

# A MOLECULAR PERSPECTIVE OF THE LAURENCIA COMPLEX (CERAMIALES, RHODOPHYTA) IN MACARONESIA REGION

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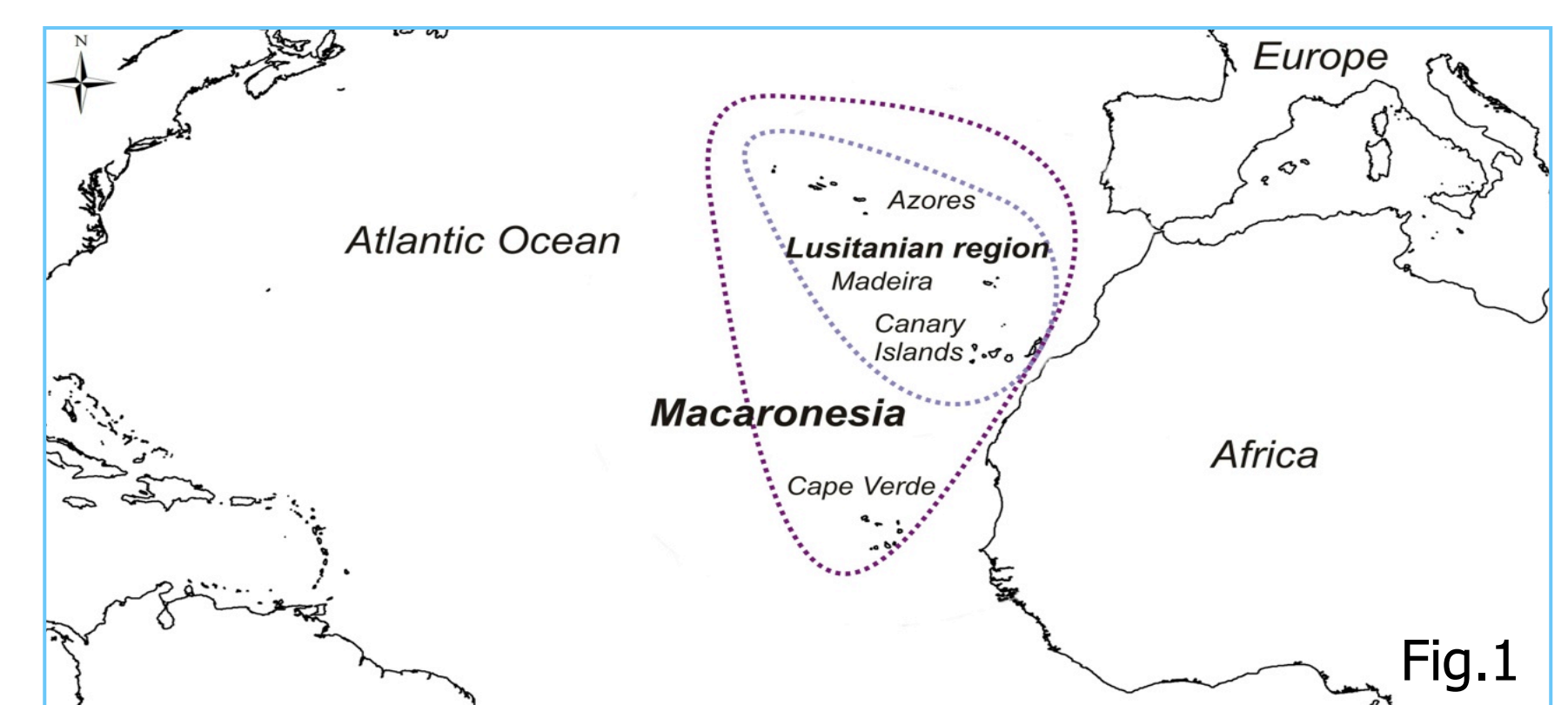


## 1. INTRODUCTION

In the present study, we undertook an integrative approach, using molecular data to assess the diversity of the *Laurencia* complex in Lusitanian Macaronesia (Azores, Madeira and Canary Islands) where speciation events are supposedly common leading to a high endemism. Identification of species of the *Laurencia* complex based on anatomical and morphological characters is extremely difficult due to phenotypic plasticity and overlaps in many morphological characters. As a consequence, among the 28 species reported so far from these Macaronesian archipelagos, 14 species records have been regarded as doubtful. In that context, DNA barcoding appears as a proficient alternative to morphological features for species assignment. We used DNA barcode data, mitochondrial COI gene and partial nuclear LSU marker, as a tool for species delimitation. A third marker, *rbcL* gene, was also studied and phylogenetic analyses were carried out using the combined data set with the aim of inferring the phylogenetic relationships and biogeographic affinities of members of the complex from Macaronesia.

## 2. MATERIALS AND METHODS

Samples of members of the *Laurencia* complex were collected in 2011-2014 in Azores, Madeira and Canary Islands (Fig. 1). Subsamples were dried and preserved in silica gel for molecular analyses. Total DNA was extracted, using the DNeasy Plant Mini Kit (Qiagen, Hilden, Germany). LSU-5' region, COI-5' region and *rbcL* gene were amplified. Sequencing reactions were performed by *Genoscope* and *Macrogen*. Phylogenetic relationships were inferred using the combined data set (LSU, COI and *rbcL*) taking into account the three partitions, with MrBayes v.3.0 beta 4 and PhyML 3.0.



## 3. RESULTS

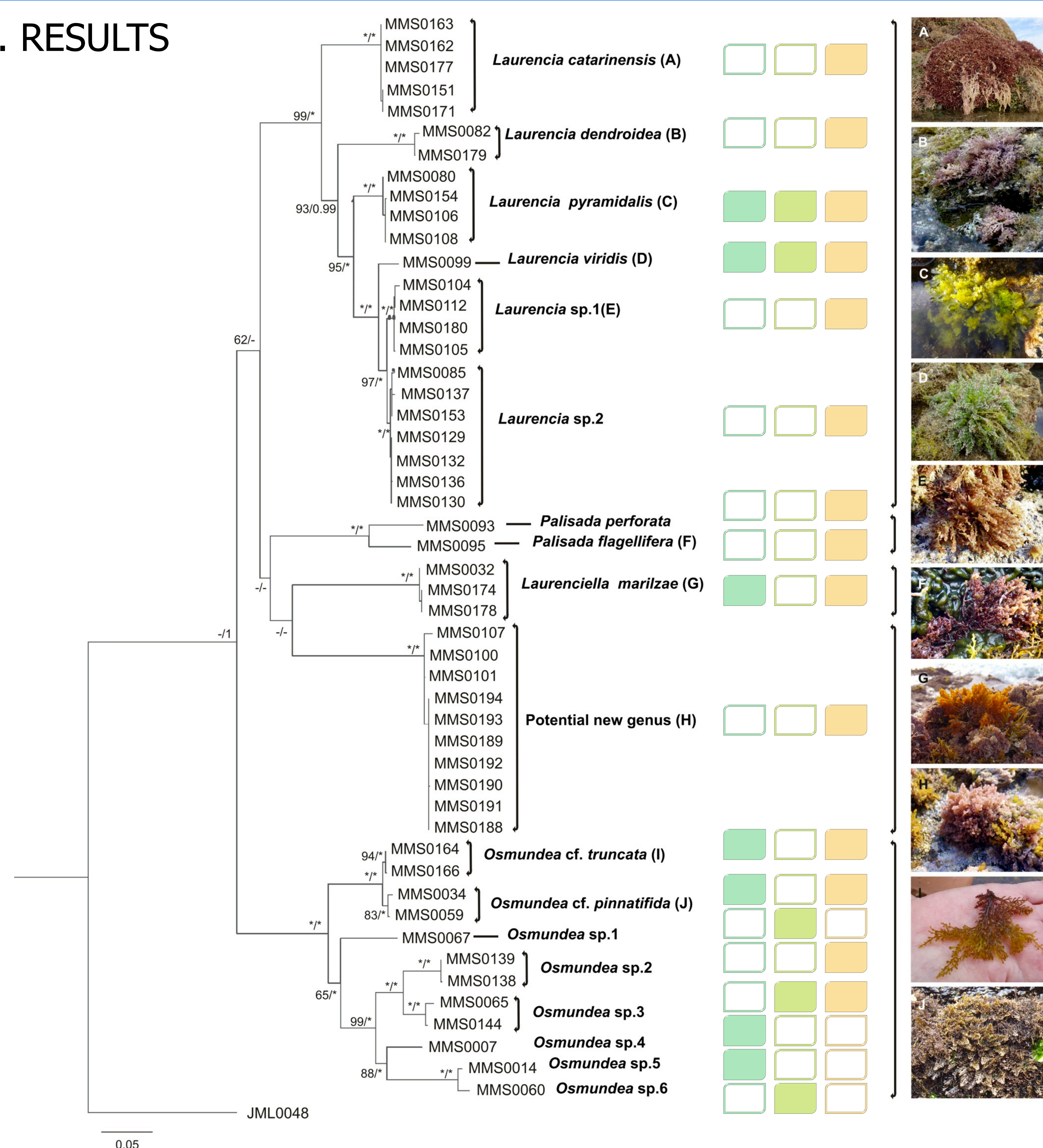


Fig.2. Phylogenetic hypothesis obtained by Maximum Likelihood inference of a data set containing three genes (partial LSU, COI and *rbcL*). Numbers at the nodes indicate Maximum Likelihood bootstrap values followed by Bayesian posterior probability; \* bootstrap=100 and posterior probability=1. Taxa present in Azores archipelago (green), Madeira Island (yellow) and Canary Islands (orange).



Fig.3. (A) Map of Azores (Portugal), C: Corvo, F: Flores, Fa: Faial, P: Pico, SM: São Jorge, G: Graciosa, T: Terceira, SM: São Miguel, Sta. M: Santa Maria. Sites where Macaronesian specimens were collected in the present study: (B) Santa Maria Island, Praia Formosa; (C) Pico Island, Barca-Madalena.



Fig.4. (A) Map of Madeira (Portugal), M: Madeira, PS: Porto Santo, D: Desertas. Sites where Macaronesian specimens were collected in the present study: (B) Porto Moniz, Madeira; (C) Madeira.



Fig.5. (A) Map of Canary Islands (Spain), P: La Palma, H: El Hierro, G: La Gomera, T: Tenerife, GC: Gran Canaria, F: Fuerteventura, L: Lanzarote. Sites where Macaronesian specimens were collected in the present study: (B) Fuerteventura Island, Garcey; (C) Tenerife Island, Puerto de la Cruz.

## 4. CONCLUSIONS

The phylogenetic reconstructions achieved from concatenated alignment of genes LSU, COI and *rbcL* show the presence of 9 known species included the genera *Laurencia*, *Osmundea* and *Palisada*, 1 putative new taxa included in the *Laurencia* genus, 6 in the *Osmundea* genus and one potential new genus.

Our results reveal that the diversity within the *Laurencia* complex has been underestimated and further phylogenetic analysis including more taxa and more markers are needed to assess relationships among different genera.

An integrative approach including morphological and chemical characters in combination with molecular characters would improve our ability to delineate species boundaries in the *Laurencia* complex.