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Shallow water sponges that associated to mangrove ecosystem at Labuhan conservation area in Sepulu, Bangkalan, Madura, East Java Province

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A B S T R A C T
Sponges (Porifera) are benthic sessile animals that have an important role in coastal ecosystems. Mangrove habitat is an example of a coastal ecosystem that can be inhabited by sponges. Research diversity on shallow water sponges in mangrove areas in Indonesia or even large scale such as the Indo-Pacific area are still relatively underestimated because of an extreme difference tidal factors. For this reason, we aimed to research a diversity of shallow water sponges in mangrove Indonesia, especially in East Java Province to enrich and inform additional data on its biodiversity. Research on shallow water sponges in mangrove was conducted from June 2017 to January 2018 at two stations located on the coast of Labuhan Village, Sepulu District, Bangkalan Regency, Madura - East Java. Mangrove areas are consisted of *Sonneratia* area in the west (B) and *Rhizophora* mangroves in the east (R). There were six sponge genera identified, namely *Spongia*, *Dysisdea*, *Lendelfedia*, *Dactylospongia*, *Cynachyrella* and one unidentified genus. *Sonneratia* mangrove was inhabited by of five genera sponges, whereas *Rhizopora* mangrove was inhabited only by one genus

Keywords: *Shallow water sponges, Diversity, Mangrove ecosystems, Sepulu Madura*

I. INTRODUCTION

Sponges are benthic animals that mostly inhabit marine ecosystem and dominate shallow and deep sea. These animals also possess a high diversity and biomass (van Soest et al. 2012). Eight thousand sponge species have been identified and it is estimated that there are still more than 6000 species have not been yet identified from a number of more than 15,000 species (Hooper & van Soest 2002; van Soest et al. 2012). Researches on sponges has been increased since two decades ago because first, sponges are first multicellular organisms that are developed from animal lineages or Animalia because of their primitive structure of tissues (Dohrmann & Wörheide 2013; Pisani et al. 2015). For this reason, sponges are suitable for research of animal evolution and origin of multicellular life. Second, sponges are chosen as a source of alternative natural drugs exploration because sponges have a potential content of bioactive or biopharmaceutical compounds (Faulkner 2002; Proksch et al. 2002). Third, sponges have an important role in marine ecosystems, especially coral reefs. Ecologically, sponges act as bio-eroders for coral restoration, recycling chemical elements in t sea, living places for symbiotic organisms such as Dinoflagellates, (Bell 2008) and sponges are supporting animals for coral survival in areas with a minimal nutrition (de Goeij et al. 2013).

Indonesia's tropical waters have been known as area that possess a high diversity of marine organism, including sponges as a component of coral reefs and marine ecosystems (Briggs 1999; Hoeksema 2004). Exploration of sponge

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diversity in this region has not been intensively carried out as in other regions like Mediterranean waters in Europe and the Caribbean in Central America (van Soest et al. 2012). The intensity study of sponge diversity in Indonesia is still dominated with researches in Coral reef triangle such as Spermonde region that covers South Sulawesi, North Sulawesi and Southeast Sulawesi waters, in addition to research in Java that has been published, namely in a Pulau Seribu waters in Jakarta (Bell et al. 2014; Cleary & de Voogd 2007; de Voogd et al. 2006; Swierts et al. 2013). For this reason, research in other parts of Java, especially in East Java is needed to enrich diversity data of sponges in Indonesia.

Preliminary research on sponge diversity in East Java Province was carried out and published in the period 2009-2013 (Abdillah et al. 2013a; Abdillah et al. 2013b; Setiawan et al. 2009). However, type of sponges that has been explored areas shallow water sponges that inhabit reef areas. Therefore, it is necessary to explore sponges that have special habitats such as shallow water sponges that associate to mangrove habitat. Mangroves have generally been known as one of coastal ecosystems that supports very a high species richness and abundance of fish (Noor et al. 1999) Some of these fish species possess a high economic value and most of the world's mangroves have obtained a status protection as nursery ground of fishes (Faunce & Serafy 2006; Redjeki 2013). Besides a place to support fish conservation, mangrove ecosystems also hold an important role for preservation of other biota like sponges (Diaz & Rutzler 2009; Wulff 2004). Recent observation study of sponges in Situbondo mangrove area was carried out in Tampora beach area in 2016 and discovered 12 sponge species associated with mangroves (Alivy et al. 2016). In this study mangrove area in Sepulu District in Bangkalan Regency, Madura Island, East Java is chosen for further research because this area has a healthy category of mangrove condition. In addition, a coastal conservation program has just been initiated in this area (Personal preliminary data and observations from Ecology Laboratory, Department of Biology, 2015).

II. Material and Methods

Sampling was carried out on 21st-23rd August 2017. Data was collected at two sampling stations located on the coast of Labuhan Village, Sepulu District, Bangkalan Regency, Madura - East Java (Figure 1). The sampling station includes *Sonneratia* mangrove area in the west (W) and *Rhizophora* mangrove in the East (E). Living form of sponges were also photographed to assist identification procedure. Sponges identification were carried out at the Zoology Laboratory and animal engineering at Biology Study Program Surabaya Department ITS Surabaya. Sponges that have been sampled were taken from apical part (volume ± 10 cm3) and were with 96% ethanol. Furthermore, sponges are preserved into vial containing 96% ethanol.

Identification stage of sponge consist of three stages. First, a sponge of ± 10 cm3 was divided into three parts, namely ± 2 cm3, ± 4 cm3 and 4 cm3. Small pieces measuring ± 2 cm3 were soaked in the bleach solution and after shedding, spicules were settled below. Spicules solution were rinsed 5x with distilled water to eliminate bleach. After bleaching solution rinsed, spicules solutions were dipped in ethanol 70% and then dry mounted in object glass with adhesive and covered with cover glass. Second, a large part ± 4 cm 3 was cut transversely and longitudinally with a hand-cutting knife about 200 microns thick. Afterwards, both tissues were also mounted in object glass directly and covered with cover glass. Third, those both spicules and skeleton slides were observed under a microscope The sponge identification guide book from Hooper & van Soest 2002 was used to determine the identity of the species or genus along with the database list of the porifera (van Soest et al. 2018) http://www.marineSpesies.org/porifera/

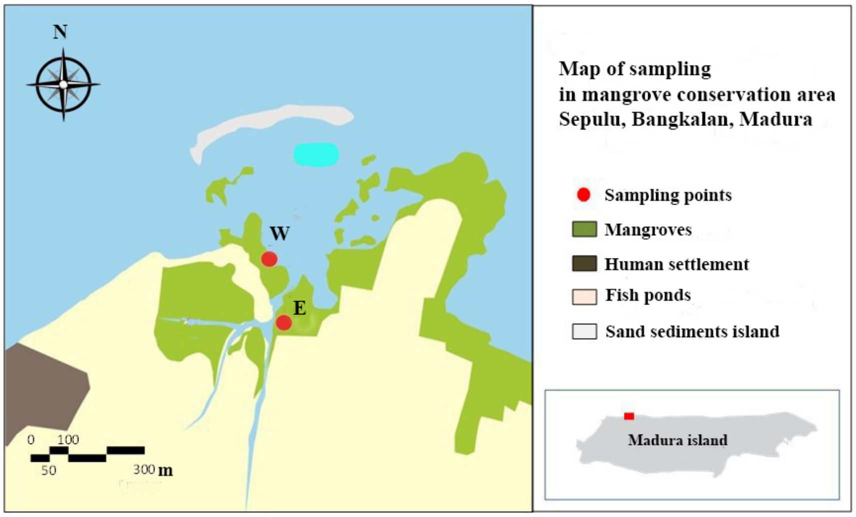

Figure 1. Sampling area of research.

III. Results

Thirteen specimens were sampled from western part of *Sonneratia* mangroves (W) and only one type of specimen only was sampled at *Rhizopora* mangroves (E). The type of specimens identified were as follows:

Class : Demospongia Sollas, 1885
Sub Class : Keratosa Grant, 1861
Order : Dictyoceratida Minchin, 1900
Family : Spongiidae Gray, 1867
Genus : *Spongia* Linnaeus, 1759
Species : *Spongia* sp.

Two *Spongi*a sp. specimens were obtained. i.e., samples 2017_SEP_B1 (Figure 2) and 2017_SEP_B8 (Figure 3). Both specimens shared a similar morphological characters, which is black on ectosome part and brownish orange in choanosome part. The colors of two specimens also turned paler when preserved in 96% ethanol. Consistency of two sponges were springy and tough. Although, 2017_SEP_B8 specimen has a very clear osculum in its ectosome. Morphological characters of skeleton tissue and sponging fibers in 2017_SEP_B8 resembles 2017_SEP_B1 in its cavity structure and absence of mineral component or spicules in skeleton.

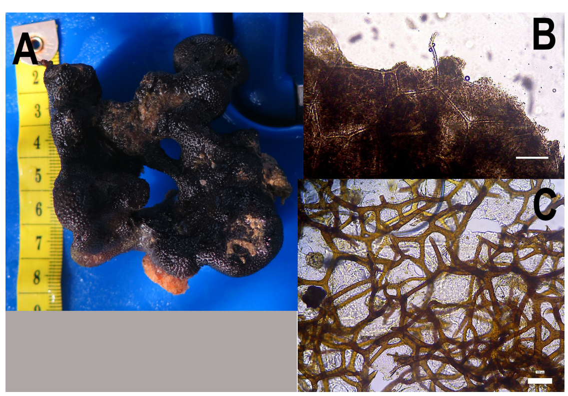

Figure 2 **A**, "Lifeform", sample 2017_SEP_B1, B, longitudinal or perpendicular section with 40x magnification, and C cross or tangential sections 40x magnification. Scale B and C =100 µm

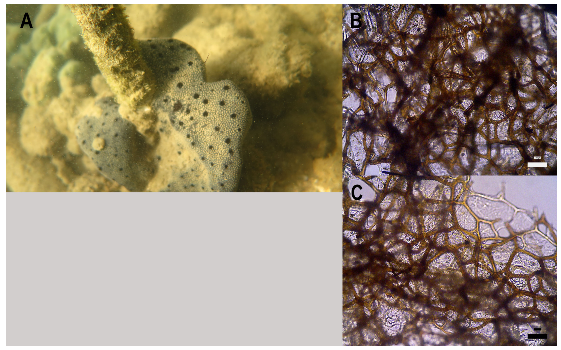

Figure 3 A, "Lifeform", sample 2017_SEP_B8, B, longitudinal or perpendicular section with 40x magnification, and C cross or tangential sections 40x magnification. Scale B and C =100 µm

Family : Dysideidae Gray, 1867
Genus : *Dysidea* Johnston, 1842
Species : *Dysidea* sp.

One *Dysidea* sp. specimen was obtained. i.e. sample 2017_SEP_B5 (Figure 4) Life colour specimen has a green color on the part of ectosome and choanosome and furthermore, possess has a shape like a thin leaf sheet. Color of specimen changed paler when preserved in 96% ethanol. The consistency of the sponge is stout. Morphological characters of skeleton tissue are lack of sponging fibers and absence of mineral spicules in skeleton.

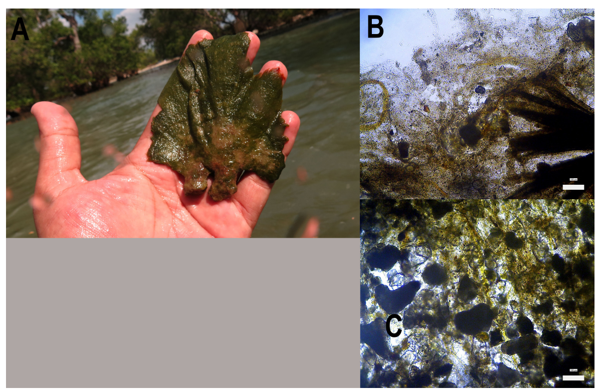

Figure 4 A, "Lifeform", sample 2017_SEP_B5, B, longitudinal or perpendicular section with 40x magnification, and C cross or tangential sections 40x magnification. Scale B and C =100 µm

Family : Thorectidae Bergquist, 1978
Sub Family: Phyllospongiinae Keller, 1889
Genus : *Lendenfeldia* Bergquist, 1980
Species : *Lendenfeldia* sp.

Two specimens of *Lendenfeldia* sp were obtained, namely samples 2017_SEP_B3 (Figure 5) and 2017_SEP_B11 (Figure 6). Live color of both specimens were similar, which is green on the part of ectosome and choanosome. Furthermore, both specimens have an encrusting growth form. Colors of two specimens also turned paler when preserved in 96% ethanol. Consistency of two specimens are not tough. Morphological characters in skeleton tissue were presence of sponging fibers resembling T-shaped sheet and fraction structure, in addition to absence of mineral components or spicules in skeleton.

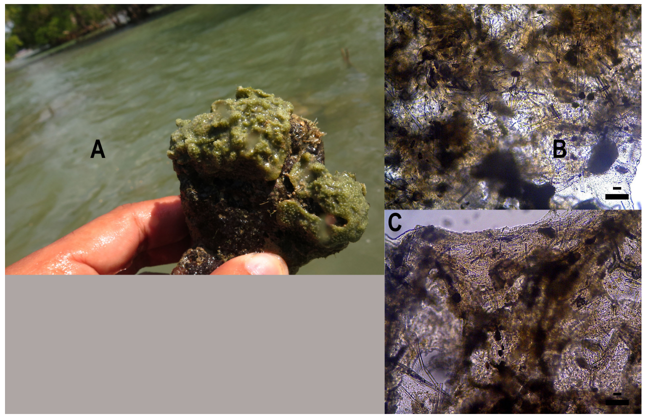

Figure 5 A, "Lifeform", sample 2017_SEP_B3, B, longitudinal or perpendicular section with 40x magnification, and C cross or tangential sections 40x magnification. Scale B and C =100 µm

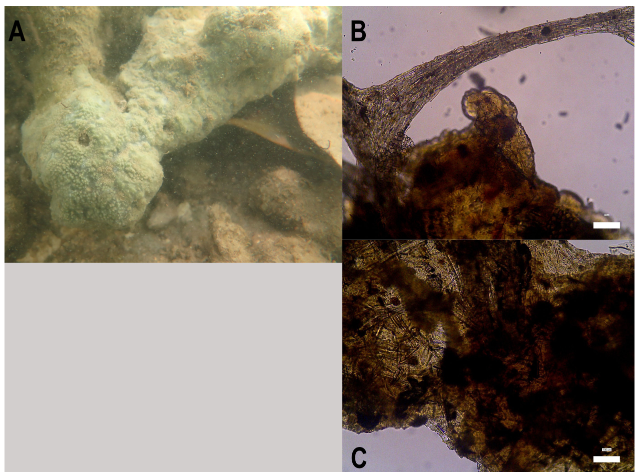

Figure 6 A, "Lifeform", sample 2017_SEP_B11, B, longitudinal or perpendicular section with 40x magnification, and C cross or tangential sections 40x magnification. Scale B and C =100 µm

Sub Family: Thorectinae Bergquist, 1978
Genus : *Dactylospongia* Bergquist, 1965
Species : *Dactylospongia* sp.

One *Dactylospongia* sp specimen, sample 2017_SEP_B4 was obtained (Figure 7). Live form is brownish yellow color and a nodule structure on its ectosome, in addition, choanosome part has a reddish-brown structure. Color of specimen changed paler when preserved in 96% ethanol. The consistency of the sponge is stout. Distinctive morphological characters in skeleton tissue are red color and compact spongin fiber components in skeleton in addition to absence of mineral components or spicules in skeleton.

Figure 7 A, "Lifeform", sample 2017_SEP_B4, B, longitudinal or perpendicular section with 40x magnification, and C cross or tangential sections 40x magnification. Scale B and C =100 µm

Subclass : Heteroscleromorpha Cárdenas, Pérez & Boury-Esnault, 2012
Order : Tetractinellida Marshall, 1876
Sub-Order: Spirophorina Bergquist & Hogg, 1969
Family : Tetillidae Sollas, 1886
Genus : *Cinachyrella* Wilson, 1925
Species : *Cinachyrella* sp.

Two *Cinachyrella* sp specimens were obtained, samples 2017_SEP_B9 (Figure 8) and 2017_SEP_B12 (Figure 9). Both specimens are similar that possess a shape like a ball. The first specimen has a yellow colored ectosomes and choanosomes while the second specimen has a brownish yellow ectosome and yellow choanosomes. Colors of two specimens also turned paler when preserved in 96% ethanol. The consistency of two sponges is stout and compressible. Distinctive morphological features in skeleton tissue are presence of spongin fiber components resemble structure of sheets and elongated parchments like a fused spicule. Spicules should be found, but in this study no mineral component or spicules was found in skeleton

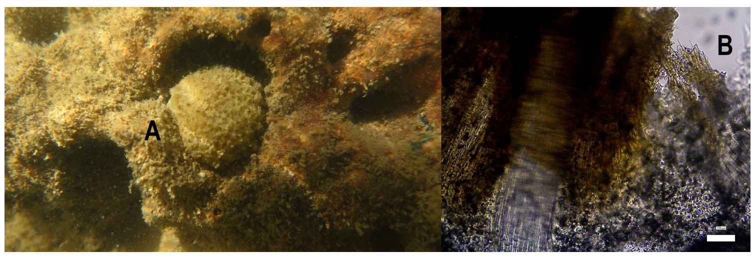

Figure 8 A, "Lifeform", sample 2017_SEP_B9, and B, longitudinal or perpendicular section with 40x magnification, Scale B =100 µm

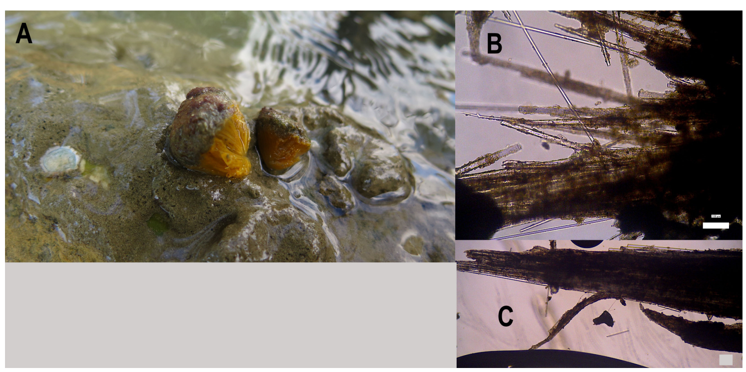

Figure 9 A, "Lifeform", sample 2017_SEP_B12, B, longitudinal or perpendicular section with 40x magnification, and C cross or tangential sections 40x magnification. Scale B and C =100 µm

Order: Haplosclerida Topsent, 1928
Family: Chalinidae Gray, 1867
Genus: *Haliclona* Grant, 1841
Sub genus: *Haliclona (Reniera)* Schmidt, 1862
Species: *Haliclona (Reniera)* sp.

Three *Haliclona (Reniera)* sp specimens, 2017_SEP_B2 (Figure 10), 2017_SEP_B6 (Figure 11) and 2017_SEP_T2 (Figure 12) were obtained. The three specimens have different morphological forms, namely; the first specimen has a honeycomb-like structure with white-colored ectosomes and choanosomes. The second specimen has a branch-like structure and a clear osculum with black colored ectosomes and choanosomes. The third specimen shaped like "encrusting" and attached to coral fragments with yellowish brown ectosomes and choanosomes. Although those specimens have a different morphology features, character of sub genus *Haliclona (Reniera)* appears in those three specimens, namely; structures that are crumble with minimal spongin fibers with type of oxea mineral spicules accompanied by spongin fibers in skeleton with a rectangular cavity structure. The color of the three specimens also turns paler when preserved in 96% ethanol. The dimensions of oxeas spicules in the first, second and third specimens of this sub genus are; 112,3-**127**-158.9 x 4,4-6-6.2 µm; 134.5-**156.5**-186.1 x 6-**8.6**-10.6 µm; 556.7-**709.6**-928.7 x 12,3-**23.3**-31.8 µm

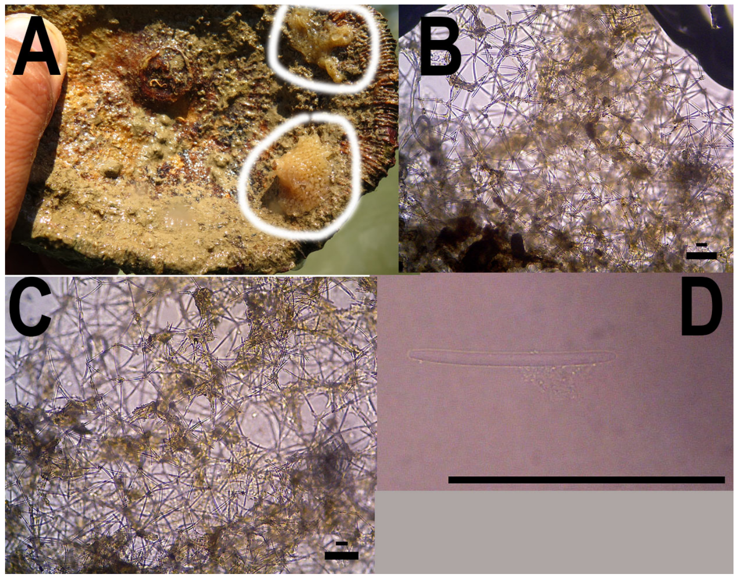

Figure 10 A, "Lifeform", sample 2017_SEP_B2, B, longitudinal or perpendicular section with 40x magnification, C cross or tangential sections 40x magnification and D, oxeas skeleton with 400x magnification. Scale B, C and D =100 µm

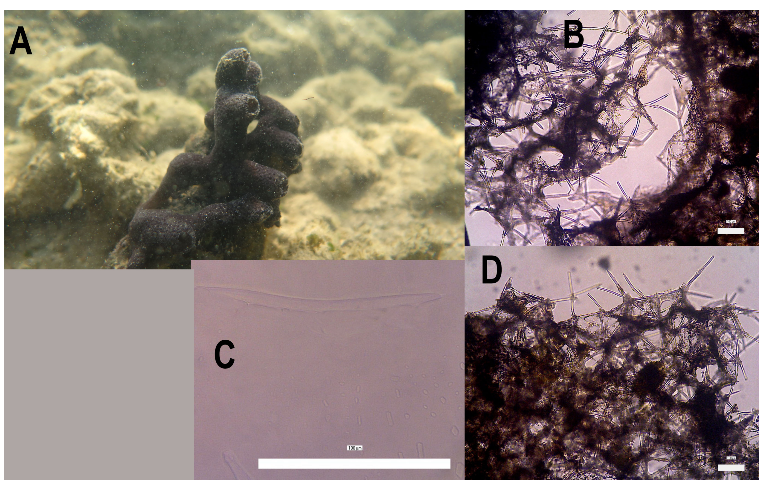

Figure 11 A, "Lifeform", sample 2017_SEP_B6, B, longitudinal or perpendicular section with 40x magnification, C, oxeas skeleton with 400x magnification and D, cross or tangential sections 40x magnification. Scale B, C and D =100 µm

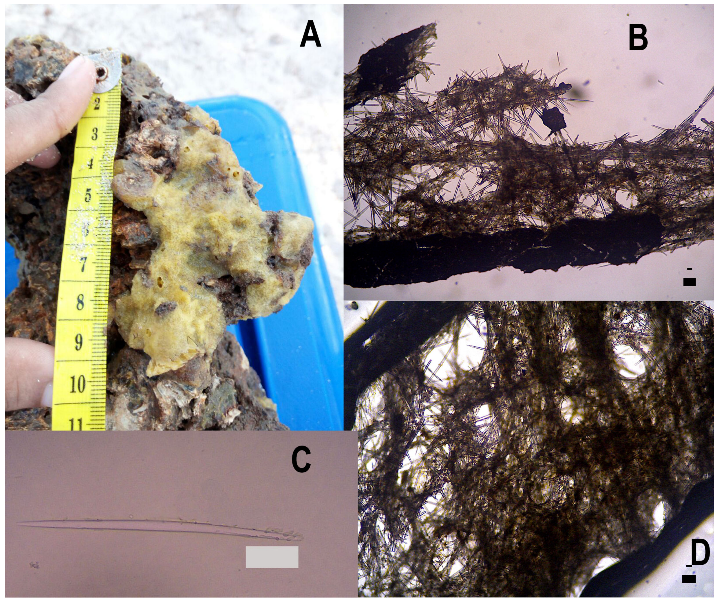

Figure 12 A, "Lifeform", sample 2017_SEP_T2, B, longitudinal or perpendicular section with 40x magnification, C, oxeas skeleton with 400x magnification and D, cross or tangential sections 40x magnification. Scale B, C and D =100 µm

There are two specimens, 2017_SEP_B10 (Figure 13) and 2017_SEP_B13 (figure 14) that only can be identified until order level. The two specimens have a different morphology feature, even though both possess encrusting. The first specimen in the unidentified group is brown, while the second specimen is white. The similarity of structure in choanosomes, which possessed a compact and shaped strands spongin tissue causes us to predict that two specimens belong to Dictyoceratida order

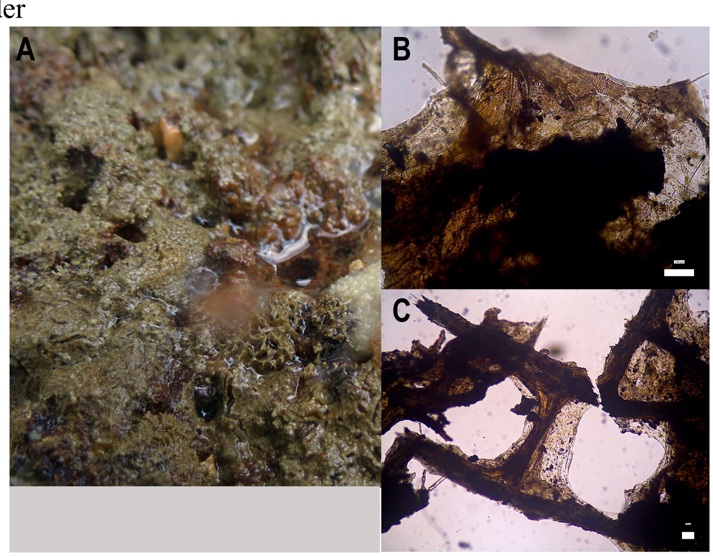

Figure 13 A, "Lifeform", sample 2017_SEP_B10, B, longitudinal or perpendicular section with 40x magnification, and C cross or tangential sections 40x magnification. Scale B and C =100 µm

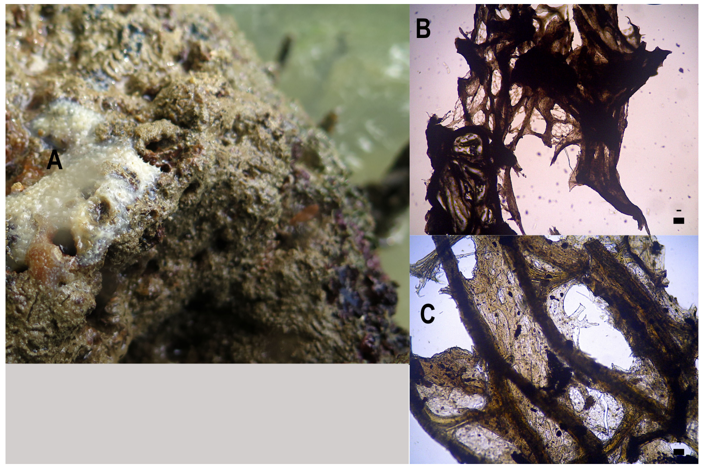

Figure 14 A, "Lifeform", sample 2017_SEP_B103, B, longitudinal or perpendicular section with 40x magnification, and C cross or tangential sections 40x magnification. Scale B and C =100 µm

IV. Discussion

The diversity of sponges between two location are different. *Sonneratia* mangrove area in the western part (W) associated with sponges of genera *Spongia, Dysidea, Lendelfedia, Dactylospongia, Cynachyrella, Haliclona* and one genus that has not been identified. On the other hand, *Rhizophora* mangrove area in the east (E) only associated to one type of genus, Spongia. The difference number of species this study might also be influenced by substrate *Sonneratia* and *Rhizopora* areas. *Sonneratia* substrate consists of sand and mud, whereas *Rhizopora* inhabit rubble coral So far, it is only known that epibion attached to the roots of Rhizopora is generally barnacles from the group of Crustaceans and shellfish from the Bivalve group. Therefore, further research is needed on the effect of substrate type and mangrove root morphology on the diversity of epibiont sponges in mangroves.

Genus *Cynachyrella* from the Tetillidae family recently found in this area was not found in previous sponge mangrove studies in Situbondo, East Java (Alivy et al. 2016), Bangka Island, North Sulawesi (Calcinai et al. 2016) and Derawan, East Kalimantan (Becking et al. 2013). Meanwhile, *Spongia* genus from Spongiidae family, *Dysidea* genus from Dysidae family, *Lendelfedia* genus, and *Dactylospongia* from Thorectidae family, and *Haliclona* genus from Chalinidae family were found well in the three previous research locations in East Java, East Kalimantan, and North Sulawesi. In this study, only five families of six genera were found because of coverage area is around 140m2, that is equal to coverage area (Alivy et al. 2016), in whichh 4 families, Biemnidae, Clionaide, Chalinidae, Dysididae are discovered. In broader a coverage area of 16,022 m2 10 sponge species from 7 families: Ancorinidae, Biemnidae, Chalinidae, Microcianidae, Niphatidae, Tethyidae and Thorectidae were found in North Sulawesi (Calcinai et al. 2016). Moreover, with a broadere coverage areas in East Kalimantan, 199,000.00 m2, 79 species of sponges from 27 families were found: Acarnidae, Ancorinidae, Biemnidae, Callyspongiidae, Chalinidae, Chondrillidae, Choelosphaeridae, Clathrinidae, Clionaidae, Darwinellidae, Dictyodendrillidae, Dictyonellidae, Dysideidae, Dysideidae , Geodiidae, Halichondriidae, Heteroxyidae, Ianthellidae, Microcionidae, Niphatidae, Petrosiidae, Placospongiidae, Spirastrellidae, Suberitidae, Tedaniidae, Tetillidae, Tethyidae and Thorectidae (Becking et al. 2013).

Other factors that influence less diversity of epibion sponges in mangroves in Indonesia or the Indo-Pacific compared to mangrove sponge studies in the Latin American Caribbean are extreme tidal differences that occur in the Indo-Pacific mangrove area compared to Caribbean waters (Maldonado et al 2015). As an example, in the study of Alivy et al. 2016, tides in the mangrove area of Tampora have a depth gradient reaching 1.1 m with a tide time of 5 hours. Other physical factors associated with tidal gradients in the Indo-Pacific region are, direct sunlight exposure also affects the growth of sponges. If the sponge is exposed to the sun's heat for too long, then the water that has been trapped in the body of the sponge will evaporate. Moreover, in Caribbean waters, in addition to low tidal gradient factor, less than 0.5 m, some sponge species in Caribbean have the ability to adapt to extreme salinity, temperature and sedimentary conditions (Calcinai et al. 2016).

Identification at the species level is still difficult because sponges have very minimal morphological features and habitat influences or "phenotypic plasticity" (Bavestrello et al. 1993; Bell & Barnes 2000; Frøhlich & Barthel 1997). This impact on morphology characters become" plastic" and changes according to habitat. Therefore, molecular identification with DNA barcoding aid are needed (Worheide & Erpenbeck 2007; Wörheide et al. 2007).

Table 1. List of Family taxa that found in this study compared to others previous study

Present family taxa	This study with 140 m^2 coverage area	Alivy et al., 2016 with 140m^2 coverage area	Calcinai et al. 2016 with 16,022 m^2 coverage area	Becking et al. 2013 with 199,000.00 m^2 coverage area
Acarnidae	-	-		√
Ancorinidae	-	-	√	√
Biemnidae	-	√	√	√
Callyspongiidae	-	-		√
Chalinidae	√	√	√	√
Chondrillidae	-	-	-	√
Choelosphaeridae	-	-	-	√
Clathrinidae	-	-	-	√
Clionaidae	-	√	-	√
Darwinellidae	-	-	-	√
Dictyodendrillidae	-	-	-	√
Dictyonellidae	-	-	-	√
Dysideidae	√	√	-	√
Geodiidae	-	-	-	√
Halichondriidae	-	-	-	√
Heteroxyidae	-	-	-	√
Ianthellidae	-	-	-	√
Microcionidae	-	-	√	√
Niphatidae	-	-	√	√
Petrosiidae	-	-	-	√
Placospongiidae	-	-	-	√
Spirastrellidae	-	-	-	√
Spongiidae	√	-	-	-
Suberitidae	-	-	-	√
Tedaniidae	-	-	-	√
Tetillidae	√	-	-	√
Tethyidae	-	-	√	√
Thorectidae	√	-	√	√

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