

Engagement of fishers in citizen science enhances the knowledge on alien decapods in Cyprus (eastern Mediterranean Sea)

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Abstract The eastern Mediterranean Sea is facing an unprecedented crisis due to the introduction and spread of alien species through the Suez Canal. Early detection and distribution monitoring of these species has been recently facilitated using social media and citizen science. The project "Is it Alien to you? Share it!!!" recorded over 840 observations for about 100 marine species in Cyprus (eastern Mediterranean Sea) primarily via a dedicated social network group. Through the utilization of morphological and molecular approaches on materials posted by a local fisher, this study provides the first records of *Ixa monodi* and *Myra subgranulata* in Cyprus and additional records of *Macrophthalmus indicus* for the island. These sightings confirm the effectiveness of citizen science

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in monitoring species distribution shifts. The cooperation of scientists with fishers has the potential to enhance surveillance, while concurrently increase awareness and position citizens as conservation leaders.

Keywords Brachyura · Coastal monitoring · Lessepsian species spreading · Levant Sea · Nonnative species

Introduction

Increasing anthropogenic pressures and climate change are threatening the Mediterranean marine ecosystems and the biodiversity that underpin ecological functions and processes (Coll et al. 2010; Katsanevakis et al. 2014). The introduction and spread of alien species are considered major threats to local marine ecosystems, economies, and human health (Bax et al. 2003; Tsirintanis et al. 2022). A major biogeographical change in the Mediterranean biota occurred with the opening of the Suez Canal in 1869, which connected the Red Sea and the Mediterranean and led to the introduction of Indo-Pacific species into the basin, a phenomenon later called as "Lessepsian immigration" (Por 1978). The subsequent expansion of the Suez Canal from 304 m² in 1869 to 5200 m² in 2010 enhanced propagule pressure and affected community composition in the Mediterranean Sea (Galil et al. 2017). Under this scenario, it is possible that the native stenothermal biota of the Mediterranean Sea might be unable to cope with climate changes and will be slowly replaced by Erythraean alien species (Moullec et al. 2019).

Under the unfolding climate change, information about alien species presence and distribution are crucial in order to understand the biological changes that occur in the Mediterranean Sea, provide insights on the species' potential ecological and economic effects, and inform management actions to limit the damages (Bariche et al. 2020; Kleitou et al. 2021a). Cyprus is the easternmost Mediterranean island, located in the Levant Sea and at the frontline of the Suez Canal, and is the first country of the European Union affected by Lessepsian immigrations (Kleitou et al. 2019). Despite its strategic position, it remains one of the countries in the region with relatively low documented numbers of alien species, mostly due to the paucity of targeted surveys (Crocetta et al. 2015; Iglésias and Frotté 2015; Kleitou et al. 2019).

The increasing use of citizen science in the island, facilitated through online platforms and mobile applications, has proved powerful and cost-effective in gathering large amount of data and overcoming limitations related to conventional monitoring (Bariche et al. 2020; Crocetta et al. 2015; Langeneck et al. 2022). Several new alien species of different phyla have been recently recorded based on the volunteered contributions of citizen-scientists. These include the jellyfish Phyllorhiza punctata von Lendenfeld, 1884 (see Kaminas et al. 2022), the sea slugs Goniobranchus obsoletus (Rüppell & Leuckart, 1830), Plocamopherus ocellatus Rüppell and Leuckart, 1828 and Haminoea cyanomarginata Heller and Thompson, 1983 (see Crocetta et al. 2015; Yokes et al. 2018; Kleitou et al. 2019), the decapods Atergatis roseus (Rüppell, 1830) and Saron marmoratus (Olivier, 1811) (see Crocetta et al. 2015; Langeneck et al. 2022), and the fish Cheilodipterus novemstriatus (Rüppell, 1838), Lutjanus argentimaculatus (Forsskål, 1775), and Oxyurichthys petersii (Klunzinger, 1871) (see Crocetta et al. 2015; Langeneck et al. 2022).

In the present study, we widen the knowledge on the alien decapod fauna of Cyprus through a combination of citizen science and laboratory approaches. Potentially interesting samples posted online were retrieved from a fisher and analysed through external morphology and (whenever possible) DNA barcoding. This led to the first record of the Lessepsian brachyuran species *Ixa monodi* Holthuis & Gottlieb 1956 and *Myra subgranulata* Kossmann, 1877 for Cyprus, and to additional sightings of *Macrophthalmus indicus* Davie 2012 for the country.

Methods

Citizen science project description and specimen recovery

The project "Is it Alien to you? Share it!!!" was established in 2016 by the Greek environmental organization iSea to facilitate citizen science through gathering data of alien species in Greece and Cyprus (Giovos et al. 2019). Since its inception, the project has actively engaged volunteers (citizen scientists) who can upload photographic material, videos, and other forms of observation on the online platform of the project and can complement their records with additional information such as specimen size, number of specimens, depth, substrate type, location, date, and type of observation (freediving, underwater photography, shore-based fishing, boat-based fishing, spearfishing, etc.). The project has expanded its reach, now encompassing a Facebook Group (www.faceb ook.com/groups/104915386661854/) and a Google Form to assist the data upload process. Additionally, the presence of taxonomists specializing in marine alien species enriches the project by identifying and validating the submitted observations (see Giovos et al. 2019). Sightings are identified to the lowest possible taxonomic level and assessed based on their validity and reliability. Once potentially interesting specimens are posted online, reporters are contacted for further information, and (whenever possible) specimens are retrieved and brought to the laboratories of iSea (Greece) or MER-Marine and Environmental Research (Cyprus) for subsequent analyses.

To date, more than 840 observations by 240 different users were posted from Cyprus, accounting for about 100 species (iSea database). Among them, crab specimens belonging to three different species were uploaded on the Facebook group of the project by a professional fisher (George Karamanos), between July 9 and September 22, 2022. Due to their morphology, and presumably accounting for alien species, some of which yet undetected for the country, they were subsequently retrieved for morphological/meristic and genetic analyses.

Morphological analyses

Retrieved specimens were identified to species level following the published literature and paying attention to species-specific diagnostic features. Then, they were weighed with a scale (accuracy 0.001 g) and morphological parameters (carapace and claws) were measured with a digital calliper (accuracy 0.01 mm).

Molecular analyses

Once morphological identifications were conducted, total genomic DNAs were extracted from the fifth pereiopod of the samples, using the DNeasy® Blood & Tissue kit (QIAGEN) and following the manufacturer's protocol. Partial sequences of the cytochrome c oxidase subunit I (COI) gene were amplified for M. subgranulata with the primers COL6b (5'-ACAAATCATAAAGATATYGG-3') and COH6 (5'-ADACTTCDGGRTGDCCAAARAAYCA-3') (Schubart and Huber 2006), whereas for M. indicus the primers dgLCO-1490 forward (5'-GGTCAACAA ATCATAAAGAYATYGG-3') and COIex reverse (5'-CCAGGTAAAATTAAAATATAAACTTC-3') (Carpenter and Wheeler 1999; Meyer 2003) were employed. For both species, polymerase chain reactions (PCRs) were conducted in 25 µl volume reactions as in Tanduo et al. (2021a). Amplifications were performed as in Tanduo et al. (2021b) with the specific annealing temperature of 48 °C for M. subgranulata and 45 °C for M. indicus. PCR products were purified and Sanger sequenced as in Tanduo et al. (2021b). Chromatograms were checked, assembled, and edited using BioEdit version 7.0.0 (Hall 2011). Obtained sequences were compared with reference sequences from the NCBI nucleotide (NT) database (Morgulis et al. 2008) and from the Barcode of Life Data Systems (BOLD) (Ratnasingham and Hebert 2007), and finally, deposited in GenBank with the codes OQ455951 and OQ455950, respectively. Molecular analyses were not carried out on the sample of I. monodi since the specimen was posted some days after its catch, and the fisher preserved it dry.

Distribution mapping and updated nomenclature

An extensive bibliographic search was conducted to document the previous records of the three alien decapods in the Mediterranean Sea. Peer-reviewed and grey literature indexed in Google Scholar were searched using the keywords: "Mediterranean", the operator AND, and the keywords (1) "Ixa monodi", (2) "Myra subgranulata", (3) "Myra fugax", (4) "Macrophthalmus indicus" and (5) "Macrophthalmus graeffei", published "anytime". The title and the abstract of the retrieved studies were then previewed individually, and each record was carefully analysed. Nomenclature and systematics were equalized to the latest nomenclature available using WoRMS (www.marinespec ies.org/, last accessed on 7 February 2023). Retrieved records of I. monodi, M. subgranulata, and M. indicus were finally divided into periods (≤ 1980 ; 1981–2000; $2001-2010; 2011-2020; \ge 2021$) and illustrated in a map using the Quantum GIS software, version 3.28.3 (www.qgis.org/en/site/).

Results

The three crabs were identified as *Ixa monodi*, *Myra subgranulata*, and *Macrophthalmus indicus*. Details about the location, habitat, gear, and depth of the citizen science sightings are reported below, together with the results of the subsequent morphological and genetic examinations that led to their identification.

Family Leucosiidae Samouelle, 1819.

Ixa monodi Holthuis & Gottlieb 1956.

Material examined: $1 \bigcirc$, 59.37×22.83 mm, Latsi (~35.053439, 32.387263), 9.VII.2022, sandy and muddy bottom, 18 m depth, trammel nets (Fig. 1a).

Brief morphological description: carapace cylindrically ovoid with two longitudinal grooves and two shallow channels on posterior part. Surface covered by tubercules, increasing in number on extremities. Bilobed frontal region. Deep orbits covering eyes. Anterolateral margin with three distinct parts. Posterior lateral margin with two submedian granular tubercules. Chelipeds long and slender with mobile part longer than others. Pereopods long and thin. Additional measurements are listed in Table 1. Carapace pinkish in colour with dark tubercles. Pinkish-orange chelipeds. Morphological features agree with those reported by Holthuis and Gottlieb (1956) and Galil et al. (2002).

Distribution: native to the Red Sea (Monod 1938), it was first recorded in the Mediterranean Sea from Mersin Bay (southern Turkey) in 1956 (Holthuis and Gottlieb 1956). It is now a widespread species in the eastern Mediterranean, with the westernmost Mediterranean record located in Egypt (Fig. 2a; Supplementary material 1). The present record constitutes the first record of the species for Cyprus.

Myra subgranulata Kossmann, 1877.

Material examined: $1 \bigcirc$, 30.82×38.55 mm, Latsi (~35.050342, 32.366850), 15.IX.2022, sandy bottom, 12 m depth, trammel nets (Fig. 1b).

Brief morphological description: carapace rounded, globular, finely granulated with poorly defined regions. Frontal region produced, curved upwards, and with V-shaped anterior margin. Deep orbits with retractable eyes. Lateral margins minutely beaded. Posterior margin with three long spines, with median longer than others. Chelipeds with cylindrical claws finely denticulate in inner margin. Pereopods long, slender, and smooth. Additional measurements are listed in Table 1. Brownish-orange carapace with darkish patches. Orange chelipeds with light dactyls. Morphological features agree with those reported by Galil (2001) and Galil et al. (2002).

Molecular results: a 606 base pairs (bp) partial sequence of the COI gene was obtained. No conspecific sequences were available in GenBank, as scores \geq 85.69% were obtained with several brachyuran species. On the other hand, BOLD queries generated a 99.34–100% score with the unpublished sequences BIM683-19.COI-5P, BIM667-19.COI-5P, BIM210-13.COI-5P, and BIM705-19.COI-5P, all deposited as *M. subgranulata* from Israel.

Distribution: native to the Western Indian Ocean, from Red Sea to Madagascar (Galil 2001), it was first recorded in the Mediterranean Sea from Israel in 1929 (Monod 1930). It is now a widespread species in the eastern Mediterranean, often misidentified in the past as *Myra fugax* (Fabricius, 1798), with the westernmost Mediterranean record located in Rhodes (Greece) (Fig. 2b; Supplementary material 1). The present record is the first documentation of the species in Cyprus.

Family Macrophthalmidae Dana, 1851.

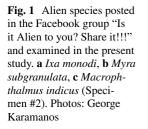
Macrophthalmus indicus Davie 2012.

Material examined: 2 3, 22.5×13.3 mm and 28.37×12.49 mm, Latsi (~35.049559, 32.371328), 22.IX.2022, sandy bottom, 12 m depth, trammel nets (Fig. 1c).

Brief morphological description: carapace wider than long with well-defined regions. Frontal region constricted medially with lateral angles pointed. Lateral margin with two distinct teeth behind exorbital tooth. Exorbital tooth slender and separated from second by a deep V-shaped notch. Remarkably long eyestalks, extending beyond lateral carapace margins. Distinct developed style at end of cornea. Chelipeds subequal. Merus with denticles on outer margin. Pereopods medium length, relatively narrow. Additional measurements are listed in Table 1. Brownish in colour with willowish patches. Morphological features agree with those reported by Davie (2012) and Gerovasileiou et al. (2017).

Molecular results: a 646 bp partial sequence of the COI gene was obtained. BLASTn queries in GenBank generated a 100% score with a sequence (OP382629) of a specimen from Rhodes (Greece) deposited as *M. indicus* and obtained from the stomach of silver-cheeked toadfish (Kondylatos et al. 2023), and a lower similarity (\leq 92.52%) with all the other sequences deposited in GenBank. BOLD queries did not provide any record of the species, nor sequences from conspecific samples were available.

Distribution: native to the Indian Ocean, including Red Sea, Gulf of Oman, Persian Gulf, and Indonesia (Davie 2012), it was first recorded in the Mediterranean Sea from Turkey in 1994 (Enzenross and Enzenross 1995). It is now a widespread species in the eastern Mediterranean, often misidentified in the past as *Macrophthalmus graeffei* A. Milne-Edwards, 1873, with the westernmost Mediterranean record located in Rhodes (Greece) (Fig. 2c; Supplementary material 1). This species was previously recorded from Cyprus by Gerovasileiou et al. (2017) and Katsanevakis et al. (2020).



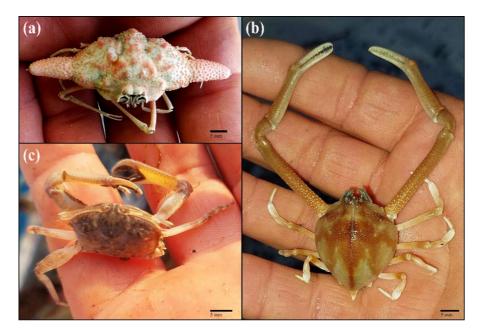


Table 1	Morphometric			
measurements of				
Ixa mono	odi, Myra			
subgrani	<i>ilata</i> , and			
Macrophthalmus indicus				
from Cy	prus			

Species	Ixa monodi	Myra subgranulata	Macrophthal- mus indicus #1	Macrophthal- mus indicus #2		
Weight (g)	4.354	11.365	1.552	1.504		
Carapace measurements (mm)						
Maximum carapace width	59.37	30.82	22.50	28.37		
Maximum carapace length	22.83	38.55	13.30	12.49		
Abdomen width	3.12	6.09	8.30	7.50		
Claw measurements (mm)						
Left: Propodus length	17.24	34.26	NA	14.38		
Left: Propodus width	1.72	7.02	NA	7.18		
Left: Propodus depth	1.65	5.59	NA	2.52		
Left: Dactyl length	4.62	NA	NA	6.64		
Right: Propodus length	18.64	34.35	NA	17.21		
Right: Propodus width	2.07	6.95	NA	6.08		
Right: Propodus depth	1.53	5.49	NA	2.64		
Right: Dactyl length	4.53	16.5	NA	7.24		

NA not available

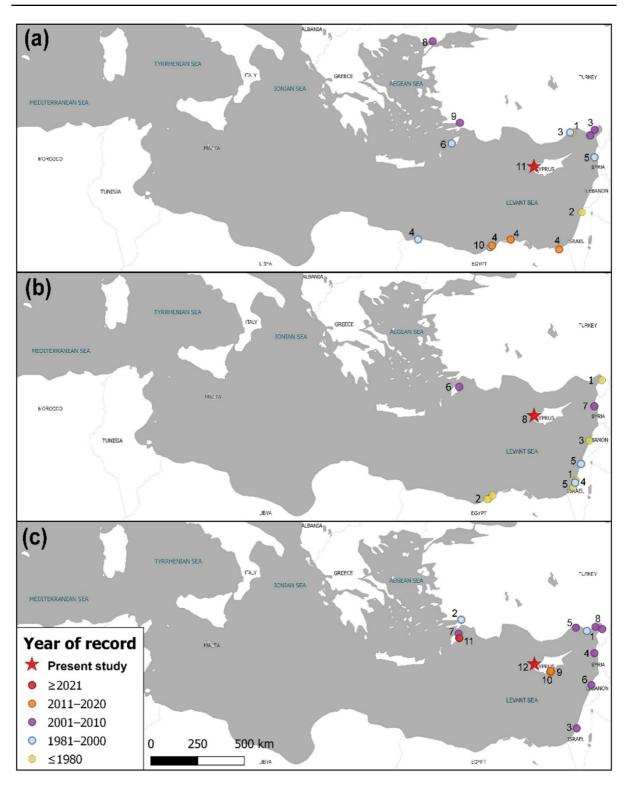


Fig. 2 Published Mediterranean records of the alien crab species examined in the present paper, divided into periods (≤ 1980 ; 1981–2000; 2001–2010; 2011–2020; ≥ 2021). a *Ixa*

monodi, **b** *Myra subgranulata*, and **c** *Macrophthalmus indicus*. Red star indicates records found in the present study. References in Supplementary material 1

Discussion

The advances in information technology infrastructure, including the availability of affordable photo and video equipment, coupled with the increasing use of social networks provide unprecedented opportunities for citizens to contribute with real data and verifiable observations about the natural world (Fraisl et al. 2022). Involving volunteers in data collection can have immediate advantages such as increased social and environmental awareness, improved data collection, and coverage of large areas at a lower cost (Giovos et al. 2019; Kleitou et al. 2021b). The present study confirms the potential of citizen science as a powerful and cost-effective monitoring tool and highlights the importance of close collaboration between fishers and scientists.

In the Mediterranean Sea, the number of alien decapods migrating through the Suez Canal is increasing at an accelerating rate, and more than 89 alien decapods have been recorded to date (Zenetos et al. 2022). The new records found in this study add substantial information on the occurrence of decapod alien species in Cypriot waters, increasing the number of alien decapods recorded up to 2022 to 20, of which 12 are brachyurans (Table 2; Supplementary material 2), whilst the specimens of *M. indicus* provide additional reports of the species in Cypriot waters and the first genetic evidence of its presence in the country.

The eastern Mediterranean is highly susceptible to marine biological invasions because of its location at the crossroad between the Ponto-Caspian and the Indian Ocean/Red Sea regions, the maritime traffic from the Indian Ocean, the widespread occurrence of fish and shellfish farms, and the high warming rates (Galil and Zenetos 2002). The Mediterranean distribution of all three species recorded in the present study is constrained in the eastern region. This distribution indicates that the introduction pathway of the species to the Mediterranean Sea was primarily the Suez Canal, whilst for Cyprus the pathway of introduction was the Suez Canal or a secondary dispersal from surrounding countries. Myra subgranulata and I. monodi were previously recorded in Rhodes (Galil and Kevrekidis 2002; Corsini and Kondilatos 2006), north-west of Cyprus, indicating that the two species might have been already present in Cyprus but overlooked, and highlighting the need for additional efforts in early detection of alien species in the island.

 Table 2
 Alien decapods recorded in Cyprus (eastern Mediterranean Sea)

#	Family	Species
1	Hippolytidae	Saron marmoratus
2	Leucosiidae	Ixa monodi
3		Myra subgranulata
4	Macrophthalmidae	Macrophthalmus indicus
5	Matutidae	Matuta victor
6	Palaemonidae	Urocaridella pulchella
7	Penaeidae	Metapenaeopsis aegyptia
8		Metapenaeus monoceros
9		Penaeus pulchricaudatus
10		Penaeus semisulcatus
11	Pilumnidae	Pilumnopeus vauquelini
12	Portunidae	Callinectes sapidus
13		Carupa tenuipes
14		Charybdis longicollis
15		Charybdis hellerii
16		Gonioinfradens giardi
17		Portunus segnis
18		Thalamita poissonii
19	Scyllaridae	Scyllarus caparti
20	Xanthidae	Atergatis roseus

Species recorded for the first time in this study are shown in bold. References in Supplementary material 2

Climate change is expected to increasingly favour pelagic, thermophilic, and generally alien species of Indo-Pacific origin (Moullec et al. 2019). More EU surveillance effort could be strategically placed in Cyprus and Levantine Sea where alien species are first introduced before they spread to the wider Mediterranean region.

Although fishery catches are intensively monitored by the national authority, local protocols might not be efficient in monitoring alien species. Indeed, fishery reporting schemes are valuable for monitoring targeted fish stocks but may not be specifically designed to effectively monitor alien species, leading to potential deficiencies in data collection and reporting. For instance, Kleitou et al. (2022) have already highlighted several limitations such as the possible exclusion of recently introduced species from the monitoring and reporting system, recording species in lower taxonomic resolution or aggregated categories, and/or misreporting species. All three species recorded here were already reported by various authors in the Mediterranean Sea, but *Ixa monodi* and *Myra subgranulata* have never been observed along the Cyprus coasts, while *Macrophthalmus indicus* was scarcely reported. The fisher reported that he occasionally encounters all three brachyuran species in his catches. The repeated observations of the three alien species indicate that they are under establishment in this region and that they have likely found appropriate conditions for their survival and reproduction. The results highlight the importance of improving current surveillance systems and the potential contribution of local ecological knowledge of fishers in monitoring species distribution shifts (Azzurro et al. 2019).

Citizen science has proved particularly effective in detecting new species, especially when combined with adequate scientific support and validation mechanisms. Learning through interaction was a critical motivation factor, according to the fisher who reported the sightings and provided the specimens. Kleitou et al. (2021b) have demonstrated that initiating close communication with divers and providing them a logbook and some basic training can yield vital sufficient data and inform about spatiotemporal changes of lionfish. Similar initiatives, dedicated to alien species and designed to monitor their detection, spread and impacts, could be promoted for fisheries. The close collaboration and engagement of fishers with the conservation sector has the potential to create long-term benefits, enhance surveillance while concurrently increasing awareness, creating new opportunities, and positioning fishers as conservation advocates (Kleitou et al. 2021c; Quintana et al. 2022). Fishers have a critical role in controlling alien species, and their awareness can secure their participation in management efforts, facilitating the implementation of EU Regulations for early monitoring of alien species.

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Data availability All data generated during this study are included in this published article and its supplementary information files.

Declarations

Conflict of interest The authors declare that they have no conflict of interests.

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