

## Taxonomy of Three Species of the Genus *Petalonema* Berkeley ex Correns (Scytonemataceae, Cyanoprokaryota/Cyanobacteria) from Maharashtra, India

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### Abstract

The genus *Petalonema* Berkeley ex Correns 1889 is a heterocytous cyanobacterium that belong to the order Nostocales and the family Scytonemataceae. Mostly, it is a terrestrial alga growing on the calcareous wet rocks, on stones, dripping walls, and on wet soils, however it has also been reported from calcareous lakes and limestone springs. Morphologically, *Petalonema* is similar to another cyanobacterial genus *Scytonema* but it differs in having certain significant features, such as irregular false branching, extremely broad and thick lamellated sheaths surrounding the trichomes, several times broader sheath than the trichomes, and funnel like divergent sheath at the ends. In India, few species of *Petalonema* are recorded from some states like Meghalaya, Karnataka, Tamil Nadu, Uttarakhand, Maharashtra, Kerala, and West Bengal. From Maharashtra, two species of *Petalonema* viz. *P. densum* and *P. alatum* are known which were recorded by Kamat in the year 1963 and 1968 respectively. Since then, researchers could not record any species of *Petalonema* from Maharashtra. In the present study, the taxonomy of three species of *Petalonema* is discussed. Of these, *P. alatum* and *P. densum* are rediscovered after decades, while *P. crassum* is a new addition to the cyanobacteria of Maharashtra.

### INTRODUCTION:

The genus *Petalonema* Berkeley ex Correns 1889 is a filamentous, falsely branched and heterocyte containing cyanobacterium. It belong to the order Nostocales and family Scytonemataceae (Komárek *et al.*, 2014). At present, only 9 species are described worldwide under this genus, and these are *P. alatum*, *P. crassum*, *P. crustaceum*, *P. densum*, *P. fluminense*, *P. incrustans*, *P. involvens*, *P. pulchrum*, and *P. velutinum* (Guiry and Guiry, 2021). *Petalonema* species are mostly terrestrial and naturally found growing on the calcareous wet rocks, on stones, dripping walls, and on wet soils, but it has also been recorded from some aquatic

habitats such as calcareous lakes, ponds and limestone springs (Kukk *et al.*, 2001; Komárek, 2013; Maree *et al.*, 2018). The type species of *Petalonema* is *P. alatum* which was first time described and illustrated in 1825 as *Oscillatoria alata* by a Scottist botanist Carmichael (Uher, 2010; Maree *et al.*, 2018). Berkeley (1833) transferred *Oscillatoria alata* to a new genus *Petalonema* (as *P. alatum*) as it contain a broader sheath surrounding the trichome. Whereas, Borzi (1879) combined *P. alatum* together with *Scytonema densum* under a new species name *Scytonema alatum*. Although *Petalonema* is morphologically similar to *Scytonema*, Komárek and Anagnostidis (1989)

considered it more closely affiliated to *Tolypothrix* based on the observed heteropolar growth. Therefore, they transferred *Petalonema* as a distinct genus under the family Microchaetaceae rather than the family Scytonemataceae, and this view was also supported by Uher (2010). On the contrary, these views were later rejected based on the molecular phylogenetic data by Mares *et al.* (2015), and supported the taxonomic placement of *Petalonema* as a separate genus distinct from *Scytonema* under the family Scytonemataceae. Some species of *Petalonema* are studied and described from different parts of India. *P. alatum* is reported from Shembaganur, Tamil Nadu (Fremy, 1942); Alibag, Maharashtra (Kamat, 1968); Kolkata, West Bengal (Gupta, 2012), and Sirsi taluk, Uttara Kannada, Karnataka. Rao (1944) described *P. alatum* var. *indicum* which was collected near Chakrata, Dehra Dun Road, Uttarakhand. Gupta (2012) reported one more species of *Petalonema* i.e. *P. crassum* from Kolkata, West Bengal. Biswas (1934) described *P. crustaceum* from Cherrapunji, Meghalaya, which was originally collected by Dr. S. P. Agharkar in 1925. Singh and Singh (2019) also described *P. crustaceum* from Sirsi taluk, Uttara Kannada, Karnataka. Kamat (1963) described *P. densum* from Takala, Kolhapur, Maharashtra, and also by Gupta (2012) from Kolkata, West Bengal. Jackson and Panikkar (2015) described *P. incrustans* from Punnathala, Thangasserry (Kollam), Kerala. In the present communication, taxonomic details of three species of the genus *Petalonema* collected from Maharashtra, India are given. Of these, *P. alatum* and *P. densum* are rediscovered after decades from Maharashtra, whereas *P. crassum* is the first report as a new cyanobacterium to the state.

#### MATERIALS AND METHODS:

While studying cyanobacterial diversity from the western Maharashtra, India, author manually collected several samples from different habitats. Sample of *P. alatum* was collected from the village Danoli near a hill station Amboli in Sindhudurg district where it was growing on calcareous rocks. *P. densum* was sampled from the wet rock at Malvan in Sindhudurg district whereas, *P. crassum* was collected from the wet rocks under trickling water at Bhandardara which is a hill station in Ahmednagar district. Field collected samples were stored in plastic sample containers and were transferred to the laboratory. Part of the samples were preserved in 4% formalin and remaining samples were separately transferred in BG 11

growth medium. For the taxonomic investigations, whole mount samples on glass slides were prepared. From each sample, about 50 thalli were observed under trinocular microscope (Leica DM2500, Germany) equipped with a Leica EC3 digital camera. Photographs were captured and processed for the morphometric measurements using LAS EZ software (Leica, Germany). Some typical characters were stressed in the samples while identifying them up to the species level. These characters includes, presence or absence of false branches, heterocyst and its positions, presence or absence of mucilaginous sheath surrounding the trichomes, its colour, thickness, and lamellations (parallel or divergent) in it. Identification was done using standard literatures that included Desikachary (1959), Uher (2010), Mares *et al.* (2015), and Komárek (2013). For the classification of identified species, Komárek *et al.* (2014) classification system was followed.

#### RESULTS AND DISCUSSION:

##### Morphology and taxonomy

After critical morphological examination, cyanobacterial samples were identified as *P. alatum*, *P. crassum* and *P. densum*. As per Komárek *et al.* (2014) classification system, these species belong to the order Nostocales and the family Scytonemataceae. Taxonomic description of each of the species is given below:

##### 1. *Petalonema alatum* Berkeley ex Kirchner 1898 (Fig. 1)

Basionym: *Scytonema alatum* Borzi ex Bornet & Flahault 1887

Filaments solitary or densely clustered in caespitose, expanded, creeping thallus on substrata, intermingled with other algae, mucilaginous, blackish brown, sometime forming woolly mass; filaments flexuous, heteropolar, 50.3 – 122.6  $\mu$  broad, usually with false branches; false branches single or mostly in pair, next to the heterocyst, free at the ends; sheath very thick, broad, firm, lamellated, distinctly funnel shaped, with divergent lamellation, colourless to yellowish brown, often inside brown, outside colourless; trichomes cylindrical, distinctly constricted at the cross walls, heteropolar, with meristematic zones at the apex, with rounded or globular end cells; cells at the apical growing region short, barrel shaped, in older parts long, cylindrical, up to twice as long as broad, pale blue green, 6.7 – 14.3  $\mu$  broad, 4.6 – 14.5  $\mu$  long; heterocyst solitary, mostly intercalary or basal, somewhat broader than the trichome,

spherical to long cylindrical, 11.2 – 19.7  $\mu$  broad, 17.1  $\mu$  long.

**2. *Petalonema crassum* [Nägeli in Kützing] Migula 1907 (Fig. 2)**

Basionym: *Scytonema crassum* Nägeli in Kützing ex Bornet & Flahault 1887

Thallus woolly, an expanding cushion like on moist rocky substratum, spongy tomentose, blackish green; filaments flexuous, erect, contorted at the base, 1 mm long, 26.3 – 47.9  $\mu$  broad; false branches free at base, erect, ascending; sheath thick, wide, ocreate, lamellated, with distinctly divergent lamellation, inner layers yellowish, middle ones brownish, outer ones colourless; trichome blue green or greenish brown in colour, 12.8 – 16.1  $\mu$  broad, constricted or unconstricted at cross walls; cells subquadrate or oblong, broader than long, 3.3 – 8.7  $\mu$  long; heterocyst globose or subquadrate, brownish, intercalary, 11.8 – 17  $\mu$  broad, 5.9 – 12.6  $\mu$  long; hormogone with terminal heterocyst at one end, sometimes on both the ends; terminal heterocyst 8.7 – 12  $\mu$  broad, 7.2 – 8.3  $\mu$  long.

**3. *Petalonema densum* A. Braun ex Migula 1907 (Fig. 3)**

Basionym: *Scytonema densum* [A. Braun] Bornet et Thuret ex Bornet & Flahault 1887

Filaments solitary or in densely aggregated clusters, cushion like, blackish brown; filaments long, more or less entangled, straight or curved, 30.8 – 47.8  $\mu$  broad, usually with false branches; false branches mostly single, sometimes geminate, free at the ends; sheath very thick, broad, firm distinct, mostly yellowish brown, at some region dark brownish, lamellated, mostly with distinct divergent lamellation, sometimes in funnel like pieces towards the ends; trichomes cylindrical, long, more or less constricted at the cross walls; cells short, barrel shaped at the ends, in older parts long, cylindrical, with bluish green content, 9.2 – 11.4  $\mu$  broad, 3.3 – 6.4  $\mu$  long; heterocyst solitary, intercalary or basal, more or less spherical, sometimes discoid, slightly broader than the trichomes, 9.4 – 16.9  $\mu$  broad, 6.6 – 15.2  $\mu$  long.

**Discussion:**

The genus *Petalonema* is separated from *Scytonema* species with *P. alatum* as the type species (Komárek, 2013; Mares *et al.*, 2015). Both the genera *Scytonema* and *Petalonema* shares common structural features (Taton *et al.*, 2006). The developmental history of the major diacritical features of the type species *P. alatum* has been given by Uher (2010). He has also provided

comparative morphometric data of most of the *Petalonema* species occurring worldwide. The morphological features of *Petalonema* such as, presence of intercalary or basal heterocyte in the filament, isopolar or heteropolar nature of the filaments, and single of geminate false branching shows close resemblance to the members of the families Scytonemataceae (e.g. *Scytonema*) and Microchaetaceae (e.g. *Tolypothrix*) (Mares *et al.*, 2015). Komárek and Anagnostidis (1989) classified *Petalonema* under the family Microchaetaceae based on the heteropolar nature of the filaments despite considering scytonematoid and geminate false branching. This classification was later rejected by Kukk *et al.* (2001), and they reclassified *Petalonema* again into *Scytonema* under the family Scytonemataceae. But Uher (2010) supported the concept of Komárek and Anagnostidis (1989). Till the year 2014, 16S rRNA gene data was available only for the species *P. cf. involvens* (Taton *et al.*, 2006; Mares *et al.*, 2015), and therefore, molecular and phylogenetic studies on the genus *Petalonema*, especially on the type species *P. alatum* was needed to confirm its valid identity (Komárek, 2013; Komárek *et al.*, 2014). Later, Komárek (2013) and Komárek *et al.* (2014) classified *Petalonema* under the family Scytonemataceae considering its morphological characters only, which separates it from other alike genera like *Scytonema* (particularly subgenus *Myochrotes*), *Tolypothrix*, and recently created genus *Chakia* (Komárková *et al.*, 2013). Thus, to resolve the taxonomic problems of *Petalonema*, Mares *et al.* (2015) recently carried a phylogenetic study on the type species *P. alatum*. Based on the results of this study, they clearly rejected all the earlier views of Uher (2010) and Komárek (2013) on *Petalonema*. Further, they separated *Petalonema* as a distinct genus from other morphologically similar genera, and treated it under the family Scytonemataceae.

Out of the three species of *Petalonema* described here, *P. alatum* was collected from the village Danoli near a hill station Amboli in Sindhudurg district. This species is previously reported only by Kamat (1968) from wall of the dam at Tinvira near the town Alibag in Riagad district, which is a non-hilly region along the coast of Maharashtra. Since there was a difference in the habitats of past and the current collection of *P. alatum*, author was interested in comparing morphology but Kamat's account was only with the list without giving any morphological description of this species. Morphometric dimensions of the current *P. alatum*

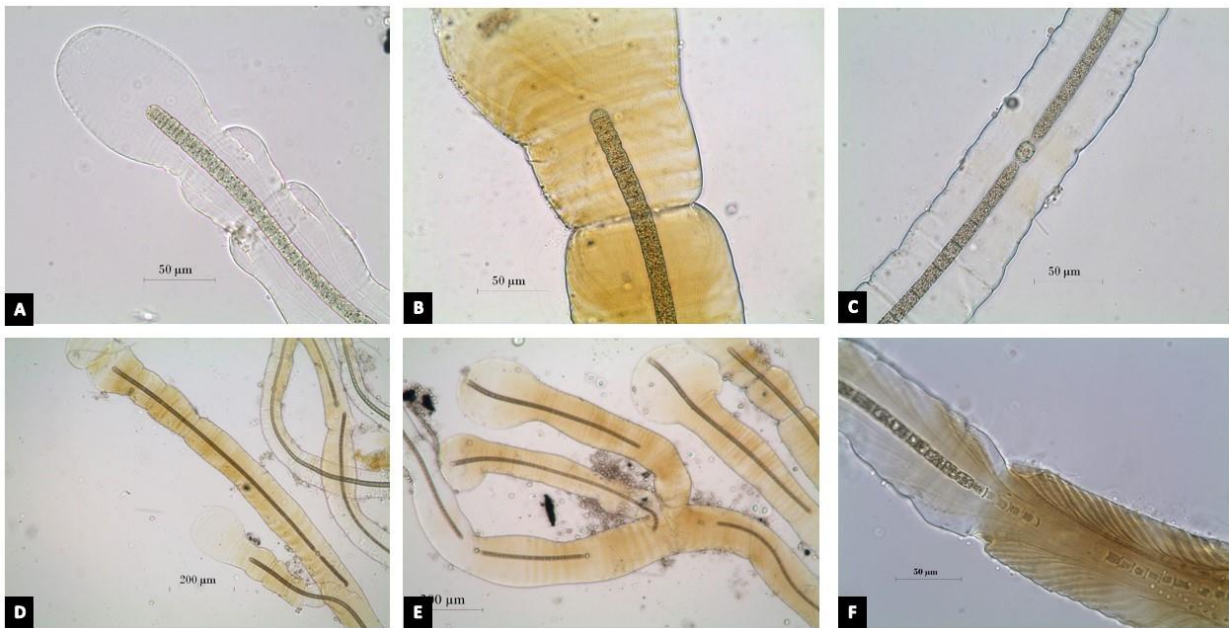


Fig. 1. *Petalonema alatum*: **A.** Apical end of filament **B.** Funnel shaped, broad and yellow coloured sheath around trichome **C.** Intercalary heterocyte in the trichome **D.** Filament with single false branch **E.** Filament with geminate false branches **F.** Filament with distinct divergent lamellations in the sheath



Fig. 2. *P. crassum*: **A.** Filament with yellow to brown broader sheath around the trichome **B.** Filament with single and geminate false branches showing blue green trichomes **C.** Filament with distinct divergent lamellations in the sheath **D.** Hormogone with terminal heterocytes (arrow).



Fig. 3. *P. densum*: **A.** Apical end of filament **B.** Filament with single false branch **C.** Filament with geminate false branches **D.** Filament with several layers in yellowish brown sheath, with oblique lamellations, and trichome with heterocyte

were found more or less similar with the dimensions of *P. alatum* mentioned by Uher (2010). The worldwide distribution of *P. alatum* is given by Maree *et al.* (2018). A species of the genus *Scytonema* i.e. *S. crassum* was merged into the genus *Petalonema* by Migula (1907) as *P. crassum*. This species was collected from wet rocks under the trickling water at Bhandardara in Ahmednagar district. The morphological characters of this species were found similar to *S. crassum* as described in Desikachary (1959) and *P. crassum* as described in Komárek (2013). There are no previous records of this species from Maharashtra, and therefore considered as a new addition to this state. Migula (1907) also separated *Scytonema densum* from the genus *Scytonema* and merged into the genus *Petalonema* as *P. densum*. From the Maharashtra, the only past record on this species is by Kamat (1963) who described it from the moist soil at Takala, Kolhapur. The author has collected *P. densum* from calcareous rocks at Malvan which

is a coastal town in Sidhurg district of Maharashtra. Distribution records of this species indicate its occurrence on humid soils, stones and rocks (Uher, 2010). Except cell length, the current *P. densum* showed slightly more cell width, filament width and heterocyte length and width than in the collected sample by Kamat (1963).

#### Conclusion

Three species of the cyanobacterial genus *Petalonema* are morphologically characterized and described, these are *P. alatum*, *P. crassum* and *P. densum*. These species are collected from calcareous rocks and wet rocks from different localities in Maharashtra (particularly from the Western Ghats). In the present study, two species of *Petalonema*; *P. alatum* and *P. densum* are rediscovered after several decades from the state. The occurrence of *P. crassum* is the first report and a new addition to the cyanobacterial diversity of Maharashtra.

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