Wood identification tool development and application, examples from Malaysia

Lee Hong Tnah, Soon Leong Lee, Chin Hong Ng, Nurul Farhanah Zakaria, Kevin KS Ng, Chai Ting Lee, Amelia Azman, Suhaila Mahruji, Zainey Abdul Kadir, Khairuddin Perdan, Mohd Nizum Mohd Nor, Bibian Diway & Eyen Khoo

FRIM, FDPM, SFC & FRC

Asia-Pacific Forestry Week 2019
Forests for Peace and Well-being
17-21 June 2019 I Incheon, Republic of Korea

BACKGROUND

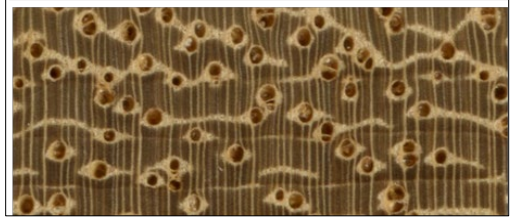
Illegal harvesting of forest resources poses a significant threat to the sustainability of forest ecosystems

BACKGROUND

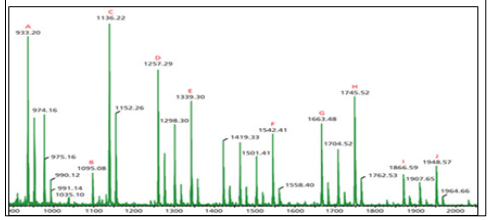
However

There are still lacking of timber tracking and identification tools that can be used for enforcement and regulation

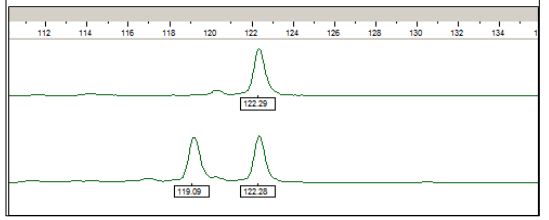
- **Image identification** (Wood anatomy & dendrochronology)



- **Chemical methods** (MS, NIRS, stable isotopes & radio-carbon)

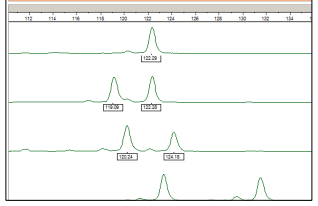


- **Genetic methods** (DNA barcodes, pop gen/phylogeo & DNA profiling)



Hence

- **Genetic methods** (DNA barcodes, pop gen/phylogeo & DNA profiling)



At FRIM, since 2007, by using genetic methods, we have developed DNA profiling and barcoding databases for TIMBER TRACKING & SPECIES IDENTIFICATION

How genetic methods are being used as an instrument for timber tracking?

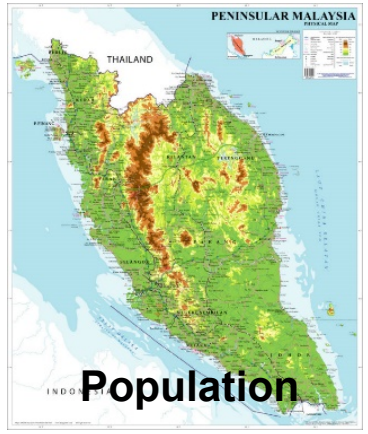
Suspected timber

Is it possible to extract quality DNA from dry wood/wood product?

Protocol

How to establish DNA database for species identification?

DNA barcoding



How to establish DNA database for population identification?

Phylogeography

How to establish DNA database for individual identification?

DNA profiling

Neobalanocarpus heimii (Chengal)

We have successfully developed a DNA-based timber tracking system for an importance timber in Malaysia

Journal of Tropical Forest Science 22(2): 214–226 (2010) — *Tnah LH et al.*

HIGHLY VARIABLE STR MARKERS OF *NEOBALANOCARPUS HEIMII* (DIPTEROCARPACEAE) FOR FORENSIC DNA PROFILING

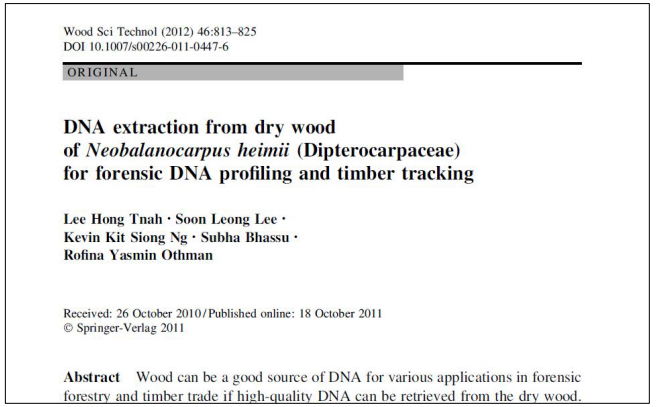
LH Tnah[1], SL Lee[1, *], KKS Ng[1], QZ Faridah[2] & I Faridah-Hanum[2]

[1]*Forest Research Institute Malaysia, 52109 Kepong, Selangor Darul Ehsan, Malaysia*
[2]*Universiti Putra Malaysia, 43400 Serdang, Selangor Darul Ehsan, Malaysia*

Received November 2009

TNAH LH, LEE SL, NG KKS, FARIDAH QZ & FARIDAH-HANUM I. 2010. Highly variable STR markers of *Neobalanocarpus heimii* (Dipterocarpaceae) for forensic DNA profiling. *Neobalanocarpus heimii*, locally known as chengal, is an important timber species in Peninsular Malaysia. Owing to the high demand for its valuable timber, *N. heimii* is subjected to illegal logging and this species may become endangered in the near future. The present study was designed to identify a set of highly polymorphic short tandem repeat (STR) markers for timber tracking of *N. heimii*. An extensive evaluation of 51 STRs developed for Dipterocarpaceae managed to identify 12 STR loci (*Nhe004, Nhe005, Nhe011, Nhe015, Nhe018, Hhu161, Ste392, Ste605, Shu014a, Shu03, Shu04* and *Shu07*), which showed specific amplification, high polymorphism, single-locus mode of inheritance, absence of null alleles and absence of mononucleotide repeat motifs in *N. heimii*. These loci can be readily used to establish a linkage between the evidentiary sample and the source, thus providing a useful set of markers for individual identification in *N. heimii*.

Keywords: Chengal, forensic science, individual identification, tropical tree species, illegal logging



Wood Sci Technol (2012) 46:813–825
DOI 10.1007/s00226-011-0447-6

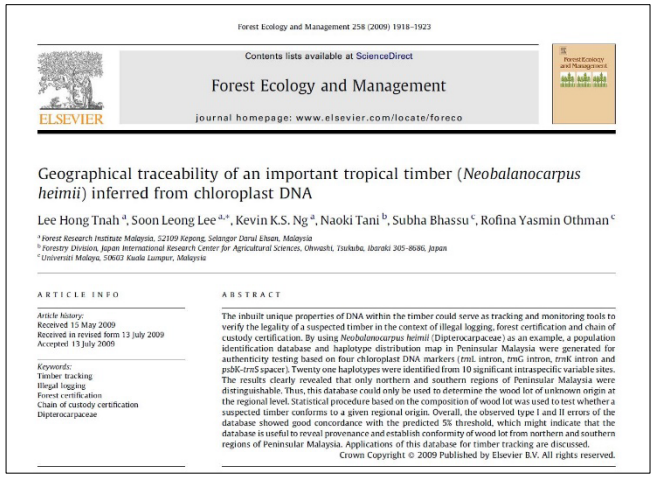
ORIGINAL

DNA extraction from dry wood of *Neobalanocarpus heimii* (Dipterocarpaceae) for forensic DNA profiling and timber tracking

Lee Hong Tnah · Soon Leong Lee · Kevin Kit Siong Ng · Subha Bhassu · Rofina Yasmin Othman

Received: 26 October 2010 / Published online: 18 October 2011
© Springer-Verlag 2011

Abstract Wood can be a good source of DNA for various applications in forensic forestry and timber trade if high-quality DNA can be retrieved from the dry wood.



Forest Ecology and Management 258 (2009) 1918–1923

Contents lists available at ScienceDirect

Forest Ecology and Management

journal homepage: www.elsevier.com/locate/foreco

ELSEVIER

Geographical traceability of an important tropical timber (*Neobalanocarpus heimii*) inferred from chloroplast DNA

Lee Hong Tnah [a], Soon Leong Lee [a,*], Kevin K.S. Ng [a], Naoki Tani [b], Subha Bhassu [c], Rofina Yasmin Othman [c]

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Timber tracking
Illegal logging
Forest certification
Chain of custody certification
Dipterocarpaceae

ABSTRACT

The inbuilt unique properties of DNA within the timber could serve as tracking and monitoring tools to verify the legality of a suspected timber in the context of illegal logging, forest certification and chain of custody certification. By using *Neobalanocarpus heimii* (Dipterocarpaceae) as an example, a population identification database and haplotype distribution map in Peninsular Malaysia were generated for authenticity testing based on four chloroplast DNA markers (*trnL* intron, *trnG* intron, *trnK* intron and *psbK–trnS* spacer). Twenty one haplotypes were identified from 10 significant intraspecific variable sites. The results clearly revealed that only northern and southern regions of Peninsular Malaysia were distinguishable. Thus, this database could only be used to determine the wood lot of unknown origin at the regional level. Statistical procedure based on the composition of wood lot was used to test whether a suspected timber conforms to a given regional origin. Overall, the observed type I and II errors of the database showed good concordance with the predicted 5% threshold, which might indicate that the database is useful to reveal provenance and establish conformity of wood lot from northern and southern regions of Peninsular Malaysia. Applications of this database for timber tracking are discussed.

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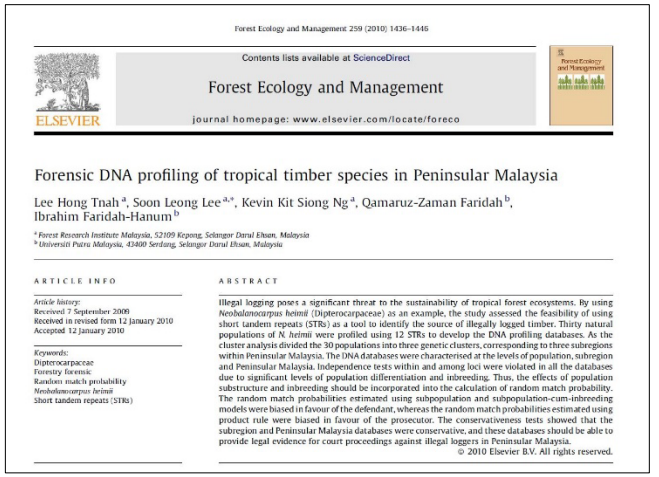

Forest Ecology and Management 259 (2010) 1436–1446

Contents lists available at ScienceDirect

Forest Ecology and Management

journal homepage: www.elsevier.com/locate/foreco

ELSEVIER

Forensic DNA profiling of tropical timber species in Peninsular Malaysia

Lee Hong Tnah [a], Soon Leong Lee [a,*], Kevin Kit Siong Ng [a], Qamaruz-Zaman Faridah [b], Ibrahim Faridah-Hanum [b]

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ARTICLE INFO

Article history:
Received 7 September 2009
Received in revised form 12 January 2010
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Keywords:
Dipterocarpaceae
Forestry forensic
Random match probability
Neobalanocarpus heimii
Short tandem repeats (STRs)

ABSTRACT

Illegal logging poses a significant threat to the sustainability of tropical forest ecosystems. By using *Neobalanocarpus heimii* (Dipterocarpaceae) as an example, the study assessed the feasibility of using short tandem repeats (STRs) as a tool to identify the source of illegally logged timber. Thirty natural populations of *N. heimii* were profiled using 12 STRs to develop the DNA profiling databases. As the cluster analysis divided the 30 populations into three genetic clusters, corresponding to three subregions within Peninsular Malaysia. The DNA databases were characterised at the levels of population, subregion and Peninsular Malaysia. Independence tests within and among loci were violated in all the databases due to significant levels of population differentiation and inbreeding. Thus, the effects of population substructure and inbreeding should be incorporated into the calculation of random match probability. The random match probabilities estimated using subpopulation and subpopulation-cum-inbreeding models were biased in favour of the defendant, whereas the random match probabilities estimated using product rule were biased in favour of the prosecutor. The conservativeness tests showed that the subregion and Peninsular Malaysia databases were conservative, and these databases should be able to provide legal evidence for court proceedings against illegal loggers in Peninsular Malaysia.

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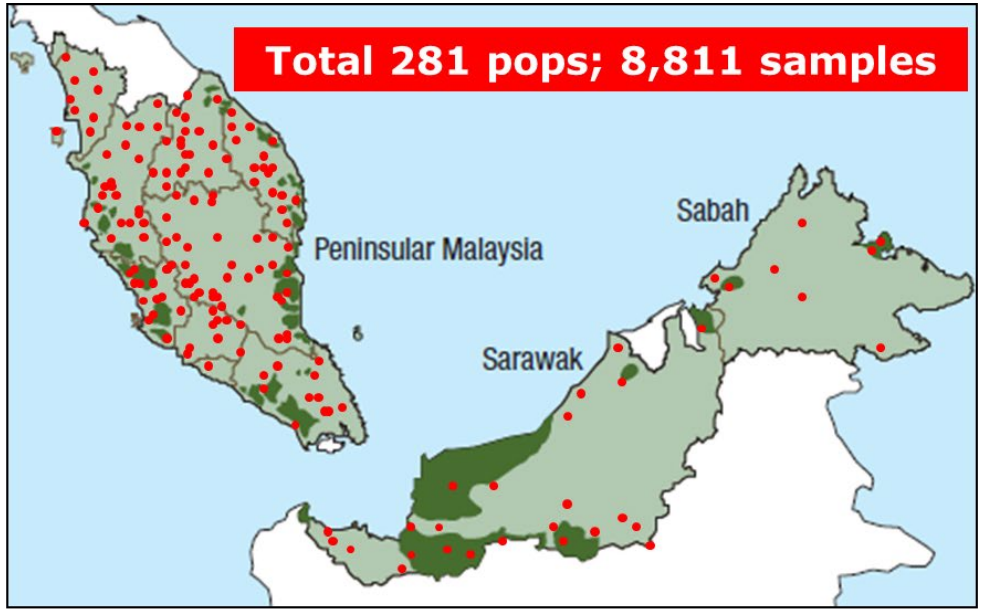
BACKGROUND



1	*Neobalanocarpus heimii*	30 pops (PM)
2	*Gonystylus bancanus*	17 pops (Ma)
3	*Koompassia malaccensis*	56 pops (Ma)
4	*Shorea platyclados*	27 pops (Ma)
5	*Aquilaria malaccensis*	35 pops (PM)
6	*Intsia palembanica*	39 pops (PM)
7	*Rhizophora apiculata*	43 pops (PM)
8	*Rhizophora mucronata*	34 pops (PM)

Forensic Science International: Genetics 23 (2016) 197–209

Contents lists available at ScienceDirect

Forensic Science International: Genetics

journal homepage: www.elsevier.com/locate/fsig

Forensic timber identification: a case study of a CITES listed species, *Gonystylus bancanus* (Thymelaeaceae)

Kevin Kit Siong Ng[a,*], Soon Leong Lee[a], Lee Hong Tnah[a], Zakaria Nurul-Farhanah[a], Chin Hong Ng[a], Chai Ting Lee[a], Naoki Tani[b], Bibian Diway[c], Pei Sing Lai[d], Eyen Khoo[e]

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Keywords:
DNA barcoding
DNA profiling
Ramin
Random match probability
Short tandem repeats (STRs)

ABSTRACT

Illegal logging and smuggling of *Gonystylus bancanus* (Thymelaeaceae) poses a serious threat to this fragile valuable peat swamp timber species. Using *G. bancanus* as a case study, DNA markers were used to develop identification databases at the species, population and individual level. The species level database for *Gonystylus* comprised of an nrDNA (ITS2) and two cpDNA (*trnH-psbA* and *trnL*) markers based on a 20 *Gonystylus* species database. When concatenated, taxonomic species recognition was achieved with a resolution of 90% (18 out of the 20 species). In addition, based on 17 natural populations of *G. bancanus* throughout West (Peninsular Malaysia) and East (Sabah and Sarawak) Malaysia, population and individual identification databases were developed using cpDNA and STR markers respectively. A haplotype distribution map for Malaysia was generated using six cpDNA markers, resulting in 12 unique multilocus haplotypes, from 24 informative intraspecific variable sites. These unique haplotypes suggest

PLOS ONE

RESEARCH ARTICLE

Geographic origin and individual assignment of *Shorea platyclados* (Dipterocarpaceae) for forensic identification

Chin Hong Ng[1☯*], Soon Leong Lee[1☯], Lee Hong Tnah[1☯], Kevin Kit Siong Ng[1☯], Chai Ting Lee[1☯], Bibian Diway[2‡], Eyen Khoo[3‡]

1 Division of Forestry Biotechnology, Forest Research Institute Malaysia, Kepong, Selangor, Malaysia, 2 Sarawak Forestry Corporation, Kuching, Sarawak, Malaysia, 3 Forest Research Centre, Sandakan, Sabah, Malaysia

☯ These authors contributed equally to this work.
‡ These authors also contributed equally to this work
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OPEN ACCESS

Citation: Ng CH, Lee SL, Tnah LH, Ng KKS, Lee CT, Diway B, et al. (2017) Geographic origin and individual assignment of *Shorea platyclados* (Dipterocarpaceae) for forensic identification. PLoS ONE 12(4): e0176158. https://doi.org/10.1371/journal.pone.0176158

Editor: Giovanni G. Vendramin, Consiglio Nazionale delle Ricerche, ITALY

Received: November 22, 2016

Abstract

The development of timber tracking methods based on genetic markers can provide scientific evidence to verify the origin of timber products and fulfill the growing requirement for sustainable forestry practices. In this study, the origin of an important Dark Red Meranti wood, *Shorea platyclados*, was studied by using the combination of seven chloroplast DNA and 15 short tandem repeats (STRs) markers. A total of 27 natural populations of *S. platyclados* were sampled throughout Malaysia to establish population level and individual level identification databases. A haplotype map was generated from chloroplast DNA sequencing for population identification, resulting in 29 multilocus haplotypes, based on 39 informative intraspecific variable sites. Subsequently, a DNA profiling database was developed from 15 STRs allowing for individual identification in Malaysia. Cluster analysis divided the 27 popu-

DNA BARCODES

DNA Barcodes

400 timber-sized species from 46 families

Adinandra dumosa	*Canarium megalanthum*	*Elaeocarpus ferrugineus*	*Knema scortechnii*	*Parashorea stellata*	*Shorea exelliptica*
Adinandra villosa	*Canarium pilosum*	*Elaeocarpus floribundus*	*Kokoona littoralis*	*Parashorea tomentella*	*Shorea faguetiana*
Agathis borneensis	*Carallia brachiata*	*Elaeocarpus griffithii*	*Kokoona reflexa*	*Parishia insignis*	*Shorea falcifera*
Aglaia argentea	*Castanopsis inermis*	*Elaeocarpus palembanicus*	*Koompassia excelsa*	*Payena lanceolata*	*Shorea kunstleri*
Aglaia odoratissima	*Ceriops tagal*	*Elaeocarpus petiolatus*	*Litsea ferruginea*	*Payena lucida*	*Shorea ladiana*
Aglaia palembanica	*Cratoxylum formosum*	*Elaeocarpus polystachys*	*Lumnitzera littorea*	*Payena maingayi*	*Shorea laevis*
Alangium ebenaceum	*Crudia curtisii*	*Elaeocarpus robustus*	*Lumnitzera racemosa*	*Payena obscura*	*Shorea lepidota*
Alstonia angustiloba	*Cyathocalyx pruniferus*	*Elaeocarpus stipularis*	*Madhuca kunstleri*	*Quercus argentata*	*Shorea leprosula*
Anisoptera costata	*Cynometra malaccensis*	*Elateriospermum tapos*	*Madhuca laurifolia*	*Rhizophora apiculata*	*Shorea longisperma*
Anisoptera curtisii	*Dacryodes costata*	*Endorspermum diadenum*	*Madhuca malaccensis*	*Rhizophora mucronata*	*Shorea lumutensis*
Anisoptera grossivenia	*Dacryodes laxa*	*Eusideroxylon zwageri*	*Madhuca motleyana*	*Rhodamnia cinerea*	*Shorea macrantha*
Anisoptera laevis	*Dacryodes rostata*	*Excoecaria agallocha*	*Madhuca penangiana*	*Rhodoleia championi*	*Shorea monticola*
Anisoptera scaphula	*Dacryodes rubiginosa*	*Gluta renghas*	*Madhuca ridleyi*	*Sandoricum koetjape*	*Shorea multiflora*
Aporosa arborea	*Dacryodes rugosa*	*Heritiera littoralis*	*Madhuca sericea*	*Schima wallichii*	*Shorea obscura*
Artocarpus elasticus	*Diospyros pillosanthera*	*Heritiera simplicifolia*	*Madhuca tubulosa*	*Shorea acuminata*	*Shorea ochracea*
Artocarpus lanceifolius	*Diospyros venosa*	*Hopea apiculata*	*Madhuca utilis*	*Shorea acuminatissima*	*Shorea ovalis*
Artocarpus rigidus	*Diospyros wallichii*	*Hopea auriculata*	*Magnolia elegans*	*Shorea agamii*	*Shorea pilosa*
Artocarpus scortechinii	*Dipterocarpus baudii*	*Hopea beccariana*	*Mesua ferrea*	*Shorea albida*	*Shorea pinanga*
Avicennia alba	*Dipterocarpus kerrii*	*Hopea bilitonensis*	*Microcos tomentosa*	*Shorea almon*	*Shorea platycarpa*
Avicennia marina	*Dipterocarpus rigidus*	*Hopea bracteata*	*Nageia motleyi*	*Shorea amplexicaulis*	*Shorea platyclados*
Avicennia officinalis	*Dipterocarpus semivestitus*	*Hopea dryobalanoides*	*Neesia altissima*	*Shorea angustifolia*	*Shorea richetia*
Azadirachta excelsa	*Dipterocarpus tempehes*	*Hopea dyeri*	*Neobalanocarpus heimii*	*Shorea argentifolia*	*Shorea roxburghii*
Baccaurea parviflora	*Dracontomelon dao*	*Hopea ferruginea*	*Neolamarckia cadamba*	*Shorea atrinervosa*	*Shorea rubra*
Bhesa paniculata	*Dryobalanops aromatica*	*Hopea glaucescens*	*Octomeles sumatrana*	*Shorea balanocarpoides*	*Shorea rugosa*
Bouea macrophylla	*Dryobalanops beccarii*	*Hopea griffithii*	*Palaquium clarkeanum*	*Shorea ciliata*	*Shorea uliginosa*
Bouea oppositifolia	*Dryobalanops lanceolata*	*Hopea sublanceolata*	*Palaquium gutta*	*Shorea collaris*	*Shorea venulosa*
Bruguiera cylindrica	*Dryobalanops oblongifolia*	*Hopea sulcata*	*Palaquium harveyi*	*Shorea collina*	*Shorea waltoni*
Bruguiera gymnorrhiza	*Dryobalanops rappa*	*Hopea wightiana*	*Palaquium hexandrum*	*Shorea confusa*	*Vatica havilandii*
Bruguiera hainesii	*Durio graveolens*	*Hunteria zeylanica*	*Palaquium hispidum*	*Shorea coriacea*	*Vatica lobata*
Bruguiera parviflora	*Durio griffithii*	*Hydnocarpus castaneus*	*Palaquium maingayi*	*Shorea crassa*	*Vatica micrantha*
Calophyllum inophyllum	*Durio malaccensis*	*Hydnocarpus filipes*	*Palaquium microphyllum*	*Shorea curtisii*	*Vatica nitens*
Calophyllum tetrapterum	*Durio zibethinus*	*Hydnocarpus woodii*	*Parartocarpus venenosus*	*Shorea dasyphylla*	*Vatica odorata*
Cananga odorata	*Dyera costulata*	*Instia bijuga*	*Parashorea densiflora*	*Shorea dealbata*	*Vatica pauciflora*
Canarium apertum	*Dysoxylum arborescens*	*Instia palembanica*	*Parashorea macrophylla*	*Shorea domatiosa*	*Xylocarpus granatum*
Canarium littorale	*Elaeocarpus angustifolius*	*Knema laurina*	*Parashorea malaanonan*	*Shorea elliptica*	*Xylocarpus moluccensis*

DNA Barcodes

20 *Gonystylus* spp. (CITES listed)

- *G. affinis var affinis*
- *G. affinis var elegans*
- *G. areolatus*
- *G. augescens*
- *G. bancanus*
- *G. borneensis*
- *G. brunnescens*
- *G. calophylloides*
- *G. calophyllus*
- *G. consanguineus*
- *G. costalis*
- *G. eximius*
- *G. forbesii*
- *G. lucidulus*
- *G. maingayi*
- *G. micranthus*
- *G. othmanii*
- *G. spectabilis*
- *G. stenosepalus*
- *G. xylocarpus*

DNA Barcodes

100 important Malaysian medicinal plants

DNA Barcodes

Aquilaria species

Seven *Aquilaria* species:

- *A. malaccensis*
- *A. subintegra*
- *A. crassna*
- *A. sinensis*
- *A. hirta*
- *A. microcarpa*
- *A. beccariana*

Food Control 95 (2019) 318–326

Contents lists available at ScienceDirect

Food Control

journal homepage: www.elsevier.com/locate/foodcont

DNA barcode database of common herbal plants in the tropics: a resource for herbal product authentication

L.H. Tnah*, S.L. Lee, A.L. Tan, C.T. Lee, K.K.S. Ng, C.H. Ng, Z. Nurul Farhanah

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ARTICLE INFO

Keywords:
DNA barcoding
Chloroplast DNA
rbcL
Species identification
trnH-psbA

ABSTRACT

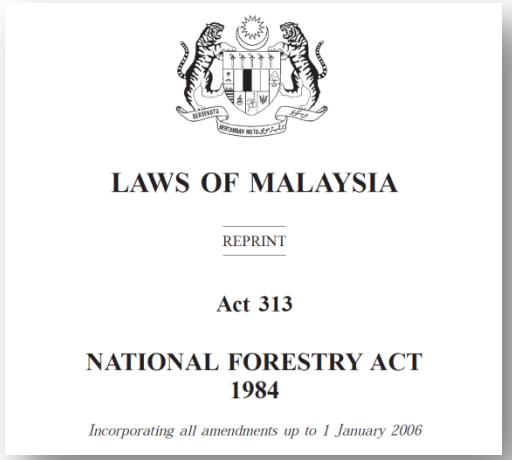
Ensuring the authenticity of raw materials used in herbal manufacturing is a key step prior to material processing. As species authentication is fundamental in the confirmation of herbal product quality, DNA barcoding techniques represent an efficient method for detecting plant-based adulterants in traded herbal products. Through this study, we established a DNA barcoding authentication system for 112 common herbal plant species in the tropics, which can be used for species identification and authentication. The DNA barcode reference database for the authentication system was generated using *rbcL* for primary differentiation, and *trnH-psbA* for secondary differentiation. The performance of the barcodes in resolving species was evaluated using similarity BLAST, phylogenetic tree reconstruction and by estimating the barcoding gap. In this study, the multigene tiered approach for DNA barcoding is proven robust with high species-level resolution (96.4%). Upon completion of the DNA barcoding authentication system, 30 herbal products from the local market were tested for their authenticity using this approach. Recovery of DNA barcodes from the herbal products was 73.4%, of which 56.7% of the products tested were authentic, whereas 10% of the herbal products were substituted with other plant taxa and 6.7% were contaminated. To this end, authentication of herbal products is challenging, but with the es-

Standard Operating Procedure (SOP)

In order for these timber tracking systems to be used:

1) To provide forensic evidences for the conviction of illegal loggers under National Forestry Act 1984

2) For timber certification to meet the international regulations such as CITES, US Lacey Act, EU and Australia Timber Regulations


LAWS OF MALAYSIA

REPRINT

Act 313

NATIONAL FORESTRY ACT 1984

Incorporating all amendments up to 1 January 2006

STANDARD OPERATING PROCEDURE (SOP)

FORENSIC DNA TESTING ON PLANT SPECIES IDENTIFICATION AND WOOD TRACKING

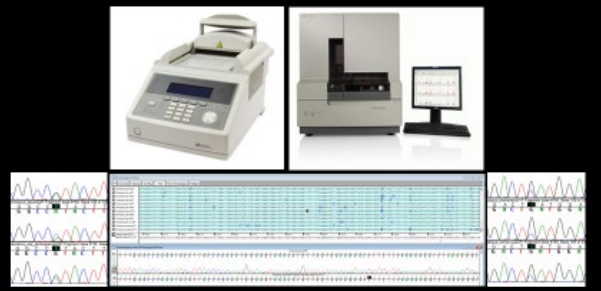
Genetics Laboratory,
Forest Research Institute Malaysia (FRIM)

Version 1.3 (1 June 2018)

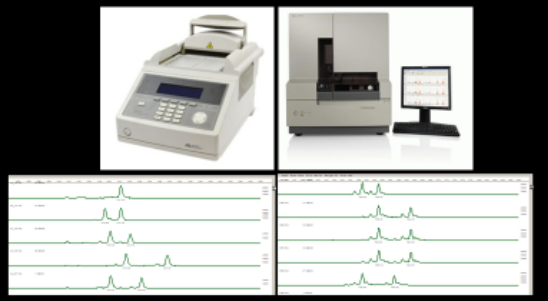
SOP1: SAMPLE COLLECTION IN THE FIELD

SOP2: DNA ISOLATION AND PURIFICATION FROM CAMBIUM & WOOD

SOP3: DNA SEQUENCING FOR SPECIES AND POPULATION IDENTIFICATION



SOP4: SHORT TANDEM REPEAT (STR) GENOTYPING FOR POPULATION AND INDIVIDUAL IDENTIFICATION

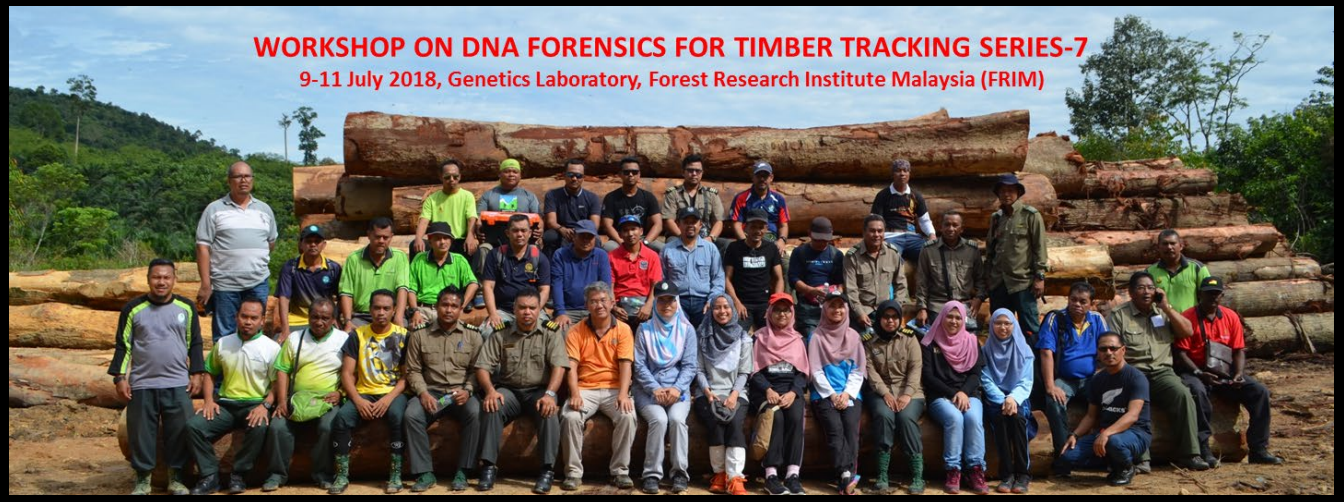


SOP1 - WoodFoKit®

CHAIN OF CUSTODY RECORD

ITEM NO.	DATE & TIME OF SEIZURE	DESCRIPTION OF EVIDENCE/PROPERTY
MGF013	13/8/2016 & 12.51pm	Barang kes kabium Chengal H.S. Hulu Langat

FROM: **Investigation Officer (FDPM)**	RELEASE SIGNATURE: Khairuddin	RELEASE DATE: 14/8/2016	DELIVERED VIA: ☐ MAIL ☒ IN PERSON ☐ OTHER:
TO: **Lab Officer (FRIM)**	RECEIPT SIGNATURE: Soonleong	RECEIPT DATE: 14/8/2016	
FROM: **Lab Officer (FRIM)**	RELEASE SIGNATURE: Soonleong	RELEASE DATE: 28/8/2016	DELIVERED VIA: ☐ MAIL ☒ IN PERSON ☐ OTHER:
TO: **Investigation Officer (FDPM)**	RECEIPT SIGNATURE: Khairuddin	RECEIPT DATE: 28/8/2016	
FROM:	RELEASE SIGNATURE	RELEASE DATE	DELIVERED VIA: ☐ MAIL ☐ IN PERSON ☐ OTHER:
TO:	RECEIPT SIGNATURE	RECEIPT DATE	


WORKSHOP ON DNA FORENSICS FOR TIMBER TRACKING SERIES-7
9-11 July 2018, Genetics Laboratory, Forest Research Institute Malaysia (FRIM)


WORKSHOP ON DNA FORENSICS FOR TIMBER TRACKING SERIES-6
19-20 Sept 2017, Genetics Laboratory, Forest Research Institute Malaysia (FRIM)


MAKMAL GENETIK
MAKMAL GENETIK


WORKSHOP ON DNA FORENSICS FOR TIMBER TRACKING SERIES-2
28 Oct 2015, Genetics Laboratory, Forest Research Institute Malaysia (FRIM)
B6

The SOPs were presented to the Council for Forest & Silvicultural, and Committee of Forest Operations & Monitoring to be implemented in Peninsular Malaysia

APPLICATIONS

FORENSIC DNA TESTING FOR PLANT SPECIES IDENTIFICATION AND TIMBER TRACKING

REPORT TO

Test Code: **MGF0115**

Dr. Lee Soon Leong, Dr. Tnah Lee Hong, Nurul Farhanah Zakaria, Dr. Kevin Ng Kit Siong, Dr. Ng Chin Hong & Dr. Lee Chai Ting

Genetics Laboratory
Forest Research Institute Malaysia (FRIM)
52109 Kepong, Selangor, Malaysia

30 September 2015

FORENSIC DNA TESTING FOR PLANT SPECIES IDENTIFICATION AND TIMBER TRACKING

REPORT TO

Test Code: **MGF0116**

Dr. Lee Soon Leong, Dr. Kevin Ng Kit Siong, Dr. Ng Chin Hong, Dr. Tnah Lee Hong, Nurul Farhanah Zakaria & Dr. Lee Chai Ting

Genetics Laboratory
Forest Research Institute Malaysia (FRIM)
52109 Kepong, Selangor, Malaysia

25 August 2016

TESTING SERVICES
DNA FORENSICS FOR TIMBER TRACKING

REPORT TO

Test Code: **MGF0117**

Dr. Lee Soon Leong, Dr. Kevin Ng Kit Siong, Dr. Ng Chin Hong, Dr. Tnah Lee Hong, Nurul Farhanah Zakaria & Dr. Lee Chai Ting

Genetics Laboratory
Forest Research Institute Malaysia (FRIM)
4 Jan 2017

FORENSIC DNA TESTING FOR PLANT SPECIES IDENTIFICATION AND TIMBER TRACKING

REPORT TO

Test Code: **MGF0817**

Dr. Lee Soon Leong, Dr. Lee Chai Ting, Dr. Kevin Ng Kit Siong, Dr. Ng Chin Hong, Dr. Tnah Lee Hong & Nurul Farhanah Zakaria

Genetics Laboratory
Forest Research Institute Malaysia (FRIM)
52109 Kepong, Selangor, Malaysia

7 June 2017

FORENSIC DNA TESTING FOR PLANT SPECIES IDENTIFICATION AND TIMBER TRACKING

REPORT TO

Test Code: **MGF0118**

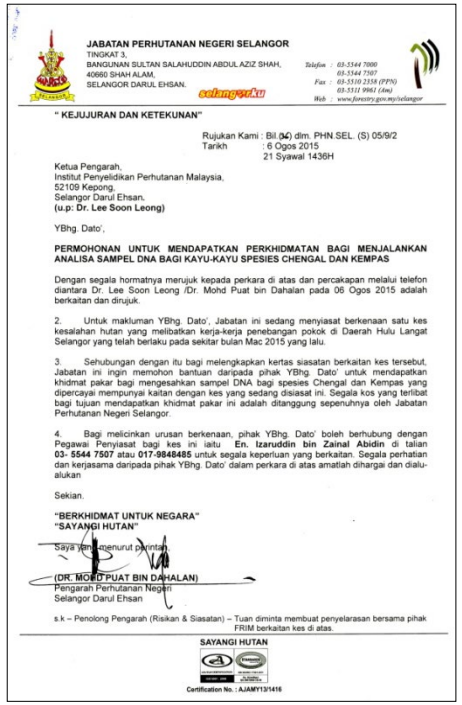
Dr. Ng Chin Hong, Dr. Lee Chai Ting, Dr. Kevin Ng Kit Siong, Dr. Tnah Lee Hong, Nurul Farhanah Zakaria and Dr. Lee Soon Leong

Genetics Laboratory
Forest Research Institute Malaysia (FRIM)
52109 Kepong, Selangor, Malaysia

10 April 2018

CASE STUDY 1

- On 6 August 2015, a request was received from State Forest Department (FD) to investigate a forest offense in a Forest Reserve



JABATAN PERHUTANAN NEGERI SELANGOR
TINGKAT 3,
BANGUNAN SULTAN SALAHUDDIN ABDUL AZIZ SHAH,
40660 SHAH ALAM,
SELANGOR DARUL EHSAN.

" KEJUJURAN DAN KETEKUNAN"

Rujukan Kami : Bil.(34) dlm. PHN.SEL. (S) 05/9/2
Tarikh : 6 Ogos 2015
21 Syawal 1436H

Ketua Pengarah,
Institut Penyelidikan Perhutanan Malaysia,
52109 Kepong,
Selangor Darul Ehsan.
(u.p: Dr. Lee Soon Leong)

YBhg. Dato',

PERMOHONAN UNTUK MENDAPATKAN PERKHIDMATAN BAGI MENJALANKAN ANALISA SAMPEL DNA BAGI KAYU-KAYU SPESIES CHENGAL DAN KEMPAS

Dengan segala hormatnya merujuk kepada perkara di atas dan percakapan melalui telefon diantara Dr. Lee Soon Leong /Dr. Mohd Puat bin Dahalan pada 06 Ogos 2015 adalah berkaitan dan dirujuk.

2. Untuk makluman YBhg. Dato', Jabatan ini sedang menyiasat berkenaan satu kes kesalahan hutan yang melibatkan kerja-kerja penebangan pokok di Daerah Hulu Langat Selangor yang telah berlaku pada sekitar bulan Mac 2015 yang lalu.

3. Sehubungan dengan itu bagi melengkapkan kertas siasatan berkaitan kes tersebut, Jabatan ini ingin memohon bantuan daripada pihak YBhg. Dato' untuk mendapatkan khidmat pakar bagi mengesahkan sampel DNA bagi spesies Chengal dan Kempas yang dipercayai mempunyai kaitan dengan kes yang sedang disiasat ini. Segala kos yang terlibat bagi tujuan mendapatkan khidmat pakar ini adalah ditanggung sepenuhnya oleh Jabatan Perhutanan Negeri Selangor.

4. Bagi melicinkan urusan berkenaan, pihak YBhg. Dato' boleh berhubung dengan Pegawai Penyiasat bagi kes ini iaitu **En. Izaruddin bin Zainal Abidin** di talian **03- 5544 7507** atau **017-9848485** untuk segala keperluan yang berkaitan. Segala perhatian dan kerjasama daripada pihak YBhg. Dato' dalam perkara di atas amatlah dihargai dan dialu-alukan

Sekian.

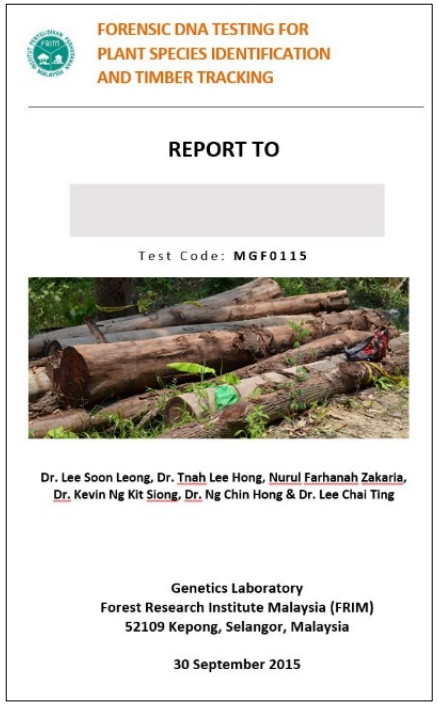
"BERKHIDMAT UNTUK NEGARA"
"SAYANGI HUTAN"

Saya yang menurut perintah,

(DR. MOHD PUAT BIN DAHALAN)
Pengarah Perhutanan Negeri
Selangor Darul Ehsan

s.k – Penolong Pengarah (Risikan & Siasatan) – Tuan diminta membuat penyelarasan bersama pihak FRIM berkaitan kes di atas.

SAYANGI HUTAN



FORENSIC DNA TESTING FOR PLANT SPECIES IDENTIFICATION AND TIMBER TRACKING

REPORT TO

Test Code: **MGF0115**

Dr. Lee Soon Leong, Dr. Tnah Lee Hong, Nurul Farhanah Zakaria, Dr. Kevin Ng Kit Siong, Dr. Ng Chin Hong & Dr. Lee Chai Ting

Genetics Laboratory
Forest Research Institute Malaysia (FRIM)
52109 Kepong, Selangor, Malaysia

30 September 2015

- The Director of the State FD was requesting a forensic examination and report for possible criminal charges

FORENSIC ACQUISITION

Guided by the officials from the State FD, sample collections were carried out in the field following SOP1: SPECIMEN COLLECTION IN THE FIELD

FORENSIC ACQUISITION

FORENSIC ACQUISITION

FORENSIC ACQUISITION

Wood tissues were collected from 13 questioned logs (Logs 1 – 13) and six potential stumps (Stumps 1 – 6) suspected to be Chengal

No	DNA ID	JPN ID	Length (m)	Diameter (cm)	Location (GPS)	Type of tissue	Collection date & time
Logs							
1	MG-F003	311	6.9	43	N3°05'31.3"E101°49'19.8"	Cambium	13/08/2015 & 11:58
2	MG-F004	314	6.2	50	N3°05'31.3"E101°49'19.8"	Cambium	13/08/2015 & 12:03
3	MG-F005	313	9.8	24	N3°05'31.3"E101°49'19.8"	Cambium	13/08/2015 & 12:11
4	MG-F006	312	5.9	30	N3°05'31.3"E101°49'19.8"	Cambium	13/08/2015 & 12:18
5	MG-F007	309	7.5	38	N3°05'31.3"E101°49'19.8"	Cambium	13/08/2015 & 12:25
6	MG-F008	315	6.1	23	N3°05'31.3"E101°49'19.8"	Cambium	13/08/2015 & 12:32
7	MG-F009	308	6.1	56	N3°05'31.3"E101°49'19.8"	Cambium	13/08/2015 & 12:36
8	MG-F010	316	9.2	20	N3°05'31.3"E101°49'19.8"	Cambium	13/08/2015 & 12:40
9	MG-F011	306	7.4	22	N3°05'31.3"E101°49'19.8"	Cambium	13/08/2015 & 12:45
10	MG-F012	305	6.2	35	N3°05'31.3"E101°49'19.8"	Cambium	13/08/2015 & 12:47
11	MG-F013	304	8.5	29	N3°05'31.3"E101°49'19.8"	Cambium	13/08/2015 & 12:51
12	MG-F014	310	5.0	36	N3°05'31.3"E101°49'19.8"	Wood core	13/08/2015 & 12:58
13	MG-F021	307	1.4	40	N3°05'31.3"E101°49'19.8"	Wood core	27/08/2015 & 11:30
Stumps							
1	MG-F001	C42	-	86	N3°05'40.3"E101°49'24.7"	Cambium	13/08/2015 & 11:08
2	MG-F002	C32	-	37	N3°05'41.7"E101°49'27.3"	Cambium	13/08/2015 & 11:29
3	MG-F015	A28	-	95	N3°05'43.0"E101°49'24.8"	Cambium	13/08/2015 & 13:40
4	MG-F018	B28	-	55	N3°05'45.2"E101°49'27.4"	Cambium	18/08/2015 & 11:22
5	MG-F019	A22	-	47	N3°05'44.2"E101°49'24.1"	Cambium	18/08/2015 & 11:48
6	MG-F020	A37	-	50	N3°05'44.6"E101°49'17.4"	Cambium	27/08/2015 & 12:44

13 questioned logs (Logs 1 – 13)

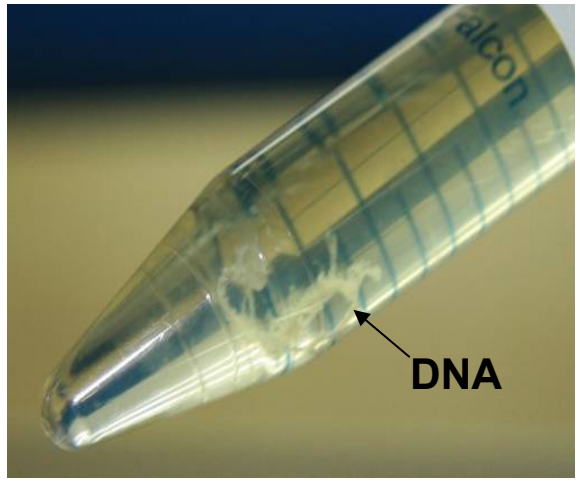
Six potential stumps (Stumps 1 – 6)

FORENSIC ANALYSIS

The total DNA of the collected tissues were extracted and purified according to SOP2: DNA ISOLATION AND PURIFICATION FROM WOOD TISSUES



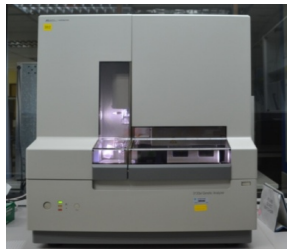


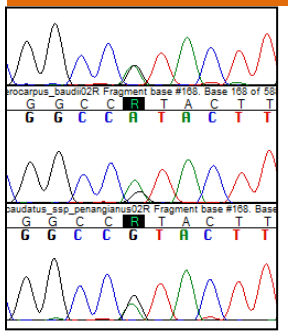
FORENSIC ANALYSIS

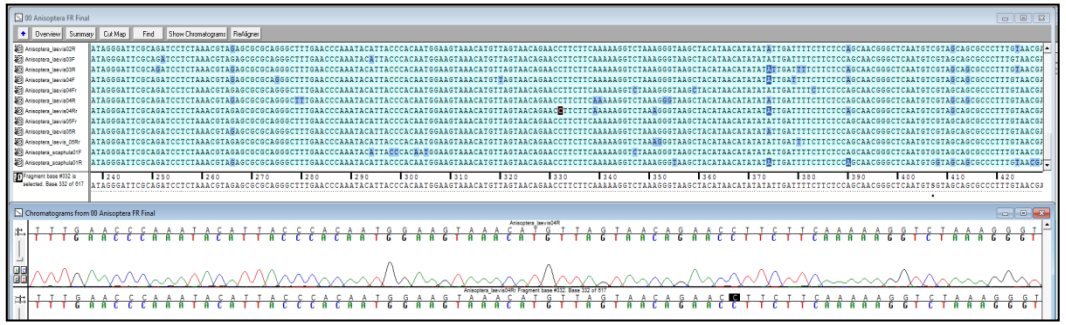
The purified DNAs obtained were subsequently sequenced on a chloroplast region (*rbc*L) following SOP3: DNA SEQUENCING FOR SPECIES AND POPULATION IDENTIFICATION

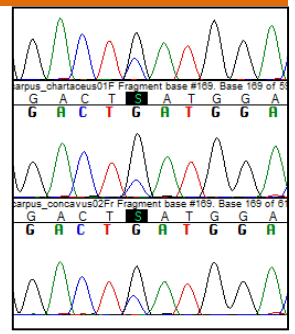
DNA sequencing for F & R primers



DNA sequences edited manually by visual inspection of electropherograms using Sequencher & aligned using ClustalW







FORENSIC ANALYSIS

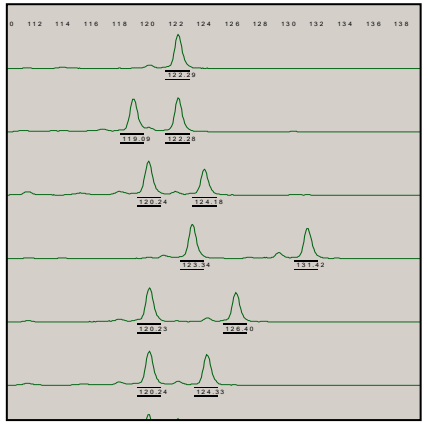
The purified DNAs obtained were further genotyped using SOP4: STR GENOTYPING FOR POPULATION AND INDIVIDUAL IDENTIFICATION

PCR

Fragment analysis

Scoring

Genotype (data)



11	260	275	121	125	162	182	147	171	131	177	214	214	142	147	108
12	260	277	103	115	182	182	147	147	141	153	214	214	139	148	102
13	260	277	117	121	188	188	147	167	141	177	214	215	138	142	102
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16	260	262	103	115	162	182	147	159	129	177	214	215	142	149	102
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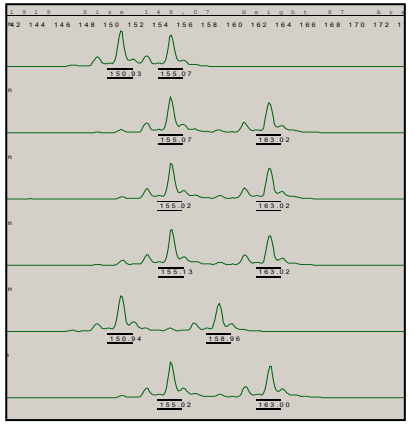
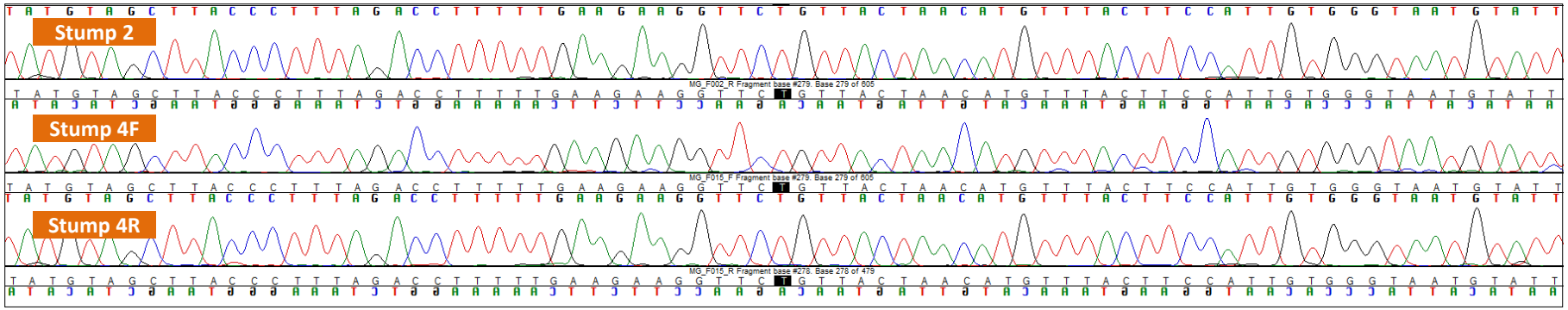
18	265	278	103	121	202	202	135	135	141	177	215	215	142	149	108
19	260	260	103	125	162	162	147	171	129	141	214	214	139	149	108
20	260	262	103	115	182	182	159	167	149	177	207	214	142	148	90
21	259	276	113	131	167	167	155	175	131	141	207	214	138	145	102
22	265	275	115	125	182	182	167	175	131	177	214	215	139	149	90
23	260	300	103	125	161	182	153	159	131	177	214	214	148	149	96
24	?	?	103	121	162	182	147	175	131	153	207	214	138	147	102
25	257	262	115	125	182	182	159	171	141	177	214	214	139	142	102
26	260	265	121	125	182	196	147	167	137	141	214	215	138	138	102
27	260	260	103	125	162	162	155	167	137	137	215	219	142	143	102
28	260	277	103	103	182	188	147	147	145	177	214	214	142	148	102



FINDINGS AND CONCLUSIONS

1) Could the questioned Logs 1 - 13 and the Stumps 1 - 6 belong to the species Chengal (*N. heimii*)?



FRIM'S DNA BARCODING DATABASE OF MAJOR TIMBERS

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>Anisoptera_costata02
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>Dipterocarpus_concavus04
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>Dipterocarpus_concavus05
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>Dryobalanops_lanceolata05
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>Dryobalanops_lanceolata06
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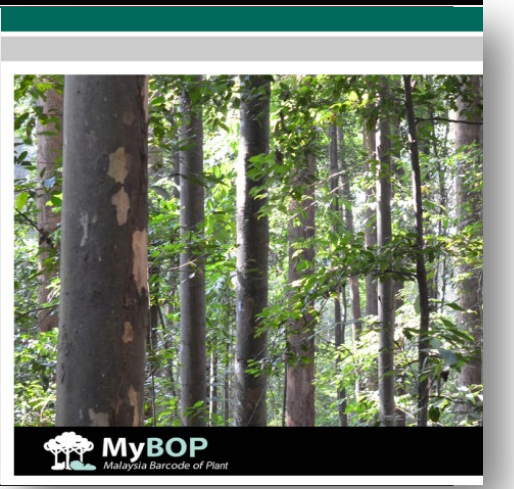
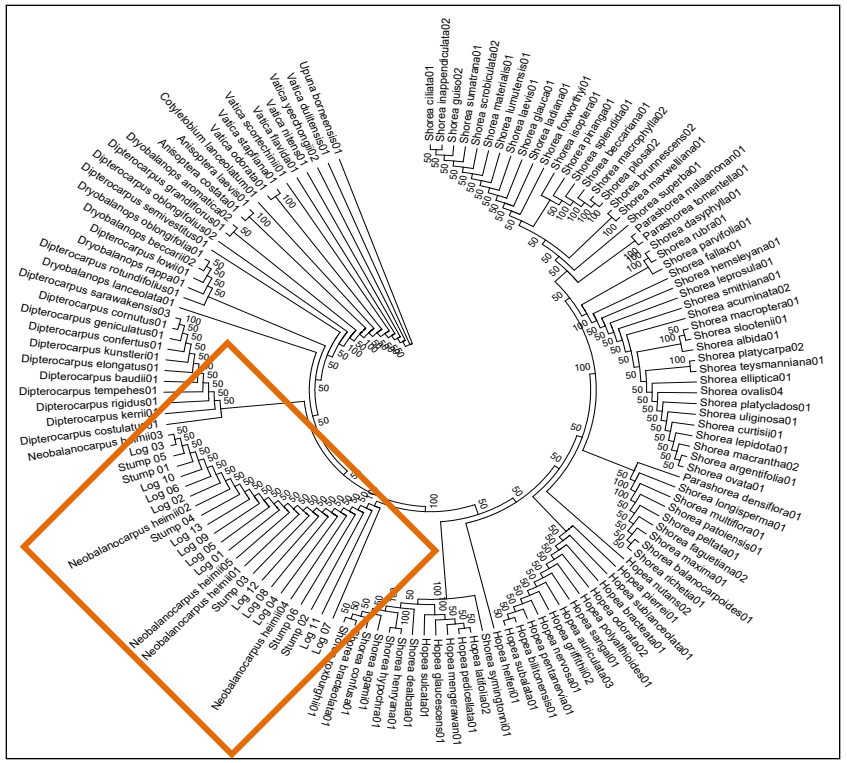


FINDINGS AND CONCLUSIONS

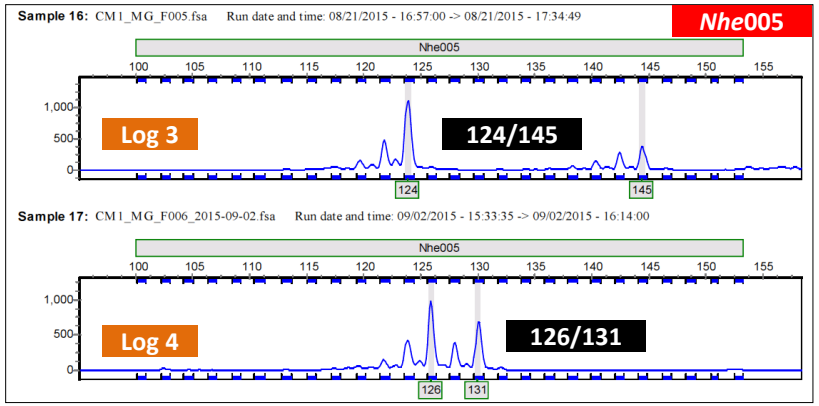
Query ID	Subject ID	% identity	Alignment length	Evalue	Bit score
Log 1	Chengal (*N. heimii*)	100.00	605	0	1081
Log 2	Chengal (*N. heimii*)	100.00	605	0	1081
Log 3	Chengal (*N. heimii*)	100.00	605	0	1081
Log 4	Chengal (*N. heimii*)	100.00	605	0	1081
Log 5	Chengal (*N. heimii*)	100.00	605	0	1081
Log 6	Chengal (*N. heimii*)	100.00	605	0	1081
Log 7	Chengal (*N. heimii*)	100.00	605	0	1081
Log 8	Chengal (*N. heimii*)	100.00	605	0	1081
Log 9	Chengal (*N. heimii*)	100.00	605	0	1081
Log 10	Chengal (*N. heimii*)	100.00	605	0	1081
Log 11	Chengal (*N. heimii*)	100.00	605	0	1081
Log 12	Chengal (*N. heimii*)	100.00	605	0	1081
Log 13	Chengal (*N. heimii*)	100.00	605	0	1081
Stump 1	Chengal (*N. heimii*)	100.00	605	0	1081
Stump 2	Chengal (*N. heimii*)	100.00	605	0	1081
Stump 3	Chengal (*N. heimii*)	100.00	605	0	1081
Stump 4	Chengal (*N. heimii*)	100.00	605	0	1081
Stump 5	Chengal (*N. heimii*)	100.00	605	0	1081
Stump 6	Chengal (*N. heimii*)	100.00	605	0	1081

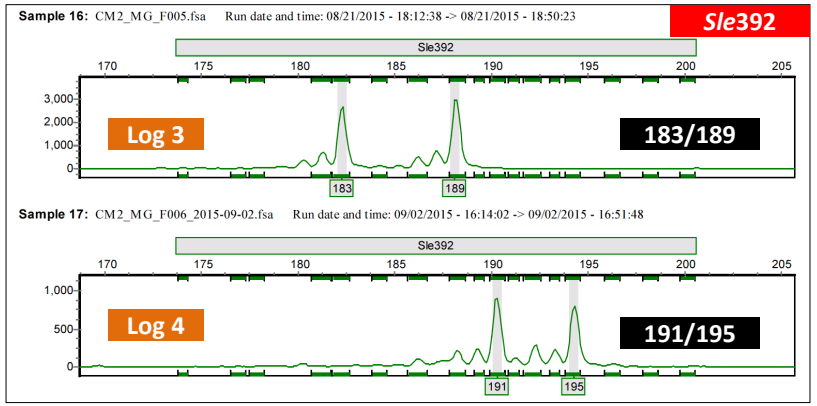


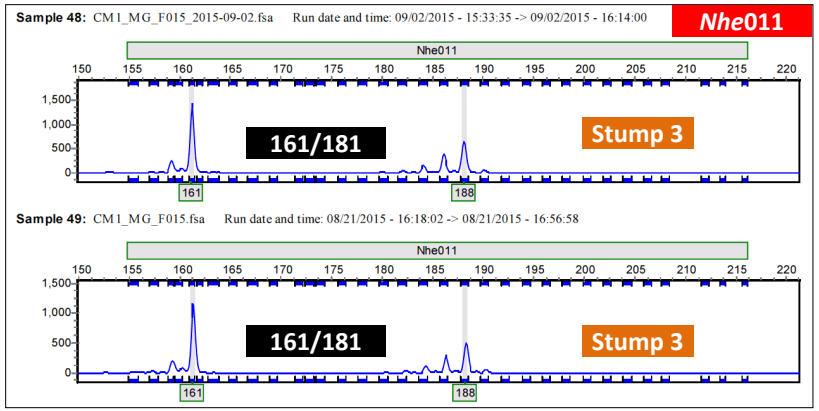
Conclude that Logs 1 - 13 and Stumps 1 - 6 belong to the species Chengal (*N. heimii*)

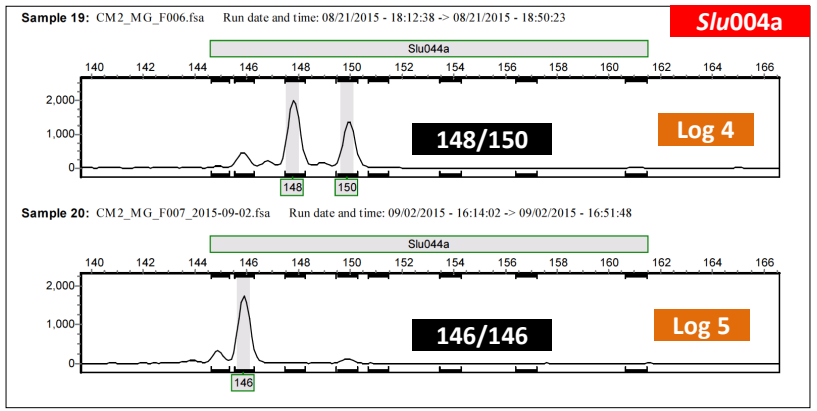
FINDINGS AND CONCLUSIONS

2) Could the questioned Logs 1 - 13 be contributed from a same tree and be originated from the Stumps 1 - 6?









FINDINGS AND CONCLUSIONS

No	*Nhe*004	*Nhe*005	*Nhe*011	*Nhe*015	*Shc*07	*Hbi*161	*Slu*044a	*Nhe*018	*Sle*392	*Shc*03
Logs										
1	258/259	103/122	172/180	130/160	141/155	98/98	146/146	139/139	189/191	124/134
2	258/264	124/145	162/168	164/176	141/155	114/114	146/146	137/137	183/189	126/136
3	258/264	124/145	162/168	164/176	141/155	114/114	146/146	137/137	183/189	126/136
4	266/277	126/131	180/186	148/150	151/161	101/104	148/150	137/137	191/195	126/132
5	258/259	103/122	172/180	130/160	141/155	98/98	146/146	139/139	189/191	124/134
6	283/283	124/145	161/170	164/170	157/157	98/107	146/150	139/139	187/189	126/128
7	258/264	124/145	162/168	164/176	141/155	114/114	146/146	137/137	183/189	126/136
8	258/259	103/122	172/180	130/160	141/155	98/98	146/146	139/139	189/191	124/134
9	258/264	124/145	162/168	164/176	141/155	114/114	146/146	137/137	183/189	126/136
10	283/283	124/145	161/170	164/170	157/157	98/107	146/150	139/139	187/189	126/128
11	258/264	124/145	162/168	164/176	141/155	114/114	146/146	137/137	183/189	126/136
12	283/283	124/145	161/170	164/170	157/157	98/107	146/150	139/139	187/189	126/128
13	258/264	124/145	162/168	164/176	141/155	114/114	146/146	137/137	183/189	126/136
Stumps										
1	258/273	122/122	180/180	148/170	171/171	101/114	146/146	137/137	189/189	126/128
2	264/283	126/128	161/161	142/164	155/161	104/114	146/150	137/137	183/185	132/132
3	264/273	122/132	161/188	148/148	151/155	98/101	146/150	137/143	183/189	128/128
4	258/259	103/122	172/180	130/160	141/155	98/98	146/146	139/139	189/191	124/134
5	264/266	128/132	161/170	162/166	141/161	98/114	146/150	133/137	187/195	126/138
6	283/283	124/145	161/170	164/170	157/157	98/107	146/150	139/139	187/189	126/128

FINDINGS AND CONCLUSIONS

FRIM'S INDIVIDUAL IDENTIFICATION DATABASE OF CHENGAL

Allele size - frequency										
*Nhe*004		*Nhe*005	*Nhe*011	*Nhe*015	*Nhe*018	*Hbi*161	*Slu*044a	*Sle*392	*Shc*03	*Shc*07
241 - 0.0032	286 - 0.0162	103 - 0.0597	156 - 0.0063	134 - 0.0096	123 - 0.0032	92 - 0.0255	146 - 0.5723	179 - 0.0032	120 - 0.0064	141 - 0.0764
243 - 0.0032	291 - 0.0097	109 - 0.0535	158 - 0.0095	136 - 0.0127	125 - 0.0064	95 - 0.4331	148 - 0.0189	181 - 0.0032	123 - 0.0064	143 - 0.0127
246 - 0.0032	294 - 0.0195	111 - 0.0063	159 - 0.0190	142 - 0.0191	131 - 0.0096	98 - 0.0701	150 - 0.3899	183 - 0.0064	124 - 0.1146	147 - 0.0159
248 - 0.0032	297 - 0.0260	114 - 0.2767	160 - 0.0570	144 - 0.0318	133 - 0.0032	100 - 0.0032	154 - 0.0157	185 - 0.1019	126 - 0.6274	151 - 0.3057
251 - 0.0130	302 - 0.0292	116 - 0.1950	161 - 0.0728	146 - 0.0382	135 - 0.0764	101 - 0.2484	160 - 0.0031	187 - 0.086	128 - 0.0732	153 - 0.0032
256 - 0.1136	309 - 0.0032	118 - 0.0189	162 - 0.0506	148 - 0.1401	137 - 0.2866	104 - 0.0191		189 - 0.2389	129 - 0.0064	155 - 0.0255
257 - 0.0065		120 - 0.0252	164 - 0.0316	150 - 0.0860	139 - 0.0510	107 - 0.1115		191 - 0.4140	130 - 0.0287	157 - 0.1146
258 - 0.0844		122 - 0.0346	166 - 0.0380	152 - 0.0860	141 - 0.0191	111 - 0.0032		193 - 0.0860	132 - 0.0605	159 - 0.1051
259 - 0.1721		124 - 0.0660	168 - 0.0316	154 - 0.1083	143 - 0.1051	114 - 0.0828		195 - 0.0287	134 - 0.0350	161 - 0.0605
261 - 0.0357		126 - 0.0535	170 - 0.0570	156 - 0.0223	145 - 0.0159	117 - 0.0032		197 - 0.0096	136 - 0.0223	163 - 0.0924
262 - 0.0325		128 - 0.0472	172 - 0.0949	158 - 0.0318	147 - 0.1019			199 - 0.0064	142 - 0.0159	165 - 0.0860
263 - 0.0065		131 - 0.0220	173 - 0.0095	160 - 0.0318	149 - 0.0191			201 - 0.0159	148 - 0.0032	167 - 0.0159
264 - 0.0974		132 - 0.0157	174 - 0.0285	162 - 0.1146	151 - 0.0159				120 - 0.0064	169 - 0.0318
265 - 0.0195		137 - 0.0409	176 - 0.0728	164 - 0.0573	153 - 0.0127				123 - 0.0064	171 - 0.0446
266 - 0.0065		141 - 0.0503	178 - 0.0791	166 - 0.0064	155 - 0.0605				124 - 0.1146	183 - 0.0032
267 - 0.0065		143 - 0.0220	180 - 0.0222	168 - 0.0350	158 - 0.0223				126 - 0.6274	187 - 0.0032
268 - 0.0195		149 - 0.0094	182 - 0.0411	170 - 0.0064	162 - 0.0414				128 - 0.0732	206 - 0.0032
269 - 0.0065		153 - 0.0031	184 - 0.0443	172 - 0.0191	164 - 0.0096					
271 - 0.013			186 - 0.0222	174 - 0.0127	166 - 0.0127					
272 - 0.0032			188 - 0.0791	176 - 0.0159	168 - 0.0096					
273 - 0.0032			190 - 0.0063	178 - 0.0096	170 - 0.0255					
274 - 0.0065			192 - 0.0127	180 - 0.0032	172 - 0.0096					
275 - 0.0909			194 - 0.0348	182 - 0.0287	174 - 0.0255					
276 - 0.0487			196 - 0.0190	186 - 0.0223	176 - 0.0127					
278 - 0.0162			198 - 0.0601	188 - 0.0064	182 - 0.0096					
279 - 0.0065				192 - 0.0032	184 - 0.0223					
280 - 0.0292				194 - 0.0064	187 - 0.0127					
281 - 0.0162				196 - 0.0096						
282 - 0.0065				198 - 0.0096						
283 - 0.0162				200 - 0.0032						
284 - 0.0065				207 - 0.0127						

Homozygote:

$$P(A_rA_i \mid A_rA_i) = \frac{[\theta + (1-\theta)p_i]}{f + (1-f)[\theta + (1-\theta)p_i]} f^2 + 2f(1-f)\frac{[2\theta + (1-\theta)p_i]}{(1+\theta)} + (1-f)^2 \frac{[2\theta + (1-\theta)p_i][3\theta + (1-\theta)p_i]}{(1+\theta)(1+2\theta)} \quad (1)$$

Heterozygote:

$$P(A_rA_j \mid A_rA_j) = 2(1-f)\frac{[\theta + (1-\theta)p_i][\theta + (1-\theta)p_j]}{(1+\theta)(1+2\theta)} \quad (2)$$

Profile frequency/Random match probability (RMP)

FINDINGS AND CONCLUSIONS

1) Logs 1, 5 and 8 had a similar profile and this profile was similar with the profile of Stump 4

Conclude that Logs 1, 5 and 8 were contributed from a same tree and these three logs were originated from Stump 4 (RMP: 1 in 5.99 x 10^6)

FINDINGS AND CONCLUSIONS

2) Logs 6, 10 and 12 had a similar profile and this profile was similar with the profile of Stump 6

Conclude that Logs 6, 10 and 12 were contributed from a same tree and these three logs were originated from Stump 6 (RMP: 1 in 3.61 x 10^6)

FINDINGS AND CONCLUSIONS

3) Logs 2, 3, 7, 9, 11 and 13 had a similar profile and this profile cannot be matched with any profiles of the six stumps

4) None of the 13 logs can be matched to Stump 1, 2, 3 & 5 – the logs were not sampled in this investigation

SUMMARY

Provide direct forensic evidence to prove that the logs had been illegally harvested from this Forest Reserve

CASE STUDY 2

- **On 3 August 2016, a request was received to investigate a forest offense case involving the felling of a Chengal tree in a Wildlife Reserve**

UNIT PENGURUSAN DAN PERLINDUNGAN REZAB KRAU
JABATAN PERLINDUNGAN HIDUPAN LIAR DAN TAMAN NEGARA (PERHILITAN)
SEMENANJUNG MALAYSIA
BUKIT RENGIT, 28500 LANCHANG PAHANG DARUL MAKMUR
TEL : 09-2803314 FAX : 09-2803332
E-mail : rezabkrau@wildlife.gov.my Laman Web : www.wildlife.gov.my

PERHILITAN

Rujukan Tuan :
Rujukan Kami : JPHL&TN(PHG) 60-5/2 IP No. 69/2016 (03)
Tarikh : 03 Ogos 2016

Ketua Pengarah,
Institut Penyelidikan Perhutanan Malaysia,
52109 Kepong,
Selangor Darul Ehsan,

YBhg. Dato',

PERMOHONAN UNTUK MENDAPATKAN PERKHIDMATAN BAGI MENJALANKAN ANALISA SAMPEL DNA BAGI BARANG KES

Dengan hormatnya, saya diarah merujuk kepada perkara diatas,

2. Adalah dimakluman YBhg. Dato', Jabatan ini sedang menyiasat berkenaan satu kes kesalahan dibawah Akta Pemuliharaan Hidupan Liar Tahun 2010 [Akta 716] yang melibatkan pencerobohan kawasan Rezab Hidupan Liar Krau yang berlaku pada 24 Julai 2016.

3. Sehubungan dengan itu, bagi melengkapkan kertas siasatan berkaitan kes tersebut. Jabatan ingin memohon bantuan dari pihak YBhg. Dato' untuk mendapatkan khidmat pakar dan analisa bagi mengesahkan sampel DNA barang kes.

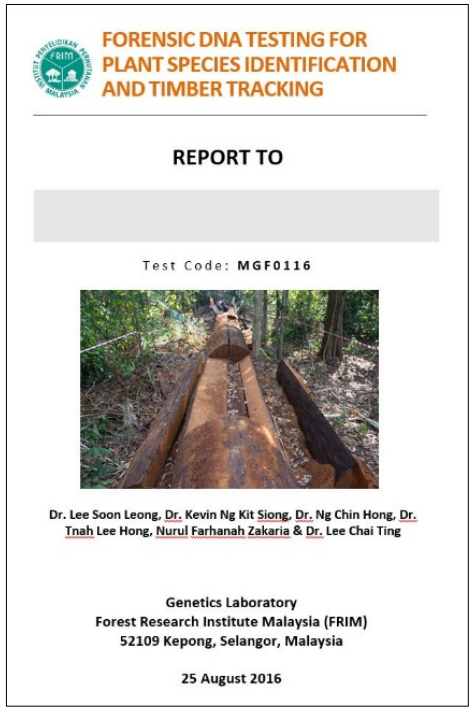
4. Untuk maklumat YBhg. Dato' kes ini akan disebut di mahkamah pada 24 Ogos 2016. Bagi melicinkan urusan berkenaan pihak YBhg. Dato' boleh berhubung terus dengan Pegawai Penyiasat iaitu En. Arsir Bin Abdul, di talian 013-4037541.

5. Segala kerjasama dan pertimbangan YBhg. Dato' amat dihargai.

Sekian, terima kasih.

"BERKHIDMAT UNTUK NEGARA"

ARSIR BIN ABDUL
Pegawai Penyiasat
Jabatan PERHILITAN



FORENSIC DNA TESTING FOR PLANT SPECIES IDENTIFICATION AND TIMBER TRACKING

REPORT TO

Test Code: **MGF0116**

Dr. Lee Soon Leong, Dr. Kevin Ng Kit Siong, Dr. Ng Chin Hong, Dr. Tnah Lee Hong, Nurul Farhanah Zakaria & Dr. Lee Chai Ting

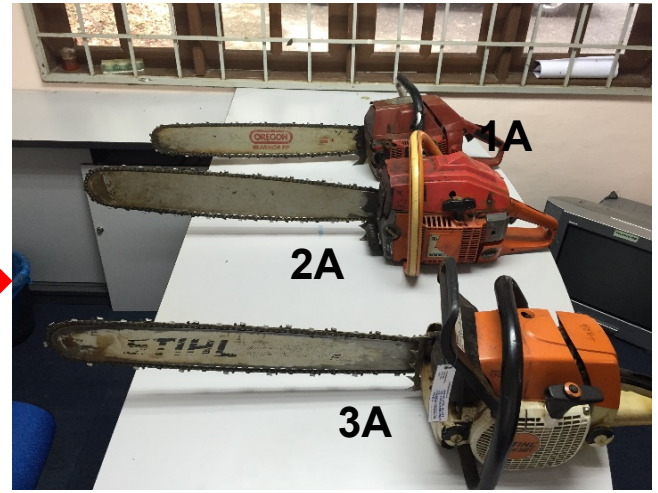
Genetics Laboratory
Forest Research Institute Malaysia (FRIM)
52109 Kepong, Selangor, Malaysia

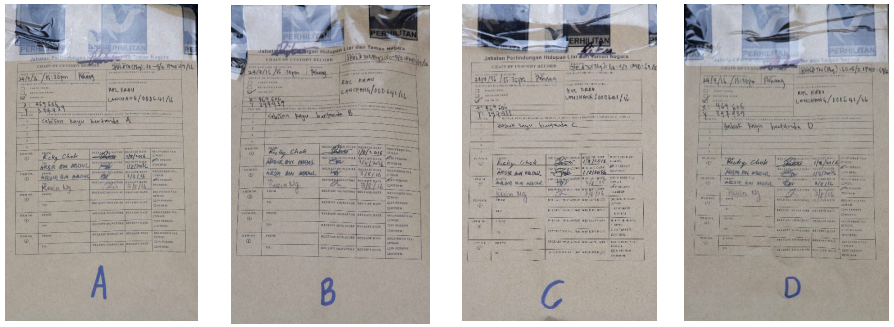
25 August 2016

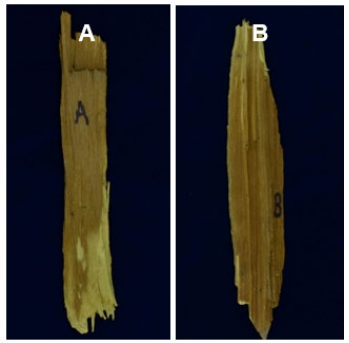

OREGON
STIHL

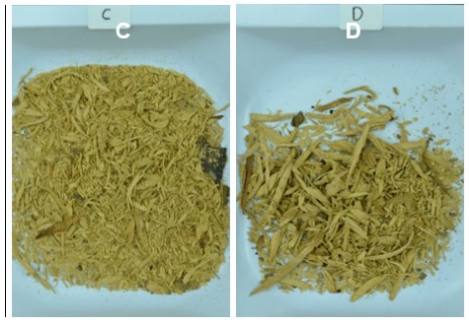

Akta Pemuliharaan
Hidupan Liar 2010
[Akta 716]
Kimwipes
WILDLIFE
FORENSIC

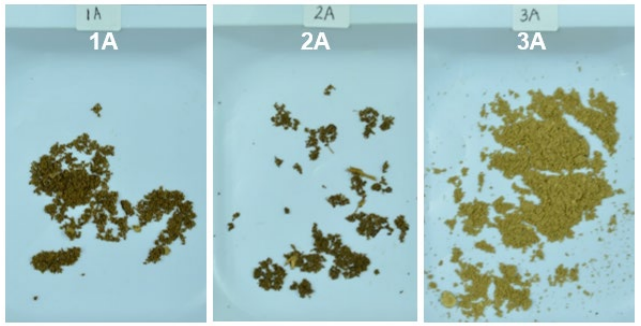
FORENSIC ACQUISITION











FINDINGS AND CONCLUSIONS

We managed to obtain consistent DNA profiles for samples A, B, C, D and 3A but not for samples 1A and 2A

The DNA extracted from samples 1A and 2A were of low quality as these samples might have been exposed to chemical which can cause DNA disintegration

FINDINGS AND CONCLUSIONS

For the samples A, B C, D and 3A, the analysis showed that all the samples shared a similar DNA profile

Sample	*Nhe*004	*Nhe*005	*Nhe*011	*Nhe*015	*Nhe*018	*Hbi*161	*Shc*03
A	259/259	114/114	170/170	144/146	137/145	101/107	130/130
B	259/259	114/114	170/170	144/146	137/145	101/107	130/130
C	259/259	114/114	170/170	144/146	137/145	101/107	130/130
D	259/259	114/114	170/170	144/146	137/145	101/107	130/130
3A	259/259	114/114	170/170	144/146	137/145	101/107	130/130

By using the FRIM's Individual Identification Database of Chengal, we can conclude that samples A, B, C, D and 3A were originated from a same tree with a RMP of 1 in 9.330×10^6

SUMMARY

Provide direct forensic evidence to prove that CHAINSAW 3A had been used to cut the Chengal tree

CASE STUDY 3

On 23 Nov 2016, a request was received from State FD to investigate a forest offense which took place in Oct 2016

TESTING SERVICES
DNA FORENSICS FOR TIMBER TRACKING

REPORT TO

Test Code: MGF0117

Dr. Lee Soon Leong, Dr. Kevin Ng Kit Siong, Dr. Ng Chin Hong, Dr. Tnah Lee Hong, Nurul Farhanah Zakaria & Dr. Lee Chai Ting

Genetics Laboratory
Forest Research Institute Malaysia (FRIM)
4 Jan 2017

Neobalanocarpus heimii


KILANG PAPAN SAM LIM SDN. BHD.
興板廠有
(COMPANY NO
SIMPANG PERTA
0048, 4920258


森林興
板廠
SAM LIM SAWMILL


KIMTECH
Kimwipes
Kimberly-Clark


Kimwipes

SUMMARY

Forest Reserve

Logging license not issued

TESTING SERVICES
DNA FORENSICS FOR TIMBER TRACKING

REPORT TO

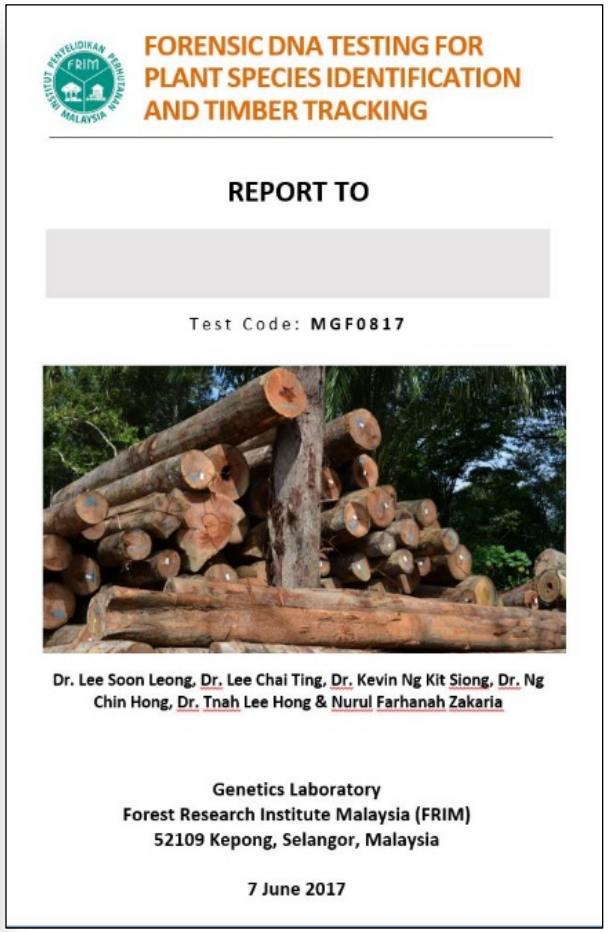
Test Code: MGF0117

Dr. Lee Soon Leong, Dr. Kevin Ng Kit Siong, Dr. Ng Chin Hong, Dr. Tnah Lee Hong, Nurul Farhanah Zakaria & Dr. Lee Chai Ting

Genetics Laboratory
Forest Research Institute Malaysia (FRIM)
4 Jan 2017

CASE STUDY 4

- **On 17 Mar 2017, a request was received to investigate a forest offense case involving the felling of valuable timber trees, inclusive of Kempas in a Wildlife Reserve**



FORENSIC DNA TESTING FOR PLANT SPECIES IDENTIFICATION AND TIMBER TRACKING

REPORT TO

Test Code: **MGF0817**

Dr. Lee Soon Leong, Dr. Lee Chai Ting, Dr. Kevin Ng Kit Siong, Dr. Ng Chin Hong, Dr. Tnah Lee Hong & Nurul Farhanah Zakaria

Genetics Laboratory
Forest Research Institute Malaysia (FRIM)
52109 Kepong, Selangor, Malaysia

7 June 2017


Samples from another 170 logs


PERHILITAN


Samples from another 22 stumps


Stump S12

SUMMARY

170 logs

Wildlife Reserve

22 stumps

FORENSIC DNA TESTING FOR PLANT SPECIES IDENTIFICATION AND TIMBER TRACKING

REPORT TO

Test Code: **MGF0817**

Dr. Lee Soon Leong, Dr. Lee Chai Ting, Dr. Kevin Ng Kit Siong, Dr. Ng Chin Hong, Dr. Tnah Lee Hong & Nurul Farhanah Zakaria

Genetics Laboratory
Forest Research Institute Malaysia (FRIM)
52109 Kepong, Selangor, Malaysia

7 June 2017

Submitted as exhibit P6 in the case of summon at the Sessions Court and the accused had pleaded guilty to the charge and fined on July 2017

CASE STUDY 5

- **On 9 Oct 2015, a request was received from a timber trading company in Singapore for wood species authentication.**

TIMBER TRADING COMPANY

Claimed supplying Chengal from Malaysia for the swimming pool decking

TESTING SERVICES
DNA FORENSICS FOR WOOD SPECIES AUTHENTICATION

REPORT TO

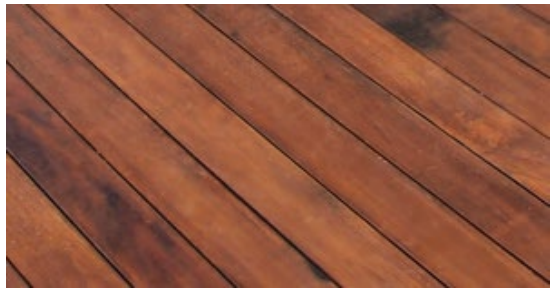
Test Code: **MGF0215**

Dr. Lee Soon Leong, Dr. Tnah Lee Hong, Dr. Kevin Ng Kit Siong, Nurul Farhanah Zakaria, Dr. Ng Chin Hong & Dr. Lee Chai Ting

Genetics Laboratory
Forest Research Institute Malaysia (FRIM)
5 November 2015

CONSTRUCTION COMPANY

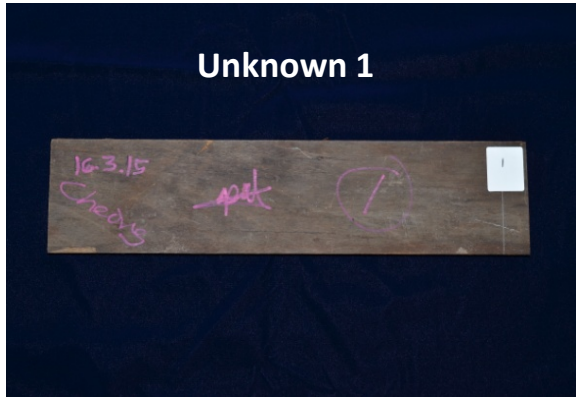
Accused not using Chengal but Giam or Balau for the swimming pool decking

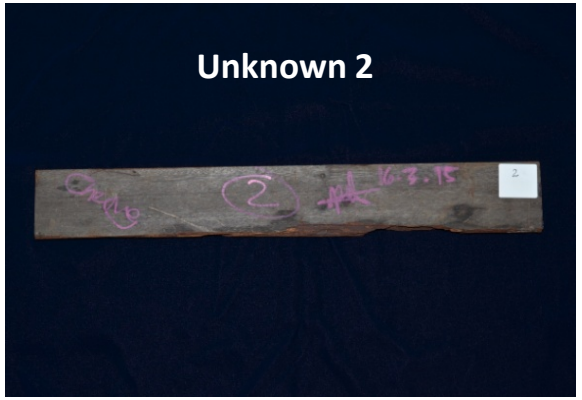


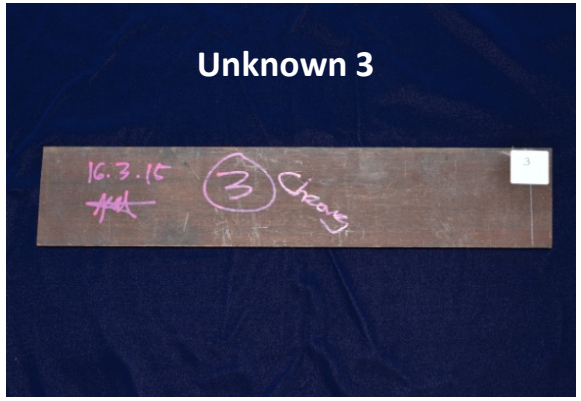
A legal case due to a dispute between the timber trading company and the construction company on the usage of wood for swimming pool decking

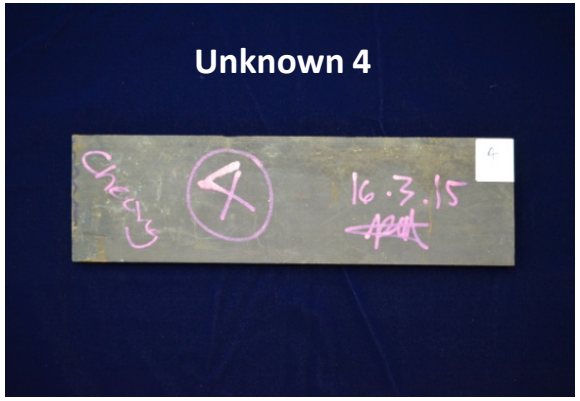
FORENSIC ACQUISITION

Four pieces of sawn wood, suspected to be Chengal were received from the company for species authentication. The samples were designated as Unknown 1, 2, 3 and 4

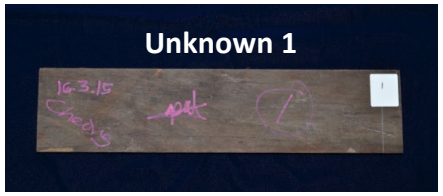


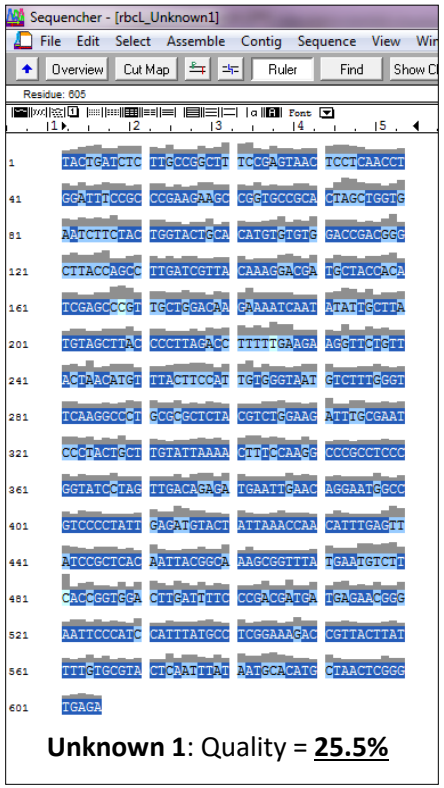




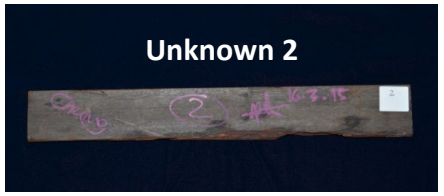


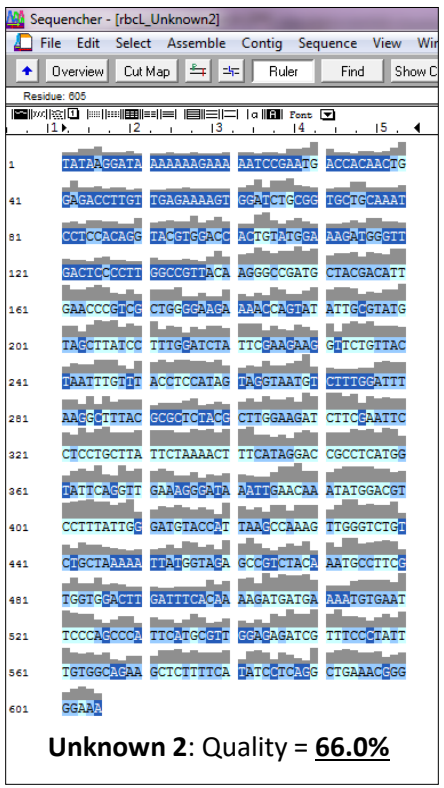
FINDINGS AND CONCLUSIONS



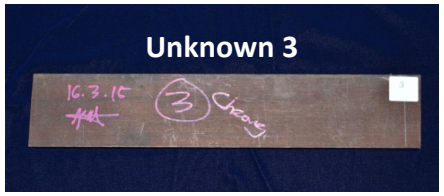


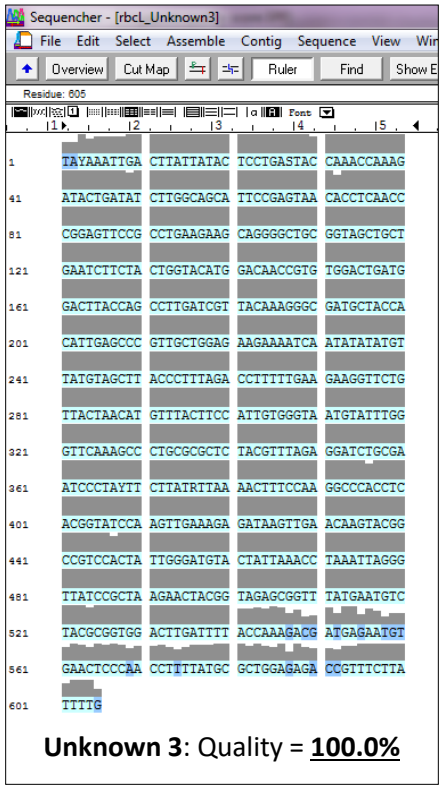
Unknown 1: Quality = **25.5%**



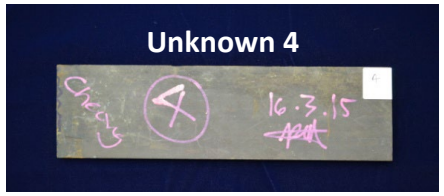


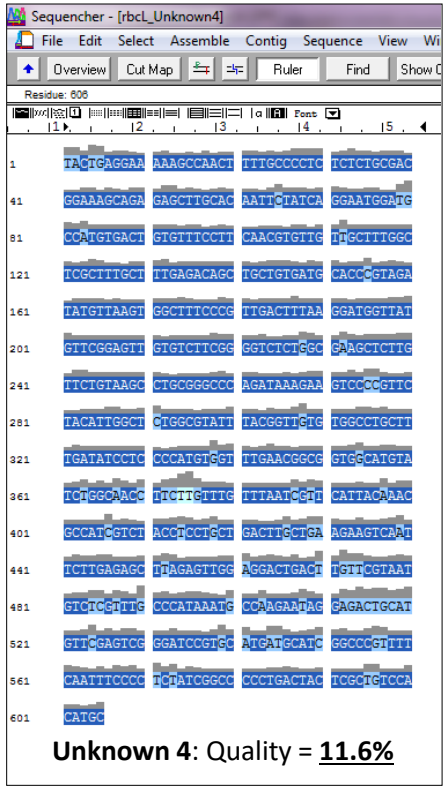
Unknown 2: Quality = **66.0%**





Unknown 3: Quality = **100.0%**

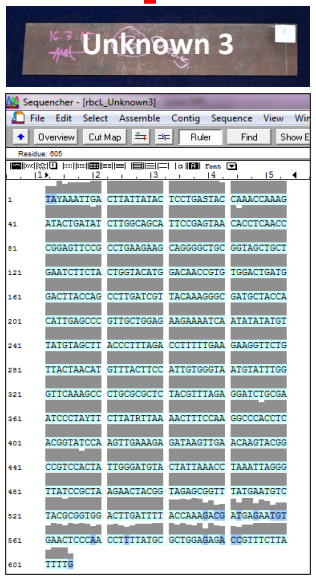




Unknown 4: Quality = **11.6%**

Quality DNA sequences for Unknown 3 but not for Unknown 1, 2 and 4

FINDINGS AND CONCLUSIONS

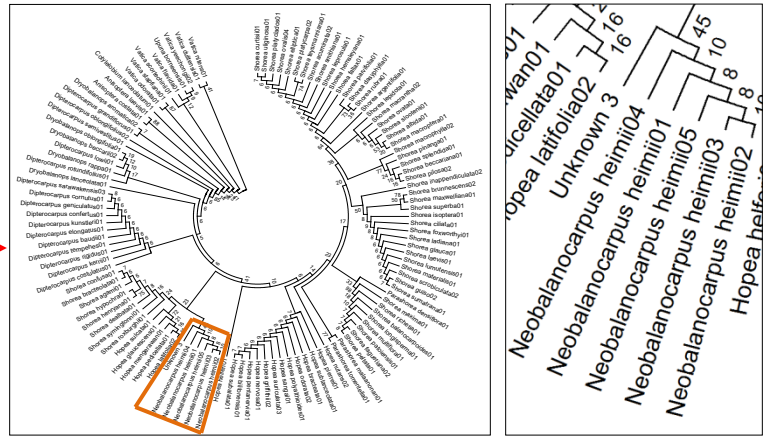


Similarity BLAST

Query ID	Subject ID (Trade name)	% identity	Mismatches
Unknown 3	*Neobalanocarpus heimii* 01 (Chengal)	100.00	0
Unknown 3	*Neobalanocarpus heimii* 02 (Chengal)	100.00	0
Unknown 3	*Hopea griffithii* (Merawan)	99.17	5
Unknown 3	*Hopea nervosa* (Merawan)	99.17	5
Unknown 3	*Shorea bracteolate* (White meranti)	99.17	5
Unknown 3	*Hopea helferi* (Giam)	99.01	6
Unknown 3	*Hopea polyalthioides* (Giam)	99.01	6
Unknown 3	*Shorea laevis* (Balau)	98.84	7

100% identity with Chengal

Maximum parsimony analysis



Well-supported clade with Chengal

SUMMARY

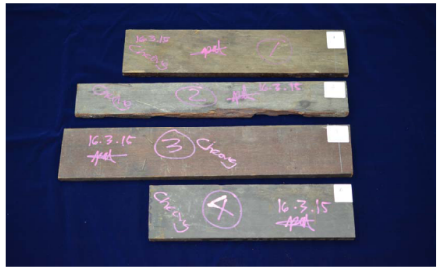
TIMBER TRADING COMPANY

Claimed supplying Chengal from Malaysia for the swimming pool decking

TESTING SERVICES
DNA FORENSICS FOR WOOD SPECIES AUTHENTICATION

REPORT TO

Test Code: **MGF0215**

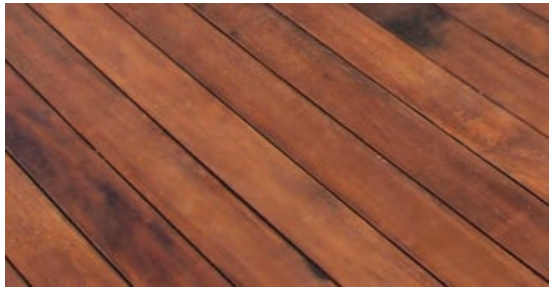


Dr. Lee Soon Leong, Dr. Tnah Lee Hong, Dr. Kevin Ng Kit Siong, Nurul Farhanah Zakaria, Dr. Ng Chin Hong & Dr. Lee Chai Ting

Genetics Laboratory
Forest Research Institute Malaysia (FRIM)
5 November 2015

CONSTRUCTION COMPANY

Accused not using Chengal but Giam or Balau for the swimming pool decking



The DNA evidence together with wood anatomy evidence have been used in court to counter the accusations by the construction company

SUMMARY

Defendant: Dr Lee Soon Leong: 1st:
IN THE STATE COURTS OF THE REPUBLIC OF SINGAPORE

Between

.. Plaintiff

And

.. Defendant

AFFIDAVIT OF EVIDENCE-IN-CHIEF

I, **DR LEE SOON LEONG** (Malaysia Passport No. A36459368) care of 52109 Kepong, Selangor, Malaysia, do affirm and say as follows:-

1. I am the head of Genetics Laboratory, Forest Biotechnology Division of the Forest Research Institute Malaysia ("**FRIM**"). I am duly authorised to make this Affidavit on the behalf of the Defendant.

2. Insofar as the matters deposed to herein, unless otherwise indicated or where the context otherwise suggest, they are based on my personal knowledge and/or documents in my possession, and are true. Insofar as the facts deposed to are not within my personal knowledge, they are true to the best of my knowledge, information and belief.

3. I have been appointed by the Defendant to prepare a Report as an expert, to identify the species of timber in respect of the timber strips provided to us by the Defendant.

4. Copies of the following are exhibited hereto marked "**LSL-1**":-

4.1 my Report to for DNA Forensics for Wood Species dated 05.11.2015 (at pages 4 to 10);

4.2 my Report to for DNA Forensics for Wood Species dated 03.02.2017 (at pages 11 to 16);

4.3 my Report to for DNA Forensics for Wood Species dated 14.04.2017 (at pages 17 to 22); and

4.4 my curriculum vitae (at pages 23 to 30).

5. In preparing my Reports and making this affidavit, my attention has been drawn to my obligation as an Expert Witness by the Defendant's solicitors. I confirm that the Reports are mine. I accept full responsibility for the same and in giving these Reports and making this affidavit, I accept that my duty is to the Honourable Court and overrides any obligation to the Defendant.

Affirmed by the abovenamed)
DR LEE SOON LEONG)
at Malaysia on this - 4 MAY 2017)
day of May 2017)

DR LEE SOON LEONG

Before me

A Notary Public

NOTARY PUBLIC
PALAKRISHNAN
A/L R. SUPPIAH
PN/WKL/HQ/25/10/2007
Expiry Date: 30-10-2017
WP. KUALA LUMPUR MALAYSIA

PALAKRISHNAN A/L R. SUPPIAH
NOTARY PUBLIC
Lot G-AB, Ground Floor, Sam Mansion
Jalan Akar / Jalan Tuba
Off Jalan Kampung Attap
50460 Kuala Lumpur
Tel: 03-22607624/5 Fax: 03-2260 6430
Email: palalaw49@gmail.com

- 4 MAY 2017

This Affidavit on filed on behalf of the Defendant.

Testify in the State Courts of the Republic of Singapore

CASE STUDY 6

Two pieces of sawn wood suspected to be *Dipterocarpus* species (KERUING) was received from a timber trading company on 26 Sep 2017.

SUMMARY

TESTING SERVICES
DNA FORENSICS FOR WOOD SPECIES AUTHENTICATION

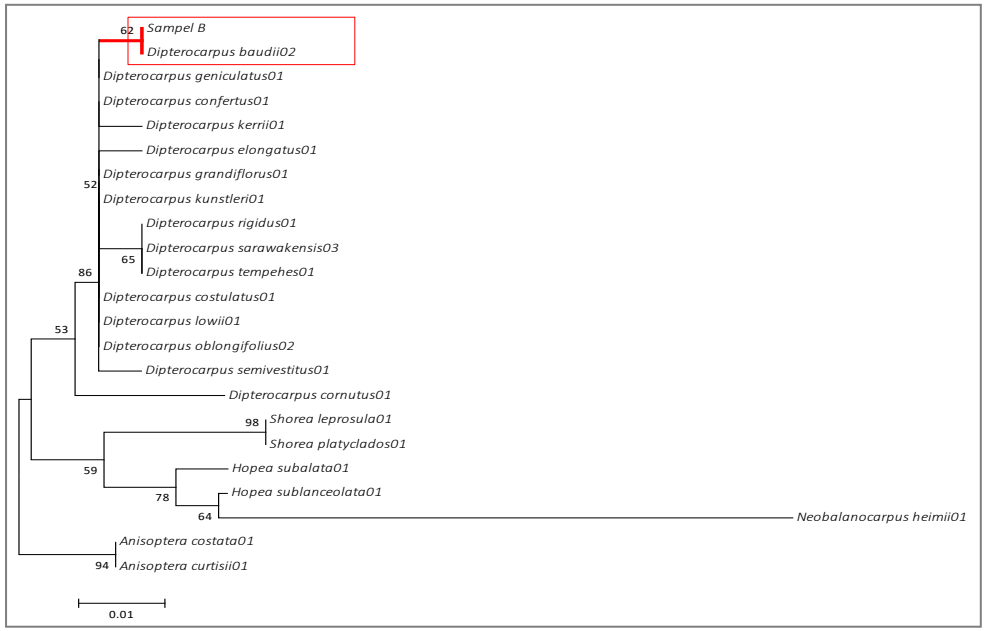
REPORT TO

Test Code: **MGF0917**

Dr. Lee Soon Leong, Dr. Tnah Lee Hong, Dr. Kevin Ng Kit Siong,
Nurul Farhanah Zakaria, Dr. Ng Chin Hong & Dr. Lee Chai Ting

Genetics Laboratory
Forest Research Institute Malaysia (FRIM)
11 October 2017

Query ID	Subject ID	% identity
Sample B	*Dipterocarpus baudii*	100.00
Sample B	*Dipterocarpus grandiflorus*	99.56
Sample B	*Dipterocarpus geniculatus*	99.56
Sample B	*Dipterocarpus oblongifolius*	99.12



Samples A & B belong to species
Dipterocarpus baudii

CONCLUSION

Based on our experience in Malaysia, we can conclude that DNA technologies are extremely useful for timber tracking and identification

We are looking forward to work with stakeholders, industries, GTTN, IAWA and scientific communities to ensure sustainable utilisation of tropical forest resources; to conserve tropical forest resources for our future generations

THANK YOU FOR YOUR ATTENTION