Morphology of *Codium arenicola* M.E.Chacana & P.C.Silva (Codiaceae, Ulvophyceae) from the Ogasawara Islands, Japan

Taiju Kitayama

Department of Botany, National Museum of Nature and Science, 4–1–1 Amakubo, Tsukuba, Ibaraki 305–0005, Japan E-mail: kitayama@kahaku.go.jp

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Abstract A benthic marine green alga, *Codium arenicola* M.E.Chacana & P.C.Silva (Codiaceae, Bryopsidales, Ulvophyceae) was collected from the mesophotic zone (46–54 m in depth) off Chichijima Island and Nishijima Island, Ogasawara Islands, Japan. This alga is different from the other species of the genus *Codium* in having an erect, terete (1–5 mm in diameter), sparsely dichotomous branches, a holdfast with sand-binding rhizoidal filaments, and cylindrical to obovate or obconical utricles (105–430 µm in diameter, 294–588 µm in length). Scatter plot of diameter versus length in the utricles revealed a wide variation in shapes of the utricles is caused by different developmental stages. It is suggested that the shape of the utricles of *C. arenicola* is more various than illustrated and stated as "claviform to muffin-shaped" in the original description.

Key words: benthic marine green alga, Codiaceae, *Codium arenicola*, Japan, Ogasawara Islands, Ulvophyceae.

The marine green algal genus Codium (Codiaceae, Bryopsidales, Ulvophyceae), which was established by Stackhouse (1797), has 120-144 species in the world (Huisman et al., 2015; Guiry and Guiry, 2020). Although the thalli of this genus are composed of unicellular siphons (multinucleate tubular units), they are very various in the external form ranging from unbranched semispherical thalli to erect branched cylinders. In Japan, 21 species of Codium have been recorded (Chacana and Silva, 2014; Yoshida et al., 2015), of which seven are composed of erect, terete, dichotomously branched thalli: C. arenicola M.E.Chacana & P.C.Silva, C. barbatum Okamura, C. contractum Kjellman, C. cylindricum Holmes, C. fragile (Suringar) Hariot, C. subtubulosum Okamura, and C. yezoense (Tokida) K.L.Vinogradova. These Japanese species are distinguishable from each other by their external forms and microscopic shapes of utricles.

The Ogasawara Islands (also called the Bonin Islands) are oceanic islands located ca. 1,000 km south of the Izu Peninsula, central Japan. In recent investigations of algal flora around the islands, several rare algae were found from the mesophotic zone or middle light zone (30-150 m in depth) using a dredging aparatus: for example, mesophotic marine algae, Discosporangia mesarthrocarpum (Meneghini) Hauck (Kitayama, 2012), Zosterocarpus ogasawaraensis Kitayama (Kitayama, 2013), Aneurianna ogasawaraensis Kitayama (Kitayama, 2014), Codium mamillosum Harvey (Kitayama, 2017), Lychaete bainesii (F.Müller & Harvey ex Harvey) M.J.Wynne (Kitayama, 2019). The presence of these algal species suggested that the Ogasawara Islands has a unique marine algal flora different from the ones of the Japanese Archipelago. In this research, Codium arenicola M.E.Chacana & P.C.Silva, which is new to the Ogasawara Islands, is observed in morphology using materials collected from the mesophotic zones off the

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Ogasawara Islands, for unraveling the algal flora of this sea area.

Materials and Methods

The green algal materials referred to as *Codium arenicola* M.E.Chacana & P.C.Silva were collected from the mesophotic zone (46–54 m in depth) off Chichijima Island and Nishijima Island in the Ogasawara Islands, Japan by a dredge using the research vessel "*The Koyo*" (87 tonnage) on 8 July 2010 and 13 July 2016, and the fishing vessel "*The 7th Ushiwo-maru*" (6.7 tonnage) on 17 July 2017. For preservation, the material was fixed and stored in 10% formalin-seawater, and specimens were mounted in glycerin jelly. Anatomical observations were made on the material by a microscope. Voucher specimens were deposited in the herbarium of National Museum of Nature and Science (TNS).

Results

Class Ulvophyceae K.R.Mattox & K.D.Stewart, 1984 Order Bryopsidales J.H.Schaffner, 1922 Family Codiaceae Kützing, 1843

Codium arenicola M.E.Chacana & P.C.Silva Nova Hedwigia 98: 249, figs. 1–3 (2014).

[Figs. 1–18]

[Type locality: Indonesia: Sulawesi: E side, Kudingareng Keke Island (Chacana and Silva, 2014: 249)]

Habitat: Grew on sandy bottoms in the mesophotic zone (46–54 m in depth).

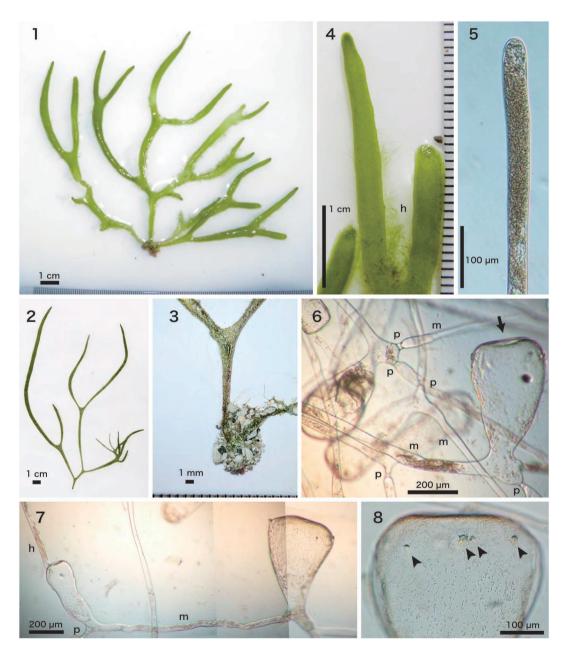
Morphology: Thalli are light green, erect, up to 23 cm in height, with a cuneate stipe (Figs. 1–3). Holdfast attached to sandy bottom by sandbinding rhizoidal filaments (Fig. 3). Branching is sparse with elongate internodes (Figs. 1–2), irregularly dichotomous or sub-dichotomous, and up to 4 orders. Branches are mostly terete, 1–5 mm in width, becoming slightly compressed at dichotomies. Hairs grow from the surface of branches (Fig. 4), green, terete, 20–52 μ m in diameter, elongated, up to several millimeters, with a round apex (Fig. 5). The thalli are composed of numerous medullary filaments and individual utricles. Medullary filaments are colorless, siphonous, elongated, and 18-64 µm in diameter (Fig. 6). Utricles are connected to other ones by 2-4 medullary filaments issued from the basal portion (Fig. 7). Apices of the utricles are rounded or flattened, often depressed, with thin walls (Fig. 6). Hair scars are abundant on the upper portion of utricles (Fig. 8). The shape of the utricles varies depending on the development (Figs. 9-17). Young utricles are cylindrical to clavate, (78-)105-194 µm in diameter, (228-) 294-560 µm in length. Mature utricles are clavate to obovate or obconical, 220-430(-486) µm in diameter, 382-588(-690) µm in length, tapering toward the base (Figs. 13-17). Plugs inside medullary filaments are located beneath the utricles, 1–3 per utricle (Figs. 9–17).

Specimens examined: The Ogasawara Islands, Japan: Off Kurazonone (53.5–53.9 m), Chichijima Island (27°30'55"N, 142°6'20"E), 17 July 2017, leg. T. Kitayama (TNS-AL 213459) (Fig. 1); Off Nishijima Island (47.9–52.3 m) (27°7'27"N, 142°10'26"E), 13 July 2016, leg. T. Kitayama (TNS-AL 213460) (Fig. 2). Off Nishijima Island (46.2–50.8 m) (27°7'14"N, 142°10'42"E), 8 July 2010, leg. T. Kitayama (TNS-AL 213461) (Fig. 3).

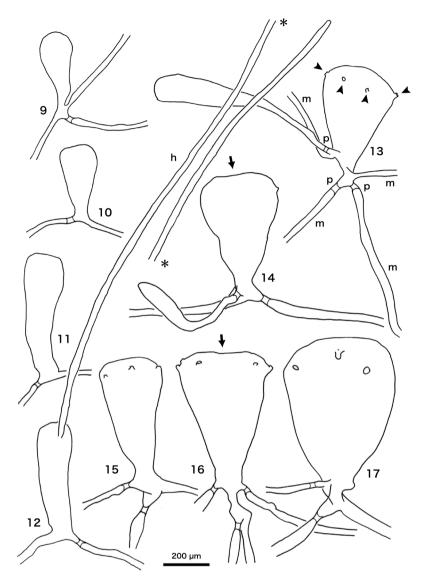
Japanese name: Otohime-miru (nov.).

Distribution: *The Indian Ocean*: Indonesia: Sulawesi (type locality); Komodo Island; Samaloa Island; Barang Lompo Island; Western Australia: Dampier Archipelago; *The Pacific Ocean*: Thailand: Songkhla; New Caledonia: Balabio Island; Philippines: Sulu; Japan: Oki Islands; Tanegashima Island; Amami Island (Chacana and Silva, 2014); the Ogasawara Islands (the present study).

A scatter plot with the variations in diameter and the variations in length in the utricles of *Codium arenicola* M.E.Chacana & P.C.Silva is shown in Fig. 18. The morphology, the habitat and distribution in the six species of *Codium* having erect, terete, dichotomously branched



Figs. 1–8. Codium arenicola M.E.Chacana & P.C.Silva from Ogasawara Islands, Japan. 1. Living thallus from Chichijima Island (TNS-AL 213459). 2. Herbarium specimen from Nishijima Island, having considerable sparse ramification (TNS-AL 213460). 3. Herbarium specimen from Nishijima Island, showing the holdfast with sand-binding rhizoidal filaments (TNS-AL 213461). 4. Apical portion of thallus with many hairs (h). 5. Apex of hair. 6. Utricles with medullary filaments (m) and plugs (p). Arrow shows a depression on the apex of utricle. 7. Young utricle and mature utricle connecting with a medullary filament (m) and hair (h) protruding from the apex of the utricle. 8. Apex of utricle with hair scars (arrowheads).



Figs. 9–17. Various shapes of utricles of *Codium arenicola* M.E.Chacana & P.C.Silva from Ogasawara Islands depending on the developmental stages. 9–11. Young utricles. 12. Young utricle with a long hair (h). 13. Mature utricle with hair scares (arrowheads), medullary filaments (m) and plugs (p). 14. Mature utricle showing a depression on the apex (arrow). 15–17. Mature utricles. Arrow shows a depression on the apex.

thalli and bearing cylindrical to conical utricles are shown in Table 1.

Discussion

The present alga collected from the mesophotic zone off the Ogasawara Islands resembles *Codium arenicola* M.E.Chacana & P.C.Silva (Chacana and Silva, 2014), *C. barbatum* Okamura (Okamura, 1930), *C. campanulatum* P.C.Silva & M.E.Chacana (Abbott and Huisman, 2004; Silva and Chacana, 2014), *Codium cicatrix* P.C.Silva (Silva, 1959), *C. profundum* P.C.Silva & M.E.Chacana (Oliveira *et al.*, 2010; Silva and Chacana, 2010) and *C. tenue* (Kützing) Kützing (Kützing, 1856; Silva, 1959; Huisman *et al.*,

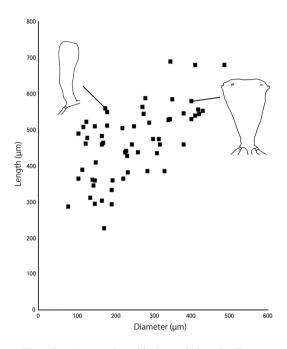


Fig. 18. Scatter plot with the variations in diameter and the variations in length in the utricles of *Codium arenicola* M.E.Chacana & P.C.Silva from Chichijima Island, Ogasawara Islands, Japan.

2015) in appearance of thalli and/or shape of utricles (Table 1). The commonality that these species share is having erect, terete, dichotomously branched thalli composed of cylindrical to obconical utricules.

In particular, the present alga relates most closely with Codium arenicola, which occurs widely in the Indo-Pacific Ocean, in having sparsely ramified branches (up to 4 orders) with long internodes and a holdfast with sand-binding rhizoids. The width of branches of the present alga may be close to ones of C. arenicola. Although branches of C. arenicola were written "Internodes terete or slightly flattened, as 6-10 mm broad, nodes flattened, cuneate, to 2 cm broad" in the original description (Chacana and Silva, 2014, fig. 2), the ones in the photograph of the holotype specimen were 1-5 mm in width. The present alga has obovate or obconical utricles in mature, while only "stout claviform to muffin-shaped" utricles were observed in the

original description of *C. arenicola* (Chacana and Silva, 2014, fig. 3). In this research, however, it was revealed that the shape of the utricles varies depending on the developmental stages (Figs. 7, 9–18). For such an alga there is a danger that partial observation on utricles may underestimate the morphological variation depending on development. In addition, the present alga agrees to the original description of *C. arenicola* in abundance of depressed apices of utricles, hairs (or hair scars), light green fronds, and "deep-water" habitat (Table 1). *C. arenicola* was recorded from the sandy bottoms at depth of 20–30 m (Chacana and Silva, 2014).

Before the definition of *Codium arenicola* by Chacana and Silva (2014), there are confusions with the other species with swollen utricles. The alga recorded as *C. arenicola* from Samoan Archipelago, South Pacific by Skelton and South (2007) is not conspecific with the genuine *C. arenicola* in having dense ramification of compressed branches, which forms often palmate fronds (figs. 702–703). Verbruggen and Costa (2015) has observed *C. arenicola* at the southern Madagascar, but it does not correspond with the species in possessing the dense ramification of compressed branches.

Chacana and Silva (2014) noted the similarity in the shape of utricles between *Codium arenicola* and the two other "deep-water" species, *C. campanulatum* (endemic to Hawaii) and *C. profundum* (Atlantic species), though they have thinner branches than the ones of *C. arenicola* and the present alga from the Ogasawara Islands: 1.5–2.3 mm in *C. campanulatum* (Abbott and Huisman, 2004) and less than 3 mm in *C. profundum* (Silva and Chacana, 2010) (Table 1).

C. tenue, which is recorded from the Indian Ocean, grows in shallow waters of estuaries and has abundant ramification with thick utricles: $(135-)200-700 \ \mu\text{m}$ in diameter (Silva, 1959) or 200–550 \ \mu\) in diameter (Huisman *et al.*, 2015). According to Chacana and Silva (2014), *C. tenue*, which Børgesen (1948) reported from Mauritius, is only *C. arenicola*. Also *C. cicatrix*, which is widely distributed in the Indian Ocean,

	Ex	External form			Utricles			
Taxon	Branching order	Height (cm)	Width (mm)	Form	Diameter (µm)	Length (µm)	Depth and substrata	Distribution
<i>Codium arenicola</i> M.E. Chacana & P.C.Silva, 2014	up to 4*	"up to 40" ca. 20*	"6-10" 1-5*	stoutly claviform to muffin-shaped	(180–)200–250(–330) (500–)600–780(–800)	(500-)600-780(-800)	20–30 m, sandy bottoms	Pacific Ocean: Indonesia (type locality), Japan, Philippines, Thailand Indian Ocean: Indonesia, Western Australia
<i>C. arenicola</i> M.E. Chacana & P.C.Silva (the present study)	up to 4	up to 23	1-5	cylindrical, clavate, obovate, obconical	(78-)105-430(-486)	(78–)105–430(–486) (228–)294–588(–680)	46–54 m, sandy bottoms	Pacific Ocean: Japan (Ogasawara Isls.)
C. barbatum Okamura, 1930	up to 10^{**} up to 10	up to 10	up to 3	cuboid, obovate, oblong	200–300	460–483	cast up ashore	Pacific Ocean: Japan, Korea
C. campanulatum P.C. Silva & M.E. Chacana (Abbott and Huisman, 2004)	up to ca. 8	12–40	1.5–2.3	stoutly cylindrical, campanulate, turbunate, pyriform, obovate, orbicular, quadrate	120-125(-360)	310-440	56-185 m	Pacific Ocean: Hawaii
<i>C. cicatrix</i> P.C.Silva, 1959	up to 9	up to 19	(2-)4-5	cylindrical, clavate	(145-)235-390	630–920	Epiphytic on <i>Cvmodocea ciliata</i>	Indian Ocean: Mozam- bique
<i>C. profundum</i> P.C.Silva & M.E.Chacana, 2010	up to 6	up to 10(-15)	up to 3	subcylindrical to pyriform, goblet- shaped, bell-shaped, muffin-shaped	(175–)200–350	350–550	up to 70 m, calcareous rubble or crustose corallines	Atlantic Ocean: Bermuda, Brazil, Canary Is., Florida, Louisiana
C. tenue (Kützing) Kützing (Silva, 1959)	up to 15	up to 45	1-5(-22)	clavate, pyriform, turbinate, quadrata, obovate, orbicular	(135–)200–700	(330–)500–800	Estuaries and lagoons, silt-covered rocks, shells, jetty piles	Indian Ocean: South Africa (Knysna to Port St. Johns)

*) Estimated from the fig. 2 in Chacana and Sliva (2014). **) Estimated from the pl. 10, f. 1 in Okamura (1930).

Table 1. Comparison of the morphology, the habitat and distribution among the species of erect, terete, dichotomously branched Codium bearing cylindrical to obconi-

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has similarity to the present alga, though the species in the original description (Silva, 1959) is different from the present alga in ramification of branches (up to 9 orders), length of utricles (up to 920 µm), and habitat (epiphytic on a seagrass, *Cymodocea ciliata* Forsskål [now *Thalassodendron ciliatum* (Forsskål) Hartog]]. On the other hand, Egerod (1974) reported "*C. cicatrix*" from Thailand (Andaman Sea), though it is not con-

able to C. barbatum and thus C. tenue is restricted to South African estuaries. As a result, the author identify the present alga specific with the species but is similar to C. areas Codium arenicola M.E.Chacana & P.C.Silva, nicola in the habitat and habit. Verbruggen and which has thin branches with sparse ramification Costa (2015) also recorded "C. cicatrix" from and cylindrical to obconical, obovate utricles. It Madagascar, which is mixed with two different is suggested that this species has a wide distribuspecies (figs. 8, 9). Both thalli do not correspond tion in mesophotic zone (20-60m in depth) in with C. cicatrix and the thallus shown in fig. 8 in the Indo-Pacific Ocean. To clarify the whole dis-Verbruggen and Costa (2015) is close to C. aretribution of this species, anatomical observations nicola in habit. Moreover, C. cicatrix recorded by and molecular analyses on more samples from Van den Heede and Coppejans (1996) is not simivarious fields and more specimens in various lar to both of C. arenicola and C. cicatrix in the herbaria are required. Presently, it is clear that small compressed thalli. Codium barbatum Okaseven species of Codium having erect, terete, mura, whose type locality (Hachijo Island) is close dichotomously branched thalli are distributed throughout Japan, and their "key to species" is to the Ogasawara Islands, bears "cuboid or obovate or oblong, short" utricles (Okamura 1930). This shown as follows.

Key to the species of Codium having erect, terete, dichotomously branched thalli in Japan

1a. Thallus growing on sandy bottoms	C. arenicola
1b. Thallus growing on rocks or boulders	2
2a. Thallus shorter, usually 10-30 cm in height	3
2b. Thallus longer, 0.3–1(–15) m in height, flattened at dichotomies	4
3a. Apices of branches clavate	C. contractum
3b. Apices of branches tapering	5
4a. Utricles 100–140 μm in diameter, 660–850 μm in length	C. subtubulosum
4b. Utricles 400–500(–900) μm in diameter, 2–3 mm in length	C. cylindricum
5a. Utricles with an acute apices	C. fragile
5b. Utricles with rounded or flattened apices	6
6a. Utricles cylindrical to clavate	C. yezoense
6b. Utricles cuboid to obovate	C. barbatum

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