Completion report

CITES-ITTO project program

REFERENCE SAMPLE COLLECTION AND IDENTIFICATION SYSTEM FOR ALL CITES-LISTED DALBERGIA SPECIES AND A FEASIBILITY STUDY FOR DIOSPYROS AND LOOK-ALIKE SPECIES

Host Government

Switzerland

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ESTABLISHMENT OF A FULLY DOCUMENTED REFERENCE SAMPLE COLLECTION AND IDENTIFICATION SYSTEM FOR ALL CITESLISTED DALBERGIA SPECIES AND A FEASIBILITY STUDY FOR DIOSPYROS AND LOOK-ALIKE SPECIES

PROJECT NUMBER

TMT-SPD 022/15

STARTING DATE

1st of October 2015

DURATION OF THE PROJECT

13 months

PROJECT COSTS

ITTO: 152,402 USD ETH (in-kind): 20,000 CHF Own fundraising: 13,193 USD

PROJECT COMPLETION REPORT

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Executive summary

This project was motivated by the problem of on-going illegal logging of precious timber species in Madagascar and the lack of identification techniques for species and provenance identification. One of the main challenges for developing such tools is the unavailability of well-documented and reliably identified reference material for all relevant species. This limitation also precludes comparisons of existing identification techniques because all studies performed to date used different and often undocumented samples.

The main objective of this project therefore was to establish a fully documented reference sample collection of CITES-listed *Dalbergia* and a feasibility study for *Diospyros* and look-alike species of both genera that can be used for developing and validating identification techniques. Only with a well-identified reference collection will we be able to develop adequate identification systems of precious timber species and provenances and assess which technique is the most adequate to answer specific questions. We therefore made field collections in five different areas in Madagascar and gathered a total of 193 samples of which 140 belong to *Dalbergia*, 32 to *Diospyros* and 21 represent look-alike species from other genera. All samples were analysed using genetic techniques and a subset of samples was used to perform wood anatomy analysis.

The original project objective was to cover all CITES-listed *Dalbergia* and a feasibility study for *Diospyros* and look-alike species of both genera by teaming up with colleagues in Guatemala who recently established their own laboratory for wood analysis through a previous CITES-ITTO project. Unfortunately, the implementation of the Nagoya protocol in Guatemala entered into force just after the project started, which led to unforeseen administrative hurdles in obtaining research and sampling permits, which finally led to the withdrawal of our partner in Guatemala. This had severe consequences for the implementation of the project. In order to fulfil the proposed project objective we contacted botanical gardens around the globe and available DNA banks to obtain material of the missing species. While unfortunately not all missing CITES-listed species could be obtained, we were nevertheless able to execute all original objectives for species from Madagascar thanks to well-established partnerships between ETH Zurich, DBEV and MBG.

All the project outputs and activities, except for those involving our partner in Guatemala, were carried out successfully, although not all of them have yet reached completion. Delays in obtaining permits at the very beginning and in exporting the samples to the lab at ETH Zurich and the underestimation of time to get all the results together for a joint publication are mainly responsible for the fact that some activities remain to be completed.

Tangible outputs from the project include the availability of the first ever fully vouchered reference collection that is suitable for analyses using different techniques. By comparing results from different techniques we will finally have the possibility to propose an identification system for precious timber species. The newly developed and field-approved sampling protocol will be useful for future efforts to establish reference collections for timber species other than those studied during this project. The valuable involvement of our partners in Madagascar increased capacity in taxonomy and wood anatomy within the country. During this project, two students received training in wood anatomical analysis as part of their master theses work. The collection trips allowed MBG field botanists to refine their identification skills for species of *Dalbergia*, *Diospyros* and look-alike species in other genera, which is important for future projects and training of young botanists.

A key lesson we learned concerning the overall implementation of this project is that for such a time-restricted project it would be wise to include only partners that have worked together before.

From the implementation of the project we conclude that a future project needs to examine more closely the problem of look-alike species. The scope of this project was too broad, which led to a substantial slowing down of the sampling work done at each site. The list of target species should be narrowed down as much as possible to the species level and should not include entire groups at the genus level.

With the established reference sample collection, additional samples and species can now be easily included in the future to expand the available database further. As the studied genera are very species-rich, both in Madagascar and globally, we recommend continuing efforts to support scientific research in order to collect and analyse all species for the reference database.

It is equally important to continue capacity building in countries of origin to perform basic genetic analyses. Although some of the necessary equipment is very expensive and requires a high level of experience for handling and maintenance and it may therefore not be possible in the near future to do all genetic analyses in the countries of origin. We therefore strongly recommend that efforts continue to build up relationships with partners that have the capacity to do all the genetic analyses.

1 Project identification

1.1 Context

Madagascar is a world biodiversity hotspot (Myers et al. 2000) and the Malagasy forests harbour a vast diversity of precious woods (Randriamalala & Liu 2010). When sold on the international market, timber from Madagascar is highly sought after and achieves high commercial values (Barrett et al. 2010). The ever-increasing demand for precious timber on the international market has led to a massive increase of illegal exploitation of rosewood, palisander and ebony species in Madagascar in recent years (Ballet et al. 2010). Controlling the international trade in illegally logged timber from Madagascar is currently not feasible, in part because species identification and provenance assignment are not possible from logs (where morphological traits used in species identification such as flowers, bark and fruits have been removed) and reliable tools for species and provenance identification remain to be improved and validated. These limitations facilitate the illegal export of precious timber from Madagascar under names of legally traded timber species.

In 2013 ITTO funded a project in Madagascar to assess the provenance of precious Malagasy timber species and the regeneration potential in various forest types (Project coordinator: Dr. Aro Vonjy Ramarosandratana). This work, combined with a literature study, allowed us to develop a fairly good idea of the species' distribution to plan the needed field collection work for the current project.

The current project is also aligned with and supports implementation of the Madagascar Action Plan that accompanied the CITES listing of all Malagasy *Dalbergia* species in 2013, which includes among others providing identification material and tests for use in CITES enforcement when implementing the Convention for species of *Dalbergia* and *Diospyros* from Madagascar. Additionally to the CITES listing a trade ban for Malagasy *Dalbergia* and *Diospyros* is currently in place (Decree No. 2010-141) to prevent illegal logging because no identification tools are yet available.

1.2 Origin and problem

To facilitate adequate implementation of regulations for CITES listed *Dalbergia* and *Diospyros* spp., reliable identification tools need to be developed and implemented. In order to do that a reference collection with well documented and correctly identified specimens is needed before identification systems can be fully developed and validated. Reliable identification of *Dalbergia*, *Diospyros* and look-alikes is currently possible only when material exhibits key morphological traits, but may become feasible in the future with available reference collections for genetic cross checking of samples. The establishment of such a reference collection for all CITES-listed *Dalbergia* species was therefore a central element of this project, along with a feasibility study for *Diospyros* spp. and lookalikes of both genera.

2 Project objectives and implementation strategy

2.1 Objectives and adjustments

The main goal of this project was to strengthen capacities of the CITES authorities to implement the convention for all species of the genera *Dalbergia* and *Diospyros* that are listed in CITES Appendix I to III and their look-alike species through the establishment of a reference sample collection.

Therefore, the main objectives intended to complete during this project were:

- To establish a fully documented reference sample collection for all *Dalbergia* species currently listed in CITES Appendix I to III that is suitable for establishing and validating different identification systems, including DNA, isotope analyses, wood anatomy, and mass spectrometry.
- 2) To analyse collected samples using DNA analysis (Dr. Sonja Hassold) and wood anatomy (Dr. Harisoa RAVAOMANALINA) for all *Dalbergia* species currently listed in CITES Appendix I to III.
- 3) To assess based on the available experience from *Dalbergia* the feasibility of analysing the species-rich ebony genus *Diospyros* with the same methods used for *Dalbergia* to establish a similar reference database as for *Dalbergia*. *Diospyros* from Madagascar (currently listed in CITES Appendix II) were used as a case study.
- 4) To evaluate whether it will be possible to efficiently include look-alike species of *Dalbergia* and *Diospyros* into a future identification system for both genera we also included samples from look-alike species in the analysis.

Adjustments: Originally we planned to exchange samples with Guatemala but due to administrative hurdles associated with the implementation of the Nagoya protocol, that were beyond our control, this was impossible. We therefore attempted to obtain material from botanical gardens and DNA banks. Unfortunately, many of the CITES-listed species occurring in Guatemala were unavailable.

2.2 Implementation strategy and adjustments

The implementation strategy consisted of:

Field collection and preparation of reference samples

- Research permits for field collections were requested and obtained.
- A standardized sampling protocol to establish a reference sample collection that can be used with different techniques was developed and botanists from Missouri Botanical Garden (MBG) in Antananarivo were trained to perform the complex sampling.
- The success of field collection work was optimized through engagement of local guides in different areas of the country who checked the phenology of the genera of interest (*Dalbergia*, *Diospyros* and look-alikes) on a regular base for us.
- Four teams were formed for the collection and only sent to the field if local guides indicated that they had spotted fruiting and/or flowering trees of the genera of interest.
- Taxonomists of MBG identified all collected samples.
- One full set of samples has been deposited in Antananarivo, Madagascar, for storage and wood anatomical analysis and another full set has been deposited at ETH Zurich in Switzerland for storage and DNA analysis. Administrative hurdles delayed the exportation of the collected samples, which also delayed the start of the genetic analysis.
- All information associated with the collected samples was captured and is archived in the Tropicos database (www.tropicos.org).

DNA analysis of samples

- The taxonomic identification was checked again prior to performing genetic analysis.
- Based on the taxonomic identifications, a core set of samples was chosen, focusing on traderelevant timber species, to perform genetic analysis.
- Samples were analysed with two different types of markers to compare species resolution.

Standard genetic markers using chloroplast DNA were used for *Dalbergia*, *Diospyros* and look-alike species, whereas specifically developed nuclear markers were used for *Dalbergia* species only.

Wood anatomical analysis of samples

- Two students were trained under the supervision of Dr. Harisoa Ravaomanalina as part of their master thesis work to perform wood anatomical analysis on a set of samples.
- A core set of samples encompassing different species from various areas was chosen to initiate the wood anatomical analysis.
- Multiple samples per species were included in the analysis and an additional focus was placed on adding more species to validate the identification key developed by Dr. Ravaomanalina's during previous work.
- The same methods were used as for the establishment of the current wood anatomical atlas (currently in press) in order to be able to compare and add results to the existing work.
- Anatomical characters used to describe the wood in juvenile twigs and branches as well as
 mature wood from the trunk were used to develop a more comprehensive view and to
 test/confirm those characters that have been observed in earlier work.
- Anatomical descriptions of look-alike species were added to those of Dalbergia and Diospyros.

2.3 Risks and assumptions

The risks identified in the project proposal concerned being able to obtain an adequate number of fertile samples in the field and potential safety issues at the collection sites. Thanks to our carefully developed sampling strategy, we were able to manage these risks successfully and to collect enough fertile material. We additionally always hired local guides on site to reduce the risk of sampling in insecure areas.

3 Project performance

The overall performance of the project was very positive and generated international attention, especially for the collected reference material, because it is the first ever collection that comprises carefully collected and vouchered sets of samples for multiple types of analyses. Such a collection is a global milestone in establishing identification systems for species in trade. The sampling strategy and sampling protocol that were developed, tested and utilized during the project could now be easily used by other parties and transferred to other species and geographic areas in the future.

3.1 Achievement of project outputs and activities

Output 1.1: Availability of a fully documented reference collection for all CITES listed *Dalbergia* spp.

Activity 1.1.0	Prepare sampling strategy and apply for collection permit for Madagascar and
	Guatemala
Changes to activity	No application for permits was sought in Guatemala due to the withdrawal of the in-
	country partner.
Realized activity	A sampling strategy was successfully developed and documented for Madagascar in order to collect a full range of material from fertile individuals in the field. All the sampling permits were received from Madagascar National Parks (MNP) and the "Direction Générale des Eaux et Forêts (DGEF)". Sampling was carefully planned and organized by our collaborator, MBG. For Guatemala we only discussed a possible
1	sampling strategy and planned sampling.

Activity 1.1.1	Coordination of sampling strategy in Madagascar and Guatemala
Changes to activity	No coordination was possible in Guatemala due to the withdrawal of the in-country
	partner.
Realized activity	The set up of the sampling strategy was crucial. We hired local guides ("our eyes in the forest"), who we knew from previous fieldwork, to track the phenology of the genera of interest for us. When these guides informed us about the emergence of flowers and fruits, one of our four collection teams from MBG then went to the field. Using this strategy, we were able to maximise our success rate in finding fertile material and at the same time minimise costs because we only went to a specific site once.

Activity 1.1.2	Field campaign for sample collection in Madagascar and Guatemala
Changes to activity	No field campaign was possible in Guatemala due to the withdrawal of the in-
	country partner.
Realized activity	Before the teams went to the field, a training session was done to ensure that everyone understood the sampling protocol and what needs to be sampled from each individual and what information needs to be recorded. Four teams were trained to collect samples in a total of five different areas of Madagascar (Appendix A). In total, 193 samples were collected of which 140 were <i>Dalbergia</i> (24 different species), 32 <i>Diospyros</i> (10 different species) and 21 look-alikes (17 different species) in other genera.

Activity 1.1.3	Expand availability of reference samples from outside Madagascar
Changes to activity	None
Realized activity	We requested available samples from botanical gardens and the Kew DNA bank in order to increase the number of available species in the reference database. This approach is an efficient way to increase available species for genetic analysis but does not work for wood anatomy, mass spectroscopy, etc. Since to date there exists no other comprehensive collection of our genera of interest, further field collection work in countries of provenance needs to be conducted.

Activity 1.1.4	Identification of the collected samples by taxonomic specialists
Changes to activity	No identification of samples from Guatemala was possible due to the withdrawal of
	the in-country partner.
Realized activity	Taxonomists from MBG identified all the collected samples from Madagascar to the
	full extent possible. In total we collected 24 different <i>Dalbergia</i> species, 10 species
	of <i>Diospyros</i> and 17 look-alike species. One set of herbarium vouchers has been
	deposited in Madagascar and one set was sent to ETH Zurich, where it will be
	integrated into the Zurich Herbaria (Z+ZT).

Activity 1.1.5	Ask for exportation permits for the collected samples
Changes to activity	No exportation was possible from Guatemala due to the withdrawal of the in-
	country partner.
Realized activity	The management authority of CITES in Madagascar granted the requested export permit. This process took more time than expected because it was not clear to the authorities if we needed a CITES permit for the type of samples we collected. We finally exported the samples from one CITES-registered institution to another.

Activity 1.1.6	Provide samples to other institutions using mass spectrometry and near infrared
	spectroscopy (NIRS).
Changes to activity	This activity was added during the project.
Realized activity	A set of samples was provided to two different institutions using the above- mentioned methods in order to generate the first common dataset based on the application of different methods to material from the same individuals.

<u>Output 1.2:</u> Availability of a fully documented reference collection for a subset of CITES listed <u>Diospyros</u> and look-alike species from Madagascar, <u>Guatemala</u> and from as many countries with natural occurrences of these species as possible

Activity 1.2.0 Prepare sampling strategy and apply for collection permits for Madagascar and Guatemala Identical to Activity 1.1.0

Activity 1.2.1 Coordination of sampling strategy in Madagascar and Guatemala Identical to Activity 1.1.1

Activity 1.2.2 Field campaign for sample collection of *Dalbergia, Diospyros* and look-alike species in Madagascar and Guatemala

Identical to Activity 1.1.2

Activity 1.2.3 Collaboration with other countries with natural occurrence of the species to expand the reference collection

Identical to Activity 1.1.3

Activity 1.2.4 Identification of the collected samples by taxonomic specialists Identical to Activity 1.1.4

Activity 1.2.5 Ask for exportation permits for the collected samples Identical to Activity 1.1.5

Activity 1.2.6 Provide samples to other institutions using mass spectrometry and near infrared spectroscopy (NIRS)

Identical to Activity 1.1.6

Output 2.1 Availability of a molecular identification method to identify CITES-listed Dalbergia species

Activity 2.1.0	Genotype all CITES-listed Dalbergia spp.
Changes to activity	None
Realized activity	140 samples have been analysed to date using three standard chloroplast markers and 20 nuclear markers (so called microsatellites). The new samples collected during the project have been integrated into the existing database in order to verify identification and to increase the genetic reference database.

Activity 2.1.1	Develop Guidelines to conduct molecular analysis (towards an applicable method)
Changes to activity	None
Realized activity	Protocols for genetic analysis are the same as used in a previous study at ETH Zurich. Guidelines will be only available when the results from the different methods are comprehensively analysed and the levels of resolution are compared. Only then will we be able to say at what point, and for which set of questions, a given method is most appropriate.

Output 2.2 Validated wood anatomy atlas that incorporates variation among individuals of the same species

Activity 2.2.0	Complete the lab work for the wood anatomy atlas including more individuals of the same species
Changes to activity	Add new species of <i>Dalbergia</i> , <i>Diospyros</i> and look-alike species to the wood anatomy atlas, trying to include more than one individual per species in the analysis.
Realized activity	In the course of this project 23 <i>Dalbergia</i> species, 9 <i>Diospyros</i> species and 15 lookalike species were analysed using standard wood anatomy techniques.

Activity 2.2.1	Complete the new catalogue for wood anatomical properties for use by customs officers
Changes to activity	An updated wood anatomical atlas for use by customs officers.
Realized activity	An updated wood anatomical atlas with more species and more replicates per species will be provided for scientific use. Additionally, the first validated identification key will be available as guidelines for use by customs officers.

Output 2.3 Generating Scientific expertise for the identification of CITES-listed Dalbergia species

Activity 2.3.0	Verify the reliability of the reference database target species
Changes to activity	None
Realized activity	Genetic analysis: For this step it was important that the taxonomic identification of the new samples was done correctly. The identification was therefore checked twice. In a second step, the samples were all genetically analysed and fed into the database. In a third step we tested whether it is possible to assign a subset of the newly collected samples to the correct species. Our results showed that this was possible, with a high level of accuracy (up to 96%). Wood anatomy: To test the reliability it was crucial to include multiple samples of the same species in the analysis. In future the updated identification key will facilitate identifications at customs.

Activity 2.3.1	Develop guidelines to implement identification techniques					
Changes to activity	It was deemed necessary to develop different guidelines for standing trees in e.g. plantations as well as wild populations (taxonomy and genetic analysis) versus for logs being traded on the international market (wood anatomy).					
Realized activity	Genetic analysis: As we focused on establishing a reliable, carefully vouchered reference database, we have only analysed leaf samples to date to provide the best available database. In the next several months we will work on guidelines that distinguish between different cases as part of the process of bringing together all results from the different analyses.					

Output 3.1 Available case study for developing an identification method for CITES listed *Diospyros* species

Activity 3.1.0	Perform experiments to check if the same protocols used for <i>Dalbergia</i> can be used for CITES listed <i>Diospyros</i> species					
Changes to activity	None					
Realized activity	Tests were successful for lab protocols and analysis using standard chloroplast markers.					

Activity 3.1.1	Start to genotype well described and documented <i>Diospyros</i> species
Changes to activity	None
Realized activity	32 samples have been analysed to date. The three standard chloroplast markers used are working reliably, but as the genus is very species rich, they will probably not be sufficient alone to distinguish among all <i>Diospyros</i> species. Substantially more genetic work and (potentially) the development of nuclear markers is needed to establish a method for <i>Diospyros</i> species identification.

Activity 3.1.2	Modify protocols and guidelines if necessary to conduct molecular analysis					
Changes to activity	None					
Realized activity	No modification was needed for standard chloroplast markers. The same protocols as for <i>Dalbergia</i> were used. Guidelines will be developed in parallel to Activity 2.3.1. when the results are brought together for publication.					

Output 4.1 Available feasibility study for developing an identification method for look-alikes of *Dalbergia* and *Diospyros* spp.

Activity 4.1.0	Perform experiments to check if the same protocols used for <i>Dalbergia</i> can be used for look-alikes of <i>Dalbergia</i> and <i>Diospyros</i> spp.
Changes to activity	None
Realized activity	Tests were successful for lab protocols and analysis using standard chloroplast markers.

Activity 4.1.1	Start to genotype well described and documented look-alike species of <i>Dalbergia</i>					
	and Diospyros					
Changes to activity	None					
Realized activity	21 samples have been analysed to date. The three standard chloroplast markers used are working reliably distinguishing look-alike species from <i>Dalbergia</i> species because sequences are very distinct. More genetic work is needed to establish a complete dataset of look-alikes.					

Activity 4.1.2	Modify protocols and guidelines if necessary to conduct molecular analysis					
Changes to activity	None					
Realized activity	No modification was needed for standard chloroplast markers. The same protocols					
	as for <i>Dalbergia</i> were used. Guidelines will be developed in parallel to Activity 2.3.1.					
	when the results are brought together for publication.					

3.2 Comparison of activity progress against schedule

Table 1 below displays the project implementation in comparison with the detailed workplan included in the monthly highlight reports sent to ITTO.

Table 1: Activity schedule in comparison with workplan. Black thick lines indicate the planned duration of the activity and blue filled cells indicate the actual time for implementation.

OUTPUTS/	RESPONSIBLE	SCHEDULE (in months)												
ACTIVITIES	PARTY	2015-2016							40					
Output 1.1		10	11	12	1	2	3	4	5	6	7	8	9	10
Activity 1.1.0	DBEV, PEG													
Activity 1.1.0	DBEV, PEG													
-	DBEV													
Activity 1.1.2														
Activity 1.1.3	PEG DBEV													
Activity 1.1.4													Ì	
Activity 1.1.5	DBEV													
Activity 1.1.6	PEG													
Output 1.2														
Activity 1.2.0	DBEV, PEG													
Activity 1.2.1	DBEV, PEG													
Activity 1.2.2	DBEV, PEG													
Activity 1.2.3	PEG													
Activity 1.2.4	DBEV, PEG													
Activity 1.2.5	DBEV,													
Activity 1.2.6	DBEV, PEG													
Output 2.1														
Activity 2.1.0	PEG													
Activity 2.1.1	PEG													
Output 2.2														
Activity 2.2.0	DBEV													
Activity 2.2.1	DBEV													
Output 2.3														
Activity 2.3.0	DBEV, PEG													
Activity 2.3.1	DBEV, PEG													
Output 3.1	DBEV, PEG													
Activity 3.1.0	DBEV, PEG													
Activity 3.1.1	PEG													
Activity 3.1.2	PEG													
Output 4.1														
Activity 4.1.0	DBEV, PEG													
Activity 4.1.1	PEG													_
Activity 4.1.2	DBEV, PEG													

3.3 Project expenditure against funds input

Total project expenditure to date by the executing agency: 186,531.5 CHF =185,595.7 USD; 14.3.2017

- Funds received from ITTO: 191,000 USD
- Funds provided by ETH Zurich (in-kind): 20,000 CHF = 19,826 USD; 14.3.2017
- Funds acquired during the project: 13,193 USD
- Funds received in total (ITTO + funds acquired): 204,193 USD
- Remaining funds (14.3.2017): 36,726.1 CHF = 36,407.1 USD

A detailed financial report can be found in Appendix B.

4 Project outcome, target beneficiaries involvement

4.1 The achievement of specific objectives

The objective of establishing a reference sample collection was fulfilled successfully with the newly developed sampling strategy. All collected samples of *Dalbergia*, *Diospyros* and look-alikes were analysed genetically and a subset was analysed using wood anatomy techniques. A good base for future projects has thus been established, but the work is not yet finished. Because the genera we are dealing with are species-rich and have wide distribution ranges, we are still missing most of species existing worldwide, in the reference database.

4.2 Situation at activity completion

- Availability of a reference sample collection for *Dalbergia* and a feasibility study for *Diospyros* and look-alike species of both genera.
- Results of different identification techniques available for a common set of samples. Results
 can be used to develop an identification strategy based on different methods to provide a
 multi-level solution.
- The results will be published in a peer-reviewed journal, which will also be important to start discussing results with other groups working on timber identification issues.
- Needs assessment report conducted by WRI, with input from project participants, which
 carefully spelled out the needed next steps in order to achieve the goal of timber
 identification in Madagascar.

4.3 Participation of the target beneficiaries

- Training of sampling teams applying the newly developed sampling protocol in the field.
- Enhanced taxonomic identification success through discussions with scientists and field botanists comparing results of genetic analysis with results from taxonomic identification.
- Participation of MBG and DBEV as collaborators in the project.
- Training of students in the wood anatomical lab of Dr. Harisoa Ravaomanalina. The students were able to complete their master thesis work using wood anatomy techniques.
- Discussions with key stakeholders in Madagascar on potential implementation strategies.
- On-going collaboration with various institutions working on the issue of identifying *Dalbergia* and *Diospyros* species.

4.4 Expectation of activity sustainability after activity completion

• Collaborations have been established with various institutions, which will promote communication and planning of future projects. The project played a key role in catalysing the communication and collaboration among Malagasy and international experts. It also helped to form a shared understanding and a vision of an integrated, multi-disciplinary approach to address the need for a reliable and accurate set of identification tools.

- The established reference sample collection will help to shape and will contribute to future projects in the course of sampling and implementation strategies.
- The project provides the basis for discussion about a possible implementation strategy to identify the species in trade. The available sample collection allows us to analyse the samples with different identification techniques. The results can then be compared and a strategy can be developed for the use of different techniques.

5 Assessment and analysis

- i) In Madagascar, the right partners were involved to ensure the smooth implementation of the project. The partner in Guatemala was also, in principle, the right one, but it turned out that that implementation of the Nagoya protocol, which came up at the same time as this project started, changed the situation substantially. The newly arising questions could not be solved in time, in part presumably because we had no yet had the time to establish personal relationships and to build trust with partners in Guatemala.
- ii) The project focused on appropriate objectives, although they could have been phrased more concisely in order to decrease repetition of activities for the different genera. Potential problems were correctly identified and were avoided thanks to a careful planning of the fieldwork.
- iii) The project partner in Guatemala withdrew due to administrative hurdles in obtaining permits to export plant material to be analysed at ETH Zurich. Future projects should officially start only once all required sampling and exportation permits have been obtained for all involved parties. At the moment it seems unlikely that any plant material can be exported from Guatemala for external analysis.
- iv) The full and successful implementation of such a project requires a running time of at least 1.5 to 2 years. Financial resources to cover the salaries of project participants in all participating countries are essential. Funding should be also available to invest in capital items (such as smaller lab machines) in order to build capacity in range states only.
- v) The main external influence that led to problems in this project was the implementation of the Nagoya protocol. This problem could not be overcome within the narrow time-frame of this project.
- vi) Partners benefited from participation in this project and were able to strengthen their skills. The beneficiaries profited from training in the lab and the field.
- vii) During this project we successfully strengthened collaboration and partnerships that will benefit future projects. The established and field-tested sampling protocol will greatly benefit future efforts to continue the collection of reference samples.
- viii) Roles and responsibilities were clarified at the beginning of the project. We have the feeling that all partners understood them and found them appropriate.

6 Lessons learned

Sampling procedure

A standardized sampling protocol is essential to ensure consistent sampling. Equally important is to provide careful training for the collecting teams and continued supervision to ensure that everybody involved understands exactly what needs to be sampled and how. A small hands-on training session before the field collection is recommended also for future projects. Further, a well-defined strategy how to select look-alike species is needed in order to identify those actually in trade rather than focusing on an entire look-alike genus.

Availability of sampling consumables

It is difficult to procure basic consumables in Madagascar. Silica gel and collecting bags were therefore brought to Madagascar by the executing agency. In order to avoid being dependent on visiting researchers to hand-carry such consumables, new ways of receiving this material need to be identified.

DNA analysis

The same protocols for DNA extraction can be used for *Dalbergia*, *Diospyros* and look-alikes. Species-specific markers are available for *Dalbergia* but remain to be developed for *Diospyros* and look-alike species.

Wood anatomy

It is very difficult to find all the needed lab consumables for wood anatomical analysis in Madagascar. Agents to stain histological sections such as Safranin O, Astra Blue and Alcian Blue, or fixation agents such as Euparal and Canada Balm need up to three months for delivery and are extremely expensive because of excessive import taxes. This fact delayed the start and hindered the performance of the lab analyses. In the future, thought must be given to an improved import strategy to reduce costs of basic consumables. One option may be to team up with hospitals or private labs to import consumables together and thereby obtain a better price.

Additionally, the wood anatomy lab does not yet have an appropriate place to store wood specimens. The space currently available is too humid and the samples are therefore vulnerable to fungi that could destroy the entire collection. In the future it will be important to build and establish a suitable place to store the valuable wood collection, which will contribute to building capacity in Madagascar.

Authorisation of research and sample collection

We encountered some difficulty obtaining research and sampling permits because the species of interest most frequently occur within protected areas managed by Madagascar National Parks. The issuance of permits took a long time, which resulted in delays in the fieldwork, thereby making it impossible to collect some species when they were thought to flower and fruit. For future projects, permits need to be available before the project starts and should include all possible field participants and all possible sampling sites. Obtaining multiple permits, as was done in the present project, should likewise be avoided as it caused quite some confusion in the field.

Collaboration

Collaborative arrangements need to be established ahead of project start. Ideally, partners should meet face to face before preparing a proposal to ensure that the needs and ideas of all parties are understood. In general, projects involving several international partners should have a longer running phase than just one year, especially if field and lab work are planned. These tasks require significant time and effort to secure research permits and to take care of all other administrative arrangements before the work can be started.

Sample logistic

Exporting samples of CITES-listed species is difficult even in established projects and under partnerships because local authorities fear making mistakes, especially with highly visible and sensitive species such as rosewoods. This can delay the process of securing export permits, which then delays downstream activities. A better and faster system will be required in the future. One possibility might be to obtain a letter of agreement from the government to conduct the proposed research and to carry out routine testing of samples abroad until local capacity is developed.

Capacity building

In order to build capacity, appropriate facilities must be available. Future projects should allow for investment in capital items such as building suitable space for labs and storage of the collection. Also, training workshops should take place at the beginning and end of the project to conclude what was learnt. We recommend implementing such an approach in future projects that involve any kind of capacity building.

7 Conclusions and recommendations

Conclusions:

- The overall project objectives were successfully achieved, although one partner dropped out and a delay of exportation permits delayed the start of some activities.
- The agreed sampling strategy was key in order to be able to collect the reference samples with flowers and/or fruits. We recommend using this strategy in future projects too.
- The development of a standardized sampling protocol that serves several techniques was a key objective in the project. This sampling protocol will be very useful for other traded timber species to set up a reliable reference database.
- It is very valuable to have the capacity of identification (MBG) and wood anatomy analysis in the country itself.

Recommendations:

- We recommend to rethink the CITES permit issuing process for scientific samples. It should also made clear for CITES-listed species whether CITES permits are required if the species is listed, but the type of samples that should be exported for analysis is not covered in the annotation of the CITES Appendices (e.g. *Dalbergia* species are listed, but the annotations do not cover leaf material. Is a CITES permit needed to export leaf material?).
- We also recommend thinking about a standard contract to comply with the Nagoya protocol.
 The availability of such a document could facilitate collaboration with range states that are reluctant to allow export of any biological sample.
- The genera of interest are very species-rich compared to other systems, which makes it
 extremely difficult to obtain suitable samples for all species. Further collection trips and
 additional scientific work is therefore needed.
- We recommend that a future project should examine more closely the problem of look-alike species. The scope of this project was too broad, which led to a substantial slowing down of the sampling work done at each site. The list of target species should be narrowed down as much as possible to the species level and should not include entire genera.
- Investment in capital items (laboratory items, lab space, etc.) to get the wood anatomy lab
 fully functioning will be important in the near future to strengthen capacity within the
 country.

We also recommend establishing a basic genetic lab in Madagascar that can handle standard
molecular analyses, including DNA extraction, PCR amplification and genotyping or
sequencing. Most of the required equipment was already donated, but actual lab space is not
available.

Responsible for the Report

Name: Dr. Sonja Hassold

Position held: Project coordinator updated: 13th of March 2017

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- Randriamalala HF, Liu Z (2010) Rosewood of Madagascar: Between democracy and conservation. *Madagascar Conservation & Development*, **5**, 11–22.

Appendix A Sampling locations in 2016

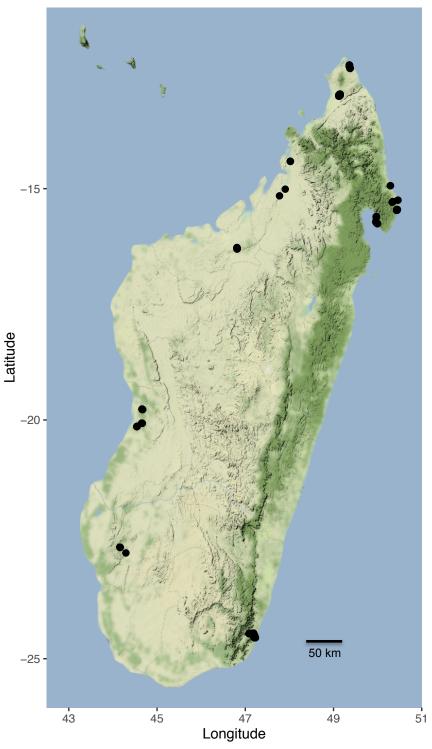


Figure 1: Black dots on the Madagascar map indicate different sampling locations in 2016. In total 193 were collected of which 140 were Dalbergia (24 different species), 32 Diospyros (10 different species) and 21 look-alikes (17 different species) in other genera.

Appendix B1 Project financial Statement March 2017

Project No. TMT-SPD 022/15 Period ending on: October 2016

<u>Project Title:</u> Establishment of a fully documented reference sample collection and identification system for all CITES-listed Dalbergia species and a feasibility study for Diospyros and look-alike species

Our accounts were handled in CHF but the budget was set up in USD. Therefore, expenditure and available are in CHF

are in	CHF.				
		Original	Updated	Expenditure To- date	Available
		Amount (A) USD	Amount (B) USD	Expended total (C) CHF	Funds (D) {B – C} CHF
10	Project Personnel				
	11. National Experts				
	12. National Consultants	32,600	32,600	16,927.2	15,672.8
	13. Other labor				C
	14. Fellowships and Training				C
	15. International Experts				C
	16. International Consultants	77,000	84,360	99,655.4	-15,295.4
	19. Component Total	109,600	116,960	116,582.7	377.3
20	Sub-contracts				0
	21. Sub-contract (with A)				C
	22. Sub-contract (with B)				C
	29. Component Total				C
30	Duty Travel				C
	31. Daily Subsistence Allowance	3,000	4,686	1851.5	2,834.7
	32. International Travel	5,000	9,103	8,374.7	728.5
	33. Transport Costs	2,000	2,043	194.5	1,849.1
					C
	39. Component Total	10,000	15,833	10,420.7	5,412.3
40	Capital Items				0
	41. Premises	18,000	18,000	18,000	18,000

	42. Land				0
	43. Vehicles				0
	44. Capital Equipment	3,000	3,000	1,551.8	1,448.2
					0
	49. Component Total	21,000		19,551.8	1,448.2
50	Consumable Items				0
	51. Raw materials (Lab materials)	60,000	60,000	22,576.1	37,423.9
	52. Spares				0
	53. Fuel and Utilities	2,800	2,800		2,800
	54. Office Supplies	5,600	5,600	3,883.2	1,716.8
					0
	59. Component Total	68,400	68,400	26,459.4	41,940.6
60	Miscellaneous				0
	61. Sundry				0
	62. Auditing	2,000	2,000		2,000
	63. Monitoring (kept at ITTO)	3,000	0		0
	64. Contingencies (kept at ITTO)	6,000	0		0
	69. Component Total	11,000	2,000		2,000
70	ETH infrastructure fees			13,516.9	-13,516.9
					0
	79. Component Total			13,516.9	-13,516.9
100	GRAND TOTAL	220,000	224,193	186,531.5	37,661.5
	Available at ETH in CHF Available at ETH in USD (14.3.2017)				36,726.1 36,407.13
	The difference (37,661.5-36,726.1= but our accounts were handled in C 203,257.62 CHF which results in a c	CHF. In total,	we received 204	_	

Differences in the budget:

- Project personnel: When I calculated the original budget I assigned 2 months of my work in Madagascar as national consultant which is obviously not correct. In total, costs in this category are not exceeded
- Consumable items: Lab analyses are not yet finished but additionally we were not able to collect as many samples as was budgeted originally.
- ETH infrastructure fees: Due on all incoming funds (10%) but we could negotiate with them that the project money that is budgeted for our collaborators are free of these infrastructure fees.

What is not yet accounted for:

- Printing and mailing expenses
- Final external audit

Appendix B2 Project cash flow March 2017

Proje	ct No. TMT-SPD 022/15	F	Period ending on: (October 2016		
CITES Our a	ct Title: Establishment of a fully docu- listed Dalbergia species and a feasibine counts were handled in CHF. Thereferred to USD during the transfer 2017; 09:38am)	lity study for Diosp r efore, many of t	yros and look-alik he budget numb	e species ers (except th	ne ones that were	
	onent	Reference	Date	Amount in USD	Local Currency (CHF)	
A. Fui	nds received from ITTO					
	1. First installment	RW01 1st payment	2.11.2015	100,000	99,970	
	2. Second installment	RW01 2 nd installment	19.5.2016	91,000	90,255.6	
	Total funds received			191,000	190,225.6	
	3. In-kind contribution ETH: lab and office space	ETH in-kind contribution	1.11.2015	20,000	20,000	
	4. Consultant work for WRI	W.R.I.	28.6.2016	11,190.5	11,017.1	
	5. Conference of WRI	W.R.I.	14.7.2016	2,002.5	1,999.2	
	6. Bank interest	ZINS	31.12.2015		15.8	
	Component total			224,193	223,257.6	
	Component difference			224,193 (budget USD) - 223,257.6 (funds received in CHF) = 935.4		
В. Ехр	penditures by executing agency					
10	Project Personnel					
	11. National Experts					
	12. National Consultants	1 st installment MBG	2.5.2016	8,742.7	8,463	
		2 nd installment MBG	28.9.2016	8,640	8,464.3	
	13. Other labor			0.00		
	14. Fellowships and Training			0.00		
	15. International Experts			0.00		
	16. International Consultants in total	14xxxxxx	monthly	98,740.9	99,655.4	

	19. Component Total			116,123.6	116,582.7
20	Sub-contracts				
	21. Sub-contract (with A)				
	22. Sub-contract (with B)				
	29. Component Total				
30	-				
	31. Daily Subsistence Allowance	See journal	Project period	1,834.5	1,851.5
	32. International Travel		Project period	8,297.9	
		See journal			8,374.7
	33. Transport Costs		Project period	192.7	194.5
	39. Component Total			10,325.1	10,420.7
40	Capital Items				
	41. Premises	ETH in-kind	1.11.2015	18,000	18,000
	42. Land				
	43. Vehicles				
	44. Capital Equipment	See journal	See journal	1,537.6	1,551.8
	49. Component Total			19,537.6	19,551.8
50	Consumable Items				
	51. Raw materials (Lab materials)	Lab analysis	See journal	22,368.9	22,576.1
	52. Spares			0.00	
	53. Fuel and Utilities			0.00	
	54. Office Supplies	See journal	See journal	3,847.6	3,883.2
	59. Component Total			26,216.5	26,459.4
60	Miscellaneous			,	·
	61. Sundry				
	62. Auditing				
	63. Monitoring				
	64. Contingencies				
	69. Component Total				
70	ETH infrastructure fees	See journal	See journal	13,392.9	13,516.9
	79. Component Total			13,392.9	13,516.9
	•			, -	,
100	GRAND TOTAL			185,595.7	186,531.5
	101. Available at ETH			36,407.1	36,726.1