Two new species and two new records of *Pterocladiella* (Gelidiales) from Malaysia based on analyses of rbcL and coxI gene sequences

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Five species of *Pterocladiella* (Gelidiales) are reported from Malaysia, identified using a combination of morphological data and partial rbcL (chloroplast) and coxI (mitochondrial) gene sequences. Two new species, *Pterocladiella phangiae* sp. nov. and *Pterocladiella megasporangia* sp. nov., are proposed based on phylogenetic analyses and distinct morphological features. *Pterocladiella phangiae* sp. nov. showed a sister relationship with *P. caerulescens*, *P. beachiae*, *P. psammophila* and *P. australafriicanensis*. *Pterocladiella megasporangia* sp. nov. formed a monophyletic clade with *P. bartlettii* and *P. melanoidea* with strong support in all analyses. Two other species are reported as new records for Malaysia and Southeast Asia. *Pterocladiella beachiae* grouped robustly with *P. beachiae* from the Caribbean Sea; *P. bartlettii* was resolved in a clade among sequences of *P. bartlettii* from the Caribbean Sea and Texas, USA. The identity of *P. caerulescens*; which had previously been reported from Malaysia based on morphological features, was verified by rbcL and coxI gene analyses as well as morphological studies. This study provides 22 new rbcL gene sequences and 18 new coxI sequences for the genus *Pterocladiella*, documents detailed morphological data for the relevant specimens and increases the number of *Pterocladiella* species known in Malaysia from two to six, including two new species.

**Key Words**: Molecular systematics, Morphology, Pterocladiaceae, *Pterocladiella phangiae*, *Pterocladiella megasporangia*

**INTRODUCTION**

The Gelidiales is an order of red algae including economically important species used for agar and agarose production. The Gelidiaceae, the largest family in the modern Gelidiales, was originally established by Kützing (1843) in the order Nemaliales (Hommersand & Fredericq 1988). The family was later elevated to the order Gelidiales by Kylin (1923) based on its diplobiontic life cycle and absence of functional auxiliary cells in the fertilization process (Santelices 1988). Fan (1961) reviewed the order Gelidiales, recognized eight genera in the Gelidiaceae and established the order Gelidiales (Santelices 1998, 1999; Santelices & Hommersand 1997) supported the establishment and validity of *Pterocladiella* and some species of *Geliidiella* and *Gelidium* were transferred (Santelices & Hommersand 1997; Santelices 1998, 2007). In recent years, various studies using a combination of morphological and molecular data have supported the monophyly of *Pterocladiella* (Bailey & Freshwater 1997; Freshwater & Bailey 1998; Shimada & Masuda 2000, 2002; Thomas & Freshwater 2001; Millar & Freshwater 2005; Nelson et al. 2006; Tronchin & Freshwater 2007).

The new family Pterocladiaceae was established for *Pterocladia* and *Pterocladiella* (Perrone et al. 2006), based on the peg-like attachment system in these genera, while *Gelidium* with brush-like tufts of rhizoids and *Geliidiella*, with unicellular rhizoids, were retained in the Gelidiaceae and Geliidiellaceae, respectively. There are currently 17 recognized species in *Pterocladiella* (Guiry & Guiry 2012), of which four, *P. caerulescens* (Kützing) Santelices & Hommersand, *P. caloglossoides* (M.A. Howe) Santelices, *P. capillacea* and *P. taylorii* (Joly) Santelices, have been reported from Southeast Asia (Pham-Hoang 1969; Silva et al. 1987, 1996; Abbott et al. 2002; Tsutsui et al. 2005; Phang et al. 2007, 2008, 2010; Santelices 2007). Of these, only *P. capillacea* has previously been reported from Malaysia based on morphological features, was verified by rbcL and coxI gene analyses as well as morphological studies. This study provides 22 new rbcL gene sequences and 18 new coxI sequences for the genus *Pterocladiella*, documents detailed morphological data for the relevant specimens and increases the number of *Pterocladiella* species known in Malaysia from two to six, including two new species.

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