

Research project of counterparts funded at IPB

Name	Counterpart	Title
Iskandar Z Siregar	Z02	Developing DNA barcoding markers for Shorea species

Background

Shorea spp are ecologically and commercially important tropical forest dipterocarp tree species in Indonesia. However, it is still a challenge to correctly identify species in the field because the species are morphologically similar at the seedling stage. I therefore carried out two main activities. One was identifying endangered Dipterocarps in Harapan Rainforest, Jambi using DNA Barcoding ITS2 (Chen et al. 2010). The other was identifying three of the main *Shorea* species in Harapan Rainforest, Jambi using ITS1 and ITS2 (Essene 2014). This study was also part of the *Shorea* project.

The OBJECTIVE of this research was to develop DNA barcoding markers able to reliably discriminate between Shorea species in Harapan Rainforest, Jambi, Sumatra, Indonesia using Internal Transcribed Spacers (ITS).

Approach

DNA samples were taken from the leaves of five endangered dipterocarps (*Shorea accuminata, S. guiso, S. leprosula, Hopea mangarawan, and H. sangal*) in Harapan Rainforest, Jambi Province, Indonesia. ILTS2 was then amplified from the DNA (Chen et al. 2010). The DNA sequences were then used to construct phylogenetic trees using Neighbour Joining (NJ) clustering.

Further, DNA sample were collected from three *Shorea* species i.e *S. ovalis, S. leprosula*, and *S. guiso*, from Harapan Rainforest, Jambi Province, Indonesia. The ILTS1 and ILTS2 were then amplified from the DNA samples. The edited sequences were assigned as barcodes to a particular taxon by comparing them with the nucleotide sequences in BLAST, NCBI.

Results and Conclusions

The phylogenetic tree constructed using ITS2 showed that ITS2 successfully differentiated *Hopea* species from *Shorea* species (figure 1). This finding indicated that ITS2 loci differentiated between these dipterocarp genera. However, it was not possible to differentiate between the three Shorea species.

ITS1 successfully identified *Shorea ovalis* at the species level, and *Shorea guiso* and *Shorea ovalis* at the genus level. While ITS2 was unable to identify the three *Shorea* species at the species level, it was able to separate the two genera. However, this study is not yet completed yet and more analysis is needed to ensure the accuracy of DNA Barcode ITS1 and ITS2 for identifying *Shorea* species. The additional analysis should include calculating the barcoding gap

and constructing a full phylogenetic tree.

This initial study showed that ITS2 (Chen at al. 2010) successfully separated Hopea and Shorea species suggesting that ITS2 was able to differentiate dipterocarps at genus level. Likewise, the preliminary study of Shorea species using ITS1 (Essene 2014) showed that the loci successfully identified Shorea ovalis at the species level and Shorea guiso and Shorea ovalis at the genus level and that ITS2 (Essene 2014) was unable successfully to identify the three Shorea species at the species level but was able successfully to identify them at the genus level.



Figure 1. Phylogenetic tree based on ITS2. HH: Hutan Harapan/Harapan Rainforest, OG: out group, cf.: confirmation.

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