



A new lichen record for the Asia: *Parmelia barroenae* Divakar, M.C. Molina & A. Crespo, Supported by Molecular Data from Turkey

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Abstract

Parmelia barroenae is reported from Turkey and Asia for the first time. Comments on its habitats, substrata, distributional data and macrophoto are provided. In addition, a phylogenetic tree was constructed by comparing the sequence data of the ITS region of closely related species.

Key words: Ascomycetes, Molecular phylogeny, *Parmelia*

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Asya için yeni bir liken kaydı: Türkiye'den moleküler veri ile desteklenen *Parmelia barroenae* Divakar, M.C. Molina & A. Crespo

Özet

Parmelia barroenae türü Türkiye ve Asya'dan ilk kez rapor edilmektedir. Türe ait habitat, substrat, dağılışı ve makro fotoğraf verilmiştir. Buna ek olarak, yakın ilişkili türlerin ITS bölgelerine ait dizi analizleri ile karşılaştırılarak bir filogenetik ağaç oluşturulmuştur.

Anahtar kelimeler: Ascomycetes, Moleküler filogeni, *Parmelia*

1. Introduction

About 1800 lichen and lichenicolous fungi species are known from Turkey (John ve Türk, 2017). In this work done by John ve Türk (2017) all the floristic articles made up to date are collected in a book. Besides lichenological research in Turkey has improved in the last thirty years with many floristic studies published recently about several regions of Turkey (e.g., John, 1992, 1995, 2007; Şenkardeşler, 2011; Halıcı and Aksoy, 2009; Kocakaya et al., 2014; Özdemir Türk et al., 2015).

The lichenized fungal genus *Parmelia* (Parmeliaceae, Lecanorales) foliose thallus, upper surface grey to grey-green or brownish grey, with or without soredia, pustules or isidia. Lower surface black, uniformly rhizinate to margins; rhizines mostly simple or sometimes squarrosely branched, black. Species formerly included in this genus are now placed in *Arctoparmelia*, *Flavoparmelia*, *Hypotrachyna*, *Melanelia*, *Melanelixia*, *Melanohalea*, *Parmelina*, *Parmelinopsis*, *Parmotrema*, *Pleurosticta*, *Punctelia* and *Xanthoparmelia* (Purvis, 1992). Only six species are known belonging to the genus *Parmelia* from Turkey; *P. discordans* Nyl., *P. omphalodes* (L.) Ach., *P. saxatilis* (L.) Ach., *P. squarrosa* Hale, *P. submontana* Nadv. ex Hale, Smiths and *P. sulcata* Taylor (John ve Türk, 2017).

2. Materials and methods

Thallus loosely adnate, 6 cm diam., lobes overlapping and imbricate, short, 2–5 mm wide, older lobes revolute. Upper surface glaucous whitish grey, becoming reticulately cracked. Pseudocyphellae laminal and marginal effigurate, numerous, soralia sparse, granular, medulla white. Rhizines on black lower surface moderately abundant, simple, not squarrose, 1–2 mm long (Figure 1). Specimen Examined: Turkey, Çorum, Laçın district, on *Quercus* sp., 40° 45' 692" N, 34° 52' 169" E., 1020 m., M. Kocakaya, MK-6795.

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Figure 1. Thallus of *Parmelia barrenoae*; MK-6795

Samples prepared from freshly collected were ground with sterile pestles. Total genomic DNA was used extracting the DNA using DNeasy Plant Mini Kit (Qiagen) following the manufacturer's protocols with minor modifications. ITS4 (TCCTCCGCTTATTGATATGC) (White et al., 1990) and ITS1-F (CTTGGTCATTTAGAGGAAGTAA), (Gardes and Bruns, 1993) were used to amplify the ITS sequence. The same regions have been successfully used by Divakar et. al., (2005) in the molecular phylogenetic analysis of the *Parmelia*. PCR-amplification was carried out, following Divakar et. al. (2005). The PCR products were visualized on 1% agarose gel as a band of approx. 500 or 600 bp. The PCR products were subsequently purified using the "ExoSAP-IT™ PCR Product Cleanup Reagent" (ThermoFisher Scientific, USA) according to the manufacturer's instructions. Sequences of PCR products obtained from *Parmelia* samples were performed using the Big Dye Terminator Cycle Sequencing v3.1 (Applied Biosystems, Foster City, CA) following the manufacturer's protocol and analysed on an ABI 3730XL Genetic Analyzer (Applied Biosystems, Foster City, CA).

ITS sequence results were analyzed automatically and manually using samples from Genbank using the Clustal W option in the BioEdit program. Compared with the nuITS rDNA sequence data of the genus-belonging species given in the previous article, which describes the *Parmelia barrenoae*. Out groups *Platismatia glauca*, *Parmelia laevior* and *P. signifera* were used. These out groups were previously described by Molina et al., (2004). Details of the samples and the GenBank accession numbers are listed in Table 1. For phylogenetic tree, MEGA 7 (Molecular Evolutionary Genetics Analysis) program was used (Kumar et al., 2016). Phylogenetic tree was constructed by Maximum Likelihood analysis using Tamura-Nei model (Figure 2) (Tamura and Nei, 1993). Pairwise deletion was performed for deleting data gaps and checking. The tree reliability was tested with 1000 bootstrap replications. Specimen is deposited in Bozok University Lichen Herbarium.

3. Results and discussion

Parmelia barrenoae was described on *Quercus* sp. from Spain (Divakar et al., 2005). These species resembles *P. sulcata*. *Parmelia sulcata* differs in having richly branched, squarrose rhizines and older lobes of *P. barrenoae* are revolute, soralia are only laminar and less developed than those of *P. sulcata* (Divakar et al., 2005). The Turkish population of *P. barrenoae* that we have examined in oak forest at 1020 m in the Central Anatolia Region.

P. barrenoae species was examined in terms of its morphological, anatomical and ecological characters. The ITS sequence of the species was successfully obtained. It was evaluated together with the sequence results from the genBank. The analysis involved 32 nucleotide sequences with outgroups. All positions containing gaps and missing data were eliminated. There were a total of 389 positions in the final dataset. The ITS sequences of *P. barrenoae* collected from the Turkey were blasted with sequences of species from Japan, Portugal and Spain. It is seen that the phylogenetic tree matches with other *P. barrenoae* species (Figure 2).

P. barrenoae is known from North America, Afrika and Poland (Hodkinson et al., 2010; Ossowska and Kukwa, 2016).

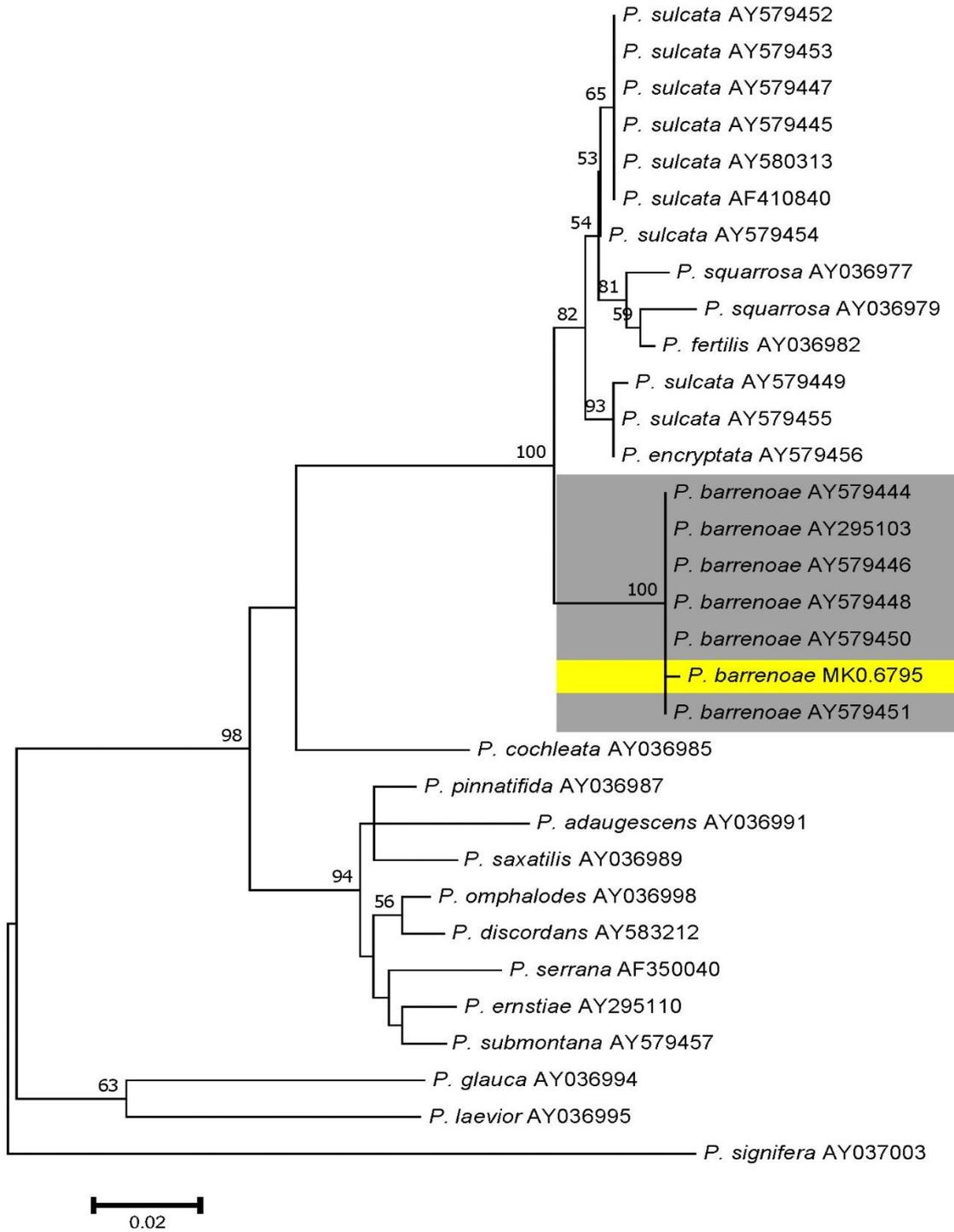


Figure 2. Maximum Likelihood (ML) analysis of the ITS region of *Parmelia barrenoae* and related species. Numbers at tree nodes indicate bootstrap values of ML (only values $\geq 50\%$).

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Table 1. The sequences used in this study were downloaded from the gene bank and the new product was indicated by bold. (The genbank number will be received after the accepted the publication)

Taxa	Locality/Source	GenBank no. (ITS)
<i>P. adaugescens</i>	Japan/GenBank	AY036991
<i>P. barrenoae</i>	Spain/GenBank	AY579444
<i>P. barrenoae</i>	Spain/GenBank	AY295103
<i>P. barrenoae</i>	Spain/GenBank	AY579446
<i>P. barrenoae</i>	Spain/GenBank	AY579448
<i>P. barrenoae</i>	Portugal/GenBank	AY579450
<i>P. barrenoae</i>	Turkey	MK0.6795
<i>P. barrenoae</i>	Spain/GenBank	AY579451
<i>P. cochleata</i>	Japan/GenBank	AY036985
<i>P. discordans</i>	UK/GenBank	AY583212
<i>P. encryptata</i>	Spain/GenBank	AY579456
<i>P. ernstiae</i>	Spain/GenBank	AY295110
<i>P. fertilis</i>	Japan/GenBank	AY036982
<i>P. omphalodes</i>	Spain/GenBank	AY036998
<i>P. pinnatifida</i>	Russia/GenBank	AY036987
<i>P. saxatilis</i>	Russia/GenBank	AY036989
<i>P. serrana</i>	Spain/GenBank	AF350040
<i>P. squarrosa</i>	USA/GenBank	AY036977
<i>P. squarrosa</i>	USA/GenBank	AY036979
<i>P. submontana</i>	Spain/GenBank	AY579457
<i>P. sulcata</i>	Spain/GenBank	AY579452
<i>P. sulcata</i>	Germany/GenBank	AY579453
<i>P. sulcata</i>	Spain/GenBank	AY579447
<i>P. sulcata</i>	Spain/GenBank	AY579445
<i>P. sulcata</i>	Spain/GenBank	AY580313
<i>P. sulcata</i>	Sweden/GenBank	AF410840
<i>P. sulcata</i>	Germany/GenBank	AY579454
<i>P. sulcata</i>	Spain/GenBank	AY579449
<i>P. sulcata</i>	Spain/GenBank	AY579455
<i>P. glauca</i>	Japan/GenBank	AY036994
<i>P. laevior</i>	Japan/GenBank	AY036995
<i>P. signifera</i>	Australia/GenBank	AY037003

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