## Taxonomic study of two *Sargassum* species (Fucales, Phaeophyceae) from the Ryukyu Islands, southern Japan: *Sargassum ryukyuense* sp. nov. and *Sargassum pinnatifidum* Harvey

### Hiromori Shimabukuro<sup>1</sup>, Ryuta Terada<sup>2,\*</sup>, Tadahide Noro<sup>1</sup> and Tadao Yoshida<sup>3</sup>

 <sup>1</sup> Education and Research Center for Marine Resources and Environment, Faculty of Fisheries, Kagoshima University, 4-50-20 Shimoarata, Kagoshima City, Kagoshima 890-0056, Japan
<sup>2</sup> Division of Fisheries Biology and Oceanography, Faculty of Fisheries, Kagoshima University, 4-50-20 Shimoarata, Kagoshima City, Kagoshima 890-0056, Japan, e-mail: terada@fish.kagoshima-u.ac.jp
<sup>3</sup> Suzaku 6-13-13, Dazaifu, Fukuoka 818-0103, Japan

\* Corresponding author

### Abstract

Species of the genus Sargassum (Fucales, Phaeophyceae) belonging to the Schizophycus group (in subgenus Sargassum sensu lato) from the Ryukyu Islands, southern Japan, were morphologically reinvestigated, and two entities were recognized: Sargassum ryukyuense sp. nov. and Sargassum pinnatifidum. The new species is characterized by: 1) compressed primary branches; 2) simple lanceolate primary and lateral leaves with an undulate and dentate margin in all stages; 3) lateral branches issuing alternate-distichously on the main branches in one plane; 4) long and flattened receptacles arranged alternate-distichously on terminal branches; 5) main and lateral branches on the stem issuing as foliar expansions in the early stage of development; and 6) vesicles with a simple coronal leaf. This new species resembles S. pinnatifidum in features 1), 3) and 5). However, it differs from S. pinnatifidum in features 2), 4) and 6). Furthermore, this new species can be distinguished from all previously described species based on the complete set of diagnostic features above and by the presence of long lingulate receptacles with short stalks. The new species is known to be endemic in the Ryukyu Islands and adjacent waters of southern Japan.

**Keywords:** Okinawa; Phaeophyceae; *Sargassum pinnatifidum*; *Sargassum ryukyuense*; taxonomy.

### Introduction

The genus *Sargassum* C. Agardh 1820 (Fucales, Phaeophyceae) is distributed widely in tropical and temperate regions of the world and has over 400 recognized species (Phillips and Fredericq 2000). The bulk of the species are distributed in the northern and southern regions of the Pacific basin, with the center of diversity in the Indian Ocean, Southeast (SE) Asia and Australia (Nizamuddin 1961, 1970). Furthermore, approximately 140 species are distributed in the subtropical and temperate zones of East Asian regions such as Japan, China and the Philippines (Silva et al. 1996, Tseng and Lu 2000).

In Japan, the taxonomy of *Sargassum* species is well understood through eminent works that focused on the species of Japan proper (Yendo 1907, Okamura 1936, Yamada 1942, Yoshida 1983, 1998). According to Yoshida et al. (2005), approximately 60 species have been recognized in this region. Yoshida (1998) also reported 13 species of *Sargassum* from the Ryukyu Islands that lie between the southern part of Kyushu Island (Japan proper) and Taiwan Island.

In the past 15 years, there have been four reports on Sargassum species found in the Ryukyu Archipelago that clarify specific characters (Ajisaka 1992, Noro et al. 1994, Ajisaka et al. 1995, 1997). For example, Ajisaka (1992) and Ajisaka et al. (1994) described the characteristic features of Sargassum polyporum Montagne. Noro et al. (1994) compared Sargassum oligocystum Montagne with two Australian Sargassum species [S. ligulatum J. Agardh and S. swartzii (Turner) C. Agardh] and a Sargassum species from Hawaii (S. echinocarpum J. Agardh). Ajisaka et al. (1995) reported ten species of Sargassum in the subgenus Sargassum (S. bulbiferum Yoshida, S. carpophyllum J. Agardh, S. crispifolium Yamada, S. glaucescens J. Agardh, S. incanum Grunow, S. longifructum Tseng et Lu, S. myriocystum J. Agardh, S. polycystum C. Agardh, and two unidentified species) from the Ryukyu Archipelago and adjacent waters.

However, information on these species in the region is incomplete, especially for some subtropical *Sargassum* species of the subgenus *Sargassum* (*sensu lato*) (Yoshida et al. 2004).

Five subgenera, *Phyllotrichia*, *Schizophycus*, *Bactrophycus*, *Arthrophycus*, and *Sargassum*, have been recognized within *Sargassum* (Agardh 1889, Setchell 1936, Yoshida 1984). According to the definition of Agardh (1889), *Phyllotrichia* and *Schizophycus* develop their main branch as "foliar expansions". The vesicles of *Phyllotrichia* are round without a coronal leaf, while species of *Schizophycus* have vesicles with a simple or divided coronal leaf. Womersley (1954) pointed out that vesicle characteristics alone are insufficient to discriminate the subgenera, and subsumed *Schizophycus* into *Phyllotrichia*. Yoshida et al. (2004) concluded that the subgenus *Schizophycus* should be merged with the subgenus *Sargassum* on the basis of molecular phylogenic analysis

using the ITS-2 region. Yoshida et al. (2004) also reassessed four "*Schizophycus*" species from Japan proper and the Ryukyu Islands, and reconfirmed the presence of *Sargassum pinnatifidum* Harvey from Okinawa (within the Ryukyus). This species was described by Harvey (1859) on the basis of a specimen collected by Charles Wright from "Loo Choo Island" (Ryukyu Island) without other information. Although Dawson (1959) reviewed the unpublished comments of Harvey (1859), the taxonomic status of this species was uncertain until the work of Yoshida et al. (2004).

Recently, we collected *Schizophycus* species, including *S. pinnatifidum*, off the Ryukyu Islands and in adjacent waters, and concluded that there are two entities of *Schizophycus* within the Ryukyus. In this paper, we give detailed descriptions of the new species *Sargassum ryukyuense* and of *S. pinnatifidum* (which has been confused with the *S. ryukyuense* in Okinawa).

### Materials and methods

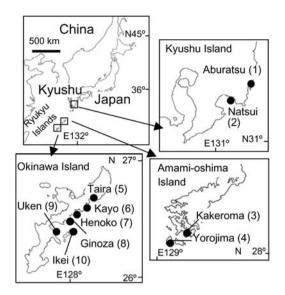
Specimens examined (*leg.* Shimabukuro unless otherwise indicated) were collected from ten locations in southern Japan. Southern part of Kyushu Island: 1) Aburatsu (31°34′ N, 131°24′ E), Nichinan City, Miyazaki Prefecture; 2) Natsui (31°24′ N, 131°8′ E), Shibushi City, Kagoshima Prefecture. Amami-oshima Island: 3) Kakeroma (28°6′ N, 129°17′ E) and 4) Yorojima (28°2′ N, 129°9′ E), Setouchi Town, both in Kagoshima Prefecture. Okinawa Island: 5) Taira (26°37′ N, 128°8′ E), Higashi Village; 6) Kayo (26°32′ N, 128°6′ E); 7) Henoko (26°31′ N, 128°1′ E), Nago City; 8) Ginoza (26°27′ N, 127°58′ E), Ginoza Village; 9) Uken (26°22′ N, 127°52′ E); and 10) Ikei (26°23′ N, 128°0′ E), Uruma City, all in Okinawa Prefecture (Figure 1).

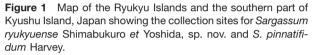
Voucher herbarium specimens were deposited in the following herbaria listed in the *Index Herbariorum* (http://sciweb.nybg.org/science2/IndexHerbariorum.asp): SAP (Hokkaido University); TNS (National Museum of Nature and Science, formerly the National Science Museum of Tokyo); and KAG (Kagoshima University Museum). Specimens fixed in 10% formalin/seawater were used for microscopic observations. Sections were prepared by hand and stained with 1% cotton blue in 50% glycerol/seawater.

### Results

# *Sargassum ryukyuense* Shimabukuro *et* Yoshida sp. nov. (Figures 2–11)

**Diagnosis** Thallus usque ad 70 cm altus, haptero discoideo vel parum conico, 10–30 mm diametro. Caulis teres, brevis, 2–5 mm longus. Rami primarii ex parte apicali caulis spiraliter exeuntes, late compressi, 4–6 mm lati. Rami laterales complanati margine integro, usque ad 250 mm longi, 3 mm lati. Folia ramorum primariorum lanceolata, margine undulato dentato, folia ramorum lateralium simplicia, lineari-lanceolata, ad apice acuta, usque ad 50 mm longa, 8 mm lata. Cryptostomata dispersa.





(1) Aburatsu, Nichinan City; (2) Natsui, Shibushi City; (3) Kakeroma Island; (4) Yorojima Island; (5) Taira, Higashi Village; (6) Kayo; (7) Henoko, Nago City; (8) Ginoza, Ginoza Village; (9) Uken; and (10) Ikei, Uruma City.

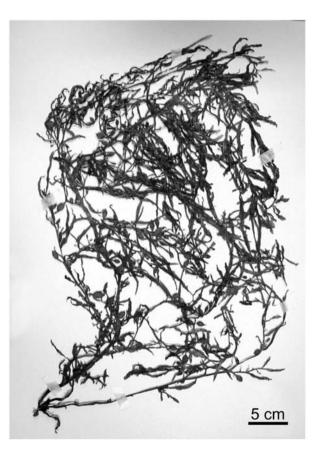
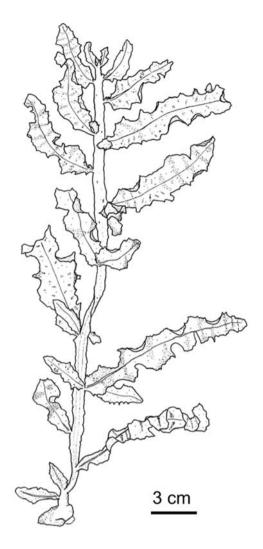


Figure 2 Holotype specimen of *Sargassum ryukyuense* Shimabukuro *et* Yoshida, sp. nov (SAP 101916, Ikei, Uruma City, Okinawa, Japan, September 10, 2002).

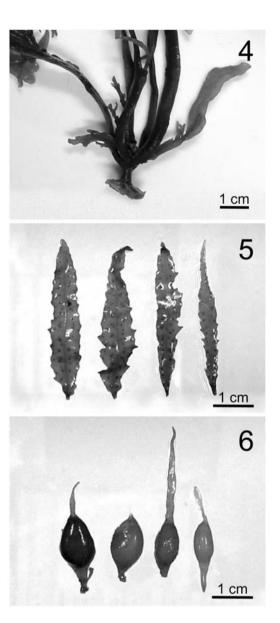


**Figure 3** Young stage of *Sargassum ryukyuense* Shimabukuro *et* Yoshida, sp. nov. showing leaves issued alternate-distichously on the main branch in one plane.

Vesiculae ellipticae vel ovoideae, usque ad 13 mm longae, 8 mm latae, breviter mucronatae vel folio margine dentato terminatae. Planta dioica. Receptacula in ramis terminalibus disposita. Receptacula masculina linearicompressa, lingulata, usque ad 25 mm longa, 4 mm lata. Receptacula foeminea lineari-compressa, lingulata, usque ad 10 mm longa, 3 mm lata. Conceptacula 200–300 µm diametro.

**Type specimens** Holotype: *SAP 101916*, Ikei, Uruma City, Okinawa Prefecture (September 10, 2002); isotypes: *SAP 101917–101919, TNS-AL 161080–161082, KAG-F 1757–1759*.

Thallus up to 70 cm tall; holdfast discoid to slightly conical, 10–30 mm in diameter; stem terete arising from center of holdfast, short, 2–5 mm long; primary branches issued spirally from apical part of the stem; primary branches compressed, 4–6 mm wide with entire margin. Lateral branches compressed and with smooth margins, issued distichously from primary branch at intervals of 10–40 mm in one plane, up to 250 mm long, 3 mm wide. Leaves of primary branches lanceolate, with obtuse or acute apex, undulate dentate margin, deciduous; leaves on lateral branches simple, acute apex, linear-lanceolate,

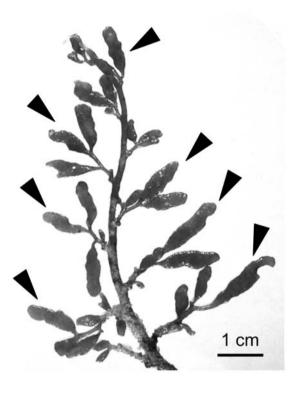


Figures 4–6 Sargassum ryukyuense Shimabukuro et Yoshida, sp. nov. (Ikei, Uruma City, Okinawa, Japan, September 10, 2002).

(4) Lower part of the thallus showing the holdfast, stem and some primary branches. (5) Leaves of various shapes. (6) Vesicles with coronal leaves.

up to 50 mm long and 8 mm wide; midrib diminishing up to apex; cryptostomata distinct and scattered on both sides of the midrib; margin irregularly dentate or smooth. Vesicles variable in shape, elliptical to ovoid, up to 13 mm long and 8 mm wide, with flattened stalk shorter than vesicle, terminated by a short mucro or a simple leaf with dentate margin; cryptostomata generally present. Short medullary filaments often present inside vesicles.

Thallus dioecious. Receptacles arranged alternate-distichously on the terminal part of branches; male receptacles simple linear, compressed, lingulate, with entire margins, 5–25 mm long and 2–4 mm wide, with short stalks; female receptacles simple linear, compressed, lingulate, with entire or small dentate margins, 3–10 mm long and 2–3 mm wide, with short stalks. Conceptacles scattered on receptacles, and 200–300  $\mu$ m in diameter. Oogonia 60–120  $\mu$ m in diameter.



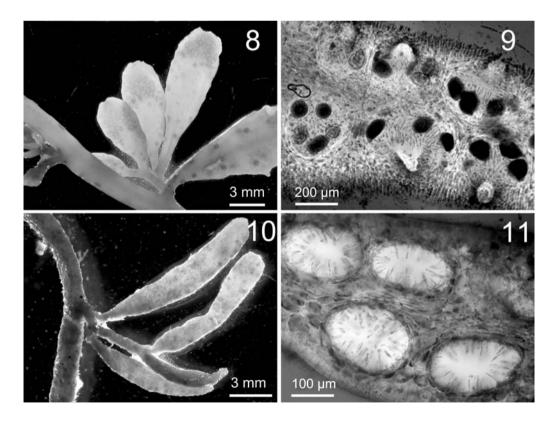
**Figure 7** Upper portion of *Sargassum ryukyuense* Shimabukuro *et* Yoshida, sp. nov. (Ikei, Uruma City, Okinawa, Japan, September 10, 2002) showing male receptacles borne on the terminal branches (arrowheads).

**Etymology** Named after the collection site: Ryukyu Islands, Japan; Japanese name: Churashima-moku (Churashima means beautiful island in the Ryukyu dialect; moku means *Sargassum*).

**Type locality** Ikei, Uruma City, Okinawa Prefecture, Japan.

**Distribution** Ryukyu Islands and Southern Kyushu Island, Japan.

Specimens examined Aburatsu, Nichinan City, Miyazaki Prefecture (August, 1940, leg. Y. Nakamura): SAP (two unnumbered specimens); Natsui, Shibushi City, Kagoshima Prefecture (June 12, 1980, leg. T. Yoshida): SAP (three unnumbered specimens); Kakeroma, Setouchi Town, Kagoshima Prefecture (June 24, 1961, leg. T. Yoshida): SAP (unnumbered specimen); Yoro, Setouchi Town, Kagoshima Prefecture (May, 1954, leg. unnamed): SAP (unnumbered specimen); Taira, Higashi Village, Okinawa Prefecture (March 26, 1962, leg. S. Kamura): SAP (two unnumbered specimens); Henoko, Nago City (July 25, 1989, leg. T. Yoshida): SAP (two unnumbered specimens); Ikei, Uruma City, Okinawa Prefecture (November 6, 2003): SAP 101920; Ikei, Uruma City, Okinawa Prefecture (November 4, 2004): SAP 101921, TNS-AL 161083, 161084; Ginoza, Ginoza Village, Okinawa Prefecture (July 26, 2003): SAP 101922, 101923, TNS-AL 161085, 161086; Ginoza, Ginoza Village, Okinawa Prefecture (June 28, 2004): KAG-F 1760, 1761.



Figures 8–11 Close-up images and transverse sections of receptacles of *Sargassum ryukyuense* Shimabukuro *et* Yoshida sp. nov. (Ikei, Uruma City, Okinawa, Japan, September 10, 2002).

(8) Close-up image of female receptacles showing the lingulate shape. (9) Transverse section of a female receptacle showing conceptacles and oogonia. (10) Close-up image of male receptacles showing the long lanceolate shape. (11) Transverse section of a male receptacle showing conceptacles.

**Habitat and seasonality** This species grows on rock in the lower intertidal to sublittoral zones at 1–3 m in depth. It can be found on the substrata from March through November. The maturation period is September.

# Sargassum pinnatifidum Harvey 1859: 327 (Figures 12–16)

Yendo 1907, Setchell 1931, 1933, Okamura 1936, Dawson 1959, Yoshida 1998, Yoshida et al. 2004.

**Type locality** "Loo Choo Island" (Ryukyu Island), Okinawa Prefecture, Japan.

**Distribution** Ryukyu Islands (Okinawa Islands), Japan.

**Specimens examined** Uken, Uruma City, Okinawa Prefecture (November 6, 2003): *SAP 101924*, *101925*, *TNS-AL 161087*, *161088*, *KAG-F 1762*; Kayo, Nago City, Okinawa Prefecture (November 17, 2003): *SAP 101926*, *TNS-AL 161089*, *KAG-F 1763*.

Thallus up to 50 cm tall; holdfast discoid to conical, 10–20 mm in diameter; stem terete arising from center of holdfast, up to 20 mm long; primary branches issuing spirally from the apical part of stem; primary and lateral branches compressed, up to 4 mm wide, margin thin, smooth or often with spines. Leaves variable in shape, expanded in same plane as branch. Leaves on basal part of thalli simple or pinnately branched, 50–110 mm long and 2–10 mm wide, margin often with sharp spines; dis-

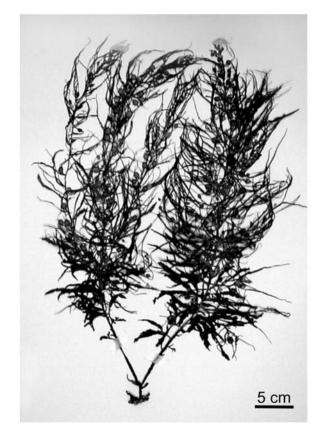


Figure 12 Herbarium specimen of *Sargassum pinnatifidum* Harvey (SAP 101925, Uken, Uruma City, Okinawa, Japan, November 6, 2003).

tinct midrib diminishing near acute apex; cryptostomata scattered on surface. Leaves on upper part of thalli pinnately branched, very narrow, with smooth margins; cryptostomata scattered on surface. Vesicles elliptical, 5–10 mm long and 3–8 mm in diameter, with stalk shorter than vesicle, coronate with simple or pinnately divided leaf, with margin smooth or denticulate. Medullary filaments present inside vesicles.

Thallus dioecious. Receptacles born on terminal branches, alternate-distichously arranged, linear, slightly compressed, with smooth margin; male receptacle 3–4 mm long and 1 mm wide; female receptacle 2 mm long and 1 mm wide. Male and female conceptacles scattered on receptacles, and 150–350  $\mu$ m in diameter. Oogonia 100–150  $\mu$ m in diameter.

**Remarks** The Chinese entity is reported to be monoecious, with male and female conceptacles present in the same receptacles (Tseng and Lu 2000). However, the Japanese entity in this study and as reported by Yoshida et al. (2004) is dioecious.

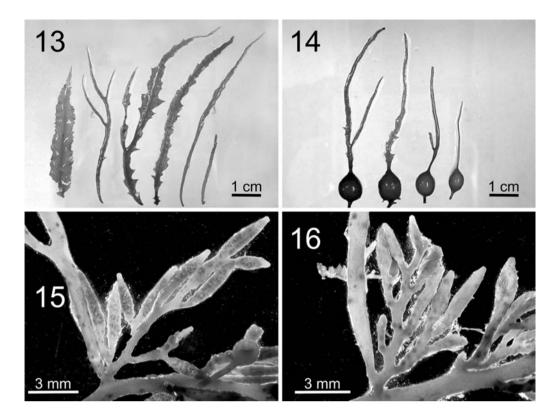
#### Discussion

Yoshida et al. (2004) reconfirmed the presence of *Sargassum pinnatifidum* described by Harvey (1859) for Okinawa Island. Formerly, although this species was placed in the subgenus *Schizophycus*, it was confused with *Sargassum patens* C. Agardh in the Ryukyu Islands. Our results reveal that the "*Schizophycus*" species from the Ryukyu Islands include two entities: *Sargassum pinnatifidum* and a new taxon. We could not find *S. patens* in this area. Our observations of *S. pinnatifidum* agree well with the description of this species by Yoshida et al. (2004).

Our new taxon is distinguished from all known species of subgenus *Sargassum* (sensu lato) from the east and southeast Asian regions by the following set of features, in addition to a diagnostic characteristic receptacle feature: 1) compressed primary branches; 2) simple lanceolate primary and lateral leaves with an undulate and dentate margin in all stages; 3) lateral branches issuing alternate-distichously on the main branches in one plane; and 4) long and flattened receptacles arranged alternate-distichously on terminal branches with short stalks (Figure 7). It also has the following features of "*Schizophycus*" species: 5) main and lateral branches issue from the stem as foliar expansions in the early stage of development (Figure 3); and 6) vesicles have a coronal leaf (Womersley 1954).

Foliar expansions of the branches of our new taxon are similar to those of *Sargassum pinnatifidum*. However, simple leaves with an undulate and dentate margin are entirely different from the narrow and filiform leaves of *S. pinnatifidum* (Table 1).

Although subgenus *Schizophycus* J. Agardh (1889) had been recognized as one of the taxa within the genus *Sargassum*, Yoshida et al. (2004) merged it with the subgenus *Sargassum* (*sensu stricto*) on the basis of the molecular phylogenic analysis of the ITS-2 region. The taxonomic status of "*Schizophycus*" species in sub-



**Figures 13–16** Sargassum pinnatifidum Harvey (Uken, Uruma City, Okinawa, Japan, November 6, 2003). (13) Leaves of various shapes. (14) Vesicles with coronal leaves. (15) Close-up image of female receptacles. (16) Close-up image of male receptacles.

genus Sargassum (sensu lato) is still uncertain. Formerly, four species of "Schizophycus" were recognized in Japan proper: Sargassum patens, Sargassum tosaense Yendo, Sargassum kashiwajimanum Yendo and Sargassum kushimotense Yendo.

Our new taxon has some resemblance to these four species in vegetative morphology, especially with lateral branches issuing alternate-distichously in one plane. However, characteristics of the leaves and receptacles of our new taxon can be distinguished from those of these four species (Table 1). Furthermore, these four previously recognized species are distributed mainly in temperate zones of Japan proper (approximately 32-40°N), and their range of distribution is different from the subtropical zone of our new taxon (approximately 26-31°N). Furthermore, the specific status of Sargassum kashiwajimanum is uncertain, because there has been no report after the original description by Yendo (1907), who used a fragment of the alga (from Kashiwajima Island in the southern part of Kochi Prefecture of the Pacific coast of central Japan) as the type material (Yoshida 1998, Yoshida et al. 2004). The irregularly branched long leaves of Sargassum tosaense (Yoshida et al. 2004) are distinguished from the simple lanceolate leaves with dentate margins of our new taxon. The narrow and filiform leaves with several branches of Sargassum patens are different from the simple lanceolate leaves of our new taxon. Leaves on the lower portion of Sargassum kushimotense have a simple, linear-lanceolate shape with dentate margins, and they are similar to those of our new taxon. However, leaves of the upper portion of S. kushimotense are narrower and more branched than those of our new taxon.

The characteristic compressed and double-edged main branch in *Sargassum ryukyuense* resembles some species of the subgenus *Sargassum* (sensu stricto): *S. oligocystum* Montagne and *S. yendoi* Okamura et Yamada (Noro et al. 1994, 1995). However, our new species, with long, flattened and lingulate receptacles, differs entirely from *S. oligocystum* and *S. yendoi*, which have digitate receptacles.

"Schizophycus" species (Sargassum kushimotense, S. pinnatifidum, S. tosaense and S. patens) have been reported from the Philippines, China and other SE Asian regions (Trono 1992, Phillips 1995, Tseng and Lu 2000). Setchell (1933), in his work on Hong Kong seaweeds, reported S. pinnatifidum, S. patens (var. patens C. Agardh, var. rodgersianum Harvey, and var. schizophyllum Yendo) and S. tosaense. Examining his description and figures, his S. pinnatifidum and S. tosaense do not seem to be identical to the Japanese entities.

Tseng and Lu (2000) recognized *Sargassum patens* and *Sargassum pinnatifidum* and provided descriptions and images. At the same time they added *Sargassum tosaense*, following the opinion of Setchell (1933). As far as can be judged from their illustrations, thalli from the Chinese coast seem to be different from *S. pinnatifidum* from the Ryukyu Islands. A more comprehensive comparison is necessary.

Accordingly, our taxon is distinguished from all known species of the subgenus *Sargassum* (*sensu lato*) from East and Southeast Asian regions by the set of features

Feature	S. ryukyuense sp. nov.	S. kashiwajimanum Yendo	S. kushimotense Yendo	S. patens C. Agardh	S. pinnatifidum Harvey	S. tosaense Yendo
Holdfast	Discoid	Unknown	Flat to conical discoid	Flat discoid	Conical discoid	Small discoid
Stem	Terete	Unknown	Terete	Terete	Terete	Terete
Length	Up to 5 mm		10–30 mm	10–20 mm	Up to 20 mm	10 mm
Diameter	Up to 4 mm		Up to 5 mm*	2 mm*	2 mm*	2 mm
Primary branches	Compressed	Compressed	Compressed	Flattened	Compressed	Compressed
Length	Up to 70 cm	Unknown	Over 1 m	Over 1 m	Over 1 m	Over 1 m
Width	4–6 mm		2–8 mm*	4 mm*	Up to 3–5 mm	2 mm
Leaves	Simple, linear-lanceolate,	Narrow and filiform,	Filiform, linear-lanceolate,	Narrow and filiform,	Filiform, simple or	Narrow and long
	undulate and dentate	branching several times	branching once or several	branching several times	branching several times,	linear-lanceolate, margin
	margin		times, margin denticulate		margin denticulate	smooth
Length	Up to 5 cm	3-5 cm**	Up to 10 cm	10–15 cm*	Up to 11 cm*	Up to 20 cm
Width	Up to 8 mm	2–4 mm**	Up to 15 mm	Up to 15 mm*	Up to 10 mm*	Up to 15 mm
Vesicles	Elliptical to ovoid	Spherical	Elliptical to ovoid	Elliptical	Elliptical	Spherical to ovoid
Length	Up to 13 mm	7-10 mm**	Up to 10 mm*	5-15 mm*	5-10 mm*	Up to 15 mm
Coronal leaf	Simple, smooth or	1-3-times furcate, smooth	Simple or once furcate,	1-3-times furcate, smooth	Simple or 1–3-times furcate,	Long simple or once
	dentate margin	margin	smooth or dentate margin	or dentate margin	smooth margin	furcate, smooth margin
Receptacles	Dioecious	Unknown	Dioecious	Dioecious	Dioecious	Dioecious
	Simple, spatulate, flattened	Unknown	Linear, furcate, compressed	Linear, furcate, compressed	Linear, compressed,	Linear, compressed
	smooth margin in male,		denticulate margin	smooth margin	smooth margin	
	flattened smooth margin or					
	sometimes with a few spines					
	in female					
Length	Up to 25 mm	Unknown	3–10 mm*	3–10 mm*	2–3 mm*	Unknown
Width	Up to 8 mm	Unknown	1–2 mm*	1–2 mm*	1 mm*	Unknown
Distribution	Ryukyu Islands	Kochi, Japan proper	China, Philippines, Japan	China, Philippines, Japan	China, Ryukyu Islands	China, Japan proper
			proper	proper		
Reference	This study	Yoshida 1998	Yoshida 1998	Yoshida 1998	Yoshida 1998	Yoshida 1998
		Yoshida et al. 2004	Yoshida et al. 2004	Yoshida et al. 2004	Yoshida et al. 2004	Yoshida et al. 2004

Table 1 Comparison of Sargassum ryukyuense with other morphologically related species.

mentioned, in addition to a diagnostic characteristic receptacle feature, and is described as *Sargassum ryu-kyuense* Shimabukuro *et* Yoshida sp. nov.

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