

Phenotypic and Genetic Diversity Evaluation of *Sengon* (*Falcataria moluccana* (Miq.) Barneby & JW Grimes) from Solomon Provenance on Progeny Trial in Cirangsad Experimental Forest, West Java

Fifi Gus Dwiyanti*, Iskandar Zulkarnaen Siregar, Ulfah Juniarti Siregar

Silviculture Department, Faculty of Forestry and Environment, IPB University, Academic Ring Road Campus IPB Dramaga, Bogor, Indonesia 16680

Received March 17, 2021/Accepted November 9, 2021

Abstract

Initial evaluation of phenotypic variability and genetic diversity was conducted on the progeny test of 2 years-old *sengon* from Solomon provenance, which derived from nine families of mother tree and were then planted in 4 blocks in the Cirangsad Experimental Forest. Phenotypic assessment on eight traits was conducted on 36 trees using a scoring system, while genetic diversity of 15 selected individuals which represented high and low-score phenotypic traits was analyzed using 5 selected RAPD primers. The result on phenotypic assessment showed that family 3 (57.25 points) has the highest average score of phenotypic quality and family 4 has the lowest average score (7.50 points). Furthermore, genetic analysis showed that the low-scoring *sengon* population had a greater mean genetic diversity ($H_e = 0.2535$) than the high-score population ($H_e = 0.2345$). The analysis of molecular variance (AMOVA) revealed a significant genetic differences (p -value < 0.001) among high and low-score populations and the dendrogram of genetic distance revealed clustering of individuals having similar superior phenotypic against those having non-superior, indicating the selection based on phenotypes in this study had succeeded in pooling the good quantitative alleles in the selected population. This evaluation results can be used as a reference in determining the best families to produce superior *sengon* (from Solomon provenance) offspring in the future that have desired adaptability, productivity, and diversity.

Keywords: *Falcataria moluccana*, genetic diversity, phenotypic variability, RAPD, *sengon*, Solomon provenance

*Correspondence author, email: fifi_dwiyanti@apps.ipb.ac.id, tel, +62 251 8626806 fax. +62 251 8626886

Introduction

Sengon (*Falcataria moluccana* (Miq.) Barneby & J. W. Grimes) is a fast-growing tree species belonging to the Fabaceae family which is native to Moluccas, New Guinea, New Britain, and the Bismarck Archipelago and Solomon Islands (Wagner et al., 1999; Setiadi et al., 2014; Baskorowati et al., 2017). The tree can grow up to 40 m tall with a clear bole up to 20 m, a diameter of up to 100 cm, and a horizontal canopy (Krisnawati et al., 2011). This species does not require specific growing requirements and is easy to adapt, thus it is widely planted and become one of the highest priority species in industrial forest plantations and community forest in Indonesia especially in Java Island as raw material for pulp, paper, and timber that most used in people's sawmills (Priadi & Hartati, 2015; Baskorowati et al., 2017; Hidayat et al., 2017a; 2017b; Mahbub et al., 2020). The provinces of Central Java and West Java are reported to have contributed more than 60% of the total number of *sengon* trees planted by community forest (Krisnawati et al. 2011). In 2019, the number of *sengon* stands harvested has produced 62,270 m³ of wood making this species one of the 10 highest-produced timber in Indonesia after *Acacia*, *Eucalyptus*, teak, *Shorea*, pine, and mahogany (BPS, 2021).

The cultivated *sengon* is very diverse in terms of growth performance and its productivity is still low (Setiadi et al., 2014; Rostini et al., 2018), therefore a *sengon* stands that has

relatively homogeneous growth and high productivity is much sought for. One of the recommended stands is the population from Solomon provenance, which is reported to have higher productivity than local *sengon* (Ikhfan & Wijayanto, 2019), which the productivity of Solomon *sengon* can reach 3 times compared to local *sengon* which are currently found in Indonesia, especially Java (Setiadi et al., 2014). However, *sengon* from Solomon provenance is very rarely cultivated by the community in Indonesia due to lack of availability of the seeds that must be imported from Solomon Islands (Setiadi et al., 2014). Therefore, the Rumpin Seed Source and Nursery Centre (RSSNC) in collaboration with State-Owned Forestry Company (Perum Perhutani) has established the progeny trial plot in the Cirangsad Experimental Forest which is intended to build seed sources, conserve genetic material and test the suitability of land on the species collected, one of which is *sengon* from Solomon provenance. Information on genetic diversity is necessary to formulate conservation strategies and marker-based assessment can be used such as DNA fragment analysis (Loera-Sánchez et al., 2019). The use of RAPD markers rather than other molecular markers for the analysis of genetic diversity in this study was based on the inexpensive, simplicity, and ease of use of these markers. In addition, this marker is dominant and does not require specific knowledge of DNA sequences (Williams et al.,

1990; Dwivedi et al., 2018). The seed source in the Cirangsad Experimental Forest will later be designated as a provider of seeds to be used for forest rehabilitation and commercial planting.

In order to select the best offspring of *sengon* from Solomon provenance planted in the progeny trial plot in Cirangsad Experimental Forest, an initial evaluation of offspring based on phenotypic variability and genetic diversity are needed. Therefore, the objective of this study was to evaluate the phenotypic variability and the genetic diversity of the Solomon *sengon* population grown in the Cirangsad Experimental Forest through Random Amplification of Polymorphic DNA (RAPD) genetic markers. The results of this evaluation study are expected to be able to recommend the best families that can be used to supply the needs of good quality Solomon *sengon* seeds in the future.

Material and Methods

Study site and plant material This study was divided into 2 activities, namely (i) fieldwork consisting of phenotypic assessment and leaf samples collection for further DNA analysis, which were carried out in the Experimental Forest of Cirangsad, and (ii) laboratory work consisting of genetic diversity analysis using Random Amplification of Polymorphic DNA (RAPD) marker, which were conducted at the Forest Genetics Laboratory, Department of Silviculture, Faculty of Forestry, IPB University, Bogor. Geographically,

the Experimental Forest of Cirangsad located in $E 106^{\circ}33'19.0771'' - E 106^{\circ}34'02.3985''$ and $S67^{\circ}30'36.6411'' - S67^{\circ}29'33.1894''$ with an altitude of 600–700 m above sea level (Figure 1). The forest is divided into several areas based on research objectives, namely progeny test, species trial, demonstration plot and untested seed orchard. Cirangsad experimental forest area has a sloping and hilly topography. The texture of the soil is clay with a CEC of 25.02 (meq 100 g^{-1} of soil). The soil is yellowish brown in color and has a pH of 4.84. The soil nitrogen content was 0.14%, while the C-organic content was 1.30%. Then the organic matter contained in the soil is 2.23%. The climate in Cirangsad is classified as type A (Schmidt & Ferguson, 1951) with an average rainfall of 2,000–2,500 mm year^{-1} .

Assessment on phenotypic variability was carried out on a 2-years old *sengon* from Solomon provenance tree stand planted in a single tree plot with a randomized complete block design at the progeny trial area of the Cirangsad Experimental Forest. The trees are located in four blocks which each block consists of nine individual trees (Figure 2). These nine individual trees are offspring from nine different mother trees (hereinafter referred to as a family), which originated from Solomon Island and were then planted in Pompok Landak Permanent Nursery located in Haurwangi Village-Bojongpicung Sub-District-Cianjur Regency-West Java Province. In total, 36 individual trees were used in this study (Table 1).

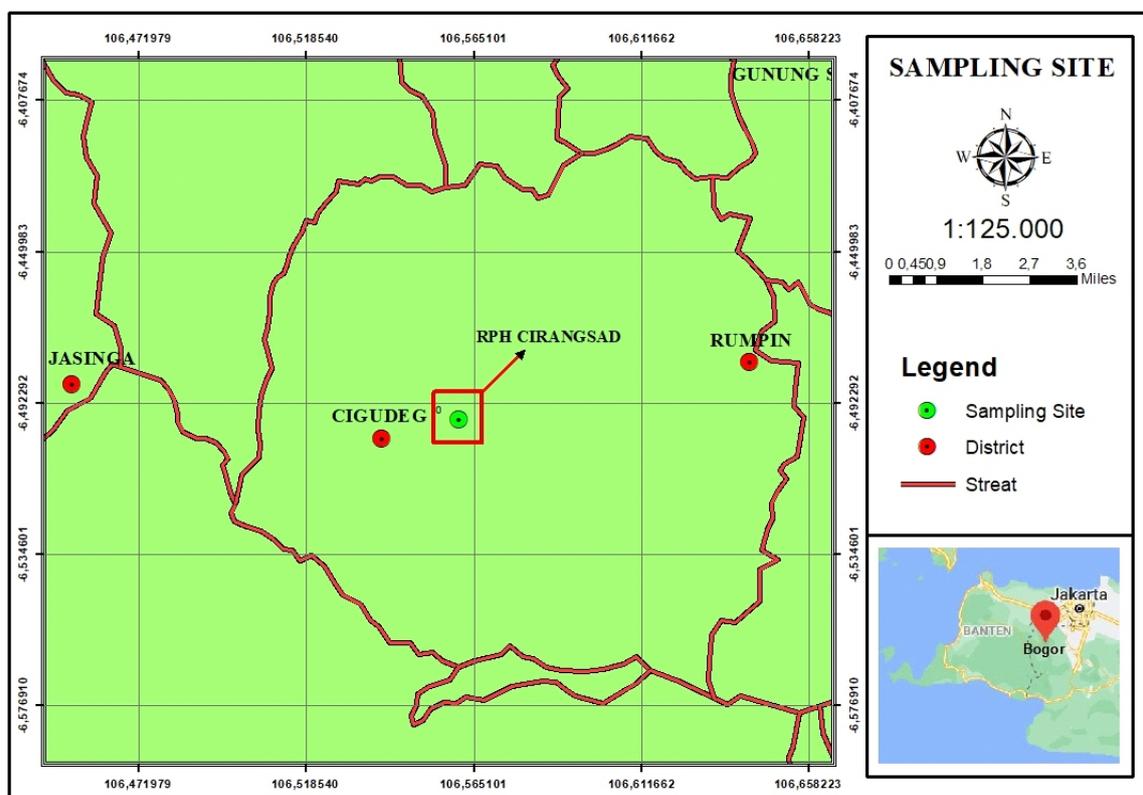


Figure 1 Map of study site in Cirangsad Experimental Forest.

Phenotypic assessment The phenotypic quality assessment of 2-years old *sengon* from Solomon provenance tree was divided into measurement of quantitative parameters and qualitative parameters, from which those parameters were selected based on desirable traits of economic interest of *sengon* tree such as stem straightness, clear bole height, disease resistance, etc. Quantitative parameters include survival percentage, tree height, clear bole height, stem diameter, branch diameter and branch angle. Meanwhile, qualitative parameters include stem shape, bark color, and resistance to pests & diseases. Tree height and branch free height were measured by 250 cm long-tree height measuring stick with a marker for every 10 cm. While, stem diameter

(diameter at breast height) and branch diameter were measured by tree caliper.

The measurement data that has been obtained was then scored based on the scoring system method with a comparison tree for several important trait according to Zobel and Talbert (1984) with some modification such as excluding crown and natural pruning parameters. In this study, 18 individuals of 2-years old common *sengon* tree and growing 4 m from the tested *sengon* tree were used as comparison trees. The comparison tree used were located near block 1 and block 3 of the tested *sengon*. Scoring on the *sengon* from Solomon provenance tree was intended to obtain a total score of all parameters measured for each individual. Each measured parameter has a specific score and the maximum total score for all parameter is 90 points (Table 2). The method of scoring analysis is by giving a rank to the average scoring rate for each family. Rank 1 shows the highest average score in the family, while ranking 9 shows the lowest average score in that family.

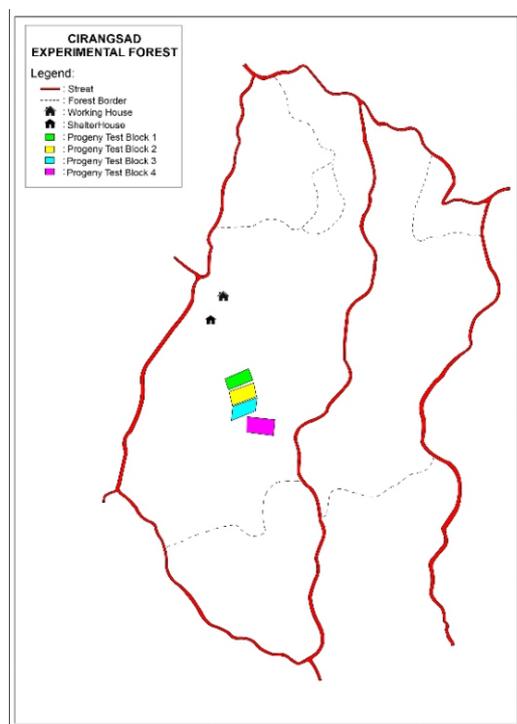


Figure 2 A sketched map of 2-years old *sengon* (from Solomon provenance) progeny trial blocks in Cirangsad Experimental Forest.

Genetic assessment Leaves from each individual *sengon* tree selected based on the phenotypic score were collected and dried in the field with silica gel for further genetic analysis. Silica gel-dried leaves were ground to a fine powder in liquid nitrogen with a mortar and pestle and subsequently used for DNA extraction. Total genomic DNA was extracted from each sampled leaf using the modified CTAB method (Weising et al., 2005).

The genomic DNA of each individual trees was amplified by Random amplified polymorphic DNA (RAPD) using five selected primers, from Operon Technologies Inc. with average size of 10 bases: OPA 05 (5' AGGGGTCTTG '3), OPA 14 (5' TCTGTGCTGG '3), OPO 10 (5' TCAGAGCGCC '3), OPU 05 (5' TTGGCGGCCT '3), and OPY 16 (5' GGGCCAATGT '3). These five primers were selected from the screening results of 23 primers kit A (OPA), B (OPB), O (OPO), U (OPU) and Y (OPY) which were successfully amplified, had clear bands and were polymorphic.

Polymerase Chain Reaction (PCR) amplification was performed in a volume of 13.5 µl, containing about 40 ng of genomic DNA, 2.5 µM of primer, and Taq DNA Polymerase (GoTaq® Green Master Mix, Promega). The reactions were

Table 1 *Sengon* (from Solomon provenance) tree codes used in this study

Family	Block			
	1	2	3	4
1	S1B1	S1B2	S1B3	S1B4
2	S2B1	S2B2	S2B3	S2B4
3	S3B1	S3B2	S3B3	S3B4
4	S4B1	S4B2	S4B3	S4B4
5	S5B1	S5B2	S5B3	S5B4
6	S6B1	S6B2	S6B3	S6B4
7	S7B1	S7B2	S7B3	S7B4
8	S8B1	S8B2	S8B3	S8B4
9	S9B1	S9B2	S9B3	S9B4

Note: S = *sengon* tree; B = block; example: S1B1 = *sengon* tree from family 1 in block 1

Table 2 Method of phenotypic quality assessment for *segon* (from Solomon provenance) trees

No	Trait	Full score (point)	Evaluation system	
			Value Scale	Score (point)
1	Stem height	10	Percentage of values above the average of control trees <5%	0
			Percentage of values above the average of control trees 5–<10%	2
			Percentage of values above the average of control trees 10–<15%	4
			Percentage of values above the average of control trees 15–<20%	6
			Percentage of values above the average of control trees 20–<25%	8
			Percentage of values above the average of control trees ≥25%	10
2	Stem diameter	20	Percentage of values above the average of control trees <5%	0
			Percentage of values above the average of control trees 5–<10%	5
			Percentage of values above the average of control trees 10–<15%	10
			Percentage of values above the average of control trees 15–<20%	15
			Percentage of values above the average of control trees ≥20%	20
3	Clear bole height	10	Percentage between clear bole height and total height <30%	0
			Percentage between clear bole height and total height <30%	2
			Percentage between clear bole height and total height <30%	4
			Percentage between clear bole height and total height <30%	6
			Percentage between clear bole height and total height <30%	8
			Percentage between clear bole height and total height <30%	10
4	Stem shape	30	Straight stem	30
			S-shaped stem	Subtract 3–20 points
			Bow-shaped stem	Subtract 3–20 points
			J-shaped stem	Subtract 2–8 points
			Spiral stem	Subtract 5–20 points
			Forked stem	Subtract 5–20 points
5	Branch diameter	5	Percentage between the diameter of the branch and the diameter of the stem where the branch is located >50%	0
			Percentage between the diameter of the branch and the diameter of the stem where the branch is located 30–50%	2
			Percentage between the diameter of the branch and the diameter of the stem where the branch is located <30%	5
6	Branch angle	5	Branch angle <50°	0
			Branch angle 50–70°	2
			Branch angle >70°	5
7	Survival rate	5	Dead tree	0
			Living tree	5
8	Pest and disease resistance	5	Unhealthy (area of signs of pest attack >20%)	0
			Healthy (area of signs of pest attack <20%)	5

carried out in a thermocycler PTC-100 Programmable Thermal Cycler (MJ Research Thermal Cycler, USA), with the use of a program following conditions: one cycle of initial denaturation at 95 °C for 10 min, then 35 cycles of denaturation at 95 °C for 1 min, annealing at 37 °C for 3 min and extension at 72 °C for 2 min, followed by a one cycle of final extension at 72 °C for 10 min. Amplified products were then separated on Agarose 2% using TAE buffer solution 1x and stained with Ethidium bromide. The electropherograms were visualized using UV Transilluminator and photographed using digital camera.

The polymorphic DNA bands were then scored to create a data matrix composed of the numerals 1 and 0 which was built on the basis of presence (1) or absence (0) of a DNA band appearing in replicates for each isolate. Furthermore, the genetic diversity indexes and parameters, including the percentage of polymorphic loci (PLP), the number of alleles observed (Na), effective allele number (Ne) (Kimura & Crow, 1964), Nei's gene diversity (Nei, 1973) (He), were calculated with POPGENE 32 (Yeh et al., 1997). For genetic similarities, the Nei's genetic distance (Nei, 1972) were

estimated using POPGENE 32 (Yeh et al., 1997) and the presence of molecular variance within and between the *segon* population structure was assessed through Analysis of molecular variance (AMOVA) by using GenAlEx 6.5 software (Peakall & Smouse, 2006). In addition, the un-weighted pair group method with arithmetic averages (UPGMA) based dendrogram was constructed using the NTSYS version 2.0 (Rohlf, 1998).

Results and Discussion

Phenotypic variability In this study, phenotypic variability of the studied *segon* was evaluated by scoring 8 phenotypic parameters on 36 individual 2-years old trees derived from 9 families. The results showed that 9 out of 36 trees died, therefore the overall full score of the parameters for these nine individuals was 0 (Table 3). The families that had a high survival rate (100%) were families 1, 2, 3, and 8. While family 4 had a lower survival rate (25%) in each block than other families indicated that individual tree from family 4 had a lack of ability to survive in low or hilly areas such as the area in the Cirangsad Experimental Forest.

The scoring results of all parameters at the family level (Table 3) showed that family 3 has the highest average score (57.25 points) and family 4 has the lowest average score (7.50 points) indicated that family 3 has superior traits in all parameters and family 4 has less superior traits in all growth parameters, especially the survival rate parameter where only 1 individual tree was still alive. Meanwhile, the score for all parameters at the individual level showed that individual tree from family 1 planted in block 2 (S1B2) received the highest score (83 points) followed by family 3 in block 3 (S3B3) and family 6 in block 3 (S6B3) which have 76 points, respectively. Additionally, individual tree from family 4 planted in block 2, 3, and 4 (S4B2, S4B3, and S4B4) received the lowest score (0 points). The results at the individual level are almost in line with the results at the family level where family 3 has a high average score and family 4 has a low average score among the other 9 families. The results of this evaluation can be used as a reference in determining the best families as seed sources as seen from the performance of the offspring.

The number of individuals of studied *sengon* that had a scoring value of ≥ 60 points was 7 individuals and the number of individuals that had a scoring value of <60 points was 28 individuals, of which there were only 8 individuals who had a score of <45 points. Based on these scoring results, 15 of the 27 living individuals in 4 blocks were selected with the highest (7 individuals) and lowest (8 individuals) scores for further genetic analysis with the RAPD marker. The selected individuals are presented in Table 4, which shows that the trees classified into the high scoring population are derive from the families 1, 3, 5, 6, 7, 8, and 9, while the trees classified into the low scoring populations are derive from

the families 1, 2, 4, 7, 8, and 9. This pattern also indicates that the mother trees from families 1, 7, 8, and 9 could produce offspring with different phenotypic performances, which may be due to outcrossing and heterozygosity.

Genetic diversity Genetic analysis of *sengon* from Solomon provenance with RAPD markers was carried out on individual trees that had been selected through phenotypic quality scoring system. The five RAPD primers used in this study showed good amplification pattern and presented a total of 63 loci (Figure 3).

The calculated genetic diversity parameters of the 2-years old studied *sengon* population were the number of alleles expected (N_a), the number of alleles observed (N_e), gene diversity (H_e) and percent polymorphic loci (PLP). The overall results of RAPD analysis of studied *sengon* population in this study showed that the expected average number of alleles (N_a) was 1.9683, the average number of alleles observed (N_e) was 1.4523, the mean value of genetic diversity (H_e) was 0.2813, and the percent polymorphic locus (PLP) was 97% (Table 5). The mean value of genetic diversity in these 2-years old studied *sengon* population provides an implicit overview of the genetic diversity of the nine parent tree families that grow in Pompok Landak Permanent Nursery. In the present study, the mean genetic diversity (H_e) of *sengon* from Solomon provenance was slightly higher when compared to the genetic diversity of the common *sengon* population ($H_e = 0.2757$) in Cianjur community forest and the genetic diversity of the *sengon* population ($H_e = 0.2349$) in nine community forests in Java Island reported by Siregar and Olivia (2012) who also used RAPD markers. In addition, this study also had a slightly

Table 3 The results of the phenotypic quality score for 2-years old *sengon* (from Solomon provenance)

Block	Scoring for 9 family of 2-years old Solomon <i>sengon</i> (point)								
	1	2	3	4	5	6	7	8	9
1	41	35	41	30	0	59	71	57	36
2	83	40	55	0	53	47	0	36	0
3	59	55	76	0	68	76	40	40	73
4	39	60	57	0	49	0	0	60	0
Average	55.50	47.50	57.25	7.50	42.50	45.50	27.75	48.25	27.25
Rank	2	4	1	9	6	5	7	3	8
Survival rate (%)	100	100	100	25	75	75	50	100	50

Table 4 List of *sengon* (from Solomon provenance) samples used for further RAPD analysis

No.	Low-score trees			No.	High-score trees		
	Family	Block	Tree code		Family	Block	Tree code
1	2	1	S2B1	1	7	1	S7B1
2	4	1	S4B1	2	1	2	S1B2
3	9	1	S9B1	3	3	3	S3B3
4	2	2	S2B2	4	5	3	S5B3
5	8	2	S8B2	5	6	3	S6B3
6	7	3	S7B3	6	9	3	S9B3
7	8	3	S8B3	7	8	4	S8B4
8	1	4	S1B4				

Note: S = *sengon* tree; B = block; example: S1B1 = *sengon* tree from family 1 in block 1

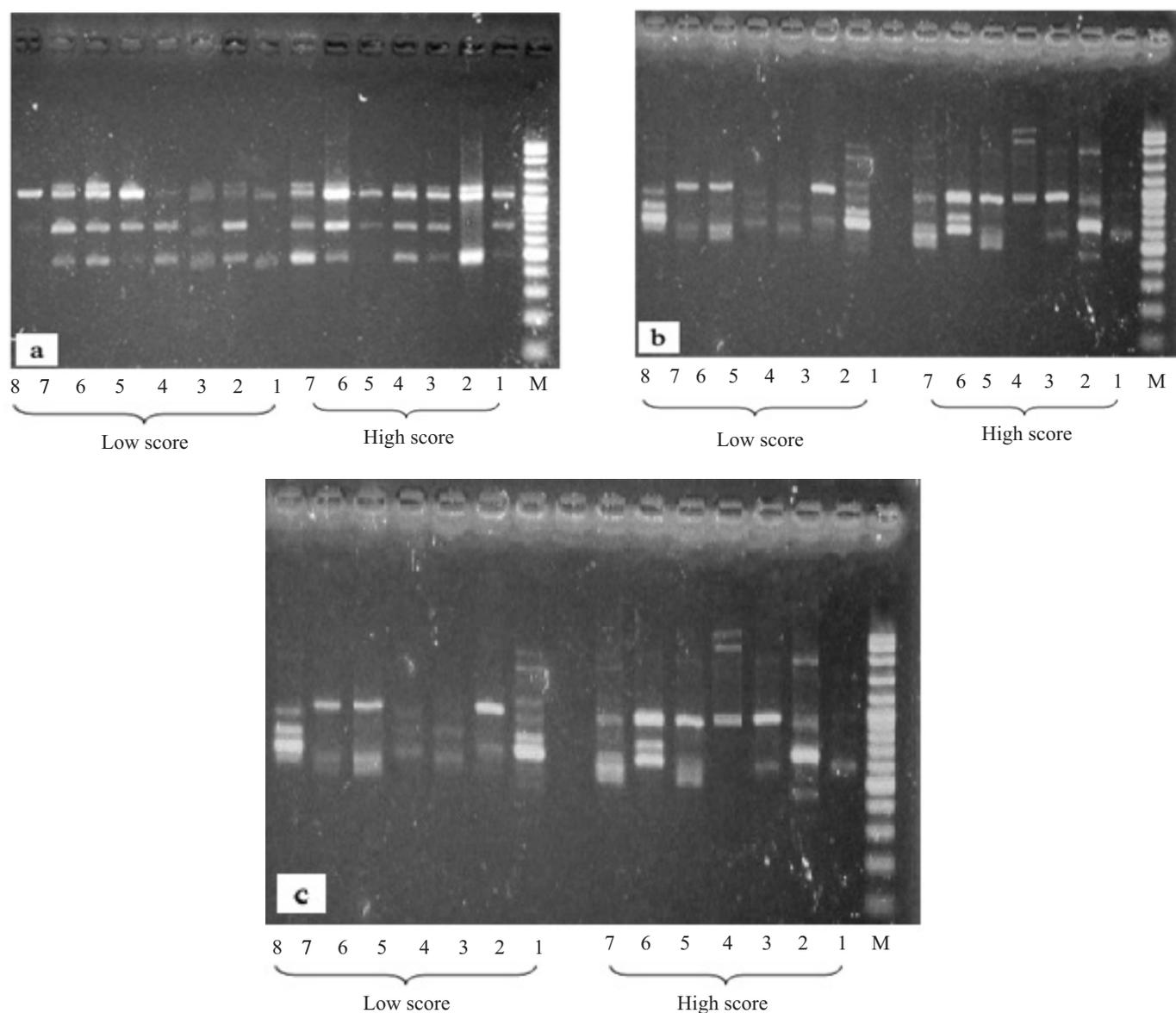


Figure 3 The example of *sengon* from Solomon provenance DNA profile using RAPD primers on 7 high-score trees and 8 low-score trees (the sample order is based on Table 4). (a) The amplified DNA of *sengon* using primer OPA-05, (b) The amplified DNA of *sengon* using primer OPA-14, and (c) The amplified DNA of *sengon* using primer OPY-16.

Table 5 Summary of genetic diversity in 2-years old *sengon* (from Solomon provenance) populations

Population	N	Na	Ne	PLP (%)	He
High score	7	1.7143 (0.4554)	1.3762 (0.3219)	71	0.2345 (0.1725)
Low score	8	1.7460 (0.4388)	1.4181 (0.3418)	75	0.2535 (0.1780)
Overall	15	1.9683 (0.1767)	1.4523 (0.3101)	97	0.2813 (0.1455)

Note: N = sample size; Na = number of alleles observed; Ne = effective allele number; He = Nei's gene diversity (Nei, 1973); and PLP = percentage of polymorphic loci. Values in parentheses are standard deviations

higher mean of genetic diversity than the *sengon* half-sib progeny population ($He = 0.244$) analyzed with the RAPD marker by Rostini et al. (2018) and genetic diversity of healthy-growing *sengon* populations ($He = 0.262$) in various locations on the Java Island which was analyzed by Lelana et al. (2018) using the RAPD marker. This pattern indicated that sufficient of the diversity source in *sengon* from Solomon provenance that grows in Pompok Landak Permanent Nursery-Haurwangi Village at this time.

In this study, genetic diversity analysis was partitioned between populations that had high phenotypic parameter scores and the other populations that had low phenotypic parameter scores (Table 5). The results of the analysis showed that the studied *sengon* population with low scores had a slightly greater mean value of genetic diversity parameters ($N_a = 1.7460$, $N_e = 1.4181$, PLP 75% and $He = 0.2535$) than the studied *sengon* population with high scores ($N_a = 1.7143$, $N_e = 1.3762$, PLP 71% and $He = 0.2345$), indicating the genetic diversity of the *sengon* from Solomon provenance having low-score population is greater when compared to the high-score population genetic diversity. The small genetic diversity in the high-score *sengon* population is expected because a selected population that has superior traits would have smaller diversity. Smaller genetics diversity means the population is getting more uniform, and if these trees will be used as plus tree in the seed orchard, the resulting offspring will have more uniform phenotypic traits as their parents.

Genetic distance The differences in genetic diversity between populations indicates the genetic distance between the two Solomon *sengon* populations. The result of the genetic distance analysis between a high-score and low-score populations (Table 6) showed that the high-score tree from family 1 in block 2 (S1B2) and low-score from family 4 in block 1 (S4B1) and also the low-score tree from family 1 in

block 4 (S1B4) and low-score from family 4 in block 1 (S4B1) have the highest genetic distance value ($d = 0.6774$, respectively) indicated that family 1 have a distant relationship with the low-score tree from family 4. Meanwhile, the low-score trees from family 8 in block 3 (S8B3) and family 7 in block 3 (S7B3) have the lowest genetic distance value ($d = 0.1358$) indicated close relationship between the two low-score families. The results of present study also revealed that the offspring derived from the same population or family do not always have close genetic distances or are in the same group (Widyastuti, 2007). For example, in this study (Table 6), it was found that the genetic distance between the studied *sengon* trees that belonged to the same family (family 9), but differed in the phenotypic trait scores, namely S9B3 (high score) and S9B1 (low score), had a large genetic distance ($d = 0.4055$). This indicates that family 9 comes from the mother tree that might be heterozygous and self crossed or might be undergoes outcrossed.

In addition, the comparison of genetic distance between high- and low-score individuals can also be described from the percentage of individuals in each genetic distance range (Table 7), where the results showed that 5.36% of the high score-low score individuals have large genetic distance (range = 0.5861–0.7361) and this was the highest percentage when compared to the percentage between individuals with the highest score (0%) or between individuals with the lowest score (3.57%) indicating high and low-score individual have more genetic distance. Moreover, 38.10% of individuals with the highest scores have a close genetic distance (range = 0.1358–0.2858) and this is the highest percentage compared to individuals with low scores or the high-low score individuals, indicating a close relationship between high score individuals. Furthermore, significant genetic differences (p -value <0.001) were detected in the analysis of molecular variance (AMOVA) among high and low-score

Table 6 Summary of mean genetic distance between *sengon* (from Solomon provenance) populations

Population	High score							Low score							
	S7B1	S1B2	S3B3	S5B3	S6B3	S9B3	S8B4	S2B1	S4B1	S9B1	S2B2	S8B2	S7B3	S8B3	S1B4
High score	S7B1	****													
	S1B2	0.3365	****												
	S3B3	0.2311	0.3589	****											
	S5B3	0.3589	0.2719	0.2390	****										
	S6B3	0.2719	0.4055	0.2513	0.2930	****									
	S9B3	0.2311	0.4543	0.3365	0.4796	0.4296	****								
	S8B4	0.1919	0.4055	0.2513	0.4296	0.3365	0.1728	****							
Low score	S2B1	0.2113	0.2113	0.3145	0.2719	0.3145	0.3145	0.2719	****						
	S4B1	0.4543	0.6774	0.4296	0.4796	0.6466	0.5322	0.4296	0.5596	****					
	S9B1	0.2930	0.3189	0.2719	0.4055	0.5596	0.4055	0.4055	0.2113	0.4543	****				
	S2B2	0.2513	0.3189	0.2719	0.4055	0.4055	0.3145	0.2719	0.1728	0.4543	0.2513	****			
	S8B2	0.2930	0.3189	0.3145	0.4543	0.5055	0.3145	0.2719	0.2113	0.4543	0.3365	0.1728	****		
	S7B3	0.2513	0.3189	0.2719	0.3145	0.4543	0.5055	0.3589	0.2513	0.4543	0.2113	0.2513	0.2930	****	
	S8B3	0.2513	0.3189	0.3145	0.3145	0.4543	0.5055	0.3589	0.2930	0.5055	0.3365	0.4296	0.4296	0.1358	****
S1B4	0.4296	0.4796	0.5055	0.5055	0.6168	0.4543	0.5055	0.3189	0.6774	0.4296	0.3365	0.3365	0.3365	0.4296	****

Note: S = *sengon* tree; B = block; example: S1B1 = *sengon* tree from family 1 in block 1

population (Table 8). AMOVA analysis comparing high and low-score population also revealed that 11% of the molecular variance existed between the population, whereas 89% was within population (Table 8).

The calculated genetic distance obtained in the present study were then used to construct a dendrogram based on UPGMA (Figure 4), which clustered the individuals and families studied. The dendrogram showed that high-score

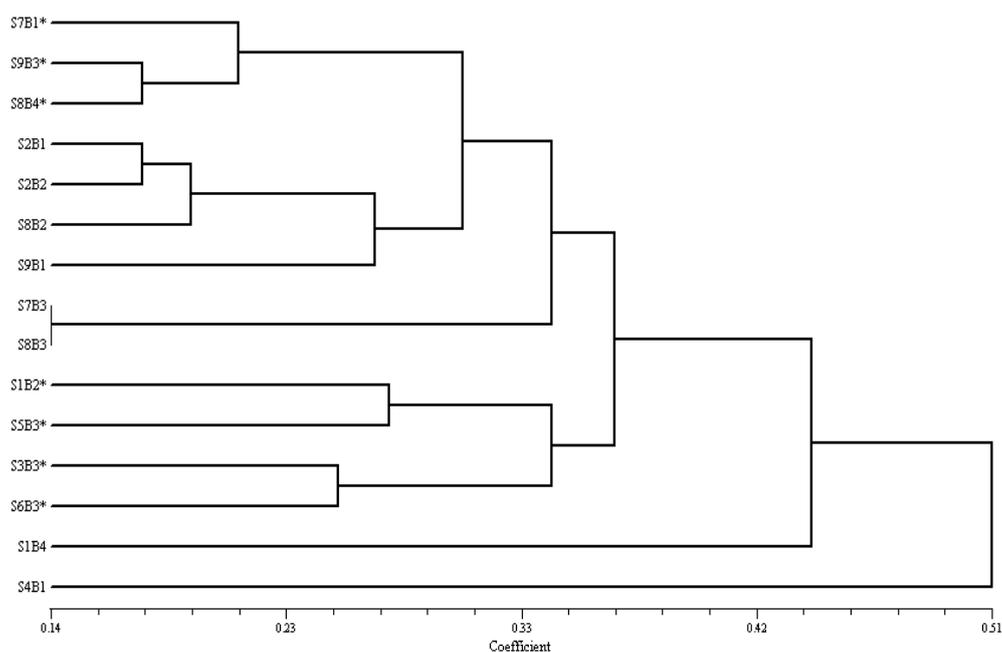
Table 7 Summary of *segon* individual percentage based on mean genetic distance range

Phenotypic score category	Mean genetic distance range	% Individu
High score–High score	0.1358–0.2858	38.10
	0.2859–0.4359	52.38
	0.4360–0.5860	9.52
	0.5861–0.7361	0.00
High score–Low score	0.1358–0.2858	21.43
	0.2859–0.4359	46.43
	0.4360–0.5860	26.79
	0.5861–0.7361	5.36
Low score–Low score	0.1358–0.2858	32.14
	0.2859–0.4359	42.86
	0.4360–0.5860	21.43
	0.5861–0.7361	3.57

Table 8 AMOVA table of molecular variation among and within high and low-score *segon* populations from GenAIEx

Source	df	Sum of squares	Mean square	Estimated varian	%	ϕ PT
Among population	1	17.415	17.415	1.142	11%	0.114***
Within population	13	115.518	8.886	8.886	89%	
Total	14	132.933		10.028	100%	

Note: df = degree of freedom, ϕ PT = recorded population genetic differentiation, and an asterisk indicates significance (p -value < 0.001).



Note: S = *segon* tree; B = block; example: S1B1 = *segon* tree from family 1 in block 1; * = high score individual tree

Figure 4 UPGMA dendrogram of genetic distance of *segon* (from Solomon provenance) families based on RAPD profiles.

tree populations tended to cluster together and do not merge with low-score tree populations. Likewise, the low-score tree population tended to form a separate cluster with the high-score tree population. This pattern suggested that even though there is no evidence that certain RAPD marker was linked or associated with the phenotypic characters observed, the RAPD marker used in this study has clustered individuals with high-scored phenotypes separately with lower scored ones, thus the selection based on phenotypes had succeeded in pooling the good quantitative alleles in the selected population.

Conclusion

The present study revealed that the family 3 has the highest average score of phenotypic quality (57.25 points) and family 4 has the lowest average score (7.50 points). This evaluation results can be used as a reference in determining the best families to produce superior *sengon* (from Solomon provenance) offspring in the future that have desired adaptability, productivity and diversity, as seen from the performance of the offspring. Furthermore, the medium level of genetic diversity ($H_e = 0.2813$) for the total *sengon* (from Solomon provenance) population in this study suggested the sufficient diversity source in *sengon* from Solomon provenance that grows in Pompok Landak Permanent Nursery. Besides that, the genetic diversity of the *sengon* from Solomon provenance having low-score population was slightly greater ($H_e = 0.2535$) when compared to the high-score population genetic diversity ($H_e = 0.2345$) indicated that the *sengon* mother tree experienced outcrossing. In addition, the comparison of genetic distance between high- and low-score individuals showed that high and low-score individual have more larger genetic distance than the high-score individuals or the low-score individuals. This pattern supported by AMOVA results that showed significant genetic differences (p -value < 0.001) among high and low-score population and also UPGMA dendrogram of genetic distance which illustrated high-score tree populations tended to cluster together and do not merge with low-score tree populations, thus the selection based on phenotypes had succeeded in pooling the good quantitative alleles in the selected population. Clustering together of individuals having similar superior phenotypic traits against those having non-superior traits indicated that those individuals have distinctive different genotype.

Acknowledgment

The authors thank Rumpin Seed and Nursery Center (RSSNC), The Directorate of Forest Tree Seed, Directorate General of Watershed Control-Forest Rehabilitation, Ministry of Environment and Forestry of the Republic of Indonesia, for providing research permission and leaf material collection permit in Cirangsad Experimental Forest. High appreciation also goes to Mr. Andik Vetriawan, B.Sc.F for his assistance during sampling in the Cirangsad Experimental Forest, and Mr. Muhammad Majiidu, B.Sc.F from Advanced Research Laboratory, IPB University for his contribution in preparing a map of the sampling location and assisting a statistical data analysis.

References

- Baskorowati, L., Bush, D., Setiadi, D., & Susanto, M. (2017). Genetic variation of growth and disease resistance traits in open-pollinated provenance-progeny trials of *Falcataria moluccana* growing on two rust-affected sites at age-18 months. *Jurnal Manajemen Hutan Tropika*, 23, 1–7. <https://doi.org/10.7226/jtjm.23.1.1>
- [BPS] Badan Pusat Statistik. (2021). Produksi perusahaan pembudidaya tanaman kehutanan menurut jenis produksi 2017–2019. Retrieved from <https://www.bps.go.id/indicator/60/1851/1/produksi-perusahaan-pembudidaya-tanaman-kehutanan-menurut-jenis-produksi.html>
- Dwivedi, S., Singh, S. Chauhan, U. K., & Tiwari, M. K. (2018). Inter and intraspecific genetic diversity (RAPD) among three most frequent species of macrofungi (*Ganoderma lucidum*, *Leucoagaricus* sp. and *Lentinus* sp.) of tropical forest of Central India. *Journal of Genetic Engineering and Biotechnology*, 16, 133–141. <https://doi.org/10.1016/j.jgeb.2017.11.008>
- Hidayat, W., Qi, Y., Jang, J. H., Febrianto, F., Lee, S. H., Chae, H. M., ..., & Kim, N. H. (2017a). Carbonization characteristics of juvenile woods of some tropical trees planted in Indonesia. *Journal of the Faculty of Agriculture, Kyushu University*, 62, 145–152.
- Hidayat, W., Sya'bani, M. I., Purwawangsa, H., Iswanto, A. H., & Febrianto F. (2017b). Effect of wood species and layer structure on physical and mechanical properties of strand board. *Jurnal Ilmu dan Teknologi Kayu Tropis*, 9, 134–140.
- Ikhfan, A. N., & Wijayanto, N. (2019). Assessing the growth of local *sengon* and Solomon *sengon* in agroforestry system. *IOP Conference Series: Earth and Environmental Science*, 394, 012028. <https://doi.org/10.1088/1755-1315/394/1/012028>
- Kimura, M., & Crow, J. F. (1964). The number of alleles that can be maintained in a finite population. *Genetics*, 49, 725–738.
- Krisnawati, H., Varis, E., Kallio, M., & Kanninen, M. (2011). *Paraserianthes falcataria* (L) Nielsen: *Ekologi, silvikultur dan produktivitas*. Bogor: CIFOR.
- Lelana, N. E., Wiyono, S., Giyanto, & Siregar, I. Z. (2018). Genetic diversity of *Falcataria moluccana* and its relationship to the resistance of gall rust disease. *Biodiversitas*, 19, 12–17. <https://doi.org/10.13057/biodiv/d190102>
- Loera-Sánchez, M., Studer, B., & Kölliker, R. (2019). DNA-based assessment of genetic diversity in grassland plant species: Challenges, approaches, and applications. *Agronomy*, 9, 881. <https://doi.org/10.3390/agronomy9120881>

- Mahbub, I. A., Tampubolon, G., & Irianto. (2020). Perbaikan beberapa sifat kimia tanah dan pertumbuhan tanaman sengon Solomon (*Falcataria moluccana* (Miq.) Barneby & Grimes) melalui pemberian kompos *Desmodium ovalifolium* pada tanah bekas tambang batubara. *Jurnal Silva Tropika*, 4, 222–228.
- Nei, M. (1972). Genetic distance between populations. *American Naturalist*, 106, 283–292. <https://doi.org/10.1086/282771>
- Nei, M. (1973). Analysis of gene diversity in subdivided populations. *Proceedings of the National Academy of Sciences of the United States of America*, 70, 3321–3323. <https://doi.org/10.1073/pnas.70.12.3321>
- Peakall, R., & Smouse, P. E. (2006). GenAlEx 6: Genetic analysis in Excel. Population genetic software for teaching and research. *Molecular Ecology Notes*, 6, 288–295. <https://doi.org/10.1093/bioinformatics/bts460>
- Priadi, D., & Hartati N. S. (2015). Viability and in vitro shoot multiplication of plus seed of sengon (*Paraserianthes falcataria*) of fresh and 4-years storage seed. *Prosiding Seminar Nasional Masyarakat Biodiversitas Indonesia*, 1, 1516–1519.
- Rohlf, F. J. (1998). *NTSYS-p.c.: Numerical taxonomy and multivariate analysis system (Version 2.0)*. Setauket, New York: Exeter Software Publishers Ltd.
- Rostini, N., Sunarya, S., Murdaningsih, H. K., Anas, & Damayanti, F. (2018). Pemanfaatan marka RAPD untuk mengestimasi diversitas genetik progeni half-sib *Paraserianthes falcataria* (L.) Nielsen hasil seleksi ketahanan boktor dan riap pertumbuhan. *Zuriat*, 29, 110–117.
- Schmidt, F. H., & Ferguson, J. H. A. (1951). *Rainfall type based on wet and dry period ratio for Indonesia with Western New Guinea*. Jakarta: Kementerian Perhubungan Djawatan Meteorologi dan Geofisika.
- Setiadi, D., Baskorowati L., & Susanto, M. (2014). Pertumbuhan sengon Solomon dan responnya terhadap penyakit karat tumor di Bondowoso, Jawa Timur. *Jurnal Pemuliaan Tanaman Hutan*, 8, 121–136.
- Siregar, U. J., & Olivia, R. D. (2012). Keragaman genetik populasi sengon (*Paraserianthes falcataria* (L) Nielsen) pada hutan rakyat di Jawa berdasarkan penanda RAPD. *Jurnal Silvikultur Tropika*, 3, 1–7.
- Wagner, W. L., Herbst, D. R., & Sohmer, S. H. (1999). *Manual of the flowering plants of Hawaii* (Vols. 1–2). Honolulu, Hawaii: University of Hawaii and Bishop Press.
- Weising, K., Nybon, H., Wolff, K., & Kahl, G. (2005). *DNA fingerprinting in plants: Principles, methods, and applications* (2nd ed.). Boca Raton, Florida: Taylor & Francis.
- Widyastuti, D. E. (2007). Keragaman genetik dengan penanda RAPD, fenotipa pertumbuhan dan pendugaan heritabilitas pada sengon (*Paraserianthes falcataria* (L.) Nielsen) [thesis]. Bogor: Institut Pertanian Bogor.
- Williams, J. G. K., Kubelik, A. R., Livak, K. J., Rafalski, J. A., & Tingey, S. V. (1990). DNA polymorphisms amplified by arbitrary primers are useful as genetic markers. *Nucleic Acids Research*, 18, 6531–6535. <https://doi.org/10.1093/nar/18.22.6531>
- Yeh, F. C., Yang, R. C., Boyle, T. B. J, Ye, Z. H., & Mao, J. X. (1997). *POPGENE, the user-friendly shareware for population genetic analysis*. Canada: Molecular Biology and Biotechnology Centre, University of Alberta.
- Zobel, B., & Talbert, J. (1984). *Applied forest tree improvement*. New York: John Willey and Sons.