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A new deep-water *Tethya* (Porifera, Tethyida, Tethyidae) from the Great Australian Bight and an updated Tethyida phylogeny

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Abstract. A new species of *Tethya* Lamarck, 1815 is described from a depth of 1000 m on the continental slope of the Great Australian Bight (GAB), southern Australia. The GAB slope was explored as part of systematic benthic surveys to understand unexplored communities in the light of current oil and gas exploration activity in the area. *Tethya irisae* sp. nov. was present at 1000 m in six of eight longitudinal depth surveys. Three molecular markers were obtained: COI, 28S (D3–D5) and ITS1-5.8S-ITS2. COI and 28S phylogenetic analyses show that the new species fits clearly within the genus *Tethya*. This is the 28th species of *Tethya* reported from Australia; it is unusual in that it has a stalk. The presence of a stalk as a morphological character to split genera in this family is questioned. The description of this new species is an opportunity to revisit the molecular phylogeny of the Tethyida Morrow & Cárdenas, 2015 using comprehensive datasets of COI and 28S markers. As in previous analyses, four *Tethya* clades were retrieved; we discuss the possibility of using external colour to support some of these clades. Despite unclear phylogenetic relationships amongst Tethyidae Gray, 1848 from Australia, our results suggest that tethyid genera *Tethytimea* Laubenfels, 1936, *Tectitethya* Sarà, 1994, *Laxotethya* Sarà & Sarà, 2002, *Stellitethya* Sarà, 1994, and *Xenospongia* Gray, 1858 derive from species of *Tethya*. We show that asters

have been secondarily lost at least twice in the Hemiasterellidae Lendenfeld, 1889: in *Liosina* Thiele, 1899 and a potential new genus from northern Australia. We formally propose the reallocation of *Liosina* from Dictyonellidae van Soest, Diaz & Pomponi, 1990 to Hemiasterellidae Lendenfeld, 1889.

Keywords. Porifera, Dictyonellidae, Hemiasterellidae, *Liosina*, marine benthos.

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Introduction

The family Tethyidae Gray, 1848, reclassified from the order Hadromerida Topsent, 1894 (order abandoned) to order Tethyida Morrow & Cárdenas, 2015, contains 14 genera of which the genus *Tethya* Lamarck, 1815 is by far the most speciose with 93 accepted species (Van Soest *et al*. 2019). Tethyidae are defined by 'stylote megascleres mainly stronglyoxeas, generally in radiate tracts and two categories of euasterose microcleres, microasters and megasters, sometimes rhadbs' (Sarà 2002). Occurrence and length of a stalk are included as key morphological characteristics in distinguishing some genera of Tethyidae (Sarà 1994, 2002; Sarà & Burlando 1994); although the development of the stalk in the family has been noted as being an adaptation to deep-water, soft sediment habitat (Sarà & Burlando 1994). The taxonomy of the Tethyidae was rearranged by Sarà (1994) into eight genera, with the new genera *Burtonitethya* Sarà, 1994 and *Tethycometes* Sarà, 1994 joining *Halicometes* Topsent, 1898, as stalked genera in the family. Cladistic analysis by Sarà & Burlando (1994) placed Tethyidae into three clades (a) the genus *Tethya*, (b) genera with stalks and (c) massive and encrusting genera.

The Australian Faunal Directory (AFD) (Hooper 2012) lists six genera of Tethyidae in Australia (*Anthotethya* Sarà & Sarà, 2002; *Laxotethya* Sarà & Sarà, 2002; *Oxytethya* Sarà & Sarà, 2002; *Stellitethya* Sarà, 1994; *Tethya* Lamarck, 1815; *Xenospongia* Gray, 1858). A seventh genus *Tethyastra* Sarà, 2002 (*Tethyastra oxyaster* (Burton, 1934), 'accepted' (van Soest *et al.* 2019)) is listed in the Codes for Australian Aquatic Biota (Rees *et al.* 1999 onwards); in the AFD this is listed as *Tethya oxyaster*.

The slope of the Great Australian Bight (GAB) was explored in 2010 as part of a preliminary exploration of deep-water communities (Currie & Sorokin 2011) and again in 2015 as part of systematic benthic surveys to understand unexplored communities in the light of current oil and gas exploration activity in the area (MNF 2015; Williams *et al.* 2018). The surveys resulted in the discovery of several new benthic species including sponges. Multiple specimens of a small stalked tethyid were found at 1000 m; here, we describe this new species, using morphological characters and molecular markers (COI, 28S (D3–D5) and ITS1-5.8S-ITS2) to consider how it fits into the family.

The description of this new species is an opportunity to revisit the molecular phylogeny of this group. Heim *et al.* (2007) and Heim & Nickel (2010) produced the first phylogenetic analyses of *Tethya*, combining COI and morphology. Since then, although large Demospongiae trees were produced that led to the creation of the order Tethyida by Morrow & Cárdenas (2015), no phylogenetic studies truly focused on the *Tethya*, Tethyidae or Tethyida. We therefore felt it was time to present an updated molecular phylogeny for COI and ran the first comprehensive 28S phylogenetic analyses focusing on the Tethyida.

Material and methods

Field collection

Tethya irisae sp. nov. was first collected, as only one specimen, in the GAB in 2010 (Currie & Sorokin 2011). More specimens were collected in November and December 2015 as part of systematic epibenthic

surveys of the central GAB slope. Stations were sampled by beam trawl along five longitudinal transects and over depths ranges from 200 m to 5000 m (Fig. 1). Benthic specimens were collected under Australian Commonwealth Area Permit No. AUCOM2015-284 and Commonwealth Marine Reserve Permit No. CMR-15-000344. *Tethya irisae* sp. nov. was found at six out of eight of the 1000 m depth stations. Specimens were photographed on board and fixed in 70% ethanol. Specimens for molecular analysis were fixed in ethanol (> 95%). Most specimens (including the holotype) are lodged at the South Australian Museum (accession prefix SAMA), Adelaide, South Australia; two specimens were deposited at the Queensland Museum (accession prefix QM), Brisbane, Australia; four specimens from lot SAMA S2039 and a thick section are deposited at the Museum of Evolution, Uppsala, Sweden (UPSZTY 178608). Collection information of specimens examined in this study is archived in the open access PANGAEA data repository (https://doi.pangaea.de/10.1594/PANGAEA.894720).

Light microscope preparation

To examine the skeleton, thick sections in resin were prepared from a specimen from collection lot SAMA S2039, following the method described by Boury-Esnault *et al.* (2002). Thin sections were also

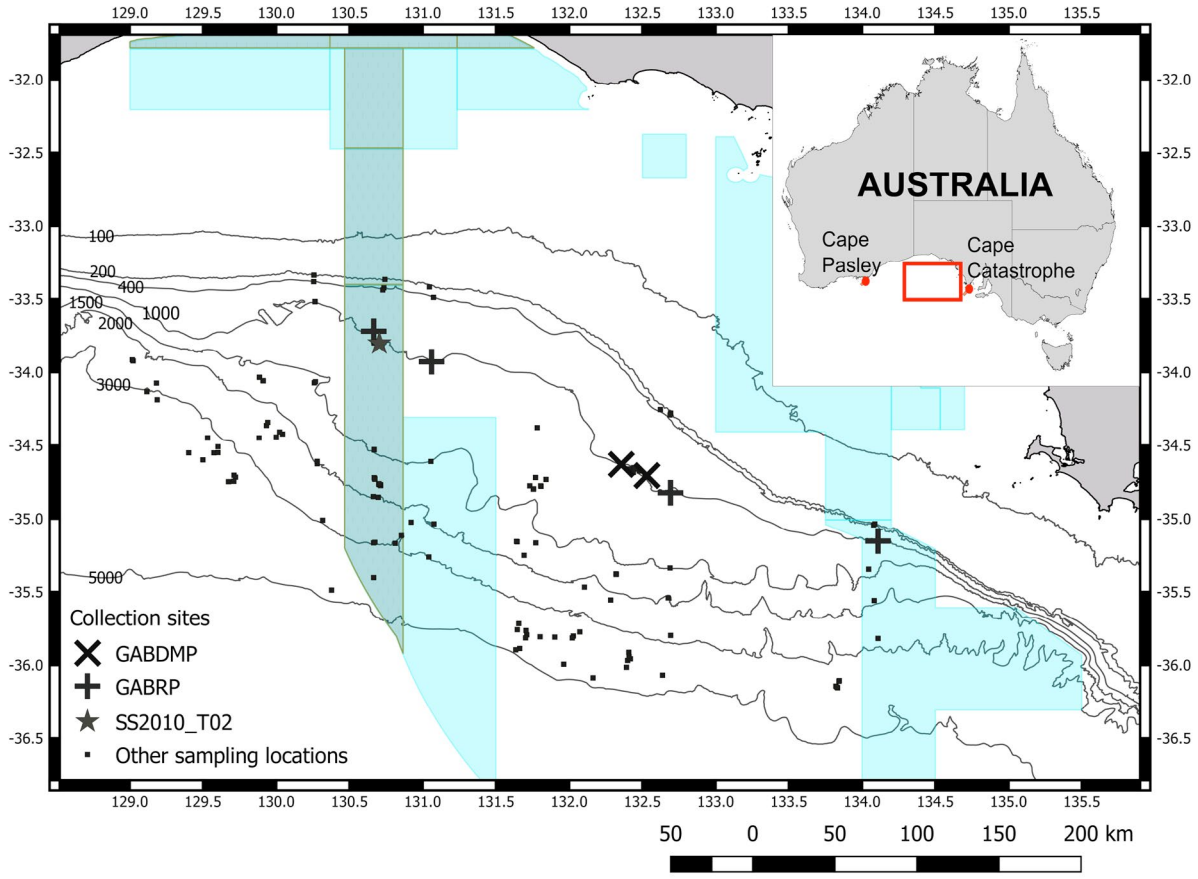


Fig. 1. Sites where *Tethya irisae* sp. nov. was collected in the Great Australian Bight, including sites that were sampled where the sponge was not found. All specimens of *Tethya irisae* sp. nov. were collected along the 1000 m contour. Light shaded polygons show the Australian Commonwealth Marine Reserves. The darker polygon strip is the GAB Marine Park Benthic Protection Zone. The 200 m contour is the edge of the continental shelf. Abbreviations: GABDMP = Great Australian Bight Deepwater Marine Program (MNF 2015); GABRP = Great Australian Bight Research Project (Williams *et al*. 2018). Cruise SS2010_T02 (Currie & Sorokin 2011).

made, with the following protocol: sections were cut perpendicular to the surface with a sharp blade and laid onto a slide, covered with a weighted coverslip and dried on a hot plate. Sections were covered with mounting media (Durcupan™) and dried overnight in an oven at 50°C. Spicule slides were prepared by dissolving a small amount (~ 2 mm^2) of sponge tissue in 3.9% sodium hypochlorite. The resultant spicules were rinsed with distilled water three times and with 95% ethanol twice before mounting on a microscope slide with DPX™ mountant.

SEM preparation

Scanning Electron Microscope (SEM) tissue preparations were made by dissolving the tissue in 12.5% sodium hypochlorite to remove the soft tissue. They were then rinsed twice in distilled water, rinsed twice in 70% ethanol and then finally twice in 98% ethanol and then air dried. SEM preparations were sputter coated in gold to improve resolution. The scanning electron micrograph photos were taken using a Hitachi TM-1000 SEM and plates assembled in Adobe Photoshop. Morphometric measurements of the spicules were done using the same Hitachi TM-1000 SEM.

Spicule terminology

Spicule terminology follows that suggested by Bergquist & Kelly-Borges (1991) for the genus *Tethya*.

Molecular studies

Whole genomic DNA was extracted from sponge tissue frozen at -80°C. A conventional hexadecyltrimethylammonium bromide (CTAB)-based protocol (Taylor *et al.* 2004) was used for isolating DNA. Briefly, the sponge tissues were ground under liquid nitrogen. The CTAB extraction buffer was applied to lyse tissues and then combined with polyvinylpyrrolidone (PVP) and β-mercaptoethanol to help remove phenolic compounds and tannins in the extract. To separate the proteins and polysaccharides from nucleic acids, phenol: chloroform: isoamyl alcohol (25:24:1) was utilised before DNA was precipitated with chilled isopropanol. The mitochondrial cytochrome c oxidase subunit 1 (COI) Folmer fragment was amplified using the universal primers LCO1490 and HCO2198 (Folmer *et al.* 1994). The D3–D5 region of 28S rRNA gene was amplified by primers NL4F and NL4R (Nichols 2005). To amplify ITS, we used primers originally designed for a unicellular eukaryote, ITSRA2 (5′-GTC CCT GCC CTT TGT ACA CA-3′) and ITS2.2 (5′-CCT GGT TAG TTT CTT TTC CTC CGC-3′) (Adlard & Lester 1995), to amplify a 753 bp long sequence including ITS1-5.8S-ITS2 and small fragments of the surrounding 28S and 18S. The thermocycler was programmed following Yang *et al.* (2017). Sequences were assembled and blasted using Geneious® ver. 8.1 (created by Biomatters, http://www.geneious.com).

All Tethyidae COI and 28S sequences from GenBank were collected and aligned with our new sequences in AliView 1.18 (Larsson 2014). No ITS alignment was made since only one other Tethyidae ITS was found on GenBank. The full COI and 28S from *Tethya wilhelma* Sarà, Sarà, Nickel & Brümmer, 2001 were assembled from the whole genome, https://bitbucket.org/molpalmuc/tethya_wilhelma-genome/src (courtesy of W.R. Francis). We included Timeidae Topsent, 1928 and Hemiasterellidae Lendenfeld, 1889 (*Adreus* Gray, 1867 and *Axos* Gray, 1867, *Hemiasterella* Carter, 1879, and *Liosina* Thiele, 1899) sequences that seemed to group close to the Tethyidae in our preliminary analyses and previous studies (Erpenbeck *et al.* 2012; Morrow *et al.* 2012; Redmond *et al.* 2013; Thacker *et al.* 2013). *Raspailia australiensis* Ridley, 1884 and *Ceratopsion axiferum* (Hentschel, 1912) 28S sequences were included as possible species of *Adreus*, as suggested by Morrow *et al.* (2019). We discarded the 28S sequence of *Timea lowchoyi* Hooper, 1986 (AY561871) and *Hemiasterella* sp. WAMZ12383 (AY561947), both from Nichols (2005) that seemed to be mis-identifications: BLAST results indicated, respectively, that they were close to *Cymbaxinella* Gazave, Carteron, Chenuil, Richelle-Maurer, Boury-Esnault & Borchiellini, 2010, and a 93% match with *Hymeniacidon heliophila* (Wilson, 1911). *Hemiasterella* sp. 1 (AY561901, QMG315767, OTU QM2839)

from Nichols (2005) was re-identified as *Adreus* sp. on SpongeMaps (http://www.spongemaps.org). *Hemiasterella* sp. (AY626310, QM G304645, OTU QM0694) from Kober & Nichols (2007) was re-identified as a tethyid by Morrow *et al.* (2012). *Hemiasterella* sp. UCMPWC1021 (AY56197) from Nichols (2005) was re-examined and re-identified as a *Jaspis* sp. (P. Cárdenas, unpublished data), we included this sequence in our outgroup. Finally, no trustworthy sequences of *Hemiasterella* were left to include in the alignment. We also included 28S (C1–D2) sequences from C. Chombard's PhD Thesis (Chombard 1998), never published or submitted to GenBank. These were copied 'by hand' from the 'Annexe B2' from her thesis, and included the following species: *Tethya* sp., *Tethya taboga* (Laubenfels, 1936), *Tectitethya crypta* (Laubenfels, 1949), *Tethya aurantium* (Pallas, 1766) and *Timea* sp. The three first samples were collected by N. Boury-Esnault in Panama in July 1995. Preliminary 28S analyses showed that the Chombard sequence of *T. aurantium* (from Chausey Islands, Normandy, France) had 1 bp difference to *T. citrina* Sarà & Melone, 1965 (HQ379237) from Wales, so we considered that the Chombard specimen had been mis-identified (both species can be difficult to discriminate but are clearly different genetically) and we thus renamed it *T.* cf. *citrina*. We discarded the 28S (D1–D2) sequence of *T. aurantium* (AY552024) from Borchiellini (2004) because it was 100% identical to that of *T. taboga* 28S (C1–D2) from Chombard (1998) and we suspected a mix-up in the sequence submitted. COI alignments were trimmed to contain only the Folmer fragment (658 bp). The 28S alignment included essentially sequences from the C1–D2, D3–D5 and/or D6–D8 regions or from the full 28S (3376 bp). When we had different 28S regions for the same species, we merged those sequences into a single one using the 'merge' option in AliView; we did this for *Timea* cf. *centrifera*, *Tethya citrina*, *Tethya hibernica* Heim, Nickel, Picton & Brümmer, 2007, *Laxotethya dampierensis* Sarà & Sarà, 2002, *Adreus micraster* (Burton, 1956), *Adreus fascicularis* (Bowerbank, 1866) and *Adreus* sp. Alignment was done using MAFFT (Katoh *et al.* 2002) implemented in AliView, then refined by eye; ambiguous regions were kept. We included in both our datasets sequences of Placospongiidae that are phylogenetically close to the Tethyida, maybe even their sister-group (Morrow *et al.* 2013; Thacker *et al.* 2013). The final COI alignment contained 39 sequences, including five outgroup sequences from *Paratimea* Hallmann, 1917 (Stelligeridae Lendenfeld, 1898), *Jaspis* Gray, 1867 (Ancorinidae Schmidt, 1870) *Cliona* Grant, 1826 (Clionaidae d'Orbigny, 1851) and *Placospongia* Gray, 1867 (Placospongiidae Gray, 1867). The 28S alignment contained 45 sequences, including three outgroup sequences from *Placospongia* and *Trachycladus* (Trachycladidae Hallmann, 1917).

Analyses were conducted with the CIPRES science gateway (http://www.phylo.org) (Miller *et al.* 2010): RAxML 8.2.10 (Stamatakis 2014) for maximum likelihood (ML) and MrBayes v. 3.2.6 (Ronquist *et al.* 2012) for Bayesian analyses. For RAxML, 1000 bootstrap iterations were run; bootstrap Bayesian analyses consisted of two runs of four chains, each for 5 000 000 generations and sampled every 1000th tree after a 25% burn-in.

Abbreviations

bp = base pairs
diam. = diameter
h = height
l = length
w = width

Collection acronyms

ABTC = Australian Biological Tissue Collection, South Australian Museum, Adelaide, Australia
AM = Australian Museum, Sydney, Australia
BMNH = The Natural History Museum (formerly British Museum of Natural History), London, UK
QM = Queensland Museum, Brisbane, Australia

SAMA = South Australian Museum, Adelaide, Australia
UPSZTY = Museum of Evolution, Uppsala, Sweden

Results

Systematics

Class Demospongiae Sollas, 1885
Order Tethyida Morrow & Cárdenas, 2015
Family Tethyidae Gray, 1848
Genus *Tethya* Lamarck, 1815

Tethya irisae sp. nov.
urn:lsid:zoobank.org:act:26151082-02AC-41F5-9E22-15EF212DBDC3
figs 1–3, 4A

Etymology

Named after the golden-winged Greek goddess Iris, grandchild of Tethys, who could reach all parts of the cosmos including the deep sea; and in memory of marine naturalist Iris Sorokin.

Material examined

Holotype

AUSTRALIA • Size 16.6 mm total height (body 11.9 mm (h) × 11.7 mm (w), stalk 4.7 mm (l) × 1.77 mm diam., raised apical osculum); Great Australian Bight (GAB); 34.822° S, 132.69° E; 1006 m depth; Great Australian Bight Research Project (GABRP) leg.; epibenthic sled; SAMA S3387.

Paratypes

AUSTRALIA • 4 specs; same collection data as for holotype; SAMA S2913, SAMA S3388, QM G305000, QM G305001 • 1 spec.; Great Australian Bight; 33.928° S, 131.06° E; 1027 m depth; GABRP leg.; epibenthic sled; UPSZTY 178608.

Additional material at South Australian Museum (sighted only)

AUSTRALIA • 1 spec.; Great Australian Bight; 33.928° S, 131.06° E; 1027 m depth; GABRP leg.; epibenthic sled; SAMA S2039 • 1 spec.; Great Australian Bight; 35.152° S, 134.109° E; 1021 m depth; GABRP leg.; epibenthic sled; SAMA S2371 • 1 spec.; Great Australian Bight; 33.718° S, 130.66° E; 1005 m depth; GABRP leg.; epibenthic sled; SAMA S2482 • 1 spec.; Great Australian Bight; 34.629° S, 132.35° E; 1021 m depth; Great Australian Bight Deepwater Marine Program (GABDMP) leg.; epibenthic sled; SAMA S2095 • 1 spec.; Great Australian Bight; 34.705° S, 132.53° E; 987 m depth; GABDMP leg.; epibenthic sled; SAMA S2096 • 1 spec.; Great Australian Bight; 33.802° S, 130.70° E; 1000 m depth; D. Currie leg.; epibenthic sled; SAMA S1461.

Comparative material

INDIA • 1 section in slide, holotype of *Burtonitethya gemmiformis* Sarà, 1994; Andaman Islands; depth unknown; BMNH 1957.7.15.1.

AUSTRALIA – **New South Wales** • 1 spec., syntype and slides of *Tethya fissurata* Lendenfeld, 1888; Port Jackson; "33°51′ S, 151°16′ E [33.85° S, 151.27° E]; depth unknown; AM G.9069 (syntype), Z6053, Z6893 (slides).

NEW ZEALAND • 1 spec., holotype (specimen and slides) of *Tethya bullae* Bergquist & Kelly-Borges, 1991; Alderman Island; "36°58′ S, 176°05′ E [36.97° S, 176.08° E]; 100 m depth; AM Z5074.

DNA barcoding

COI (MH518072), 28S (D3–D5) (MH511148), ITS1-5.8S-ITS2 (MH511149). All sequences came from the same individual from lot SAMA S2913, although this is a different individual than the type specimens. A tissue sample from this voucher is deposited at the Australian Biological Tissue Collection at the South Australian Museum, Adelaide (ABTC145318).

Description

A small, spherical to oval, stalked, sponge (Fig. 2). The sponge body is 11–14 mm diam., with the stalk approximately the same length as the diameter of the sponge. The surface is covered in polygonal plate-like tubercules (2–3 mm diam.) separated by grooves (0.5 mm wide, 0.25–0.5 mm deep). The sponge is firm to hard and spiculose. Grey/white in life and in ethanol. There is a single raised apical osculum. No sign of any budding.

Skeleton. A stalk of dense megascleres supports the sponge. There is a 'nucleus' where the stalk meets the centre of the sponge body, and although the stalk may divide and/or flatten and thicken externally it emanates from the same point at the base of the sponge. From the nucleus dense bundles (0.3–0.7 mm in

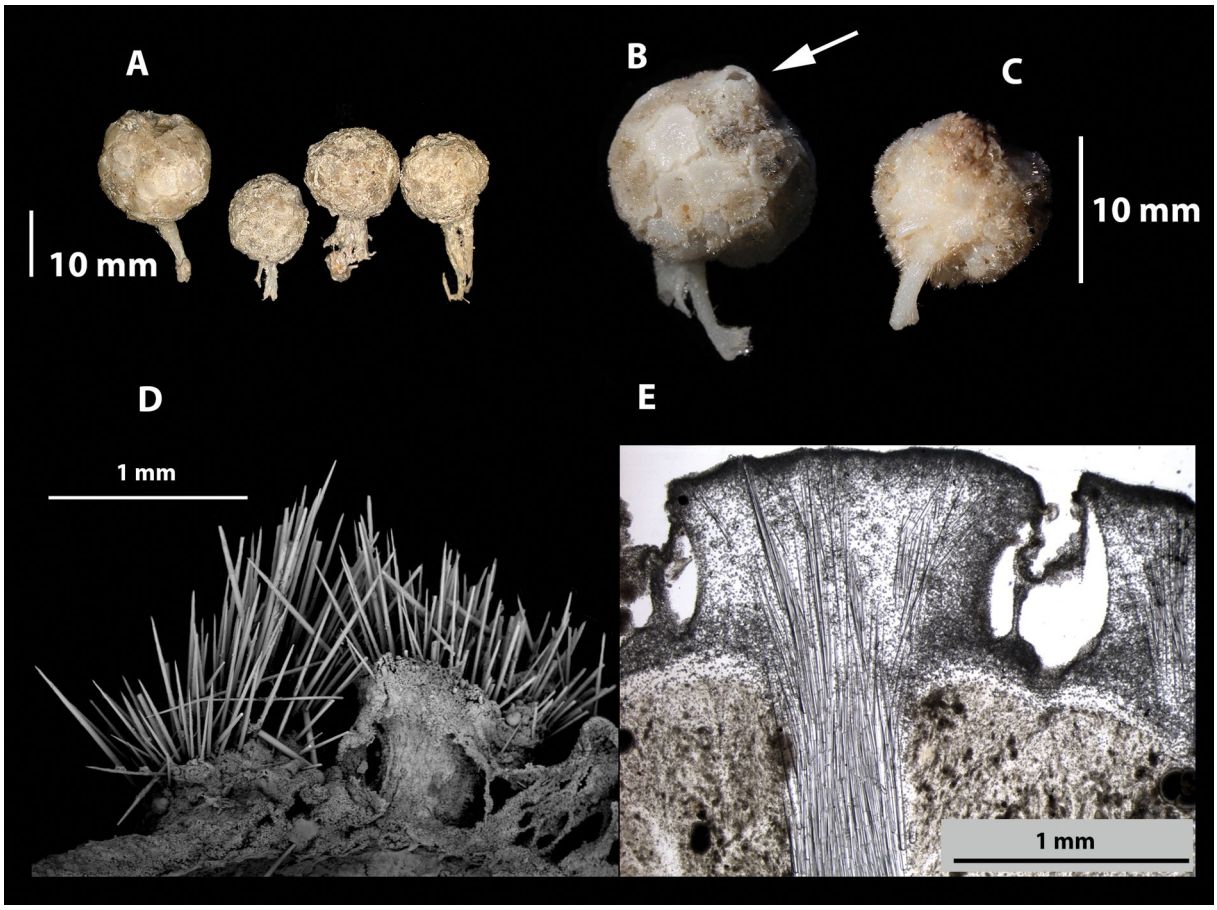


Fig. 2. A. Freshly collected specimens (lot SAMA S2096) of *Tethya irisae* sp. nov. **B**. Paratype (QM G305000) showing single apical oscule (arrow), and tessellated plate-like polygonal tubercules. **C**–**D**. Holotype (SAMA S3387), entire specimen and SEM showing surface tubercules with emerging megascleres. **E**. Section of UPSZTY 178608, showing the well-developed cortex and cortical canals around the tubercules.

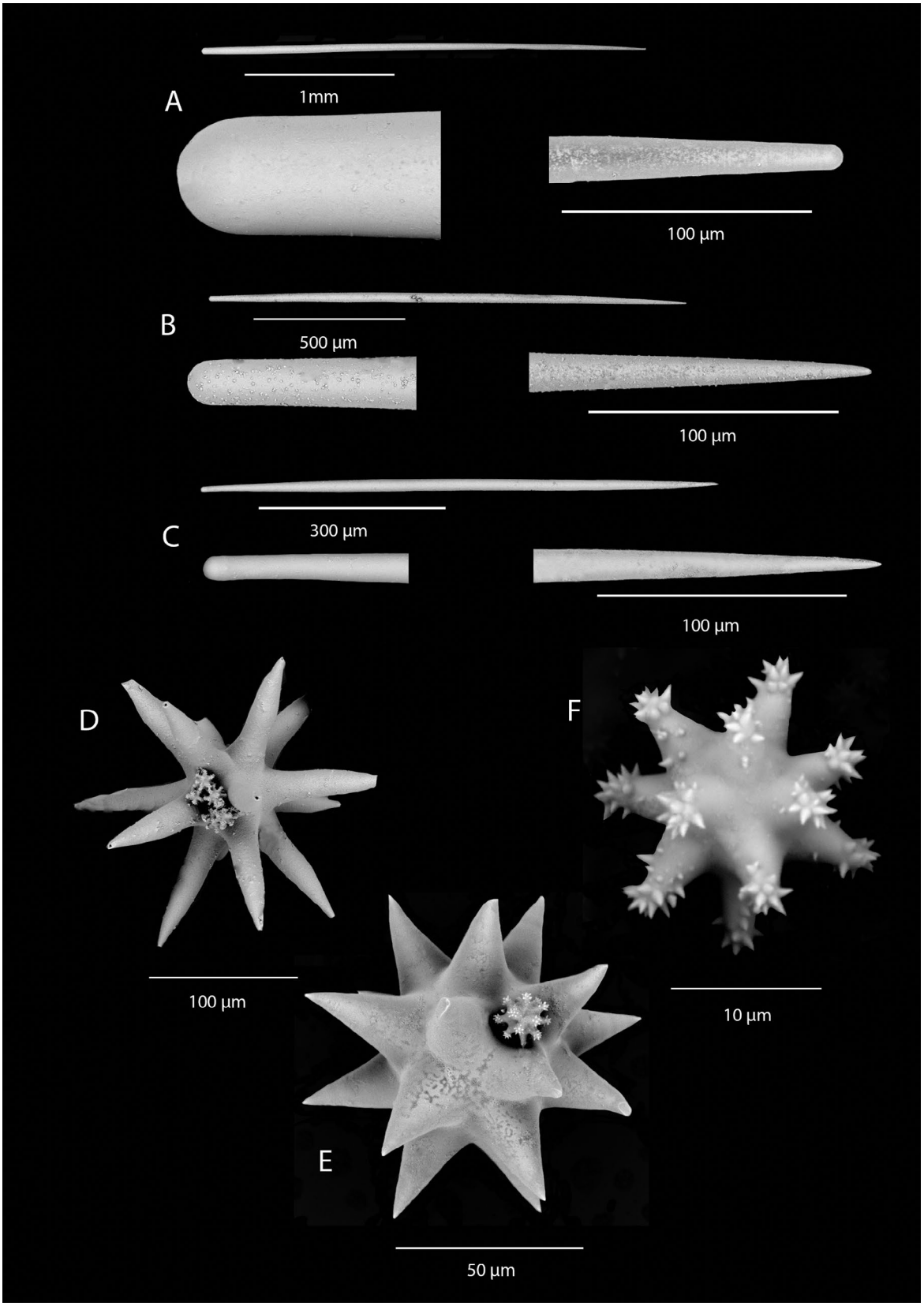


Fig. 3. *Tethya irisae* sp. nov. spicules. **A**–**B**. Straight style/strongyloxeas. **C**. Subtylostyle. **D**. Long-rayed oxyspheraster. **E**. Short-rayed oxyspheraster with small acanthooxyspheraster. **F**. Acanthooxyspheraster.

diameter) of megascleres radiate through the choanosome to the surface tubercles; the bundles slightly fan out in the tubercles. The cortex is a dense layer of microscleres and oxyspherasters interspersed with megascleres emerging through the tubercules, making the surface microscopically hispid (Fig. 2D–E). The cortex is well developed and follows the contours of the tubercules, 1–1.7 mm thick. Large cortical canals are visible between tubercles (Fig. 2E). A thin fibrous layer is below the cortex, it has microscleres in a much lower density. Large oxyspherasters are especially found at the base of the cortex. The megascleres of the stalk are covered in a layer of microscleres and regularly interspersed with short-rayed oxyspherasters. The choanosome is rich in sediment-like particles; there are some microscleres and rare oxyspherasters. Foraminifera (*Globigerina* d'Orbigny, 1826) and Radiolaria are common in the cortical canals and the choanosome.

Spicules. Megascleres are straight style/strongyloxeas (size range 900–3060 × 17–52 μm) (Table 1, Fig. 3A–B) the proximal end is smooth and rounded, the distal end is tapered (not stepped) and either rounded or pointed. There are auxiliary thinner styles to subtylostyles in the medulla between the main styles (260–900 × 7–22 μm) (Fig. 3C). Megaster microscleres are two types of oxyspherasters: long-rayed oxyspherasters (120–185 μm) (Fig. 3D) have ~15 rays that can be bent towards the oxeote tips (ray profile is conical); short-rayed oxyspherasters (53–154 μm) (Fig. 3E) have a larger centrum ~17 rays with a conical profile and oxeote tips. Micraster microscleres are acanthooxyspherasters (12–20 μm) (Fig. 3F), with a centrum and spined tips, and lightly spined on the rays.

Ecology and distribution

Found on the continental slope in the Great Australian Bight at a depth of 1000 m, in soft sediment (clay/silt).

Remarks

The morphology as well as molecular markers confirm that our new sponge is a *Tethya*. Table 2 shows morphological comparisons between other species of *Tethya* from Australia and New Zealand. The external appearance of *Tethya irisae* sp. nov. is similar to *T. fissurata* from Port Jackson, New South Wales, Australia, which is spherical with polygonal tubercules and has a stalk. However, *T. fissurata* differs from *T. irisae* sp. nov. in body size (~ 4 cm diam.), tubercule shape, and number of oscula (2–4). *Tethya fissurata* has megascleres with stepped ends unlike *T. irisae* sp. nov., which are smooth and *T. fissurata* lacks the short-rayed oxyspherasters seen in *T. irisae* sp. nov. Although we do not know the exact depth at which *T. fissurata* was collected, Port Jackson (viz. Sydney Harbour) is not deeper than 45 m (Johnston *et al.* 2015), so this is presumably a shallow species. *Tethya bullae* is a deep-water (100 m) sponge that is of comparable size to *T. irisae* sp. nov., although it has prominent raised tubercules rather than the flat plate-like tubercules of *T. irisae* sp. nov. (Fig. 4). The holotype from the Australian Museum does not include a stalk but the description and photograph in Bergquist & Kelly-Borges (1991) shows "basal flattened branched rooting processes". The long-rayed oxyspherasters of *T. irisae* sp. nov. are similar to those of *T. bullae*. The short-rayed oxyspherasters in *T. irisae* sp. nov. do not fork as those of *T. bullae*. *Tethya irisae* sp. nov. has lightly spined acanthooxyspherasters compared to the completely spined acanthooxyspherasters of *T. bullae*. In addition to *T. fissurata* and *T. bullae*, other *Tethya* with rooting processes/stolons are shown in Table 2 (descriptions in bold text). It is difficult to tell how similar the rooting processes are to each other but these species differ in spicule forms and dimensions from *T. irisae* sp. nov. For example: species with megascleres < 2000 μm (*T. acuta* Sarà & Sarà, 2004, *T. bergquistae* Hooper in Hooper & Wiedenmayer, 1994, *T. burtoni* Sarà & Sarà, 2004, *T. dendyi* Sarà & Sarà, 2004, *T. robusta* (Bowerbank, 1873), *T. seychellensis* (Wright, 1881), *T. stolonifera* Bergquist & Kelly-Borges, 1991); species with megasters not of a 'spheraster' form (*T. amplexa* Bergquist & Kelly-Borges, 1991, and *T. fastigata* Bergquist & Kelly-Borges, 1991); species with very different microscleres (*T. ingalli* Bowerbank, 1858, *T. flexuosa* Sarà & Sarà, 2004 and *T. monstrosa* (Burton, 1924)).

Table 1. Spicule dimensions of *Tethya irisae* sp. nov. holotype (SAMA S3387) and paratypes (QM G305000, QM G305001).

Spicule type	Specimen #	n	Size range (µm) Mean value underlined
Strongyloxeas (l × w)	S3387	34	897–1361–3060 × 17–27–52
	G305000	26	1110–2310–3130 × 19–42–72
	G305001	36	1270–2495–3160 × 20–39–64
Styles (l × w)	S3387	13	262–675–1090 × 7–16–23
	G305000	16	830–1067–1730 × 11–20–33
	G305001	8	1250–1525–1970 × 19–27–34
Short-rayed oxyspherasters (diam.)	S3387	75	53–81–154
	G305000	41	44–99–177
	G305001	59	49–98–133
Long-rayed oxyspherasters (diam.)	S3387	21	120–153–185
	G305000	17	154–196–253
	G305001	2	131–134–136
Acanthooxyspherasters (diam.)	S3387	55	12–15–20
	G305000	43	13–15–19
	G305001	32	13–16–19

In addition, *T. irisae* sp. nov. is collected at the start of the bathyal zone (~1000 m). The deepest of the *Tethya* is *T. compactus* Bergquist, 1961 (402 m), which has very different external morphology.

It occurred to us that when using the key to genera of Tethyidae (Sarà 2002), *Tethya irisae* sp. nov. appears closest to the monospecific genus *Burtonitethya*, a tethyid with a stalk of equal length to the diameter of the sponge. The type of *Burtonitethya* (*B. gemmiformis*), was collected from the Andaman Sea at an unknown depth (Sarà 1994). *Burtonitethya gemmiformis* was originally assigned to *Tethya* (labelled as *Tethya gemmiformis* Burton & Rao, 1957 on the NHM microscope slide) but was re-assigned to a new genus *Burtonitethya* by Sarà (1994) on account of the stalk, the conspicuous nucleus with strongyles, the reduced lacunar cortex, the specialised surface tubercules and the giant oxyaster megasters. Our new species clearly differs from this species in having different microscleres and does not have the giant megasters present in *B. gemmiformis*. As there is no specimen of the type species of *Burtonitethya* and thus no potential to sequence the sponge, we cannot test if *Burtonitethya* is a junior synonym of *Tethya*.

As seen above, the genus *Tethya* shows many different modes of attachment including basal stolons, basal roots, curved peduncles, flattened rooting processes as well as attachment discs and narrow skirts of tissue. Our results suggest that the stalk may not be a good genus-defining character within the family. Heim *et al.* (2007) in their analysis of *Tethya* species, for which they used morphological characters and molecular markers, suggest that characters pertaining to ecological influences may have developed several times. Similarly, we suggest that some of the external morphological characters used to separate genera of Tethyidae are homoplasious, probably appearing several times in different clades of *Tethya* and we question whether they should be grouped as definitive characters in morphological identifications. In the same way the genus *Amphitethya* Lendenfeld, 1907 (Family Tetillidae Sollas, 1886) was created based on its stalk, but phylogenies show it is a *Cinachyrella* Wilson, 1925 (Szitenberg *et al.* 2013; Schuster *et al.* 2017).

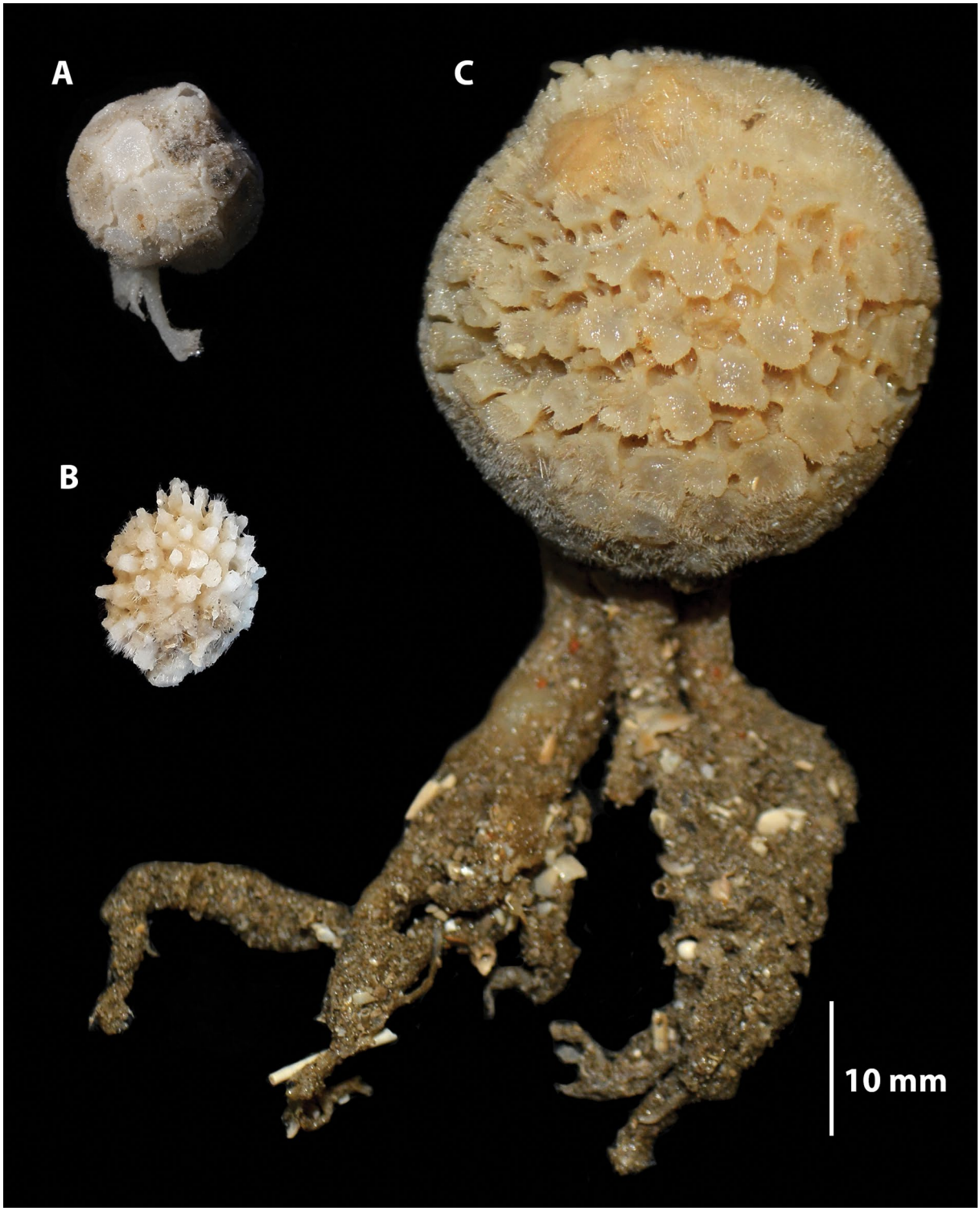


Fig. 4. Comparative sizes and external morphology of species of *Tethya* Lamarck, 1815. **A**. *Tethya irisae* sp. nov., holotype (SAMA S3387). **B**. *Tethya bullae* Bergquist & Kelly-Borges, 1991, part of the holotype (AM Z5074). **C**. *Tethya fissurata* Lendenfeld, 1888, syntype (AM G.9069) Note: the original photo of *T. bullae* (Bergquist & Kelly-Borges 1991) shows rooting processes.

Table 2 (continued on five next pages)**.** Comparison of morphology between species of *Tethya* Lamarck, 1815 from Australia and New Zealand.

Species	**External appearance** Significant attachment in bold	**Body diam. Colour**	**Skeletal arrangement** Megasclere tracts (diam. in µm) & interstitial brushes	**Spicules** **Ms** = megascleres, **me** = megasters, **mi** = microsters (size in µm)	**Type locality**	**Depth** (m)
T. irisae sp. nov.	Spherical; plate-like polygonal tubercules separated by grooves; apical osculum; hard. **Stalk, same length as body diam.**	11 mm Grey/white	Tracts of megasclere bundles (300–700) are uniform from choanosome to cortex and fan slightly in tubercules.	**Ms:** strongyloxeas (to 3000 × 70), styles (to 1970 × 34). **me:** long & short-rayed oxyspherasters (44–177) **mi:** acanthooxyspherasters (12–20)	Great Australian Bight	1000
T. acuta Sarà & Sarà, 2004[1]	Spherical with small, distinct tubercules; firm. **Buds and rhizoid protuberances.**	13 mm Unknown*	Thick tracts of megasclere bundles (250) fanning out in the cortex.	**Ms:** strongyloxeas, anisostrongyles, styles (to 1600 × 20) **me:** oxyspherasters (15–50) **mi:** oxyasters (8–20)	Pt Phillip Heads, Victoria, Australia	Unknown*[a]
T. amplexa Bergquist & Kelly-Borges, 1991[2]	Spherical to subspherical; surface encrusted with animals. Broad flat tubercules. Apical oscule. **Long curved flexible peduncle.**	30–70 mm Yellow	Robust bundles (440–800) diverging into branches in cortex. Brushes of interstitial megascleres.	**Ms** anisostrongyloxeas (to 2300 × 32) **me:** oxyasters (16–35) **mi:** various, incl**.** acanthotylasters acanthochiasters, acanthooxyspherasters (10–20)	Mimiwhangata, New Zealand	7
T. bergquistae Hooper in Hooper & Wiedenmayer 1994[1,2]	Spherical to ovate; surface mammillate to tessellated with buds arising on filaments; firm compressible. Oscules in groups of 3–4). **Root-like processes.**	20–25 mm Rose pink	Widely separated thin flexuous tracts of megasclere bundles (171–612) radiating into tertiary branches at surface. Interstitial megasclere brushes.	**Ms:** anisostrongyloxeas (to 1617 × 18) **me:** oxyspherasters (18–52), oxyasters (22–31) **mi:** acanthotylasters/acantho-strongylasters (10–17), micro-oxyasters (7–10).	Whitianga, New Zealand	27
T. bullae Bergquist & Kelly-Borges, 1991[2]	Spherical to ovate; large blunt well separated tubercules. **Basal flattened branched rooting processes.**	10–14 mm Yellow grey	Tracts of megasclere bundles (500) uniform from choanosome to cortex, where they radiate slightly to support the raised tubercules.	**Ms:** anisostrongyloxeas (to 2225 × 30) **me:** long and short-rayed oxyspherasters **mi:** completely microspined acanthooxyspherasters & acanthostrongylasters (11–15), microoxyspherasters (9–10)	Alderman Islands, New Zealand	100
T. burtoni Sarà & Sarà, 2004[1]	Spherical to hemispherical; tubercules conulated, flat or inconspicuous. Sponges clump together. **Small marginal stolons.**	10–13 mm Pale orange brown	Tracts of megasclere bundles (500–750) unbranched. Brushes of auxiliary interstitial megascleres.	**Ms:** strongyloxeas/anisostrongyles to (1800 × 40) + interstitial styles **me:** oxy-/spherasters (40–80) **mi:** tylasters (8–14), chiasters or tylote oxyasters (12–15)	Tauranga, New Zealand	1

Table 2 (continued)**.** Comparison of morphology between species of *Tethya* Lamarck, 1815 from Australia and New Zealand.

Species	**External appearance**	**Body diam. Colour**	**Skeletal arrangement**	**Spicules**	**Type locality**	**Depth** (m)
T. compacta Bergquist, 1961[3]	Rhomboid, with lateral fan-like projections. Stony.	8 mm Mid brown (in ethanol)	Tracts (356–722) (in ectosome). Endosome with radiate structure.	**Ms:** strongyloxeas (to 1574 × 21) **me:** spherasters (25–43) **mi:** tylasters (7–12)	Chatham Rise, New Zealand	402
T. communis Bergquist & Kelly-Borges, 1991[2]	Spherical to subspherical; growing in clumps.	7–12 mm Dull grey pink (in ethanol)	Flexuous tracts of megasclere bundles (125–300) have elaborate tertiary branching.	**Ms:** anisostrongyloxeas (to 1225 × 12) **me:** oxyspherasters (38–55), oxyasters (7–12) **mi:** includes polyrhabds	Cape Banks, New South Wales, Australia	Intertidal
T. dendyi Sarà & Sarà, 2004[1]	Body covered in small, flattened tubercules. **Rhizoid-like stolons.**	20 mm Pale buff (in ethanol)	Tracts of megasclere bundles (250–500) branch in choanosome and form fans in cortex.	**Ms:** strongyloxeas and styles (to 980 × 15) **me:** spherasters 20–30 **mi:** tylasters/chiasters/oxyasters (10–16)	Pt Phillip Heads, Victoria, Australia	Unknown[a]
T. expansa Sarà & Sarà, 2004[1]	Irregular shape, very hard, irregular labyrinthine tubercules.	20–30 mm Unknown	Tracts of megasclere bundles (250–500) form secondary branching in cortex with fans at surface.	**Ms:** strongyloxeas and anisostrongyles (1400 × 28) **me:** spherasters/oxyasters (30–50) **mi:** tylasters/chiasters/oxyasters (7–13)	Ahipara Bay, New Zealand	Unknown
T. fastigata Bergquist & Kelly-Borges, 1991[2]	Ovoid; 1–3 large raised apical oscules; surface conulose. **Short, thick stolons.**	40–60 mm Bright orange	Fine, widely separated tracts of megasclere bundles (275–539) radiate slightly towards surface.	**Ms:** anisostrongyloxeas (to 2425 × 35) **me:** oxyasters (18–39) **mi:** various incl. acanthotylasters/chiasters (9–20)	Poor Knights Isl. New Zealand	25–40
T. fissurata Lendenfeld, 1888[4,5]	Spherical; tubercules separated by deep grooves. **Thick stalk which divides into roots.**	40 mm Beige (in ethanol)	Tracts of megasclere bundles branch in the tubercules, supporting the edge of the tubercules and spicules extend through the surface.	**Ms:** strongyloxeas (to 4000 × 80) **me:** short-rayed spherasters (160), long-rayed spherasters (240) **mi:** oxyasters (55), tylasters (19)	Pt Jackson, New South Wales, Australia	Unknown[b]
T. flexuosa Sarà & Sarà, 2004[1]	Irregular spherical shape. Tubercules rounded and slightly conical and carry thin filaments. Elastic consistency. **Basal rhizoid stolons.**	18–35 mm Brownish-white	Tracts of megasclere bundles (250–750) branch into secondary and tertiary branches in cortex.	**Ms** strongyloxeas and anisostrongyles (to 2600 × 50) **me:** spherasters & oxyspherasters (30–80) **mi:** tylasters (10–15), oxyasters (10–50)	NW Australia	30–62
T. gigantea (Lendenfeld, 1888)[4]	Spherical; surface with 7 mm high mounds; tuberculose.	60–90 mm Orange-red	Widely spaced tracts of megasclere bundles (500–700), separating only slightly in surface tufts.	**Ms:** strongyles (2000 × 35) **me:** oxyspherasters (30–60) **mi:** chiasters/oxyasters (10–20) & polyrhabds	Pt Jackson, New South Wales, Australia	Unknown[b]

Table 2 (continued)**.** Comparison of morphology between species of *Tethya* Lamarck, 1815 from Australia and New Zealand.

Species	**External appearance**	**Body diam. Colour**	**Skeletal arrangement**	**Spicules**	**Type locality**	**Depth** (m)
T. gunni Sarà & Sarà, 2004[1]	Smooth with small conulated tubercules.	25 mm Faded purple (in ethanol)	Tracts of megasclere bundles (250–500) branch in the choanosome ending in compact fans in tubercules.	**Ms** strongyloxeas (to 1850 × 35), styles (to 800 × 10) **me:** spherasters (20–55) **mi:** tylasters/chiasters/oxyasters	Tasmania, Australia	Unknown
T. hooperi Sarà & Sarà, 2004[1]	Body ellipsoid; smooth surface, no tubercules.	20–30 mm Ochraceous (in ethanol)	Tracts of megasclere bundles (250–500) radiate in cortex with secondary and tertiary branching.	**Ms:** strongyloxeas (to 1530 × 20) **me:** spherasters (30–60) **mi:** tylasters (8–14) oxyasters (15–25)	Heron, Is. Great Barrier Reef, Australia	12
T. ingalli Bowerbank, 1858[1]	Spherical/sub-spherical; flattened often inconspicuous tubercules. **Basal stolons.**	30–50 mm Pinkish	Tracts of megasclere bundles split into secondary and tertiary branches in cortex.	**Ms:** strongyloxeas/anisostrongyles (to 2130 × 30) **me:** oxyspherasters (70–90) **mi:** tylasters (10–15), chiasters, oxyasters (20–30)	SW Australia	30–50
T. japonica Sollas, 1888[6]	Spherical; conules rounded; gemmiferous.	13–22 mm Greyish white	[No details on tracts.]	**Ms:** strongyloxeas (to 1500 × 26) **me:** spherasters (67) **mi:** chiasters (12)	Eastern Philippines	32
T. laevis (Lendenfeld, 1888)[4]	Spherical/ovate; surface smooth to tuberculate.	30–45 mm Light brown (in ethanol)	Tracts of megasclere bundles (500–800) expand to surface tufts.	**Ms:** strongyloxeas (to 2000 × 36) **me:** spherasters (30–60) **mi:** chiasters/oxyasters (10–20), tylasters (10–15), polyrhabds	Pt Jackson, New South Wales, Australia	Unknown[2]
T. magna Kirkpatrick, 1903[1,7]	Oval or spherical; conulated surface (in young specimens), expanding to polygonal plates. **Tree-like rootlets.**	70 mm Purple-brown (in ethanol)	Tracts of stout megasclere bundles (500–1000), unbranched but with terminal fans.	**Ms:** strongyloxeas (to 4805 × 75) **me:** spherasters (120) **mi:** asters (35–45), chiasters (12–17)	Natal, South Africa	62
T. microstella Sarà, 1990[8]	Hemispherical/cushion-like; tubercules vary from conules to flat papillae.	10 mm Brownish/ light yellow	Tracts of megasclere bundles in choanosome are accompanied by styles in cortex.	**Ms:** strongyloxeas (to 1600 × 15) **me:** spherasters (30–80) **mi:** tylasters (4–6) & larger 'other microsters'	Pioneer Bay, Orpheus Is, Queensland, Australia	0–1
T. monstrosa (Burton, 1924)[9]	Spherical; sub-glabrous, a few small buds attached. **Rooting processes well developed.**	14–25 mm Nut brown (in ethanol)	Radial tracts of megasclere bundles spread out into fans at surface.	**Ms:** strongyloxeas **me:** spherasters (48) **mi:** tylasters (20) & oxyasters (32). Many microscleres have abnormalities where the tyles & spines are reduced to knobs or absent leaving just a sphere.	Tasmania	Unknown

Table 2 (continued)**.** Comparison of morphology between species of *Tethya* Lamarck, 1815 from Australia and New Zealand.

Species	**External appearance**	**Body diam. Colour**	**Skeletal arrangement**	**Spicules**	**Type locality**	**Depth** (m)
T. mortoni Bergquist & Kelly-Borges, 1991[2]	Ovoid; low tubercules. Buds on erect filaments.	6–8 mm Maize yellow	Fine tracts of megasclere bundles (122–490) uniform from choanosome to cortex, expand slightly in cortex. Interstitial spicules radial.	**Ms:** anisostrongyloxeas (to1092 × 21) **me:** long-rayed oxyspherasters (26–62), short rayed oxyspherasters (28–52) **mi:** incl. acanthostrongylasters & acanthotylasters (8–13)	Manukau Harbour, Auckland, New Zealand	0.5
T. multistella Lendenfeld, 1888[4,1]	Spherical to subspherical; regularly tuberculose.	20–40 mm Red, flesh or rose	Radial tracts of megasclere bundles (300–500) are trumpet-like in shape and spread to secondary and tertiary branches in tubercules.	**Ms:** strongyloxeas (to 3500 × 18) **me:** spherasters (20–50) **mi:** tylasters (12–16), oxyasters (5–10)	Pt. Jackson, New South Wales Australia	Unknown
T. orphei Sarà, 1990[8]	Subspherical; lengthened but apically flattened tubercules at top of sponge, smooth elsewhere.	5 mm Brown yellowish	[Tracts/bundles not described.]	**Ms:** strongyloxeas, (to 1600 × 15) styles (to 1200 × 10) **me:** spherasters (10–50) **mi:** chiasters & tylasters (8–12), oxyasters (10–30)	Pioneer Bay, Orpheus Is, Queensland, Australia	0–1
T. pellis Bergquist & Kelly-Borges, 1991[2]	Flattened subspherical, tubercules divided by pore grooves; firm but compressible.	40 mm Flesh rose	Tracts of megasclere bundles widely spaced – branch in top third of cortex into fans.	**Ms:** anisostrongyloxeas (to 2075 × 30) **me:** oxyspherasters (25–120), oxyasters (20–25) **mi:** acantho-chiasters/tylasters/ oxyspherasters (13–18), microoxyasters (5–11)	Fairlight, Sydney Harbour, New South Wales, Australia	Intertidal
T. popae Bergquist & Kelly-Borges, 1991[2]	Irregular subspherical; ill-defined tubercules or smooth; 2–8 apical oscules; sponges can cluster connected by stolons.	8–16 mm Bright deep orange	Flexuous tracts of megasclere bundles have elaborate tertiary branching.	**Ms:** anisostrongyloxeas (to 910 × 10) **me:** oxyspherasters (20–55) **mi:** acanthochiasters & acanthostrongylasters (7–13), microoxyasters (2–10)	Cape Banks, New South Wales, Australia	Intertidal
T. pulitzeri Sarà & Sarà, 2004[1]	[No description, species designation from slides only.]	Unknown Unknown	Unknown.	**Ms**: strongyloxeas, anisostrongyles (to 1100 × 15) **me:** oxyspherasters (30–50) **mi:** tylasters & chiasters (8–10), oxyasters (10–15)	Heron Island, Great Barrier Reef, Australia	Unknown
T. robusta (Bowerbank, 1873) [1]	Hemispherical; polygonal areas separated by grooves **Attached by short broad robust basal stolons.**	30–50 mm Yellow-grey	Slim tracts of megasclere bundles expand slightly in cortex.	**Ms:** anisostrongyloxeas (to 1850 × 32) **me:** oxyspheraster (30–90), oxyasters (11–22) **mi:** acantho-tylasters/strongylasters (7–12), microoxyspherasters (4–5)	Australia	0.2–30

Table 2 (continued)**.** Comparison of morphology between species of *Tethya* Lamarck, 1815 from Australia and New Zealand.

Species	**External appearance**	**Body diam. Colour**	**Skeletal arrangement**	**Spicules**	**Type locality**	**Depth** (m)
T. seychellensis (Wright, 1881)[1,2]	Spherical or hemispherical; smooth with flattened polygonal tubercules separated by grooves; firm compressible. **Attached by fine stolons.**	15–25 mm Brown red	Tracts of megasclere bundles (250–500) have secondary branching in cortex – spicules protrude well beyond surface.	**Ms:** anisostrongyloxeas (to 1500 × 30) **me:** oxyspherasters (17–76), oxyasters (32–50) **mi:** acanthochiasters & acanthostrongylasters (7–13), microasters (7–17)	Dampier, Western Australia	Intertidal
T. stellodermis Sarà & Sarà, 2004[1]	[Shape not described]; surface of very flattened tubercules; very hard.	25 mm Pale purple brown (in ethanol)	Thick tracts of megasclere bundles (300–500) branch into secondary and tertiary tracts in cortex.	**Ms:** strongyloxeas (to 1400 × 28) **me:** spherasters/oxyasters (10–40) **mi:** various incl. tylasters (20–25), oxyasters (5–10)	Pt Phillip Heads, Victoria	Unknown
T. stolonifera Bergquist & Kelly-Borges, 1991[2]	Spherical to subspherical; circular or polygonal tubercules; soft compressible. **Extensive stolons.**	3–11 mm Reddish yellow	Narrow well separated tracts of megasclere bundles (200–425) radiate in cortex forming conical brushes.	**Ms:** anisostrongyloxeas (to 1225 × 26) **me:** oxyspherasters (13–76); **mi:** acanthostrongylasters, acanthotylasters (8–14) rare microoxyspherasters (3–10)	Waitemata Harbour, Auckland, New Zealand	0. 5–1.5
T. tasmaniae Sarà & Sarà, 2004[1]	Spherical with small tubercules.	20 mm Unknown	Tracts of megasclere bundles (500–1000) branch in choanosome and end in cortex as wide cones.	**Ms:** strongyloxeas/anisostrongyles (to 1200 × 20) **me:** spherasters 20–50 **mi:** various e.g., tylasters, oxyasters (3 types), chiasters	Blackmans Bay, Tasmania, Australia	Unknown

Sources. [1]Sarà & Sarà (2004); [2]Bergquist & Kelly-Borges (1991); [3]Bergquist (1961); [4]Lendenfeld (1888); [5]Hallmann (1914); [6]Sollas (1888); [7]Kirkpatrick (1903); [8]Sarà (1990); [9]Burton (1924).

Notes. * unknown, or not noted in sources. [a] Maximum depth of Pt Phillip Heads is 50 m (Barton *et al.* 2012); [b] Maximum depth of Pt Jackson is 45 m (Johnston *et al.* 2015). *Tethya diploderma* Schmidt, 1870 is listed as occurring in New Zealand by Kelly *et al.* (2009), but is not included as this is currently shown as inaccurate by van Soest (2019). *Tethya compacta* Bergquist, 1961 is not included in the list of Australia and New Zealand species by Sarà & Sarà (2004) due to its synonymisation by Bergquist with *T. aurantium* (Pallas, 1766), however they note that the taxonomic status of *T. compacta* "remains uncertain"; it is included here as it is shown as accepted by van Soest (2019). The occurance of *T. aurantium* in New Zealand is shown as inaccurate (van Soest 2019) and is not included here.

Results of the phylogenetic analyses

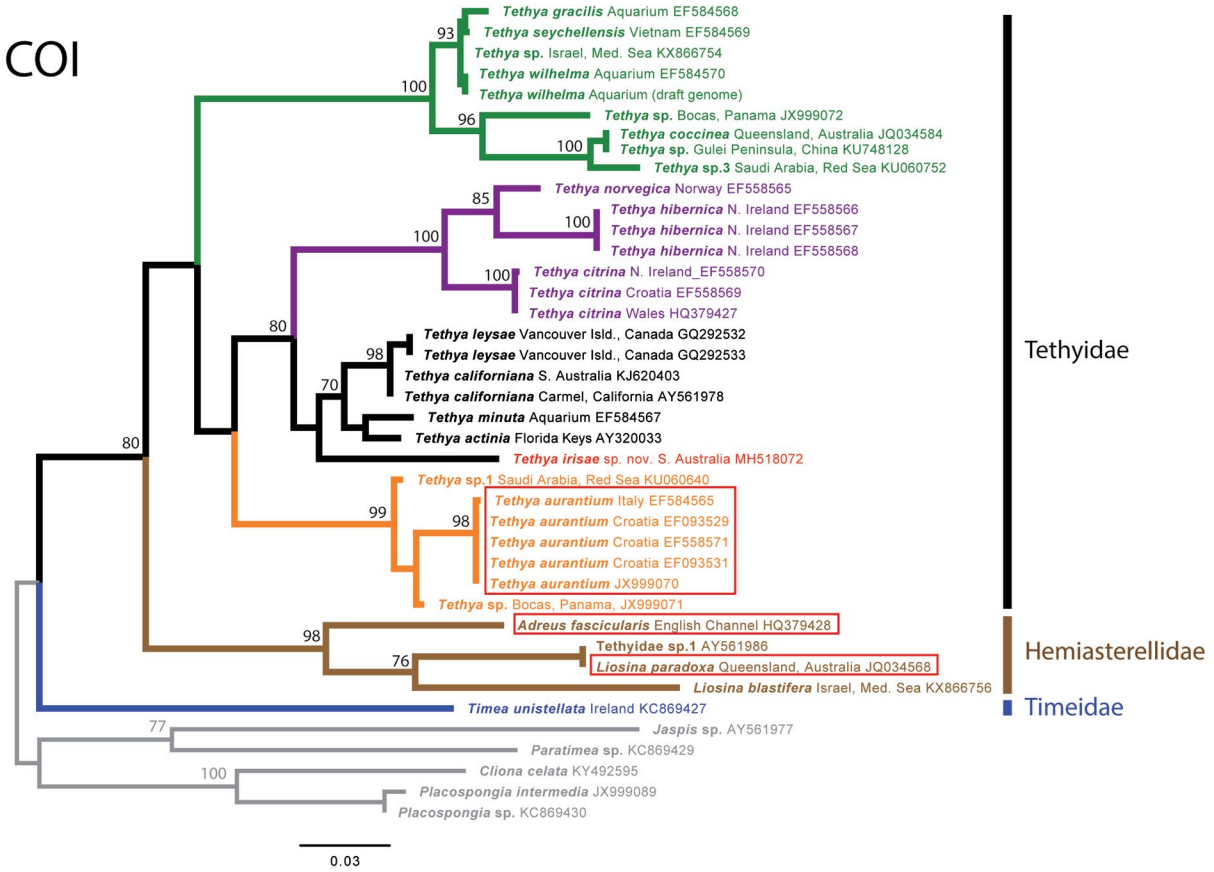
28S and COI trees had similar topologies (Fig. 5). The monophyly of the Tethyida was not supported (28S, bootstrap of 12; COI, bootstrap of 33) with Timeidae sister to a moderately (COI) to poorly supported (28S) Tethyidae + Hemiasterellidae clade. *Timea* sp. from the '3PP cave', La Ciotat, France, (Chombard 1998) did not group with the rest of the Timeidae, but its paraphyletic position was poorly supported. The Hemiasterellidae (*Adreus*, *Axos*, *Liosina*) seemed to group in a moderately to well-supported clade (28S, 69; COI, 98). The 28S tree suggested that the Australian *Laxotethya dampieriensis* (Tethyidae) and a Tethyida sp. from Ireland with no obvious genus assignment (C. Morrow, pers. comm.) had more ambiguous positions: in RaxML analyses they grouped together (poorly supported), while in MrBayes analyses Tethyida sp. branched between the Timeidae and the rest of the Tethyida. *Liosina paradoxa* Thiele, 1899 and *Liosina blastifera* Vacelet, Bitar, Carteron, Zibrowius & Perez, 2007 did not group together in the 28S trees, while *Adreus* was polyphyletic with two clades (one from Australia, the other from the North Atlantic). With COI, where we only have *Tethya* sequences, we could identify four moderately to well-supported clades: 1) the *T. aurantium* clade, 2) the *T. wilhelma* clade, 3) the *T. citrina* clade and 4) the *T. actinia* Laubenfels, 1950 clade. Our new species *T. irisae* sp. nov. branched as the sister taxa to clade 4 (in RaxML and MrBayes) but this is poorly supported. In our 28S dataset where we found the same four clades, all other Tethyidae genera (*Tethytimea* Laubenfels, 1936, *Stellitethya*, *Xenospongia* and *Tectitethya* Sarà, 1994) were mixed with *Tethya* species, especially in clade 4. *T. irisae* sp. nov. grouped with *Tectitethya* in clade 4 (in RaxML and MrBayes analyses) but this is poorly supported. *T. wilhelma* was originally described from a tropical aquarium in Germany but its original geographical location is unknown. In *Tethya* clade 2, we noticed several species were very close genetically to *T. wilhelma*: with COI, *Tethya* sp. from Israel (Mediterranean Sea) had only 1 bp difference with *T. wilhelma*; with 28S, *T. taboga* from Panama had 0–2 bp difference with *T. wilhelma* (uncertainty due to two ambiguous base pairs) while *Tethya* sp. 2 from Saudi Arabia had 1 bp difference with *T. wilhelma*.

Discussion

Tethya irisae sp. nov. is a new and distinctive sponge from the slope of the Great Australian Bight (GAB). This is the 28th *Tethya* species reported from Australia, and the deepest, being found in the bathyal zone at around 1000 m deep. With uncertainties in the validity of genera divisions based on morphological characters e.g., possession of a stalk, a reappraisal of the genera within Tethyidae based on molecular sequencing is needed.

Phylogeny of Tethyidae

The first phylogenetic analyses of *Tethya*, using COI and morphology (Heim *et al.* 2007; Heim & Nickel 2010) revealed four main clades: 1) the *seychellensis-wilhelma* complex, 2 + 3) the *citrinia-actinia* complex divided in two subclades (European species and western Atlantic species + eastern Pacific) and 4) the *aurantium* clade. Our COI and 28S analyses with extended datasets retrieve these four clades, but with a higher biogeographical diversity. The *seychellensis-wilhelma* complex now includes specimens from Israel, Vietnam, Panama, China and Queensland; the *aurantium* clade now includes species from the Mediterranean Sea, the Red Sea and Panama. All clades are well-supported in the COI tree except for clade 3, the western Atlantic/Pacific clade. This is precisely the group joined by the COI sequence of *T. irisae* sp. nov.; its position within this group, however, remains unclear. These same four clades are not as clear in our 28S tree, their inter-relationships are also different, and not supported at all. This may be due to the fact that our 28S alignment is a mix of different 28S domains and different sampling than COI, both of which may influence some of the groupings. The *seychellensis-wilhelma* and *aurantium* clades are well supported with 28S as well. On the other hand, the *citrinia* and *actinia* subclades are unclear, and this is probably due to the addition in this dataset of many different genera of Tethyidae (*Tethytimea*, *Tectitethya*, *Stellitethya*, *Xenospongia* Gray, 1858, *Laxotethya*). As suggested by Sarà *et al*.


COI
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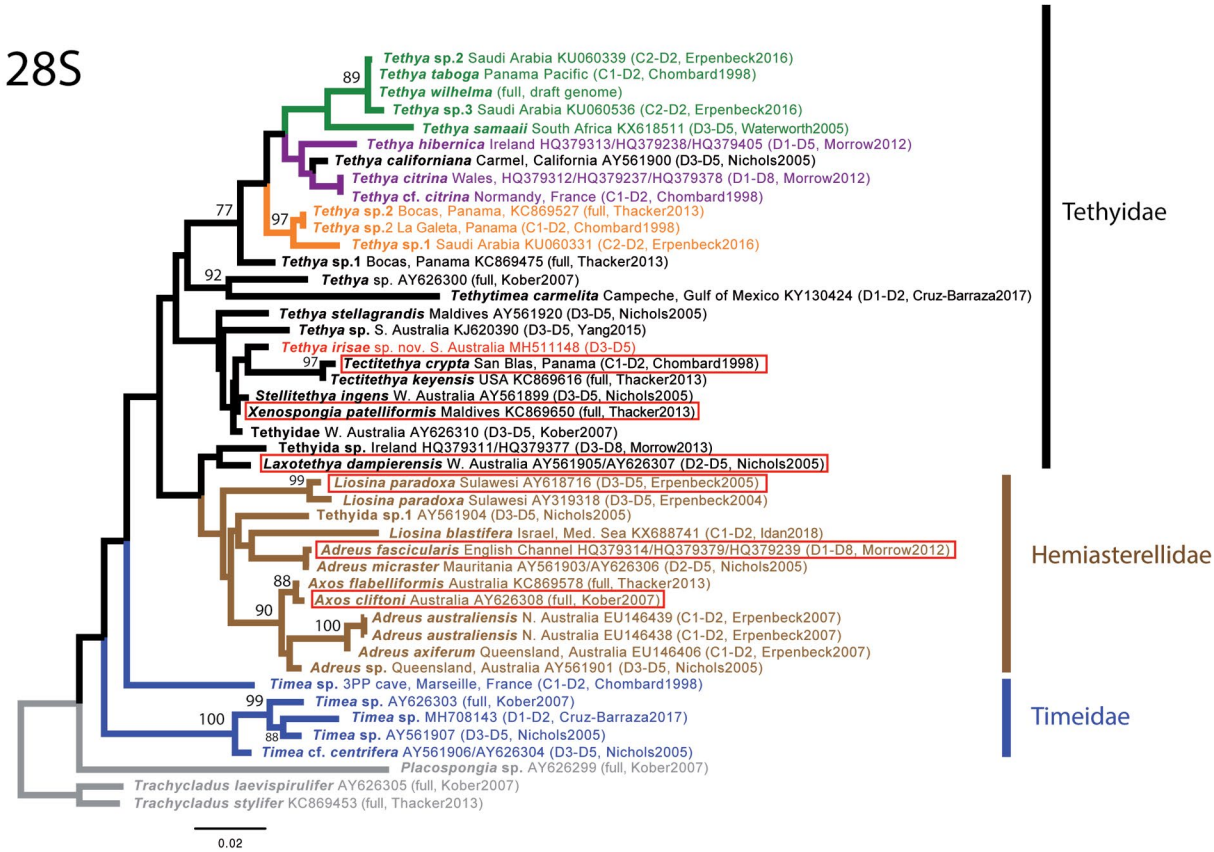
Tethya seychellensis Vietnam EF584569
Tethya sp. Israel, Med. Sea KX866754
Tethya wilhelma Aquarium EF584570
Tethya wilhelma Aquarium (draft genome)
Tethya sp. Bocas, Panama JX999072
Tethya coccinea Queensland, Australia JQ034584
Tethya sp. Gulei Peninsula, China KU748128
Tethya sp.3 Saudi Arabia, Red Sea KU060752
Tethya norvegica Norway EF558565
Tethya hibernica N. Ireland EF558566
Tethya hibernica N. Ireland EF558567
Tethya hibernica N. Ireland EF558568
Tethya citrina N. Ireland_EF558570
Tethya citrina Croatia EF558569
Tethya citrina Wales HQ379427
Tethya leysae Vancouver Isld., Canada GQ292532
Tethya leysae Vancouver Isld., Canada GQ292533
Tethya californiana S. Australia KJ620403
Tethya californiana Carmel, California AY561978
Tethya minuta Aquarium EF584567
Tethya actinia Florida Keys AY320033
Tethya irisae sp. nov. S. Australia MH518072
Tethya sp.1 Saudi Arabia, Red Sea KU060640
Tethya aurantium Italy EF584565
Tethya aurantium Croatia EF093529
Tethya aurantium Croatia EF558571
Tethya aurantium Croatia EF093531
Tethya aurantium JX999070
Tethya sp. Bocas, Panama, JX999071
Adreus fascicularis English Channel HQ379428
Tethyidae sp.1 AY561986
Liosina paradoxa Queensland, Australia JQ034568
Liosina blastifera Israel, Med. Sea KX866756
Timea unistellata Ireland KC869427
Jaspis sp. AY561977
Paratimea sp. KC869429
Cliona celata KY492595
Placospongia intermedia JX999089
Placospongia sp. KC869430
0.03
Tethyidae
Hemiasterellidae
Timeidae


28S
Tethya sp.2 Saudi Arabia KU060339 (C2-D2, Erpenbeck2016)
Tethya taboga Panama Pacific (C1-D2, Chombard1998)
Tethya wilhelma (full, draft genome)
Tethya sp.3 Saudi Arabia KU060536 (C2-D2, Erpenbeck2016)
Tethya samaaii South Africa KX618511 (D3-D5, Waterworth2005)
Tethya hibernica Ireland HQ379313/HQ379238/HQ379405 (D1-D5, Morrow2012)
Tethya californiana Carmel, California AY561900 (D3-D5, Nichols2005)
Tethya citrina Wales, HQ379312/HQ379237/HQ379378 (D1-D8, Morrow2012)
Tethya cf. citrina Normandy, France (C1-D2, Chombard1998)
Tethya sp.2 Bocas, Panama, KC869527 (full, Thacker2013)
Tethya sp.2 La Galeta, Panama (C1-D2, Chombard1998)
Tethya sp.1 Saudi Arabia KU060331 (C2-D2, Erpenbeck2016)
Tethya sp.1 Bocas, Panama KC869475 (full, Thacker2013)
Tethya sp. AY626300 (full, Kober2007)
Tethytimea carmelita Campeche, Gulf of Mexico KY130424 (D1-D2, Cruz-Barraza2017)
Tethya stellagrandis Maldives AY561920 (D3-D5, Nichols2005)
Tethya sp. S. Australia KJ620390 (D3-D5, Yang2015)
Tethya irisae sp. nov. S. Australia MH511148 (D3-D5)
Tectitethya crypta San Blas, Panama (C1-D2, Chombard1998)
Tectitethya keyensis USA KC869616 (full, Thacker2013)
Stellitethya ingens W. Australia AY561899 (D3-D5, Nichols2005)
Xenospongia patelliformis Maldives KC869650 (full, Thacker2013)
Tethyidae W. Australia AY626310 (D3-D5, Kober2007)
Tethyida sp. Ireland HQ379311/HQ379377 (D3-D8, Morrow2013)
Laxotethya dampierensis W. Australia AY561905/AY626307 (D2-D5, Nichols2005)
Liosina paradoxa Sulawesi AY618716 (D3-D5, Erpenbeck2005)
Liosina paradoxa Sulawesi AY319318 (D3-D5, Erpenbeck2004)
Tethyida sp.1 AY561904 (D3-D5, Nichols2005)
Liosina blastifera Israel, Med. Sea KX688741 (C1-D2, Idan2018)
Adreus fascicularis English Channel HQ379314/HQ379379/HQ379239 (D1-D8, Morrow2012)
Adreus micraster Mauritania AY561903/AY626306 (D2-D5, Nichols2005)
Axos flabelliformis Australia KC869578 (full, Thacker2013)
Axos cliftoni Australia AY626308 (full, Kober2007)
Adreus australiensis N. Australia EU146439 (C1-D2, Erpenbeck2007)
Adreus australiensis N. Australia EU146438 (C1-D2, Erpenbeck2007)
Adreus axiferum Queensland, Australia EU146406 (C1-D2, Erpenbeck2007)
Adreus sp. Queensland, Australia AY561901 (D3-D5, Nichols2005)
Timea sp. 3PP cave, Marseille, France (C1-D2, Chombard1998)
Timea sp. AY626303 (full, Kober2007)
Timea sp. MH708143 (D1-D2, Cruz-Barraza2017)
Timea sp. AY561907 (D3-D5, Nichols2005)
Timea cf. centrifera AY561906/AY626304 (D3-D5, Nichols2005)
Placospongia sp. AY626299 (full, Kober2007)
Trachycladus laevispirulifer AY626305 (full, Kober2007)
Trachycladus stylifer KC869453 (full, Thacker2013)
0.02
Tethyidae
Hemiasterellidae
Timeidae

(2001) and Heim *et al.* (2007), *Tethya wilhelma* and *T. gracilis* Sarà, Sarà, Nickel & Brümmer, 2001, both described from aquaria in Germany belong to the *seychellensis-wilhelma* complex. There is only 1 bp difference between the COI of *T. wilhelma* and *Tethya* sp. (Mediterranean Sea, Israel) so this specimen should be re-examined to see if it could be conspecific with *T. wilhelma*. Heim *et al.* (2007) showed that the most reliable characters for *Tethya* taxonomy were morphometric spicule data, but none could actually make good morphological synapomorphies for the two *Tethya* clades supported with COI and 28S. New characters (e.g., chemical compounds, specialized cells, associated microbes) must be explored in order to find independent support for these groups.

External colour may be a reliable character to discriminate those clades, as shown previously in some calcareous sponges (Rossi *et al.* 2011). Indeed, most shallow water *Tethya* species have a yellow, orange to red surface colour, probably due to different carotenoids (Tanaka *et al.* 1982) some of which they can synthesise themselves (Liaaen-Jensen *et al.* 1982) and therefore have a genetic basis. All species currently in the *citrina* subclade (*T. norvegica* Bowerbank, 1872, *T. citrina* and *T. hibernica*) are light-yellow coloured. Species from the *actinia* subclade and *aurantium* clade are usually bright yellow to orange to bright orange, except for the 'aquarium' species *T. minuta* Sarà, Sarà, Nickel & Brümmer, 2001 (white, in artificial conditions at least). Finally, the *seychellensis-wilhelma* clade seems to include especially bright red/carmine surface-coloured species (*T. seychellensis*, *Tethya* sp. from Bocas, *T. coccinea* Bergquist & Kelly-Borges, 1991, *Tethya* sp. 3 from Saudi Arabia, *T. taboga*, *T. samaaii* Ribeiro & Muricy, 2011), except for the *Tethya* sp. from Israel which was more light orange, and except again for the 'aquarium' species (*T. wilhelma* and *T. gracilis*). In red surface-coloured species, the choanosome is usually orange. However, more colours exist: some *Tethya* can be green (e.g., *Tethya brasiliana* Ribeiro & Muricy, 2004), dark blue (e.g., *Tethya cyanea* Ribeiro & Muricy, 2004), or pink (e.g., *Tethya bergquistae*) but none of these species have been sequenced yet. We can probably dismiss the green colour. It is found in species that can also be orange; Laubenfels (1950) suggested the green colour of *T. actinia* in Bermuda was due to symbiotic algae (a specimen may "turn orange" when fixed in alcohol, as the chlorophyll is extracted). More problematic are species with varying colours, from yellow to orange and red (e.g., *Tethya fastigata*). As for the few deep-sea species of *Tethya*, some have lost their colours (e.g., *Tethya irisae* sp. nov.) while others have retained them: e.g., *Tethya levii* Sarà, 1988 from New Caledonia is light orange, and groups in the *actinia* clade, in accordance with our hypothesis (P. Cárdenas, unpublished data). This grouping-by-colour hypothesis should be further tested with the sequencing of new species of *Tethya*. Other genera of Tethyidae included in our dataset have usually irregular massive forms or are disc-shaped (instead of subspherical forms), and all have dark colours: black-brownish for *Tectitethya* spp., beige-gray for *Xenospongia*, and whitish-brown in ethanol for *Laxotethya* and *Stellitethya* (the live colour is unknown). Since all except *Laxotethya* are sister group to a bright orange *Tethya* sp. from South Australia (possibly in the *actinia* clade) (Fig. 5, 28S tree), we suppose the common ancestor of these other genera lost its yellow-orange colours, and so its capacity to produce carotenoids.

Our COI and 28S dataset include type species of four Tethyidae genera (of the 14 valid genera): *Tethya* (*T. aurantium*, COI), *Tectitethya* (*T. crypta*, 28S), *Xenospongia* (*X. patelliformis*, 28S) and *Laxotethya* (*L. dampierensis*, 28S). In addition to that, two other Tethyidae genera are represented in our 28S tree: *Stellitethya* and *Tethytimea*. All these genera are essentially defined by different skeletal structures and therefore body shape; all these genera have an indistinct or ill-defined cortex (vs a distinct thick cortex for *Tethya*) and an irregular massive or encrusting shape (vs (sub)spherical shape in *Tethya*). Our

Fig. 5 (opposite page)**.** Tethyida COI and 28S maximum-likelihood (RaxML) trees. ML bootstrap supports (1000 bootstrap replicates) > 70 are indicated. After the species name, locality of the specimen is given (when known), followed by the GenBank accession number(s). For 28S, we also indicated the 28S region that was sequenced as well as the first author + date of the publication where the sequence first appeared. Type species of genera are in red boxes while *Tethya irisae* sp. nov. appears in red.

28S tree suggests that *Xenospongia*, *Stellitethya*, *Tectitethya* and *Tethytimea* are grouping with *Tethya* (Fig. 5), while *Laxotethya* groups with Hemiasterellidae, albeit with no support. *Tethytimea carmelita*, *Tectitethya* and *Stellitethya/Xenospongia* evolved independently within *Tethya* thus suggesting that the loss of a distinct cortex and of the subspherical shape happened several times. More sequences from Australian *Tethya* are needed to understand the origin and relationships of these other Tethyidae genera. One clade that is moderately supported (bootstrap of 69) is the sister-group relationship of *Xenospongia* and *Stellitethya*, with a 5–6 bp difference in 28S (D3–D5). Both genera have a poorly defined cortex but different shapes: discoid for *Xenospongia*, massive irregular for *Stellitethya*. These two genera also share megasters reaching large sizes (> 150 µm), as in *T. irisae* sp. nov., grouping nearby (bootstrap of 62) with *Tectitethya* (which does not have very large megasters).

Phylogeny of Tethyida

The Tethyida also include Hemiasterellidae and Timeidae. Since the ex-hemiasterellid genera *Stelligera* Gray, 1867 and *Paratimea* have been reallocated to the Stelligeridae Lendenfeld, 1898, order Axinellida (Morrow *et al.* 2012), the Hemiasterellidae include four genera: *Adreus*, *Axos*, *Hemiasterella* and the monospecific *Leptosastra* Topsent, 1904. The sister position of Hemiasterellidae *Adreus* and *Axos* with the Tethyidae has been repeatedly shown by previous COI (Morrow *et al.* 2012, 2013), 28S (Thacker *et al*. 2013; Cruz-Barraza *et al.* 2017) and 18S analyses (Redmond *et al.* 2013). *Liosina*, a genus with only four species and a loose arrangement of oxeas/styles, had been tentatively assigned to the family Dictyonellidae, order Bubarida (van Soest *et al.* 2002). The grouping of *Liosina paradoxa*, type species, with the Hemiasterellidae was revealed for the first time by Morrow *et al.* (2012) with 28S. Our study confirms this grouping for the first time with COI sequences (*L. paradoxa* and *Liosina blastifera*), as well as a 28S sequence of *L. blastifera*. Its unambiguous grouping with the Hemiasterellidae suggests that species of *Liosina* are in fact Hemiasterellidae that have secondarily lost their euasters. Furthermore, *Liosina* often have a polygonal surface pattern, and/or pores in shallow grooves, a character present in most Tethyidae and Timeidae, which often gives rise to the characteristic surface tubercles. Since we have sequenced the type species of *Liosina*, we formally propose the reallocation of *Liosina* from Dictyonellidae to Hemiasterellidae. However, it is unclear in our trees whether *Liosina* is polyphyletic. The small polygons from *Liosina*, also called tubercles or microconules, are also present in the three *Adreus* species from Australia (*A. australiensis* (Ridley, 1884), *A. axiferum* (Hentschel, 1912) and *Adreus* sp.) thereby confirming their reallocation. These two branching species also share with the Tethyida the typical radial bundles of megascleres fanning out closer to the surface. *Adreus australiensis* and *A. axiferum* were previously grouped in Raspailidae (Hooper 1991) although they lacked echinating megascleres, before Erpenbeck *et al.* (2007) showed with 28S that they clustered instead with Hemiasterellidae and Tethyidae. These two species also secondarily lost their asters, while the vase-shaped *Adreus* sp. from Queensland (QM G315767) still has tylasters. This second case of aster loss in the Tethyida suggests that, similarly as in the Astrophorina (Cárdenas *et al.* 2011), more genera or species without asters, can be expected to be reallocated to the Tethyida once they are sequenced. Since this *Adreus* clade does not cluster with the clade of *Adreus fascicularis* (type species of the genus), they potentially represent a new genus in the Hemiasterellidae. To conclude, the Hemiasterellidae now include *Adreus*, *Axos*, *Hemiasterella*, *Leptosastra*, *Liosina*, and a potential new genus. So far, all GenBank *Hemiasterella* sp. sequences are doubtful and failed to cluster with the Hemiasterellidae (cf. Material and methods). The type specimen of *Hemiasterella typus* Carter, 1879, has not been revised and sequenced so that the phylogenetic position of *Hemiasterella* remains to be tested. We note, however, that *H. typus* does not share with most of the Tethyida 1) a surface with pores in grooves around tubercles/plates or 2) bundles of megascleres fanning out at the surface.

To sum up the main findings of the phylogenetic analysis.

1. Four *Tethya* clades were retrieved (as in previous analyses) for which no synapomorphies are currently known; we, however, discuss the possibility of using external colour to support some of these clades.
2. Despite unclear phylogenetic relationships amongst Tethyidae from Australia, our results suggest that Tethyidae genera *Tethytimea*, *Tectitethya*, *Laxotethya*, *Stellitethya*, and *Xenospongia* derive from species of *Tethya*, which may challenge their validity in the future.
3. Our results suggest that Hemiasterellidae is the sister-group of Tethyidae while the position of Timeidae is still ambiguous (not supported).
4. We show that asters have been secondarily lost at least twice in the Hemiasterellidae: in *Liosina* and a potential new genus from northern Australia. We formally propose the reallocation of *Liosina* from Dictyonellidae to Hemiasterellidae.

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References

Adlard R.D. & Lester R.J.G. 1995. Development of a diagnostic test for *Mikrocytos roughleyi*, the aetiological agent of Australian winter mortality of the commercial rock oyster, *Saccostrea commercialis* (Iredale & Roughley). *Journal of Fish Diseases* 18: 609–614.
https://doi.org/10.1111/j.1365-2761.1995.tb00365.x

Barton J., Pope A. & Howe S. 2012. *Marine Natural Values Study Vol 2: Marine Protected Areas of the Victorian Embayments Bioregion, Part 1 Port Phillip Bay*. Parks Victoria Technical Series No. 77. Parks Victoria, Melbourne.

Bergquist P.R. 1961. Demospongiae (Porifera) of the Chatham Islands and Chatham Rise, collected by the Chatham Islands 1954 Expedition. *New Zealand Oceanographic Institute Memoir* 13 (139): 169–206.

Bergquist P.R. & Kelly-Borges M. 1991. An evaluation of the genus *Tethya* (Porifera: Demospongiae: Hadromerida) with descriptions of new species from the southwest Pacific. *The Beagle. Records of the Northern Territory Museum of Arts and Sciences* 8: 37–72.

Borchiellini C., Alivon E. & Vacelet J. 2004. The systematic position of *Alectona* (Porifera, Demospongiae): a Tetractinellid sponge. *Bollettino dei musei e degli istituti biologici dell'Universita di Genova* 68: 209–217.

Boury-Esnault N., Marschal C., Kornprobst J.M. & Barnathan G. 2002. A new species of *Axinyssa* Lendenfeld, 1897 (Porifera, Demospongiae, Halichondrida) from the Senegalese coast. *Zootaxa* 117: 1–8. https://doi.org/10.11646/zootaxa.117.1.1

Burton M. 1924. A revision of the sponge family Donatiidae. *Proceedings of the Zoological Society of London* 1924(4): 1033–1045. https://doi.org/10.1111/j.1096-3642.1924.tb03326.x

Cárdenas P., Xavier J.R., Reveillaud J., Schander C. & Rapp H.T. 2011. Molecular phylogeny of the Astrophorida (Porifera, Demospongiae) reveals an unexpected high level of spicule homoplasy. *PLoS ONE* 6: e18318. https://doi.org/10.1371/journal.pone.0018318

Chombard C. 1998. *Les Demospongiae à asters: phylogénie moléculaire et homologie morphologique.* Ph.D thesis, *Muséum national d'Histoire naturelle, Paris.*

Cruz-Barraza J.A., Vega C., Ávila E., Vázquez-Maldonado L.E. 2017. Integrative taxonomy reveals the first record and a new species for the previously monotypic genus *Tethytimea* (Tethyida: Tethyidae) in the Gulf of Mexico. *Zootaxa* 4226: 113–125. https://doi.org/10.11646/zootaxa.4226.1.6

Currie D. & Sorokin S. 2011. *A Preliminary Assessment of the Deepwater Benthic Communities of the Great Australian Bight Marine Park*. Report to the South Australian Department of Environment and Natural Resources and the Commonwealth Department of Sustainability, Environment, Water, Populations and Communities. Vol. No. F2011/000526-1. SARDI Aquatic Sciences Publication, Adelaide. Available from: http://www.pir.sa.gov.au/__data/assets/pdf_file/0018/232092/A_preliminary_assessment_of_the_deepwater_benthic_communities_of_the_Great_Australian_Bight_Marine_Park.pdf [accessed 7 Sep. 2018].

Erpenbeck D., List-Armitage S., Alvarez B., Degnan B.M., Wörheide G. & Hooper J.N.A. 2007. The systematics of Raspailiidae (Demospongiae: Poecilosclerida: Microcionina) re-analysed with a ribosomal marker. *Journal of the Marine Biological Association of the United Kingdom* 87: 1571–1576. https://doi.org/10.1017/S0025315407058201

Erpenbeck D., Hall K.B., Büttner G., Sacher K., Schätzle S., Schuster A., Vargas S., Hooper J.N.H. & Wörheide G. 2012. The phylogeny of halichondrid demosponges: Past and present re-visited with DNA-barcoding data. *Organisms Diversity and Evolution* 12: 57–70. https://doi.org/10.1007/s13127-011-0068-9

Folmer O., Black M., Hoeh W., Lutz R. & Vrijenhoek R. 1994. DNA primers for amplification of mitochondrial cytochrome c oxidase subunit I from diverse metazoan invertebrates. *Molecular Marine Biology and Biotechnology* 3: 294–299.

Hallmann E.F. 1914. A revision of the monaxonid species described as new in Lendenfeld's 'Catalogue of the Sponges in the Australian Museum'. Part I. *Proceedings of the Linnean Society of New South Wales* 39: 263–315.

Heim I., Nickel M. & Brümmer F. 2007. Phylogeny of the genus *Tethya* (Tethyidae: Hadromerida: Porifera): molecular and morphological aspects. *Journal of the Marine Biological Association of the United Kingdom* 87: 1615–1627. https://doi.org/10.1017/S0025315407058419

Heim I. & Nickel M. 2010. Description and molecular phylogeny of *Tethya leysae* sp. nov. (Porifera, Demospongiae, Hadromerida) from the Canadian Northeast Pacific with remarks on the use of microtomography in sponge taxonomy. *Zootaxa* 2422: 1–21. https://doi.org/10.11646/zootaxa.2422.1.1

Hooper J. 1991. Revision of the family Raspailiidae (Porifera: Demospongiae), with description of Australian species. *Invertebrate Systematics* 5: 1179–1481.

Hooper J. 2012. *Porifera*. Australian Faunal Directory. Australian Biological Resources Study, Canberra. Available from https://biodiversity.org.au/afd/taxa/PORIFERA [accessed 4 Apr. 2018].

Johnston E.L., Mayer-Pinto M., Hutchings P.A., Marzinelli M., Ahyong S.T., Birch G., Booth D.J., Creese R.G., Doblin M.A., Figueira W., Gribben P.E., Pritchard T., Roughan M., Steinberg P.D. & Hedge L.H. 2015. Sydney Harbour: what we do and do not know about a highly diverse estuary. *Marine and Freshwater Research* 66: 1073–1087. https://doi.org/10.1071/MF15159

Katoh K., Misawa K., Kuma K. & Miyata T. 2002. MAFFT: a novel method for rapid multiple sequence alignment based on fast Fourier transform. *Nucleic Acids Research* 30: 3059–3066. https://doi.org/10.1093/nar/gkf436

Kelly M., Edwards A.R., Wilkinson M.R., Alvarez B., Cook S. de C., Bergquist P.R., Buckeridge St J., Campbell H.J., Reiswig H.M., Valentine C., Vacelet J. 2009. Phylum Porifera: sponges. *In*: Gordon D.P. (ed.) *New Zealand inventory of biodiversity: 1. Kingdom Animalia: Radiata*, *Lophotrochozoa*, *Deuterostomia*: 23–46. Canterbury University Press.

Kirkpatrick R. 1903. Descriptions of South African Sponges. Part III. *Marine Investigations in South Africa* 2 (14): 233–264.

Kober K.M. & Nichols S.A. 2007. On the phylogenetic relationships of hadromerid and poecilosclerid sponges. *Journal of the Marine Biological Association of the United Kingdom* 87: 1585–1598. https://doi.org/10.1017/S0025315407058237

Larsson A. 2014. AliView: a fast and lightweight alignment viewer and editor for large datasets. *Bioinformatics* 30: 3276–3278. https://doi.org/10.1093/bioinformatics/btu531

Laubenfels M.W. de. 1950. The Porifera of the Bermuda Archipelago. *Transactions of the Zoological Society of London* 27: 1–154. https://doi.org/10.1111/j.1096-3642.1950.tb00227.x

Lendenfeld R. von. 1888. *Descriptive Catalogue of the Sponges in the Australian Museum*, *Sidney*. Taylor & Francis, London.

Liaaen-Jensen S., Renstrøm B., Ramdahl T., Hallenstvet M. & Bergquist P. 1982. Carotenoids of marine sponges. *Biochemical Systematics and Ecology* 10: 167–174.

Miller M.A., Pfeiffer W. & Schwartz T. 2010. Creating the CIPRES Science Gateway for inference of large phylogenetic trees. *In*: *Proceedings of the Gateway Computing Environments Workshop (GCE)*: 1–8. https://doi.org/10.1109/GCE.2010.5676129

MNF. 2015. *RV* Investigator *Voyage Summary: IN2015_C01 – GAB Deep Water Geological and Benthic Ecology Program*. Marine National Facility, Australia.

Morrow C.C., Picton B.E., Erpenbeck D., Boury-Esnault N., Maggs C.A., Allcock A.L. 2012. Congruence between nuclear and mitochondrial genes in Demospongiae: A new hypothesis for relationships within the G4 clade (Porifera: Demospongiae). *Molecular Phylogenetics and Evolution* 62: 174–190. https://doi.org/10.1016/j.ympev.2011.09.016

Morrow C.C., Redmond N.E., Picton B.E., Thacker R.W., Collins A.G., Maggs C.A., Sigwart J.D. & Allcock A.L. 2013. Molecular phylogenies support homoplasy of multiple morphological characters used in the taxonomy of Heteroscleromorpha (Porifera: Demospongiae). *Integrative and Comparative Biology* 53: 428–446. https://doi.org/10.1093/icb/ict065

Morrow C. & Cárdenas P. 2015. Proposal for a revised classification of the Demospongiae (Porifera). *Frontiers in Zoology* 12: 7. https://doi.org/10.1186/s12983-015-0099-8

Morrow C., Cárdenas P., Boury-Esnault N., Picton B., McCormack G., van Soest R., Redmond N., Collins A., Maggs C., Sigwart J. & Allcock L.A. 2019. Integrating morphological and molecular taxonomy with the revised concept of Stelligeridae (Porifera: Demospongiae). *Zoological Journal of the Linnean Society* zlz017. https://doi.org/10.1093/zoolinnean/zlz017

Nichols S.A. 2005. An evaluation of support for order-level monophyly and interrelationships within the class Demospongiae using partial data from the large subunit rDNA and cytochrome oxidase subunit I. *Molecular Phylogenetics and Evolution* 34: 81–96. https://doi.org/10.1016/j.ympev.2004.08.019

Redmond N.E., Morrow C.C., Thacker R.W., Diaz M.C., Boury-Esnault N., Cárdenas P., Hajdu E., Lobo-Hajdu G., Picton B.E., Pomponi S.A., Kayal E. & Collins A.G. 2013. Phylogeny and systematics of Demospongiae in light of new small-subunit ribosomal DNA (18S) sequences. *Integrative and Comparative Biology* 53: 388–415. https://doi.org/10.1093/icb/ict078

Rees A.J.J., Yearsley G.K., Gowlett-Holmes K. & Pogonoski J. 1999 onwards. *Codes for Australian Aquatic Biota (on-line version)*. CSIRO Marine and Atmospheric Research.

Ronquist F., Teslenko M., van der Mark P., Ayres D.L., Darling A., Höhna S., Larget B., Liu L., Suchard M.A. & Huelsenbeck J.P. 2012. MrBayes 3.2: Efficient Bayesian phylogenetic inference and model choice across a large model space. *Systematic Biology* 61: 539–542. https://doi.org/10.1093/sysbio/sys029

Rossi A.L., Russo C.A.M., Solé-Cava A.M., Rapp H.T. & Klautau M. 2011. Phylogenetic signal in the evolution of body colour and spicule skeleton in calcareous sponges. *Zoological Journal of the Linnean Society* 163: 1026–1034. https://doi.org/10.1111/j.1096-3642.2011.00739.x

Sarà M. 1990. Australian *Tethya* (Porifera, Demospongiae) from the Great Barrier Reef with description of two new species. *Bolletino di Zoologia* 57: 153–157. https://doi.org/10.1080/11250009009355691

Sarà M. 1994. A rearrangement of the family Tethyidae (Porifera Hadromerida) with establishment of new genera and description of two new species. *Zoological Journal of the Linnean Society* 110: 355–371. https://doi.org/10.1111/j.1096-3642.1994.tb01479.x

Sarà M. 2002. Family Tethyidae Gray, 1848. *In*: Hooper J.N.A. & van Soest R.W.M. (eds) *Systema Porifera. A Guide to the Classification of Sponges*: 245–267. Kluwer Academic/Plenum Publishers, New York.

Sarà M. & Burlando B. 1994. Phylogenetic reconstruction and evolutionary hypotheses in the family Tethyidae (Demospongiae). *In*: van Soest R.W.M., Van Kempen T.M.G. & Braeckman J.C. (eds) *Sponges in Time and Space*: 111–116. Balkema, Rotterdam.

Sarà M. & Sarà A. 2004. A revision of Australian and New Zealand *Tethya* (Porifera: Demospongiae) with a preliminary analysis of species-groupings. *Invertebrate Systematics* 18: 117–156. https://doi.org/10.1071/IS03008

Sarà M., Sarà A., Nickel M. & Brümmer F. 2001. Three new species of *Tethya* (Porifera: Demospongiae) from German aquaria. *Stuttgarter Beiträge zur Naturkunde A (Biologie)* 631: 1–15.

Schuster A., Lopez J.V., Becking L.E., Kelly M., Pomponi S.A., Wörheide G., Erpenbeck D. & Cárdenas P. 2017. Evolution of group I introns in Porifera: new evidence for intron mobility and implications for DNA barcoding. *BMC Evolutionary Biology* 17: 82. https://doi.org/10.1186/s12862-017-0928-9

Sollas W.J. 1888. Report on the Tetractinellida collected by *H.M.S. Challenger*, during the years 1873–1876. *Report on the Scientific Results of the Voyage of H.M.S. Challenger during the years 1873–76. Zoology* 25 (part 63): 1–458. https://doi.org/10.5962/bhl.title.6513

Stamatakis A. 2014. RAxML version 8: A tool for phylogenetic analysis and post-analysis of large phylogenies. *Bioinformatics* 30: 1312–1313. https://doi.org/10.1093/bioinformatics/btu033

Szitenberg A., Becking L.E., Vargas S., Fernandez J.C.C., Santodomingo N., Wörheide G., Ilan M., Kelly M. & Huchon D. 2013. Phylogeny of Tetillidae (Porifera, Demospongiae, Spirophorida) based on three molecular markers. *Molecular Phylogenetics and Evolution* 67: 509–519. https://doi.org/10.1016/j.ympev.2013.02.018

Tanaka Y., Yamamoto A., Katayama T. 1982. Two natural 7-cis aromatic carotenoids from sea sponge *Tethya amamensis*. *Nippon Suisan Gakkaishi* 48: 1651–1655.

Taylor M.W., Schupp P.J., Dahllöf I., Kjelleberg S. & Steinberg P.D. 2004. Host specificity in marine sponge-associated bacteria, and potential implications for marine microbial diversity. *Environmental Microbiology* 6: 121–130. https://doi.org/10.1046/j.1462-2920.2003.00545.x

Thacker R.W., Hill A.L., Hill M.S., Redmond E., Collins A.G., Morrow C.C., Spicer L., Carmack C.A, Zappe M.E., Pohlmann D., Hall C., Diaz M.C. & Bangalore P.V. 2013. Nearly complete 28S rRNA gene sequences confirm new hypotheses of sponge evolution. *Integrative and Comparative Biology* 53: 373–387. https://doi.org/10.1093/icb/ict071

van Soest R.W.M., Erpenbeck D. & Alvarez B. 2002. Family Dictyonellidae van Soest, Diaz & Pomponi, 1990. *In*: Hooper J.N.A.& van Soest R.W.M. (eds) *Systema Porifera. A Guide to the Classification of Sponges*: 773–786. Kluwer Academic/Plenum Publishers, New York.

van Soest R.W.M., Boury-Esnault N., Hooper J.N.A., Rützler K., de Voogd N.J., Alvarez, B., Hajdu E., Pisera A.B., Manconi R., Schönberg C., Klautau M., Picton B., Kelly M., Vacelet J., Dohrmann M., Díaz M.-C., Cárdenas P., Carballo J.L., Ríos P. & Downey R. 2019. *World Porifera Database. Tethyidae Gray, 1848*. Available from http://www.marinespecies.org/porifera/porifera.php?p=taxdetails&id=131677 [accessed 28 Mar. 2019].

Williams A., Althaus F., MacIntosh H., Loo M., Gowlett-Holmes K., Tanner J.E., Sorokin S.J. & Green M. 2018. Characterising the invertebrate megafaunal assemblages of a deep-sea (200–3000 m) frontier region for oil and gas exploration: the Great Australian Bight, Australia. *Deep Sea Research* 157–158: 78–91. https://doi.org/10.1016/j.dsr2.2018.07.015

Yang Q., Franco C.M.M., Sorokin S.J. & Zhang W. 2017. Development of a multilocus-based approach for sponge (phylum Porifera) identification: refinement and limitations. *Scientific Reports* 7: 41422. https://doi.org/10.1038/srep41422

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