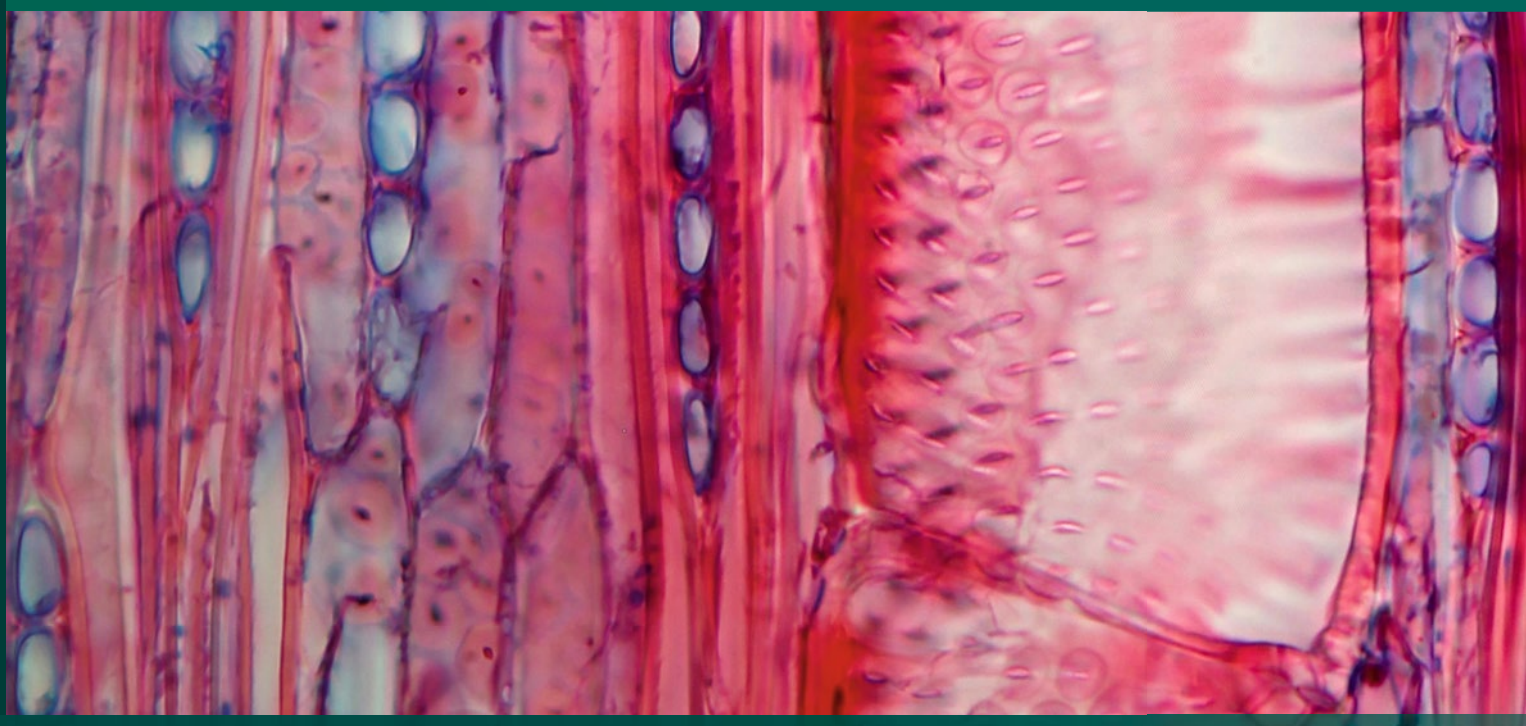

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with a Special Focus on Wood Identification

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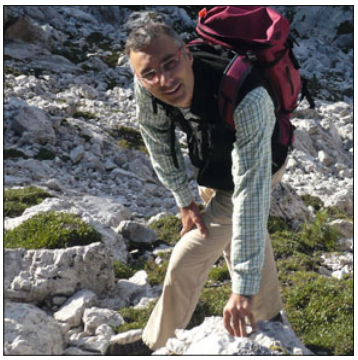
For this book, she collected the plant material, prepared the slides, realized the anatomical descriptions and photomicrographs, and managed the graphic design of the atlas.



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For this book, he contributed to the anatomical descriptions, and to the layout of the atlas.

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He supervised the material preparation, anatomical descriptions, and atlas design of the present book.

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This work was inspired by the results of my long work as Permanent Secretary within the CITES Scientific Authority at the Department of Plant Biology and Ecology, set up by the listing of Malagasy *Dalbergia* and *Diospyros* in Appendix II.

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Bako Harisoa Ravaomanalina

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1. Introduction

Background

Madagascar has an exceptional biodiversity which is characterized by a high level of endemisms with a remarkable variety of bioclimates and habitats, including humid forests, dry deciduous forests, dry thicket forests, littoral forests, coastal zones and central high mountains. Malagasy natural forests are renowned for precious woods, which are a main forest product with a considerable added value on both international and national markets. The precious woods of Madagascar are mainly produced by members of 2 genera, *Dalbergia* and *Diospyros*. A total 48 species of *Dalbergia* are recognized, only one of which is non endemic (Bosser & Rabevohitra 2005), and some of which produce rosewood and palisander timber.

The taxonomy of *Diospyros* is currently being revised. A total of 85 described species are currently recognized, 82 of which endemic and 3 are not endemic, as summarized in the catalogue of Vascular Plant of Madagascar (www.tropicos.org/project/mada), and at least 130 additional species remain to be named and de scribed (Pete Lowry, pers. comm.)

These timbers have a significant economic role in Madagascar. Villagers use *Dalbergia* species as fuel. Rosewood and palisander species are exported as raw logs or finished products as furniture and handicrafts. Ebony wood (*Diospyros* spp.) is used for the manufacture of luxury goods, cutlery, brushes, marquetry, canes, lutes, wind musical instruments and piano keys. This black wood is highly demanded on the international market because of its colour, sturdy, hardness and resistance.

The continuing increase of international demand encourages illegal trade and harvesting of these species. Selective and excessive harvesting of mature, seed-bearing specimens leads to rarefaction of large-diameter trees. Extensive harvesting initiated small-diameter logging of trees for the handcraft products and musical instruments. Wood constitutes nearly the total energy supply especially in the region of Morondava.

Over 90% of exported products are logs and sawn wood and only 10% of wood are sold on local markets.

Timber identification on the international market is not possible from logs because morphological traits used in species identification such as flowers, bark and fruits have been removed. Therefore, it is possible for customs authorities to distinguish legally from illegally harvested logs. For these reasons, Malagasy precious woods *Dalbergia* and *Diospyros* have received protection under CITES Appendix II in 2013. The listing of these species helps to ensure the legality of trade, the traceability of wood and wood products, and it should help to ensure that the exploitation is not detrimental to the survival of these species. Identification is the basic element of CITES enforcement. To facilitate adequate implementation of the Appendix II listing of *Dalbergia* spp. and *Diospyros* spp., Madagascar presented an action plan in which establishment of reliable and fast identification techniques for logs and wood products need to be developed and implemented.

Considerable effort occurred to enhance anatomical, chemical and physical methods to address this problem in all countries with natural occurrences of these species. In general, wood anatomy is the most used and appropriate method to identify timber species. Gasson *et al*. (2010) pursued quantitative anatomical characters using PCA and Bayes comparisons, and managed to identify all *Dalbergia nigra* samples. Miller and Wiemann (2006) compared the wood of *Dalbergia nigra* and *Dalbergia spruceana* and used the different fluorescence of water extracts and wood density to separate the two. Kite *et al*. (2010) found the flavonoid that is unique to *Dalbergia nigra*, at least in the 15 American, African and Asian species

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compared. Pigozzo *et al*. (2010) have suggested to distinguish *Dalbergia* species, especially *Dalbergia nigra* and *Dalbergia spruceana* by using infrared spectrometry. A rapid chemical method of analysis was developed by Lancaster and Espinoza (2012) to differentiate commonly traded species, among them *Dalbergia baronii* and *Dalbergia madagascariensis*. Mass spectrometry was applied to distinguish different species of *Dalbergia* at the international level, however species from Madagascar need more detailed analyses (Espinoza et al. 2015; McClure et al. 2015; Lancaster & Spinoza, 2012).
However, except the wood anatomical descriptions recently given by P. Détienne, available on InsideWood (Wheeler, 2011), extensive studies of Malagasy *Dalbergia* species in relation to wood identification are still lacking. Anatomical descriptions of *Diospyros lotus* and *Diospyros kaki* have been made by Schweingruber (1990) and Saldari *et al*. (2008). Changes in ray arrangement of *Diospyros lotus* was analyzed by Myśkow and Zagórska-Marek (2012) and some physical, biometry and mechanical strength properties were determined by Kiaei and Bakhshi (2014). Anatomical descriptions of some Malagasy *Diospyros* species are available on the wood database InsideWood. Moreover, not all the descriptions are based on vouchered wood samples, and we have to take in mind that some woods may not have been correctly identified (IAWA, 2011) in addition to that, the genus *Diospyros* is currently on taxonomic revision. All descriptions studied in this book are based on vouchered wood samples and they are available in collections. The species identification was made by the taxonomy specialists Georges Schatz and Pete Lowry II, Missouri Botanical Garden, France and US.

About the manual

This manual replies to the expectations of the action plan for *Diospyros* and *Dalbergia* about the establishment of a reference collection and identification system for CITES-listed species. Since this is the first time that an anatomical atlas about Malagasy precious wood was planned we decided to give an overview about xylem and bark from twigs, branches and stems. Special focus was given on stem wood identification. This study will address a limited number of species, relevant to the international trade. The monographic presentation are arranged by genera, and within them by species in alphabetical order.
A key for the identification of *Dalbergia* and *Diospyros* species is provided on the base of stem wood anatomical features.
This atlas covers 19 *Dalbergia* species and 31 *Diospyros* species. Among the described species, *Dalbergia bracteolata* and *Diospyros ferrea* are the only two species which are not endemic to Madagascar. Ten *Diospyros* species were not described and published taxonomically (*Diospyros acutiflora*, *D. antongiliensis*, *D. brevipedicellata*, *D. chitinophora*, *D. maculata*, *D. maxima*, *D. olacinoides*, *D quadrangularis*, *D. rubripetiolata*, *D. tropophylloides*). The codification of anatomical features is based on one individual, represented by the stem, branch and twig. Since *Dalbergia* and *Diospyros* species grow in different vegetation formations in Madagascar, ecological adaptations are expressed by various anatomical structures.

2. Materials and Methods

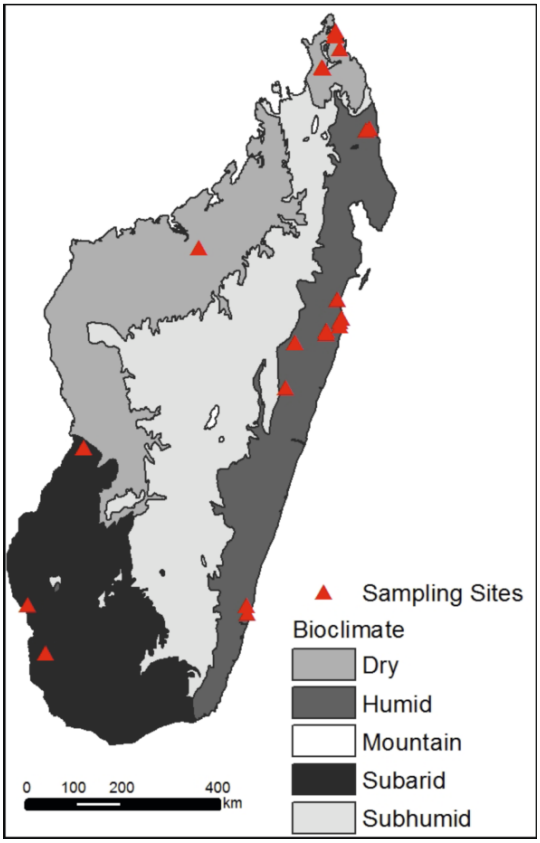
Origin of the material studied

The sampling of plant material was carried out in Madagascar in July 2013 and January 2014. The sampling sites are indicated on the bioclimatic map. All the species were collected in their natural habitat. Species identification was carried out by the taxonomists of Missouri Botanical Garden both in Madagascar and in Missouri. General botanical information were carried out by Missouri Botanical Garden Madagascar (2009, 2014) and the Department of Plant Biology and Ecology (2010, 2011). Data distributions were derived from the Catalogue of Vascular Plant of Madagascar (www.tropicos.org). Consequently, database per specimens used in the atlas are available on the same web site. Herbarium specimens of the presented material can be consulted at the international herbaria Missouri (MBG), Paris (MNHN), Brussels (RMCA) and Zürich (Botanical Garden) while wood specimens were available from wood collection of Tervuren in Brussels (RMCA).

The sampled species are summarized on the Table 1a and 1b. The number of the wood samples, deposited in the Tervuren Wood collection of the Royal Museum for Central Africa (Belgium) is indicated with Tw. Slides collection are also available at WSL Birmensdorf and at the Department of Plant Biology and Ecology in Antananarivo (Madagascar).

Ten *Diospyros* species on bold showed in Table 1b don't have the authors name because they are already identified by the specialist but not yet officially published (Schatz and Lowry comm. pers.).

Botanical descriptions and the use of plants are briefly given per species. Local names in English (EN), in French (FR) and in Malagasy (ML) are provided. Ecological conditions of the natural habitat as elevation, vegetation and bioclimate are extremely useful for the identification. The presence of the species on the defined previously protected areas (PA) and the new one (NPA) contributes to the protection of these precious woods. The origin of the sampled species is always mentioned on the distribution map.



Sampling sites of Malagasy *Dalbergia* and *Diospyros* species.

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Table 1a. Descriptive information on *Dalbergia* samples analysed.

1) Collector (CR: Charles Rakotovao; RAF: Rakotoarison Fenonirina; RBE: Roger Bernard; RIR: Richard Randrianaivo; RZK: Richardson Razakamalala; T: Department of Plant Biology and Ecology, University of Antananarivo); 2) Sample identification number; 3) Scientific name; asterisks indicate species not endemic to Madagascar; 4) Author name; 5) Locality of sampling; 6) Forest type from which the plant was sampled (D.: Deciduous); 7) Identification number of the specimens available at the Tervuren Xylarium, TW no.

1) Collector	2) Sample	3) Species	4) Author	5) Locality	6) Forest type	7) Tw no.
T	12	*Dalbergia baronii*	Baker	Didy	Humid	66656
RZK	7709	*Dalbergia bathiei*	R.Vig.	Tampolo	Humid	66551
CR	6513	**Dalbergia bracteolata*	Baker	Kirindy	Dry, D.	66454
RZK	7712	*Dalbergia chapelieri*	Baill.	Tampolo	Humid	66553
T	24	*Dalbergia emirnensis*	Benth.	Mt Français	Dry, D.	66657
RIR	2416	*Dalbergia greveana*	Baill.	Ankarana	Dry, D.	66477
RIR	2430	*Dalbergia lemurica*	Bosser & R.Rabev.	Oronjia/Rigny	Dry, D.	66490
RBE	2247	*Dalbergia louvelii*	R. Vig.	Betampona	Humid	55506
RAF	13	*Dalbergia madagascariensis*	Vatke	Makirovana	Humid	55566
RZK	7704	*Dalbergia maritima*	R. Vig.	Betampona	Humid	66546
RIR	2478	*Dalbergia mollis*	Bosser & R.Rabev.	Ankarafantsika	Dry, D.	66530
T	37	*Dalbergia monticola*	Bosser & R.Rabev.	Didy	Humid	66658
CR	6512	*Dalbergia neoperrieri*	Bosser & R.Rabev.	Kirindy	Dry, D.	66453
RZK	7699	*Dalbergia orientalis*	Bosser & R.Rabev.	Betampona	Humid	66541
RIR	2440	*Dalbergia peltieri*	Bosser & R.Rabev.	Oronjia/Rigny	Dry, D.	66499
T	25	*Dalbergia pervillei*	Vatke	Mt Français	Dry, D.	66659
RIR	2418	*Dalbergia purpurascens*	Baill.	Ankarana	Dry, D.	66479
RIR	2446	*Dalbergia suaresensis*	Baill.	Mt Français	Dry, D.	66505
RIR	2470	*Dalbergia trichocarpa*	Baker	Ankarafantsika	Dry, D.	66551

Table 1b. Descriptive information on *Diospyros* samples analysed.

1) Collector (CR: Charles Rakotovao; EME: Emeline; RAF: Rakotoarison Fenonirina; RBE: Roger Bernard; RIR: Richard Randrianaivo; RNA; Razafimamonjy Nivo Alisoa; RZK: Richardson Razakamalala; T: Department of Plant Biology and Ecology, University of Antananarivo); 2) Sample identification number; 3) Scientific name; asterisk indicate species not endemic to Madagascar; 4) Author name (ined.: inedited); 5) Locality of sampling; 6) Forest type from which the plant was sampled (D.: Deciduous, T.: Thicket); 7) Identification number of the specimens available at the Tervuren Xylarium, TW no.

1) Collector	2) Sample	3) Species	4) Authors	5) Locality	6) Forest type	7) Tw no.
T	33	*Diospyros aculeata*	H.Perrier	Beheloka	Dry, T.	66660
RIR	7724	*Diospyros acutiflora*	ined.	Andranotsara	Humid	66562
T	27	*Diospyros analamerensis*	H.Perrier	Mt Français	Dry, T.	66661
RIR	2405	*Diospyros ankifiensis*	H.Perrier	Ankarana	Dry, D.	66468
RAF	6	*Diospyros antongiliensis*	ined.	Makirovana	Humid	66517
RIR	2461	*Diospyros bernieriana*	(Baill.) H.Perrier	Sahafary	Dry, D.	66517
RBE	2248	*Diospyros brevipedicellata*	ined.	Betampona	Humid	66368
RBE	2264	*Diospyros calophylla*	Hiern	Analalava	Humid	66384
RIR	2403	*Diospyros chitinophora*	ined.	Ankarana	Dry, D.	66466
RZK	7714	**Diospyros ferrea*	(Willd.) Bakh.	Andranotsara	Humid	66555
RZK	7703	*Diospyros fuscovelutina*	H.Perrier	Betampona	Humid	66545
CR	6516	*Diospyros humbertiana*	H.Perrier	Kirindy	Dry, D.	66457
RNA	1	*Diospyros latispathulata*	H.Perrier	Ranobe	Dry, T.	66533
RZK	7700	*Diospyros leucocalyx*	Hiern	Betampona	Humid	66542
RBE	2240	*Diospyros maculata*	ined.	Betampona	Humid	66360
T	42	*Diospyros mangabensis*	Aug.DC.	Andasibe	Humid	66662
RBE	2239	*Diospyros maxima*	ined.	Betampona	Humid	66359
EME	21	*Diospyros microrhombus*	Hiern	Mahabo Mananivo	Humid	66408
RIR	2411	*Diospyros occlusa*	H.Perrier	Ankarana	Dry, D.	66452
RIR	2439	*Diospyros olacinoides*	ined.	Oranjia/Rigny	Dry, D.	66498
T		*Diospyros parifolia*	(Schltr.) Bakh.	Didy	Humid	66663
RIR	2452	*Diospyros perrieri*	Jum.	Mt Français	Dry, D.	66511
EME	20	*Diospyros pervilleana*	(Baill.) G.E.Schatz & Lowry	Mahabo Mananivo	Humid	66407
CR	6504	*Diospyros platycalyx*	Hiern	Kirindy	Dry, D.	66445
RZK	7710	*Diospyros quadrangularis*	ined.	Tampolo	Humid	66552
RZK	7715	*Diospyros rubripetiolata*	ined.	Andranotsara	Humid	66556
RBE	2253	*Diospyros sclerophylla*	H. Perrier	Tampolo	Humid	66373
RZK	7713	*Diospyros squamosa*	Bojer ex A.DC.	Andranotsara	Humid	66554
T	32	*Diospyros tropophylloides*	ined.	Mt Français	Dry, D.	66664
EME	4	*Diospyros velutipes*	(H. Perrier) G.E.Schatz & Lowry	Mahabo Mananivo	Humid	66394
RIR	2407	*Diospyros vescoi*	Hiern	Ankarana	Dry, D.	66470

Plant material preparation

The individual chosen in its natural habitat appeared normal and healthy. A portion of the main stem was cut near the plant's basis. Twig and branch samples were also collected from the same individual. Branch samples were collected from the first branch with the height dependent on the position of the branch and ranging from 5 to 20 m.

Plant material was stored in a sealed plastic bag to which several drops of 40 % ethanol were added. Each sample was labelled with a permanent marker pen containing the collector codes and the identification number of the sample(s).

In the laboratory, from each branch and stem sample, a 1 cm^3 sample was split from the outermost wood, including the bark. For twig samples, special care was taken to preserve the bark and the pith on the same sample. The thin sections (15 - 25 µm) have been made using disposable blades and GLS microtome (Gärtner and Scweingruber, 2013). The section were stained with Astra blue and Safranin O, dehydrated with alcohol and xylene, and mounted in Canada balsam (Chaffey 2002, Gärtner and Schweingruber, 2013). Sections with bark were initially bleached in Eau de Javel for 10 to 15 minutes before staining.

A transmission-light microscope was used to observe slides while polarized filter were applied for observing crystal presence and cell wall thickness. Sections were microscopically inspected using magnifications of 20-1000 times and photographed by using a digital camera mounted on the microscope (Olympus BX41). Magnifications are indicated in µm (0,001 mm) above a black scale bar in each picture.

For all species the anatomy of pith, primary xylem, branch xylem, twig and stem bark and stem xylem were described. Differences between branch and stem xylem were determined.