Journal of the Marine Biological Association of the United Kingdom

cambridge.org/mbi

Original Article

Cite this article: Ordines F, Ramírez-Amaro S, Fernandez-Arcaya U, Marco-Herrero E, Massutí E (2019). First occurrence of an Ophiohelidae species in the Mediterranean: the high abundances of *Ophiomyces grandis* from the Mallorca Channel seamounts. *Journal of the Marine Biological Association of the United Kingdom* **99**, 1817–1823. https://doi.org/10.1017/S0025315419000808

Received: 4 June 2019
Revised: 23 August 2019
Accepted: 27 August 2019
First published online: 30 September 2019

Key words:
Biodiversity; brittle stars; DNA barcoding; Mallorca Channel; seamounts; western Mediterranean

Author for correspondence:
Francesc Ordines, E-mail: xisco.ordinas@ieo.es

First occurrence of an Ophiohelidae species in the Mediterranean: the high abundances of *Ophiomyces grandis* from the Mallorca Channel seamounts

Francesc Ordines, Sergio Ramírez-Amaro, Ulla Fernandez-Arcaya,
Elena Marco-Herrero and Enric Massutí

Instituto Español de Oceanografía, Centre Oceanogràfic de les Balears, Moll de Ponent s/n, 07015 Palma, Spain

Abstract

The first record of the ophiuroid family Ophiohelidae from the Mediterranean Sea is reported. It consists of the description of the new record of *Ophiomyces grandis* from the Mallorca Channel seamounts in the Balearic Islands, western Mediterranean, where it shows high abundances. We present both the morphological description of the individuals collected and, for the first time, the cytochrome oxidase subunit I (COI) sequence of this species. The morphological traits of our specimens match the available descriptions of *O. grandis*. On the other hand, molecular analyses show a large genetic distance between *O. grandis* and *Ophiomyces delata*, the two species being very similar morphologically. Despite the high abundances of *O. grandis* reported here, previous surveys in the Mallorca Channel seamounts using ROV did not detect it, emphasizing the importance of beam trawl sampling to improving the biodiversity description of these geomorphological sea bottom features.

Introduction

The Ophiuroidea (brittle stars and basket stars) is the most speciose class of echinoderms, with at least 2100 described species (Stöhr *et al.*, 2012). The ophiuroid family Ophiohelidae Perrier, 1893, only represented by 19 species, has been recently resurrected by Parameswaran *et al.* (2015). These authors followed Thuy & Meyer (2013) who conclusively separated the genera *Ophiomyces* Lyman, 1869, *Ophiohelus*, Lyman, 1880, *Ophiotholia* Lyman, 1880 and *Ophiothauma* H. L. Clark, 1938, from the family Ophiacanthidae Ljungman, 1867 and placed them back into the family Ophiomycetidae Verrill, 1899, considered by Parameswaran *et al.* (2015) as a junior synonym of Ophiohelidae. More recently, O'Hara *et al.* (2017) placed Ophiacanthidae and Ophiohelidae in different orders: Ophiacanthida O'Hara, Hugall, Thuy, Stöhr & Martynov, 2017 and Ophioscolecida O'Hara, Hugall, Thuy, Stöhr & Martynov, 2017, respectively.

The species belonging to Ophiohelidae are typically deep-water dwellers, not found shallower than 250 m depth except for isolated records from seamounts and the exceptional shallow occurrences of *Ophiomyces delata* Koehler, 1904, at 56 m depth off the Andaman Islands, in the northern Indian Ocean (Parameswaran *et al.*, 2015), and *Ophiomyces papillospinus* Litvinova, 2001, at 75 m depth off Shirihama, Japan, in the Pacific Ocean (Okanishi *et al.*, 2016). Nine species are presently known in the genus *Ophiomyces* (Parameswaran *et al.*, 2015), with none of them recorded so far from the Mediterranean (Koukouras *et al.*, 2007; Mecho *et al.*, 2014; Öztoprak *et al.*, 2014).

Ophiomyces grandis Lyman, 1879 was described from a sample collected at 1860 m depth off Tristan da Cunha. Although it has also been recorded off Tasmania at 1630 m depth, in south-eastern Australia, this species is considered predominantly distributed in the Atlantic Ocean, where it has been recorded, apart from the type locality, from the Rockall Trough to Gibraltar at depths ranging from 230 to 800 m (Paterson, 1985).

Due to the presence of this species in the Pacific Ocean overlapping with the distribution range of *O. delata* and the morphological resemblance of both species, Parameswaran *et al.* (2015) proposed the possibility that they could be synonymous or that together they represent a multi-species complex. Such taxonomic uncertainties have promoted the application of molecular genetics approaches such as DNA barcoding (Hebert *et al.*, 2003), a method that has successfully been used for species discrimination or the discovery of cryptic species complexes (e.g. Baric & Sturmbauer, 1999; Boissin *et al.*, 2008).

The aim of the present work is to describe in detail the specimens of *O. grandis* collected in the Mallorca Channel seamounts, western Mediterranean. We also provide for the first time its COI barcode and compare it genetically to *O. delata*.

Materials and methods

Study area and sampling

The Balearic Islands (western Mediterranean) are located in the Balearic Promontory, a structural elevation 348 km in length, 105 km wide and from 1000 to 2000 m high with respect to the surrounding basins. In the Mallorca Channel that separates the two subunits of this promontory (Mallorca-Menorca and Eivissa-Formentera), three seamounts are located (Figure 1): Ses Olives, with its summit at around 290 m depth, and the Ausias March and Emile Baudot with summits around 90 m depth. While Ses Olives and Ausias March are of continental origin, the Emile Baudot is of volcanic origin (Acosta *et al.*, 2004).

In the Mediterranean, the scientific knowledge about seamounts is marked by large gaps, mainly on the eastern basin, but also by an asymmetry between the amount of geological studies and the biological ones (Würtz *et al.*, 2015). Currently, Ses Olives, Ausias March and Emile Baudot seamounts are being studied within the LIFE IP INTEMARES project. The objective is to improve the scientific knowledge on habitats and species, and human activities, to include these seamounts in the European network of marine Natura 2000 sites.

The first INTEMARES survey in the seamounts of the Mallorca Channel was conducted during July 2018 on board the RV 'Ángeles Alvariño'. A total of 18 samples were taken in daytime (Figure 1) with a Jennings type beam trawl of 2 and 0.5 m horizontal and vertical openings, respectively, and a 5 mm mesh size cod-end. The efficiency of beam trawl for sampling epibenthos has been estimated by Reiss *et al.* (2006). A SCANMAR system was used to control the arrival and departure of this gear to the bottom. Beam trawl samples were of 5 min duration at around 2 knots. Samples were washed on a 1 mm mesh sieve, sorted and identified to species or to the lowest possible taxonomic category. Specimens of unidentified species were preserved for further identification in the laboratory by taxonomic specialists. Specimens of *Ophiomyces* sp. were preserved in absolute ethanol. Abundance and biomass of species were standardized to 500 m^2.

Individuals were analysed under a stereomicroscope Leica M165C equipped with a camera Leica MC170. Identification was done according to Lyman (1869, 1879), Mortensen (1927), Ziesenhenne (1940), Paterson (1985), Litvinova (2001) and Parameswaran *et al.* (2015). The terminology applied for the morphological description follows Stöhr *et al.* (2012) and Hendler (2018). The *O. grandis* individuals were deposited in the Marine Fauna Collection (http://www.ma.ieo.es/cfm/) based at the Centro Oceanográfico de Málaga (Instituto Español de Oceanografía) with the identification reference numbers CFM7020–CFM7029.

Molecular analyses

Total DNA was extracted from a small piece of arm (~2 cm) of one individual of *O. grandis* using the DNeasy Blood and Tissue Extraction kit (QIAGEN). Polymerase chain reaction (PCR) was used to amplify the mitochondrial DNA barcoding fragment (cytochrome oxidase I, COI) with universal primers for Echinodermata EchinoF1/EchinoR1 (Ward *et al.*, 2008). PCR was performed in 25 µl volume (17.2 µl ddH$_2$O, 2.5 µl Mangobuffer (Bioline), 1 µl DNTPs, 1.75 MgCl$_2$, 0.5 µl BSA, 0.5 µl each primer (each 10 pmol), 0.05 µl TAQ (Bioline) and 1 µl DNA). The PCR thermal profile was: initial stage of 96 °C for 5 min; then 35 cycles at 94 °C for 60 s, 50 °C for 60 s and 72 °C for 60 s, followed by a final extension at 72 °C for 10 min. PCR products were purified using the QIAquickR PCR Purification Kit (QIAGEN). Both heavy and light strands were sequenced on an ABI 3130 sequencer using the ABI Prism Terminator BigDye® Terminator Cycle Sequencing Reaction Kit (Applied Biosystems).

Sequences were imported into BioEdit 7.0.5.2. (Hall, 1999) and checked for quality and accuracy with nucleotide base assignment. Multiple sequence alignments (MSA) were obtained with ClustalW (Thompson *et al.*, 1994). The DNA sequences obtained were deposited in the GenBank database (http://www.ncbi.nlm.nih.gov/genbank/) under accession number MK934137. In order to compare our *O. grandis* specimen with other species belonging to genus *Ophiomyces* and to another species with a different genus but within the Ophiohelidae family, we downloaded the COI sequences of the following species from GenBank (https://www.ncbi.nlm.nih.gov/genbank/) and Barcode of Life Data (BOLD; http://www. boldsystems.org; Ratnasingham & Hebert, 2007): *O. delata* (GenBank IDs: KU895350 and HM900450) and *Ophiotholia spathifer* (Lyman, 1879) (GenBank ID: KU895353). In addition, a COI sequence of *Ophiophrura liodisca* H.L. Clark, 1911 (GenBank ID: KU895351), which belongs to the Ophioscolecidae family, a sister family of Ophiohelidae, was included as outgroup. Percentage of genetic distance (*p*-distance) and number of base differences between pairs of DNA sequences were estimated with MEGA v.6 (Tamura *et al.*, 2013). Overall similarities were also assessed with neighbour-joining, using Kimura 2-parameter model (K2P; Kimura, 1980). The tree was produced using MEGA v.6 performing 1000 bootstrap replicates.

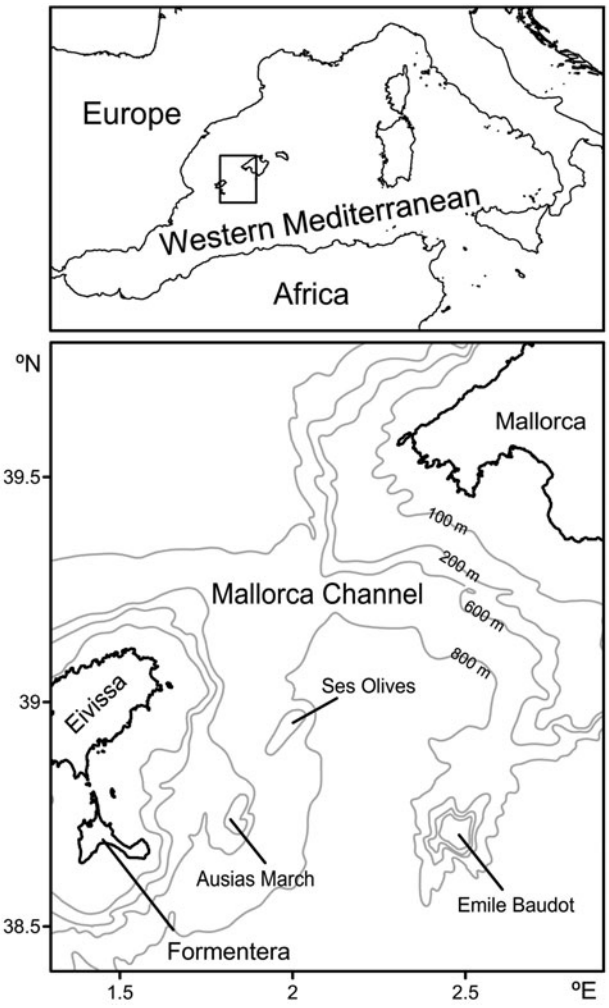

Fig. 1. Map of the study area showing the Mallorca Channel seamounts.

Results

Material

Ophiomyces grandis appeared in the three seamounts surveyed in the Mallorca Channel. A total of 204 specimens of this species were collected from seven beam trawl samples. For the purpose of a detailed description, the 10 individuals in a better state of preservation were selected: three from the Ses Olives seamount (RV 'Ángeles Alvariño', survey INTEMARES-A22B_0718; Stn. 20, beam trawl; 38°56.10′N 01°57.97′E, 275 m depth), three from the Ausias March seamount (same survey; Stn. 37, beam trawl; 38°45.89′N 01°47.36′E, 124 m depth) and four from the Emile Baudot seamount (same survey; Stn. 60, beam trawl; 38° 43.22′N 02°29.49′E, 138 m depth).

SYSTEMATICS

Class OPHIUROIDEA Gray, 1840
Order OPHIOSCOLECIDA O'Hara, Hugall, Thuy, Stöhr & Martynov, 2017
Family OPHIOHELIDAE Perrier, 1893
Genus *Ophiomyces* Lyman, 1869
Ophiomyces grandis Lyman, 1879
Ophiomyces grandis Lyman, 1879: 46, Figures 383–385.
Ophiomyces peresi Reys, 1961: 154, Figures 3–5, photographs a, b.

Diagnosis

Disc sack-like, lacking radial shields, completely covered with scales and with fine spines present. Sack usually raised up. Arms often bent upwards in both alive and preserved individuals. Oral plates completely covered by two rows of flattened oral papillae; six papillae present on the edge row on each oral plate, whereas four papillae are present on the ventral surface row on each oral plate. Fourth tentacle pore with three flattened spatulate tentacle scales on each side of the ventral arm plate. Sixth to 13th–14th tentacle pores with two spatulate tentacle scales on the lateral arm plate and one flat rounded scale on the ventral arm plate. Near the tip of the arm, tentacle pores with only one leaf-like scale, present on the lateral arm plate. Lateral arm plates meeting dorsally and ventrally except in the first two segments where they are separated dorsally by the dorsal arm plates. Eleven arm spines proximally, 9–10 distally, which completely cover the dorsal surface of the arms except for the two first segments.

These characters distinguish our specimens from the rest of valid species in the *Ophiomyces* genus. The arm spines completely covering the dorsal surface of the arms distinguish them from *Ophiomyces altissimus* Litvinova, 2001, *Ophiomyces nadiae* Litvinova, 2001, *O. papillospinus* and *Ophiomyces danielae* Litvinova, 2001. The number of tentacle scales attached to the ventral arm plates, up to three pairs in our individuals, distinguishes them from *Ophiomyces multispinus* Ziesenhenne, 1940, up to four pairs, and *Ophiomyces frutectosus* Lyman, 1869, up to two pairs. The lateral arm plates meeting dorsally beyond the second segment in our specimens also distinguishes them from *O. multispinus*, for which lateral arm plates meet dorsally beyond the third segment. The number of tentacle scales attached to the ventral arm plates and the shape of distal tentacle scales attached to the lateral arm plate (leaf-like in our specimens) distinguishes our specimens from *O. delata*, which proximally has up to two tentacle scales attached to the ventral arm plates, whereas distally, tentacle scales attached to the lateral arm plate are conical and spine-like. The number and relative size of the arm spines in our specimens, up to 11 very unequal in size, distinguish them from *Ophiomyces mirabilis* Lyman, 1869, with up to six arm spines nearly equal in size.

Description

Disc sack-like, globose, completely covered by fine scales, with pointed spines in the inter-radial areas, smaller but more broadly distributed on the ventral side (Figure 2A–D). Radial shields absent. Disc diameter 4.9–6.5 mm, disc height 3.4–5.2 mm, length of the arms 3–3.2 times disc diameter. Jaw width similar to jaw length. Teeth large, flat and broad, resembling a leaf, distally ending in a point (Figure 2E). Below teeth, there are three spine-like crest papillae, the one in the centre follows the alignment of teeth and is flanked by the other two which are in line with the outer row of oral papillae covering the edge of each oral plate. Another row of oral papillae covers the ventral surface of each oral plate. First (most proximal) oral papilla on the outer row of papillae with similar shape to crest papillae, then second, third, fourth, fifth and sixth become progressively larger and flatter, the sixth much wider than the fifth, almost triangular and located between the edge and ventral surface rows of oral papillae; ventral surface row of papillae with four oral papillae, similar in size and shape to fourth papillae in the edge row, blunt and slightly flattened; outer row of oral papillae oriented upwards and perpendicular to oral plate edge, while oral papillae on the ventral surface of the oral plate usually oriented upwards and backwards to the disc edge (Figure 2E). After removing mouth papillae, a small diamond shaped oral shield can be seen; adoral shields small and triangular, not touching. Arms often rising up to the dorsal side, sometimes covering the sack-like disc in some of the preserved individuals. Proximally, dorsal arm plates about 2.5 times wider than long, shortly separated from one another (Figure 2F); distally almost as wide as long, more rounded and widely separated. Lateral arm plates meeting dorsally and ventrally except in the first two segments where they are separated dorsally by the dorsal arm plates (Figure 2F, G). Arm spines present on lateral arm plates, which completely cover the dorsal surface of the arms except for the two first segments; from the third segment, the gap of spines where the lateral arm plates meet is very narrow and similar to gaps between any two contiguous spines; 11 arm spines proximally, dorsalmost first to sixth smaller than the rest and gradually increasing in size but seventh almost twice as long as sixth, eighth the longest, 8–11 the largest ones and a little flattened; distal arm segments usually with 9–10 arm spines, gradually decreasing the difference between the length of inner and outer ones, all arm spines very similar in length from the 15–16 segment onwards (Figure 2H). First to third ventral arm plates almost contiguous, third to fourth clearly separated and becoming gradually more separated distally, 10th and 11th already separated by around half of the ventral arm plate length; ventral arm plates with very concave sides, distally axe shaped, distal width slightly larger than length of the plate. Third tentacle pores with two large flattened spatulate tentacle scales laterally projecting over the tentacle pore from each side of the ventral arm plate, a fifth scale is on the apex of the ventral plate, projecting to the mouth opening; fourth tentacle pores with three large flattened spatulate tentacle scales projecting laterally over the tentacle pores from each side of the ventral arm plate (Figure 2I); fifth tentacle pores with two tentacle scales projecting laterally over the tentacle pores from each side of the ventral arm plate and two smaller spatulate tentacle scales, distally oriented, on the lateral arm plate; sixth to 13th–14th tentacle pores with one tentacle scale projecting laterally over the tentacle pores from each side of the ventral arm plate and two tentacle scales on the lateral arm plate (Figure 2J); beyond, ventral arm plates do not bear any tentacle scale while two tentacle scales are present on the lateral arm plate, one of which is lost closer to the arm tips where only one leaf-like tentacle scale on the lateral arm plate is left (Figure 2F). Colour of individuals alive generally white but lateral arm plates and upper part of the disc orange, particularly the inter-radial areas; colour of specimens preserved in ethanol is white-yellowish (Figure 2A, B).

Remarks

The examined specimens from the Mallorca Channel seamounts are similar to the description of the *O. grandis* holotype from

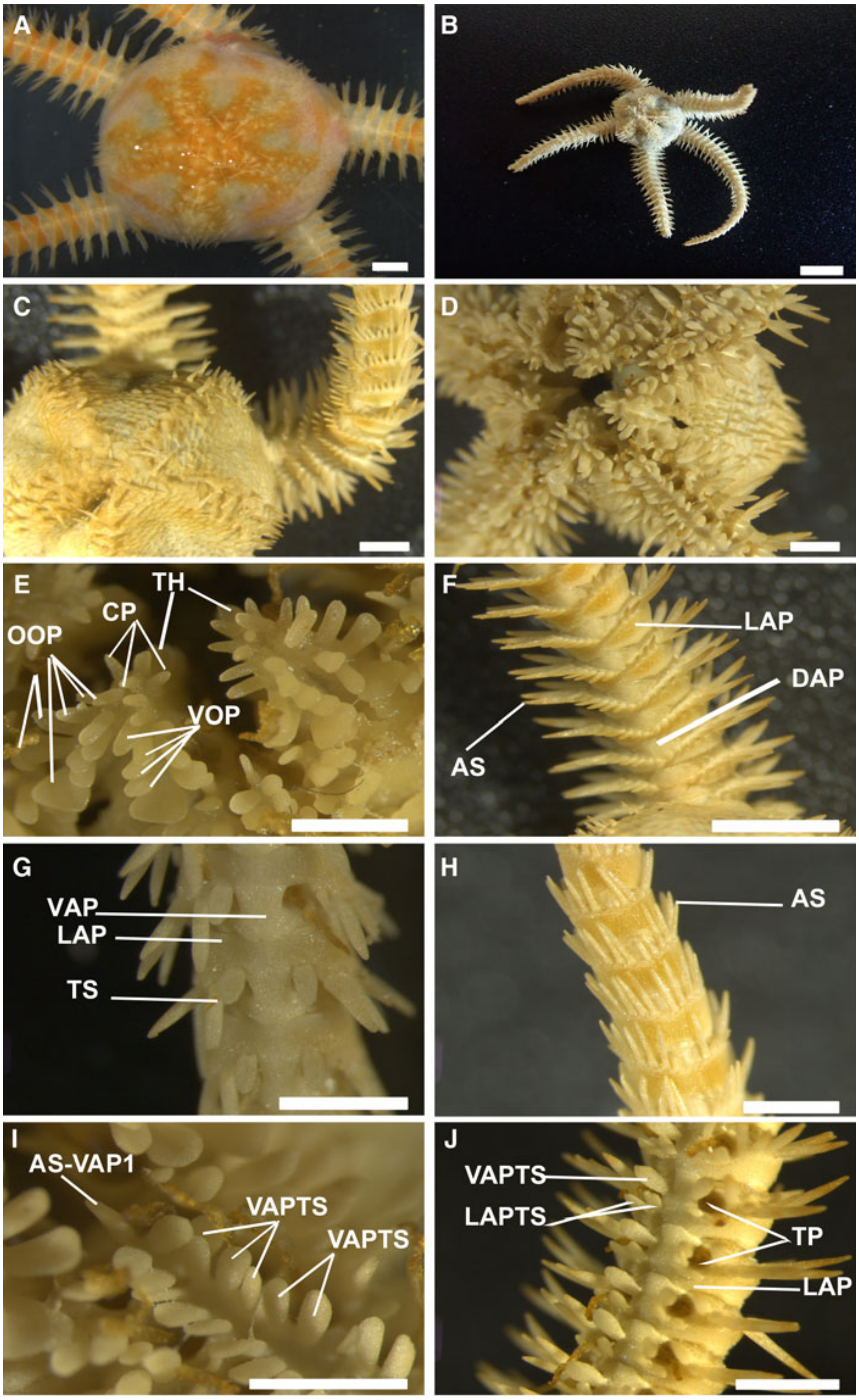

Fig. 2. Photographs of the *Ophiomyces grandis* specimen CFM7020 collected from the Mallorca Channel seamounts showing: the individual alive (A), the individual preserved in ethanol (B), dorsal view of the disc (C), ventral view of the disc (D), jaws (E), dorsal view of the proximal portion of the arm (F), ventral view of the distal portion of the arm (G), dorsal view of the distal portion of the arm (H), ventral view of the proximal portion of the arm (I) and the ventral view of the middle portion of the arm (J). Scale bar represents 1 mm in all photographs except in 2B (30 mm). AS, arm spines; CP, crest papillae; DAP, dorsal arm plates; LAP, lateral arm plate; LAPTS, lateral arm plate tentacle scale; OOP, outer row of oral papillae; TH, teeth; TP, tentacle pore; TS, tentacle scale; VAP, ventral arm plate; AS-VAP1, apical scale on first ventral arm plate; VOP, row of oral papillae on the ventral surface of the oral plate; VAPTS, ventral arm plate tentacle scale.

Tristan da Cunha by Lyman (1879), and its redescription by Paterson (1985). It differs from Paterson's (1985) redescription in the proportions of the ventral arm plates, longer than wide in that case and slightly wider than long in our samples. However, Lyman (1879) for the holotype described it as 'about as broad as long' on the fourth ventral arm plate. Our specimens differ also from Paterson (1985) in the number of arm spines, nine in the holotype, whereas we counted 11 proximally and 9–10 distally. Lyman (1879) reported 11 arm spines. Our specimens match well with the description of the Tasmanian record by O'Hara (1990) except for the number of papillae on the ventral side row of the oral plates, two in O'Hara's description *vs* four in our specimens.

Genetics

A fragment of 530 base pairs (bp) of the COI mitochondrial gene was sequenced for the specimen studied. The nucleotide frequencies were T = 29.06, C = 27.17, A = 24.72 and G = 19.06. High values of genetic distance as well as a high number of base pairs differences were detected between *O. grandis* and *O. delata* specimens (*p*-distance = 17.3/17.7% and bp differences = 92/94). Larger values were detected when comparing *O. grandis* and *O. spathifer* (*p*-distance = 18.8% and bp differences = 100). The neighbour-joining tree showed two clades, with an 80% bootstrap support, within the *Ophiomyces* genus, corresponding to *O. delata* and *O. grandis* (Figure 3). These results supported the morphological identification of *O. grandis*.

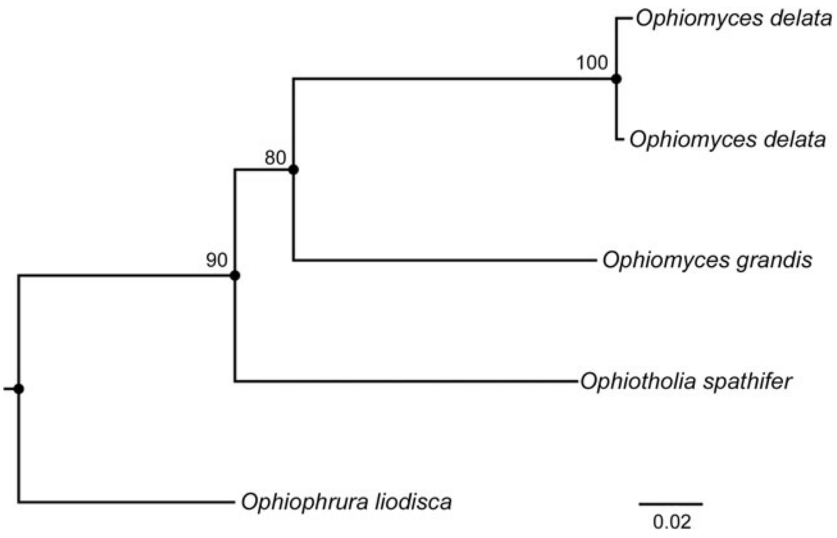

Fig. 3. Neighbour-joining tree based on the cytochrome c oxidase subunit I (COI) fragment including the *Ophiomyces grandis* specimen CFM7020 collected from the Mallorca Channel seamounts. Bootstrap values are shown as percentages and are indicated near the nodes.

Ecology

The species was collected on the three seamounts sampled. The bathymetric distribution of its occurrences ranged from 124 to 312 m. None of the five beam trawl samples collected deeper (i.e. from 446 to 759 m depth) caught any specimen of *O. grandis*. When present, the abundance of *O. grandis* ranged between 3 and 66 specimens per 500 m^{-2}. The highest abundances, 23, 53 and 66 specimens per 500 m^{-2}, were recorded at 275, 242 and 312 m depth at Ses Olives, Ausias March and Emile Baudot seamounts, respectively.

The associated fauna accompanying *O. grandis* varied depending on the depth and seamount. In Ses Olives seamount, the most abundant accompanying species in the sampling stations at 275 and 290 m depth were Porifera (*Thenea muricata* (Bowerbank, 1858) and some species pending to determine) and the decapod crustaceans *Ebalia nux* A. Milne-Edwards, 1883, and *Plesionika edwardsii* (Brandt, 1851), the echinoderms *Dictenophiura carnea* (Lütken, 1858), *Ophiocten abyssicolum* (Forbes, 1843) and *Pseudostichopus occultatus* von Marenzeller, 1893, and the Polychaeta *Hyalinoecia tubicola* (O.F. Müller, 1776).

On the Ausias March seamount, 67 individuals of *O. grandis* were collected at 242 m depth, representing more than half of the total number of individuals of all species collected in that sample. The most abundant accompanying benthic species at this station were the decapod crustaceans *Munida speciosa* von Martens, 1878 and *Plesionika antigai* Zariquiey Álvarez, 1955, and the Polychaeta *H. tubicola*. In contrast, on this seamount, but at 124 m depth, where only four specimens of *O. grandis* were collected, they were mainly accompanied by the decapod crustaceans *Aegaeon lacazei* (Gourret, 1887), *Ergasticus clouei* A. Milne-Edwards, 1882 and *Pagurus prideaux* Leach, 1815, the Polychaeta *Spirobranchus triqueter* (Linnaeus, 1758) and the echinoderm *O. abyssicolum*.

On the Emile Baudot seamount, the most abundant accompanying species of *O. grandis* in the samples collected at 138 and 146 m depth were the decapod crustaceans *Inachus dorsettensis* (Pennant, 1777), *E. clouei* and *E. nux*, the brachiopod *Gryphus vitreus* (Born, 1778), and the echinoderm *Cidaris cidaris* (Linnaeus, 1758). In this seamount, at 312 m depth, the deepest sample station where *O. grandis* was collected, it was the most abundant species with 89 specimens caught (21% of the total number of individuals of all species). At this station *O. grandis* was accompanied mainly by the echinoderms *P. occultatus*, *D. carnea* and *Amphiura filiformis* (O.F. Müller, 1776), and the decapod crustaceans *E. clouei* and *P. antigai*.

Discussion

According to Galil & Zibrowius (1998), the first scientists who studied the biological communities of seamounts in the Mediterranean, these geomorphological structures can be considered as islands, biogeographically separated from the continental coasts by great depths. These authors suggested that seamounts may serve as isolated refuges for relict populations of species that have disappeared from a previously larger distribution range. This may be the case of *Ophiomyces grandis* in the Mediterranean. However, it may also be that seamount communities are more connected through dispersal than previously thought, as suggested by Clark *et al.* (2010) (and references therein), and that *O. grandis* has evolved to live in these geomorphological isolated features. Its reported distribution is almost exclusively from isolated islands or seamounts mainly in the Atlantic Ocean such as Tristan da Cunha, Rockall Trough or the Galicia Bank (Lyman, 1879; Paterson, 1985; Litvinova, 2001; Serrano *et al.*, 2017), although it has also been reported from the Bass Strait in south-east Australia (O'Hara, 1990).

The high abundances of *O. grandis* detected in the present work are evidence that the population is well established in the Mallorca Channel seamounts and that the Mediterranean should be considered within species' distribution range. It also suggests that *O. grandis* is probably present in other Mediterranean seamounts, although it seems to be absent from Mediterranean continental deep-sea areas (Koukouras *et al.*, 2007; Mecho *et al.*, 2014; Öztoprak *et al.*, 2014; Moya-Urbano *et al.*, 2016). The abundance of seamount-like features in the Mediterranean is considered large and was estimated to be composed of about a hundred of these structures that occupy a large area of about 89,000 km^2 (Morato *et al.*, 2013). Despite their importance, Mediterranean seamounts are poorly known, with *in situ* information only available for less than 10% of them and mainly consisting of geological investigations, with almost no biological and ecological studies until the end of the 20th century, except for the Erastothenes seamount in the Levant basin (Galil & Zibrowius, 1998; Danovaro *et al.*, 2010).

This situation has changed during the last decade when research surveys have been conducted in scattered seamounts mainly located in the Alboran Sea, around the Balearic Islands and the Tyrrhenian Sea (Danovaro *et al.*, 2010; Bo *et al.*, 2011; Sabatini *et al.*, 2011; Aguilar *et al.*, 2013; De la Torriente *et al.*, 2018). These surveys mainly used Remote Operated Video (ROV) and grabs in order to sample the benthic and demersal communities, although bottom trawl was also used in the Baronie seamount, in the Tyrrhenian Sea, central Mediterranean (Sabatini *et al*., 2011). These studies have considerably increased the benthic biodiversity knowledge of Mediterranean seamounts, particularly for the Seco de los Olivos, Seco de Palos and Mallorca Channel seamounts in the western Mediterranean and for the Vercelli seamount in the Tyrrhenian Sea (Anon, 2011; Bo *et al*., 2011; Aguilar *et al*., 2013; Maldonado *et al*., 2015). Despite the abundance of *O. grandis* reported in the present work, none of the faunistic lists resulting in the mentioned works reported the presence of *O. grandis*, neither in Ses Olives, Ausias March nor Emile Baudot seamounts, where more than 20 hours of video were recorded (OCEANA, 2011). Perhaps a hiding behaviour could explain the absence of this species from ROV images, or maybe it was one of the Ophiuroidea species reported as no identified ophiuroids (OCEANA, 2011). To our knowledge, beam trawl has only been used once before to sample seamounts in the Mediterranean, in the Erastothenes seamount, where a single one hour beam trawl sample was performed (Galil & Zibrowius, 1998). Only one echinoderm species, *Odontaster mediterraneus* (von Marenzeller, 1893), was reported from that sample. Contrastingly, in our beam trawl samples, the echinoderms were the most abundant (a total of 983 individuals were collected) and diverse (35 species) group of mobile benthic fauna after crustaceans (1878 individuals belonging to 49 species), evidence that the Mallorca Channel seamounts represent a suitable environment for echinoderms, and particularly for *O. grandis*, which was among the dominant species in several of the samples where it appeared. It is also remarkable that the shallowest depth of occurrence of *O. grandis* in the Mallorca Channel seamounts, 124 m, may be the shallowest record ever reported for this species, and also, one of the shallowest records for the *Ophiomyces* genus, except for the exceptionally shallow records of *O. delata* and *O. papillospinus*, from the Andaman Islands and Japan, respectively (Parameswaran *et al*., 2015; Okanishi *et al*., 2016).

The overlapping distribution of *O. grandis* and *O. delata* in the Pacific Ocean and the large coincidences in their morphological traits led Parameswaran *et al*. (2015) to suggest that these two species were synonyms or that they represented a species complex. However, the genetic differences observed in the present study between *O. grandis* and *O. delata* confirm the validity of both species.

The assessment of benthic species richness on seamounts can be strongly influenced by the sampling methodology applied (Pitcher *et al*., 2007), with extractive sampling yielding broader estimation of biodiversity, as well as the possibility of obtaining individuals for genetic analyses (Williams *et al*., 2015). Our results emphasize the importance of beam trawl sampling in seamounts biodiversity research. This is not always possible due to the often rough geomorphology of seamounts. However, when possible, in sedimentary bottoms, this type of sampling can be a useful tool, which in combination with ROV and other image acquisition methodologies can provide a more realistic picture of the biodiversity of seamounts' benthic communities.

Acknowledgements. This research has been developed within the project LIFE IP INTEMARES. We are very grateful to Dr Ben Thuy and two anonymous reviewers whose comments and suggestions have greatly contributed to improve this manuscript. We also thank all participants in the survey INTEMARES MALLORCA0818 and the Captain and crew of the RV 'Ángeles Alvariño'.

Financial support. This study was funded by the project LIFE IP INTEMARES. Sergio Ramírez-Amaro is supported by a postdoctoral contract co-funded by the Regional Government of the Balearic Islands and the European Social Fund.

References

Acosta J, Ancochea E, Canals M, Huertas MJ and Uchupi E (2004) Early Pleistocene volcanism in the Emile Baudot Seamount, Balearic Promontory (western Mediterranean Sea). *Marine Geology* **207**, 247–257.

Aguilar R, Pastor X, Garcia S, Marin P and Ubero J (2013) Importance of seamounts-like features for Mediterranean marine habitats and threatened species. *Rapport Comité Internationale Mer Méditerranée* **40**, 716.

Anon (2011) *Campañas oceanográficas Proyecto LIFE+ INDEMARES. Seco de los Olivos. OCEANA*, 76 pp.

Baric S and Sturmbauer C (1999) Ecological parallelism and cryptic species in the genus *Ophiothrix* derived from mitochondrial DNA sequences. *Molecular Phylogenetics and Evolution* **11**, 157–162.

Bo M, Bertolino M, Borghini M, Castellano M, Covazzi Harriague A, Gioia Di Camillo C, Gasparini GP, Misic C, Povero P, Pusceddu A, Schroeder K and Bavestrello G (2011) Characteristics of the mesophotic megabenthic assemblages of the Vercelli Seamount (North Tyrrhenian Sea). *PLoS ONE* **6**(2), e16357.

Boissin E, Feral JP and Chenuil A (2008) Defining reproductively isolated units in a cryptic and syntopic species complex using mitochondrial and nuclear markers: the brooding brittle star, *Amphipholis squamata* (Ophiuroidea). *Molecular Ecology* **17**, 1732–1744.

Clark MR, Rowden AA, Schlacher T, Williams A, Consalvey M, Stocks KI, Rogers AD, O'Hara TD, White M, Shank TM and Hall-Spencer JM (2010) The ecology of seamounts: structure, function, and human impacts. *Annual Reviews in Marine Science* **2**, 253–278.

Danovaro R, Company JB, Corinaldesi C, D'Onghia G, Galil B, Gambi C, Gooday AJ, Lampadariou N, Marco Luna G, Morigi C, Olu K, Polymenakou P, Ramirez-Llodra E, Sabbatini A, Sardà F, Sibuet M and Tselepides A (2010) Deep-sea biodiversity in the Mediterranean Sea: the known, the unknown, and the unknowable. *PLoS ONE* **5**(8), e11832.

De la Torriente A, Serrano A, Fernández-Salas LM, García M and Aguilar R (2018) Identifying epibenthic habitats on the Seco de los Olivos Seamount: species assemblages and environmental characteristics. *Deep-Sea Research Part I* **135**, 9–22.

Galil B and Zibrowius H (1998) First benthos samples from Erastothenes Seamount, eastern Mediterranean. *Senckenbergiana Maritima* **28**(4/6), 111–121.

Hall TA (1999) Bioedit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT. *Nucleic Acid Symposium Series* **41**, 95–98.

Hebert PDN, Cywinska A, Ball SL and de Waard JR (2003) Biological identifications through DNA barcodes. *Proceedings of the Royal Society B: Biological Sciences* **270**, 313–322.

Hendler G (2018) Armed to the teeth: a new paradigm for the buccal skeleton of brittle stars (Echinodermata: Ophiuroidea). *Contributions in Science* **526**, 189–311.

Kimura M (1980) A simple method of estimating evolutionary rate of base substitutions through comparative studies of nucleotide sequences. *Journal of Molecular Evolution* **16**, 111–120.

Koukouras A, Sinis AI, Bobori D, Kazantzidis S and Kitsos M-S (2007) The echinoderm (Deuterostomia) fauna of the Aegean Sea, and comparison with those of the neighbouring seas. *Journal of Biological Research* **7**, 67–92.

Litvinova NM (2001) The ophiuroid genus *Ophiomyces* (Echinodermata, Ophiuroidea). In Kuznetsov AP and Zezina ON (eds), *Composition and Structure of the Marine Bottom Biota: Collected Proceedings*. Moscow: VNIRO Publishing House, pp. 145–158.

Lyman T (1869) Preliminary report on the Ophiuridae and Astrophytidae dredged in deep water between Cuba and Florida reef. *Bulletin of the Museum of Comparative Zoology at Harvard College* **1**, 309–354.

Lyman T (1879) Ophiuridae and Astrophytidae of the exploring voyage of H.M.S. "Challenger", under Prof. Sir Wyville Thomson, F.R.S. Part II. *Bulletin of the Museum of Comparative Zoology at Harvard College* **6**(2), 17–83.

Maldonado M, Aguilar R, Blanco J, García S, Serrano A and Punzón A (2015) Aggregated clumps of lithistid sponges: a singular, reef-like bathyal habitat with relevant paleontological connections. *PLoS ONE* **10**(5), e0125378.

Mecho A, Billett DSM, Ramírez-Llodra E, Aguzzi J, Tyler PA and Company JB (2014) First records, rediscovery and compilation of deep-sea echinoderms in the middle and lower continental slope of the Mediterranean Sea. *Scientia Marina* **78**(2), 281–302.

Morato T, Kvile KØ, Taranto GH, Tempera F, Narayanaswamy BE, Hebbeln D, Menezes GM, Wienberg C, Santos RS and Pitcher TJ (2013) Seamount physiography and biology in the north-east Atlantic and Mediterranean Sea. *Biogeosciences (Online)* **10**, 3039–3054.

Mortensen T (1927) *Handbook of the Echinoderms of the British Isles*. Oxford: Oxford University Press.

Moya-Urbano E, Moya F, Mateo-Ramírez Á, Gallardo-Núñez M, Ordines F, Farias C, Manjón-Cabeza M, Urra J, Rueda J and Ruiz CG (2016). Preliminary characterization of echinoderm assemblages in circalittoral and bathyal soft bottoms of the northern Alboran Sea. *Frontiers in Marine Science. Conference Abstract: XIX Iberian Symposium on Marine Biology Studies*. doi: 10.3389/conf.FMARS.2016.05.00102.

OCEANA (2011) Montañas submarinas de las Islas Baleares: Canal de Mallorca. Propuesta de protección para Ausias March, Emile Baudot y Ses Olives. *OCEANA*, 64 pp.

O'Hara TD (1990) New records of Ophiuridae, Ophiacanthidae and Ophiocomidae (Echinodermata: Ophiuroidea) from south-eastern Australia. *Memoirs of Museum Victoria* **50**, 287–305.

O'Hara TD, Hugall AF, Thuy B, Stöhr S and Martynov AV (2017) Restructuring higher taxonomy using broad-scale phylogenomics: the living Ophiuroidea. *Molecular Phylogenetics and Evolution* **107**, 415–430.

Okanishi M, Sentoku A, Fujimoto S, Jimi N, Nakayama R, Yamana Y, Yamauchi H, Tanaka H, Kato T, Kashio S, Uyeno D, Yamamoto K, Miyaza K and Asakura A (2016) Marine benthic community in Shirahama, southwestern Kii Peninsula, central Japan. *Publications of the Seto Marine Biological Laboratory* **44**, 7–52.

Öztoprak B, Doğan A and Daği̇ E (2014) Checklist of Echinodermata from the coasts of Turkey. *Turkish Journal of Zoology* **38**, 892–900.

Parameswaran UV, Jaleel KUA, Gopal A, Sanjeevan VN and Vijayan AK (2015) On an unusual shallow occurrence of the deep-sea brittle star *Ophiomyces delata* in the Duncan Passage, Andaman Islands (Northern Indian Ocean). *Marine Biodiversity* **46**(1), 151–156.

Paterson GLJ (1985) The deep-sea Ophiuroidea of the North Atlantic Ocean. *Bulletin of the British Museum (Natural History)* **49**, 1–162, figs 1–59.

Pitcher TJ, Morato T, Hart PJ, Clark MR, Haggan N and Santos RS (eds) (2007) *Seamounts: Ecology, Fisheries and Conservation*. Oxford: Blackwell Publishing.

Ratnasingham S and Hebert PDN (2007) BOLD: the barcode of life data system (www.barcodinglife.org). *Molecular Ecology Notes* **7**, 355–364.

Reiss H, Kröncke I and Ehrich S (2006) Estimating the catching efficiency of a 2-m beam trawl for sampling epifauna by removal experiments. *ICES Journal of Marine Science* **63**, 1453–1464.

Reys JP (1961) Deux ophiures nouvelles: *Amphipholis tissieri* et *Ophiomyces peresi*. *Recueil des Travaux de la Station Marine d'Endoume* **22**, 153–157.

Sabatini A, Follesa MC, Locci I, Matta G, Palmas F, Pendugiu AA, Pesci P and Cau A (2011) Demersal assemblages in two trawl fishing lanes located on the Baronie Seamount (Central Western Mediterranean). *Journal of the Marine Biological Association of the United Kingdom* **91**, 65–75.

Serrano A, Cartes JE, Papiol V, Punzón A, García-Alegre A, Arronte JC, Ríos P, Lourido A, Frutos I and Blanco M (2017) Epibenthic communities of sedimentary habitats in a NE Atlantic deep seamount (Galicia Bank). *Journal of Sea Research* **130**, 154–165.

Stöhr S, O'Hara TD and Thuy B (2012) Global diversity of brittle stars (Echinodermata: Ophiuroidea). *PLoS ONE* **7**, e31940.

Tamura K, Stecher G, Peterson D, Filipski A and Kumar S (2013) MEGA6: Molecular Evolutionary Genetics Analysis Version 6.0. *Molecular Biology and Evolution* **30**, 2725–2729.

Thompson JD, Higgins DG and Gibson TJ (1994) CLUSTAL w: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position specific gap penalties and weight matrix choice. *Nucleic Acid Research* **22**, 4673–4680.

Thuy B and Meyer CA (2013) The pitfalls of extrapolating modern depth ranges to fossil assemblages: new insights from Middle Jurassic brittle stars (Echinodermata: Ophiuroidea) from Switzerland. *Swiss Journal of Palaeontology* **132**(1), 5–21.

Ward RD, Holmes BH and O'Hara TD (2008) DNA barcoding discriminates echinoderm species. *Molecular Ecology Resources* **8**, 1202–1211.

Williams A, Althaus F and Schlacher TA (2015) Towed camera imagery and benthic sled catches provide different views of seamount benthic diversity. *Limnology and Oceanography: Methods* **13**(2), 62–73.

Würtz M, Rovere M and Bo M (2015) Introducing the Mediterranean seamount atlas: general aspects. In Würtz M and Rovere M (eds), *Atlas of the Mediterranean Seamounts and Seamount-Like Structures*. Málaga: IUCN, pp. 11–19.

Ziesenhenne FC (1940) New Ophiurans of the Allan Hancock Pacific expeditions. *Allan Hancock Pacific Expeditions* **8**(2), 5–58.