

Determining *Impatiens* Phylogeny Using Multi-Gene Concatenated Sequences

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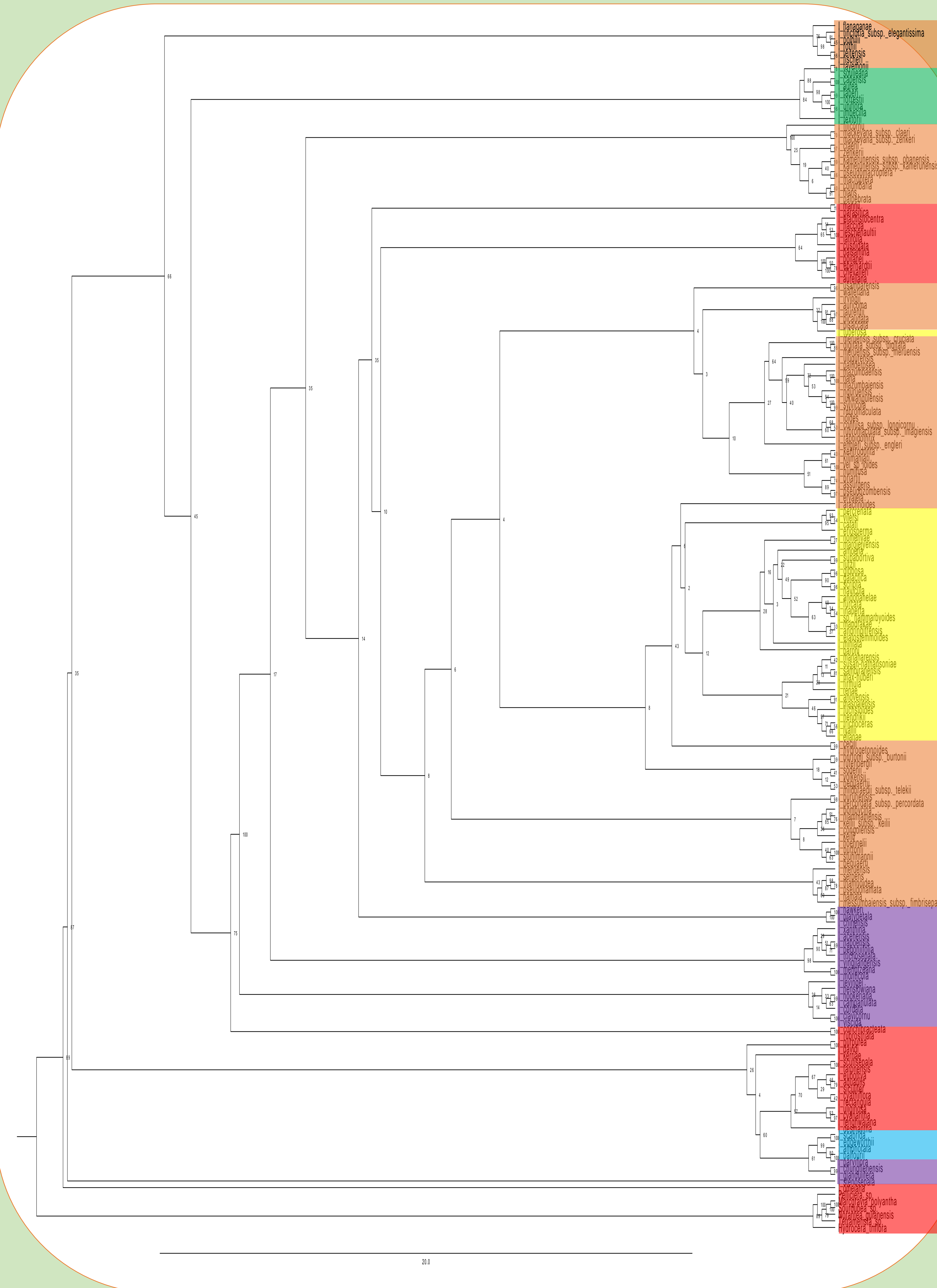
Introduction

The *Impatiens* genus composes the majority of the *Balsaminaceae* family, consisting of over 1000 species in tropical regions of the globe alone. The diversity of the genus is prominent in the Asian and African tropics, with large focus in the Eastern Arc Mountains of Tanzania, where much of the data collection used in this project occurred. Little is known on the diversification process and origins of the *Impatiens* genus, and how the vast differences in pollinators and morphology arose. Diversification has been further outlined and linked to origins in East Asia, followed by spread to Africa on multiple occasions, through previous research (Janssens 2009). Continued research on this topic aims to improve understanding of the evolutionary relationships in this highly diverse genus, through a more definitive phylogenetic tree. With each addition of sequences by a new researcher, there is a clearer understanding of the radiation of *Impatiens* that occurred throughout history. After extraction from leaf tissue samples, the genes focused on for this purpose are known as the Internal Transcriber Spacer (ITS), RNA Polymerase Degradation Factor 1 (DEF1), and RNA Polymerase Degradation Factor 2 (DEF2) genes. Combining our new sequences with already published sequence data (Yuan et al 2004, Janssens et al 2009, Ravelivololona 2018), we were able to establish a more robust phylogenetic tree for many of the species in the genus *Impatiens*.

Methods

Molecular Genetics: Silica dried leaves previously collected in Tanzania were selected by species and put through the CTAB DNA extraction protocol alongside herbarium specimens from the Missouri Botanical Garden. The protocols set forth by ²Janssens et al (2009) and ³Yuan et al (2004) were implemented for primer additions, thermocycling, Big Dye cycle sequencing reaction, and PCR cleanup. At the Field Museum in Chicago, IL the Applied Biosystems 3730 DNA Analyzer was utilized for sequencing.

Data Analysis: CodonCodeAligner (CodonCode Corporation) was used for chromatogram reading and generating consensus sequences. Sequences were aligned using MAFFT (Katoh, Rozewicki 2002) and sequences were visualized using AliView (Larson, A. 2014). Sequences were concatenated using Seqkit (Shen et al 2016) and a maximum likelihood tree search was performed with three partitions under GTRCAT model of evolution using RaXML (Stamatakis 2014) via CIPRES (Miller et al 2010) and the maximum likelihood tree was visualized and edited using FigTree v1.4.3 (Rambaut 2007).



Key:

- E & SE Asia
- Africa
- Madagascar
- N America
- Europe
- S India & Sri Lanka



Hypothesis

- The genus *Impatiens* will be determined to be monophyletic rather than polyphyletic, with more than one lineage of origin for the approximately 200 species used for this phylogenetic tree
- Dispersal of *Impatiens* into the Eastern Arc Mountains should have occurred more than once across the history of the genus, making the *Impatiens* of that region polyphyletic

Discussion

Regarding major clades and distributions, the results here are relatively similar when combined to those organized previously by Janssens et al and Yuan et al. However, through concatenation of multiple genes, this phylogenetic tree gives a fuller picture than analysis of a single gene that had been done previously. While this phylogenetic tree does not include the entirety of African or global *Impatiens*, it is a large enough sample size to determine at least some of the dispersals of the genus from Asia to Africa and Madagascar. The continued study of the process of lineage diversification is not only critical to the understanding of the *Impatiens* genus but to the broader spectrum of plant evolution, and further understanding of speciation of such a genus such as this could give us a clearer picture of how the planet will look after potential large geographical shifts in the future.



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Literature Cited

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