PROJECT PROPOSAL TO THE
INTERNATIONAL TROPICAL TIMBER ORGANISATION (ITTO)
Submitted by Ministry of Forestry and Wildlife of Cameroon

Optimising DNA verification tests in supply chains of *Prunus africana* and *Pericopsis elata* in Cameroon

SUMMARY

Genetic markers will be used to determine the optimal supply chain verification sampling location and intensity for African Cherry, *Prunus africana* (i.e. annual harvesting plot Prunus allocation unit or PAUs), and also to support/increase the confidence of the existing CITES licensing system for *Prunus africana* and African teak, *Pericopsis elata*.

This project seeks to build on previous projects: Pilot Implementation of a DNA traceability system for *Prunus africana* in Prunus Allocation Units in Cameroon and the Democratic Republic of Congo (DRC) (Project PP-A/39-162A); and Pilot Implementation of a DNA traceability system for *Pericopsis elata* in forest concessions and sawmills in Cameroon and Congo. The project will either develop new single nucleotide polymorphism (SNP) markers (for *P. Africana*) or utilize previously developed SNP markers for *P. elata* and the newly developed genetic markers for *P. africana*. These markers are easier to integrate into new laboratories than microsatellite markers and, as a result, can be performed in laboratories in Africa in as well as in Australia.

This activity supports the ITTO-CITES project output of a cost-effective regulatory system for the trade in CITES listed tree species. The main outputs are: (1) Determine the optimal supply chain verification sampling location and intensity (2) Implementation of DNA verification in supply chains at optimal location and intensity in Cameroon.

EXECUTING/IMPLEMENTING AGENCY	Ministry of Forestry and Wildlife (MINFOF)/Department of Forests/CITES MA
COLLABORATING AGENCIES	University of Adelaide, Australia Double Helix Tracking Technologies, Singapore Faculty of Sciences, University of Douala (UD), Cameroon Syndicates of Industries in charge of Harvesting, Processing, and Exportation of Special Products (SIHPESP), Cameroon Association of Timber and Forest Industries (ATFI), Cameroon
DURATION	24 months
START DATE	February 1st, 2021

BUDGET AND PROPOSED SOURCE OF FINANCE

(a)	ITTO Contribution (in US$)	**US$ 264,600**
(b)	MINFOF/UD (in-kind contribution for sampling)	US$ 25,000

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PART I: CONTEXT

Origin/Background

This project focusses on two of Africa's timber trees that are at risk of illegal logging, African cherry, *Prunus africana*, and African teak, *Pericopsis elata*.

Prunus africana occurs naturally in Afromontane forest habit and is geographically widespread (Stewart, 2003). Due to the popularity of *P. africana* bark, for its medicinal properties, and timber for the manufacture of axe and hoe handles (Stewart, 2003), *P. africana* is listed as vulnerable on IUCN Red List of Threatened species (IUCN 2020).

Pericopsis elata occurs naturally in Côte d'Ivoire, Ghana, Nigeria, Cameroon, Republic of the Congo and The Democratic Republic of the Congo (DRC) (Martial et al., 2019; Bourland et al., 2012), and is listed as endangered on the IUCN Red List of Threatened Species (IUCN 2020). The highly valuable timber from *P. elata* is used for boat building, joinery and flooring, making this species vulnerable to illegal logging.

Paper-based methods of supply chain certification leave room for fraudulent activity. False documentation can be used to hide the true species and/or origin of timber (Lowe et al., 2016). Developing DNA based methods of identifying timber species and origin is paving the way to a more reliable ways of proving species/origin claims (Lowe and Cross, 2011).

Pilot studies to develop genetic markers for the verification of timber origin for *P. elata* and *P.africana* trees have shown that bark/sawn timber can successfully be matched to source individuals with a high degree of statistical confidence. However, practical applications of these tests have shown that the chain of custody of bark/sawn timber is not completely intact. Samples taken at different points in the supply chain were shown to have been sourced from a mixed origin, when a single origin was claimed. However, in many cases the correct source could be determined retrospectively from the pool of samples, indicating that the observed inconsistencies were unlikely to be the result of illegal activity, but rather demonstrate the difficulty with strict chain of custody application at the individual tree level. Hence, relying on individual matching is potentially a limitation of implementing genetic markers in this manner.

The pilot studies also showed evidence of spatial genetic structure in *P. africana* and although not explicitly tested for *P. elata*, life history traits strongly indicate this would also be the case. Therefore, using genetic markers to identify the population of origin may be a more feasible method of enforcing compliance in *P. elata* and *P.africana* supply chains, as it does not depend on individual tree level supply chain control.

Application of any scientific methods for timber identification to supply chain monitoring and compliance, must consider not only the technicalities of the tests in question, but also the context in which they are to be applied. The tests must be feasible technically, logistically, and provide information that is useful in supporting legal and regulatory compliance.

Building on the success of the previous project to develop microsatellite markers in *P. africana*, we propose an extension that will develop single nucleotide polymorphism (SNP) markers for the same species. This project will assess the utility of SNP markers for determining timber/bark origins of *P. elata* and *P. africana* at various population

levels (i.e., annual harvesting plot, Prunus allocation unit or PAU, five-year forest bloc). Further, we will assess feasibility with respect to local context including sampling logistics, specific legal/regulatory requirements, and implementation capacity.

Cameroon is a producer country member of the International Tropical Timber Organization (ITTO) and a party to the Convention on International Trade on endangered species of Fauna and Flora (CITES). Cameroon has already been supported by the two organizations to sustain two endangered species including *P. elata* and *P. africana*, in the frame of the ITTO-CITES Programme. A total of nine (9) projects were funded by the ITTO-CITES Programme in Cameroon. Those projects assisted Cameroon in developing management plans and non-detriment findings (NDF) reports for these species, as well as a pilot study of the applicability of DNA traceability to these species.

This project is aligned with the Cameroonian national strategy for SFM, growth and employment. The project is consistent with Cameroon's forest law, to the elements related to improve forest governance to be precise. The project is also in accordance with the 2020-2023 Priority Action Plan of the Cameroon Ministry of Forestry and Wildlife/Programme Management and renewal of the forest resource/Action 2: legal provision of markets/Activity 2: to ensure the traceability of the CITES listed tree species.

The Faculty of Sciences of the University of Douala has always assisted Cameroon CITES authorities in research with view to develop management plans and Non-Detriment Findings (NDF) for threatened tree species listed in the CITES appendix II, including *P. africana* and *P. elata*. Research team guided by Prof Jean Lagarde Betti, the Regional coordinator (RC) for Africa of the ITTO-CITES Programme from 2008-2015 and the current RC for Africa of the ongoing CITES Tree Species Programme (CTSP) have published many works related to the biology, ecology and management of the two tree species. With this, the Faculty of Sciences of the University of Douala has skills to coordinate at the local level this project..

PART II: THE PROJECT

1. Project Objective

Implement an effective DNA traceability system to control trade in *Prunus africana* and *Pericopsis elata.* Routine application of this approach will enable random verification of traceability documentation to detect log substitution and fraud to a high level of confidence.

2. Justification

2.1 Problems to be addressed

The pilot study under the ITTO-CITES Programme showed that *P. africana* bark could successfully be matched to the source individual with a high degree of statistical confidence, and that *P. elata* timber could successfully be matched to the source individuals. However, practical applications of these tests have shown that there are some errors in the chain of custody of bark and timber. Samples were often shown to be from mixed origin when a single origin was claimed. However, in many cases the correct source could be determined retrospectively from the pool of samples. This indicated that tracing bark/timber to a population rather than an individual would prove to be a more useful strategy for ensuring that the chain of custody remains unbroken. This project will

determine the optimal sampling location and intensity (i.e., annual harvesting plot, Prunus allocation unit or PAU, five-year forest bloc) for such a strategy.

SNP markers have already been developed for *P. elata.* SNP marker development for *P. africana* will identify new markers that are easier to integrate into new laboratories than microsatellite markers and, as a result, can be performed in laboratories in Africa in as well as in Australia. Additionally, to analyse SNPs, much shorter DNA products are amplified compared with microsatellites. Therefore, SNP-based genotyping can be more reliably used when dealing with the degraded DNA extracted from bark.

2.2 Intended situation after project completion

At the completion of this project, SNP markers will be developed for *P. africana* and the optimal sampling location and intensity for implementing these markers and the previously designed markers for *P. elata* will be identified. The proposed DNA traceability systems for *P. africana* and *P. elata* can then be put into effect to secure controlled supply chains. Scientific verification of existing chain-of-custody documentation will identify and deter attempts to substitute or mis-declare bark from non-authorised areas in order to circumvent controls or exceed quotas.

2.3 Target beneficiaries

The target beneficiaries of this activity include:

i) The Government of Cameroon that will gain international recognition for prudent implementation of CITES.
ii) Industry (Syndicates of Industries in charge of Harvesting, Processing, and Exportation of Special Products (SIHPESP)) in Cameroon will benefit from increased confidence in sustainable supply of *Prunus africana* and a stable regulatory framework.
iii) The Association of Timber and Forest Industries (ATFI) will benefit from increased confidence in sustainable supply of *P. elata* and a stable regulatory framework.
iv) The scientific institutes concerned in Cameroon will have reliable and sound data on the DNA of *Prunus africana* and *P. elata*. Cameroon highland forest communities will gain international recognition and credibility for sound supply chain management.
v) Scientists, conservationists and non-governmental organizations (NGOs) interested in the sustainable management of *P. africana* and *P. elata*.

The information from this project will be disseminated through various types of publications, workshops/seminars and communication media.

2.4 Risks

Field work is not feasible in the North West and South West regions of Cameroon at this moment due to security risks. This risk can be mitigated by avoiding activities in these regions and instead sampling in the Adamawa, Centre and Littoral regions.

International travel has become a risk due to COVID-19. This risk will be mitigated by training being performed remotely, workshops being conducted online and local personnel in Cameroon being used as much as possible.

3. Outputs

Objective 1: Implement an effective DNA traceability system to control trade in Prunus africana.

- Output 1.1: Development of SNP markers for *Prunus africana* suitable for DNA fingerprinting of bark (differentiation of bark between individual trees of the same species).
- Output 1.2: Determine the optimal supply chain verification sampling location and intensity for *P. africana* and *P. elata*.
- Output 1.3: Implementation of DNA verification in supply chains at for *P. africana* and *P. elata at* optimal location and intensity (as determined in Output 1.1) in Cameroon.
- Output 1.4: Project report and draft publication

4. Activities

4.1 Output 1.1

- *Activity 1.1.1 Population sampling*
  - Cameroon was selected as the country of focus for this project because of the resources we already have there. This is of particular importance in the risk management for this project due to travel restrictions as a consequence of COVID-19

    For *P. africana,* five hundred (500) samples will be collected from 5 different Prunus Allocation Units (PAUs), that are managed on five-year half rotations (ten-year rotations), within Cameroon. Twenty (20) samples will be collected from each of the five annual harvesting/debarking plots within each PAU (100 samples per PAU). This sampling strategy will allow us to determine whether it is possible to differentiate between trees from different PAUs or, at a finer level, represented by the individual annual harvesting/debarking plot. The PAUs will be selected in the Adamawa (Adamawa 1, 3, 5), Centre (Mount Banda Banda) and Littoral (Mount Kupe) regions.

    For *P. elata,* five hundred (500) samples will be collected from 5 different five-year forest blocs (logging units) within Cameroon. Twenty (20) samples will be collected from each of the 5 annual cutting plots within each five-year bloc (100 samples per five-year forest bloc). This sampling strategy will allow us to determine whether it is possible to differentiate between trees from different five-year forest blocs or, at a finer level, represented by the individual annual cutting plots. Samples will be collected in the Forest management Unit (FMU) number 10021, located in the Boumba and Ngoko division, East region of Cameroon. The five-year forest blocs will be selected after discussing with the forest concessionaire

- *Activity 1.1.2 DNA extraction and genetic marker development*
  - The *P. africana* samples from the above populations will be processed in the laboratory to identify 'polymorphic' genetic markers. These markers will be able to further explore the spatial genetic structure of *P. africana* by identifying population specific differences which can be used when verifying the geographical origin of timber/bark.

4.2 Output 1.2

- *Activity 1.2.1 Analysis with genetic markers*
  - The newly developed genetic markers for *P. africana* will be used to determine whether it is possible to differentiate between PAUs and individual annual harvesting/debarking plot. The previously developed genetic markers will be used to determine whether it is possible to differentiate between five-year forest blocs and individual annual cutting plots. This will be done in the laboratory facilities at The Advanced DNA, Identification and Forensic Facility (ADIFF) at the University of Adelaide.

4.3 Output 1.3

- *Activity 1.3.1 Workshops for stakeholder consultation and field training*

  An opening workshop is recommended to conduct training of staff in wood sampling techniques. At the completion of this project, a training workshop will be given to strengthen CITES trade compliance for *P. africana* and *Pericopsis elata*

- *Activity 1.3.2 Preparation of training "tool-box"*

  Preparation of a "tool-box" consisting of a technical training guide and other materials will be prepared to assist in integrating the findings of this project into practice

4.4 Output 1.4

- *Activity 1.4.1 Preparation of final report and draft publication*

  At the completion of the proposed work, a report detailing the findings of the DNA analysis will be provided. Additionally, a draft will be written up for publication in a scientific journal

5. Work Plan

Outputs/Activities	Responsible party	Month																							
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24
Output 1.1																									
1.1.1 Population sampling	MINFOF / UD																								
1.1.2 DNA extraction and genetic marker development for *P. africana*	The University of Adelaide																								
Output 1.2																									
1.2.1 Analysis with genetic markers (*P. africana* and *P. elata*)	The University of Adelaide																								
Output 1.3																									
1.3.1 Workshops for stakeholder consultation and field training	DoubleHelix / MINFOF / UD																								
1.3.2 Preparation of training “tool-box”	DoubleHelix / UD																								
Output 1.4																									
1.4.1 Preparation of final report and draft publication	The Universty of Adelaide / DoubleHelix/UD																								

6. Budget

6.1 Total Project Budget by Activity

The detailed project budget by specific activity (or by inputs necessary to conduct specific each activity) is shown in Table 1.

Table 1: ITTO budget by inputs for conducting each activity (US$)

N°	ITEM	TOTAL (US$) TO:		
		MINFOF	U. OF ADELAIDE	TOTAL
10.	**Project Personnel**			
	11. International experts (University of Adelaide as the Lead Consultant and DoubleHelix as the Project Manager) For breakdown see Annex A		169 100	169 100
	12. National Technical Committee	4 800		4 800
	13. National coordinator (Ministry of Forestry and Wildlife as the Executing Agency-EA)	5 400		5 400
	14 National Technical Assistance (Faculty of Sciences of the University of Douala, Cameroon, as the scientific and technical advisor of the EA)	10 800		10 800
	15. Labour (technicians, workers recruited) (Cameroon)	13 000		13 000
	16. Students (Cameroon)	9 000		9 000
	18. Component Total	**43 000**	**169 100**	**212 100**
20.	**Sub-contracts**	**-**		**-**
30.	**Duty Travel**			
	31. Daily Subsistence Allowance (National Technical Committee, missions and field trips) (Cameroon)	15 000		15 000
	32. Transport Costs (Car hire and driver) (Faculty of Sciences of the University of Douala, Cameroon)	5 000		5 000
	35. Component Total	**20 000**		**20 000**
40.	**Capital Items**			
	41. Offices	Gov		Gov
	42.Capital equipment (laptop, telephone, GPS, printer) (Faculty of Sciences of the University of Douala, Cameroon)	4 000		4 000
	44. Component Total	**4 000**		**4 000**
50.	**Consumable Items**			
	51. Raw materials software and shape files (Faculty of Sciences of the University of Douala, Cameroon)	-		-
	52. Fuel and utilities (National Herbarium of Cameroon)	3 000		3 000
	53. Office supplies (ink, papers, block notes, air-time) (Faculty of Sciences of the University of Douala, Cameroon)	2 000		2 000
	57. Component Total	**5 000**		**5 000**
60.	**Miscellaneous**			
	61. Communication (phone and internet) (Faculty of Sciences of the University of Douala, Cameroon)	1 500		1 500
	62. workshops and meetings (Cameroon)	15 000		15 000
	65. Component Total	**16 500**		**16 500**
70.	**Audit Costs (to be retained by ITTO)**	**7 000**		**7 000**
100.	**GRAND TOTAL**	**95 500**	**169 100**	**264 600**

6.2 Project Budget by Source

The project budget by source is as summarized in Table 2.

Table 2: Activity budget by source (US$)

Item	ITTO	MINFOF/UD*	Total (US$)
10. Activity Personnel	212 100		**212100**
20. Sub-contracts			
30. Duty Travel	20 000		**20000**
40. Capital Items	4 000		**4000**
50. Consumable Items	5 000		**5000**
60. Miscellaneous	16 500		**16500**
70. Audit Costs	5 000		**7 000**
Total	**262 600**		**264 600**

*Note: MINFOF/UD will provide equivalent of $25,000 in personnel time (in-kind) to help offset costs of personnel involved in sampling activities.

6.3 Details on Personnel budget in Cameroon.

Table 3 presents the consolidated personnel budget for the proposed work in Cameroon.

Table 3: Personnel budget details

ITEMS	UNIT PRICE	NUMBER UNITS	DURATION (months)	TOTAL ($ US)
National Technical Committee				
8 members (DSA)	300	8	2	4 800
Component total				**4 800**
Permanent team				
National coordinator (MINFOF)	450	1	12	5 400
Assistant national coordinator (UD)	450	1	24	10 800
Secretary-accountant (UD)	200	1	12	2 400
Component total				**18 600**
Labour Cost				
Forest technicians	250	2	10	5 000
Workers	200	4	10	8 000
Students	200	3	15	9 000
Component total				**22 000**
Workshops and meetings				
Workshops and meetings	10 000	1	1,5	15 000
Component total				**15 000**
Missions and field trips				
Missions and field trips	5 000	1	3	15 000
Component total				**15 000**

PART III: OPERATIONAL ARRANGEMENTS

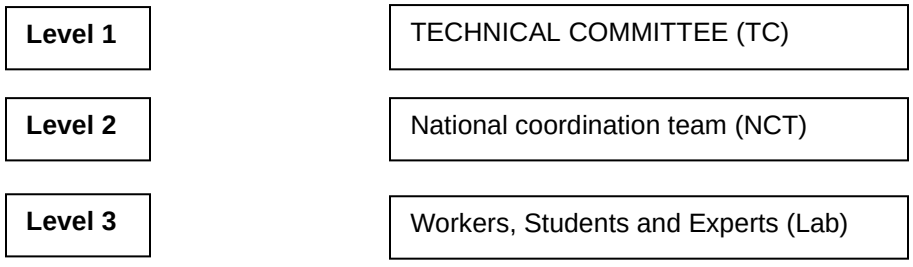
Management Structure

The Project will be implemented by the Ministry of Forestry and Wildlife, Directorate of Forestry, the CITES Management Authority (MA) as the Executing Agency (EA), with the Scientific and Technical Assistance of the Faculty of Sciences of the University of Douala, Cameroon, in collaboration with the University of Adelaide, Australia, DoubleHelix Tracking Technologies, Singapoore and the Syndicates of Industries in charge of Harvesting, Processing, and Exportation of Special Products (SIHPESP). At the National Level, the Faculty of Sciences of the University of Douala will assist the Cameroon CITES MA in the selection of project sites, and collection of field data on a scientific basis. The University of Adelaide and DoubleHelix will assist the Cameroon's forest administration through the Faculty of Sciences of the University of Douala in developing the tracking system based on Single Nucleotide Polymorphism (SNP) markers. Field activities will be carried in three regions including: Adamawa, Centre, East and Littoral.

A Technical Committee under the auspices of the MINFOF will be established to oversee the execution of the activity. The Technical Committee will provide guidance on technical matters and ensure that the specific activities are carried out according to the Work Plan.

The members of the Technical Committee will comprise staff from the CITES management authority, CITES scientific authority, the PAUs and the Institute for Agricultural Research for Development (IRAD). The management structure is illustrated in Figure 1 with 3 levels.

Figure 1 Management structure of the Activity



Monitoring, Reporting and Evaluation

The progress of the activity will be monitored by the Technical Committee.

DoubleHelix will be responsible for monthly progress reports based on the achievements of outputs/specific activities of the Work Plan and a final completion report will be prepared within 2 months of the activity completion for submission to ITTO.

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Annex A Detailed budget of University of Adelaide (Lead Consultant) and Double Helix Tracking Technologies Pte Ltd (Project Manager)

Table A1 breaks down the budget allocated to the University of Adelaide and DoubleHelix. It covers costs of development and implementation in Cameroon.

Table A1: Detailed budget by output and activity (US$)

N°	OUTPUT / ACTIVITY	TOTAL (US$)
10.	**Output 1.1 Development of SNP markers for *Prunus africana* suitable for DNA fingerprinting of bark (differentiation of bark between individual trees of the same species)**	
	11. Activity 1.1.1 Population sampling	In country partner
	12. Activity 1.1.2 Genetic marker development	5,100
	13. Component Total	**5,100**
20.	**Output 1.2 Determine the optimal supply chain verification sampling location and intensity.**	
	21. Activity 1.2.1 Analysis with genetic markers	120,000
	22. Component Total	**120,000**
30.	**Output 1.3 Implementation of DNA verification in supply chains at optimal location and intensity (as determined in Output 1.1) in Cameroon**	
	31. Activity 1.3.1 Workshops for stakeholder consultation and field training	In country partner
	32. Activity 1.3.2 Preparation of training “tool-box”	In country partner
	33. Component Total	In country partner
40.	**Output 1.4: Project report and draft publication**	
	41. Preparation of final report and draft publication	-
50.	**Project coordination and reporting**	
	51. Lead consultant	-
	52. Project manager	34,000
	53. Administrative support	10,000
	54. Component Total	**44 000**
100.	**GRAND TOTAL**	**169,100**