Research projects of counterparts funded at IPB University in 2021

Name	Counterpart	Title
Iskandar Z. Siregar, Sri Rahayu, Ulfah Juniarti Siregar, Essy Harnelly, Fifi Gus Dwiyanti, Bambang Irawan, Muhammad Majiidu	**B14**	Genetic diversity of forest reproductive materials (FRM) produced from seeds (*Archidendron pauciflorum* Benth.) and clone (*Peronema canescens* Jack.)

Background and Objectives

The success of ecosystem restoration through a variety of planting activities can be influenced by several factors, one of which is the planting material. The genetic diversity of seed or other reproductive materials needs to be studied since it is crucial to determine success rate of plantation. Jalonen *et al.* (2017) found that about 40 % of forest plants reproductive materials originated from fragmented populations with insufficient quality. Considering the importance of trees growing in the *EFForTS-BEE* plots as future seed sources, we studied the reproductive materials from two species as examples. High quality Jengkol seeds and Sungkai clones are needed with adequate genetic diversity for future use in ecosystem restoration. This objective of this study is to determine the genetic diversity of Jengkol and Sungkai on *EFForTS-BEE* experimental plots.

Methods

Novel microsatellite markers (SSR) were developed based on short read sequence data. The following step of SSR analysis: i) DNA extraction by the CTAB Method (Doyle & Doyle, 1990), ii) SSR primer amplification (Ide *et. al.*, 2014), and iii) Fragment analysis based on ABI Genetic Analyzer 3500.

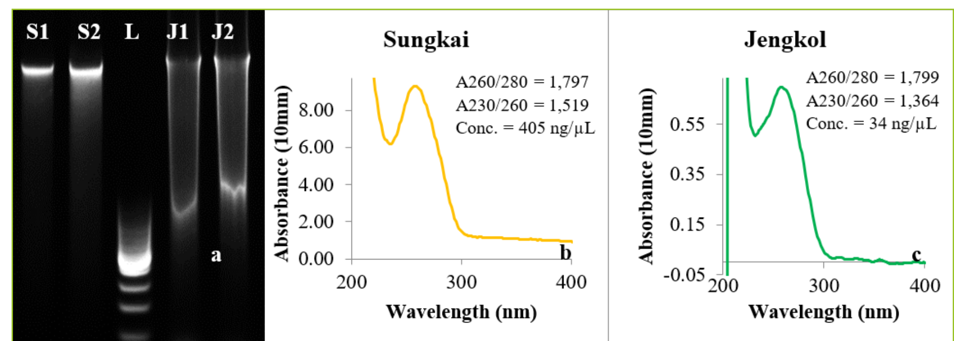

Figure 1. DNA extraction results. DNA electrophoresis (a); DNA quantity and quality of Sungkai (b); DNA quantity and quality of Jengkol; Sungkai (S); Jengkol (J); DNA Ladder 100 bp (L); Concentration (Conc.)

DNA from Sungkai and Jengkol were successfully extracted using the CTAB method. The DNA band in the gel electrophoresis is shown in figure 1a. The DNA concentration of Sungkai was 405 ng/µL and had a purity of 1.797 for A260/280 and 1.519 for A230/260 as shown in figure 1b, respectively. In contrast, The DNA Jengkol concentration was lower, 34 ng/µL, and had a purity of 1.799 for A260/280 and 1.364 for A230/260 as presented in figure 1c, respectively. It is sufficient for SSR-PCR Amplification. The specific SSR primers of Sungkai and Jengkol were designed using whole-genome data sequences.

Short read sequences

Data analysis resulted in sequence information as shown in tables 1 and 2.
According to table 4, 10.055 SSR primer candidates were founded in 9.460 sequences of Jengkol, while for Sungkai, more SSR primer candidates included 42.728 in 32.121 sequences. Of these candidates, we tested 20 loci for Sungkai and 20 loci for Jengkol using the PCR amplification assay as shown in figures 2 and 3.
Figure 56 shows that almost all SSR primer candidates of Sungkai were successfully amplified, and only one locus failed to be amplified. On the other side, all SSR primer candidates of Jengkol were successfully amplified at an annealing temperature of 60 °C. These SSR primers can be used for polymorphism testing on the respective FRMs.

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Table 1. Sequence data information of Jengkol and Sungkai

Species	N of sequences examined	N of examines sequences (bp)	N of identified SSRs	N of SSR containing sequences	N of sequences containing more than 1 SSR	N of SSRs present in coumpound formation
Archidendron pauciflorum	389.266	168.548.255	10.055	9.640	394	205
Peronema canescens	151.448	231.541.369	42.728	32.121	7.736	1.865

Table 2. SSR candidates ofJengkol and Sungkai

	Species	Unit size					
		2	3	4	5	6	7
Number of SSRs	*Archidendron pauciflorum*	3.771	5.403	763	62	42	14
	Peronema canescens	27.732	11.427	2.742	590	352	65

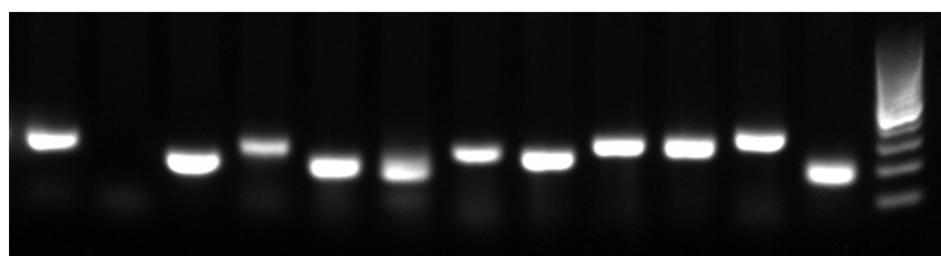

Figure 2. PCR amplification test of Sungkai. DNA Ladder 100 bp (L); locus (1-20).

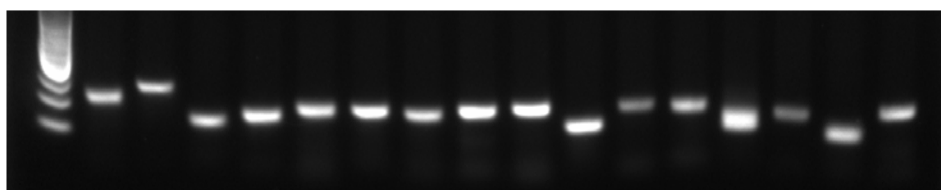

Figure 3. PCR amplification test of Jengkol. DNA Ladder 100 bp (L); locus (1-20).

References

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