

## ORIGINAL ARTICLE

# Fast and novel botanical exploration of a 320-km transect in eastern Amazonia using DNA barcoding

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## ABSTRACT

We explored a 320-km transect in the Tumucumaque mountain range along the border between southern French Guiana and Brazil, sampling all trees and lianas with DBH  $\geq 10$  cm in seven 25 x 25-m plots installed near seven boundary milestones. We isolated DNA from cambium tissue and sequenced two DNA barcodes (*rbcLa* and *matK*) to aid in species identification. We also collected fertile herbarium specimens from other species (trees/shrubs/herbs) inside and outside the plots. The selected DNA barcodes were useful at the family level but failed to identify specimens at the species level. Based on DNA barcoding identification, the most abundant families in the plots were Burseraceae, Fabaceae, Meliaceae, Moraceae, Myristicaceae and Sapotaceae. One third of the images of sampled plants posted on the iNaturalist website were identified by the community to species level. New approaches, including the sequencing of the ITS region and fast evolving DNA plastid regions, remain to be tested for their utility in the identification of specimens at lower taxonomic levels in floristic inventories in the Amazon region.

**KEYWORDS:** DNA barcoding, French Guiana-Brazil border, *matK*, *rbcLa*, tree inventory, Tumucumaque

# Exploração botânica rápida e inovadora de um transepto de 320 km no leste da Amazônia usando código de barras de DNA

## RESUMO

Um transepto de 320 km foi explorado na Serra do Tumucumaque, ao longo da fronteira entre o sul da Guiana Francesa e o Brasil por meio da amostragem de todas as árvores e lianas com DAP  $\geq 10$  cm em sete parcelas de 25 x 25 m instaladas perto de sete marcos fronteiriços. Isolamos DNA de tecido cambial e sequenciamos dois códigos de barra de DNA (*rbcLa* e *matK*) para auxiliar na identificação das espécies. Também coletamos espécimes de herbário férteis de outras espécies (árvores/arbustos/ervas) dentro e fora das parcelas. Os códigos de barra de DNA selecionados foram úteis em nível de família, mas não conseguiram identificar espécimes em nível de espécie. Com base na identificação de DNA *barcoding*, as famílias mais abundantes nas parcelas foram Burseraceae, Fabaceae, Meliaceae, Moraceae, Myristicaceae e Sapotaceae. Um terço das imagens de plantas amostradas postadas no website iNaturalist foram identificadas em nível de espécie. Novas abordagens, incluindo o sequenciamento da região ITS e regiões de DNA plastidial de rápida evolução, ainda precisam ser testadas quanto à sua utilidade na identificação de espécimes até níveis taxonômicos mais baixos em inventários florísticos na região amazônica.

**PALAVRAS-CHAVE:** código de barras de DNA, fronteira Guiana Francesa-Brasil, inventário de árvores, *matK*, *rbcLa*, Tumucumaque

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## INTRODUCTION

The Amazon region harbours the richest flora of the planet, yet many areas remain under-collected (Prance *et al.* 2000; Hopkins 2007). As a result, there is still considerable uncertainty about the total number of tree species occurring in this region (Cardoso *et al.* 2017; Ter Steege *et al.* 2019). In eastern Amazonia, the upper Jarí River is one of the most poorly sampled areas, which is mostly due to the challenging terrain of the Tumucumaque mountain range (Ter Steege *et al.* 2013; Zizka *et al.* 2018), where the tallest trees of Amazonia were recently detected (Gorgens *et al.* 2019).

In 2015, a survey team hiked 320 km along the border between French Guiana and Brazil, from the trijunction point with Suriname towards the Oiapoque River (Figure 1). The goals of the expedition were to clarify the exact location of the border (Le Tourneau *et al.* 2016), to explore the Tumucumaque mountain range, and to test the feasibility of a rapid botanical inventory based primarily on tissue collections and techniques requiring only lightweight equipment. In the past, progress in the knowledge of Amazonian flora has been

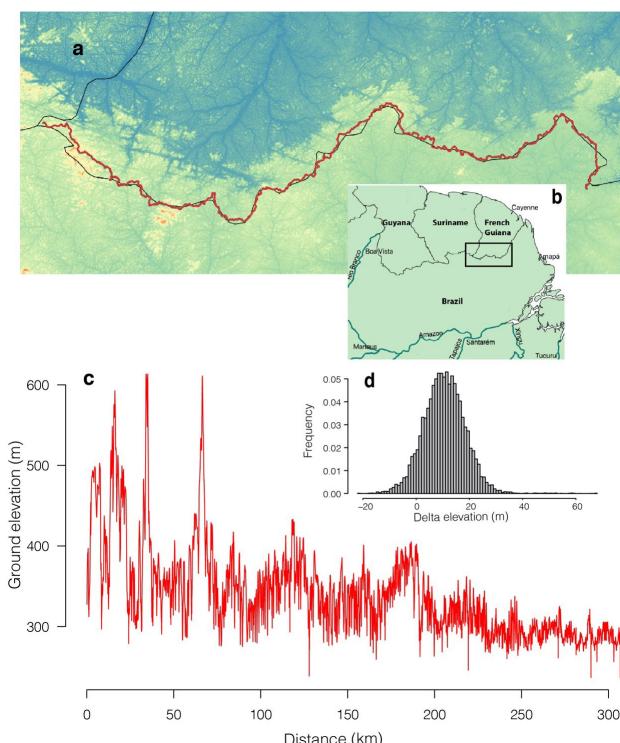
obtained by establishing transects (Tuomisto *et al.* 2003; Pitman *et al.* 2008) and by setting up permanent sampling plots (Blundo *et al.* 2021). However, the French Guiana-Brazil border remains largely underexplored.

In the RAINFOR network of permanent sampling sites (<http://www.rainfor.org/en/map>), there is a dearth of data between the mouth of the Jari River and the Nouragues Research Station, which are more than 500 km apart. The Amazon Tree Diversity Network includes more information on the tree diversity of the Tumucumaque mountain range but is limited to the trijunction region (<https://atdn.myspecies.info/node/2456>). This part of the border between Brazil and French Guiana had been surveyed by the French National Geographical Institute (Institut Géographique National - IGN) in 1956-57. Later, in 1961-62 the binational Brazil/France border commission oversaw the construction of seven milestones on selected sites to demarcate the border (Le Tourneau 2017).

One method to facilitate the identification of sterile plant material is DNA barcoding, which consists of extracting and sequencing short orthologous DNA sequences for each collected plant, and comparing the obtained sequences to a publicly available reference database (GenBank, maintained by the National Center for Biotechnology Information; <https://www.ncbi.nlm.nih.gov/genbank/>). This method has proven to be effective for the identification of animal species (Hebert *et al.* 2003), due to the existence of a mitochondrial DNA region called cytochrome oxidase 1 (CO1). CO1 is a good DNA barcode because it is short enough for Sanger sequencing; it can be sequenced using the same pair of primers flanking the sequence in a wide range of taxonomic groups, and it is variable enough to discriminate between sister species (Hebert *et al.* 2003).

In plants, the search for universal DNA barcodes has been more difficult, and several strategies have been tested specifically for Neotropical plants (Gonzalez *et al.* 2009; Kress *et al.* 2009). The goal of this contribution is not to debate the utility of DNA barcodes for plant identification, but rather to use this approach to aid the taxonomic identification process. Hollingsworth *et al.* (2009) recommended the use of a combination of two plastid DNA regions, the first part of the *rbcL* gene (henceforth *rbcLa*), and a large fragment of the *matK* gene. Recently, Lima *et al.* (2018) conducted a survey on the publicly available DNA sequences from tree species of the flora of São Paulo state in Brazil and generated new sequences of three of the most widely used plant DNA barcodes (*rbcL*, *matK* and ITS) for 609 tree species of that flora. However, they did not assess the identification potential of the DNA barcodes they surveyed.

The primary goal of this contribution was to evaluate the potential of the DNA barcoding approach to aid in the taxonomic identification process in a poorly known tropical



**Figure 1.** Environmental context of the 320-km transect. A – Transect route (in red) overlaid on a topographic map of Eastern Amazonia (SRTM product at 1 arc second resolution; downloaded from the USGS Earth Explorer website); blue-to-red colour ranges from 125 to 775 m a.s.l. within the study area; B – Location of the transect area along the border between southern French Guiana and Brazil; C – Ground elevation along the transect measured by a hand-held GPS unit; D – Plot of the difference between remote-sensed ground elevation (SRTM) and ground elevation (mean difference =  $10.6 \pm 7.9$  standard deviation). This figure is in colour in the electronic version.

area, and to highlight the use of samples from cambium tissue for this purpose. Cambium tissue, as an alternative to collecting inaccessible canopy leaves, has already been used in previous DNA barcoding studies (Colpaert *et al.* 2005; Tibbits *et al.* 2006; Gonzalez *et al.* 2009; Novaes *et al.* 2009). We report on the results of our botanical exploration in this little explored area of Amazonia along the border of Brazil and French Guiana, including environmental conditions, forest structure and tree inventory. We explored the utility of extracting DNA from cambium tissue and the botanical identification potential of two widely used plant DNA barcodes, namely *rbcLa* and *matK*.

## MATERIAL AND METHODS

The 2015 expedition involved 20 people (including 15 from the 3<sup>rd</sup> Infantry Regiment of the Foreign Legion, part of the Forces Armées de Guyane) and lasted six weeks, with a weekly re-supply of food (Kew youtube 2016). Milestones were separated by 23–68 km, and given the rugged terrain, the team moved about 10 km per day. The route of the expedition was pre-planned but had to be adapted daily depending on local terrain conditions.

A hand-held GPS unit (Garmin 62) logged the location of the route. The elevation data provided a good opportunity to test the altimetry data from the Shuttle Radar Topography Mission (SRTM) at 30 m resolution in a little-explored area, and on rugged terrain. Using a Tinytag data logger (temperature and humidity; Gemini data loggers, Scientific House, Terminus Rd, Chichester, UK), we monitored environmental conditions throughout the route.

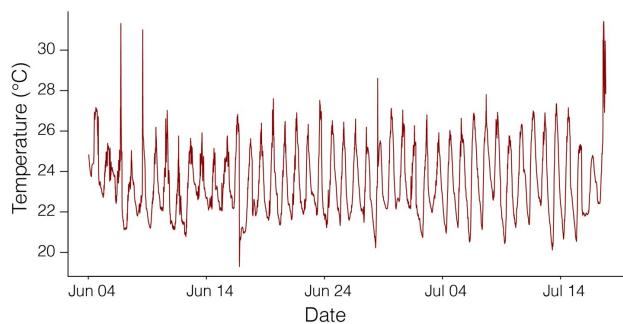
The route of the expedition is plotted in Figure 1 (a-b). Elevation measurements recorded with the GPS unit show that the terrain of the Tumucumaque range is rugged, with elevation varying between 200 m to 600 m a.s.l. (Figure 1c). Visual impression of the forest along the route are shown with panoramic photographs (Supplementary Material, Figure S1). Comparing ground data and SRTM data showed that the match in elevation was generally quite good, within 7.9 m (standard deviation), but with a systematic bias: SRTM tends to overestimate the elevation by about 10.6 m (Figure 1d). The Tinytag data logger showed that mean temperature did not display a trend along the transect, but varied principally due to daily variations, from 21°C to 27°C, with a few peaks above 30°C when the team reached tabletop inselbergs (Figure 2). Air humidity was consistently high, reaching 100% at night with a minimum around 80% at mid-day (Figure 3).

Seven randomised 25 × 25-m (0.0625 ha) plots were established around the seven country-boundary milestones. In each plot, each free-standing stem ≥ 10 cm DBH was sampled for cambium using a cleaned knife, as a rapid alternative to collecting herbarium specimens from trees for identification.

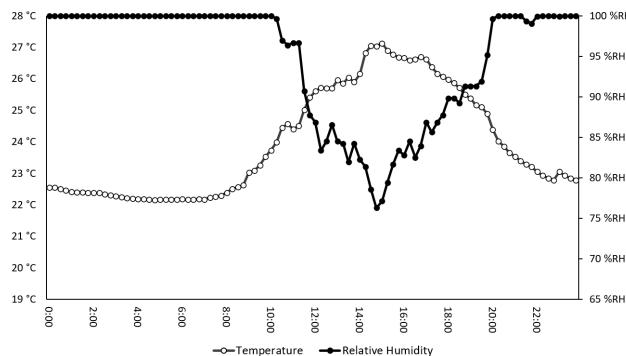
Each sampled tree or liana was measured for DBH, and the tree height was recorded on a visual estimation.

We also sampled fertile material (herbarium specimens) from plants inside and outside the plots, during the march, and from some trees that were felled for a helicopter landing. The herbarium vouchers were deposited at Kew (K) and Cayenne (CAY). The herbarium specimens were directly identified by taxonomists based on morphological features. Field images associated with 167 herbarium specimens were also placed on the iNaturalist website ([www.inaturalist.org/observations/willmilliken](http://www.inaturalist.org/observations/willmilliken)) to determine whether the specimens could be identified by other experts without knowledge of the corresponding herbarium vouchers. As we did not collect fertile specimens from the trees in the plots, we could not use herbarium identifications to support DNA identification.

The DNA samples from the cambium of trees and lianas in the plots were used for preliminary identification. These samples did not include the cork, but they did include the cork cambium, the phloem and the vascular cambium together, as recommended by Tibbits *et al.* (2006). Samples were individually wrapped in tea-filter paper, numbered, and immediately stored in an airtight plastic container with dried silica gel, following a procedure previously described in Gonzalez *et al.* (2009). A bark slash on each sampled tree,



**Figure 2.** Air temperature along the transect measured by a Tinytag unit, with daily fluctuations.



**Figure 3.** Example of daily fluctuation of temperature and humidity during the transect route, measured by a Tinytag unit. The data are from 29 Jun 2015, at an altitude of 387 m a.s.l.

with the tree number, was also photographed. After the end of the trip, samples were shipped for DNA extraction and sequencing.

For DNA analysis, up to 30 mg of dry tissue of each cambium sample was ground for two minutes in a TissueLyser mixer-mill disruptor (Qiagen, California, USA) using tungsten beads. Lysis incubation was carried out at 65 °C for two hours, using a CTAB 1% PVP buffer. Total DNA extraction was performed with a Biosprint 15 workstation (Qiagen, CA) following the manufacturer's protocols. PCR amplification was performed for the two plastid DNA barcoding regions selected. The *rbcL* marker is the first half of the *rbcL* gene and was amplified using classic primers: 1F and 724R (Gonzalez *et al.* 2009). The *matK* region was amplified using two combinations of primers: 390F and 1326R (Cuénoud *et al.* 2002); 3F\_Kim and 1R\_Kim (Dunning and Savolainen 2010; Lima *et al.* 2018). The PCR reaction mix included 0.2 µl of GoTaq 51 U/µl (Promega), 10 µl of 5 x buffer, 1 µl of 20 µM for each primer, 1 µl of dNTP 10 µM, 1 ml of DNA template and H<sub>2</sub>O for a final volume of 50 µl. PCR products were purified with a MinElute PCR Purification Kit (Qiagen, CA).

Cycle sequencing reactions were performed in 10 µl reactions using 1 µl of BigDye Terminator cycle sequencing chemistry (v3.1; ABI; Warrington, Cheshire, UK) and run on an ABI sequencer. The two genetic regions were sequenced in both forward and reverse directions. DNA fragments were visually inspected and assembled with Geneious v.8 and curated manually if necessary. The DNA sequences were then matched with BLAST against the NCBI reference nucleotide collection using Megablast, a plugin available in Geneious. Default options of Megablast were used, and for each sequence, the top hit was visually inspected in the resulting lookup table. The 193 *rbcL* sequences were 328–681 nucleotides (nt) in length (three were less than 500 nt). Two sequences had a pairwise sequence similarity < 97%, and they were removed from subsequent analyses. The 227 *matK* sequences were 123–804 nucleotides in length (nine were less than 500 nt). Two

sequences had a pairwise sequence similarity < 97%, and they were also removed from subsequent analyses. All sequences were submitted to NCBI, and GenBank accession numbers are available in the Supplementary Material, Tables S1, S2.

## RESULTS

Overall, 279 trees were sampled in a total area of 0.4375 ha over the seven plots (Table 1), resulting in an estimated density of 642 trees with DBH ≥ 10 cm per hectare. In addition, 15 lianas were sampled from the plots. Average tree height was 16 m, and average basal area was 38.2 (24 - 58.5) m<sup>2</sup> ha<sup>-1</sup>, with a tendency to smaller basal area in plots at higher elevations (387–556 m a.s.l.) than the plots at lower elevations (285–366 m a.s.l.). A brief description of the plots is provided in the Supplementary Material (Figure S3).

We extracted DNA sequences for 235 individuals (84.2% of the cambium samples). Mean DNA concentration was 12 ng µL<sup>-1</sup>, range = 3 - 25 ng µL<sup>-1</sup>. Of the 235 samples, 197 (83.8%) were amplified for *rbcL*, and 220 (93.6%) for *matK*, with 170 samples (72.3%) amplified for both markers. Matching to the NCBI reference database revealed that most of the specimens could be confidently identified to family.

Comparing the identifications based on either *rbcL* and *matK* revealed a corresponding match per sample in 98.8% at the family level, 59.1% at the generic level and 10.7% at the species level. Tree taxa identified at the species level were *Balizia pedicellaris* (DC.) Barneby & J.W.Grimes, *Diospyros tetrandra* Hiern, *Leonia glycyarpa* Ruiz & Pav., *Ormosia arborea* (Vell.) Harms, *Pseudopiptadenia suaveolens* (Miq.) J.W.Grimes, *Rhabdodendron amazonicum* (Spruce ex Benth.) Huber, *Siparuna decipiens* (Tul.) A.DC, *Theobroma cacao* L. and *Trymatococcus oligandrus* (Benoist) Lanj., and one species of liana (*Hippocratea volubilis* L.). Interestingly, one sample was identified by both barcodes as *Pouteria campechiana* (Kuhn) Baehni, which is not native to the Amazon region (Awang-Kanak and Bakar 2018). Due to the low taxonomic resolution of the DNA barcoding, the overall botanical results are reported at family resolution.

The most abundant family across the tree samples was Burseraceae (21%), all of them attributed to genus *Protium* (Daly and Fine 2018), followed by Fabaceae (11%), Meliaceae (8%), Moraceae (8%), Myristicaceae (6%), Sapotaceae (6%) and Vochysiaceae (5%) (Table 2). Together, these families contributed over 50% of all tree individuals in the seven plots. We did not include a survey of lianas, due to the small sample size (Supplementary Material, Tables S1 and S2).

Thirty-five species (31.8% of cambium samples sequenced with one or both markers) were given tentative identifications (Table 3), based on: 1) DNA barcodes (removing alternative identification for species that are not present in French Guiana or neighbouring countries); 2) conformation from the bark slash (Figure 4) by one of the authors (J. Engel); 3) the species

**Table 1.** Basal area, maximal DBH, number of trees ≥ 10 cm DBH, and average tree height in each of seven plots (25 m × 25 m) sampled along a 320-km transect on the Brazil-French Guiana border. GPS coordinates of the plots are in WGS 84.

Plot	Basal area (m <sup>2</sup> ha <sup>-1</sup> )	DBH max (cm)	Tree count	Average tree height (m)	Longitude	Latitude
1	29.95	65.5	47	15	-54.436817	2.209524
2	27.47	44.5	41	17	-54.190377	2.176846
3	52.53	38.3	50	18	-53.973027	2.207217
4	23.96	68.0	34	15	-53.774145	2.368937
5	37.13	110.0	33	15	-53.550010	2.251592
6	37.54	105.0	42	14	-53.359604	2.344864
7	58.58	120.0	36	19	-53.281768	2.187473
Mean	38.2		40.1	16.1		

**Table 2.** List of tree families identified in seven 25x25-m plots along the Brazil-French Guiana border. The numbers refer to individual tree counts; INDET refers to trees that could not be identified through DNA barcoding.

Family	Plot							Total
	1	2	3	4	5	6	7	
Burseraceae	16	8	5	2	7	8	10	56
Fabaceae	2	2	8	6	4	5	2	29
Meliaceae	3	2	7	3	3	2	1	21
Moraceae	4	2	5	2	3	5		21
Myristicaceae			3	6	3		4	16
Sapotaceae	10	1			1	4		16
Vochysiaceae		2	3			7		12
Malvaceae	4	1	1		2	2		10
Apocynaceae	1	1	2	1	2	1		8
Olacaceae	1	1	2			4		8
Arecaceae	1	3				3		7
Anacardiaceae	2		3			1		6
Urticaceae			2			2	2	6
Chrysobalanaceae	3			1	1			5
Lauraceae	1	1	2	1				5
Lecythidaceae	1			2	1			4
Ebenaceae					3			3
Euphorbiaceae	2			1				3
Nyctaginaceae	1	1			1			3
Salicaceae			1		1		1	3
Siparunaceae				3				3
Annonaceae	1	1						2
Rhabdodendraceae			2					2
Rubiaceae	2							2
Sapindaceae				2				2
Violaceae	1				1			2
Clusiaceae					1			1
Erythroxylaceae		1						1
Humiriaceae						1	1	
Lacistemataceae			1					1
Myrtaceae	1							1
Ochnaceae	1							1
Putranjivaceae					1			1
Simaroubaceae	1							1
Ulmaceae					1			1
INDET	3	3	4	2	1	6		19

was also collected as a herbarium specimen on the expedition, though not from within the plots. Burseraceae and Meliaceae, however, could only be confidently assigned to family level.

Of the 289 herbarium vouchers collected during the expedition, 160 were identified by botanists from CAY and K to species (55%) and 27 to genus only (9%). Of the 167 images of vouchered specimens placed on iNaturalist in 2015, 57 (34%) were identified to species in 2021 and 58 (35%) to genus only (Supplementary Material, Figure S2). Among the determined species, 48 were marked as 'Research Grade', meaning that two experts or knowledgeable people have reviewed the observation and agreed.



**Figure 4.** Examples of bark slash sampled from trees along the Brazil-French Guiana border for DNA barcoding analysis from cambium tissue. A – *Protium* sp.; B – Arecaceae; C – *Helicostylis pedunculata* Benoit; D – *Swartzia* sp. (possibly *Swartzia* cf. *canescens* Torke, based on the bark slash); E – Sapotaceae; F – Sapotaceae; G – Vochysiaceae; H – *Conopeiba guianensis* Aubl.; I – *Brosimum alicastrum* S

## DISCUSSION

The tree composition (at family level) in our plots was comparable with that of the Nouragues Station in Central French Guiana (Poncy *et al.* 2001), but differs from that of Northern French Guiana, with an under-representation of Chrysobalanaceae and Lecythidaceae. This corresponds to other observations on the decrease in Lecythidaceae species-richness, and the increase in Burseraceae, from north to south in French Guiana (Guitet *et al.* 2015). Comparing data with other plot surveys in the Amazon, the average basal area was high (Phillips *et al.* 2004). In other parts of French Guiana, basal areas of 30–35 m<sup>2</sup> ha<sup>-1</sup> are not uncommon, while lower basal areas (25–30 m<sup>2</sup> ha<sup>-1</sup>) are sometimes found in ancient anthropogenic areas (Odonne *et al.* 2019). A discussion about the herbarium specimens collected on the expedition (available on request from the authors), and ecology relating to these specimens, are described in Le Tourneau *et al.* (2016) and in the Supplementary Material (Figure S1, Figure S3).

Collection of cambium samples for DNA barcoding is quicker, particularly for larger trees, than leaf tissue collection. It also means that leafless trees can be surveyed, e.g., in the dry season. Compared to wood, it was found that the cambium had a higher concentration of DNA than the heartwood or sapwood, although it also had larger amounts of PCR reaction inhibitors (Tang *et al.* 2011). The cambium samples that we collected were rapidly dried with silica gel, corresponding to the best long-term approach for DNA preservation (Mangaravite *et al.* 2020), and we indeed found that DNA

**Table 3.** Species identifications (likely determination) of species sampled in the Tumucumaque range (Brazil-French Guiana border) using *rbcL*a and *matK* DNA barcodes. 'Collected' refers to numbered W. Milliken specimens stored at Kew (K) and Cayenne (CAY); Slash ID is J. Engel's preliminary identification based on images of the bark slash.

Likely determination	Family	<i>rbcL</i> a	<i>matK</i>	Slash ID	Collected
<i>Balizia pedicellaris</i> (DC.) Barneby & J.W.Grimes	Fabaceae	<i>Balizia pedicellaris</i>	<i>Balizia pedicellaris</i>		
<i>Brosimum alicastrum</i> Sw.	Moraceae	<i>Brosimum rubescens</i>	<i>Brosimum alicastrum</i>	<i>Brosimum</i>	
<i>Brosimum guianense</i> (Aubl.) Huber ex Ducke	Moraceae	<i>Brosimum rubescens</i>	<i>Brosimum guianense</i>	<i>Brosimum</i>	
<i>Brosimum lactescens</i> (S.Moore) C.C.Berg	Moraceae	<i>Brosimum alicastrum</i>	<i>Brosimum lactescens</i>	<i>Brosimum</i>	
<i>Carapa guianensis</i> Aubl.	Meliaceae	<i>Swietenia mahagoni</i>	<i>Carapa guianensis</i>	<i>C. guianensis</i>	
<i>Casearia javitensis</i> Kunth.	Salicaceae	<i>Casearia javitensis</i>		<i>C. javitensis</i>	
<i>Chaunochiton kappleri</i> (Sagot ex Engl.) Ducke	Olaceae		<i>Chaunochiton kappleri</i>	<i>C. kappleri</i>	
<i>Conceveiba guianensis</i> Aubl.	Euphorbiaceae	<i>Conceveiba terminalis</i>	<i>Conceveiba martiana</i>	<i>C. guianensis</i>	
<i>Cupania scrobiculata</i> Rich.	Sapindaceae	<i>Cupania scrobiculata</i>	<i>Synima cordieri</i> *	<i>Cupania</i>	
<i>Diplotropis purpurea</i> (Rich.) Amshoff	Fabaceae	<i>Diplotropis purpurea</i>		<i>D. pupurea</i>	
<i>Erythroxylum macrophyllum</i> Cav.	Erythroxylaceae	<i>Erythroxylum novogranatense</i>	<i>Erythroxylum macrophyllum</i>		5263
<i>Geissospermum argenteum</i> Woodson	Apocynaceae	<i>Vallesia antillana</i> *	<i>Geissospermum laeve</i>	<i>G. argenteum</i>	5261
<i>Guarea sylvatica</i> C.DC.	Meliaceae	<i>Guarea sylvatica</i>	<i>Guarea pterorhachis</i>	<i>G. sylvatica</i>	
<i>Gustavia hexapetala</i> (Aubl.) Sm.	Lecythidaceae	<i>Gustavia hexapetala</i>	<i>Grias cauliflora</i> *	<i>G. hexapetala</i>	
<i>Helicostylis pedunculata</i> Benoist	Moraceae	<i>Helicostylis pedunculata</i>	<i>Castilla elastica</i>	<i>Moraceae</i>	
<i>Hippocratea volubilis</i> L. **	Celastraceae	Celastraceae sp.	<i>Hippocratea volubilis</i>	<i>Celastraceae</i>	
<i>Hymenaea courbaril</i> L.	Fabaceae		<i>Hymenaea courbaril</i>		
<i>Iryanthera sagotiana</i> (Benth.) Warb.	Myristicaceae	<i>Iryanthera sagotiana</i>	<i>Haematoxylon glabrum</i> *	<i>I. sagotiana</i>	
<i>Lacistema aggregatum</i> (P.J.Bergius) Rusby	Lacistemataceae	<i>Lacistema robustum</i> *	<i>Lacistema aggregatum</i>		
<i>Leonia glycycarpa</i> Ruiz & Pav	Violaceae	<i>Leonia glycycarpa</i>	<i>Leonia glycycarpa</i>	<i>L. glycycarpa</i>	
<i>Macoubea guianensis</i> Aubl.	Apocynaceae		<i>Macoubea guianensis</i>	<i>M. guianensis</i>	
<i>Maquira calophylla</i> (Poep. & Endl.) C.C.Berg	Moraceae	<i>Maquira calophylla</i>	<i>Castilla elastica</i> *	<i>Moraceae</i>	
<i>Minquartia guianensis</i> Aubl.	Olaceae		<i>Minquartia guianensis</i>	<i>Minquartia</i>	
<i>Naucleopsis guianensis</i> (Mildbr.) C.C.Berg	Moraceae	<i>Naucleopsis guianensis</i>	<i>Castilla elastica</i> *	<i>Moraceae</i>	
<i>Pouteria campechiana</i> (Kunth) Baehni *	Sapotaceae	<i>Pouteria campechiana</i> *	<i>Pouteria campechiana</i> *	<i>Pouteria</i>	
<i>Protium excelsior</i> Byng & Christenh.	Burseraceae	<i>Protium excelsior</i>		<i>Burseraceae</i>	
<i>Pseudopiptadenia suaveolens</i> (Miq.) J.W.Grimes	Fabaceae	<i>Pseudopiptadenia suaveolens</i>	<i>Pseudopiptadenia suaveolens</i>		
<i>Ptychopetalum olacoides</i> Benth.	Olaceae	<i>Ptychopetalum olacoides</i>	<i>Ptychopetalum petiolatum</i> *	<i>P. olacoides</i>	
<i>Rhabdodendron amazonicum</i> (Spruce ex Benth.) Huber	Rhabdodendraceae	<i>Rhabdodendron amazonicum</i>	<i>Rhabdodendron amazonicum</i>		5291
<i>Simarouba amara</i> Aubl.	Simaroubaceae	<i>Simarouba amara</i>		<i>S. amara</i>	
<i>Siparuna decipiens</i> (Tul.) A.DC.	Sipuarunaceae	<i>Siparuna decipiens</i>	<i>Siparuna decipiens</i>	<i>S. decipiens</i>	
<i>Sterculia pruriens</i> (Aubl.) K.Schum.	Malvaceae	<i>Sterculia pruriens</i>	<i>Sterculia apetala</i>	<i>S. pruriens</i>	5252
<i>Tapirira obtusa</i> (Benth.) J.D.Mitch.	Anacardiaceae	<i>Tapirira obtusa</i>	<i>Tapirira guianensis</i>	<i>Tapirira</i>	5234
<i>Touroulia guianensis</i> Aubl.	Ochnaceae	<i>Touroulia guianensis</i>			5314
<i>Trymatococcus oligandrus</i> (Benoist) Lanj.	Moraceae	<i>Trymatococcus oligandrus</i>	<i>Trymatococcus oligandrus</i>	<i>Trymatococcus</i>	5297
<i>Virola michelii</i> Heckel	Myristicaceae	<i>Myristica fragrans</i> *	<i>Virola michelii</i>	<i>V. michelii</i> (or <i>V. kwatae</i> )	

\* Not present in French Guiana or neighbouring countries

\*\* Liana

quality was good, despite the fast collection approach adopted in this work.

Our low success in species identification using DNA barcodes from cambium samples nevertheless confirm that the selected markers do not fully resolve plants down to species level in all plant families (Gonzalez *et al.* 2009). Our inability to identify Burseraceae and Meliaceae beyond family level was probably since both families include clades that radiated recently (Fine *et al.* 2014; Koenen *et al.* 2015), and therefore the DNA barcodes selected in this study are unable to discriminate the species in these clades sufficiently.

One illustration of this problem is the identification of one of our specimens as *Pouteria campechiana*. In NCBI, many species in the genus *Pouteria* have the same *matK* and *rbcL* sequences, perhaps due to a relatively recent radiation of this clade within Sapotaceae, subfamily Chrysophylloideae (De Faria *et al.* 2017). Our BLAST search against NCBI selected one of the possible species and happened to select one that does not occur in our study area, highlighting one of the problems of relying too much on DNA barcodes for species identification. To avoid this type of geographical bias, we could have downloaded the full NCBI database, select only the species known to occur in the study region, and then run the BLAST search on the regional subset. However, several species of *Pouteria* cooccur in this region, and this procedure would merely reduce geographical inconsistencies, and not resolve the issue with species identification for Sapotaceae.

Adding more DNA barcodes to the ones selected here, such as the Internal Transcribed Spacers region of nuclear ribosomal DNA (ITS) or the plastid *trnH-psbA* intergenic spacer might, in some families, increase the rate of correct identification (Gonzalez *et al.* 2009; Hollingsworth *et al.* 2009; Costion *et al.* 2011; Bolson *et al.* 2015). In an analysis of plant fragments from Brazilian caves (mainly roots), the ITS2 spacer was believed to be the best marker for identification (Ramalho *et al.* 2018). More recent studies have shown that ITS2 is likely to become recognised as the standard DNA barcode for plants (Moorhouse-Gann *et al.* 2018; Miao *et al.* 2019). However, the ITS region presents specific challenges for plants: the ribosomal cluster which carries the ITS region is present in multiple copies in the plant cell, and many of these copies are non-functional, but appear to be retained in the cell (Feliner and Rosselló 2007; Group *et al.* 2011). Non-functional ITS copies appear to have a lower GC content and are preferentially selected during PCR and sequencing, creating potential biases (Besnard *et al.* 2009).

Identification of trees in forest plots, using herbarium specimens, continues to be problematic. In an analysis of 60 plots in Western Amazonia, over the last 30 years, 25% of specimens were misidentified, and in some difficult genera 50% were incorrect (Baker *et al.* 2017). One of the issues with species identification within our plots (through DNA

barcoding) is that there are large numbers of plant species that have not yet been placed in the NCBI reference collection. Only 31% of known plants have sequences in Genbank, and these were fewer near the Equator (Cornwell *et al.* 2019), where our survey was carried out. Of the species-rich flora of São Paulo (southeastern Brazil), 58% of tree species have at least one barcoding sequence available, including 35.5% with ITS data (Lima *et al.* 2018). Based on current accumulation rates, it is possible that 100% species coverage will be achieved within the next 20 years for the São Paulo tree flora, but nevertheless this may not be enough for complete identification in specific taxonomic groups of communities with closely related taxa (Lima *et al.* 2018). Southern Brazil has been more densely explored and studied than Amazonia, so a high coverage of DNA sequences for the flora of our study area is far from being reached.

Accurate iNaturalist identification of plant images in a poorly known Amazonian region requires trained researchers. Good-quality images, as shown here, can improve the knowledge of species distribution without collecting herbarium specimens. However, given that our images were available to researchers over five years, and less than half are now identified to species, this is not a rapid way to assess biodiversity. New computer-based image identification resources (AI/machine learning) will probably improve and accelerate biodiversity knowledge (Wäldchen and Mäder 2018), but this will require more ‘training’ of images from poorly known taxa (Van Horn *et al.* 2018).

The under-sampling of inter-fluvial areas of Amazonia remains a major hurdle to biodiversity discovery, and future research should prioritize these less accessible areas in a more systematic way to improve conservation planning. In terms of sampling plant diversity, technological development in communication and automated monitoring could bring down the costs of sampling in the future (Mulatu *et al.* 2017; Draper *et al.* 2020).

## CONCLUSIONS

Our study demonstrates that lightweight expeditions can benefit from the advances in novel biodiversity monitoring, yet the impossibility to collect herbarium specimens from trees in such conditions is an impediment to species discovery. Identification of trees using DNA from cambium samples and two DNA barcodes (*rbcL* and *matK*) yielded low success at the species level. Our identifications at the family level are insufficient for comparable surveys across Amazonia. Using DNA barcoding to aid species identification will require further development, not only of sampling methods but also the necessary knowledge to support it (a baseline of accurate and reproducible DNA barcodes). We hope that this research, and the discovery of new techniques, will stimulate increased research in eastern Amazonia.

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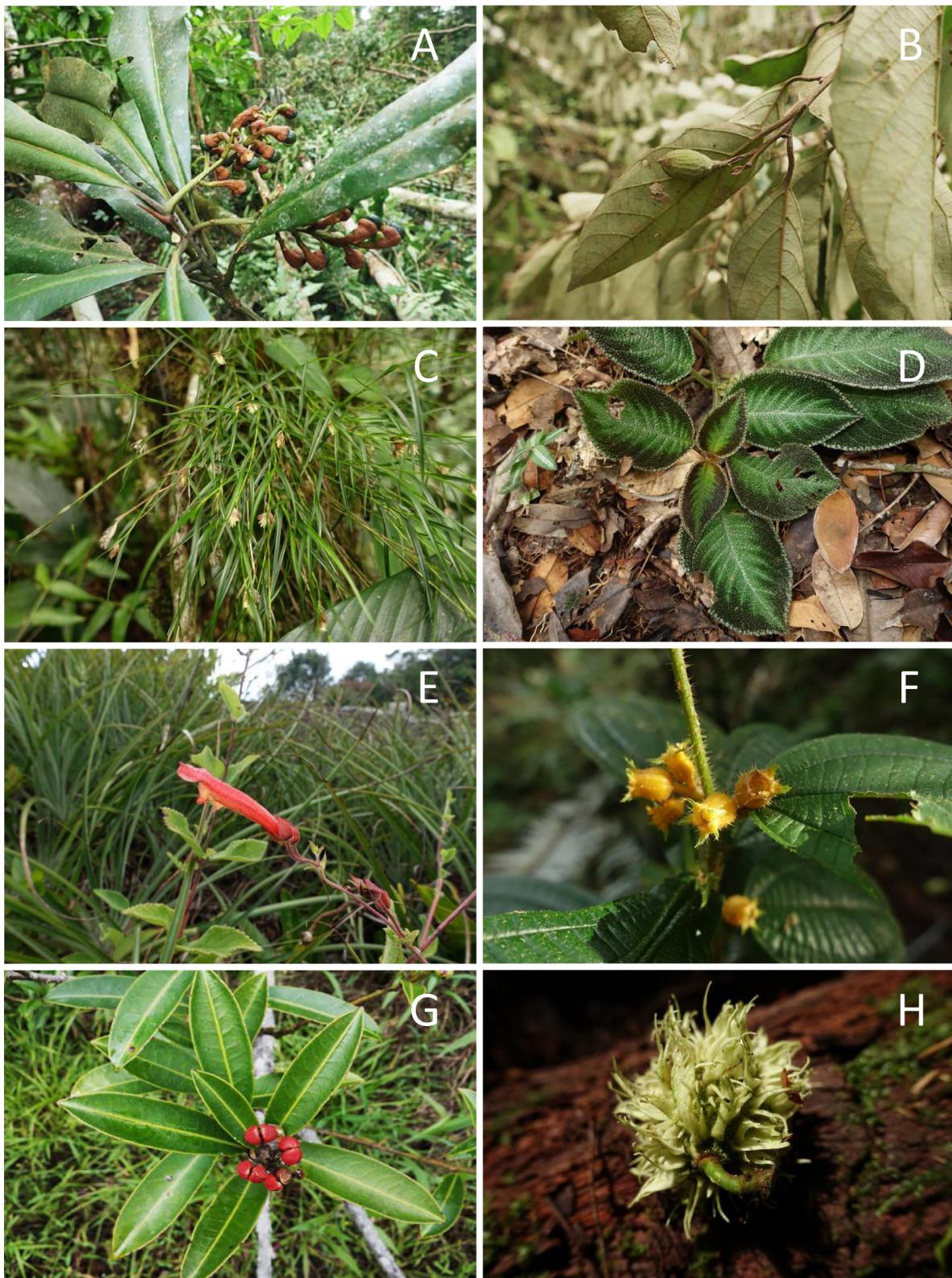
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**SUPPLEMENTARY MATERIAL** (only available in the electronic version)

Milliken *et al.* Fast and novel botanical exploration of a 320-km transect in eastern Amazonia using DNA barcoding



**Figure S1.** Examples of panoramas of the vegetation along the 320-km transect along the border between southern French Guiana and Brazil. **A** – 2.324275N, 54.5391W, creek between hills, with *Euterpe oleracea* Mart., *Socratea exorrhiza* (Mart.) H.Wendl. (Arecaceae), *Sympomia globulifera* Lf. (Clusiaceae) and *Rapatea paludosa* Aubl. (Rapateaceae); **B** – 2.279289N, 54.5253W, granite inselberg, with *Mandevilla surinamensis* (Pulle) Woodson (Apocynaceae), *Oreopanax capitatus* (Jacq.) Decne. & Planch. (Araliaceae), *Topoeba parasitica* Aubl. (Melastomataceae), *Clusia palmicida* Rich. (Clusiaceae) and *Sapium argutum* (Müll. Arg.) Huber (Euphorbiaceae); **C** – 2.256044N, 54.4826W, forest on ridge, with *Astrocaryum sciophyllum* (Miq.) Pulle (Arecaceae); **D** – 2.207178N, 54.438W, granite inselberg. In damp areas on the rock the species included *Sipanea wilson-brownii* R.S. Cowan (Rubiaceae), *Paepalanthus oyapockensis* Herzog (Eriocaulaceae), *Rhynchospora subdicephala* T. Koyama (Cyperaceae), *Utricularia hispida* spp. (Lentibulariaceae), *Sinningia incarnata* (Aubl.) D.L.Denham (Gesneriaceae), among others; **E** – 2.168647N, 54.3391W, forest on ridge, with *Astrocaryum sciophyllum* (Miq.) Pulle (Arecaceae); **F** – 2.166903N, 54.2006W, forest on ridge, with the massive *Huberodendron swieteniooides* (Gleason) Ducke (Malvaceae); **G** – 2.207217N, 53.973027W, forest on ridge, dominated by Fabaceae and Meliaceae. Fluted trunks on the left are *Minquartia guianensis* Aubl. (Olaceaceae); **H** – 2.300701N, 53.885363W, forest on ridge with young stems of *Oenocarpus* sp. (Arecaceae); **I** – 2.347966N, 53.804337W, swamp forest dominated by *Euterpe oleracea* Mart. (Arecaceae); **J** – 2.369397N, 53.772392W, hilltop forest on inselberg, close to Milestone 4, with *Ananas comosus* (L.) Merr. (Bromeliaceae), and *Syagrus inajai* (Spruce) Becc. (Arecaceae) which are possible clues of past human occupations.



**Figure S2.** Examples of specimen identification by other botanists, five years after the images became available on iNaturalist (<https://www.inaturalist.org/home>). A – *Rhabdodendron amazonicum* (Spruce ex Benth.) Huber; B – *Styrax pallidus* A.DC.; C – *Elleanthus graminifolius* (Barb.Rodr.) Løjtnant; D – *Nautilocalyx pictus* (Hook.) Sprague; E – *Sinningia incarnata* (Aubl.) D.L.Denham; F – *Maieta poeppigii* Mart. ex Cogn.; G – *Sapium argutum* (Müll.Arg.) Huber; H – *Carpotroche longifolia* (Poepp.) Benth.



**Figure S3.** Brief description of the seven 25x25-m plots sampled for tree cambium along a 320-km transect along the Brazil-French Guiana border (further details in Table 1). **1** – Forest approximately 100 m from an open granite inselberg, on shallow soil, with few large trees and a low canopy (approx. 15 m). The most abundant tree families were Burseraceae (*Protium* spp.) and Sapotaceae; **2** – Trees larger than in Plot 1 (DBH and height), and more diverse in families and species and with deeper soil. Several trees were felled next to the clearing (for a helicopter landing), where collections were made. These included: *Aspidosperma excelsum* Benth. (Apocynaceae), *Oenocarpus bacaba* Mart. (Arecaceae), *Protium morii* Daly, *Protium robustum* (Swart) D.M. Porter, *Protium spruceanum* (Benth.) Engl. (Burseraceae), *Caryocar microcarpum* Ducke (Caryocaraceae), *Dicorynia guianensis* Amshoff, *Ormosia amazonica* Ducke, *Swartzia panacoco* (Aubl.) R.S.Cowan (Fabaceae), *Goupiac glabra* Aubl. (Goupiaceae), *Licaria debilis* (Mez) Kosterm (Lauraceae), *Eschweilera coriacea* (DC.) S.A.Mori (Lecythidaceae), *Trichilia micrantha* Benth. (Meliaceae), *Trymatococcus oligandrus* (Moraceae), *Iryanthera* sp. (Myristicaceae), *Touroulia guianensis* Aubl. (Ochnaceae), *Rhabdodendron amazonicum* (Spruce ex Benth.) Huber (Rhabdodendraceae), *Talisia carinata* Radlk., *Toulacia* sp. (Sapindaceae), *Manilkara huberi* (Ducke) Standl. (Sapotaceae), *Styrax cf. macrophyllus* Schott ex Pohl (Styracaceae), *Coussapoa angustifolia* Aubl. and *Pououma minor* Benoit (Urticaceae). It is likely that *Protium* sp. in the DNA identifications may have been one of the three species collected, that all the Arecaceae (DNA) were *Oenocarpus bacaba*, and that *Swartzia* sp. (DNA) was *S. panacoco* (identified correctly by matK); **3** – This was the plot highest in tree density, and second for basal area, with  $52.53 \text{ m}^2 \text{ ha}^{-1}$ , mostly small trees (DBH max = 38.3 cm) of Fabaceae and Meliaceae. Located on top of a little plateau, partly on a slope, and likely an old secondary forest, with few species represented by several individuals, but *Protium* sp. (Burseraceae), (identified as *P. decandrum* (Aubl.) Marchand by rbcLa determination) appeared three times and *Rhabdodendron amazonicum* (Spruce ex Benth.) Huber (Rhabdodendraceae) twice; **4** – Located at the top of an inselberg, but on a draining substrate (not directly on the rocky outcrop), with an open understory and a low tree density ( $544 \text{ trees ha}^{-1}$ ). The largest tree was measured on this plot. Dominated by Fabaceae and Myristicaceae, with three individuals of *Carapa guianensis* Aubl. (Meliaceae); **5** – Located on a ridge. Plot with the fewest trees (density of  $528 \text{ tree ha}^{-1}$ ). Dominated by Burseraceae, with both *Protium* spp. and *Protium excelsior* Byng & Christenh. (rbcLa); **6** – Plot with the lowest canopy, with open understory and many small diameter stems. Dominated by Burseraceae and Vochysiaceae, in which *Erisma uncinatum* Warm. (rbcLa) is probably the most abundant species; **7** – Low-density plot ( $576 \text{ tree ha}^{-1}$ ) on a well-drained flatland close to the seventh milestone. Dominated by Burseraceae, with both *Protium* spp. and *Protium excelsior* (rbcLa), as in Plot 5.

**Table S1.** Blast results for *rbcLa* DNA barcodes against the NCBI online database for samples from seven 25-m<sup>2</sup> plots along a 320-km transect on the Brazil-French Guiana border. 'Query' refers to the tree label as given in the field. The two last columns report the best hit sequence (organism name and accession) to the DNA barcode.

Query	Query coverage	Sequence length	% Identical sites	% Pairwise identity	%GC	Bit-score	Grade (%)	Organism	Accession number
1.1	100.00	681	100.0	100.0	43.0	1258.69	100.0	<i>Trichilia</i> sp.	KC628372
1.13	99.41	677	100.0	100.0	41.7	1251.3	99.7	<i>Trymatococcus oligandrus</i>	FJ038126
1.15	99.71	679	100.0	100.0	43.7	1255	99.9	<i>Pouteria decorticans</i>	FJ038177
1.16	99.71	679	100.0	100.0	41.8	1255	99.9	<i>Helicostylis pedunculata</i>	FJ038121
1.18	100.00	681	99.7	99.7	43.3	1247.61	99.9	<i>Chrysophyllum oliviforme</i>	L12607
1.2	99.71	679	100.0	100.0	43.6	1255	99.9	<i>Chrysophyllum sanguinolentum</i>	FJ038163
1.21	99.27	676	100.0	100.0	42.8	1249.46	99.6	<i>Swartzia</i> sp.	FJ038056
1.23	99.85	680	99.9	99.9	44.3	1251.3	99.9	<i>Ocotea venulosa</i>	KF981236
1.27	99.71	679	99.9	99.9	43.6	1249.46	99.8	<i>Chrysophyllum sanguinolentum</i>	FJ038163
1.3	99.71	679	100.0	100.0	43.9	1255	99.9	<i>Sterculia pruriens</i>	FJ038104
1.32	99.71	679	99.6	99.6	41.8	1238.38	99.6	<i>Helicostylis pedunculata</i>	FJ038121
1.33	100.00	681	99.7	99.7	43.5	1247.61	99.9	<i>Manilkara zapota</i>	EU980807
1.34	99.27	676	100.0	100.0	42.8	1249.46	99.6	<i>Swartzia</i> sp.	FJ038056
1.36	99.71	679	100.0	100.0	43.6	1255	99.9	<i>Chrysophyllum prieurii</i>	GQ428633
1.4	99.71	679	100.0	100.0	43.6	1255	99.9	<i>Chrysophyllum sanguinolentum</i>	FJ038163
1.41	100.00	681	99.9	99.9	42.7	1253.15	99.9	<i>T.guianensis chloroplast</i>	Z75690
1.43	91.48	623	97.8	98.8	45.1	1099.88	95.1	<i>Fusaea longifolia</i>	GQ428542
1.48	100.00	681	99.9	99.9	43.8	1253.15	99.9	<i>Theobroma cacao</i>	JQ228389
1.7	99.85	680	99.9	99.9	43.1	1251.3	99.9	<i>Abarema brachystachya</i>	KF981222
2.1	100.00	681	100.0	100.0	42.9	1258.69	100.0	<i>Hirtella suffulta</i>	KX180070
2.11	100.00	681	99.6	99.6	42.9	1242.07	99.8	<i>Trichilia emetica</i>	AY128244
2.12	100.00	681	99.9	99.9	43.2	1253.15	99.9	<i>Hexopetion mexicanum</i>	JX903251
2.13	99.71	679	100.0	100.0	43.6	1255	99.9	<i>Chrysophyllum prieurii</i>	GQ428633
2.17	100.00	681	99.9	99.9	43.4	1255	100.0	<i>Ferdinandusa speciosa</i>	AM117226
2.18	100.00	681	99.6	99.6	42.9	1242.07	99.8	<i>Trichilia emetica</i>	AY128244
2.19	99.71	679	99.7	99.7	42.7	1243.92	99.7	<i>Erisma uncinatum</i>	FJ038209
2.2	92.22	628	98.9	99.4	45.2	1134.96	95.8	<i>Fusaea longifolia</i>	GQ428542
2.2	99.71	679	100.0	100.0	43.9	1255	99.9	<i>Protium decandrum</i>	FJ037977
2.21	95.89	653	100.0	100.0	43.0	1206.98	97.9	<i>Hirtella suffulta</i>	KX180070
2.23	99.71	679	99.9	99.9	42.9	1249.46	99.8	<i>Pachycormus discolor</i>	GU935437
2.24	97.65	665	100.0	100.0	42.7	1229.14	98.8	<i>Swartzia</i> sp.	FJ038056
2.28	95.89	653	99.7	99.8	42.6	1199.6	97.8	<i>Brosimum rubescens</i>	GQ428590
2.3	99.71	679	100.0	100.0	43.9	1255	99.9	<i>Protium decandrum</i>	FJ037977
2.3	100.00	681	99.9	99.9	43.6	1253.15	99.9	<i>Leonia glycycarpa</i>	FJ670179
2.31	99.71	679	99.7	99.7	43.9	1245.76	99.7	<i>Protium decandrum</i>	FJ037977
2.32	99.85	680	100.0	100.0	42.4	1256.84	99.9	<i>Aparisthium cordatum</i>	KF981218
2.33	99.71	679	99.9	99.9	43.9	1249.46	99.8	<i>Protium decandrum</i>	FJ037977
2.35	99.71	679	100.0	100.0	43.9	1255	99.9	<i>Sterculia pruriens</i>	FJ038104
2.36	99.71	679	100.0	100.0	43.9	1255	99.9	<i>Protium decandrum</i>	FJ037977
2.37	100.00	681	99.1	99.2	42.9	1227.3	99.6	<i>Loxopterygium huasango</i>	GU935431
2.38	100.00	681	99.9	99.9	43.2	1253.15	99.9	<i>Hexopetion mexicanum</i>	JX903251
2.39	100.00	681	100.0	100.0	44.3	1258.69	100.0	<i>Simarouba amara</i>	EU043036

**Table S1.** Continued

Query	Query coverage	Sequence length	% Identical sites	% Pairwise identity	%GC	Bit-score	Grade (%)	Organism	Accession number
2.4	99.41	677	99.9	99.9	41.7	1245.76	99.6	<i>Trymatococcus oligandrus</i>	FJ038126
2.4	99.71	679	100.0	100.0	43.9	1255	99.9	<i>Protium decandrum</i>	FJ037977
2.42	99.71	679	100.0	100.0	43.9	1255	99.9	<i>Protium decandrum</i>	FJ037977
2.5	100.00	681	100.0	100.0	42.9	1258.69	100.0	<i>Hirtella suffulta</i>	KX180070
2.6	99.71	679	99.9	99.9	43.9	1249.46	99.8	<i>Protium decandrum</i>	FJ037977
2.7	99.71	679	100.0	100.0	43.3	1255	99.9	<i>Himatanthus</i> sp.	GQ428618
2.8	91.78	625	99.4	99.6	42.9	1140.5	95.7	<i>Diplotropis purpurea</i>	GQ428606
2.9	99.71	679	99.9	99.9	43.9	1249.46	99.8	<i>Protium decandrum</i>	FJ037977
2B*	100.00	681	98.2	98.2	41.1	1192.21	99.1	<i>Millettia leptobotrya</i>	KJ440056
2C*	100.00	681	99.4	99.4	43.8	1236.53	99.7	<i>Bignonia capreolata</i>	HQ384884
2D*	100.00	681	99.4	99.4	44.3	1236.53	99.7	<i>M.sanderi chloroplast</i>	X91764
2G*	100.00	681	99.3	99.3	43.6	1234.68	99.7	<i>Martinella obovata</i>	L36444
2H*	100.00	681	99.9	99.9	44.2	1253.15	99.9	<i>Securidaca bialata</i>	EU644682
2I*	99.71	679	100.0	100.0	43.9	1255	99.9	<i>Arrabidaea pubescens</i>	AF102641
2J*	100.00	681	99.4	99.4	44.1	1236.53	99.7	<i>Maripa paniculata</i>	AY101046
2K*	100.00	681	99.9	99.9	44.1	1253.15	99.9	<i>Eriandra fragrans</i>	AM234170
3.1	99.71	679	100.0	100.0	43.0	1255	99.9	<i>Guarea silvatica</i>	FJ038158
3.11	99.71	679	100.0	100.0	45.1	1255	99.9	<i>Iryanthera sagotiana</i>	FJ038128
3.16	99.71	679	100.0	100.0	43.4	1255	99.9	<i>Pourouma tomentosa</i>	FJ038203
3.18	99.41	677	100.0	100.0	41.7	1251.3	99.7	<i>Trymatococcus oligandrus</i>	FJ038126
3.2	100.00	681	99.9	99.9	43.5	1253.15	99.9	<i>Proxburghii chloroplast</i>	Z70152
3.24	99.71	679	100.0	100.0	43.9	1255	99.9	<i>Protium decandrum</i>	FJ037977
3.25	100.00	681	99.3	99.3	42.9	1230.99	99.6	<i>Loxopterygium huasango</i>	GU935431
3.26	99.71	679	100.0	100.0	43.9	1255	99.9	<i>Sterculia pruriens</i>	FJ038104
3.27	99.71	679	100.0	100.0	44.2	1255	99.9	<i>Protium sagotianum</i>	FJ037982
3.3	100.00	681	99.9	99.9	42.1	1253.15	99.9	<i>Brosimum alicastrum</i>	AF500346
3.3	100.00	681	100.0	100.0	43.0	1258.69	100.0	<i>Trichilia</i> sp.	KC628372
3.31	99.71	679	99.4	99.4	43.9	1232.84	99.6	<i>Protium decandrum</i>	FJ037977
3.32	100.00	681	99.1	99.1	43.9	1225.45	99.6	<i>Erythroxylum novogranatense</i>	KX256287
3.34	99.71	679	100.0	100.0	43.4	1255	99.9	<i>Aspidosperma cruentum</i>	FJ037963
3.35	100.00	681	99.1	99.1	44.2	1225.45	99.6	<i>Rhabdodendron amazonicum</i>	Z97649
3.36	99.71	679	98.4	99.0	44.5	1214.37	99.4	<i>Licaria guianensis</i>	GQ428566
3.37	99.71	679	100.0	100.0	43.0	1255	99.9	<i>Guarea silvatica</i>	FJ038158
3.39	99.71	679	100.0	100.0	42.7	1255	99.9	<i>Diplotropis purpurea</i>	GQ428606
3.4	100.00	681	99.4	99.4	42.9	1236.53	99.7	<i>Swartzia cardiosperma</i>	AM234259
3.41	99.71	679	100.0	100.0	42.3	1255	99.9	<i>Brosimum guianense</i>	GQ428589
3.42	99.71	679	100.0	100.0	42.7	1255	99.9	<i>Swartzia</i> sp.	FJ038056
3.43	99.71	679	100.0	100.0	43.0	1255	99.9	<i>Guarea silvatica</i>	FJ038158
3.44	99.71	679	99.3	99.3	43.9	1229.14	99.5	<i>Protium decandrum</i>	FJ037977
3.46	99.71	679	100.0	100.0	43.0	1255	99.9	<i>Guarea silvatica</i>	FJ038158
3.47	99.41	677	100.0	100.0	41.7	1251.3	99.7	<i>Trymatococcus oligandrus</i>	FJ038126
3.48	100.00	681	99.4	99.4	42.9	1236.53	99.7	<i>Swartzia cardiosperma</i>	AM234259
3.49	99.71	679	100.0	100.0	43.0	1255	99.9	<i>Guarea silvatica</i>	FJ038158

**Table S1.** Continued

Query	Query coverage	Sequence length	% Identical sites	% Pairwise identity	%GC	Bit-score	Grade (%)	Organism	Accession number
3.5	100.00	681	99.1	99.1	44.2	1225.45	99.6	<i>Rhabdodendron amazonicum</i>	Z97649
3.5	99.41	677	100.0	100.0	41.7	1251.3	99.7	<i>Trymatococcus oligandrus</i>	FJ038126
3.6	100.00	681	100.0	100.0	44.1	1258.69	100.0	<i>Tapirira obtusa</i>	GU935446
3.7	99.71	679	100.0	100.0	43.4	1255	99.9	<i>Pououma tomentosa</i>	FJ038203
3.8	99.27	676	100.0	100.0	44.1	1249.46	99.6	<i>Tapirira obtusa</i>	GU935446
3.9	99.85	680	99.9	99.9	43.1	1251.3	99.9	<i>Abarema brachystachya</i>	KF981222
4.1	99.71	679	100.0	100.0	42.3	1255	99.9	<i>Neea floribunda</i>	FJ038135
4.1	100.00	681	100.0	100.0	44.9	1258.69	100.0	<i>Myristica fragrans</i>	AY298839
4.11	99.71	679	100.0	100.0	42.3	1255	99.9	<i>Brosimum rubescens</i>	GQ428590
4.12	100.00	681	100.0	100.0	43.9	1258.69	100.0	<i>Swietenia mahagoni</i>	FN599465
4.13	99.71	679	100.0	100.0	45.1	1255	99.9	<i>Iryanthera sagotiana</i>	FJ038128
4.14	99.71	679	100.0	100.0	43.9	1255	99.9	<i>Protium decandrum</i>	FJ037977
4.15	100.00	681	100.0	100.0	44.2	1258.69	100.0	<i>Lacistema robustum</i>	JX664056
4.16	99.71	679	100.0	100.0	42.3	1255	99.9	<i>Gustavia hexapetala</i>	FJ038089
4.18	99.85	680	100.0	100.0	41.9	1256.84	99.9	<i>Ormosia arborea</i>	KF981227
4.19	99.71	679	100.0	100.0	45.4	1255	99.9	<i>Siparuna decipiens</i>	FJ038200
4.2	100.00	681	100.0	100.0	43.9	1258.69	100.0	<i>Swietenia mahagoni</i>	FN599465
4.2	99.71	679	99.9	99.9	42.9	1249.46	99.8	<i>Swartzia benthamiana</i>	FJ038055
4.22	99.71	679	100.0	100.0	45.4	1255	99.9	<i>Siparuna decipiens</i>	FJ038200
4.23	100.00	681	100.0	100.0	44.9	1258.69	100.0	<i>Myristica fragrans</i>	AY298839
4.25	99.71	679	100.0	100.0	45.4	1255	99.9	<i>Siparuna decipiens</i>	FJ038200
4.26	100.00	681	100.0	100.0	42.9	1258.69	100.0	<i>Hirtella suffulta</i>	KX180070
4.27	99.85	680	100.0	100.0	42.8	1256.84	99.9	<i>Balizia pedicellaris</i>	KF981225
4.29	100.00	681	100.0	100.0	43.8	1258.69	100.0	<i>M.laxum chloroplast</i>	X91765
4.3	100.00	681	100.0	100.0	44.9	1258.69	100.0	<i>Myristica fragrans</i>	AY298839
4.3	99.71	679	100.0	100.0	43.4	1255	99.9	<i>Aspidosperma cruentum</i>	FJ037963
4.31	99.71	679	99.9	99.9	42.9	1249.46	99.8	<i>Swartzia benthamiana</i>	FJ038055
4.32	99.71	679	100.0	100.0	45.1	1255	99.9	<i>Iryanthera sagotiana</i>	FJ038128
4.33	99.71	679	100.0	100.0	44.8	1255	99.9	<i>Virola kwatae</i>	FJ038129
4.4	99.71	679	100.0	100.0	43.3	1255	99.9	<i>Ptychopetalum olacoides</i>	FJ038139
4.6	100.00	681	100.0	100.0	42.6	1258.69	100.0	<i>Gavarretia terminalis</i>	AY794953
4.7	99.71	679	100.0	100.0	42.3	1255	99.9	<i>Brosimum rubescens</i>	GQ428590
4.8	100.00	681	100.0	100.0	43.9	1258.69	100.0	<i>Swietenia mahagoni</i>	FN599465
4.9	99.71	679	99.9	99.9	42.9	1249.46	99.8	<i>Swartzia benthamiana</i>	FJ038055
5.1	100.00	681	99.3	99.3	44.2	1230.99	99.6	<i>Matisia cordata</i>	AJ233117
5.1	99.71	679	99.4	99.4	42.1	1232.84	99.6	<i>Naucleopsis guianensis</i>	GQ428596
5.13	100.00	681	100.0	100.0	43.2	1258.69	100.0	<i>Diospyros tetrandra</i>	EU980756
5.15	50.22	342	99.4	99.6	46.5	625.288	74.9	<i>Casearia javitensis</i>	JQ626018
5.16	100.00	681	99.9	99.9	43.5	1253.15	99.9	<i>Proxburghii chloroplast</i>	Z70152
5.17	100.00	681	100.0	100.0	44.9	1258.69	100.0	<i>Myristica fragrans</i>	AY298839
5.19	99.71	679	100.0	100.0	43.3	1255	99.9	<i>Diospyros carbonaria</i>	FJ038021
5.2	99.71	679	99.4	99.4	42.1	1232.84	99.6	<i>Naucleopsis guianensis</i>	GQ428596
5.2	99.71	679	100.0	100.0	43.6	1255	99.9	<i>Tetragastris altissima</i>	FJ037987

**Table S1.** Continued

Query	Query coverage	Sequence length	% Identical sites	% Pairwise identity	%GC	Bit-score	Grade (%)	Organism	Accession number
5.21	100.00	681	99.6	99.6	42.9	1242.07	99.8	<i>Trichilia emetica</i>	AY128244
5.22	99.71	679	100.0	100.0	43.3	1255	99.9	<i>Diospyros carbonaria</i>	FJ038021
5.23	99.71	679	100.0	100.0	44.2	1255	99.9	<i>Protium sagotianum</i>	FJ037982
5.24	100.00	681	99.9	99.9	42.1	1253.15	99.9	<i>Brosimum alicastrum</i>	AF500346
5.26	48.16	328	95.4	96.6	42.7	551.422	72.4	<i>Trichilia euneura</i>	JQ625863
5.29	99.71	679	100.0	100.0	43.7	1255	99.9	<i>Pouteria decorticans</i>	FJ038177
5.3	53.01	361	94.5	96.5	45.4	590.202	74.8	<i>Tilia x</i>	KX163059
5.31	100.00	681	99.0	99.0	44.8	1219.91	99.5	<i>Aganosma marginata</i>	AJ419730
5.32	100.00	681	99.3	99.3	44.2	1230.99	99.6	<i>Matisia cordata</i>	AJ233117
5.33	99.71	679	100.0	100.0	44.2	1255	99.9	<i>Protium sagotianum</i>	FJ037982
5.34	99.71	679	100.0	100.0	43.6	1255	99.9	<i>Tetragastris altissima</i>	FJ037987
5.35	99.71	679	100.0	100.0	43.9	1255	99.9	<i>Protium decandrum</i>	FJ037977
5.36	100.00	681	99.1	99.1	43.8	1225.45	99.6	<i>Vallesia antillana</i>	AJ419767
5.4	99.71	679	100.0	100.0	43.6	1255	99.9	<i>Tetragastris altissima</i>	FJ037987
5.5	100.00	681	99.9	99.9	43.5	1253.15	99.9	<i>Sclerolobium sp.</i>	AM234242
5.6	100.00	681	99.9	99.9	42.9	1253.15	99.9	<i>Hirtella suffulta</i>	KX180070
5.7	99.71	679	100.0	100.0	44.2	1255	99.9	<i>Protium sagotianum</i>	FJ037982
5.8	99.71	679	100.0	100.0	43.9	1255	99.9	<i>Cupania scrobiculata</i>	FJ038156
6.1	99.71	679	100.0	100.0	43.6	1255	99.9	<i>Tetragastris altissima</i>	FJ037987
6.1	99.85	680	99.3	99.3	41.9	1229.14	99.6	<i>Ormosia arborea</i>	KF981227
6.12	99.71	679	99.4	99.4	42.1	1232.84	99.6	<i>Naucleopsis guianensis</i>	GQ428596
6.13	99.71	679	100.0	100.0	43.7	1255	99.9	<i>Aspidosperma marcgravianum</i>	FJ037965
6.15	99.71	679	100.0	100.0	43.4	1255	99.9	<i>Pourouma tomentosa</i>	FJ038203
6.16	99.71	679	100.0	100.0	42.9	1255	99.9	<i>Ruizterania albiflora</i>	FJ038212
6.17	99.85	680	99.9	99.9	42.5	1251.3	99.9	<i>Guapira opposita</i>	KF981271
6.19	99.71	679	100.0	100.0	43.4	1255	99.9	<i>Pourouma tomentosa</i>	FJ038203
6.2	99.71	679	100.0	100.0	43.6	1255	99.9	<i>Tetragastris altissima</i>	FJ037987
6.20	97.21	662	99.7	99.7	41.8	1212.52	98.5	<i>Trymatococcus oligandrus</i>	FJ038126
6.22	100.00	681	100.0	100.0	43.0	1258.69	100.0	<i>Cotinus obovatus</i>	GU935422
6.23	99.41	677	100.0	100.0	43.1	1251.3	99.7	<i>Trichilia sp.</i>	KC628372
6.24	99.71	679	99.6	99.6	42.7	1240.22	99.7	<i>Erisma uncinatum</i>	FJ038209
6.25	100.00	681	100.0	100.0	43.5	1258.69	100.0	<i>Manilkara zapota</i>	EU980807
6.27	99.71	679	99.6	99.6	42.7	1240.22	99.7	<i>Erisma uncinatum</i>	FJ038209
6.28	100.00	681	97.8	98.1	42.1	1188.52	99.1	<i>Ampelocera hottleyi</i>	AF500335
6.3	100.00	681	100.0	100.0	43.8	1258.69	100.0	<i>Pachira aquatica</i>	AJ233119
6.3	100.00	681	100.0	100.0	43.5	1258.69	100.0	<i>Sclerolobium sp.</i>	AM234242
6.31	100.00	681	100.0	100.0	43.5	1258.69	100.0	<i>Sclerolobium sp.</i>	AM234242
6.32	99.71	679	100.0	100.0	42.9	1255	99.9	<i>Ruizterania albiflora</i>	FJ038212
6.33	99.71	679	99.6	99.6	42.7	1240.22	99.7	<i>Erisma uncinatum</i>	FJ038209
6.34	99.71	679	99.6	99.6	42.7	1240.22	99.7	<i>Erisma uncinatum</i>	FJ038209
6.35	100.00	681	99.9	99.9	43.3	1253.15	99.9	<i>Pouteria campechiana</i>	KX426215
6.36*	99.71	679	99.7	99.7	43.3	1243.92	99.7	<i>Celastraceae sp.</i>	FJ037994
6.38	99.41	677	99.9	99.9	41.7	1245.76	99.6	<i>Maquira calophylla</i>	FJ038123

**Table S1.** Continued

Query	Query coverage	Sequence length	% Identical sites	% Pairwise identity	%GC	Bit-score	Grade (%)	Organism	Accession number
6.4	99.71	679	100.0	100.0	42.3	1255	99.9	<i>Brosimum guianense</i>	GQ428589
6.41	99.71	679	100.0	100.0	43.6	1255	99.9	<i>Tetragastris altissima</i>	FJ037987
6.42*	99.71	679	99.7	99.7	43.3	1243.92	99.7	<i>Celastraceae sp.</i>	FJ037994
6.43	100.00	681	100.0	100.0	43.5	1258.69	100.0	<i>Sclerolobium sp.</i>	AM234242
6.44	100.00	681	100.0	100.0	43.8	1258.69	100.0	<i>Pachira aquatica</i>	AJ233119
6.45	100.00	681	100.0	100.0	43.5	1258.69	100.0	<i>Manilkara zapota</i>	EU980807
6.6	99.71	679	100.0	100.0	44.2	1255	99.9	<i>Protium sagotianum</i>	FJ037982
6.7	99.71	679	99.9	99.9	41.5	1249.46	99.8	<i>Maquira calophylla</i>	FJ038123
6.8	99.71	679	100.0	100.0	42.9	1255	99.9	<i>Ruizterania albiflora</i>	FJ038212
7.1	100.00	681	100.0	100.0	43.8	1258.69	100.0	<i>M.laxum chloroplast</i>	X91765
7.1	97.65	665	100.0	100.0	43.8	1229.14	98.8	<i>Pseudopiptadenia suaveolens</i>	FJ038053
7.13	95.89	653	100.0	100.0	43.2	1206.98	97.9	<i>Trichilia emetica</i>	AY128244
7.15	99.71	679	100.0	100.0	45.1	1255	99.9	<i>Iryanthera sagotiana</i>	FJ038128
7.16	99.71	679	100.0	100.0	42.6	1255	99.9	<i>Lecythis idatimon</i>	FJ038090
7.19	98.97	674	100.0	100.0	44.1	1245.76	99.5	<i>Protium decandrum</i>	FJ037977
7.2	99.71	679	100.0	100.0	43.9	1255	99.9	<i>Protium decandrum</i>	FJ037977
7.26	99.71	679	99.9	99.9	43.6	1251.3	99.8	<i>Tetragastris altissima</i>	FJ037987
7.27	99.71	679	100.0	100.0	43.3	1255	99.9	<i>Ptychopetalum olacoides</i>	FJ038139
7.3	99.71	679	100.0	100.0	43.6	1255	99.9	<i>Tetragastris altissima</i>	FJ037987
7.4	99.71	679	100.0	100.0	45.1	1255	99.9	<i>Iryanthera sagotiana</i>	FJ038128
7.5	97.21	662	100.0	100.0	44.0	1223.6	98.6	<i>Leonia glycycarpa</i>	FJ670179
7.7	97.36	663	99.5	99.7	45.2	1214.37	98.5	<i>Iryanthera sagotiana</i>	FJ038128

\* = liana

**Table S2.** Blast results for *matK* DNA barcodes against the NCBI online database for samples from seven 25-m<sup>2</sup> plots along a 320-km transect on the Brazil-French Guiana border. 'Query' refers to the tree label as given in the field. The two last columns report the best hit sequence (organism name and accession) to the DNA barcode.

Query	Query coverage	Sequence length	% Identical sites	% Pairwise identity	%GC	Bit-score	Grade (%)	Organism	Accession number
1.1	83.51	770	99.6	99.7	35.1	1410.11	91.6	<i>Trichilia martiana</i>	JQ588367
1.1	30.91	285	99.3	99.6	38.9	520.029	65.3	<i>Protium costaricense</i>	GQ982071
1.11	30.91	285	99.3	99.6	38.9	520.029	65.3	<i>Protium costaricense</i>	GQ982071
1.12*	86.88	801	100.0	100.0	34.2	1480.29	93.4	<i>Forsteronia acouci</i>	EF456339
1.13	82.86	764	99.9	99.9	31.5	1408.27	91.4	<i>Trymatococcus oligandrus</i>	FJ037932
1.16	84.49	780	99.5	99.5	32.4	1417.5	92.0	<i>Castilla elastica</i>	KU856438
1.18	84.16	777	99.7	99.7	33.5	1423.04	92.0	<i>Pouteria campechiana</i>	KX426215
1.2	58.35	538	97.0	98.1	31.8	933.678	78.2	<i>Sterculia multiovula</i>	JQ626455
1.2	83.73	772	99.7	99.8	33.2	1417.5	91.8	<i>Pouteria campechiana</i>	KX426215
1.21	82.32	759	99.6	99.7	31.5	1387.95	91.0	<i>Swartzia panacoco</i>	KT876194
1.22	84.27	777	99.9	99.9	36.3	1432.27	92.1	<i>Carapa guianensis</i>	NC_037442
1.23	83.73	772	99.9	99.9	36.3	1421.19	91.8	<i>Lindera benzoin</i>	MG220609
1.27	83.95	774	99.7	99.7	33.2	1419.35	91.8	<i>Pouteria campechiana</i>	KX426215
1.28	80.91	746	99.9	99.9	33.6	1373.18	90.4	<i>Pouteria campechiana</i>	KX426215
1.29	84.27	777	99.1	99.5	34.9	1410.11	91.9	<i>Protium pallidum</i>	AY594476
1.3	83.19	768	99.0	99.2	33.4	1378.72	91.2	<i>Sterculia apetala</i>	GQ982103

**Table S2.** Continued

Query	Query coverage	Sequence length	% Identical sites	% Pairwise identity	%GC	Bit-score	Grade (%)	Organism	Accession number
1.3	84.27	777	98.6	99.3	34.9	1395.34	91.8	<i>Protium pallidum</i>	AY594476
1.32	81.24	749	99.7	99.7	32.3	1373.18	90.5	<i>Castilla elastica</i>	KU856438
1.33	83.41	770	99.2	99.2	33.2	1387.95	91.3	<i>Pouteria campechiana</i>	KX426215
1.34	84.27	777	99.1	99.4	31.7	1406.42	91.8	<i>Swartzia panacoco</i>	KT876194
1.35	84.27	777	99.9	99.9	33.5	1430.43	92.1	<i>Pouteria campechiana</i>	KX426215
1.36	84.27	777	100.0	100.0	33.5	1435.97	92.1	<i>Pouteria campechiana</i>	KX426215
1.39	82.97	765	99.1	99.2	34.9	1380.57	91.1	<i>Protium costaricense</i>	JQ587172
1.4	84.27	777	100.0	100.0	33.5	1435.97	92.1	<i>Pouteria campechiana</i>	KX426215
1.43	84.27	777	99.7	99.9	36.3	1428.58	92.1	<i>Guatteria ouregou</i>	KP859342
1.47	30.69	283	99.3	99.6	39.2	516.336	65.2	<i>Protium costaricense</i>	GQ982071
1.48	82.75	763	99.6	99.7	32.6	1395.34	91.2	<i>Theobroma cacao</i>	MF350235
1.5	66.81	616	97.7	98.7	34.3	1083.26	82.8	<i>Sterculia pruriens</i>	FJ514606
1.5	80.26	740	100.0	100.0	35.1	1367.64	90.1	<i>Protium pallidum</i>	AY594476
1.6	83.62	771	99.0	99.5	34.6	1395.34	91.6	<i>Protium pallidum</i>	AY594476
1.7	84.27	777	99.4	99.7	31.9	1417.5	92.0	<i>Balizia</i> sp.	KX302319
1.8	79.18	730	98.2	99.0	35.2	1299.31	89.1	<i>Trichilia martiana</i>	JQ588367
2.1	82.75	763	98.8	98.8	32.6	1360.25	90.8	<i>Eugenia uniflora</i>	KR867678
2.1	83.84	773	99.7	99.9	31.7	1421.19	91.9	<i>Hirtella macrosepala</i>	KX180068
2.12	75.60	697	100.0	100.0	33.4	1288.24	87.8	<i>Sclerosperma mannii</i>	AM114629
2.13	83.62	771	99.9	99.9	33.2	1421.19	91.8	<i>Pouteria campechiana</i>	KX426215
2.15	80.80	745	99.2	99.3	32.7	1347.33	90.0	<i>Chaunochiton kappleri</i>	DQ790179
2.16	83.19	771	98.3	98.4	32.7	1356.56	90.8	<i>Phytelephas aequatorialis</i>	KT312924
2.16	75.60	697	100.0	100.0	33.4	1288.24	87.8	<i>Sclerosperma mannii</i>	AM114629
2.17	81.45	751	99.6	99.6	34.9	1371.33	90.5	<i>Ferdinandusa chlorantha</i>	FJ905361
2.18	80.69	744	99.9	99.9	35.8	1369.49	90.3	<i>Trichilia prieureana</i>	KC627568
2.19	84.27	777	99.6	99.7	33.7	1421.19	92.0	<i>Vochysia ferruginea</i>	GQ982128
2.2	84.27	777	99.7	99.8	36.4	1426.73	92.0	<i>Guatteria</i> cf.	KP859347
2.2	19.74	182	99.5	99.7	39.6	333.517	59.7	<i>Bursera fagaroides</i>	KF224981
2.21	78.20	721	99.2	99.4	31.8	1310.39	88.8	<i>Hirtella macrosepala</i>	KX180068
2.21	69.63	642	98.4	99.1	30.7	1149.74	84.4	<i>Hirtella macrosepala</i>	KX180068
2.23	82.54	761	98.3	98.4	35.5	1336.25	90.4	<i>Toxicodendron succedaneum</i>	HQ427343
2.24	84.27	777	99.5	99.7	31.7	1419.35	92.0	<i>Swartzia panacoco</i>	KT876194
2.26	84.60	780	99.6	99.7	33.6	1426.73	92.1	<i>Vochysia ferruginea</i>	GQ982128
2.27	84.60	780	99.5	99.5	31.3	1419.35	92.0	<i>Conceveiba martiana</i>	FJ670011
2.28	84.82	782	99.1	99.2	31.5	1410.11	92.0	<i>Brosimum alicastrum</i>	GQ981947
2.31	82.21	758	100.0	100.0	36.3	1400.88	91.1	<i>Ocotea catharinensis</i>	KF555429
2.32	83.62	771	98.6	99.0	31.3	1376.87	91.3	<i>Conceveiba martiana</i>	FJ670011
2.33	83.08	766	98.6	99.1	34.9	1369.49	91.1	<i>Protium costaricense</i>	GQ982071
2.34	84.49	779	99.4	99.4	34.3	1411.96	91.9	<i>Chimarrhis parviflora</i>	GQ981964
2.35	83.19	768	99.2	99.3	33.4	1386.11	91.2	<i>Sterculia apetala</i>	GQ982103
2.36	83.95	774	99.7	99.8	34.8	1421.19	91.9	<i>Protium costaricense</i>	GQ982071
2.37	84.82	782	99.0	99.4	34.9	1411.96	92.1	<i>Astronium graveolens</i>	JQ586469

**Table S2.** Continued

Query	Query coverage	Sequence length	% Identical sites	% Pairwise identity	%GC	Bit-score	Grade (%)	Organism	Accession number
2.38	75.60	697	100.0	100.0	33.4	1288.24	87.8	<i>Sclerosperma mannii</i>	AM114629
2.4	77.98	719	99.3	99.4	31.7	1303.01	88.7	<i>Trymatococcus oligandrus</i>	FJ037932
2.4	80.80	745	99.5	99.7	35.2	1362.1	90.3	<i>Protium costaricense</i>	GQ982071
2.42	19.74	182	98.4	99.2	39.6	326.131	59.5	<i>Bursera fagaroides</i>	KF224981
2.5	87.20	804	99.8	99.9	31.8	1478.44	93.5	<i>Hirtella macrosepala</i>	KX180068
2.6	83.95	774	99.4	99.5	34.8	1406.42	91.7	<i>Protium costaricense</i>	GQ982071
2.7	83.73	773	99.9	99.9	34.9	1421.19	91.8	<i>Himatanthus bracteatus</i>	EF456366
2.9	81.56	752	99.3	99.4	35.2	1365.79	90.5	<i>Protium costaricense</i>	JQ587172
2A*	83.51	770	99.4	99.4	33.8	1395.34	91.4	<i>Salacia oblonga</i>	KX573076
2B*	84.92	783	99.1	99.2	27.8	1411.96	92.1	<i>Deguelia negrensis</i>	JX506607
2C*	84.92	783	99.5	99.6	34.7	1426.73	92.2	<i>Adenocalymma validum</i>	MG831871
2D*	85.68	790	98.9	98.9	33.9	1411.96	92.3	<i>Forsteronia acouci</i>	EF456339
2G*	83.62	772	98.7	98.8	34.3	1371.33	91.2	<i>Anemopaegma foetidum</i>	NC_037230
2H*	84.38	778	99.9	99.9	30.3	1432.27	92.1	<i>Securidaca diversifolia</i>	JQ588835
2I*	81.89	756	99.7	99.7	34.5	1384.26	90.8	<i>Bignoniaceae sp.</i>	JQ586988
2J*	82.21	758	96.0	97.6	34.4	1277.16	89.9	<i>Maripa nicaraguensis</i>	JQ587303
2K*	75.49	697	96.3	96.3	32.4	1142.35	85.9	<i>Carpolobia conradsiana</i>	JX517551
3.1	82.65	762	97.9	98.8	35.0	1343.63	90.7	<i>Guarea pterorhachis</i>	JQ588347
3.11	80.48	742	99.9	99.9	36.3	1367.64	90.2	<i>Haematodendron glabrum</i>	AY220447
3.12	84.27	777	99.9	99.9	31.3	1432.27	92.1	<i>Tachigali sp.</i>	KX538536
3.12	84.27	777	99.5	99.6	31.3	1421.19	91.9	<i>Tachigali sp.</i>	KX538536
3.13	82.00	756	99.2	99.4	36.2	1373.18	90.7	<i>Ocotea catharinensis</i>	KF555429
3.14	80.15	739	99.5	99.5	32.8	1341.79	89.8	<i>Qualea rosea</i>	JQ626462
3.15	84.27	777	99.7	99.7	32.2	1424.89	92.0	<i>Guapira riedeliana</i>	FN597630
3.16	82.54	761	99.5	99.6	33.4	1387.95	91.1	<i>Pououma bicolor</i>	GQ982067
3.18	82.86	764	100.0	100.0	31.5	1411.96	91.4	<i>Trymatococcus oligandrus</i>	FJ037932
3.26	82.32	760	98.3	98.8	33.2	1345.48	90.6	<i>Sterculia apetala</i>	GQ982103
3.27	78.96	728	99.2	99.5	35.6	1319.63	89.2	<i>Protium costaricense</i>	GQ982071
3.28	80.91	746	99.7	99.7	33.6	1367.64	90.3	<i>Vochysia ferruginea</i>	GQ982128
3.29	80.15	739	99.5	99.5	32.8	1341.79	89.8	<i>Qualea rosea</i>	JQ626462
3.3	81.67	753	99.7	99.7	31.7	1380.57	90.7	<i>Brosimum lactescens</i>	KU856472
3.3	78.42	723	99.3	99.6	35.4	1315.93	89.0	<i>Trichilia martiana</i>	JQ588367
3.31	78.52	724	99.0	99.4	35.5	1308.55	89.0	<i>Protium costaricense</i>	GQ982071
3.32	80.04	738	99.7	99.7	32.8	1352.87	89.9	<i>Erythroxylum macrophyllum</i>	GQ981986
3.33	79.39	732	99.0	99.3	35.9	1327.01	89.3	<i>Haematodendron glabrum</i>	AY220447
3.33	77.98	719	98.7	99.2	35.7	1295.62	88.6	<i>Haematodendron glabrum</i>	AY220447
3.34	80.26	743	98.4	98.5	35.4	1306.7	89.4	<i>Aspidosperma triternatum</i>	AM295077
3.35	79.61	734	99.6	99.7	33.4	1341.79	89.6	<i>Rhabdodendron amazonicum</i>	JQ844136
3.36	66.92	617	99.2	99.6	37.0	1122.04	83.3	<i>Licaria chrysophylla</i>	JQ626395
3.37	80.69	744	98.4	99.0	35.2	1325.17	89.8	<i>Guarea pterorhachis</i>	JQ588347
3.39	13.34	123	94.3	97.0	32.5	202.405	55.1	<i>Minquartia guianensis</i>	KU247535
3.4	81.24	749	100.0	100.0	33.0	1384.26	90.6	<i>Inga paraensis</i>	KX374525

**Table S2.** Continued

Query	Query coverage	Sequence length	% Identical sites	% Pairwise identity	%GC	Bit-score	Grade (%)	Organism	Accession number
3.4	80.69	744	98.9	99.0	31.6	1332.55	89.8	<i>Swartzia panacoco</i>	KT876194
3.41	81.56	752	99.5	99.6	31.7	1371.33	90.6	<i>Brosimum alicastrum</i>	GQ981947
3.42	75.16	693	99.9	99.9	31.9	1277.16	87.5	<i>Swartzia canescens</i>	JQ626472
3.43	80.80	745	98.9	99.3	35.3	1341.79	90.0	<i>Guarea pterorhachis</i>	JQ588347
3.44	73.43	677	98.8	99.3	35.9	1218.06	86.3	<i>Protium opacum</i>	JQ626503
3.45	58.89	543	98.5	98.8	32.8	972.458	78.9	<i>Guarea grandifolia</i>	GQ982002
3.45	60.41	557	98.2	99.0	33.0	990.925	79.7	<i>Guarea grandifolia</i>	GQ982002
3.46	82.75	763	98.7	99.1	35.1	1367.64	91.0	<i>Guarea pterorhachis</i>	JQ588347
3.47	81.89	755	99.9	99.9	31.5	1391.65	90.9	<i>Trymatococcus oligandrus</i>	FJ037932
3.48	83.30	768	98.6	98.8	31.3	1365.79	91.1	<i>Swartzia panacoco</i>	KT876194
3.49	84.16	777	99.5	99.5	35.1	1411.96	91.8	<i>Guarea grandifolia</i>	GQ982002
3.5	77.77	717	99.6	99.7	33.5	1310.39	88.7	<i>Rhabdodendron amazonicum</i>	JQ844136
3.5	82.86	764	100.0	100.0	31.5	1411.96	91.4	<i>Trymatococcus oligandrus</i>	FJ037932
3.6	80.69	744	96.9	98.4	36.0	1290.08	89.6	<i>Tapirira guianensis</i>	KF981295
3.7	82.32	759	99.3	99.5	33.5	1380.57	90.9	<i>Pououma bicolor</i>	GQ982067
3.9	80.59	743	99.5	99.7	31.6	1358.41	90.2	<i>Balizia sp.</i>	KX302319
4.1	84.27	777	99.9	99.9	32.2	1430.43	92.1	<i>Guapira riedeliana</i>	FN597630
4.1	84.27	777	100.0	100.0	36.2	1435.97	92.1	<i>Virola michelii</i>	AY220454
4.11	84.82	782	99.1	99.1	31.5	1408.27	92.0	<i>Brosimum alicastrum</i>	GQ981947
4.12	84.27	777	99.9	99.9	36.3	1430.43	92.1	<i>Carapa guianensis</i>	NC_037442
4.13	84.27	777	100.0	100.0	36.0	1435.97	92.1	<i>Haematodendron glabrum</i>	AY220447
4.15	85.57	789	99.2	99.2	31.9	1424.89	92.4	<i>Lacistema aggregatum</i>	FJ670025
4.16	84.27	777	97.8	97.8	32.2	1341.79	91.0	<i>Grias cauliflora</i>	MF359952
4.17	84.16	776	99.7	99.7	36.5	1423.04	92.0	<i>Cinnamomum aromaticum</i>	MF627719
4.18	84.16	776	99.9	99.9	30.0	1428.58	92.0	<i>Ormosia arborea</i>	KX816384
4.19	79.72	735	100.0	100.0	34.8	1358.41	89.9	<i>Siparuna decipiens</i>	JQ626498
4.2	84.27	777	99.9	99.9	36.3	1430.43	92.1	<i>Carapa guianensis</i>	NC_037442
4.2	83.73	772	100.0	100.0	31.2	1426.73	91.9	<i>Swartzia panacoco</i>	KT876194
4.21	80.59	743	99.6	99.7	30.1	1360.25	90.1	<i>Ormosia arborea</i>	KX816384
4.21	79.50	733	99.0	99.4	30.2	1327.01	89.4	<i>Ormosia arborea</i>	KX816384
4.22	79.83	736	100.0	100.0	34.9	1360.25	89.9	<i>Siparuna decipiens</i>	JQ626498
4.23	84.27	777	100.0	100.0	36.2	1435.97	92.1	<i>Virola michelii</i>	AY220454
4.25	79.83	736	100.0	100.0	34.9	1360.25	89.9	<i>Siparuna decipiens</i>	JQ626498
4.26	87.20	804	100.0	100.0	31.8	1485.83	93.6	<i>Hirtella macrosepala</i>	KX180068
4.27	84.27	777	100.0	100.0	32.0	1435.97	92.1	<i>Balizia pedicellaris</i>	KF981315
4.28	82.75	763	99.9	99.9	34.5	1404.57	91.3	<i>Minquartia guianensis</i>	KU247535
4.29	85.90	792	100.0	100.0	34.0	1463.67	93.0	<i>Macoubea guianensis</i>	GU973901
4.3	84.27	777	100.0	100.0	36.2	1435.97	92.1	<i>Virola michelii</i>	AY220454
4.3	83.62	774	98.7	98.7	35.5	1371.33	91.2	<i>Aspidosperma triternatum</i>	AM295077
4.31	83.73	772	100.0	100.0	31.2	1426.73	91.9	<i>Swartzia panacoco</i>	KT876194
4.32	84.27	777	100.0	100.0	36.0	1435.97	92.1	<i>Haematodendron glabrum</i>	AY220447
4.33	84.27	777	100.0	100.0	35.8	1435.97	92.1	<i>Virola nobilis</i>	GQ982126
4.34	83.95	774	99.6	99.6	34.8	1413.81	91.8	<i>Protium costaricense</i>	GQ982071

**Table S2.** Continued

Query	Query coverage	Sequence length	% Identical sites	% Pairwise identity	%GC	Bit-score	Grade (%)	Organism	Accession number
4.6	84.92	783	99.9	99.9	31.5	1441.51	92.4	<i>Conceveiba martiana</i>	FJ670011
4.7	84.82	782	99.1	99.1	31.5	1408.27	92.0	<i>Brosimum alicastrum</i>	GQ981947
4.8	84.27	777	99.9	99.9	36.3	1430.43	92.1	<i>Carapa guianensis</i>	NC_037442
4.9	83.73	772	100.0	100.0	31.2	1426.73	91.9	<i>Swartzia panacoco</i>	KT876194
5.1	83.30	768	99.6	99.6	33.2	1402.73	91.5	<i>Patinoa sphaerocarpa</i>	AY589074
5.1	81.89	755	99.7	99.8	32.2	1386.11	90.8	<i>Castilla elastica</i>	KU856438
5.11	84.27	777	99.5	99.5	35.4	1413.81	91.9	<i>Synima cordieri</i>	AY724333
5.13	84.27	777	100.0	100.0	33.2	1435.97	92.1	<i>Diospyros tetrandra</i>	DQ924058
5.14	84.27	777	100.0	100.0	32.3	1435.97	92.1	<i>Hymenaea courbaril</i>	KX538511
5.16	83.62	771	99.2	99.2	31.8	1386.11	91.4	<i>Parkia multijuga</i>	EU362018
5.17	84.27	777	100.0	100.0	36.2	1435.97	92.1	<i>Virola michelii</i>	AY220454
5.18	76.03	701	100.0	100.0	36.5	1295.62	88.0	<i>Virola michelii</i>	JQ626468
5.19	84.27	777	99.9	99.9	33.5	1430.43	92.1	<i>Diospyros dichroa</i>	DQ924011
5.2	83.95	774	99.6	99.6	34.8	1413.81	91.8	<i>Protium costaricense</i>	GQ982071
5.22	84.27	777	99.9	99.9	33.5	1430.43	92.1	<i>Diospyros dichroa</i>	DQ924011
5.23	83.95	774	99.5	99.5	34.8	1408.27	91.7	<i>Protium costaricense</i>	GQ982071
5.24	85.25	786	99.9	99.9	31.8	1447.05	92.6	<i>Brosimum lactescens</i>	KU856472
5.25	74.95	691	99.7	99.8	36.8	1269.77	87.4	<i>Carapa guianensis</i>	NC_037442
5.29	84.27	777	99.9	99.9	33.5	1430.43	92.1	<i>Pouteria campechiana</i>	KX426215
5.30	54.12	499	97.0	98.5	34.7	867.199	76.3	<i>Iryanthera sagotiana</i>	JQ626420
5.31	85.90	792	100.0	100.0	35.4	1463.67	93.0	<i>Odontadenia perrottetii</i>	EF456272
5.32	83.30	768	99.6	99.6	33.2	1402.73	91.5	<i>Patinoa sphaerocarpa</i>	AY589074
5.33	83.95	774	99.5	99.5	34.8	1408.27	91.7	<i>Protium costaricense</i>	GQ982071
5.34	83.95	774	99.6	99.6	34.8	1413.81	91.8	<i>Protium costaricense</i>	GQ982071
5.36	84.27	777	99.2	99.4	34.8	1408.27	91.8	<i>Geissospermum laeve</i>	DQ660517
5.4	77.01	710	98.2	98.6	35.9	1260.54	87.8	<i>Protium costaricense</i>	JQ587172
5.5	82.75	763	99.6	99.6	31.1	1395.34	91.2	<i>Tachigali</i> sp.	KX538536
5.6	87.20	804	100.0	100.0	31.8	1485.83	93.6	<i>Hirtella macrosepala</i>	KX180068
5.7	75.05	692	97.4	98.6	36.0	1210.68	86.8	<i>Protium costaricense</i>	GQ982071
5.7	77.11	711	96.5	97.7	35.6	1212.52	87.4	<i>Protium costaricense</i>	GQ982071
5.8	84.27	777	99.5	99.5	35.4	1413.81	91.9	<i>Synima cordieri</i>	AY724333
6.1	69.85	644	98.6	99.1	35.8	1155.28	84.5	<i>Geissospermum laeve</i>	DQ660517
6.1	82.97	765	99.3	99.3	30.2	1386.11	91.2	<i>Ormosia coutinhoi</i>	KY079016
6.13	83.95	774	99.0	99.0	35.5	1386.11	91.5	<i>Aspidosperma triternatum</i>	AM295077
6.14	83.95	774	99.6	99.6	34.8	1413.81	91.8	<i>Protium costaricense</i>	GQ982071
6.15	85.90	792	99.6	99.6	33.6	1447.05	92.8	<i>Pououma bicolor</i>	GQ982067
6.16	84.60	780	98.6	98.7	32.6	1382.41	91.6	<i>Qualea grandiflora</i>	AF368216
6.17	84.27	777	99.7	99.7	32.2	1424.89	92.0	<i>Guapira riedeliana</i>	FN597630
6.19	85.90	792	99.6	99.6	33.6	1447.05	92.8	<i>Pououma bicolor</i>	GQ982067
6.2	81.34	750	98.4	99.0	35.2	1336.25	90.2	<i>Protium costaricense</i>	JQ587172
6.2	59.87	552	99.5	99.6	33.0	1007.54	79.7	<i>Brosimum alicastrum</i>	GQ981947
6.22	85.47	789	98.4	98.4	34.9	1389.8	92.0	<i>Pachycormus discolor</i>	AY594493
6.24	80.59	743	99.7	99.7	33.5	1362.1	90.2	<i>Vochysia ferruginea</i>	GQ982128

**Table S2.** Continued

Query	Query coverage	Sequence length	% Identical sites	% Pairwise identity	%GC	Bit-score	Grade (%)	Organism	Accession number
6.25	84.27	777	99.9	99.9	33.6	1430.43	92.1	<i>Micropholis gnaphaloclados</i>	JQ413918
6.26	77.98	719	99.6	99.6	35.5	1312.24	88.8	<i>Protium costaricense</i>	GQ982071
6.27	84.60	780	99.6	99.7	33.6	1426.73	92.1	<i>Vochysia ferruginea</i>	GQ982128
6.3	83.30	768	99.7	99.7	33.1	1408.27	91.5	<i>Eriotheca macrophylla</i>	HQ696713
6.3	70.17	647	99.2	99.4	32.2	1175.59	84.8	<i>Tachigali</i> sp.	KX538536
6.31	84.27	777	99.7	99.8	31.3	1426.73	92.0	<i>Tachigali</i> sp.	KX538536
6.32	84.60	780	98.6	98.7	32.6	1382.41	91.6	<i>Qualea grandiflora</i>	AF368216
6.33	84.60	780	99.7	99.7	33.6	1430.43	92.2	<i>Vochysia ferruginea</i>	GQ982128
6.34	84.60	780	99.7	99.7	33.6	1430.43	92.2	<i>Vochysia ferruginea</i>	GQ982128
6.35	84.27	777	99.9	99.9	33.5	1430.43	92.1	<i>Pouteria campechiana</i>	KX426215
6.36*	84.27	777	99.9	99.9	33.5	1430.43	92.1	<i>Hippocratea volubilis</i>	HM230173
6.38	80.69	744	98.9	98.9	32.7	1330.71	89.8	<i>Castilla elastica</i>	KU856438
6.39	83.84	774	99.5	99.5	31.0	1410.11	91.7	<i>Tachigali</i> sp.	KX538536
6.4	31.34	289	99.7	99.7	32.2	531.109	65.5	<i>Brosimum guianense</i>	JQ626530
6.4	83.95	774	99.4	99.4	34.8	1406.42	91.7	<i>Protium costaricense</i>	GQ982071
6.41	82.43	760	99.5	99.5	35.0	1384.26	91.0	<i>Protium costaricense</i>	GQ982071
6.42*	83.84	773	99.7	99.8	33.5	1419.35	91.8	<i>Hippocratea volubilis</i>	HM230173
6.44	83.30	768	99.7	99.7	33.1	1408.27	91.5	<i>Eriotheca macrophylla</i>	HQ696713
6.45	84.27	777	99.9	99.9	33.6	1430.43	92.1	<i>Micropholis gnaphaloclados</i>	JQ413918
6.6	83.95	774	99.7	99.7	34.8	1419.35	91.8	<i>Protium costaricense</i>	GQ982071
6.7	51.08	471	98.9	99.0	33.8	845.039	75.1	<i>Castilla elastica</i>	KU856438
6.8	84.60	780	98.7	98.7	32.6	1386.11	91.7	<i>Qualea grandiflora</i>	AF368216
6.9	83.51	770	98.3	98.5	35.1	1362.1	91.0	<i>Trichilia martiana</i>	JQ588367
6.9	83.51	770	98.3	98.7	35.1	1362.1	91.1	<i>Trichilia martiana</i>	JQ588367
7.1	85.90	792	100.0	100.0	34.0	1463.67	93.0	<i>Macoubea guianensis</i>	GU973901
7.1	81.02	747	99.7	99.8	31.9	1371.33	90.4	<i>Pseudopiptadenia suaveolens</i>	DQ790637
7.12	80.59	743	99.9	99.9	35.4	1367.64	90.2	<i>Minquartia guianensis</i>	KU247535
7.13	82.21	758	99.9	99.9	35.2	1395.34	91.0	<i>Trichilia martiana</i>	JQ588367
7.14	78.52	724	100.0	100.0	35.5	1338.09	89.3	<i>Protium opacum</i>	JQ626503
7.15	84.27	777	100.0	100.0	36.0	1435.97	92.1	<i>Haematodendron glabrum</i>	AY220447
7.16	82.10	757	99.7	99.7	32.8	1387.95	90.9	<i>Lecythis pneumatophora</i>	MF359953
7.19	83.95	774	99.7	99.7	34.8	1419.35	91.8	<i>Protium costaricense</i>	GQ982071
7.2	78.09	720	99.7	99.8	35.6	1323.32	88.9	<i>Protium costaricense</i>	GQ982071
7.24	84.27	777	100.0	100.0	36.2	1435.97	92.1	<i>Virola michelii</i>	AY220454
7.26	83.95	774	99.2	99.3	34.8	1402.73	91.6	<i>Protium costaricense</i>	GQ982071
7.27	80.69	744	99.2	99.2	35.1	1341.79	89.9	<i>Ptychopetalum petiolatum</i>	KC627490
7.3	83.95	774	99.5	99.5	34.8	1410.11	91.7	<i>Protium costaricense</i>	GQ982071
7.4	84.27	777	100.0	100.0	36.0	1435.97	92.1	<i>Haematodendron glabrum</i>	AY220447
7.5	76.14	702	100.0	100.0	33.5	1297.47	88.1	<i>Leonia glycycarpa</i>	JQ626572
7.6	78.42	723	99.9	99.9	35.1	1330.71	89.1	<i>Minquartia guianensis</i>	KU247535
7.7	84.27	777	99.5	99.6	36.0	1421.19	91.9	<i>Haematodendron glabrum</i>	AY220447

\* = liana