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How many Lauraceae species occur in the ARIE Biological Dynamics of Forest Fragments Project (BDFFP)?

Projeto em andamento 2017 - 2019

About the project

In Brazilian Amazon, Lauraceae is represented by 441 species, distributed in 24 genera (Quinet et al., 2016) which is certainly an underestimate given the deficits in Amazonian botanical knowledge (Hopkins, 2007). High diversity and low taxonomic knowledge of Lauraceae is one of the greatest challenges in forest inventories and in taxonomic, ecological and evolutionary studies in the Amazon. Recent studies of Lauraceae show that the circumscription of species in this group is virtually impossible without the use of molecular data, and that the boundaries between species in the Manaus region are still unreliable (Carvalho, 2017; da Mata, 2015; Rivera-Parada, 2017).

Inside 94 ha of plots belonging to ARIE Biological Dynamics of Forest Fragments Project (BDFFP) have been studied forestry dynamics, and effects of forest fragmentation in the last 30 years (Laurance et al., 2011). Currently, BDFFP data has 10469 recorded plants and 1172 specimens of Lauraceae, belonging to 14 genera and 160 species. Of this data base, 64 morphotypes, 11 of which not even the genus is known. However, even with all the data currently collected for Lauraceae, there is a considerable deficiency in the number of samples per species in silica gel. Extremely difficult identification of Lauraceae individuals at genera and species level by the traditional method of identifying samples (Gomes et al., 2013), in one of the region's best known botanically of the Amazon as Manaus, indicates that is imperative adding other sources of information (Near Infrared Spectroscopy-NIR and Leaf Geometric Morphometric Analyses-SHAPE) which could accompany complementary the molecular data. In this context, It is proposed to review local delimitation of Lauraceae species occurring in the permanent plots of ARIE Biological Dynamics of Forest Fragments Project (BDFFP) with the aid of molecular, spectral and morphological data, improving process of species identification and quality of the data monitored in the BDFFP to contribute to the taxonomy of Amazonian Lauraceae.

Left: Ocotea rhynchophylla, bark. Right: Ocotea cernua, leaf

Specifically, we address two questions:

1. Are Lauraceae species in BDFFP correctly delimited and identified?
2. Do these species correspond to distinct phylogenetic lineages?

To address these questions, the goals are:

1. Review the local delimitation and taxonomy of Lauraceae species occurring in the permanent plots of ARIE Biological Dynamics of Forest Fragments Project (BDFFP), adding molecular, spectral and morphological evidence.
2. Produce a molecular phylogeny for the Lauraceae species occurring in the BDFFP.
3. Review the taxonomy of Lauraceae species delimited in BDFFP and describe new species.
4. Produce an illustrated identification guide for these species.

Given that in the BDFFP area many Lauraceae species coexist, molecular differences will be robust evidence of reproductive isolation between species, because the problem of geographic variation is inexistent (Mayr, 1992). Therefore, this study will adopt the perspective of the biological concept of species (BCS) to review the delimitation of Lauraceae species. In this context, if the samples of the same species form a monophyletic clade, the morphology, monophyly itself and their coexistence will clearly indicate species limits, since they will together indicate reproductive isolation.

This research is supported by Biological Dynamics of Forest Fragments Project (BDFFP) and CTFS ForestGEO of Smithsonian Tropical Research Institute.

Field site

All research is conducted at the ARIE Biological Dynamics of Forest Fragments Project (BDFFP), in the Manaus city, Amazonas state of Brazil. The site was established in 1985 by Instituto Nacional de Pesquisas da Amazônia (INPA)Instituto Nacional de Pesquisas da Amazônia and Smithsonian Institution (SI)BDFFP plots. The plots around 94 ha are located on lowland forest. BDFFP is the most

active research sites in the Central Amazon and has had more than 200 research projects to date.

Tool's project

Morphological data - SHAPE. To date, BDFFP database has shown that we have around, 160 species, in 14 genera and 10469 individuals os Lauraceae sampled. This numbres including 96 published names and 64 morphotypes. In addition, we have all information added from other Lauraceae projects, such as in *Aniba*, *Licaria*, *Ocotea* genera, and others projects about morphological information worked inside BDFFP project. In this context, one of our perpective is to use and to take advantage of several vegetative exsicates with along its information, and then to integrate it to improve the delimitation and identification process.

Left: Patricia Lira selecting the best leaf - complete leaf to SHAPE process. Right: Printing leaf as first part of SHAPE process

On this part, Patricia Lira trying to select the best sample, due to is important to have "complete leaf" in the best condition - format, apex, base, nerves- and to give it a same scale for each leaf worked, and finally to complete SHAPE process.

After that, we are going to integrate the SHAPE leaf, based on Fourier elliptic coefficient implemented in **SHAPE software**
for trying to differentiate Lauraceae species, and the same time adding morphological data through **Labotam-Plantas PDBFF**
as leaf's format, leaf's apex, leaf's base, nerves, tricomes, and another ones

Spectral data - NIR The Amazon forest has a high diversity, principally tree families as Lauraceae,

where many species coexistence and are closely related at local scale. Particularly, Lauraceae has many species difficult to delimit and identify. On this context, novelties tecnologies and less invasive such Fourier-Transform Near-Infrared (FT-NIR) spectroscopy is a very utility technique (Foley et al., 1998; Durgante et al. 2013) and generating a reliable discriminate analysis to identify species based on spectral data, through of prediction models that can to be express in different graphic forms. In the most of botanical studies has been used since bark and leaf (fresh and/or dry). Recently studies such Prata et al. (2018) are showing there are high correlation between phylogenetic and spectral distances, support a strong phylogenetics information.

Left: Exsicates samples BDFFP project-INPA Brazil. Right: FT-NIR office - INPA Herbarium (acervo II)

On this project are being used many botanical material collected through different related project to BDFFP, which main a good opportunity to take advantage to spectral data and integrate another vegetative data like **SHAPE software**
to generate identifications best supported.

We are using Fourier Transform Near-Infrared (FT-NIR) Spectroscopy Analyser from Thermo Fisher Scientific model Antaris II controlled by Result Software, localated in the INPA Herbarium (Manaus, Brazil). We are sampling 4 spectral data (2 abaxial leaf, 2 adaxial leaf) for each exsicate, and 8 individuals as minimal sample for species/morphotype.

Molecular data - SILICA

Field Work 2018

On this 2018, we did only one field work inside ARIE-BDFFP, on Porto Alegre Camp. We collected 55 Lauraceae samples (exsicates and silica), which only 3 were collected with flowers and bottom flowers: TAG 3114-350, 3114-7143, 3209-214. All others are sterile. After many time, it was collected species like: Ocotea sandwithii and Williamodendron spectabile, which it has few individuals in other areas.

The team

Marisabel Ureta Adrianzén

She is a M.Sc. student of PPG-Botânica/INPA at Instituto Nacional de Pesquisas da Amazônia. She received her Master's degree in Biodiversity in Tropical Areas and Conservation in 2013 (UIMP-Spain). Her master's research focused on Myristicaceae family and its aboveground biomass input in Peruvian amazon. Nowadays, Marisabel has returned to the Amazonia, in this time in Brazil, and has begun a challenging study in Lauraceae family, considered the most complicated family at the taxonomic level. Her study, tries to integrate three kinds of tools such as molecular, spectral and morphological data from all species of Lauraceae collected on BDFFP project. She is conducting her dissertation research "How many Lauraceae species occuring on permanent plots of BDFFP?" at the ARIE Biological Dynamics of Forest Fragments Project, where almost all permanent plots are inside of terra firme forest.

Alberto Vicentini

(Advisor) He is botanical researcher at Instituto Nacional de Pesquisas da Amazônia, PhD on Ecology, evolution and systematic from University of Missouri (UMSL-USA). His research focuses on delimiting species in *Pagamea* (Rubiaceae) from context of phylogenetic and phenetics patterns to geographical context. His current research focuses on the flora on white-sand forest related of Amazonia, studying species complexes in *Pagamea* and Lauraceae principally, interested in permanent plots of terra firme forest on BDFFP, managementing software and database to improve the availability of taxonomic data of flora and fungi, and driving to apply the near infrared spectroscopy as a effective tool on identification of Amazonian flora.

Patricia de Azevedo Lira

(Associate student) She is a undergraduate student conducting an associated research project about Leaf geometrical morphometric analyses of Lauraceae. Currently, she already has collecting more of 300 leaf shape of Lauraceae species from BDFFP, in order to understand and to answer how this morphological information could allow to differentiate among species, which would be a great and extra goal in difficult species in its identification as Lauraceae. This research will also serve to her Biology graduation thesis (TCC). Leaf shape information from Patricia's project and another vegetative and reproductive information already available from BDFFP, will help improve the circumscription such as efficient morphological tool for support of the project.

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Last update: **27/46/2018 16:46**