Phylogenetic relationships of the Bangiales (Rhodophyta) from King George Island, Antarctica : Investigating the evolutionary nature of the Antarctic and sub-Antarctic Bangiales

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Abstract

Members of the Bangiales (Rhodophyta) are distributed worldwide from tropic to Antarctic and Arctic waters. Three species of the Bangiales; *Bangia* sp. (as *B. atropurpurea*), *Pyropia endiviifolia* (as *Porphyra endiviifolium*) and *Wildemania plocamiestris* (as *Porphyra plocamiestris*), have been reported in the Antarctic. Morphological and molecular data were investigated for the Bangiales from the Antarctic and its adjacent waters. Molecular data from over 150 taxa of the Bangiales worldwide including previously published sequences, indicated that the genera *Bangia*, *Dione*, *Porphyra*, *Pyropia*, *Wildemania* and other related genera be recognized in the Bangiales as in the previous molecular study. The *Bangia* sp. from the Antarctic was strongly allied to *B.* sp. from Atlantic Canada, interestingly. *Pyropia endiviifolia* is olive green in color and it allied to a clade with at least three *Pyropia* species from Falkland Islands, Navarino Island, Rio Seco and Punta Arenas, Chile. *Wildemania plocamiestris* growing on other macroalgae in sub-tidal zone grouped into the genus *Wildemania* with the species having one or two cell layers in molecular data. The diversity, taxonomic issues, phylogenetic relationships, distribution and the divergence times of the Antarctic members of the Bangiales were discussed.

Introduction

Bangia and *Porphyra* belonging to the order **Bangiales** are distributed world wide from the Arctic or Antarctic to tropical waters. Three species of the Bangiales have been reported from the Antarctic: *Bangia* sp. (as *B. fuscopurpurea),* *Porphyra plocamiestris* and *Pyropia endiviifolia*, and several species have been added from sub-Antarctic waters (Clayton *et al.* 1997, Kim *et al.* 2001). Recently, the studies of materials from New Zealand, South Africa and sub-Antarctic islands have revealed unexpectedly high generic diversity in members of the **Bangiales** from the southern hemisphere regions (Nelson *et al*. 2006, Sutherland *et al*. 2011). In this study, plastid *rbc*L and mitochondrial *cox*1 gene sequences were examined for six entities of *Bangia* and *Porphyra* sensu lato collected from the Antarctic and Chile in order to get some implications for the phylogenetic relationships with other related members and to estimate the divergence times of the Antarctic Bangiales.

Methods

Maximum likelihood (ML) tree of 168 taxa of the Bangiales and its bootstrap probabilities were estimated with RAxML (v. 8.2.4).

In order to estimate the divergence times of the **Bangiales**, we used three red algal fossil dates, (a) 1,222–1,174 Ma for a stem taxon; i.e. the filamentous and spore-bearing red alga *Bangiomorpha*, (b) 633–551 Ma for Doushantuo fossil-coralline algae, and (c) 137-114 Ma for Cenozoic corallines. Using the ML tree topology obtained with RAxML based on *rbc*L data, we estimated divergence times of ingroup taxa with MCMCTREE.

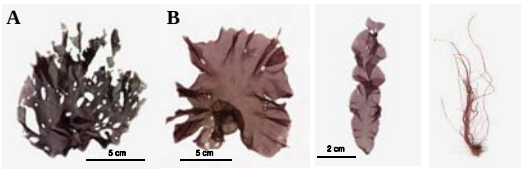

Fig. 1. *Pyropia endiviifolia* and *P. sp.* A: A plant from King George Island, Antarctica. B: A plant from Punta Arenas, Chile.

Fig. 2. *Porphyra woolhousiae* and from Punta Arenas, Chile.

Fig. 3. *Bangia* sp. from the Antartic

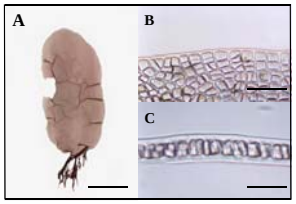

Fig. 4. *Wildemania plocamiestris* from King George Island, Antarctica. A: Habit, B: Surface view of vegetative cells and margin of blade, C: Cross-sectional view of vegetative cells. Scale bars: 2 cm (A), 40 ㎛ (B-C)

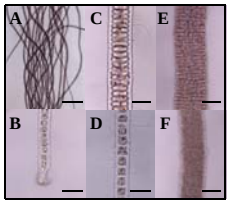

Fig. 5. *Bangia* sp. from King George Island, Antarctica. A: Habit, B: Rhizoidal cells, C: Cells of middle parts, D: Cells of lower part, E: Zygotosporangia, F: Spermatangia. Scale bars = 200 ㎛ (A), 50 ㎛ (B-D), 100 ㎛ (E-F)

Results & Discussion

1. Phylogenetic relationships of the Antarctic Bangiales

The maximum likelihood (ML) phylogeny inferred from the *rbc*L data was shown in Fig. 6. The ML indicated that the genera *Bangia*, *Dione*, *Pyropia*, *Porphyra*, *Wildemania* and other related genera be recognized in the Bangiales as in the previous molecular study.

Interestingly, *Bangia* sp. from the Antarctic was strongly allied to *B.* sp. from Atlantic Canada. It was also different from *B. fuscopurpurea* by 1-2 bp in *rbc*L and from *B. fuscopurpurea* from north Pacific (Korea and Japan) by 11-12 bp in *cox*1 data.

Porphyra woolhousiae from Antarctica, Chile and Falkland Islands grouped into a clade with *Porphyra* spp. from South Africa and Chile.

Pyropia endiviifolia from the Antarctic was different from the material of *Pyropia* sp. from Chile by 18-19 bp in *rbc*L and 28 bp in *cox*1 gene sequences, showing that two materials would be the different species. This species grouped into a clade with *Pyropia* spp. from South Africa, *Py. cinnamomea* and *Py. viridientata* from New Zealand and *Pyropia* spp. from Chile and Falkland Islands based on *rbc*L data.

Wildemania plocamiestris from the Antarctic and Chile which has one cell layer of blade grouped into a clade with *Wildemania* sp. from Falkland Islands and Chile and *P. amplissima* from north Atlantic having two cell layer based on *rbc*L data.

2. Taxonomic issues

Important taxonomic characters such as cell layer, sexuality (monoecious or dioecious), arrangement of reproductive cells (mixed or sectored vertically) which have been concerned as key characters for taxonomy of the Bangiales do not reflect the molecular phylogeny.

3. Divergence times of the Antarctic Bangiales

In running MCMCTREE, the divergence time of the Bangiales from a common ancestor with the Bangiophyceae and the Florideophyceae was calculated as 835 (a, 95% HPD: 794–881) Ma, a mid-Neoproterozoic (Fig. 7).

The major divergences within the Bangiales occurred during the mid–Mesozoic to late–Mesozoic eras beginning with the genus *Porphyra*, with an estimated divergence time of 164 (b, 95% HPD: 137–196 for *Porphyra*) to 110 (c, 95% HPD: 91–126 for *Pyropia*/*Wildemania*) Ma, late-Julassic, Mesozoic.

The divergence times of the Antarctic entities, *Pyropia endiviifolia* and *Wildemania plocamiestris* from a common ancestor with the southern hemispheric species was calculated as 10.83 (d, 95% HPD: 6.33-16.09 for *Pyropia endiviifolia*) and 8.56 (e, 95% HPD: 4.56–12.74 for *Wildemania plocamiestris*) Ma, late-Miocene, Cenozoic. *Bangia* sp. and *Porphyra woolhousiae*, however, would recently diverge from each of their common ancestors during 5.11 (f, 95% HPD: 2.25–8.58) Ma and 1.48 (g, 95% HPD: 0.58–3.41) Ma, late-Cenozoic, respectively.

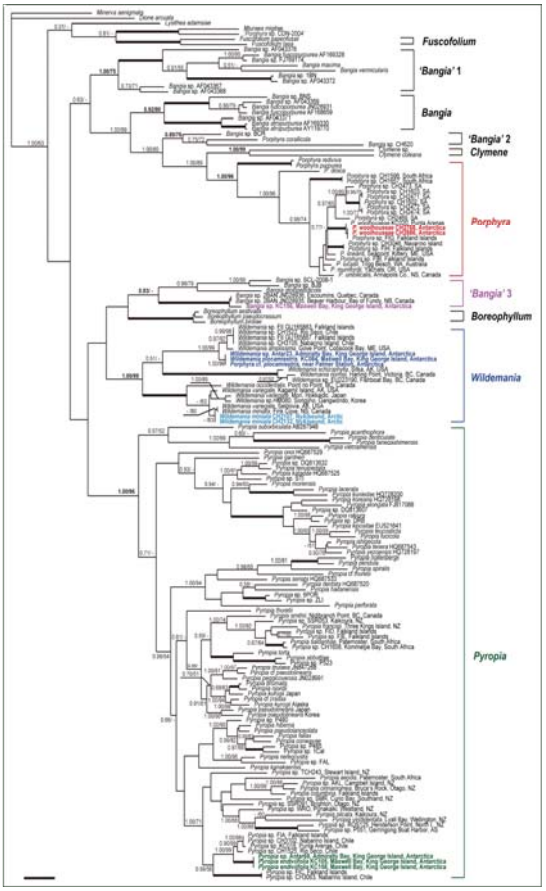

Fig. 6. Maximum likelihood tree constructed with RAxML for the plastid *rbc*L data set (GTR+I+G model). Values at branches represent Bayesian posterior probabilities (left value) and 2000 bootstrap replicates for maximum likelihood (right values) analysis. Branches lacking values received less than 50% support. Scale bar = 0.01 substitutions/site.

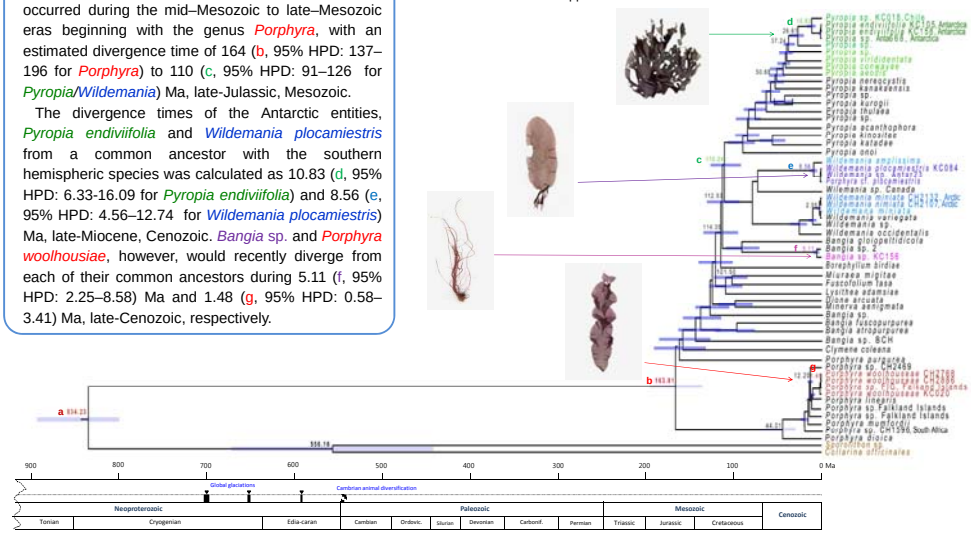

Fig. 8. Divergence times and 95% credibility interval estimated with global clock assumption. Estimated times of major divergences based on *rbc*L gene clock analysis using the best RAxML tree. Branch lengths are proportional to divergence time (i.e., millions of years ago, Ma). Numbers in the right side of each node indicate divergence times [post mean time (95% CI lower and upper time)]. The geologic timeline is given under the chronological timeline in a million year scale. Three global glaciations were hypothesized to have occurred 716-670, 645-635, and 581-579 Ma (three arrowheads in the Neoproterozoic Era). The Cambrian animal diversification occurred approximately 520-543 Ma, at the beginning of the Paleozoic Era.

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