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DATA NOTE

Complete plastome sequences of two *Psidium* species from the Galápagos Islands [version 1; peer review: 2 approved]

Bryan Reatini¹, Maria de Lourdes Torres², Hugo Valdebenito², Todd Vision ¹¹Department of Biology, University of North Carolina at Chapel Hill, Chapel Hill, North Carolina, 27514, USA²Universidad San Francisco de Quito, Quito, Ecuador

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Abstract

We report the complete plastome sequences of an endemic and an unidentified species from the genus *Psidium* in the Galápagos Islands (*P. galapageium* and *Psidium* sp. respectively).

Keywords

plastome, *Psidium*, Galapagos, guayabillo

Open Peer Review

Approval Status

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version 1
30 Aug 2018	view	view
1. Michael O. Dillon , The Field Museum, Chicago, USA		
2. Carolyn Proença , University of Brasília (UnB), Brasília, Brazil		

Any reports and responses or comments on the article can be found at the end of the article.

Corresponding author: Bryan Reatini (reati1bs@gmail.com)

Author roles: **Reatini B:** Conceptualization, Data Curation, Funding Acquisition, Investigation, Validation, Visualization, Writing – Original Draft Preparation, Writing – Review & Editing; **Torres MdL:** Conceptualization, Project Administration, Supervision, Writing – Review & Editing; **Valdebenito H:** Conceptualization, Investigation, Project Administration, Supervision, Writing – Review & Editing; **Vision T:** Conceptualization, Funding Acquisition, Methodology, Project Administration, Supervision, Writing – Review & Editing

Competing interests: No competing interests were disclosed.

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Introduction

Over a quarter of all vascular plant species are endemic to islands, making them hotspots of plant diversity and conservation (Kreft *et al.*, 2008). In the Galápagos Islands, there are roughly 560 native species of plants of which approximately 32% are endemic (Lawesson *et al.*, 1987). However, many of these endemic species have remained relatively unstudied since they were originally given scientific descriptions, making the study of the evolutionary histories of these unique taxa difficult. In the present study, we constructed the complete plastome sequences of two species of *Psidium* (guava) from the Galápagos Islands, one endemic and one currently unidentified in hopes of facilitating future work on the evolutionary relationships of these species.

Methods

This research is authorized under the permit: MAE-DNB-CM-2016-004 in compliance with Ecuadorian regulations.

Leaf samples were collected during May of 2017 from the Galápagos endemic *Psidium galapageium* Hook (commonly known as guayabillo) on the island of San Cristobal (0.89094°S, 89.43769°W) and from an unidentified *Psidium* species on the island of Santa Cruz (0.62313°S, 90.38581°W). Based on morphological similarity, the *Psidium* sp. individual is suspected to be *P. acidum* (Landrum, 2016), but no reference or barcode sequence from *P. acidum* is available for confirmation.

Leaf tissue was desiccated immediately after harvesting using silica gel. DNA extractions were performed using a Qiagen DNeasy Plant mini kit (Qiagen, Inc.). Sequence data was generated in the form of paired-end, 150 bp reads using a KAPA library prep kit (Roche Sequencing) and sequenced on an Illumina HiSeq 4000 platform (Illumina, Inc.).

Reads were quality and adapter trimmed using *Trim Galore!* version 0.4.3 with a minimum *phred* score value of 20 and minimum read length of 50 bp. Filtered reads were then aligned to the *Psidium guajava* plastome reference available at NCBI (Accession: KX364403) using the *mem* function within *BWA* version 0.7.15 (Li & Durbin, 2009). Consensus plastome sequences were generated using the *mpileup* function within *samtools* version 1.8 followed by the *call* and *consensus* functions within *bcftools* with a minimum depth of coverage of 10x (Li *et al.*, 2009). Using *IRscope* (Amiryousefi *et al.*, 2018), the *P. galapageium* and *Psidium* sp. plastomes respectively were confirmed to contain a large single copy of 88,268 bp and 87,747 bp and a small single copy of 18,465 bp and 18,490 bp separated by two inverted repeats of 26,071 bp and 26,360 bp for total lengths of 158,875 bp and 158,957 bp (Figure 1).

Annotations were generated using the program *Plann* (Huang & Cronk, 2015). Of the 132 gene features annotated previously in the *Psidium guajava* (guava) chloroplast genome on NCBI (Accession: KX364403), all were recovered in the *Psidium* sp. and *P. galapageium* plastome sequences. The non-identity of the two taxa sampled is evidenced by the absolute pairwise sequence divergence of the concatenated sequences of three conserved genes (MatK, psbA, and rbcL) which have been successfully used as barcodes previously in *Psidium* (Kress *et al.*, 2009). Sequences were aligned using MUSCLE within MEGA version 7.0.26 (Tamura *et al.*, 2007), and the number of nucleotide differences were counted between these alignments to estimate divergence. A total of 35 differences were observed among 4011 sites (0.87% uncorrected divergence) between *P. guajava* (Accession: KX364403) and *P. galapageium*, 45 differences (1.1%) between *P. guajava* and *Psidium* sp., and 40 differences (0.99%) between *P. galapageium* and *Psidium* sp.

Figure 1. Genomic structure of junction sites between the long single copy (LSC, light blue), short single copy (SSC, light green), and inverted repeat (IRa and IRb, orange) regions. Proximate genes are shown. The circular genomes have been linearized for illustration.

Data availability

Voucher specimens for *P. galapageium* and *Psidium* sp. are available at the Charles Darwin Research Station herbarium (Index Herbariorum code CDS) with accession numbers 3053515 and 3053562, respectively. The corresponding plastome sequences for *P. galapageium* and *Psidium* sp. are available at NCBI with accession numbers MH491846 and MH491847, respectively.

Competing interests

No competing interests were disclosed.

Grant information

This work was supported by a Louise Coker Fellowship from the University of North Carolina at Chapel Hill (UNC).

All funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

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Open Peer Review

Current Peer Review Status:

Version 1

Reviewer Report 03 October 2018

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Carolyn Proença

Department of Botany , Institute of Biological Sciences, University of Brasília (UnB), Brasília, Brazil

This appears to be a sound paper. Methods are modern, standard and referenced with versions cited when appropriate. Sequences are vouchered by herbarium specimens and have been deposited in Genbank. I have checked the accession numbers given and they are correct. Figure 1 is very clear and detailed, and the legend is adequate and complete.

My most important comment is to point out that very recently, Landrum (2017, in Canotia 13:1-101) has treated *Psidium galapageium* Hook.f. as a synonym of widespread *P. oligospermum* DC. He comments in his paper that "As recognized here *Psidium oligospermum* is a widespread and variable species" and that "A geographically broad study with molecular techniques of *Psidium oligospermum*, including related species ... would be valuable." Reatini's paper relates directly to this comment by Landrum (2017) and the fact that the independent species status of *P. galapageium* is not accepted by all should be made. Other studies however (Proença *et al.* 2014 Flora de Sergipe: Myrtaceae; and Tuler *et al.* 2017, Flora of Espírito Santo: *Psidium* (Myrtaceae). Rodriguesia 68:1791-1805) do not include *P. galapageium* within *P. oligospermum*.

A minor quibble is with authorities for scientific names in the paper, that are either erroneous or missing. *Psidium galapageium* Hook.f., is the correct form (authority erroneously given in the paper as Hook who is actually a different botanist from Joseph Hooker who described the species). *Psidium acidum* (Mart. ex DC.) Landrum (authorities absent in the paper) is how this species should be cited.

Is the rationale for creating the dataset(s) clearly described?

Yes

Are the protocols appropriate and is the work technically sound?

Yes

Are sufficient details of methods and materials provided to allow replication by others?

Partly

Are the datasets clearly presented in a useable and accessible format?

Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Myrtaceae Systematics, Plant Phylogenetics

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

Reviewer Report 03 October 2018

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Michael O. Dillon

Integrative Research Center, The Field Museum, Chicago, IL, USA

This paper reports the results of sequencing efforts of a species native to the Galapagos Islands. The ability to distinguish new species and especially "crypto-species" that are not always obvious due to similar comparative morphologies. The paper should be published and this is the appropriate venue.

Is the rationale for creating the dataset(s) clearly described?

Yes

Are the protocols appropriate and is the work technically sound?

Yes

Are sufficient details of methods and materials provided to allow replication by others?

Yes

Are the datasets clearly presented in a useable and accessible format?

Yes

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Vascular plant systematist

I confirm that I have read this submission and believe that I have an appropriate level of expertise to confirm that it is of an acceptable scientific standard.

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