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# First record of *Sporolithon ptychoides* Heydrich (Sporolithales, Corallinophycidae, Rhodophyta) from Thailand

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**Abstract** – *Sporolithon ptychoides* Heydrich (Sporolithaceae, Sporolithales), the type species of the genus Sporolithon, is newly reported for Thai waters based on specimens collected from the Gulf of Thailand and the Andaman Sea. A detailed morphological and anatomical account is provided, including comparisons with published data of *S. ptychoides* and its related species. Epithallial cells are flared. Cells of adjacent filaments connect laterally mostly by secondary pit connections. Tetrasporangia are grouped in sori that occur in patches over the thallus surface. Sori are buried in distinct rows in the thallus. Details of male, female and carposporangial conceptacles of *S. ptychoides* are described for the first time. Gametangial thalli are monoecious with spermatangia and carposporangia born in uniporate conceptacles. Dendroid spermatangial branches occur on the floor, walls and roof of the male conceptacle chamber. Carpogonial branch consists of a hypogenous cell and a carpogonium. Central fusion cell is absent on the floor of the carposporangial conceptacle chamber.

**Non-geniculate coralline red algae / *Sporolithon ptychoides* / taxonomy / Thailand**

**Résumé (traduit par la rédaction)** – *Sporolithon ptychoides* Heydrich (Sporolithaceae, Sporolithales), généritype de *Sporolithon*, est reporté pour la première fois dans les eaux Thaïlandaise (Golfe de Thaïlande et mer d'Andaman). L'anatomie et la morphologie des spécimens thaïlandais sont décrites et comparées avec les observations publiées sur *S. ptychoides* et les autres espèces du genre *Sporolithon*. Les cellules épithéliales sont évasées. Les cellules des filaments adjacents se connectent latéralement, principalement par des synapses secondaires. Les tétraspocystes sont regroupés en sores à la surface du thalle. Les sores forment des lignes distinctes sous-jacentes. Les conceptacles carposporangiaux

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mâles et femelles sont ici décrits pour la première fois pour *S. ptychoides*. Les gamétophytes sont monoïques avec les carpospocystes et les spermatocystes provenant de conceptacles unipores. Les rameaux portant les spermatocystes se développent à la surface du plancher, des murs et du toit de la chambre du conceptacle male. Le rameau carpogonial consiste en une cellule hypogène et un carpogonium. Les fusions centrales sont absentes du plancher de la chambre du conceptacle des carposporocystes.

**Coralline non articulée / algues rouges / *Sporolithon ptychoides* / taxonomie / Thaïlande**

## INTRODUCTION

Non-geniculate coralline red algae (Corallinophycidae, Rhodophyta) are very noticeable components on Indo-Pacific reefs (Marsh, 1970; Littler, 1973; Adey *et al*., 1982). Despite of their ecological importance, coralline algae from these reefs have received little attention compared to other marine algae (Ballesteros & Afonso-Carrillo, 1995; Ringeltaube & Harvey, 2000). In Thailand, taxonomic information on non-geniculate coralline red algae is limited. After Foslie (1901) reported eight species (nine species after revision, Foslie, 1901; Woelkerling *et al.*, 2005) from Koh Chang during the Danish expedition to Thailand ("Siam") in 1899-1900, two additional species have been reported by Nateewathana *et al.* (1981) and Lewmanomont & Ogawa (1995). Woelkerling *et al.* (1996) have long argued that most non-geniculate corallines cannot be identified only based on growth form, and that anatomical observations of reproductive features are critical for accurate identification.

To date, only one species of *Sporolithon* has been recorded from Thai waters: *S. schmidtii* (Foslie) G.D.Gordon, T.Masaki *et* Akioka by Foslie (1901, as *Archaeolithothamnion*) from Koh Kradat, Gulf of Thailand.

During surveys of the algal diversity on coral reefs of Thailand (2006-2011) we collected numerous coralline red algae. This study reports on the first record of *Sporolithon ptychoides* Heydrich (1897), from the Gulf of Thailand and the Andaman Sea. We compare vegetative and reproductive characteristics with other species of *Sporolithon*. In addition, new information on spermatangial (male), carpogonial (female) and carposporangial conceptacles are reported for *S. ptychoides.*

## MATERIALS AND METHODS

Specimens were collected from the coral reefs at Koh Man and Koh Tan in the Gulf of Thailand, Similan Islands National Park and Koh Rasha in the Andaman Sea, and preserved in 5% formalin/seawater. Voucher specimens were deposited at Kasetsart University Museum of Fisheries (KUMFAL).

For microscopic observations, selected fragments of preserved specimens were decalcified in 0.6 M $HNO_3$ and rinsed with distilled water. Tissue embedding process modified from Sass (1966) was used for histological and microslide preparation. The tissue was sectioned at 10-15 μm and dyed with 1% aniline blue and mounted with 50% Karo Syrup. For scanning electron microscopy, fractured

pieces were mounted on stubs using carbon adhesive, stored in a desiccators for at least 24 hr prior to examination, coated with gold for 2-3 min in an Eiko Engineer IB-2 ion coater and examined with a Joel JSM 5600LV scanning electron microscope (SEM), at 10kV accelerating voltage. Tetraporangia, tetrasporangial compartments, tetrasporangial pores and number of tetrasporangial compartment paraphyses were measured and counted following Keats & Chamberlain (1993). Conceptacle measurements follow Adey *et al.* (1982). Growth form and anatomical terminology follows Woelkering *et al.* (1993, 1996).

## OBSERVATIONS

***Sporolithon ptychoides* Heydrich, 1897: 67-69**

**Basionym:** *Sporolithon ptychoides* f. *ptychoides* (as *Sporolithon ptychoides* f. *dura* Heydrich, 1897: 67-69).

**Synonyms:** *Sporolithon mediterraneum* Heydrich, 1899: 131; *Archaeolithothamnion mediterraneum* (Heydrich) Foslie, 1900: 8; *Archaeolithothamnion dimotum* Foslie *et* Howe, 1906: 128; *Sporolithon dimotum* (Foslie *et* Howe) Yamaguishi-Tomia *ex* Wynne, 1986: 2243.

**Lectotype:** TRH (not seen, considering that Verheij (1993) has presented a description), El Tor, Red Sea, no number (as *Sporolithon ptychoides* f. *dura*) designated by Woelkerling & Townsend (Woelkerling, 1988: 204, Fig. 239).

**Specimens Examined:**

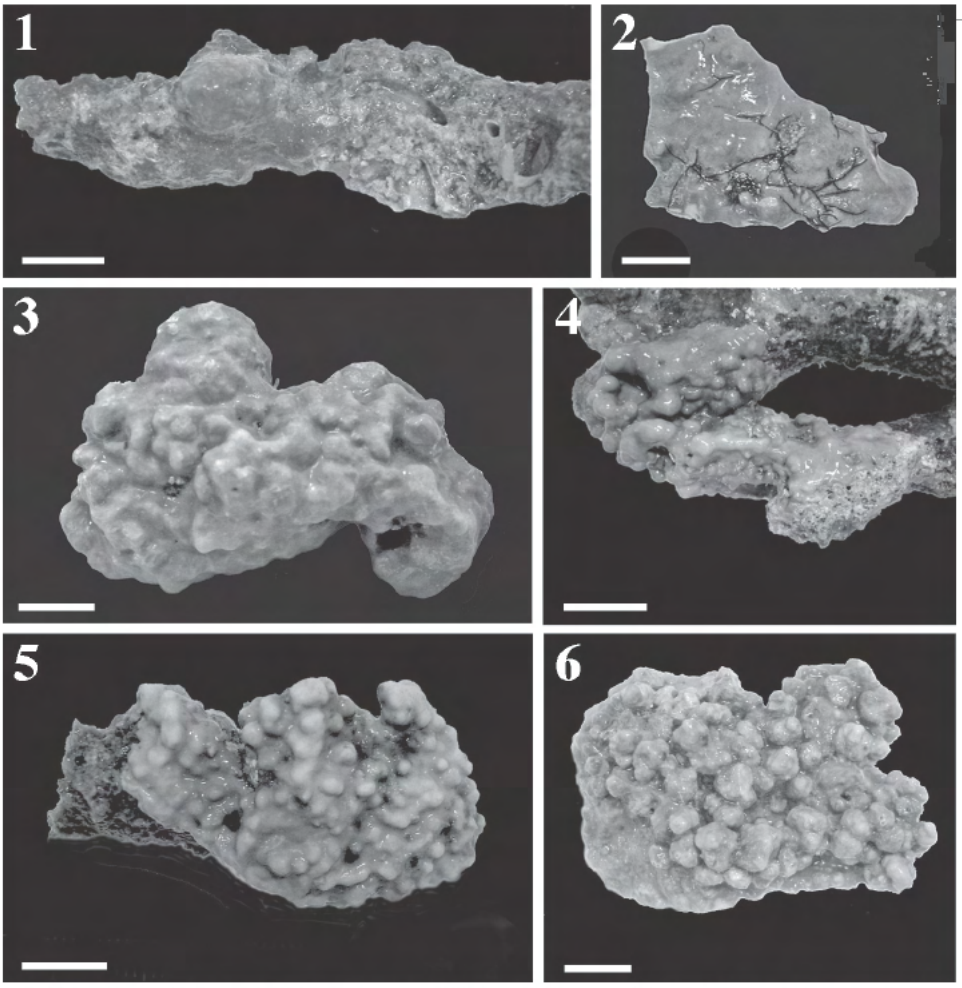
**Gulf of Thailand:** Hin Yuan (12°34′ 40.46″E; 101°43′ 03.31″N), Koh Man, Rayoung Province, *leg. C. Kaewsuralikhit*, 12.xii.2007, 7 m depth, KUMFAL 0001. Koh Man Klang (12°36′ 04.35″E; 101°41′ 36.72″N), Rayoung Province, *leg. C. Kaewsuralikhit*, 13.xii.2007, 5 m depth, KUMFAL 0023, KUMFAL 0027. Koh Tan (12°16′ 11.14″E; 99°56′ 45.68″N), Surat Thani Province, *leg. C. Kaewsuralikhit*, 22. iv.2008, 4 m depth, KUMFAL 0133.

**Andaman Sea:** Koh Pha (8°54′ 47.06″E; 98°13′ 20.85″N), Phang Nga Province, *leg. T. Duangdee*, 1.vii.2009, 5 m depth, KUMFAL 0124. Koh Noung (8°29′ 05.32″E; 97°38′ 41.12″N), Similan Is. National Park, Phang Nga Province, *leg. T. Noiraksa*, 4.iv.2011, 6 m depth, KUMFAL 0045, KUMFAL 0047. Koh Meang (8°34′ 19.03″E; 97°38′ 10.40″N), Similan Is. National Park, Phang Nga Province, *leg. T. Noiraksa*, 5.iv.2011, 5 m depth, KUMFAL 0085, KUMFAL 0086, KUMFAL 0088. Bacon reef (8°38′ 35.55″E; 97°39′ 07.18″N), Similan Is. National Park, Phang Nga Province, *leg. T. Noiraksa*, 6.iv.2011, 6 m depth, KUMFAL 0073. Koh Gao (8°40′ 32.52″E; 97°39′ 11.83″N), Similan Is. National Park, Phang Nga Province, *leg. T. Noiraksa*, 7.iv.2011, 8 m depth, KUMFAL 0074.

### Vegetative structure morphology

Thallus smooth to lumpy and strongly adherent to the substratum (Figs 1-6). Colour in living plants vary from yellowish green, greenish brown, pinkish to gray-pink. Thallus pseudoparenchymatous; cells of adjacent filaments connected laterally, mostly by secondary pit connections rather than cell fusions (Fig. 7).

The crustose part of the thallus dorsiventral and monomerous (Fig. 8) in which a single branched system consisting of ventral filaments that are more or less parallel to the substratum (cells 4-8 µm in diameter and 10-35 µm long) curve towards the thallus surface (cells 4-8 µm in diameter and 6-13 µm long). Outward

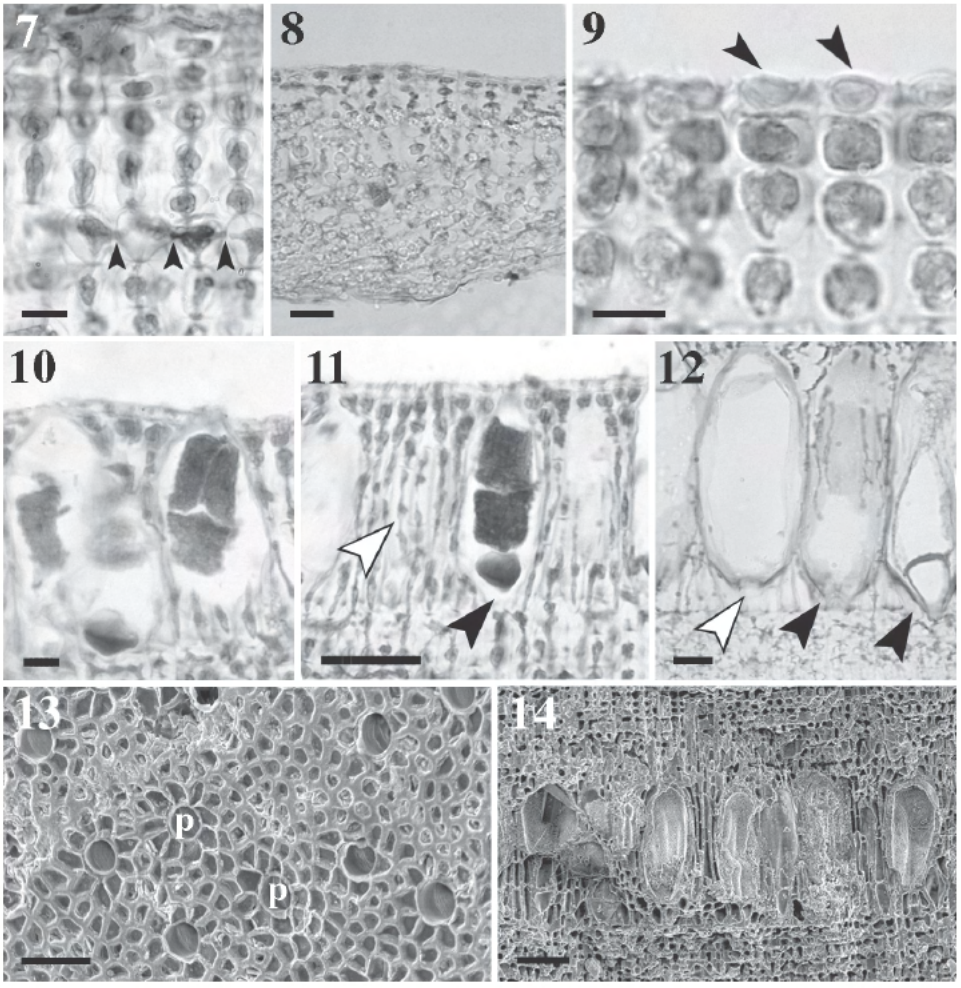



Figs 1-6. Growth forms of *Sporolithon ptychoides*. **1, 2.** Smooth form (Fig. 1, KUMFAL 0088; Fig. 2, KUMFAL 0045). **3-6.** Lumpy form (Fig. 3, KUMFAL 0124; **4.** KUMFAL 0047; **5.** KUMFAL 0001; **6.** KUMFAL 0023) (All scale bars, 1 cm).

curving filaments terminating in a single layer of epithallial cells that are more or less elliptical but with flared, thick outer-walls and tiny cell lumen. Epithallial cells measure 4-8 μm in diameter and 3-5 μm in length (Fig. 9).

## Reproductive morphology

Sori irregularly shaped, raised 2-5 cells above the surrounding vegetative thallus, and bearing tetrasporangial compartments in dense clusters. Individual tetrasporangial chambers uniporate and measuring 65-100 μm in height and 30-56 μm in diameter. Chambers containing a single cruciately arranged tetrasporangium measuring 55-85 μm long and 25-50 μm in diameter (Fig. 10), subtended by a single large stalk cell. Zonately arranged bisporangia (Fig. 11) infrequent.




Figs 7-14. Vegetative and tetrasporangial anatomy of *Sporolithon ptychoides*. **7**. Vertical section of thallus showing secondary pit connections (arrowheads) between cells of contiguous filaments (KUMFAL 0124) (Scale bar, 5 µm). **8**. Vertical section of the vegetative thallus showing the monomerous construction (KUMFAL 0088) (Scale bar, 10 µm). **9**. Vertical section of the outer vegetative thallus showing flared epithallial cells (arrowheads) (KUMFAL 0124) (Scale bar, 10 µm). **10.** Vertical section showing a cruciately arranged tetrasporangium (KUMFAL 0124) (Scale bar, 10 µm). **11**. Vertical section showing a bisporangium, paraphyses (white arrowhead) between the tetrasporangial compartment and a single large stalk cell (black arrowhead) (KUMFAL 0124). (Scale bar, 40 µm). **12**. Tetrasporangial compartment developed on an elongated basal cell layer (white arrowhead) and not on an elongated basal cell layer (black arrowheadss) (KUMFAL 0045). (Scale bar, 10 µm). **13**. SEM image of the surface of a tetrasporangial sorus showing tetrasporangial chamber pores (p) surrounded by 8-9 rosette cells (KUMFAL 0045) (Scale bar, 20 µm). **14**. SEM image showing a buried tetrasporangial sorus (KUMFAL 0045) (Scale bar, 50 µm).

Tetrasporangial chambers subtended by a basal layer of elongated cells (Figs 10-12). Adjacent tetrasporangial chambers separated by calcified paraphyses, 5-9 cells long and 1-7 filaments bearing between tetrasporangial chamber.

Tetrasporangia bearing an apical pore plug with a pore 9-12 μm in diameter and surrounded by 8-9 rosette cells (Fig. 13). Groups of senescent tetrasporangial chambers become buried in distinct layers in the thallus (Fig. 14).

Gametangial thalli monoecious with spermatangia and carposporangia born in uniporate conceptacles. Male conceptacles chambers measuring 40-80 μm in height and 83-216 μm in diameter, clustered in dense aggregations and distinctly visible under stereoscope and SEM (Figs 15, 16). The first spermatangial initials transformed to intercalary meristematic cells, which become elongated as Verheij (1992) described for *S. episoredion.* At early stage of male conceptacle development, the elongated cells, which are the initials of the spermatangial mother cells, connect from the floor to the roof of conceptacle and divide the young conceptacle to many compartments (Figs 17, 18). Dendroid spermatangia occuring on the floor, walls and roof of the male conceptacles. Old male conceptacles becoming buried in the growing thallus.

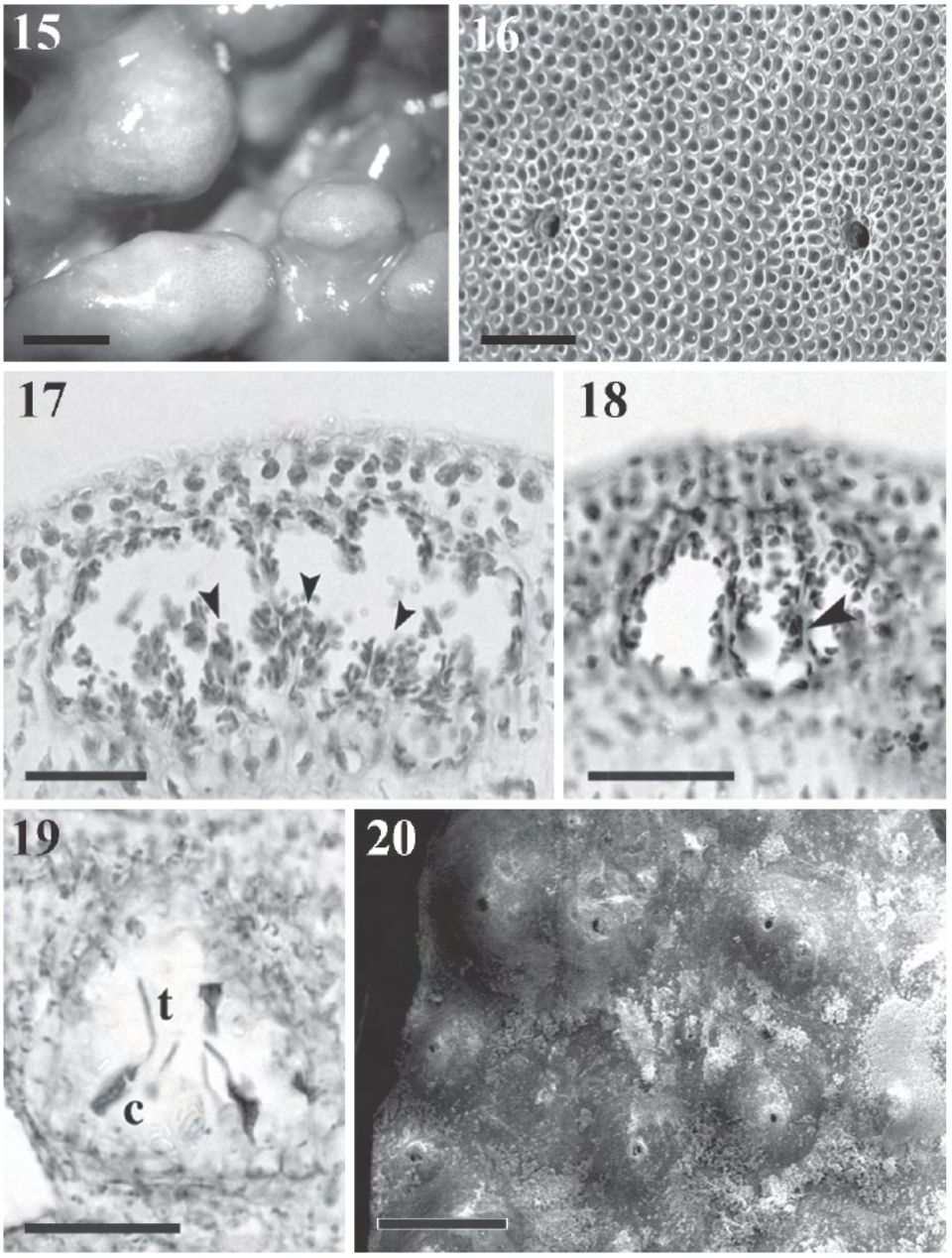
One unfertilized female conceptacle, measuring 46 μm in chamber height and 38 μm in chamber diameter, was found buried in gametangial plant (Fig. 19). In the female conceptacle, the carpogonial branch consisted of a hypogenous cell and a carpogonium that extended into a trichogyne.

After presumed karyogamy, carposporophytes developing within female conceptacles and forming carposporangial conceptacles. Carposporangial conceptacles grouped in clusters and raised above the surrounding thallus surface (Fig. 20). Their chambers measuring 166-185 μm in height and 150-183 μm in diameter. We were unable to observe the gonimoblast filaments in the carposporangial conceptacle chamber. Carpospores measuring 57-70 μm long and 25-30 μm in diameter, found in matured and buried conceptacles (Fig. 21). The central fusion cell is absent (Fig. 22). The cells surrounding the carposporangial conceptacle are well stained and flatten by the development of the conceptacle. Male and carposporangial conceptacles are found on the same crust (Figs 23, 24).

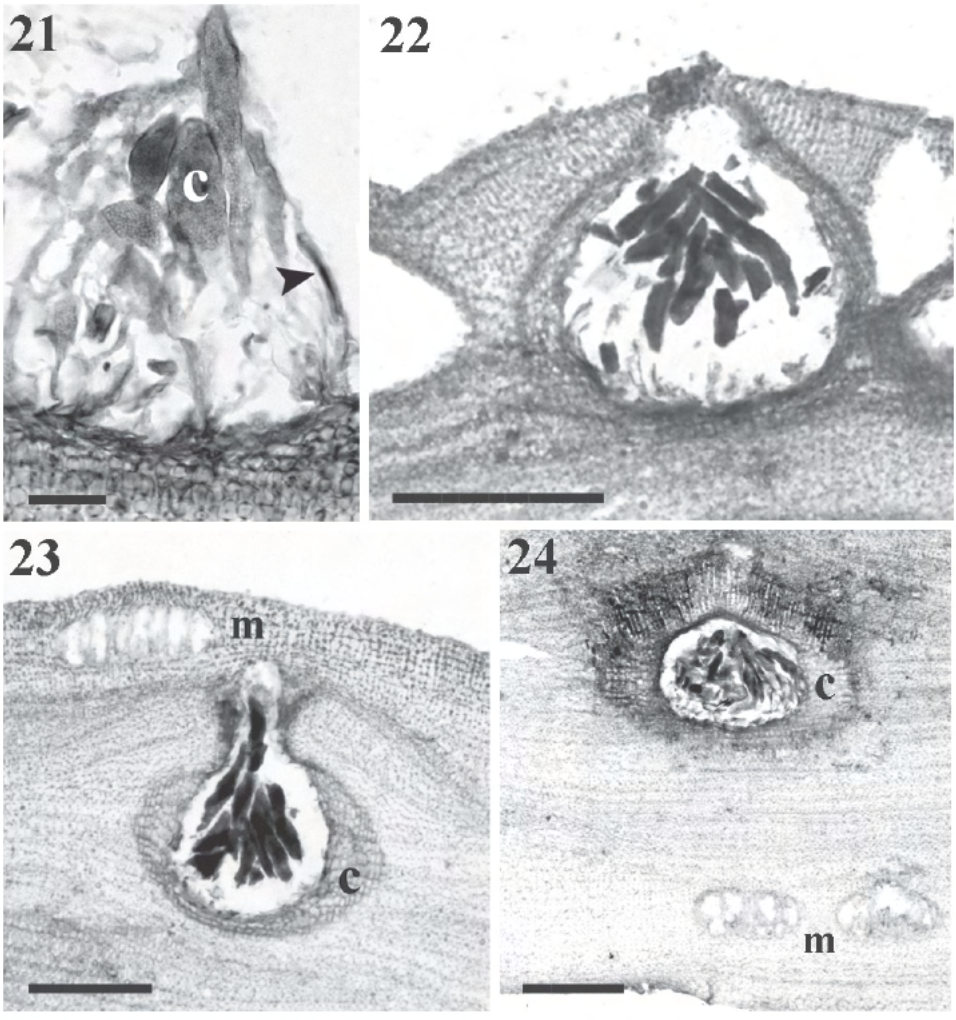
## DISCUSSION

The features observed for our specimens correspond to *Sporolithon ptychoides* described by Verheij (1993), Keats & Chamberlain (1993), Alongi *et al.* (1996) and Bahia *et al.* (2011) (Table 1). The specimens examined in this study showed slight differences, however, from those reported in these studies. Differences reported here refer mainly to (i) the size of the tetrasporangia which

Figs 15-20. Gametangial anatomy of *Sporolithon ptychoides*. **15**. Male conceptacles on the tips of protuberances as seen under a stereoscope (KUMFAL 0047) (Scale bar, 1 cm). **16**. SEM image of two male conceptacles showing conceptacle roofs flush with the surrounding thallus surface (KUMFAL 0047) (Scale bar, 50 μm). **17**. Vertical section through male conceptacle showing dendroid spermatangial branches (arrowheads) produced around the entire conceptacle chamber (KUMFAL 0047) (Scale bar, 10 μm). **18**. Vertical section through an early stage male conceptacle showing elongated spermatangial mother cells initials (arrowhead) connected from the chamber floor to the roof divided the young conceptacle into compartments (KUMFAL 0047) (Scale bar, 10 μm). **19**. Vertical section through the thallus showing a buried female conceptacle. Note the unfertilized carpogonia (c) extending into a trichogyne (t) (KUMFAL 0073) (Scale bar, 10 μm). **20**. SEM image of aggregated carposporangial conceptacles showing conceptacle roofs protruding above the thallus surface (KUMFAL 0073) (Scale bar, 500 μm).




are generally smaller in range; and (ii) tetrasporangia mainly formed on a basal layer of elongate cell. The size of tetrasporangia and tetrasporangial compartments in this study was measured following the method described by Keats & Chamberlain (1993). The differences in tetrasporangial dimensions are possibly due to the different measurement methods. For example, the sporangial size of




Figs 21-24. Gametangial anatomy of *Sporolithon ptychoides*. **21**. Magnified view of carposporangial conceptacle showing mature carpospores (c) and an unfertilized carpogonial branch (arrowhead) (KUMFAL 0073) (Scale bar, 10 μm). **22**. Vertical section through a carposporangial conceptacle showing elongated carposporangia in the conceptacle chamber (KUMFAL 0073) (Scale bar, 50 μm). **23**. Vertical section of the thallus showing a buried carposporangial conceptacle (c) and a male conceptacle (m) at the thallus surface (KUMFAL 0073) (Scale bar, 50 μm). **24**. Vertical section of the thallus showing buried carposporangial (c) and male (m) conceptacles (KUMFAL 0073) (Scale bar, 50 μm).

*S. schmitdii* (as *A. schmidtii*) reported by Foslie (1901) was based on measurements of the sporangial chamber, rather than the sporangia itself (Foslie, 1904: 41). To prevent such misinterpretation, we suggested measuring both the tetrasporangia and tetrasporangial compartments separately to clarify the descriptive details amongst species of the Sporolithales.

Foslie (1901) described *Sporolithon schmidtii* (as *A. schmidtii* Foslie) from Koh Kradat, Gulf of Thailand. After studying the lectotype collection of

Table 1 Comparative analysis of characters of Sporolithon ptychoides and its related species

| *Character* | *S. ptychoides*[1] | *S. ptychoides*[2] | *S. ptychoides*[3] | *S. ptychoides*[4] | *S. ptychoides*[5] | *S. schmidtii*[6] | *S. schmidtii*[7] | *S. molle*[3] |
|---|---|---|---|---|---|---|---|---|
| Locality | Gulf of Thailand, Andaman Sea | Atlantic Ocean, Brazil | Red Sea, Indian Ocean, Indonesia, Hawaii | Indian Ocean, South Africa | Mediterranean Sea | Gulf of Thailand | Guam | Red Sea, Indian Ocean, Indonesia |
| Cell connection | Mainly secondary pit connection | Mainly secondary pit connection | Mainly secondary pit connection | Mainly cell fusions | Secondary pit connection and cell fusion | No data | Cell fusion | Mainly secondary pit connection |
| Tetrasporangial compartment length (μm) | 65-100 | No data | No data | 77-108 | No data | No data | 62-85 | No data |
| Tetrasporangial compartment diameter (μm) | 30-56 | No data | No data | 29-53 | No data | No data | 25-44 | No data |
| Tetrasporangial compartment pore diameter (μm) | 9-12 | 10-14 | No data | 10-14 | 12-15 | No data | No data | No data |
| Tetrasporangia length (μm) | 55-85 | 75-105* | 85-105* | 49-70 | 85-130* | 50-65* | 36-64 | 70-85* |
| Tetrasporangia diameter (μm) | 25-50 | 40-55** | 35-45** | 37-50 | 40-60** | 30-40** | 20-31 | 25-45** |
| No. of rosette cells surrounding tetrasporangial compartment pore | 8-9 | 10-11 | No data | 8-11 | No data | No data | No data | No data |
| Layer of elongate cells at the base of tetrasporangia | Mainly present | Present | Present | Present | Present | No data | No data | Absent |
| No. of cells to which tetrasporangial sorus is raised above vegetative thallus | 2-5 | 2-4 | 1-2 | 2-8 | No data (raised) | No data | No data | 0 |
| No. of cells in tetrasporangial paraphyses | 5-9 | 3-5 | 3-5 | 7-9 | 4-6 | No data | No data | 3-4 |
| Male conceptacle<br>Conceptacle chamber height (μm)<br>Conceptacle chamber diameter (μm) | Found<br>40-80<br>83-216 | Not found | Found<br>No data<br>No data | Not found | Not found | Not found | Not found | Found<br>100-250<br>450-700 |
| Spermatangia system- type and distribution | Dendroid on floor and roof | | Dendroid on floor and roof | | | | | Dendroid on floor and roof |
| Female conceptacle<br>Conceptacle chamber height (μm)<br>Conceptacle chamber diameter (μm) | Found<br>46<br>38 | Not found | Not found | Not found | Not found | Not found | Not found | Not found |
| Carposporangial conceptacle<br>Conceptacle chamber height (μm)<br>Conceptacle chamber diameter (μm) | Found<br>166-185<br>150-183 | Not found | Not found | Not found | Not found | Not found | Not found | Not found |

**Information sources:** 1. this study. 2. Bahia *et al.* (2011). 3. Verheij (1993). 4. Keat & Chamberlain (1993). 5. Alongi *et al.* (1996). 6. Foslie (1901). 7.Gordon *et al.* (1976).

**Remarks:** * Tetrasporangia length might be confused with the tetrasporangial compartment length due to the measurement method.

** Tetrasporangia diameter might be confused with the tetrasporangial compartment diameter due to the measurement method.

*S. schmidtii*, Verheij (1993) concluded that this species is closely related to *S. ptychoides* based on the burying of senescent tetrasporangial chambers and the formation of tetrasporangia on a layer of elongated cells. In order to clarify the relationship between these two species, a detailed study of newly collected materials for *S. schmidtii* is needed.

Verheij (1993) pointed out that the presence of tetrasporangia on an elongated basal cell layer is one of the important characteristics for distinguishing *S. ptychoides* from *Sporolithon molle* (Heydrich) Heydrich. However, in our specimens of *S.ptychoides* there are some variations of the development of tetrasporangia on a basal layer of elongated cells. The basal layer of elongate cells was present or absent at the base of tetrasporangia in the same thalli (Fig. 12). In this case, the position of tetrasporangia on the elongated cells may depend on the direction of cutting in regions where tetrasporangia and paraphyses developed or their natural variations, which need further confirmations.

Male conceptacles have been reported for *S. ptychoides* (Segonzac, 1983; 1984 as *A. erythreum*; Verheij, 1993), *S. episoredion* (Verheij, 1992, 1993) and *S. durum* (Townsend *et al.*, 1995). Segonzac (1983, 1984) reported and illustrated an empty male conceptacle. Later, Verheij (1993: 191) reported on the dendroid spermatangia occurring on the roof as well as the floor of male conceptacles in *S. ptychoides*, but without illustration. Our study confirms the observations of Verheij (1993). For carposporangial conceptacles within the genus *Sporolithon*, Verheij (1992) reported gonimoblast filaments produced from a fusion cell for *S. episoredion* while Townsend *et al.* (1995) showed carposporangia apparently developing directly from fertilized carpogonia in *S. durum* and so a central fusion cell is absent for this latter species. Incidentally, in carposporangial plants thus far reported in the related genus *Heydrichia,* carposporangia apparently also development directly from fertilized carpogonia resulting in short gonimoblast filaments scattered across the floor and walls of the female conceptacle, and the absence of a central fusion cell (Townsend *et al.*, 1994; Harvey *et al.*, 2002; Maneveldt and van der Merwe, 2012). This study is the first to report female and carposporangial conceptacles for *S. ptychoides.*

This is the first report of *S. ptychoides* in Thai waters from the Gulf of Thailand and the Andaman Sea. Our study expands the geographic distribution of this species and confirms the taxonomic features associated with gametangial conceptacle structure. Not only the tetrasporangia and tetrasporangial conceptacle characters, but also the additional details of gametangial and carposporangial conceptacles described in this paper can be used to distinguish *S. ptychoides* from other species of *Sporolithon.*

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