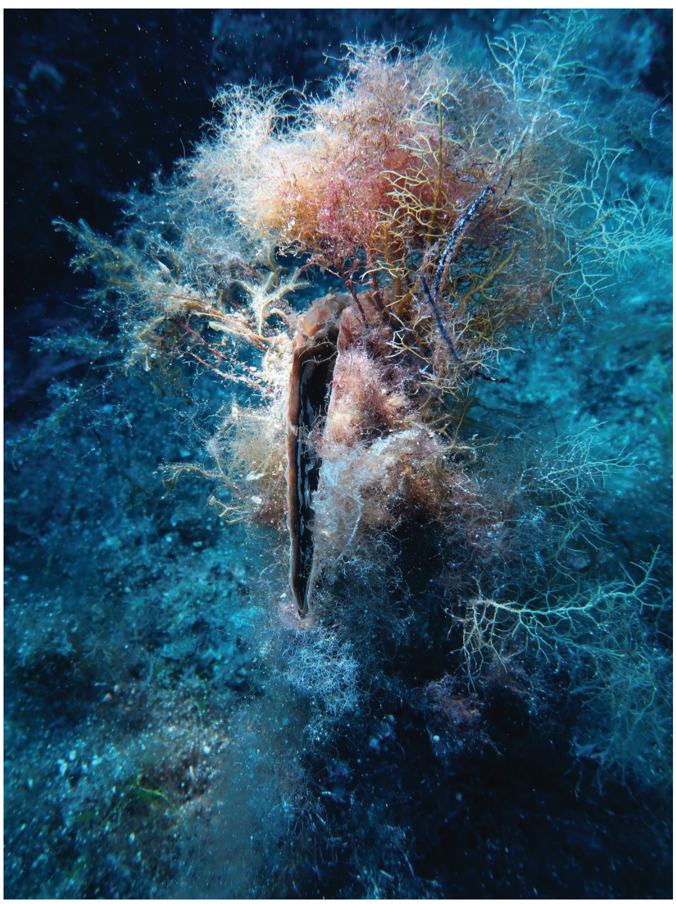

Fig. 4: This picture shows the only *P. nobilis* specimen found during the Interreg MED AMare project Citizen Science campaign.

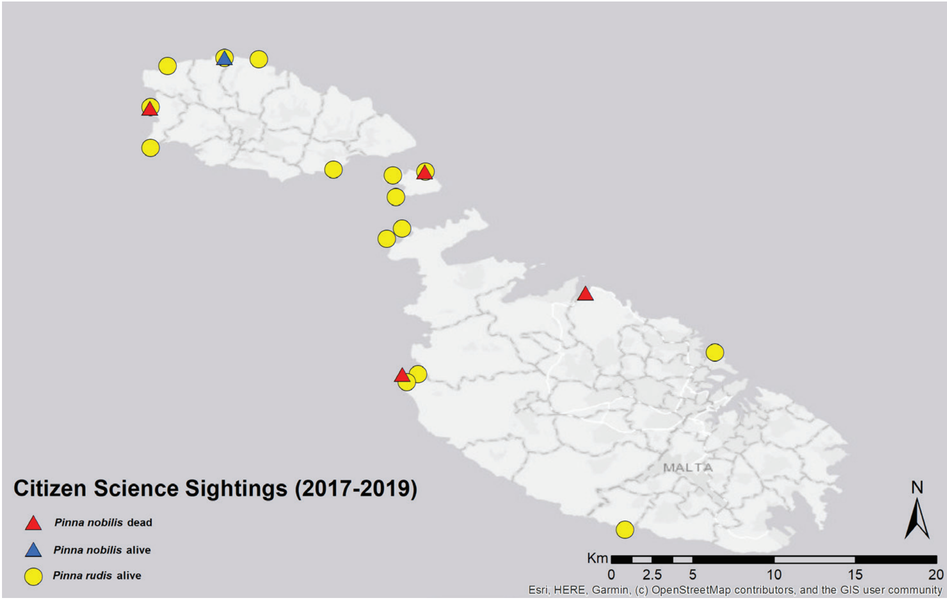

Fig. 5: Citizen science records for *P. nobilis* and *P. rudis* submitted by SCUBA divers for the years 2017-2019.

values from Tunisia, in fact, ranged between 0.02 and 10 individuals/100 m^2 (Rabaoui *et al*., 2008). This result might suggest that anthropogenic impacts were already affecting the Maltese *P. nobilis* populations before 2006, given their low recorded densities, although site-specific abiotic and biotic differences between Maltese and Tunisian waters might be responsible for this observation. A 100% mortality rate was recorded within the same *P. nobilis* populations surveyed in Maltese waters over a span of five years, since none of the specimens tagged in 2006 were found in 2012 and the populations recorded in 2012 were constituted by young individuals only, with an average recorded density of 0.2 individuals/100 m^2. At site 2, which had supported a population of live *P. nobilis* in 2006, only empty shells were recorded during the 2012 survey, representing a dramatic crash of the species at the same site.

This population crash took place before 2016, when the MME was first recorded within the Mediterranean, and the reason for this mortality event should be ascribed to other causes. Öndes *et al*. (2020) report identified, through a stakeholder survey, fishing gears, poaching, pollution and boat anchoring as the main causes of fan mussel population decline.

In 2018, no live individuals of *P. nobilis* were recorded. Since the current study is a semi-quantitative one, we are not in the position to conclude whether this recent population crash was caused by the MME or by human impacts given that no individuals with symptoms of the disease (slow reaction to valves' closure) were recorded. Through the contribution of citizen scientists, new benthic areas were explored within Maltese waters, without the need of additional costly surveys. Even in this case, no signs of infected *P. nobilis* individuals were observed, since the only recorded living specimen was in good health. The higher occurrence of *P. rudis* compared to that of *P. nobilis* recorded by SCUBA divers during the 2018 citizen science survey may suggest that *P. rudis* is more resistant to disturbance and/or infections than *P. nobilis*. In fact, Vázquez-Luis *et al*. (2017) reports that *P. rudis* was not or was less affected by the *P. nobilis* MME. The low number of recorded empty *P. nobilis* valves might be explained by the fact that the sites explored by the citizen scientists are profoundly affected by touristic activities, including SCUBA diving itself, boating and snorkelling, with empty *P. nobilis* valves being commonly collected as decorative items. From the observations documented within this study, a relentless decline in the abundance of both *Pinna* species within Maltese waters since 2006 emerges, in particular for *P. nobilis.* This work also shows how citizen science can play an important role in marine ecological monitoring programmes, such as those for *P. nobilis* and *P. rudis* (Kelly *et al*., 2020). Despite the high scientific value of conventional underwater visual censuses, circular transect sampling (the technique we adopted in this study) might not have been adequate to track the occurrence of these low-density species (Irigoyen *et al*., 2018).

With the assistance of citizen scientists, new potential refuges for *P. nobilis* and *P. rudis* were identified. The only living *P. nobilis* specimen recorded during the 2018 citizen science survey (Fig. 4) was discovered along the northern coast of Gozo, in close vicinity to Wied il-Għasri (36.080736°N, 14.229153°E) at a depth of 25 m, an area not specifically explored by the underwater visual census surveys conducted in 2012 and 2018. All the *P. rudis* sightings recorded in this study were made at sites not included within the 2006, 2012 and 2018 visual census survey study areas. This confirms the importance of citizen science as a tool for scouting extensive benthic areas of interest, for providing precious information in terms of suitable habitats for *P. nobilis* and *P. rudis* and for supplementing the information collected through conventional and formal visual census surveys.

Increasing the training and the participation of citizen scientists can result in an increment in submitted report quality and in quantitative data sampling (McKinley *et al*., 2017). As emerged from the recent findings of Vázquez-Luis *et al*. (2021), an increased monitoring effort for the populations of *P. nobilis* and *P. rudis* has become imperative, given that the persistence of the highly endangered *P. nobilis* might depend on its possible natural hybridization with *P. rudis* which in turn might potentially render it immune to *H. pinnae*). For this reason, a more comprehensive spatial and depth-segregated dataset on the real distribution of both species is required. Moreover, Katsanevakis *et al.* (2021) have pointed out the dire necessity of perseverance in underwater exploration efforts, in order to identify unknown populations which are essential for a potential natural recovery of individual abundance values. This goal can be only achieved through a broadening of the knowledge held by expert underwater scientists and citizen scientists. Moreover, while only SCUBA divers were involved in the Interreg MED AMAre project, according to Dalby *et al.* (2021), a larger community inclusivity should be aimed at. The key to the success of a citizen science project is the involvement of a wider spectrum of stakeholders (e.g. fishers, tourists, schools etc.) rather than just that of a narrow spectrum of final users (e.g. SCUBA divers) (Dalby *et al.*, 2021). Fishers, for example, have an unparalleled knowledge on the sea, and with their activities they can provide records of specimens not accessible to SCUBA divers (Öndes *et al*., 2020). The Interreg MED AMare project may represent, in Malta, the first stepping stone in this process of citizen involvement and awareness-generation in terms of the current noble pen shell MME. In this way, stakeholders and users may influence the activities of decision-makers by assisting in the formulation of more effective marine conservation plans and in the establishment of productive MPAs within Maltese waters. In conclusion, this work represents the first attempt at fostering the awareness of citizens on the conservation-related challenges facing the endangered *P. nobilis* and *P. rudis* species, in a drive to nurture an ocean-literate public as prescribed by the UN's Ocean Sciences Decade.

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