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Discrimination of taxonomic identity at species, genus and family levels using Fourier Transformed Near-Infrared Spectroscopy (FT-NIR)

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ABSTRACT

Fourier Transformed Near-Infrared Spectroscopy (FT-NIR) has previously been shown to be effective in species discrimination of plant species, this prompted us to ask if higher taxonomic levels could also be discriminated, and if discrimination based on branch pieces would be equally efficient or better than based on leaves. We tested this with a sample of 384 branches and 349 leaves of 40 Amazonian species. We obtained spectral readings of dry branch and leaf material, and compared the rate of correct predictions of species, genera and family with a classifier based on Linear Discriminant Analysis (LDA). Discrimination of species, genus and family with Fourier Transformed Near-Infrared Spectroscopy (FT-NIR) was good using either branches or leaves. We obtained an average of 90.8% correct species identifications over all species based on branch FT-NIR profiles, and 94.1% based on leaves. Also, we obtained more than 95% correct genus and family identifications. Most of the identification errors occurred among species, genera and families of distinct clades. Near-infrared spectroscopy has great potential for discriminating species from branch samples and is suitable to discriminate a diverse range of genera and families of Amazonian trees.

1. Introduction

Conservation of tropical forests and trees, as much as sustainable management (Sarmiento et al., 2011) require accurate species identification (Hopkins, 2005). However, correct species identification requires a high level of taxonomic expertise, confirmation by specialists and the presence of reproductive material (Mori and Cunha, 1995), which hinder large scale accurate identification of plants in hyper-diverse areas such as the tropics (Hopkins, 2005). Most plant collections from plot-based inventories are sterile specimens, what increases the probability of misclassification (Gomes et al., 2013). According to Ferreira and Hopkins (2004), it is common that in inventories many different species are grouped under a single name due to high morphological similarity.

The classic use of morphological traits for species identification is limited since it does not consider either phenotypic plasticity or the existence of cryptic taxa (Gomes et al., 2013). Therefore, it is necessary to develop new research tools to improve identification. One such tool is Fourier Transformed Near-Infrared Spectroscopy (FT-NIR) (Pastore et al., 2011; Lang et al., 2015), a highly cost-effective non-destructive technique, that is fast and requires no pre-treatment of samples (Pastore et al., 2011; Fernández et al., 2011).

Applications of Fourier Transformed Near-Infrared Spectroscopy (FT-NIR) can be found in all areas of science. In forestry science, FT-NIR has been used to predict the chemical, physical, and mechanical properties of wood (Schimelck et al., 2003; Tsuchikawa and Kobori, 2015). The technique has also been successfully used to determine plant geographical provenance (Sandak et al., 2011; Li et al., 2012). Recently, FT-NIR has been revealed as a promising tool in the discrimination and identification of species in many biological groups. Close animal species (Tolleson et al., 2005; Johnson et al., 2013) can be discriminated by NIR spectroscopy of their faeces. FT-NIR spectroscopy of plant leaves has been shown to be very accurate for discrimination of plant species (Krajsek et al., 2008; Castillo et al., 2008; Fan et al., 2010; Durgante et al., 2013), even at different developmental stages (Lang et al., 2015). The method also has also been demonstrated as a tool to assist discrimination of woods (Muñiz et al., 2012), even within complex and highly similar groups such as mahogany (Pastore et al., 2011; Braga et al., 2011). Furthermore, other spectroscopic techniques based on spectral data have been used to identify species (Asner and Vitousek, 2005; Féret and Asner, 2011) or their chemical properties (Asner et al., 2014).

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Table 1
Results of discriminant analysis for the species level. The third and fourth columns are the number of specimens used to obtain FT-NIR spectra. Accuracy of predictions shows the percentage of correct identifications for each species, based on leaf or branch models that used all wavelengths of the NIR spectrum. The average accuracy over all species is shown in last line of the table.

Linear Discriminant Analyses (LDAs) Model: Function generated with 2/3 of the total sample, branches or leaves. Validation: the remaining 1/3 of the total sample

		#specimens		Prediction accuracy	
Family	Species	Branch	Leaf	Branch model	Leaf model
Annonaceae	*Duguetia flagellaris Huber*	24	23	96.43	95.5
Burseraceae	*Protium grandifolium Engl.*	5	5	100	97
	Protium hebetatum D.C. Daly	35	33	94.2	93.33
	Protium pilosissimum Engl.	7	7	99	100
	Protium pilosum (Cuatrec.) Daly	7	4	60.5	100
	Tetragastris panamensis (Engl.) Kuntze	6	7	84	100
Chrysobalanaceae	*Licania caudata Prance*	5	5	34	100
	Licania hirsuta Prance	6	6	29	84
	Licania micrantha Miq.	13	13	92.33	96.67
	Licania occultans Prance	8	8	71	74.5
Euphorbiaceae	*Senefeldera macrophylla Ducke*	16	4	100	100
Fabaceae	*Inga obidensis Ducke*	10	10	93	83
	Zygia ramiflora (F. Muell.) Kosterm.	6	5	88.67	100
Lecythidaceae	*Corythophora alta R. Knuth*	7	6	83	89.33
	Eschweilera coriacea (DC.) S.A. Mori	7	7	75	88
	Eschweilera pedicellata (Rich.) S.A. Mori	5	5	68.5	72.75
	Eschweilera tessmannii R. Knuth	8	7	97	100
	Eschweilera truncata A.C. Sm.	16	16	94	100
	Eschweilera wachenheimii (Benoist) Sandwith	9	11	89	84.5
Malvaceae	*Theobroma silvestre Spruce ex K. Schum.*	10	10	89.67	100
Melastomataceae	*Henriettella caudata Gleason*	11	11	100	100
Moraceae	*Brosimum rubescens Taub.*	10	10	88.67	91.33
	Helianthostylis sprucei Baill.	12	12	98.33	98.33
	Naucleopsis caloneura (Huber) Ducke	7	7	99	100
Myristicaceae	*Iryanthera coriacea Ducke*	7	7	100	97
	Virola calophylla (Spruce) Warb.	6	6	99	100
	Virola sebifera Aubl.	10	9	81	59
Myrtaceae	*Eugenia* sp. *nov.floraducke*	10	10	100	100
Rubiaceae	*Psychotria astrellantha Wernham*	7	7	100	99.5
Rutaceae	*Adiscanthus fusciflorus Ducke*	16	14	95.25	93.25
Salicaceae	*Ryania pyrifera (Rich.) Uittien & Sleumer*	5	5	97.5	100
	Ryania speciosa Vahl	8	8	95	100
Sapotaceae	*Chrysophyllum sanginolentum (Pierre) Baehni*	14	6	97	100
	Micropholis guyanensis (A. DC.) Pierre	10	8	94.67	99.5
Siparunaceae	*Siparuna cristata (Poepp. & Endl.) A. DC.*	7	6	100	100
	Siparuna decipiens (Tul.) A. DC.	4	4	100	95
	Siparuna poeppigii (Tul.) A. DC.	4	4	100	100
Urticaceae	*Pourouma ovata Trécul*	9	8	98	100
Violaceae	*Paypayrola grandiflora Tul.*	7	7	83	100
	Rinorea racemosa (Mart.) Kuntze	10	8	85.67	100
	Total of specimens	384	349		
	Percentage of correct identification over all species			90.8	94.1

The previous successes in the use of FT-NIR spectroscopy for species discrimination prompted us to ask if higher taxonomic levels could also be discriminated. Some studies indicate that this is possible for classes or clades in angiosperms (Carballo-Meilan et al., 2016; Cavender-Bares et al., 2016), and a small demonstration (only three genera) of the possibility of separating genera (Gorgulu et al., 2007). Still open is the question of how much consistency there is in the signal of genera and families so these can also be predicted by FT-NIR spectroscopy. These previous studies also have examined few clades, and therefore a generalization of the discrimination capacity of FT-NIR spectra for higher levels of phylogeny is still lacking.

Most analyses of plants for discrimination have been based on leaves. The use of branch samples for taxonomic analysis in FT-NIR may provide some advantages compared to leaves, as leaves are generally more variable and plastic than wood. It is well known that leaves at different developmental stages may differ in cell wall thickness, and chemical composition (Raven et al., 2001; Dhugga, 2001), and these could change the spectral response (Lang et al., 2015; Wu et al., 2016). Beyond variability, it is well known that throughout development leaves are cumulatively contaminated by fungi and bacteria, both internally and externally (Eigenbrode and Espelie, 1995; Flaishman et al., 1995; Bringe et al., 2006), and this contamination can affect the structure and chemical composition, thus modifying leaf spectral properties (Ashourloo et al., 2014). Branches on the other hand are expected to have lower incidence of contamination, since their inner tissues are protected by bark, what would result in a cleaner spectral response compared to leaves.

As for the leaves, spectral FT-NIR measurements of branches can be obtained from dry material, which allows larger number samples already collected and deposited in herbarium collections to be analyzed or reanalyzed, as only a small branch piece (~0.5 cm in length and ~0.35 cm in diameter) is needed, which will not cause damage to voucher specimens. The advantage of branch samples for spectral readings of herbarium samples is that a piece can be obtained from samples that have been glued or tightly sewn, what is difficult or impossible for leaves.

Considering the importance of correct identification for conservation, sustainability and for knowledge about biodiversity, this study aims to analyze the potential of FT-NIR spectroscopy to discriminate species, genera and families of a diverse group of Amazon trees using

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