



# *Lipkea ruspoliana* Vogt, 1886 (Cnidaria: Staurozoa) in Portugal: the contribution of citizen science to range extension and taxonomic discussion of rare species

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**Abstract** Citizen science online platforms are increasingly making important sources of biological information available at the click of a button, allowing the interaction of volunteers and scientists to report and identify the world's diversity. In this work, we combined the data available in citizen science platforms (iNaturalist and GelAvista) and the effort and collaboration of a diverse team of scuba-divers,

scientists, and underwater photographers to report for the first time the presence of the stauromedusa *Lipkea* (Cnidaria: Staurozoa) in Portugal. Based on DNA (COI and 16S), we identified the species as *Lipkea ruspoliana*. However, the marginal lappets of some Portuguese specimens are similar to those of *Lipkea sturdzii*, a species described based on a single specimen in 1893 and never reported again. These results suggest that *L. sturdzii* is a synonym of *L. ruspoliana*, although further taxon and habitat sampling for genetic studies and investigations of intraspecific morphological variation are necessary. In addition, we provided data on species ecology, reviewed the records of Staurozoa from Portugal, and mapped the world geographic distribution of *Lipkea*. Staurozoa is a cryptic group of cnidarians, and *Lipkea* seems to be a particularly rare genus. Citizen science has proved to be a valuable contribution to studies on the group, enabling biological discussions that otherwise would be more challenging.

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## Introduction

The engagement of citizens in documenting world diversity has allowed successful advances in scientific achievements (Bonney et al. 2009; Silvertown 2009). Moreover, the current needs to meet international

targets toward biodiversity conservation benefits from the integration of multiple sources of data. Citizen science and community-based monitoring programs are emerging as relevant sources of data (namely large-scale data on species distribution or population abundance), which can complement scientific research (Chandler et al. 2017; Magalhães et al. 2020). Citizen science initiatives in the marine field have gradually increased in recent years, contributing to expand research possibilities, both spatially and temporarily (Thiel et al. 2014; Giovos et al. 2019; Earp & Liconti 2020; Kelly et al. 2020; Garcia-Soto et al. 2021). Examples are the role of citizens in detecting marine invasions (Scyphers et al. 2015), species management and conservation (Cranswick et al. 2022), identifying shifts in species distribution (Tanaka et al. 2021), and even reporting marine litter (Kawabe et al. 2022). Citizen science data combined with plankton and oceanographic surveys (Pires et al. 2018) and with laboratory studies (Gueroun et al. 2021) are also increasing the knowledge on species ecology and biology. The contribution of citizens to scientific projects can occur in a study delineated with a specific goal (e.g., Kaminas et al. 2022) or more broadly, in online social media platforms designed for sharing nature observations. A leading platform contributing to biodiversity knowledge and conservation is iNaturalist (e.g., Roberts et al. 2022), making species records available to researchers, including information on rare and threatened species (e.g., Maňko et al. 2017; Wilson et al. 2020; Cranswick et al. 2022). The citizen science project GelAvista (<https://gelavista.ipma.pt/>) is monitoring the occurrence of jellyfish species in Portugal since 2016 and promoting scientific data on the life cycle and ecology of these animals (Pires et al. 2018; Gueroun et al. 2021).

A group lacking information about their diversity and distribution along most coastlines are the Staurozoa, also known as stalked jellyfishes, rare marine benthic cnidarians generally reported in temperate and polar waters (Mills and Hirano 2007; Miranda and Marques 2016; Miranda et al. 2018). Their current diversity encompasses about 50 species in 11 genera (Miranda et al. 2018). Staurozoan species have a distinctive life cycle in which a sexual stage with polypoid and medusoid traits lives attached to the substrate by a peduncle (Wietrzykowski 1912; Hirano 1986; Kikinger and Salvini-Plawen 1995; Miranda et al. 2010, 2018). The species are often camouflaged

against the background of algae (Larson 1980; Daly et al. 2007; Miranda et al. 2018) and some of them are seasonal (Corbin 1979; Miranda et al. 2012), which further hampers their observation and collection by researchers. Because of their biology and historical research bias, Staurozoa is one of the least studied classes among Cnidaria (Miranda et al. 2018), and the scarcity of information about species richness and geographic distribution challenges the discussions about their conservation (Miranda and Marques 2016). In this context, the engagement of volunteers in the work of researchers has already proved to be a valuable approach to identify new species and records (Miranda et al. 2017). Citizen science platforms thus represent a powerful tool to provide new insights about the diversity and ecology of cnidarians (Anthony et al. 2023; Rojas-Cruz et al. 2023) and is especially promising for staurozoans.

On June 24, 2020, the author L.S.M. contacted the co-author S.T. regarding a picture posted on the iNaturalist platform (<https://www.inaturalist.org/observations/48823432>; Table 1); a sighting that was also sent to the GelAvista project. The species in the image could be easily identified as belonging to the genus *Lipkea* because of the presence of marginal lappets (lobes) (Vogt 1886; Zagal et al. 2011; Miranda et al. 2016a, b). However, currently there are only two species of Staurozoa formally reported from Portugal: (1) *Haliclystus auricula* James-Clark, 1863, recorded in Moledo do Minho, Foz do Douro, and Granja; and (2) *Calvadosia campanulata* Lamouroux, 1815, recorded in Sines (Nobre 1931; Rodrigues et al. 2020) (Table 2). Therefore, the aim of this study is to report the first occurrence of *Lipkea* in Portugal and to identify the species based on the material collected, discussing the systematics of the genus.

## Material and methods

### Sampling

After the contact of L.S.M. and knowing that the record had a scientific value, S.T. organized a field trip to collect samples for molecular diagnosis, under the Biomares program aimed at the conservation and management of the Professor Luiz Saldanha Marine Park (Arrábida Natural Park). A

**Table 1** New records of *Lipkea ruspoliana* from Portugal

Date	Sites	Observers	Latitude, longitude	Depth (m)	Number of individuals observed in the field	Link iNaturalist
July 11, 2012	Ponta da Passagem Cabo Espichel, Arrábida Natural Park, Portugal	João Pedro Silva	38.409591, –9.203538	~15	1	<a href="https://www.inaturalist.org/observations/3072947">https://www.inaturalist.org/observations/3072947</a>
June 7, 2020	Ponta da Passagem Cabo Espichel, Arrábida Natural Park, Portugal	Sílvia Tavares, Mário Rolim	38.409759, –9.202670	10	1	<a href="https://www.inaturalist.org/observations/48823432">https://www.inaturalist.org/observations/48823432</a>
August 14, 2020*	Ponta da Passagem Cabo Espichel, Arrábida Natural Park, Portugal	Sílvia Tavares, Mário Rolim, Emanuel Gonçalves, Sylvie Dias, Ester Serrão	38.409759, –9.202670	10	5	–
December 17, 2020	Baía da Armação, Arrábida Natural Park, Portugal	Sílvia Tavares, Mário Rolim	38.439372, –9.061268	10	1	<a href="https://www.inaturalist.org/observations/66781645">https://www.inaturalist.org/observations/66781645</a>
June 13, 2021	Catrapona, Arrábida Natural Park, Portugal	Sílvia Tavares, Mário Rolim	38.413309, –9.241278	35	1	<a href="https://www.inaturalist.org/observations/91516927">https://www.inaturalist.org/observations/91516927</a>
August 15, 2021	Ponta da Passagem Cabo Espichel, Arrábida Natural Park, Portugal	Sílvia Tavares, Mário Rolim	38.409759, –9.202670	10	4–5	<a href="https://www.inaturalist.org/observations/91516372">https://www.inaturalist.org/observations/91516372</a>
September 26, 2021	Ponta da Passagem Cabo Espichel, Arrábida Natural Park, Portugal	Sílvia Tavares, Mário Rolim, Emanuel Gonçalves, Sylvie Dias	38.409759, –9.202670	–	–	–
October 24, 2021	Ponta da Passagem Cabo Espichel, Arrábida Natural Park, Portugal	Sílvia Tavares, Mário Rolim, Emanuel Gonçalves, Sylvie Dias	38.409759, –9.202670	10	7	–
July 24, 2022	Ponta da Passagem Cabo Espichel, Arrábida Natural Park, Portugal	Sílvia Tavares, Mário Rolim, Emanuel Gonçalves, Sylvie Dias	38.409759, –9.202670	10	18	<a href="https://www.inaturalist.org/observations/127814975">https://www.inaturalist.org/observations/127814975</a>

**Table 1** (continued)

Date	Sites	Observers	Latitude, longitude	Depth (m)	Number of individuals observed in the field	Link iNaturalist
September 11, 2022	Baía da Armação, Arrábida Natural Park, Portugal	Sílvia Tavares, Mário Rolim, Emanuel Gonçalves, Sylvie Dias	38.439372, -9.061268	8	1	<a href="https://www.inaturalist.org/observations/134754996">https://www.inaturalist.org/observations/134754996</a>
November 12, 2022	Jardim das Gorgônias, Arrábida Natural Park, Portugal	Sílvia Tavares	38.434615, -9.062685	10	1	–

\*One specimen collected for DNA analyses

**Table 2** Staurozoans from Portugal based on the literature (excluding the records from this study, Table 1)

Species	Localities	Latitude, longitude*	Reference
<i>Haliclystus auricula</i>	Moledo do Minho, Portugal	41.849817, -8.868967	Nobre 1931
	Foz do Douro, Portugal	41.150760, -8.678641	Nobre 1931
	Granja, Portugal	41.040320, -8.651305	Nobre 1931
<i>Calvadosia campanulata</i>	Sines, Portugal	37.957860, -8.894234	Nobre 1931

\*Estimated, based on the localities from the literature

diverse team of scuba-divers, scientists, and underwater photographers were involved. Unfortunately, it was not possible to collect individuals for morphological analysis since the specimens of *Lipkea* were strongly attached to the rocky substrate, and the manipulation of the specimens caused an intense contraction. However, different individuals were photographed in high resolution using super-macro photography before collection (August 14, 2020: 5 individuals observed and 40 images analyzed; October 24, 2021: 7 individuals observed and 9 images analyzed; July 24, 2022: 18 individuals observed and 7 images analyzed; see also additional images available in iNaturalist, Table 1), to register morphological characteristics.

One specimen (Table 1) was collected, preserved in 90% ethanol and transported at low temperature to the Portuguese Institute for Sea and Atmosphere (IPMA) national laboratory (Lisbon), where it was immediately preserved in 96% ethanol by A.d.S. and stored at -20 °C until it was transferred to the Centre of Marine Sciences (CCMAR, Faro, Portugal), for molecular analysis. The sample has been deposited at CCMAR (voucher ID: 20-0194).

### Geographical distribution

We reviewed the current known geographic distribution of Staurozoa in Portugal (Table 1) and the world distribution of *Lipkea*, including the new records from this study (Tables 1, 3). We mapped the records using Google Earth Pro (version 7.3) to obtain the coordinates from site names described in the literature (Tables 2, 3). The maps were constructed using SimpleMappr (Shorthouse 2010).

### DNA extraction, amplification and sequencing

Genomic DNA was extracted with the DNeasy Blood and Tissue kit (Qiagen) following the manufacturer's instructions with minor changes. Specifically, the purified DNA was eluted from the spin column in four successive steps of 50 µl each using buffer AE warmed to 65 °C. The quality, purity, and yield of the DNA extract were assessed using gel electrophoresis and nanodrop spectrophotometer. We amplified fragments of two mitochondrial genes, the cytochrome *c* oxidase subunit I (COI) and the large subunit rRNA (16S), using

**Table 3** World distribution of *Lipkea* (excluding the records from this study, Table 1)

Species	Localities	Latitude, longitude*	References
<i>L. ruspoliana</i>	Alghero, northwestern Sardinia, Mediterranean Sea	40.555171, 8.314531	Vogt 1886, 1887
	Aquarium of the Oceanographic Museum of Monaco	43.730312, 7.425644	Pisani et al. 2007
<i>L. sturdzii</i>	Near the Blue Grotto, Island of Capri, Bay of Naples, Italy	40.546158, 14.242154	Antipa 1893
<i>L. stephensoni</i>	Near Cape Town, Stil Bay, South Africa	−34.374674, 21.429941	Carlgren 1933
	Onrust River, Overberg, Western Cape, South Africa	−34.4196, 19.1801	Miranda et al. 2017
	Smitswinkel Bay, False Bay, Cape Town, Western Cape, South Africa	−34.2743, 18.4728	Miranda et al. 2017
	Between Partridge Point and Castle Rocks, False Bay, Cape Town, Western Cape, South Africa	−34.2457, 18.4795	Miranda et al. 2017
<i>Lipkea</i> sp.	In aquarium, Chiba, Japan	35.596151, 140.076973	Miranda et al. 2016a
	Poor Knights Islands, New Zealand	−35.497141, 174.74951	Cairns et al. 2009
	Fiordland, New Zealand	−45.603043, 166.60995	Cairns et al. 2009
	Jervis Bay, New South Wales, Australia	−35.06712, 150.681813	Zagal et al. 2011

\*Estimated, based on the localities from the literature

the primers jgHCO2198 and jgLCO1490 (Geller et al. 2013), and FiMod and R2 (Cartwright et al. 2008), respectively. Two PCRs were run for each molecular marker in 25 µl final volume reactions containing 0.4 mM dNTPs, 2 mM MgCl<sub>2</sub>, 0.5 µM of each primer, 1X GoTaq Flexi buffer (Promega), 5 U GoTaq® Flexi DNA Polymerase (Promega) and 5 µl of genomic DNA (1 ng or 1:5 dilution of stock DNA). Amplification conditions followed those used in Miranda et al. (2016a). The PCR products of each barcode were then pooled, purified with Agencourt AMPure XP magnetic beads (Beckman Coulter) and sequenced at CCMAR's Sequencing Platform with an Applied Biosystems 3130xl Genetic Analyzer using BigDye®Terminatorv3.1 chemistry and POP7 polymer (forward and reverse directions). The forward and reverse sequences from each molecular marker were confirmed using the BLAST software (Basic Local Alignment Search;

available at <https://blast.ncbi.nlm.nih.gov/Blast.cgi>) and deposited in GenBank (Table 4).

#### Molecular analyses

Sequences were assembled and edited using Geneious Prime (Biomatters, Auckland, New Zealand) and then aligned with other staurozoan sequences from GenBank (Table 4) using MAFFT (Katoh et al. 2002). Missing ends of the alignments were removed based on the shortest sequence length, totaling 506 bp for COI and 523 bp for 16S (1029 bp in the combined alignment).

Phylogenetic analyses were performed based on maximum likelihood (ML) criteria using PhyML 3.0 online execution tool (Guindon et al. 2010), with default settings for tree searching. The model of sequence evolution GTR+I+G was selected for the COI alignment, the model GTR+G for 16S, and the

**Table 4** Species used in the phylogenetic analysis and respective GenBank accession numbers of mitochondrial molecular markers (COI and 16S)

Suborder	Family	Species	GenBank accession numbers	
			COI	16S
Amyostaurida	Kishinouyeidae	<i>Calvadosia campanulata</i>	KU257483	KU257508
Myostaurida	Halicystidae	<i>Halicystus antarcticus</i>	KU257475	EU294003
		<i>Halicystus auricula</i>	MF346311	MF322735
	Lucernariidae	<i>Lucernaria sainthilareii</i>	KU257482	KU257507
	Lipkeidae	<i>Lipkea ruspoliana</i>	KU257480	KU257503
		<i>Lipkea</i> sp. Japan	KU257493	KU257520
		<i>Lipkea</i> sp. Portugal	OQ108502*	OQ117049*

\*Sequences obtained in this study

model GTR+G for the combined COI and 16S alignment, using the Smart Model Selection Tool (SMS, Lefort et al. 2017) available in PhyML, based on the Akaike information criterion (AIC). Branch support was estimated with bootstrap using 1,000 replicates. FigTree (<http://tree.bio.ed.ac.uk/software/figtree/>) was used to visualize and edit the resulting trees.

## Results

### Notes on morphology (Figs. 1, 2)

Calyx broad and funnel-shaped (Figs. 1a, 2a–e). Peduncle of difficult visualization, strongly attached to rocks (Fig. 2d). Four interradial septa generally visible in calyx (Fig. 1a). One specimen exhibited an unusual symmetry with five interradial septa (Fig. 2b). Six to 10 marginal lappets (Figs. 1, 2). Marginal lappets more or less smooth, some specimens (at least two) presenting a clear row of ~15–25 short filiform projections, which are fused to one another by a web, flap-like structure (Fig. 1b–d). Manubrium with four perradial lips (Figs. 1a, 2a, b, f). One specimen with five lips in manubrium (unusual symmetry) (Fig. 2b). Gonads embedded in gastrovascular cavity of subumbrella, vesicles pink or white in color, occupying the region close to manubrium (confined to basal part of calyx), around the interradial infundibula (Figs. 1a, 2a, b, f). Perradial and interradial anchors absent. Pad-like adhesive structures absent. Secondary tentacles also absent. Coronal muscle entire (Fig. 1a). White spots of nematocysts on subumbrellar surface, margin of calyx, marginal lappets, above the gonads, and deep in infundibula (Figs. 1a, 2b). Body translucent / pinkish or whitish (Figs. 1, 2). Total calyx diameter about 10–20 mm.

### Records

*Lipkea* sp. has been observed at different localities within the Professor Luiz Saldanha Marine Park (Arrábida Natural Park) (Table 1; Fig. 3), a marine protected area established in 1998. In addition to our observations and based on entries at the iNaturalist platform, *Lipkea* has already been observed in Portugal since at least July 11, 2012 (<https://www.inaturalist.org/observations/3072947>) (Table 1), although initially misidentified as a sea anemone. The species

has been observed from June to December (2014, 2020–2022) (Table 1).

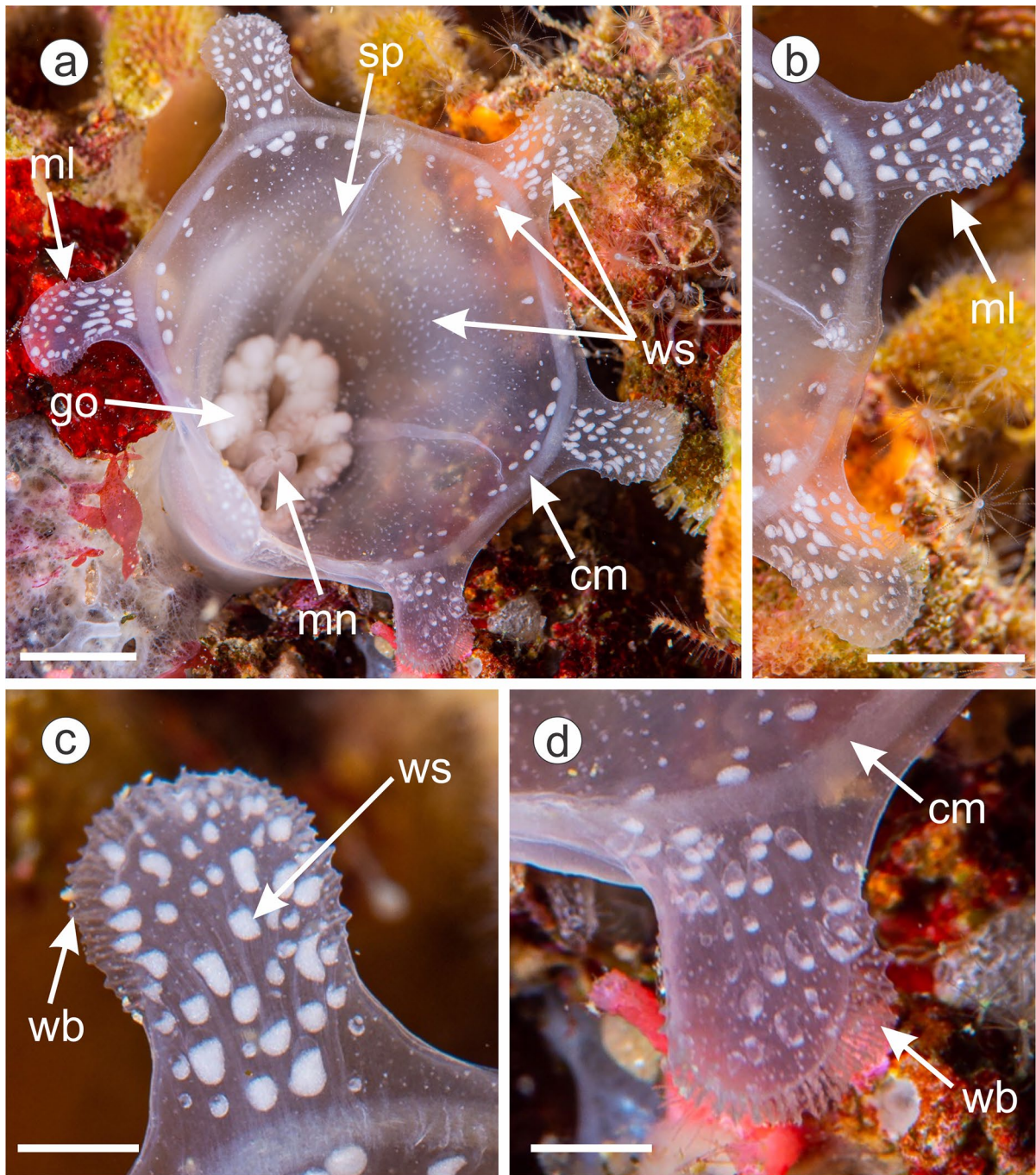
### Molecular inferences

The ML phylogenetic reconstructions for the COI (Fig. S1), 16S (Fig. S2), and combined COI+16S alignments resulted in the same tree topology (Fig. 4). The genus *Lipkea* is closely related to *Lucernaria*, with the *Lipkea* sp. from Portugal closely related to *Lipkea ruspoliana* (with high/full support), from the aquarium of the Oceanographic Museum of Monaco (Fig. 4).

Based on the DNA distance matrix, we observed that, for COI, there is a genetic difference of 1.19% between *Lipkea* sp. from Portugal and *Lipkea ruspoliana* (Table 5), and about 20% between both lineages and *Lipkea* sp. from Japan. The COI sequences of *Halicystus antarcticus* and the closely related *Halicystus auricula* differ by 3.87% (Table 5). The genetic distance of *Lipkea* sp. from Portugal to the other species analyzed ranged from 24.43 to 27.29% for COI (Table 5). For 16S, the genetic distances were smaller, with a 0.19% divergence between *Lipkea* sp. from Portugal and *Lipkea ruspoliana*, and 12.71% between both lineages relative to *Lipkea* sp. from Japan (Table 6). Within *Halicystus*, the genetic difference between *Halicystus antarcticus* and *Halicystus auricula* was 1.17% (Table 6). The genetic distance of *Lipkea* from Portugal to the other species ranged from 30.83 to 31.93% for 16S (Table 6).

## Discussion

In this study, we reported for the first time the presence of *Lipkea* in Portugal. Based on the molecular results for two mitochondrial barcodes (COI and 16S), we conservatively identified the species as *Lipkea ruspoliana*. The percentage of sequence divergence between our specimen and an individual of *L. ruspoliana* sequenced by Miranda et al. (2016a) is within the range of intraspecific sequence variability reported by previous studies on staurozoan species for both the COI (0–2.16%) and 16S (0–0.75%), as well as within the range of the closely related *H. antarcticus* and *H. auricula* (COI: 3.87% and 16S: 1.17%) also included in our analyses (Holst et al. 2019) (Tables 5 and 6). Indeed, the small difference

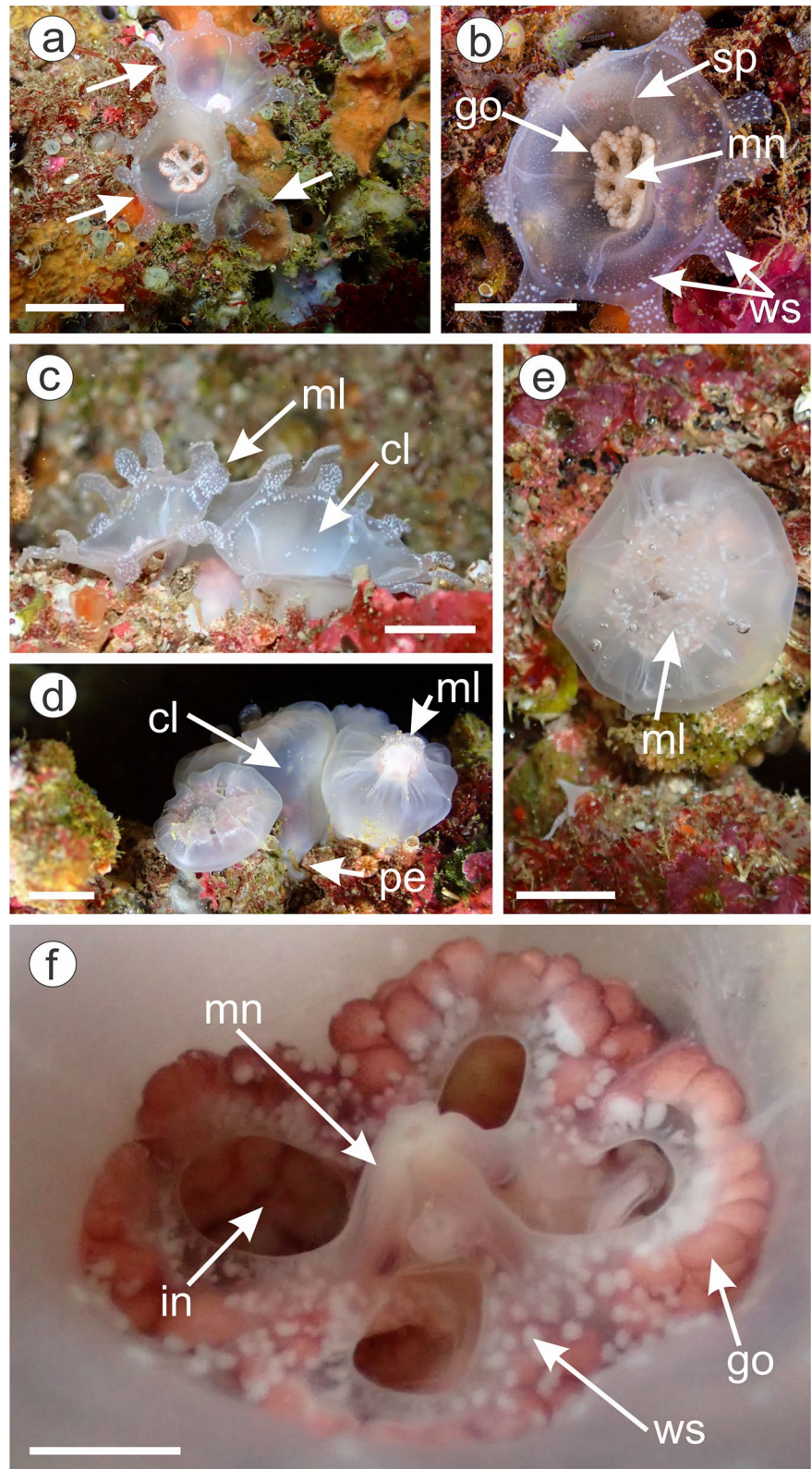


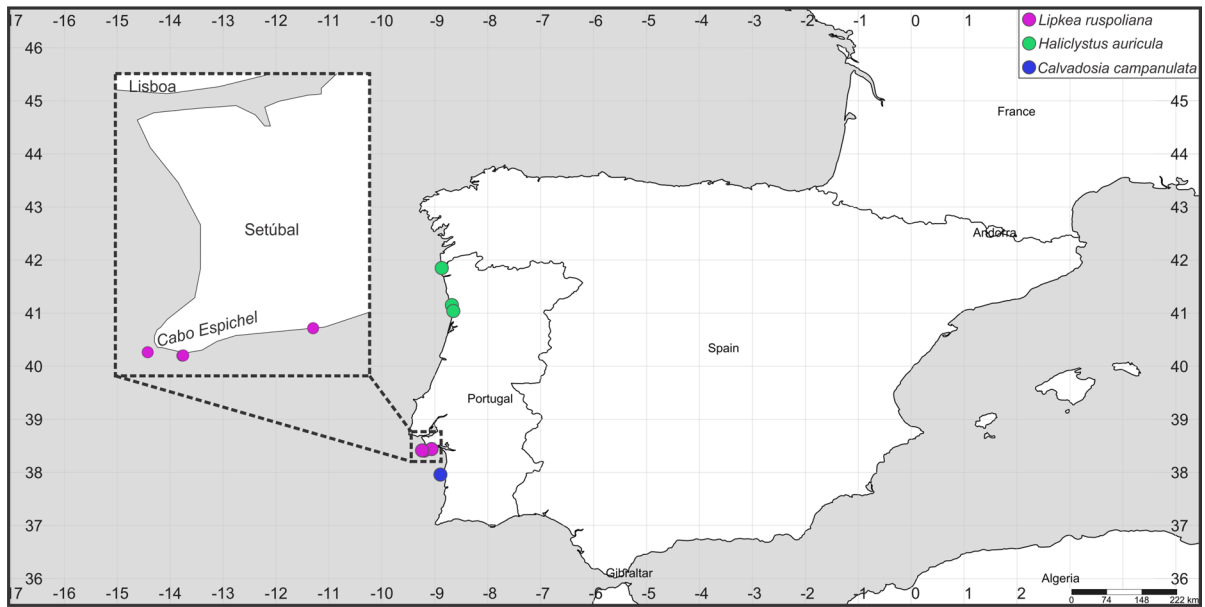
**Fig. 1** Specimens of *Lipkea* from Portugal, evidencing the marginal lappets. **a** General view of the animal in the field; **b** marginal lappets; **c** and **d** detail of marginal lappets with a row of short filiform (possibly tentacular) projections, which are fused to one another by a web. Abbreviations: cm, coro-

nal muscle; go, gonad; ml, marginal lappet; mn, manubrium; sp, septum; wb, web with filiform projections; ws, white spot. Scale = **a**, **b** 5 mm; **c** and **d** 2 mm. Photo credits: Emanuel Gonçalves

**Fig. 2** General morphology of *Lipkea* from Portugal.

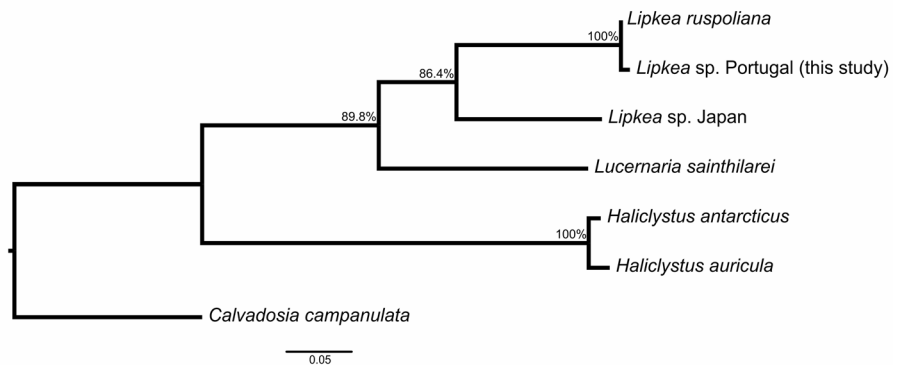
**a** Aggregation of different specimens, in subumbrellar and lateral views; **b** abnormal individual with 5 infundibula and 5 septa; **c** lateral view of the animals in the field; **d** and **e** animals contracted against the substrate; **f** detailed view of the manubrium (oral region), evidencing the infundibula and gonads. Abbreviations: cl, calyx; go, gonad; in, infundibulum; ml, marginal lappet; mn, manubrium; pe, peduncle; sp, septum; ws, white spot. Scales: **a** 15 mm; **b–e** 7.5 mm; **f** 1 mm. Photo credits: Emanuel Gonçalves (**b**) and Sílvia Tavares (**a, c–f**)





**Fig. 3** Geographic distribution of stauromedusae from Portugal (see Tables 1, 2)

**Fig. 4** Maximum likelihood phylogenetic hypothesis including the specimen from Portugal. Analysis based on combined data of mitochondrial markers COI and 16S. The tree was rooted on the node *Calvadosia campanulata* (Amyostaurida). Bootstrap indices under maximum likelihood at each node. See Table 4 for additional information



**Table 5** Uncorrected pair-wise p-differences of mitochondrial COI from the stauzoan species included in this study

#	Species	1	2	3	4	5	6	7
1	<i>C. campanulata</i>	0.0000						
2	<i>H. antarcticus</i>	0.2506	0.0000					
3	<i>H. auricula</i>	0.2558	0.0387	0.0000				
4	<i>L. sainthilarei</i>	0.2645	0.2386	0.2490	0.0000			
5	<i>L. ruspoliana</i>	0.2562	0.2781	0.2545	0.2413	0.0000		
6	<i>Lipkea</i> sp. Japan	0.2578	0.2543	0.2538	0.2421	0.2048	0.0000	
7	<i>Lipkea</i> sp. Portugal	0.2669	0.2729	0.2549	0.2443	0.0119	0.2026	0.0000

previously found between these two *Haliclystus* species raised the possibility that they should be taxonomically reassessed (Holst et al. 2019). The genetic

divergence found between the *Lipkea* specimen from Portugal and *Lipkea ruspoliana* is even lower for both COI (1.19%) and 16S (0.19%). However, genetic

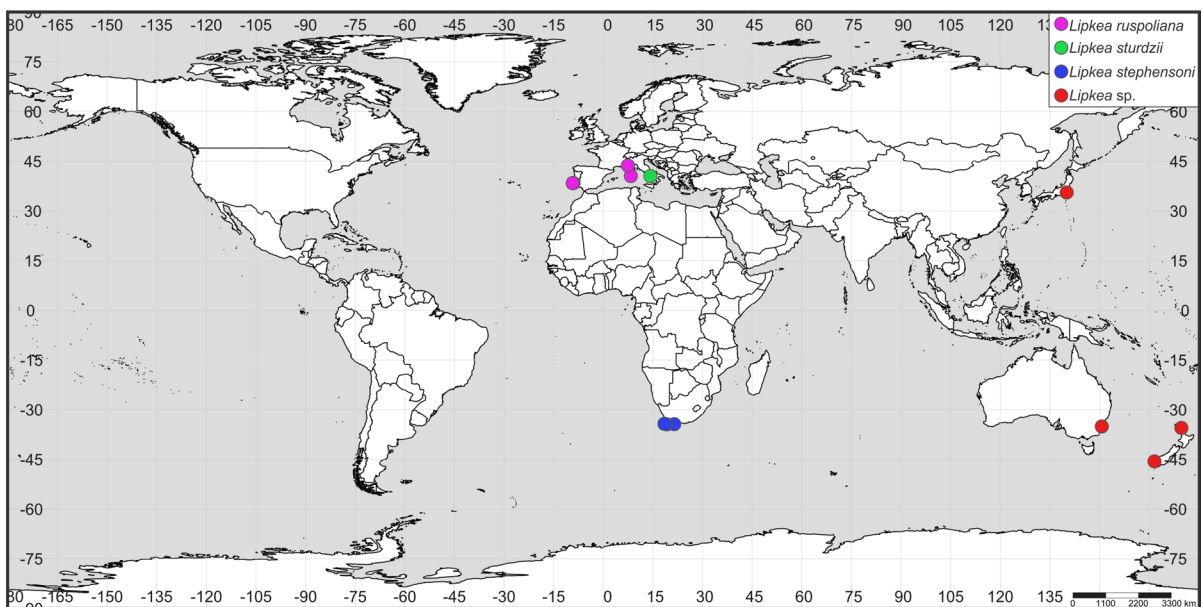
**Table 6** Uncorrected pair-wise p-differences of mitochondrial 16S from the staurozoan species included in this study

#	Species	1	2	3	4	5	6	7
1	<i>C. campanulata</i>	0.0000						
2	<i>H. antarcticus</i>	0.3355	0.0000					
3	<i>H. auricula</i>	0.3381	0.0117	0.0000				
4	<i>L. sainthilarei</i>	0.3152	0.3111	0.3165	0.0000			
5	<i>L. ruspoliana</i>	0.3083	0.3196	0.3222	0.1695	0.0000		
6	<i>Lipkea</i> sp. Japan	0.2828	0.3219	0.3317	0.1685	0.1271	0.0000	
7	<i>Lipkea</i> sp. Portugal	0.3083	0.3167	0.3193	0.1720	0.0019	0.1271	0.0000

distance alone should not be used as a proxy to identify species (Pires and Marinoni 2010; Zamani et al. 2022; Zhang and Bu 2022) and these data should be interpreted with caution as Staurozoa still lacks a comprehensive characterization of intraspecific genetic divergence to establish the limits between species (Lutz et al. 2006; Miranda et al. 2016a).

The genus *Lipkea* is currently represented by three species: *Lipkea ruspoliana* Vogt 1886, *Lipkea sturdzii* (Antipa 1893), and *Lipkea stephensoni* Carlgren 1933 (Table 3; Fig. 5), all of which were originally described from single specimens. The description of *Lipkea ruspoliana* was based on a specimen from Alghero, on the Sardinian coast of the Mediterranean Sea (Vogt 1886, 1887), and has been recently recorded in aquaria of the Oceanographic Museum

of Monaco (Pisani et al. 2007). *Lipkea sturdzii* was described from a specimen from Capri Island, Gulf of Naples, Italy. Originally, it was described as *Capria sturdzii* (Antipa 1893), but later Carlgren (1933) synonymized the genera *Lipkea* and *Capria*. The species has not been reported again since its original description. In contrast, *Lipkea stephensoni*, originally described from a specimen from Still Bay, Eden, South Africa (Carlgren 1933), was later reported in several other regions of South Africa (Miranda et al. 2017). In addition to these three species, unidentified species of *Lipkea* have been recorded from New Zealand, Australia, and Japan (Cairns et al. 2009; Zagal et al. 2011; Miranda et al. 2016a). Because of our insufficient knowledge on the geographic distribution of stalked jellyfishes (Miranda et al. 2018) and



**Fig. 5** World geographic distribution of *Lipkea* (see Tables 1, 3)

the unexpected new records of some species (Holst and Laakmann 2019; Ames et al. 2021), it is not possible to infer whether or not *Lipkea ruspoliana* is native in Portugal or an exotic/invasive occurrence. The Arrábida Natural Park is considered a hotspot for marine biodiversity and a Natura 2000 site (Cunha et al. 2014) having species with Mediterranean, subtropical, and temperate affinities (Henriques et al. 2009), being mainly studied for the management of fisheries resources and restoration of seagrass habitats, due to anthropogenic pressures. As the assessment of the biodiversity of the Arrábida Natural Park is still incomplete, it is not possible to reach a conclusion regarding the nature of the species in Portugal. However, regular monitoring may clarify this issue in the future.

There is scarce information in the literature regarding morphological intraspecific variation in *Lipkea*. The major trait that differentiates the species is the morphology of the marginal lappets, a trait that is unique to *Lipkea* despite its homology within the Staurozoa being disputed. *Lipkea ruspoliana* was originally described with eight perradial and interradial lappets, which were suggested to be homologous to the eight primary tentacles (Vogt 1886, 1887)—or later in the development to the eight anchors/rhopalioids (Hirano 1986; Miranda et al. 2016a, b). However, additional observations of *L. ruspoliana* identified 8–12 adradial lappets, which were then interpreted as modified tentacles with an adradial position (Pisani et al. 2007). *Lipkea sturdzii* and *L. stephensoni* were also described with adradial lobes (Antipa 1893; Carlgren 1933), a similar position to the usual eight adradial arms in other stauromedusae. Indeed, Zagal et al. (2011) regarded the lappets as “short, rounded arms.” Studies on the development of *Lipkea* would be helpful to elucidate the homology of the marginal lappets.

In addition to the number of lappets, which have been shown to vary in several *Lipkea* species [*L. ruspoliana*: 6–12 (Vogt 1886, 1887; Pisani et al. 2007; this study); *L. stephensoni*; 8 lappets based on Carlgren 1933, variable number based on Miranda et al. 2017; and *Lipkea* sp. from Australia: 9 or 11 (Zagal et al. 2011)], the morphology of the lappets also appears to be taxonomically informative for the genus. Originally, *L. ruspoliana* was described with smooth marginal lappets (Vogt 1886, 1887; Pisani et al. 2007), whereas *Lipkea sturdzii* has a row of

16–20 toothlike or short finger-shaped rudimentary tentacles, which are fused to one another by a web (Antipa 1893; Mayer 1910). *Lipkea stephensoni* was characterized by short lappets with reduced tentacles in one row along their margin (Carlgren 1933). More recent observations on *L. stephensoni* (Miranda et al. 2017) revealed that specimens at about 23 m of depth had rudimentary tentacles in the lappets, whereas intertidal and subtidal specimens had smooth marginal lappets, similarly to *L. ruspoliana* (Miranda et al. 2017). The original description of *L. stephensoni* (Carlgren 1933) was based on an intertidal specimen, which raises questions regarding the possible influence of the environment on the morphological variation of the marginal lappets. *Lipkea* sp. from Australia was described with marginal lappets “with a single row of 18–27 rudimentary toothlike tentacles” (Zagal et al. 2011), similar to some *L. stephensoni* (Carlgren 1933; Miranda et al. 2017).

Within this context, at least two specimens from Portugal photographed in closer view (collected at the same time and in the same locality as the sample collected for DNA in this study; August 14, 2020, Ponta da Passagem Cabo Espichel, Arrábida Natural Park, Portugal; Table 1) have very similar marginal lappets to those described for *L. sturdzii*, with a clear row of short filiform projections, possibly from tentacular origin, fused to one another by a web (Fig. 1). That evidence, in association with our molecular results and a relatively close geographic occurrence (Fig. 5), strengthens the hypothesis that *L. sturdzii* should be synonymized with *L. ruspoliana*, the name with priority. Broader taxon and habitat sampling for genetic studies and investigations of intraspecific morphological variation (e.g., marginal lappet), including observations at different stages of development, are required to clarify this discussion. Specifically, sampling at the type locality of *L. sturdzii* is encouraged, which can be facilitated by citizen science initiatives such as iNaturalist and GelAvista.

In this study, we reinforce the value of free access, online platforms for scientific purposes. A simple image posted on iNaturalist, followed by a crucial effort of divers and researchers to collect samples for DNA analysis to support image-based (morphological) identification, was decisive to report the first record of *Lipkea* in Portugal and to shed some light on a taxonomic discussion regarding a species (i.e., *L. sturdzii*) not reported for 130 years, i.e., since its

original description (Antipa 1893). The use of these platforms, with an environment that stimulates active collaboration and engagement of the general public in the identification/validation of species records and occurrences, is particularly helpful for rare species such as staurozoans. Furthermore, iNaturalist species records are automatically transferred to the Marine Park biodiversity database ([https://www.inaturalist.org/observations?project\\_id=biodiversity-of-arrabida-s-marine-park](https://www.inaturalist.org/observations?project_id=biodiversity-of-arrabida-s-marine-park)), which is important for local diversity knowledge, scientific dissemination, and conservation.

As a note of caution, it should be highlighted that iNaturalist data is not free of errors (Koo et al. 2022), and these errors need to be cautiously assessed in order to guarantee the scientific reliability of resulting publications (Kosmala et al. 2016). Although online platforms for recording and sharing biodiversity observations can be used to address issues regarding species in inaccessible habitat or with unpredictable occurrences (Cranswick et al. 2022), including taxonomic discussions such as in this study (see also Winterton 2020), photographs cannot replace voucher specimens deposited in collections, a reference for species description and further identifications (Ceríaco et al. 2016; Buckner et al. 2021). Collection of a complete specimen was not possible in this study mainly because of difficult field access and animal contraction, but we succeeded in reporting a new record in a protected area, providing data on species seasonality, recognizing important structures based on photographs, and discussing systematics based on DNA and aspects of morphology, a significant advance that would go unnoticed without the contribution of citizen science.

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#### Declarations

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

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