



Seasonality of the Alien *Lamprohaminoea ovalis* (Pease, 1868) (Gastropoda: Cephalaspidea) along the Central-eastern Coasts of Sicily, Central Mediterranean Sea

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Abstract: *Lamprohaminoea ovalis* is an alien species from the Indo-West Pacific, which has colonised almost all the Mediterranean Sea in the last twenty years. Although this species has been widely reported from the invaded area, its biology and ecology are still poorly known. The aim of the present study was to investigate the seasonality of this species in an invaded area, the central-eastern coast of Sicily (Italy). An underwater visual census with scuba diving has been carried out at three stations (Santa Tecla, Santa Maria La Scala and Catania) in 2017–2020 to collect data on seasonality of *L. ovalis*. Populations appear at the end of summer (August), reach their peak in autumn, decrease at the beginning of winter and disappear at the beginning of spring. This seasonal trend could depend on the fact that this species may have a warm water affinity and, thus, it may fail to survive in cold periods. In addition, breeding activities have been documented each year.

Key words: non-indigenous species, *Haminoea cyanomarginata*, Ionian Sea, Italy

Introduction

The genus *Lamprohaminoea* Habe, 1952 includes colourful species usually living in tidal and shallow waters of tropical and subtropical areas of the Indo–West Pacific (GOSLINER et al. 2008, 2015, APTE & DESAI 2017, TIBIRIĆA & MALAQUIAS 2017). One of them, *Lamprohaminoea ovalis* (Pease, 1868) (widely reported in the previous literature as *Haminoea cyanomarginata* Heller & T. E. Thompson, 1983), has invaded the Mediterranean Sea, where it is now widespread (ZENETOS et al. 2004, RAGKOUSIS et al. 2020). *Lamprohaminoea ovalis* has a whitish-translucent elongated body with purple lines along the edges of the cephalic shield and of the parapodial and the pallial lobes, sometimes also showing yellow rounded blotches all over the body (OSKARS & MALAQUIAS 2020). The cephalic shield is deeply bifurcate posteriorly and divided into a pair of well-

developed flaps (CROCETTA & VAZZANA 2009). The eyes are spaced and the pericocular area is sometimes pigmented with colourful blotches (OSKARS & MALAQUIAS 2020). Its parapodial lobes are short and do not meet dorsally (OSKARS & MALAQUIAS 2020).

The original distribution of this species is in the Indo-West Pacific, from the Red Sea (off Sudan, Eritrea, Saudi Arabia, Daymaniyat Islands and Oman) to the western and central Pacific (off Philippines, Vanuatu, Line Islands, Okinawa, Japan, Marshall Islands, Mariana Islands and Guam, eastern Australia, Queensland and New South Wales, Tahiti, French Polynesia and Hawaii) (OSKARS & MALAQUIAS 2020). In the Mediterranean (Table 1), this species has been recorded for the first time in 2001 off Greece (RUDMAN 2003, ZENETOS et al. 2005). Later on, it has been found in 2002 off Turkey (RUDMAN 2003, Çinar et al. 2011), in 2006 at Malta (MIFSUD 2007), in 2007 off Italy (CROCETTA

& VAZZANA 2009, CROCCETTA et al. 2009), in 2016 in the Adriatic Sea off the Croatian coast (PONTES et al. 2012–2020), in 2017 and 2020 at Spain (at Mallorca, Nova Tabarca Island and Ibiza) (FERNÁNDEZ-VILERT et al. 2018, RAGKOUSIS et al. 2020), in 2018 at Cyprus (YOKEŞ et al. 2018) and in the same year off Libya (RIZGALLA et al. 2018). Along Italian coasts, this species has been reported at Calabria, Campania and Sicily and is generally considered as an established alien species (SERVELLO et al. 2019). In particular, *L. ovalis* has been recorded for the first time at Calabria in 2007 at Saline Joniche (Reggio Calabria) (CROCCETTA & VAZZANA 2009). Later, in 2009 *L. ovalis* has been found at Lazzaro (Reggio Calabria) (CROCCETTA et al. 2009) and then again at Saline Joniche (Reggio Calabria) and at Scilla (Reggio Calabria) in 2013 (STASOLLA et al. 2014). Off Campania, *L. ovalis* has been reported in 2014 at Massa Lubrense (Naples) (KATSANEVAKIS et al., 2014) and at Palinuro (TIBERTI et al., 2015). At Sicily, *L. ovalis* has been reported in 2008 at Castellammare del Golfo (Trapani), in 2011 at Pantelleria Island (Trapani), in 2013 at Aci Castello (Catania) (STASOLLA et al. 2014); in 2017–2020, it has been found several times at Santa Maria La Scala and Santa Tecla (Acireale), Aci Trezza (Aci Castello) and Catania (LOMBARDO & MARLETTA 2020).

Although this species has been widely reported, its biology and ecology are poorly known (ZENETOS et al. 2004). Hitherto, *L. ovalis* has been found from just below the sea level to 30 m of depth (RUDMAN 2003, CROCCETTA & VAZZANA 2009, FERNÁNDEZ-VILERT et al. 2018, RIZGALLA et al. 2018, LOMBARDO & MARLETTA 2020) on sandy and rocky substrates covered by algal film, Corallinaceae, *Cystoseira s.l.*, sponges and hydrozoans (CROCCETTA & VAZZANA 2009, CROCCETTA et al. 2009, STASOLLA et al. 2014, RIZGALLA et al. 2018). This species, as the other congeneric species, feeds on diatoms, cyanobacteria and filamentous algae (ZENETOS et al. 2004, CRUZ-RIVERA & PAUL 2006). Nevertheless, there are no data on the seasonality and the life history of *L. ovalis* in both Mediterranean Sea and in Indo–West Pacific area.

The aim of the present study is to document the seasonality of this species in an area of its invaded geographical range, the central-eastern coast of Sicily (Italy).

Materials and Methods

The seasonality of *L. ovalis* has been studied from 2017 to 2020 at different sites located along the central-eastern coast of Sicily (Fig. 1A, B). This area is geologically constituted by a complex of subvolcanic

rocks and effusive marine products forming fields of pillow lavas (SCIUTO et al. 2017). Accumulated along the coast and at the base of shallow cliffs, there is a belt of blocks up to a few metres in size, caused by rocky outcrops dissected by fault systems and partially dismantled (SCIUTO et al. 2017). From the coastline to down to 10–15 m depth, the bottom has a sloping topography and consists of the basaltic bedrock covered with large volcanic blocks (SCIUTO et al. 2017). Offshore, the bottom of sediments becomes progressively muddier at major depths with rocky outcrops locally exposed (ROSSO 2001). Up to 10–15 m of depth, the macrophytobenthos consists mainly of photophilic seaweeds of the orders Sphacelariales and Dictyotales, geniculate coralline algae and turf-forming algae (CATRA et al. 2019), while at deeper localities encrusting Rhodophyta are dominant in the coralligenous habitats (COSTANZO et al. 2021).

The study areas were selected according to the different level of anthropogenic impact. Two sites, Ognina (37°31'50.4" N – 15°07'10.8" E) and Bellatrix (37°32'03.2" N – 15°07'35.2" E), both in the municipality of Catania, were the most anthropogenically-affected among the study areas. Since they were near to one another, they were considered as a single site named Catania. Two other sites, Acque Fredde (37°38'15.7" N – 15°10'52.1" E) and Scalo Pennisi (37°38'23.2" N – 15°11'04.6" E), both located in the hamlet of Santa Tecla (Acireale) and presenting the most natural environmental conditions, were considered as a single site designated as Santa Tecla. Finally, the site of Santa Maria La Scala (37°36'46.5" N – 15°10'31.4" E) (Acireale), located between Catania and Santa Tecla and exhibiting intermediate conditions between the former two sites, was also included in the observations. In these last three sites, there were several springs due to the flow of freshwaters from Etna to the sea (CATRA et al. 2006).

Data on *L. ovalis* were collected through underwater visual census by scuba diving. A total of 305 dives (98 in Santa Maria La Scala, 117 in Catania and 90 in Santa Tecla) were carried out all year round (marine-weather conditions permitting), twice a week, during daylight, between 9:00 and 11:30 am. In each scuba dive (in a range of depth between 0–45 m, according to the seabed geomorphology), the same path was followed and all *L. ovalis* specimens were photographed with an Olympus TG-4 underwater camera and counted *in situ*; the depths in which they had been encountered were registered. Through analysing photographic data, it was also possible to determine the substrates, on which the specimens were found as well as their breeding activities. For each station, graphics and data were processed

Table 1. Records of *Lamprohaminoea ovalis* in the Mediterranean Sea.

Country	Location	Year	References
Greece	Korinthiakos	2001	RUDMAN (2003), ZENETOS et al. (2005)
Turkey	-	2002	RUDMAN (2003), Çinar et al. (2011)
Malta	Id-Delli; Gnejna Bay; Fomm ir-Rih Bay	2006	MIFSUD (2007)
Italy	Saline Joniche	2007	CROCETTA & VAZZANA (2009)
Italy	Castellamare del Golfo	2008	STASOLLA et al. (2014)
Italy	Lazzaro	2009	CROCETTA et al. (2009)
Italy	Pantelleria	2011	STASOLLA et al. (2014)
Italy	Saline Joniche; Scilla; Aci Castello	2013	STASOLLA et al. (2014)
Italy	Massa Lubrese; Palinuro	2014	KATSANEVAKIS et al. (2014), TIBERTI et al. (2015)
Croatia	Kašuni	2016	PONTES et al. (2012-2020)
Spain	Mallorca	2017	FERNÁNDEZ-VILERT et al. (2018)
Italy	Santa Maria La Scala; Santa Tecla; Catania; Aci Trezza	2017–2020	LOMBARDO & MARLETTA (2020)
Cyprus	Protraras	2018	YOKEŞ et al. (2018)
Libya	Tripoli	2018	RIZGALLA et al. (2018)
Spain	Nova Tabarca Island; Ibiza	2020	RAGKOUSIS et al. (2020)

**Fig. 1.** A. Geographical area where this study has been carried out. B. Study areas within the central-eastern coast of Sicily.

through EXCEL. To evaluate the demographic trend of *L. ovalis*, data were calculated as mean number of specimens per season. The seasons were considered as follows: winter (December – February), spring (March – May), summer (June – August) and autumn (September – November). The experimental design adopted was completely randomised and replicated four times. Data were processed to analysis of variance (ANOVA). Owing to the Covid-19 pandemic situation, it was not possible to carry out diving activities in April 2020; therefore, the mean number of *L. ovalis* specimens for this month was calculated through the average number of specimens between March 2020 and May 2020.

Results

Santa Tecla

At Santa Tecla (Fig. 2, Table 2), we observed an overall mean of specimens corresponding to 2.71 ($p < 0.01$) considering all the years of study. In particular, in this site, during 2017, between winter and spring, a decrease in the average number of specimens equal to -86.41% ($p < 0.01$) was noticed. Between this last season and autumn, there was an abrupt increase in the average number of specimens amounting to 5251% ($p < 0.01$). Instead, in 2018, there was an almost stable trend with a low average number of individuals. In 2019, winter was the season with the highest average

Table 2. Raw data showing the average number of specimens in each season and year in Santa Tecla ($p < 0.01$).

Years	Winter	Spring	Summer	Autumn	Overall Means
2017	2.43	0.33	3.00	17.67	5.86
2018	1.67	2.00	1.00	2.00	1.67
2019	5.33	0.33	0.33	1.33	1.83
2020	0.33	0.00	0.00	5.67	1.50
Overall Means	2.44	0.67	1.08	6.67	2.71

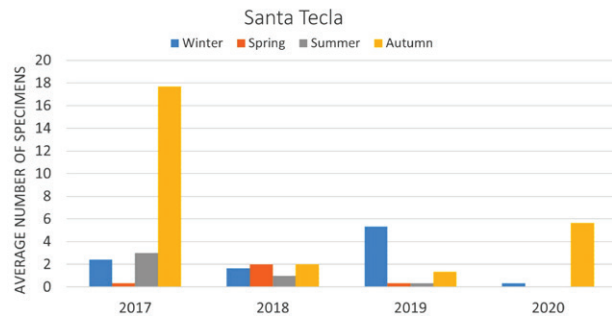


Fig. 2. Oscillations in the average number of *Lamprohaminoea ovalis* specimens found in Santa Tecla during the four years' trial ($p < 0.01$).

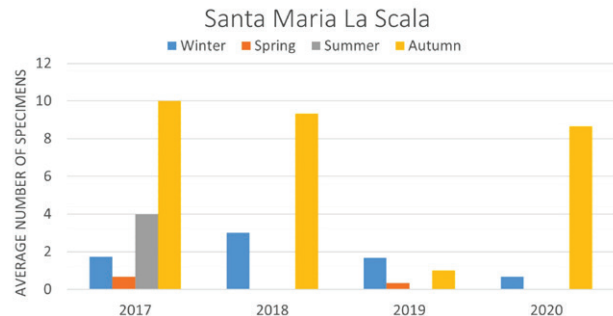


Fig. 3. Oscillations in the average number of *Lamprohaminoea ovalis* specimens found in Santa Maria La Scala during the four years' trial ($p < 0.01$).

Table 3. Raw data showing the average number of specimens in each season and year in Santa Maria La Scala (S. M. L. S.) ($p < 0.01$).

Years	Winter	Spring	Summer	Autumn	Overall Means
2017	1.73	0.67	4.00	10.00	4.10
2018	3.00	0.00	0.00	9.33	3.08
2019	1.67	0.33	0.00	1.00	0.75
2020	0.67	0.00	0.00	8.67	2.33
Overall Means	1.77	0.25	1.00	7.25	2.57

number of specimens (5.33), while spring and summer presented an equal trend, characterised by the almost total absence of individuals. Between winter and these last seasons, it could be noted a decrease in the average number of specimens corresponding to -93.80% ($p < 0.01$). From summer to autumn, there was a rise of 303% ($p < 0.01$) in the average number of specimens. In 2020, the only season with a remarkable presence of individuals was autumn (5.66). Breeding activity of *L. ovalis* at this site was documented in autumn of both 2017 and 2020. Moreover, from 2017 to 2020, no egg masses were found. In Santa Tecla, the specimens were observed in a range of depth of 5.5–24.3 m. In this site, from 2017 to 2020, there was a general decrease in the average number of specimens, from 5.85 in 2017 to 1.5 in 2020 ($p < 0.01$).

Santa Maria La Scala

At Santa Maria La Scala (Fig. 3, Table 3), we recorded an overall mean of specimens considering all the years of study equal to 2.57 ($p < 0.01$).

In this area, between winter and spring of 2017, a decrease in the average number of specimens corresponded to -61.84%. Subsequently, from spring to autumn, there was a gradual rise in the average number of specimens, which generated an evident autumnal peak [1415% ($p < 0.01$)]. In 2018, there was a total lack of specimens in spring and summer while, in winter, the average number of specimens was 3. The autumn was the season with the highest average number of individuals reaching to 9.33. The year with the lowest average number of specimens for this station (0.75) was 2019. Between winter and spring 2019, there was a fall in the average number of individuals equivalent to -80.12% ($p < 0.01$), followed by the total absence of animals in summer; the autumn of 2019 was with an average number of specimens equal to 1. In 2020, the seasonal trend of *L. ovalis* was similar to that of 2018, albeit with a lower average number of individuals. Breeding activities at this site were in summer and autumn of 2017, only in autumn of

2018, in winter and autumn of 2019 and only in autumn of 2020. In this area, no egg masses were found. In Santa Maria La Scala, specimens were observed in a range of depth 5–23.8 m. Moreover, at this site, the average number of specimens decreased from 2017 (4.1) to 2019 (0.75) and then increased in 2020 to 2.33 ($p < 0.01$).

Catania

Overall, among the study areas, Catania (Fig. 4, Table 4) was the site exhibiting the lowest overall mean of specimens considering all the years of study (1.32, $p < 0.05$). In this site, during 2017, there was a difference between the winter trend, which presented an average number of specimens of 0.2 and summer and autumn trends with an average number of specimens of 5.33 and 5.66, respectively. In fact, between winter and summer-autumn peak, there was an increase in *L. ovalis*' population equal to 2675% ($p < 0.05$). Instead, in 2018, almost no *L. ovalis* specimens have been found, with a few individuals observed only in autumn (0.33). In 2019, the seasons in which the presence of *L. ovalis* was registered were winter and autumn, with an average number of specimens of 0.33 and 2.33, respectively. The year 2020 was the only one in which *L. ovalis* specimens were observed throughout all seasons. Winter and spring presented the same trend, with an average number of individuals of 0.33. Between the spring and the summer, an increase in the average number of specimens

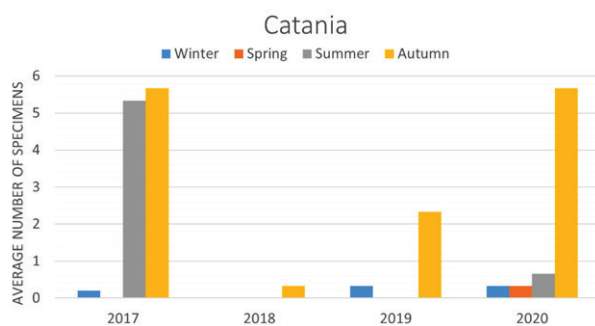


Fig. 4. Oscillations in the average number of *Lamprohaminoea ovalis* specimens found in Catania during the four years' trial ($p < 0.05$).

equal to 100% happened and after it, there was an autumnal peak of 757.57% ($p < 0.05$). In Catania, *L. ovalis*' reproductive activities were seen in summer and autumn 2017 and in autumn 2020. In this site, no egg masses were registered. In Catania, *L. ovalis* specimens were observed in a depth range of 9.8–23.4 m. In this site since 2017 to 2020, there was a general decrease in the average number of specimens, which started from 2.8 in 2017 to an almost disappearance in 2018 and 2019 and, finally, an increase to 1.75 in 2020 ($p < 0.05$).

Other observations

Overall, in the study areas, of the various morphs of *L. ovalis* described by OSKARS & MALAQUIAS (2020), all specimens found represented either the typical *L. cyanomarginata* morph (Fig. 5A) or the morph with the yellow blotches scattered on the body (Fig. 5B). During this study, a faded specimen was also found (Fig. 5C). Furthermore, *L. ovalis* specimens were found, often in numerous groups, on the following substrates: detritus, sand, *Codium bursa* (Olivi) C. Agardh, *Peyssonellia* spp., *Jania* sp., turf-forming Rhodophyta, *Dictyota* spp., *Halopteris scoparia* (Linnaeus) Sauvageau and *Padina pavonica* (Linnaeus) Thivy. Most of specimens were seen exhibiting the typical *L. ovalis* trailing behaviour (Fig. 5D) as documented by RIZGALLA et al. (2018). In addition, a specimen was observed while suspending its body by a thread of mucus to move down.

Discussion

The seasonality of *L. ovalis* has been hereby first explored worldwide, with dense populations mostly found during autumn. Indeed, *L. ovalis* populations appeared at the end of summer (August), reached their peak in autumn, decreased at the beginning of winter and, finally, disappeared at the beginning of spring. This seasonal trend has been uniformly observed at all the studied sites. This is in agreement with observations held in nearby areas (Calabria), where *L. ovalis* was often found in large numbers especially during autumn, e.g., Saline Joniche (70 specimens found in September, see CROCETTA &

Table 4. Raw data showing the average number of specimens in each season and year in Catania ($p < 0.05$).

Years	Winter	Spring	Summer	Autumn	Overall Means
2017	0.20	0.00	5.33	5.67	2.80
2018	0.00	0.00	0.00	0.33	0.08
2019	0.33	0.00	0.00	2.33	0.67
2020	0.33	0.33	0.67	5.67	1.75
Overall Means	0.22	0.08	1.50	3.50	1.32



Fig. 5. A. Dorsal view of the typical *Lamprohaminoea cyanomarginata* morph (Photo G. Marletta). B. Dorsal view of the morph type with the yellow blotches scattered on the body (Photo A. Lombardo). C. Dorsal view of the faded specimen (Photo A. Lombardo). D. An example of the typical trailing behaviour (Photo A. Lombardo). E. Breeding activity (Photo G. Marletta).

VAZZANA 2009) and Scilla (50 specimens found in November, see STASOLLA et al. 2014). This seasonal trend could depend on the fact that this species may have a warm-water affinity and therefore it may fail to survive in the colder periods.

Among the study areas, a north-south gradient of the total average number of specimens was found between the investigated localities: 2.71 in Santa Tecla, 2.57 in Santa Maria La Scala and 1.32 in Catania.

The success of *L. ovalis* in colonizing the Mediterranean is probably related to its chemical toxicity and aposematism. MOLLO et al. (2008) have observed that, unlike the Mediterranean species of the family Haminoeidae Pilsbry, 1895, this alien species has a metabolite with high activity as a feeding deterrent. Furthermore, through its aposematic colouration, *L. ovalis* gives predators the possibility to learn soon to avoid it (MOLLO et al. 2008). In fact, the colour pattern that distinguishes *L. ovalis* from the cryptically coloured Mediterranean *Haminoea* spp. is linked to the presence of a chemical defence that probably has resulted in the successful establishment of this species in its invasive geographical range in the Mediterranean Sea.

Indeed, although *L. ovalis* has so far only been reported through records, the present study is the first being conducted over a long period of

time. It demonstrates that this species is now well-established outside its native range. As evidence of this, in all the study areas, breeding activities (Fig. 5E) have been documented each year between 2017 and 2020. Nevertheless, no egg masses have been found during this study, probably because they have a cryptic whitish-translucent aspect, as reported by CROCETTA & VAZZANA (2009, see fig. 1D). Thus, such egg masses are not easily observable. In fact, since most of the specimens have been seen among turf-forming algae, probably the egg masses were covered from them.

In conclusion, through the present study, it has been demonstrated that *L. ovalis* is a well-established species along the central-eastern coast of Sicily and here its presence is remarkable, especially during autumn. The impacts of this invasive alien species on marine ecosystems are still little known. Therefore, further studies will be needed to evaluate the effects of *L. ovalis* on native marine heterobranchian fauna and the local ecosystems.

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