



A new genus and species of abyssal sponge commonly encrusting polymetallic nodules in the Clarion-Clipperton Zone, East Pacific Ocean

Swee-Cheng Lim, Helena Wiklund, Adrian G. Glover, Thomas G. Dahlgren & Koh-Siang Tan

To cite this article: Swee-Cheng Lim, Helena Wiklund, Adrian G. Glover, Thomas G. Dahlgren & Koh-Siang Tan (2017) A new genus and species of abyssal sponge commonly encrusting polymetallic nodules in the Clarion-Clipperton Zone, East Pacific Ocean, *Systematics and Biodiversity*, 15:6, 507-519, DOI: [10.1080/14772000.2017.1358218](https://doi.org/10.1080/14772000.2017.1358218)

To link to this article: <http://dx.doi.org/10.1080/14772000.2017.1358218>



Published online: 24 Sep 2017.



Submit your article to this journal [↗](#)



Article views: 1338



View related articles [↗](#)



View Crossmark data [↗](#)



Citing articles: 1 View citing articles [↗](#)

Research Article



A new genus and species of abyssal sponge commonly encrusting polymetallic nodules in the Clarion-Clipperton Zone, East Pacific Ocean

SWEE-CHENG LIM¹, HELENA WIKLUND², ADRIAN G. GLOVER², THOMAS G. DAHLGREN^{3,4} & KOH-SIANG TAN¹

¹Keppel-NUS Corporate Laboratory, Tropical Marine Science Institute, National University of Singapore, 18 Kent Ridge Road, Singapore 119227

²Life Sciences Department, The Natural History Museum, Cromwell Road, London SW7 5BD, UK

³Uni Research, Thormøhlensgate 49B, Bergen, Norway

⁴Gothenburg Global Biodiversity Centre, Department of Marine Sciences, University of Gothenburg, Box 463, 40530 Gothenburg, Sweden

(Received 19 January 2017; accepted 4 July 2017; published online 25 September 2017)

The Clarion-Clipperton Zone (CCZ) in the East Pacific is a vast region targeted for deep-sea mineral exploration, for which there are almost no published taxonomic data. Here we describe *Plenaster craigi* gen. nov. sp. nov. from depths of ~4000 m in the eastern CCZ polymetallic nodule province. Despite over 40 years of intense exploration in the area, we reveal that *P. craigi* sp. nov. is the most abundant sponge and the most common metazoan encrusting on nodules in our study area at the eastern CCZ. It has a mean abundance of 15.3 ± 8.9 individuals per m² across 11 stations in a 30 × 30 km study site nested within the Singapore exploration area. The white encrusting sponge is filled with spherocyasters with occasional styles protruding the surface. *Plenaster craigi* sp. nov. is morphologically similar to genera from three different families in two orders: *Timea* (Timeidae; Tethyida); *Hemiasterella* and *Leptosastra* (Hemiasterellidae; Tethyida); and *Paratimea* (Stelligeridae; Axinellida). However, based on the molecular (COI and 28S) phylogenetic trees generated in this study, *P. craigi* sp. nov. was located in the Order Axinellida and appeared to be distant to *Timea*, *Hemiasterella*, *Leptosastra*, and *Paratimea*. We propose a new genus for our material to be placed provisionally in the family Stelligeridae, as it is the only family in the order Axinellida whose members possess euasters. This provisional placement may change when sequences of the type specimens of these genera and advanced phylogenetic reconstruction methods become available in the future. However, we have shown clearly that *Plenaster* gen. nov. is unique and distinct from all currently known taxa. *Plenaster craigi* sp. nov. being an abundant metazoan encrusting on nodule and easily identified filter-feeding animal is a potentially indicator species for future mining impacts in the eastern CCZ, and possibly across the entire CCZ.

LSID: urn:lsid:zoobank.org:act:99D2DDDBA-4643-4752-8C8A-AFF314E6B6E3

Key words: 28S, abyss, deep-sea mining, manganese nodule, new species, *Plenaster* gen. nov., Porifera

Introduction

The Clarion-Clipperton Zone (CCZ) in the Eastern Pacific Ocean (Fig. 1) has vast quantities of polymetallic nodules (as nodules hereafter) on the seafloor at abyssal depths of 4000–6000 m. The International Seabed Authority (ISA) has designated the area, approximately between longitudes 115°–160° W and latitudes 5°–20°N, for nodule exploration and eventual exploitation. These nodules are

rock concretions containing manganese, iron, copper, nickel, cobalt, zinc, silver, and numerous other metals, which can be over 20 cm in size but are usually 5–10 cm in diameter (see Glasby, Li, & Sun 2015; Thiel, Angel, Foell, Rice, & Schriever, 1998). Nodules at the CCZ often exhibit a smooth upper surface resulting from hydroge- netic growth (Halbach, Friedrich, & von Stackelberg, 1988; Thiel et al., 1998). They lie on the seafloor, generally half buried, providing a small hard surface for growth of sessile organisms such as foraminifera, bryozoans, cni- darians, polychaetes, and sponges (see Gooday, Goineau,

Correspondence to: Swee-Cheng Lim. E-mail: sponging@gmail.com

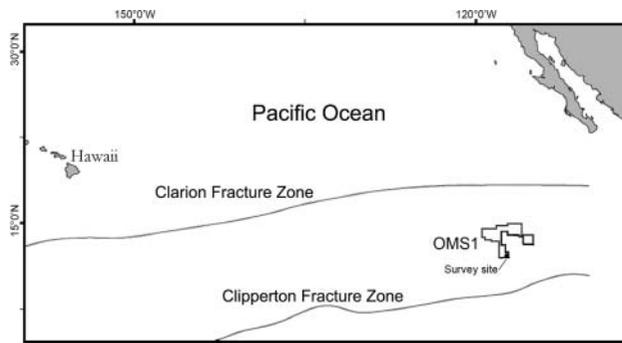


Fig. 1. Location of Ocean Mineral Singapore exploration area and the ABYSSLINE cruise AB02 survey site (30 × 30 km) in the Clarion-Clipperton Zone (East Pacific Ocean).

& Voltski, 2015; Kamenskaya, Gooday, Tendal, & Melnik, 2015; Mullineaux, 1987; Tilot, 2006; Veillette *et al.*, 2007; Wendt, 1974). The environment at the abyssal depth is characterized by perpetual darkness, low temperature of less than 4°C, immense hydrostatic pressure and low nutrient levels, supporting a highly diverse but numerically impoverished marine fauna (Bruun, 1956; Frankenberg & Menzies, 1968; Glover, Dahlgren, Wiklund, Mohrbeck, & Smith, 2016a; Hessler & Jumars, 1974).

There are over 500 species of sponges reported from depths exceeding 500 m worldwide (Glover, Higgs, & Horton, 2017); this is approximately 5% of the total sponge species described. Historically, the abyssal depths of the North Pacific Ocean have only been sporadically sampled for sponges due to high costs and technical difficulties. As a result, very little is known about the sponge fauna throughout the abyssal North Pacific Ocean, and even less is known of the sponges of the Clarion-Clipperton Zone (see Downey & Janussen, 2015; Koltun, 1970; Lendenfeld, 1915; Lévi, 1964; Ridley & Dendy, 1887; Reiswig & Stone, 2013; Schulze, 1887; Vacelet, 2006). Recent efforts to establish biodiversity baselines in the CCZ (see Amon *et al.*, 2016; Dahlgren *et al.*, 2016; Glasby, 2000; Glover *et al.*, 2016a, 2016b; ISA, 1999, 2015; Vanreusel, Hilario, Ribeiro, Menot, & Arbizu, 2016) by countries or commercial entities, with the ultimate objective of assessing the commercial feasibility of collecting nodules, have given us a better picture of the abyssal environment and fauna. However, detailed sponge records are lacking, despite more than 100 scientific cruises to the central east Pacific Ocean in the last 40 years (ISA, 2015; ISA, pers. comm., 2015; Nimmo, Morgan, & Banning, 2013).

The majority of the sponge records are from photographic assessments of the seafloor (see Amon *et al.*, 2016; Bluhm, 1994; Foell & Pawson, 1986; Owen, Sanders, & Hessler, 1967; Tilot, 2006; Vanreusel *et al.*, 2016) with the purpose of documenting nodule density and the associated fauna. Such sponge records only provide a

cursory view of the diversity of sponges present. Small sponges less than 5 cm wide were often ignored, as they would appear as indeterminate spots on imaging devices.

In 2013, on a research cruise to the eastern CCZ UK-1 exploration area, a very small but highly abundant white, nodule-dwelling sponge was photographed and recorded during careful processing of a large number of nodules from samples collected as part of the project ‘ABYSSLINE’. During the second ABYSSLINE cruise in 2015, additional sites were visited including the Ocean Mineral Singapore (OMS) exploration area and a large number of box core samples were collected and examined. To the best of our knowledge, this abundant new sponge species has never been documented from previous CCZ cruises. Here we describe a common and abundant sponge encrusting on nodules in the eastern CCZ. Morphological and molecular data support the erection of a new genus *Plenaster* gen. nov. for this sponge. We believe the sponge to be of significant ecological importance in understanding the potential impacts of future mineral extraction in the eastern CCZ.

Materials and methods

Material was collected during the ABYSSLINE 02 Cruise at a 30 × 30 km study area in the OMS exploration area in the East Pacific Ocean (Fig. 1) between 12 Feb–25 Mar 2015 on R/V *Thomas G. Thompson*. An Ocean Instruments USNEL spade box core, with core size of 0.25 m² (50 cm × 50 cm area) was used to sample the sea floor at 11 stations at the following coordinates and depths: S01, 12 07.06233N, 117 20.62027W, 4183 m; S02, 12 04.90838N, 117 10.69245W, 4072 m; S03, 12 10.86618N, 117 15.65712W, 4115 m; S04, 12 00.56092N, 117 10.69237W, 4148 m; S05, 12 13.03998N, 117 19.52705W, 4093 m; S06, 12 08.69268N, 117 19.52758W, 4041 m; S07, 12 08.15358N, 117 12.90369W, 4082 m; 4172 m; S09, 12 05.98863N, 117 11.79682W, 4051 m; S10, 12 03.27587N, 117 15.10293W, 4100 m; S11, 12 00.559N, 117 22.819W, 4148 m; S12, 12 01.64111N, 117 19.51136W, 4127 m. All nodules brought up by the box core were examined carefully for encrusting organisms following the protocol described by Glover *et al.* (2016a). All visible organisms encrusting on the nodules were documented and sampled for their abundance. Sponge specimens were photographed on nodules kept under cold filtered seawater, carefully removed from the surface of the nodules with a scalpel and photographed again through a stereomicroscope. Sponge specimens were preserved in 80% molecular-grade ethanol and kept chilled.

For morphological examination, thick sections of sponge preserved in ethanol were made with a surgical blade and these sections were soaked in xylene-phenol solution before mounting on glass slides in DPX[®]. These

preparations were examined under the light microscope at 40–400× magnification. Dissociated spicule suspensions were obtained by boiling sections in concentrated nitric acid. These spicules were subsequently mounted on brass stubs for examination under a JEOL 6510 scanning electron microscope (SEM). Residues of the spicule suspensions were also mounted on glass slides in DPX® for measurements of spicule dimensions under the light microscope. Spicule size data are given as minimum–average–maximum based on a minimum of 25 measurements for each spicule type unless otherwise indicated. Systematics and the order of treatment of sponge families and genera follow *Systema Porifera* (Hooper & Van Soest, 2002) and the World Porifera Database (Van Soest et al., 2017). Voucher specimens were deposited at the Lee Kong Chian Natural History Museum (Singapore) and The Natural History Museum, London (NHMUK).

DNA sequencing

DNA extractions were performed on a Hamilton Microlab STAR Robotic Workstation at the Sequencing Facility, NHM, using Qiagen DNeasy Blood and Tissue Kit. Approximately 660 bp of the mitochondrial gene COI were amplified using the primers LCO1490 5′-GGTCAA-CAAATCATAAAGATATTGG -3′ and HCO2198 5′-TAAACTTCAGGGTGACCAAAAAATCA -3′ (Folmer, Black, Hoeh, Lutz, & Vrijenhoek, 1994), and approximately 800 bp of the nuclear gene 28S rDNA (D1-D2 region) were amplified using C1′ 5′-ACCCGCTGAATT-TAAGCAT-3′ (Lê, Lecointre, & Perasso, 1993) and D1R 5′-AACTCTCTCMTTCARAGTTC-3′ (Colgan, Ponder, Beacham, & Macaranas, 2003). PCR mixtures contained 1 µL of each primer (10 µM), 2 µL of DNA template, and 21 µL of Red Taq DNA Polymerase 1.1X MasterMix (VWR). The PCR profile was as follows: 94°C/300s, (94°C/60s, 55°C/60s, 72°C/120s)×35 cycles, 72°C/300s. PCR purification was done using a Millipore Multiscreen 96-well PCR Purification System, and sequencing was performed on an ABI 3730XL DNA Analyser (Applied Biosystems) at the NHM Sequencing Facility, using the primers mentioned above. Overlapping sequence fragments were concatenated into consensus sequences using Geneious v.6.1.7 (<https://www.geneious.com>, Kearse et al., 2012).

Phylogenetic analyses

In total 70 terminal taxa from a range of families and genera with asterose microscleres were used in the phylogenetic analyses of 28S, and 101 taxa for COI. Sequences downloaded from GenBank were aligned with the new species using MAFFT (Katoh, Misawa, Kuma, & Miyata, 2002) for the 28S and MUSCLE (Edgar, 2004) for COI,

both programs as plug-ins in Geneious, with default settings. Bayesian phylogenetic analyses (BA) were conducted with MrBayes 3.2 (Ronquist et al., 2012), analyses were run three times for 10 million generations of which 2.5 million generations were discarded as burn-in. The evolutionary model, GTR+I+G, used for the molecular data in BA was obtained using jModelTest (Posada, 2008).

Results and Systematics

Phylum Porifera Grant, 1836
 Class Demospongiae Sollas, 1885
 Subclass Heteroscleromorpha Cárdenas, Perez, & Boury-Esnault, 2012
 Order Axinellida Lévi, 1953
 Family Stelligeridae Lendenfeld, 1898
Plenaster gen. nov. Lim & Wiklund

Diagnosis. Skeleton comprising mainly of dense spheroxyasters, with occasional styles arranged in a haphazard fashion and, with most protruding the surface. Thinly encrusting growth form with hispid surface. Megascleres are exclusively styles. Microscleres are euasters with numerous tapering rays (>20) with recurved spines. Type species. *Plenaster craigi* sp. nov. (designation herein).

Etymology. The generic name *Plenaster* is used as a noun, derived from the Latin *plenus* (plenty, filled) and *aster* (star), in reference to the abundant spheroxyasters (star-shaped microscleres) making up the entire sponge.

Remarks

Stelligeridae is the only family in Order Axinellida that has members bearing styles and euasters like *Plenaster*, and this new genus is provisionally placed in this family. There are currently four genera (*Halicnemia*, *Higginsia*, *Paratimea*, and *Stelligera*) assigned to the family Stelligeridae according to Van Soest et al. (2017). Members of *Halicnemia* and *Higginsia* do not have euasters and instead have microxeas that are acanthose, centrangulate or straight. Only *Paratimea* and *Stelligera* have euasters with smooth rays. *Paratimea* can be further distinguished from *Plenaster* in having tylostyles and centrotylote tornotes or oxeas. *Stelligera* has oxeas in addition to styles as megascleres and a more complex skeleton comprising a lightly compressed axial skeleton, reticulated extra-axial skeleton and styles that radiate from the core of the sponge to the periphery.

Plenaster craigi sp. nov. Lim & Wiklund
 Figs (2–6)



Fig. 2. Individuals of *Plenaster craigi* sp. nov. encrusting on polymetallic nodules (indicated by arrows) obtained from a box core sample at sampling site S07, Ocean Mineral Singapore exploration area in the Clarion-Clipperton Zone (East Pacific Ocean). Scale bar = 10 mm.

Holotype. ZRC.POR.0280, ABYSSLINE 02 Cruise (OMS), 27 February 2015, 4148 m depth, on polymetallic nodule in box core (BC10) at study site (S04), 12°00.560'N 117°10.692'W. Coll: T.G. Dahlgren, H. Wiklund, A.G. Glover, M.J. Brasier, K.S. Tan, and S.C. Lim.

Paratype. NHMUK2017.3.1.1, ABYSSLINE 02 Cruise (OMS), 25 March 2015, 4146 m depth, on polymetallic nodule in box core (BC25) at study site (S11), 12°00.559'N 117°22.819'W. Coll: T.G. Dahlgren, H. Wiklund, A.G. Glover, M.J. Brasier, K.S. Tan, and S.C. Lim.

Description

Sponge thinly encrusting on the top surfaces of polymetallic nodules, typically 1–2 mm in thickness (not more than 4 mm) and 5 mm wide (not more than 10 mm) (Figs 2, 3). Sponge irregularly shaped, snow-white in colour; surface appears smooth and glistening from afar but hispid when observed in detail. Oscules not observed in all the specimens brought up to surface.

Skeleton. Consists of dense mass of spheroxyasters and a small number of styles with the pointed end protruding through the surface (Fig. 4), in excess of 500 μm in length. Numerous interconnecting canals 10–100 μm in diameter in the choanosome.



Fig. 3. *Plenaster craigi* sp. nov. holotype after it was retrieved from a box core sample at sampling site S04 in Ocean Mineral Singapore exploration area, Clarion-Clipperton Zone (East Pacific Ocean). The hispid surface of the sponge is due to the protruding styles. Scale bar = 1 mm.

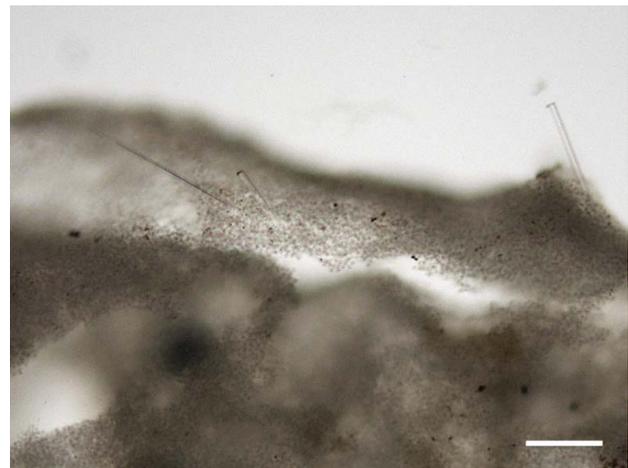


Fig. 4. *Plenaster craigi* sp. nov. cross-section of skeleton. Scale bar = 100 μm .

Spicules. Comprises of styles and spheroxyasters. Styles (Fig. 5) straight, sometimes slightly bent, with a fairly large size range, 1500–1850–2200 μm \times 10–14.7–17 μm (N = 10). Spheroxyasters 11.6–12.5–13.9 μm in diameter

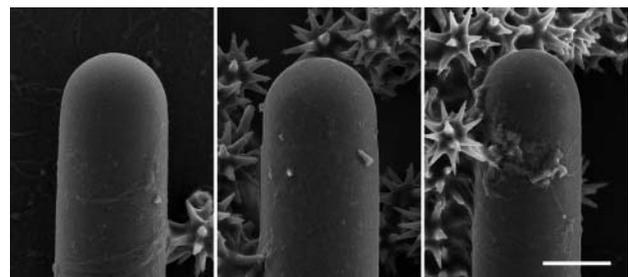


Fig. 5. Styles in *Plenaster craigi* sp. nov. Scale bar = 10 μm .

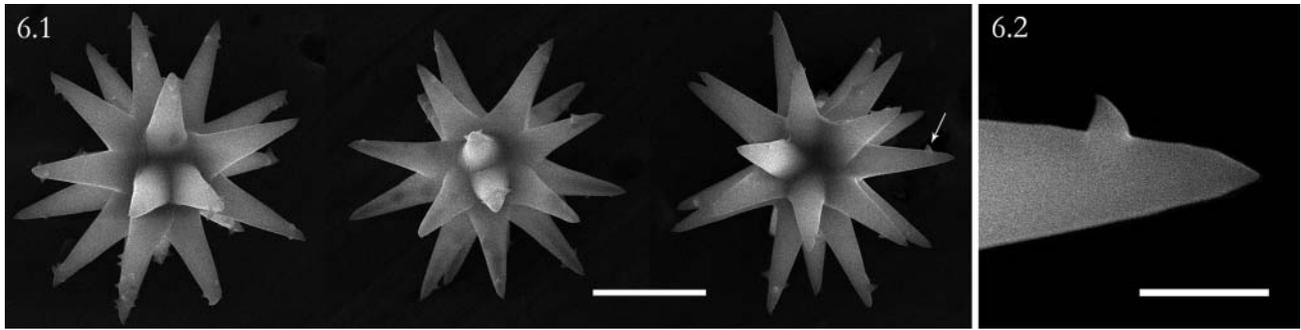


Fig. 6. 6.1, Spheroxyasters from holotype of *Plenaster craigi* sp. nov. Scale bar = 5 µm. 6.2, Magnified photograph shows a recurved spine (arrow). Scale bar = 1 µm.

bearing 22–27 rays, each ray may bear up to five recurved spines at the distal end (Fig. 6).

Molecular identification. The sequences of material have been deposited in GenBank with the accession numbers KY748655–57 for 28S (Paratype KY748657) and KY748658–63 for COI (Holotype KY486558; Paratype KY748661). Three of the sequenced specimens, including the paratype, have been deposited at NHMUK (NHMUK 2017.3.1.1–3).

Etymology. The specific epithet; *craigi*; is in honour of Prof. Craig R. Smith, University of Hawaii, who worked tirelessly on both long research cruises of the ABYSS-LINE programme (2013–2016) as Chief Scientist responsible for the highly successful sampling programme.

Distribution and abundance. So far known only from the eastern region of the Clarion Clipperton Zone, encrusting on polymetallic nodules at abyssal depths of between 4041–4183 m. A total of 42 individuals were obtained from 11 box cores in our 900 km² study area with a mean density of 15.3±8.9 (minimum = 4, maximum = 32) individuals per m². The nodule coverage in the 11 box cores was between 20.1–42.8% with an average of 32.8±7.7%. It should be noted that estimated nodule coverage is based on nodules visible on the surface.

Remarks

In morphological terms, *Plenaster craigi* sp. nov. is most similar to species in the genus *Timea* (Timeidae, Tethyida). All *Timea* species are known to be thinly encrusting (1 mm thick, or thinner), with tracts of tylostyles and/or styles rising from the substratum toward and beyond the surface (ectosome) and dense euasters forming a protective crust at the surface (see Carballo & Cruz-Baraza, 2006; Rützler, 2002). Perhaps the closest Indo-Pacific species in morphological terms is *Timea tethya* (de Laubenfels, 1954), which has only styles as megascleres (700 × 14 µm) and three categories of spheroxyasters

(38 µm, 20 µm, and 5–7 µm in diameter). *Plenaster craigi* sp. nov. is clearly distinct, having much longer styles and possessing only a single category of spheroxyasters (11.6–13.9 µm). There are four *Timea* species that have both styles and tylostyles as megascleres; *T. trigonostellata* (Carter, 1880) from Sri Lanka, *T. lophastrea* Hentschel, 1909 from Australia, *T. authia* de Laubenfels, 1930 from California (see de Laubenfels (1932) for full description), and *T. intermedia* (Lévi, 1958) from the Red Sea. Apart from the conspicuous absence of tylostyles, *P. craigi* sp. nov. possess considerably longer styles (up to 2200 µm in length) than *Timea* species. The preceding four species can be further differentiated from *P. craigi* sp. nov. by the following characters: *T. authia* has tylasters 6–23 µm in diameter; *T. trigonostellata* has distinctive tylasters 12.7 µm in diameter with four rays; *T. lophastrea* has oxes and lophasters with a branching broad distal end of each ray; and *T. intermedia* has subtylostyles and tylasters. In the Atlantic Ocean, *Timea chondrilloides* (Topsent, 1904) from the Azores also contains only styles as megascleres but it has much larger spherasters (27–42 µm in diameter) instead of spheroxyasters and those spherasters have much fewer rays. The recently described *Timea clippertoni* (Van Soest, Kaiser, & Van Syoc, 2011) from shallow waters off the Clipperton Island at 55 m depth, has distinctive calthrope-like asters (Van Soest et al., 2011).

Three species of *Hemiassterella* (*H. aristoteliana*, *H. digitata*, and *H. typus*) from the family Hemiassterellidae (Tethyida) are also similar in spicular composition to our material, while the rest of the *Hemiassterella* members have additional megascleres. *Plenaster craigi* sp. nov. differs from *H. aristoteliana*, *H. digitata*, and *H. typus* in having spheroxyasters which have a much larger centrum and pointed rays with prominent, robust recurved spines, while all three members of *Hemiassterella* have oxyasters with strongylote rays. All members of the genus *Hemiassterella* have exclusively vasiform or cup-shaped growth forms and more-or-less plumo-reticulate choanosomal skeletons (Hooper, 2002). Many thinly encrusting species are related to erect forms (Morrow et al., 2012) and

similarities in overall body shape do not always imply relatedness. The monotypic *Leptosastra* (Hemiasterellidae), is similar to *P. craigi* sp. nov.; *Leptosastra constellata* Topsent, 1904 (from the Atlantic Ocean) is a white and thinly encrusting sponge with a loosely constructed or a sub-hymedesmioid skeleton. However it has small acanthostyles (80–100 × 7 µm) and rhabds (135 × 2 µm) whereas *P. craigi* sp. nov. only has large styles. *Paratimea*, a former member of Hemiasterellidae before it was transferred to Stelligeridae (Axinellida) by Morrow and Cárdenas (2015), is also an encrusting sponge with a loosely constructed or a sub-hymedesmioid skeleton, but all *Paratimea* species have centrotylote tornotes or oxeas which are absent in *P. craigi* sp. nov.

Based on our molecular analyses, both 28S and COI trees (Figs 7, 8) *Plenaster craigi* sp. nov. was nested in Axinellida *sensu lato* clade and was sister to Axinellidae, Heteroxyidae, Raspailiidae, and Stelligeridae clades. The Axinellida *s.l.* clade was well supported in the 28S tree (Fig. 7) and to a lesser extent also in the COI tree (Fig. 8). This is not surprising, as 28S sequences are widely known to have higher variability and phylogenetic signal in lower taxonomic level than those of COI sequences (see Cárdenas, Menegola, Rapp, & Diaz, 2009; Chombard, Boury-Esnault, & Tillier, 1998; Erpenbeck *et al.*, 2007, 2012, 2016; Morrow *et al.*, 2013; Schuster *et al.*, 2015). For example, the families of Order Bubarida were well resolved in the 28S tree but not in the COI tree (Figs 7, 8). *Plenaster craigi* sp. nov. has the closest affinity to Hemiasterellidae sp. ‘KC869615’ but there was another Hemiasterellidae clade comprising of *Adreus fascicularis* ‘HQ379239’, *Axos cliftoni* ‘AY626308’, *Hemiasterella* sp. ‘AY561901’, and Tethyidae sp. ‘HQ379236’ was also present in Tethyida in the 28S tree. Similarly, in the COI tree, *P. craigi* sp. nov. displayed the closest affinity to a Hemiasterellidae clade comprising *Adreus fascicularis* ‘HQ379428’ and Tethyidae sp. ‘AY561986’ in Tethyida. Our results showed that there is an *Adreus* + *Axos* clade in Tethyida for Hemiasterellidae (Tethyida) but also a *Hemiasterella*/Hemiasterellidae clade in another order, the Axinellida *s.l.* These results suggest that Hemiasterellidae is a polyphyletic clade, as also shown by Morrow and Cárdenas (2015). *Plenaster craigi* sp. nov. is a sister taxon to a clade comprising *Cymbastela coralliophila* ‘JQ034537’ (Axinellidae, Axinellida) and *Petromica pacifica* ‘LN624194’ (Desmanthidae, Bubarida). All *Timea* sequences were distant from *Plenaster* clade in both trees. Timeidae formed a polyphyletic group in both trees. In the 28S tree, a clade comprising three *Timea* species (*Timea* sp. ‘AY626303’, *Timea* sp. ‘AY561907’ and *Timea* cf. *centrifera* ‘AY626304’), while *Timea lowchoyi* ‘AY561871’ clustered with Tethyidae. Similarly, in the COI tree, *Timea* sp. ‘AY561968’ and *Timea unistellata* ‘KC869427’ did not cluster together.

All families bearing euasters and most other families bearing asters were represented in our trees (Figs 7, 8) and

these were largely congruent with major demosponge phylogeny studies (Morrow & Cárdenas, 2015; Morrow *et al.*, 2012, 2013; Redmond *et al.*, 2013; Thacker *et al.*, 2013) as well as the current demosponge classification adopted in the World Porifera Database (Van Soest *et al.*, 2017). We attempted to include as many sequences of taxa with asterose microscleres as possible in our trees in view of the homoplasious nature of asterose microscleres (see Cárdenas, Xavier, Reveillaud, Schander, & Rapp, 2011; Morrow *et al.*, 2012, 2013). Euasters are asters with rays that radiate from a central point whilst asters have rays projecting from an axis that can be straight or spiral (Boury-Esnault & Rützler, 1997 and see also Cárdenas *et al.*, 2011). Not all the genera in the families above contain asterose microscleres, e.g., only *Paratimea* and *Stelligera* in Stelligeridae contain asterose microscleres but not the other two genera, *Higginsia* and *Halicnemia*. The Trachycladidae clade presented in Morrow *et al.*'s (2013) work on the distribution of asterose microscleres is absent in our trees but it is not critical as its members only have a very peculiar form of asterose microscleres known as spined vermiform spinispirae and not true euasters. It seems asterose microscleres can be very variable and plastic. Sterrasters in Geodiidae are derived from euasters, whilst those in Placospongiidae are from spirasters (Vosmaer & Vernhout, 1902) and a vastly different acanthoxea could have been derived from asterose microscleres (Topsent, 1897). It was also necessary to include Bubarida (an order that does not contain taxa with asterose microscleres) in our trees because of its close affinity to Axinellida exhibited as shown by Morrow, Picton, Erpenbeck, Boury-Esnault, Maggs, and Allcock, (2012, 2013), Redmond *et al.* (2013), and Thacker *et al.* (2013).

Discussion

Despite over 100 cruises to the CCZ since the start of polymetallic nodule exploration in the late 1970s (Nimmo *et al.*, 2013; ISA pers. comm., 2015; ISA, 2015), it appears that one of the most abundant and potentially ecologically important nodule-dwelling species has been overlooked. We make this taxonomic description and associated molecular and morphological data available to facilitate future studies in the region, and to encourage the establishment of a sustainable conservation strategy.

Plenaster craigi sp. nov., being simple in skeletal structure and bearing only two types of spicules, could be related to taxa from three different families in two orders; *Timea* (Timeidae; Tethyida); *Hemiasterella* and *Leptosastra* (Hemiasterellidae; Tethyida); and *Paratimea* (Stelligeridae; Axinellida). However our molecular data (Figs 7, 8) suggest that *P. craigi* sp. nov. is sister to Axinellidae, Heteroxyidae, Raspailiidae, and Stelligeridae nesting in Axinellida *s.l.* clade. Taxa with the closest affinity to

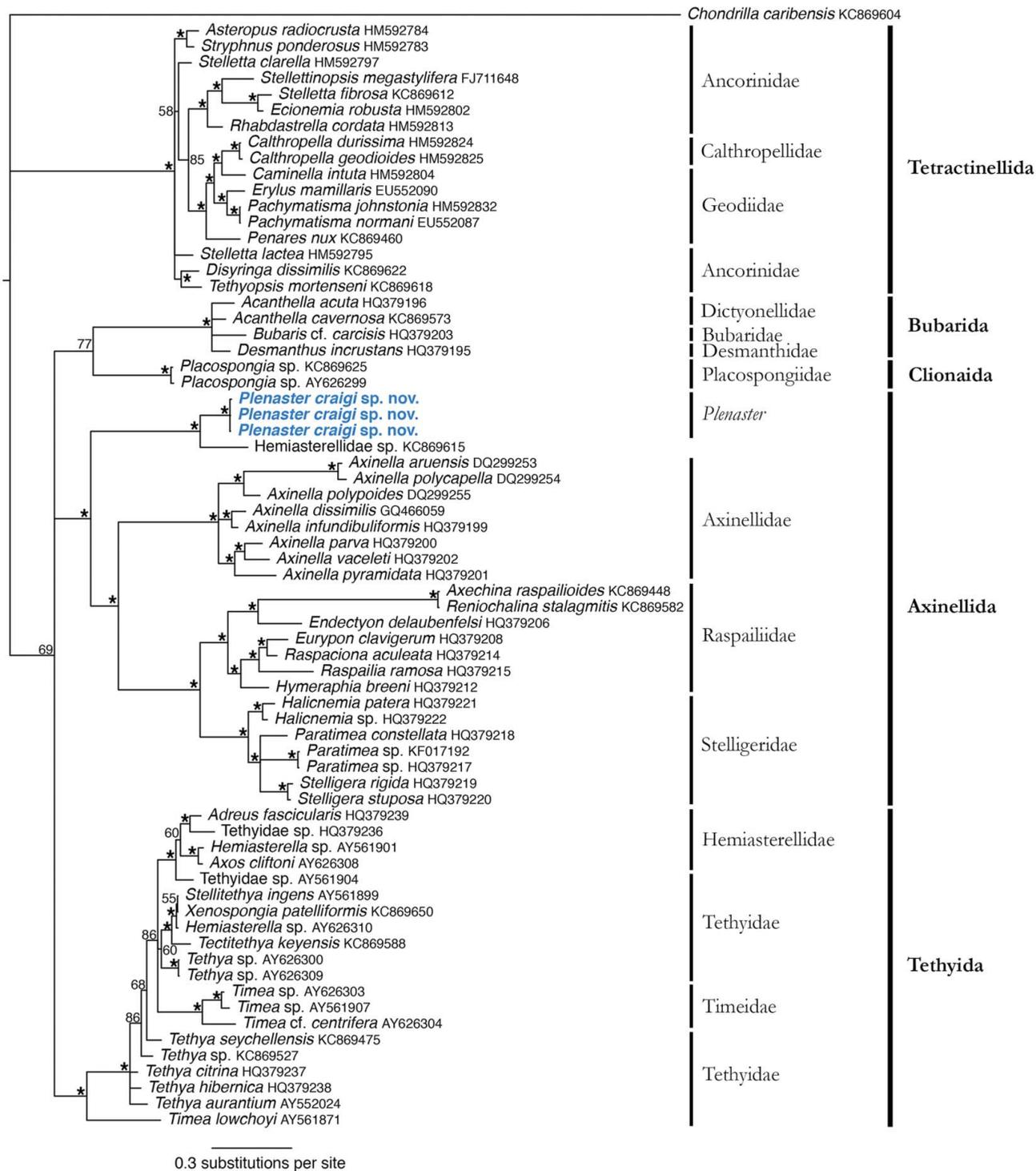


Fig. 7. Consensus tree from Bayesian analysis of 28S rRNA and 70 species in all the families and genera containing taxa (as far as possible) with both asterose microscleres. Asterisks denote posterior probabilities of 95 or higher. Taxon names are followed by their GenBank accession numbers.

P. craigi sp. nov. were revealed in both trees. In the 28S tree (Fig. 7), *Hemiasporellidae* sp. ‘KC869615’ is a sister species to *P. craigi* sp. nov. with good support. Similarly in the COI tree (Fig. 8), another *Hemiasporellidae* member, *Hemiasporella* sp. ‘AY561977’, was sister to *P. craigi*

sp. nov. Although we were not able to examine the morphological characters of these undetermined hemiasporellids, we have compared the morphological characters of four taxa (*Hemiasporella aristoteliana*, *H. digitata*, *H. typus*, and *Leptospora constellata*) in *Hemiasporellidae*

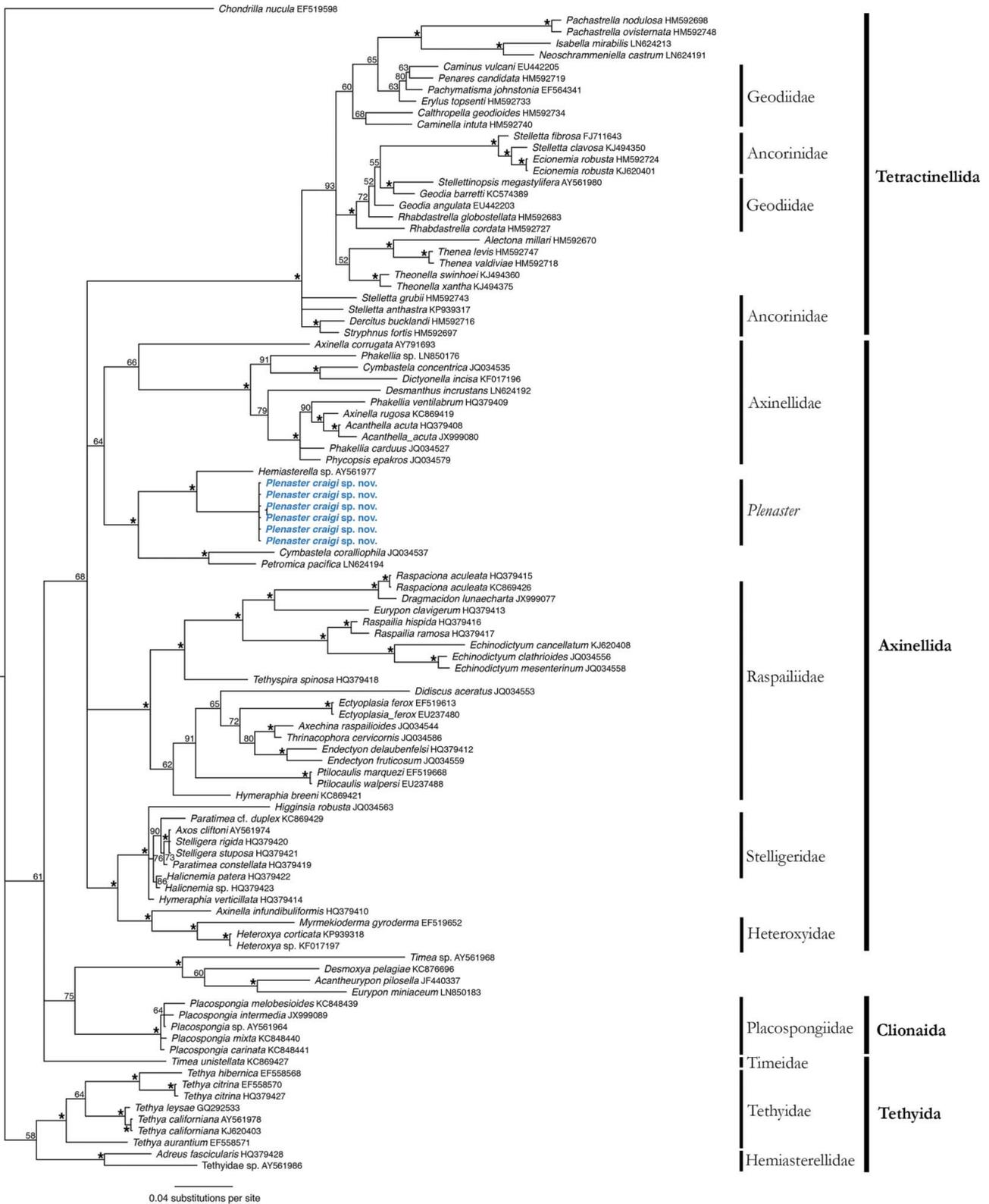


Fig. 8. Consensus tree from Bayesian analysis of COI and 101 species in all the families and genera containing taxa (as far as possible) with both asteroose microscleres. Asterisks denote posterior probabilities of 95 or higher. Taxon names are followed by their GenBank accession numbers.

Downloaded by [71.206.48.162] at 12:37 17 November 2017

most similar to *P. craigi* sp. nov., and they are clearly distinct from *P. craigi* sp. nov. Our trees based on 28S and COI sequences also showed that Hemiasterellidae is polyphyletic (Figs 7, 8) in agreement with Morrow and Cárdenas (2015) and Morrow et al. (2012). There is one clade comprising undetermined Hemiasterella/Hemiasterellidae species that clustered with *P. craigi* sp. nov. and another clade comprising of *Adreus* + *Axos* clade in Tethyida. Currently, only the clade in Tethyida is recognized (see Morrow & Cárdenas, 2015; Van Soest et al., 2017). However, the sequence for the type species for *Hemiasterella*, i.e., *H. typus* Carter, 1879, and other *Hemiasterella* species have not been sequenced to determine the true phylogeny of Hemiasterellidae. All *Timea* sequences were distant from *P. craigi* sp. nov. but Timeidae is polyphyletic and the type species for *Timea*, *T. stellata* Bowerbank, 1866, has not been sequenced. In our COI tree, there are two taxa, *Cymbastela coralliophila* 'JQ034537' and *Petromica pacifica* 'LN624194', forming a sister clade to *P. craigi* sp. nov. + *Hemiasterella* sp. 'AY561977'. Both *Cymbastela* (Axinellidae, Axinellida) and *Petromica* (Desmanthidae, Bubarida) are very different morphologically to each other and very different from *Plenaster*. *Cymbastela* are cup-shaped sponges having exclusively oxeas and without asters, and *Petromica* are massive sponges with desmas and without microscleres. It is important to note that *Cymbastela coralliophila* 'JQ034537' is unrelated to other *Cymbastela* species (Erpenbeck et al., 2012) so its generic assignment is uncertain. *Petromica pacifica* 'LN624194' formed a Desmanthidae clade in a Axinellida *s.l.* clade together with two other *Petromica* sequences in Schuster et al. (2015) but sequences of three other genera including the type species were absent (*Desmanthus*, *Paradesmanthus*, and *Sulcastrella*), so it is not a robust Desmanthidae clade. Examination of these materials is required to understand their affinities with *P. craigi* sp. nov.

Plenaster craigi sp. nov. has the closest affinity to Axinellidae in Axinellida *s.l.* clade (Figs 7, 8) amongst the four families but only Stelligeridae has members (*Paratimea* and *Stelligera*) containing styles and euasters like *Plenaster*. It is important to note that the classification of the current four families in the Order Axinellida is still equivocal (see Alvarez, De Voogd, Van Soest, & R. W., 2016; Morrow & Cárdenas, 2015; Morrow et al., 2012; Morrow et al., 2013; Redmond et al., 2013; Thacker et al., 2013). The family Stelligeridae Lendenfeld, 1898 was erected recently by Morrow et al. (2012) with *Halicnemia* and *Higginsia* (formerly in the now obsolete family Desmoxiidae), and *Paratimea* and *Stelligera* (formerly from Hemiasterellidae). Order Axinellida Lévi, 1953 was also recently resurrected by Morrow and Cárdenas (2015). It comprises existing Axinellidae (formerly from Order Halichondrida) and Raspailiidae (formerly from Order Poecilosclerida) and also the newly resurrected families

Stelligeridae and Heteroxyidae. The hypotheses derived from the above studies have consistently demonstrated that Axinellidae contains non-monophyletic genera with members that are unrelated to each other, including *Axinella*, the type genus of the family (Alvarez et al., 2016). Alvarez et al. (2016) were not convinced that the use of the Order Axinellida is appropriate as it appears not definable by clear morphological markers and has no practical use yet for classifying sponges without homologous DNA sequences. The erection of Order Axinellida and its families are based on phylogenetic analyses, which did not include all the type species of the genera. Morrow and Cárdenas (2015) acknowledged that a large degree of incongruence still exists between sponge phylogenies reconstructed on the basis of molecular sequences and those based on cladistic analysis of morphological characters (see also Morrow et al., 2012, 2013). There is still much work to be done in estimating phylogenetic hypotheses with sequences from type specimens and reconciling differences in morphological characters and molecular data. Many groupings proposed will continue to change with descriptions and sequencing of new species, use of new datasets and improvements in phylogenetic reconstruction methods (Morrow & Cárdenas, 2015).

Plenaster has an entire skeleton comprising of dense spheroxyasters with recurved spines, which is a strong apomorphy for a new genus or even a new family. However, *Plenaster* is a phylogenetic 'orphan' with no similar species and the erection of a new family may be better supported after similar taxa are discovered, perhaps from other abyssal environments. When the sequences of *Hemiasterella* type species, *H. typus*, and other *Hemiasterella* species are known, they might cluster with the existing or unknown *Hemiasterella*/Hemiasterellidae species in our trees in Axinellida *s.l.* clade to establish the validity of Hemiasterellidae in Order Axinellida. However, placing *Plenaster* in the current Hemiasterellidae which is in the Tethyida in the light of its location in Axinellida *s.l.* clade, is akin to placing *Plenaster* further away from its true phylogenetic position. With all these considerations, we have refrained from erecting a new family in the order Axinellida. Instead we have provisionally placed the new species in the family Stelligeridae, until more sequences and advanced phylogenetic reconstruction methods are available to reveal the true phylogeny of *Plenaster*.

Our study has also shown that *Plenaster craigi* sp. nov. was by far the most common metazoan organism attached to the nodules in our study area with a mean abundance of 15.3 ± 8.9 individuals per m^2 . Data (in preparation) from the UK-1 exploration area that is adjacent to the OMS area also suggests that *P. craigi* is the most common metazoan encrusting the nodules. This is corroborated by observations from subsequent research cruises to other areas in the eastern CCZ, suggesting that this species is possibly the most abundant metazoan on the nodules

(D. Jones, pers. comm., 2017; S. Taboada, pers. comm., 2017). High-definition imagery is now increasingly used to determine megafaunal abundance, but the available resolution is still insufficient to allow positive identification of small organisms such as *P. craigi*. For example, megafaunal abundance based on imagery at the adjacent UK-1 area averaged only 1.48 individuals per m² (Amon *et al.*, 2016), much less abundant than *P. craigi*. The likely reason that *P. craigi* was not reported previously is not for the lack of sampling (there have been many sampling cruises to the CCZ) but due clearly to insufficient observational and taxonomic work being carried out (e.g., Glover *et al.*, 2016a), and the fact that the species is quite small and requires careful specimen handling. In some CCZ areas, nodules can cover up to 99% of the seafloor surface area (ISA, 2010) which is much higher than the average nodule coverage (32.8±7.7%) in our study. The abundance of *P. craigi* could be higher at these higher nodule density areas as it is expected to correlate positively with the surface area of nodules available for attachment. Besides seamounts and occasional magmatic outcrops, the nodules are the only hard substratum available for sessile species to attach to in the CCZ (see Wendt, 1974), as there are hardly any other forms of hard substrata on the soft and muddy seafloor. The abundant and valuable nodules that might be extracted from CCZ in the future (see Glover & Smith, 2003; ISA, 2015) could result in loss of substratum for *P. craigi* and other sessile organisms, and consideration of this fauna is vital to making conservation decisions in the deep sea. As filter-feeders, *P. craigi* are also likely to be affected by sediment plumes generated from nodule collection. Owing to their abundance and wide distribution, they may prove to be an ideal 'indicator' species for future monitoring operations at the eastern CCZ, and possibly across the entire CCZ. Future work could also include determining the ecological role of *P. craigi*, as well as monitoring abundance and diversity before, during, and after harvesting activities in adjacent areas. Knowledge of the taxonomy, species structure, biogeography, and basic natural history of deep-sea animals is clearly essential for accurate assessment of the risk of species extinctions from large-scale mining (Glover & Smith, 2003).

Acknowledgements

Lim SC and Tan KS thank the National Research Foundation, Keppel Corporation and National University of Singapore for supporting this work done in the Keppel-NUS Corporate Laboratory. They would like to further acknowledge the St. John's Island National Marine Laboratory for providing the facilities necessary for conducting the research. The Laboratory is a National Research Infrastructure under the National Research Foundation

Singapore. AG, HW, and TD were supported by UK Seabed Resources Ltd (UKSRL), which is gratefully acknowledged. The conclusions put forward reflect the views of the authors alone, and not necessarily those of the institutions within the Corporate Laboratory or UKSRL. We thank Craig Smith, the Chief Scientist, for his efforts in coordinating the ABYSSLINE cruises and the overall programme. We would like to acknowledge the support of Madeleine Brasier from the Natural History Museum team in sampling on cruise AB02 and Magdalena Georgieva on cruise AB01. We are very grateful to the UKSRL supported-scientists with us on the cruise for sharing their knowledge and their help. Support for this work was received from JPI Oceans 'Mining Impact' and the Swedish Research Council Formas (TGD). AG was also partially supported by the European Union Framework 7 project 'Managing Impacts of Deep-sea Resource Exploitation' (MIDAS) project #603418. The crew of R/V Thomas G. Thompson were instrumental in making the survey a very successful one. The manuscript benefited greatly from constructive comments provided by two anonymous reviewers, to whom we extend our appreciation for their time and effort.

Disclosure statement

No potential conflict of interest was reported by the authors.

Funding

This work was supported by the Keppel-NUS Corporate Laboratory [grant number R-261-507-020-281]; UK Seabed Resources Ltd [grant number NHM_SBU_13002; UNI_Research_SRD100500]; European Union Framework 7 project [grant number 603418]; Swedish Research Council FORMAS [grant number 210-2014-1737].

References

- Alvarez, B., De Voogd, N. J., & Van Soest, R. W. M. (2016). Sponges of the family Axinellidae (Porifera: Demospongiae) in Indonesia. *Zootaxa*, 4137, 451–477. doi:10.11646/zootaxa.4137.4.1
- Amon, D. J., Ziegler, A. F., Dahlgren, T. G., Glover, A. G., Goineau, A., Gooday, A. J., ... Smith, C. R. (2016). Insights into the abundance and diversity of abyssal megafauna in a polymetallic-nodule region in the eastern Clarion-Clipperton Zone. *Scientific Reports* 6, 30492. doi:10.1038/srep30492
- Bluhm, H. (1994). Monitoring megabenthic communities in abyssal manganese-nodule sites of the East Pacific-Ocean in association with commercial deep-sea mining. *Aquatic Conservation-Marine and Freshwater Ecosystems*, 4, 187–201. doi:10.1002/aqc.3270040302
- Boury-Esnault, N., & Rützler, K. (Eds.). (1997). Thesaurus of Sponge Morphology. *Smithsonian Contributions to Zoology*, Vol. 596 (pp. 1–55). Washington DC: Smithsonian Institution Press.

- Bowerbank, J. S. (1866). *A Monograph of the British Spongiadae*, Vol. 2. London: Ray Society.
- Bruun, A. F. (1956). The abyssal fauna: Its ecology, distribution and origin. *Nature*, *177*, 1105.
- Carballo, J. L., & Cruz-Barraza, J. A. (2006). The genus *Timea* (Porifera: Timeidae) in the Eastern Pacific Ocean: Revision of type material, new species and intraspecific variability. *Cahiers de Biologie Marine*, *47*, 301–320.
- Cárdenas, P., Menegola, C., Rapp, H. T., & Diaz, M. C. (2009). Morphological description and DNA barcodes of shallow-water Tetractinellida (Porifera: Demospongiae) from Bocas del Toro, Panama, with description of a new species. *Zootaxa*, *2276*, 1–39.
- Cárdenas, P., Perez, T., & Boury-Esnault, N. (2012). Sponge systematics facing new challenges. In M. A. Becerro, M. J. Uriz, M. Maldonado, & X. Turon (Eds.), *Advances in Sponge Science: Phylogeny, Systematics, Ecology*, Vol. 61, (pp. 79–102). London, Amsterdam, Waltham, San Diego: Academic Press, Elsevier.
- Cárdenas, P., Xavier, J. R., Reveillaud, J., Schander, C., & Rapp, H. T. (2011). Molecular Phylogeny of the Astrophorida (Porifera, Demospongiae(rho)) Reveals an Unexpected High Level of Spicule Homoplasy. *Public Library of Science One*, *6*, 1–18. doi:10.1371/journal.pone.0018318
- Carter, H. J. (1879). On *Holastrella*, a Fossil Sponge of the Carboniferous Era, and on *Hemiasterella*, a new Genus of Recent Sponges. *Annals and Magazine of Natural History*, (5) *3*(14), 141–150, pl. XXI.
- Carter, H. J. (1880). Report on Specimens dredged up from the Gulf of Manaar and presented to the Liverpool Free Museum by Capt. W.H. Cawne Warren. *Annals and Magazine of Natural History*, (5) *6*(31), 35–61, pls. IV–VI; 129–156, pls. VII–VIII.
- Chombard, C., Boury-Esnault, N., & Tillier, S. (1998). Reassessment of homology of morphological characters in tetractinellid sponges based on molecular data. *Systematic Biology*, *47*, 351–366. doi:10.1080/106351598260761
- Colgan, D. J., Ponder, W. F., Beacham, E., & Macaranas, J. M. (2003). Gastropod phylogeny based on six segments from four genes representing coding or non-coding and mitochondrial or nuclear DNA. *Molluscan Research*, *23*, 123–148. doi:10.1071/MR03002
- Dahlgren, T. G., Wiklund, H., Rabone, M., Amon, D. J., Ikebe, C., Watling, L., ... Glover, A. G. (2016). Abyssal fauna of the UK-1 polymetallic nodule exploration area, Clarion-Clipperton Zone, central Pacific Ocean: Cnidaria. *Biodiversity Data Journal*, *4*, e9277. doi:10.3897/BDJ.4.e9277
- De Laubenfels, M. W. (1930). The Sponges of California. (Abstracts of dissertations for the degree of doctor of philosophy). *Stanford University Bulletin*, *5*(98), 24–29.
- De Laubenfels, M. W. (1932). The marine and fresh-water sponges of California. *Proceedings of the United States National Museum*, *81*(2927), 1–140.
- De Laubenfels, M. W. (1954). Occurrence of sponges in an aquarium. *Pacific Science*, *8*, 337–340.
- Downey, R. V., & Janussen, D. (2015). New insights into the abyssal sponge fauna of the Kurile-Kamchatka plain and Trench region (Northwest Pacific). *Deep-Sea Research Part II-Topical Studies in Oceanography*, *111*, 34–43. doi:10.1016/j.dsr2.2014.08.010
- Edgar, R. C. (2004). MUSCLE: Multiple sequence alignment with high accuracy and high throughput. *Nucleic Acids Research*, *32*, 1792–1797. doi: 10.1093/nar/gkh340
- Erpenbeck, D., Duran, S., Rutzler, K., Paul, V., Hooper, J. N. A., & Wörheide, G. (2007). Towards a DNA taxonomy of Caribbean demosponges: A gene tree reconstructed from partial mitochondrial CO1 gene sequences supports previous rDNA phylogenies and provides a new perspective on the systematics of Demospongiae. *Journal of the Marine Biological Association of the United Kingdom*, *87*, 1563–1570. doi:10.1017/s0025315407058195
- Erpenbeck, D., Hall, K., Alvarez, B., Buettner, G., Sacher, K., Schaetzle, S., ... Wörheide, G. (2012). The phylogeny of halichondrid demosponges: Past and present re-visited with DNA-barcoding data. *Organisms Diversity & Evolution*, *12*, 57–70. doi:10.1007/s13127-011-0068-9
- Erpenbeck, D., Voigt, O., Al-Aidaros, A. M., Berumen, M. L., Buttner, G., Catania, D., ... Wörheide, G. (2016). Molecular biodiversity of Red Sea demosponges. *Marine Pollution Bulletin*, *105*, 507–514. doi:10.1016/j.marpolbul.2015.12.004
- Foell, E. J., & Pawson, D. L. (1986). Photographs of invertebrate megafauna from abyssal depths of the Northeastern Equatorial Pacific-Ocean. *Ohio Journal of Science*, *86*, 61–68.
- 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.
- Frankenberg, D., & Menzies, R. J. (1968). Some Quantitative Analyses of Deep-Sea Benthos Off Peru. *Deep-Sea Research*, *15*, 623–626.
- Glasby, G. P. (2000). Economic geology - Lessons learned from deep-sea mining. *Science*, *289*, 551–553. doi:10.1126/science.289.5479.551
- Glasby, G. P., Li, J., & Sun, Z. L. (2015). Deep-Sea Nodules and Co-rich Mn Crusts. *Marine Georesources & Geotechnology*, *33*, 72–78. doi:10.1080/1064119x.2013.784838
- Glover, A. G., Dahlgren, T., Wiklund, H., Mohrbeck, I., & Smith, C. R. (2016a). An End-to-End DNA Taxonomy Methodology for Benthic Biodiversity Survey in the Clarion-Clipperton Zone, Central Pacific Abyss. *Journal of Marine Science and Engineering*, *4*, 2. doi:10.3390/jmse4010002
- Glover, A. G., Higgs, N., & Horton, T. (2017). World Register of Deep-Sea species. Retrieved from <http://www.marinespecies.org/deepsea> (accessed 11 January 2017)
- Glover, A. G., & Smith, C. R. (2003). The deep-sea floor ecosystem: Current status and prospects of anthropogenic change by the year 2025. *Environmental Conservation*, *30*, 219–241. doi:10.1017/s0376892903000225
- Glover, A. G., Wiklund, H., Rabone, M., Amon, D., Smith, C. R., O'Hara, T., ... Dahlgren, T. (2016b). Abyssal fauna of the UK-1 polymetallic nodule exploration claim, Clarion-Clipperton Zone, central Pacific Ocean: Echinodermata. *Biodiversity Data Journal* *4*: E7251. doi: 10.3897/BDJ.4.e7251
- Gooday, A. J., Goineau, A., & Voltzki, I. (2015). Abyssal foraminifera attached to polymetallic nodules from the eastern Clarion Clipperton Fracture Zone: A preliminary description and comparison with North Atlantic dropstone assemblages. *Marine Biodiversity*, *45*, 391–412. doi:10.1007/s12526-014-0301-9
- Grant, R. E. (1836). Animal Kingdom. In R. B. Todd (Ed.), *The Cyclopaedia of Anatomy and Physiology*, Vol. 1. (pp. 107–118). London: Sherwood, Gilbert, and Piper.
- Halbach, P., Friedrich, G., & von Stackelberg U. (Eds.). (1988). *The manganese nodule belt of the Pacific Ocean: Geological environment, nodule formation, and mining aspects*. Stuttgart: F. Enke Verlag.
- Hessler, R. R., & Jumars, P. A. (1974). Abyssal community analysis from replicate box cores in central North Pacific. *Deep-*

- Sea Research*, 21, 185–209. doi:10.1016/0011-7471(74)90058-8
- Hentschel, E. (1909). Tetraxonida. I. Teil. In W. Michaelsen & R. Hartmeyer (Eds.), *Die Fauna Südwest-Australiens. Ergebnisse der Hamburger Südwest-Australischen Forschungsreise 1905*, Vol 2(21). (pp. 347–402, pls. XXII–XXIII). Jena: Fisher.
- Hooper, J. N. A. (2002). Family Hemiasterellidae Lendenfeld, 1889. In J. N. A. Hooper & R. W. M. Van Soest (Eds.), *Systema Porifera. A guide to the classification of sponges*, Vol. 1 (pp. 186–195). New York, Boston, Dordrecht, London, Moscow: Kluwer Academic/ Plenum Publishers.
- Hooper, J. N. A., & Van Soest, R. W. M. (Eds.). (2002). *Systema Porifera: A Guide to the Classification of Sponges*. New York, Boston, Dordrecht, London, Moscow: Kluwer Academic/Plenum Publishers.
- International Seabed Authority. (1999). *Deep-seabed Polymetallic Nodule Exploration: Development of Environmental Guidelines: Proceedings of the International Seabed Authority's Workshop Held in Sanya, Hainan Island, People's Republic of China, 1-5 June 1998*. Kingston: International Seabed Authority.
- International Seabed Authority. (2010). *A Geological Model Of Polymetallic Nodule Deposits In The Clarion-Clipperton Fracture Zone. ISA Technical Study: No. 6*. Kingston: International Seabed Authority.
- International Seabed Authority. (2015). *Deep Sea Macrofauna of the Clarion-Clipperton Zone Taxonomic Standardization Workshop Ulsan, the Republic of Korea, 23-30 November 2014. ISA Technical Study: No. 13*. Kingston: International Seabed Authority.
- Kamenskaya, O. E., Gooday, A. J., Tendal, O. S., & Melnik, V. F. (2015). Xenophyophores (Protista, Foraminifera) from the Clarion-Clipperton Fracture Zone with description of three new species. *Marine Biodiversity*, 45, 581–593. doi:10.1007/s12526-015-0330-z
- 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.
- Kearse, M., Moir, R., Wilson, A., Stones-Havas, S., Cheung, M., Sturrock, S., ... Drummond, A. (2012). Geneious Basic: An integrated and extendable desktop software platform for the organization and analysis of sequence data. *Bioinformatics*, 28, 1647–1649. doi:10.1093/bioinformatics/bts199
- Koltun, V. M. (1970). Sponge fauna of the northwestern Pacific from the shallows to the hadal depths. In V. G. Bogorov (Ed.), *Fauna of the Kurile-Kamchatka Trench and its environment*. Institute of Oceanology of the Academy of Sciences of the U.S.S.R., Vol. 86 (pp. 165–221). Moscow: Akademiya Nauk SSSR. Trudy Instituta Okeanologii in P.P. Shishov and Izdatelstvo Nauka.
- Lê, H. L., Lecointre, G., & Perasso, R. (1993). A 28S rRNA-based phylogeny of the gnathostomes: First steps in the analysis of conflict and congruence with morphologically based cladograms. *Molecular Phylogenetics and Evolution*, 2, 31–51. doi:10.1006/mpev.1993.1005
- Lendenfeld, R. Von (1898). Die Clavulina der Adria. *Nova acta Academiae Caesareae Leopoldino Carolinae germanicae naturae curiosorum*, 69, 1–251, pls. I–XII.
- Lendenfeld, R. Von (1915). The Sponges. 3. Hexactinellida. In: Reports on the Scientific Results of the Expedition to the Eastern Tropical Pacific, in charge of Alexander Agassiz, by the U.S. Fish Commission Steamer 'Albatross', from October, 1904, to March, 1905, Lieut. Commander L.M. Garrett, U.S.N., Commanding, and of other expeditions of the 'Albatross', 1891-1899 (29). *Memoirs of the Museum of Comparative Zoology at Harvard College*, 42, 1–396, pls. 1–109.
- Lévi, C. (1953). Sur une nouvelle classification des Démosponges. *Compte rendu hebdomadaire des séances de l'Académie des sciences. Paris*, 236, 853–855.
- Lévi, C. (1958). Résultats scientifiques des Campagnes de la 'Calypso', Campagne 1951–1952 en Mer Rouge (suite). 11. Spongiaires de Mer Rouge recueillis par la 'Calypso' (1951-1952). *Annales de l'Institut Océanographique*, 34, 3–46.
- Lévi, C. (1964). Spongiaires des zones bathyale, abyssale et hadale. *Galathea Report. Scientific Results of The Danish Deep-Sea Expedition Round the World, 1950–52*, 7, 63–112, pls. II–XI.
- Morrow, C. C., & Cárdenas, P. (2015). Proposal for a revised classification of the Demospongiae (Porifera). *Frontiers in Zoology*, 12. doi:10.1186/s12983-015-0099-8
- 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. doi: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., ... 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. doi:10.1093/icb/ict065
- Mullineaux, L. S. (1987). Organisms living on manganese nodules and crusts - distribution and abundance at 3 North Pacific sites. *Deep-Sea Research*, 34, 165–184. doi:10.1016/0198-0149(87)90080-x
- Nimmo, M., Morgan, C., & Banning, D. (2013). *NI 43-101 Technical Report. Clarion-Clipperton Zone Project, Pacific Ocean*. Queensland: Golder Associates.
- Owen, D. M., Sanders, H. L., & Hessler, R. R. (1967). Bottom photography as a tool for estimating benthic populations. In J. B. Hersey (Ed.), *Deep-Sea Photography. Johns Hopkins Oceanographic Studies*, Vol. 3 (pp. 229–234). Baltimore: Johns Hopkins Press.
- Posada, D. (2008). jModelTest: Phylogenetic model averaging. *Molecular Biology and Evolution*, 25, 1253–1256. doi:10.1093/molbev/msn083
- Redmond, N. E., Morrow, C. C., Thacker, R. W., Diaz, M. C., Boury-Esnault, N., Cárdenas, P., ... 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. doi:10.1093/icb/ict078
- Reiswig, H. M., & Stone, R. P. (2013). New glass sponges (Porifera: Hexactinellida) from deep waters of the central Aleutian Islands, Alaska. *Zootaxa*, 3628, 1–64.
- Ridley, S. O., & Dendy, A. (1887). Report on the Monaxonida collected by H.M.S. 'Challenger' during the years 1873-1876. *Report on the Scientific Results of the Voyage of H.M.S. 'Challenger', 1873–1876. Zoology*, 20(59), i-lxviii, pp. 1–275, pls. I–LI, 1 map.
- Ronquist, F., Teslenko, M., van der Mark, P., Ayres, D. L., Darling, A., Höhna, S., ... Huelsenbeck, J. P. (2012). MrBayes 3.2: Efficient Bayesian phylogenetic inference and model choice across a large model space. *Systematic Biology*, 61, 539–542. doi:10.1093/sysbio/sys029

- Rützler, K. (2002). Family Timeidae Topsent, 1928. In J. N. A. Hooper & R. W. M. Van Soest (Eds.), *Systema Porifera. A guide to the classification of sponges*, Vol. 1 (pp. 266–267). New York, Boston, Dordrecht, London, Moscow: Kluwer Academic/ Plenum Publishers.
- Schulze, F. E. (1887). Report on the Hexactinellida collected by H.M.S. ‘Challenger’ during the years 1873–1876. *Report on the Scientific Results of the Voyage of H.M.S. ‘Challenger’, 1873-1876. Zoology*, 21 (Text), 1–514.
- Schuster, A., Erpenbeck, D., Pisera, A., Hooper, J., Bryce, M., Fromont, J., & Worheide, G. (2015). Deceptive Desmas: Molecular Phylogenetics Suggests a New Classification and Uncovers Convergent Evolution of Lithistid Demosponges. *Public Library of Science One*, 10. doi:10.1371/journal.pone.0116038
- Sollas, W. J. (1885). A Classification of the Sponges. *Annals and Magazine of Natural History*, (5) 16(95), 395.
- Thacker, R. W., Hill, A. L., Hill, M. S., Redmond, N. E., Collins, A. G., Morrow, C. C., ... Bangalore, P. V. (2013). Nearly Complete 28S rRNA Gene Sequences Confirm New Hypotheses of Sponge Evolution. *Integrative and Comparative Biology*, 53, 373–387. doi:10.1093/icb/ict071
- Thiel, H., Angel, M. V., Foell, E. J., Rice, A. L., & Schriever, G. (1998). *Marine Science and technology-environmental risks from large-scale ecological research in the deep sea. A desk study*. Luxembourg: Office for Official Publications of the European Communities.
- Tilot, V. (2006). *Biodiversity and distribution of the megafauna. Vol. 1. The polymetallic nodule ecosystem of the Eastern Equatorial Pacific Ocean. Intergovernmental Oceanographic Commission. Technical Series 69*. Belgium: Project UNESCO COI/Min Vlanderen.
- Topsent, E. (1897). Sur le genre *Halicnemis* Bowerbank. *Mémoires de la Société zoologique de France*, 10, 235–251.
- Topsent, E. (1904). Spongiaires des Açores. *Résultats des campagnes scientifiques accomplies par le Prince Albert I. Monaco*, 25, 1–280, pls. 1–18; 113–114; pl. XII Figure 9a–9c.
- Vacelet, J. (2006). New carnivorous sponges (Porifera, Poecilosclerida) collected from manned submersibles in the deep Pacific. *Zoological Journal of the Linnean Society*, 148, 553–584. doi:10.1111/j.1096-3642.2006.00234.x
- Van Soest, R. W. M., Boury-Esnault, N., Hooper, J. N. A., Rützler, K., de Voogd, N. J., Alvarez de Glasby, B., ... Cárdenas, P. (2017). *World Porifera database*. Retrieved from <http://www.marinespecies.org/porifera> (accessed 11 January 2017)
- Van Soest, R. W. M., Kaiser, K., & Van Syoc, R. (2011). Sponges from Clipperton Island, East Pacific. *Zootaxa*, 2839, 1–46.
- Vanreusel, A., Hilario, A., Ribeiro, P. A., Menot, L., & Arbizu, P. M. (2016). Threatened by mining, polymetallic nodules are required to preserve abyssal epifauna. *Scientific Reports*, 6, 26808. doi:10.1038/srep26808
- Veillette, J., Sarrazin, J., Gooday, A. J., Galeron, J., Caprais, J. C., Vangriesheim, A., ... Juniper, S. K. (2007). Ferromanganese nodule fauna in the Tropical North Pacific Ocean: Species richness, faunal cover and spatial distribution. *Deep-Sea Research Part I-Oceanographic Research Papers*, 54, 1912–1935. doi:10.1016/j.dsr.2007.06.011
- Vosmaer, G. C. J., & Vernhout, J. H. (1902). The Porifera of the Siboga-Expedition. I. The genus *Placospongia*. In M. Weber (Ed.), *Siboga-Expeditie. Uitkomsten op zoologisch, botanisch, oceanographisch en geologisch gebied verzameld in Nederlandsch Oost-Indië 1899–1900 aan boord H.M. ‘Siboga’ onder commando van Luitenant ter zee le kl. G.F. Tydeman. 9(Monographie VIa)*. (pp. 1–17, pls. I–V). Leiden: Brill EJ.
- Wendt, J. (1974). Encrusting Organisms in Deep-Sea Manganese Nodules. In K. J. Hsü & H. C. Jenkyns (Eds.), *Pelagic Sediments: On Land and under the Sea* (pp. 437–447). Oxford: Blackwell Publishing Ltd.

Associate Editor: Joanne Porter