Growth diversity, heritability, and genetic correlation of 4-years-old of *Alstonia angustiloba* progeny test in Wonogiri, Central Java

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**Abstract.** *Alstonia angustiloba* is a local tree species that have potential for community forest plantation; therefore, it is important to provide improved seeds. This study aimed to determine the diversity of growth, estimate the value of heritability, and the genetic correlation of the 4-years-old *A. angustiloba* progeny test. Randomized Completely Block Design with two factors (population and family) were used in this study. In this study, families are nested in the population. The population factors consisted of 4 levels, and family factors consisted of 43 families. The analysis of variance showed that height and stem diameter growth were significantly different between populations and families at four years old. The best height and stem diameter growth at the population level was obtained from the Pendopo population, 4.45 m and 7.71 cm, respectively. At the family level, the best height growth was obtained from 9 families (4.46-5.06 m), and the best stem diameter growth was obtained from 11 families (7.48-8.72 cm). The estimated individual heritability value for height was 0.41, and stem diameter was 0.23. Estimated family heritability values were 0.66 for height and 0.50 for stem diameter. The genetic correlation between height and stem diameter was 0.97.

1. Introduction

Wood demand for industrial raw materials increases continuously. Meanwhile, wood products from natural forests outside Java decrease. [1] stated that wood production from natural forests in 2015 was 8.3 million m$^3$, but production decreased to 5.7 million m$^3$ in 2018. To overcome decreasing wood production from natural forests problem, the development of high productivity plantations is very important. One of the potential tree species for the development of plantation forests is *Alstonia angustiloba* (*pulai darat*).

In Indonesia, this species is naturally distributed in Sumatera, Bangka, Java, Kalimantan/Borneo, and Sulawesi [2]. Due to the large range of its natural distribution, it suggests that *A. angustiloba* will adapt to many areas' environmental conditions. In the timber industry, this wood can be used to manufacture chests, matches, heels, handicraft items such as *wayang golek* and masks, concrete molds, pencil slate, and pulp. There are several industries that have developed and required a supply of raw material for *pulai darat* wood, including the pencil slate industry in Lubuk Linggau, Musi Rawas district, South Sumatra province, and the center for wood carving craft industry in Patuk, Gunung Kidul, Yogyakarta. In the pencil slate industry in Musi Rawas, South Sumatra, raw materials were supplied...
from the community forests but only supported ±50% of the required capacity [3]. For the carving 
handicraft industry in Gunung Kidul, the supply of pulai wood comes from natural stands in East Java, 
and there is still a shortage of ± 60 m³ per month [4].

Therefore, the development of pulai darat plantation forests with high productivity is needed, which 
needs the support of improved seeds. The progeny test of A. angustiloba had been established in 
Wonogiri, Central Java, and will be converted to seed orchard after final selections. Periodic evaluations 
are carried out to determine the genetic performance of A. angustiloba progeny test. This study aims to 
determine the diversity of growth, the estimated value of individual and family heritability, and the 
genetic correlation of the 4-years-old A. angustiloba.

2. Material and Methods

2.1. Material

Progeny test of 4-years-old A. angustiloba in KHDTK (Forest Area with Specific Purpose) Wonogiri, 
Central Java, was used for this study. This established progeny test used seeds originated from Carita-
Banten, Pendo-Muara Enim, Lubuk Linggau-Musi Rawas, and Solok-West Sumatera. The 
geographic location, altitude, and rainfall of 4 populations of the natural distribution of A. angustiloba 
are presented in Table 1. Plant height measurement used gauges, stem diameter measurement using 
calipers, and tally sheets to record measurement results.

Table 1. Geographical location, altitude, and rainfall of 4 populations of the natural distribution of A. 
angustiloba progeny test in Wonogiri, Central Java.

| Seed Origin | Province/District | Geographic Location | Altitude (m asl) | The average of rainfall (mm year⁻¹) |
|-------------|-------------------|---------------------|-----------------|----------------------------------|
| Carita      | Banten            | 105º53’ – 106º01’ BT | 6º14’ – 6º25’ LS | 30-100                           | 2000                           |
|             | Muara Enim        | 103º34’ – 103º58’ BT | 3º20’ – 3º32’ LS | 90-150                           | 2780                           |
|             | Musi Rawas        | 102º44’ – 103º01’ BT | 3º15’ – 3º24’ LS | 120-200                          | 2760                           |
|             | Sumatera Barat    | 100º20’ – 101º00’ BT | 0º35’ – 0º50’ LS | 500-600                          | 2800                           |

Source: [3].

2.2. Methods

2.2.1. Research design. A Completely Randomized Block Design with two factors (seed origin and family) 
was used in this study. In this study, families were nested in the seed origin. The seed origin factors consisted of 4 levels, as listed in Table 1. Family factors consisted of 43 families, namely 15 
from the Carita-Banten population, nine from the Pendo-Muara Enim population, 15 from the Lubuk 
Linggau-Musi Rawas, and four from the Solok-West Sumatera population. Each family has planted four 
individuals and repeated six times; hence the total plants were 1032.

2.2.2. Research procedure. The research was carried out by measuring the height and diameter of the 
stems of A. angustiloba progeny test with 100% sampling intensity. Plant height was measured from 
the stem base to the top of the plant, and the stem diameter was measured at the breast height (1.3 m 
(dbh)) from the base of the stem. The tally sheet was printed and arranged according to the plant design 
for the proper record. The observed parameters in this study were tree/plant height and stem diameter.

2.3. Data analysis

Height and stem diameter were analyzed using individual data; with the linear model as following [5]:

\[ h = \beta_0 + \beta_1 x + \epsilon \]

\[ d = \beta_0 + \beta_1 x + \epsilon \]

where:
- \( h \) is the plant height (cm)
- \( d \) is the stem diameter (cm)
- \( x \) is the age of the plant (years)
- \( \beta_0, \beta_1 \) are the regression coefficients
- \( \epsilon \) is the residual error

The results obtained from the analysis were then used to determine the genetic performance of the 
progeny test, heritability, and genetic correlation.
\[ Y_{ijkl} = \mu + B_i + P_j + F_k(P_j) + B_i*F_k(P_j) + E_{ijkl} \]

Notes:
- \( Y_{ijkl} \) = observations in i-th block, j-th population, k-th family, and l-th individual;
- \( \mu \) = general mean of observations;
- \( B_i \) = effect of the i-th block;
- \( P_j \) = effect of the j-th population (seed origin);
- \( F_k(P_j) \) = the influence of the k-th family nested in the j-th population;
- \( B_i*F_k(P_j) \) = the effect of the interaction of the i-th block with the k-th family nested in the j-th population;
- \( E_{ijkl} \) = random error.

The Duncan Multiple Range Test (DMRT) is used to analyze variance is significant at the 1% or 5% test level. Individual and family heritability on tree height and stem diameter characters were estimated using the following equation:

\[
\begin{align*}
    h_i^2 &= \frac{3\sigma_f^2}{\sigma_f^2 + \sigma_{fb}^2 + \sigma_e^2} \\
    h_f^2 &= \frac{\sigma_f^2}{\sigma_f^2 + \sigma_{fb/B}^2 + \sigma_{e/NB}^2}
\end{align*}
\]

Notes:
- \( h_i^2 \) = individual heritability value,
- \( h_f^2 \) = family heritability value,
- \( \sigma_f^2 \) = variance component of family,
- \( \sigma_{fb}^2 \) = variance component of family and block interaction,
- \( \sigma_e^2 \) = variance component of error,
- \( B \) = harmonic mean of the number of blocks,
- \( N \) = harmonic mean of the number of individuals per plot.

The variance component of the family (\( \sigma_f^2 \)) in the individual heritability equation is assumed to be 1/3 of the additive genetic variance (\( \sigma_A^2 \)) because the potential for single or relative mating of pulai plants in nature is quite high [6]. The estimated genetic correlation between traits was calculated using the following formula [7]:

\[
r_g = \frac{\sigma_f(x,y)}{\sqrt{\sigma_f(x)^2 + \sigma_f(y)^2}}
\]

Notes:
- \( r_g \) = genetic correlation,
- \( \sigma_f(x,y) \) = covariance component of family for characters x and y,
- \( \sigma_f(x) \) = variance component of family for character x,
- \( \sigma_f(y) \) = variance component of family for character y.

3. Result and Discussion

3.1. Growth diversity

Variation of height and stem diameter was found at the 4-years-old progeny test of A. angustiloba in Wonogiri, Central Java. The average plant height is 3.91 ± 0.78 m, and the stem diameter is 6.71 ± 1.62 cm. Therefore, the analysis of variance was carried out to determine the diversity of growth in height and stem diameter, as presented in Table 2.

The analysis of variance (Table 2) showed that the treatment of seed origin and family varied significantly in height and stem diameter growth, reflecting the diversity between populations and
between families. As represented, this progeny test at 1 and 3 years old was in the same test plot [3, 6]. These results indicate that the growth (height and stem diameter) of A. angustiloba from 1 to 4 years old remains diverse even though the progeny test experienced drought when it reached 1.5 to 2 years old; a resulted number of plants experienced death with a survival rate of 82.07% at the age of 2 years [8, 9]. This information shows that drought stress with a relatively high mortality rate does not significantly affect the growing diversity in height and stem diameter of A. angustiloba. A previous study [6] informed that drought stress had the same relative impact on the survival rate of A. angustiloba between populations and between families. Plants that can survive drought stress are generally through the response of transpiration control, control of turgor pressure, the formation of small and succulent leaves, and increased proline accumulation [10, 11, 12, 13]. [6] reported that the response of A. angustiloba to drought stress occurred through the mechanism of increased proline accumulation because increased proline accumulation can reduce the osmotic potential of cells. Hence, cell turgor pressure can be maintained [13].

Table 2. Analysis of variance of height and stem diameter of 4-years-old A. angustiloba progeny test in Wonogiri, Central Java.

| Source of Variation                  | Degree of Freedom | Height       | Stem Diameter |
|--------------------------------------|-------------------|--------------|---------------|
| Block                                | 5                 | 49.5670      | 145.9718      |
| Population                           | 3                 | 51.1560**    | 140.0160**    |
| Family (population)                  | 39                | 3.6893**     | 14.8677**     |
| Block*family (population)            | 199               | 4.4572**     | 20.1793**     |
| Error                                | 560               | 0.6351       | 2.6236        |
| Total                                | 806               |              |               |

Note: "*" Significantly different at the level of 0.01

The diversity of Alstonia was also reported by [14] with RAPD markers, which stated that some populations in Indonesia have sufficient genetic diversity (0.247). The genetic diversity value is higher when compared to the average genetic diversity of the sengon population in Java Island with the RAPD marker of 0.235 [15] and kulim (Scorodocarpus borneensis) originated from Riau with an RAPD marker of 0.226 [16]. The genetic diversity values in this study were similar to the genetic diversity of Cordia africana in Ethiopia, which was 0.220-0.320 [17]; meranti (Shorea leprosula) from several populations in West Kalimantan, Central Kalimantan, and East Kalimantan with an isozyme marker of 0.247 [18]. However, the genetic diversity value in this study was lower when compared to the genetic diversity value of Gyrinops versteegii in Kebar, Papua [19], Diospyros rumphii in North Sulawesi [20], mindi (Melia azedarach) in West Java [21], Eucalyptus urophylla in Indonesia [22], Guaiacum sanctum in Costa Rica [23] and Dalbergia monticola in Madagascar [24]. Low genetic diversity in this study is probably due to the high exploitation of the Alstonia in their natural habitat. Hence, the stands in nature tend to decline, resulting in decreased genetic diversity.

To find out the difference in growth between treatments, the DMRT was then carried out. The results of the DMRT on the growing diversity in height and stem diameter between population origins are presented in Figure 1, and between families are presented in Table 3.
Figure 1. DMRT height and stem diameter between seed origin of 4 years old *A. angustiloba*. Seed origin: 1) Carita-Banten, 2) Pendopo-Muara Enim, 3) Lubuk Linggau-Musi Rawas, and 4) Solok-West Sumatera.

The DMRT (Figure 1) showed that the height and stem diameter growth of the 4-years-old *A. angustiloba* from the Pendopo population is better than the Lubuk Linggau, Carita, and Solok populations. Meanwhile, the growth in height and stem diameter of *A. angustiloba* from the Lubuk Linggau and Carita populations were better than the Solok population. The diversity of growth between populations is in accordance with the previous results, i.e., 1 and 3 years old observations [3, 6]. A previous study with the RAPD marker [14] informed that the genetic distance of the *A. angustiloba* from the Pendopo, Lubuk Linggau, and Carita populations were grouped in one cluster, while the Solok population was included in another cluster. In addition, the difference in altitude (elevation) of the Solok population with other populations is quite large (Table 1). Therefore it exhibited the lowest growth compared to other populations. According to [25], such a large difference in elevation can interfere with the physiological activity of plants affecting the growth.

Table 3. DMRT of families to height and stem diameter of 4 years old *A. angustiloba* progeny test at Wonogiri, Central Java.

| No. | Family | The mean of height (m) | No. | Family | The mean stem diameter (cm) |
|-----|--------|------------------------|-----|--------|-----------------------------|
| 1   | 21     | 5.06 a                 | 18  |        | 8.72 a                      |
| 2   | 17     | 4.88 ab                | 2   | 27     | 8.70 a                      |
| 3   | 36     | 4.80 abc               | 3   | 21     | 8.45 ab                     |
| 4   | 27     | 4.76 abcd              | 4   | 6      | 8.29 abc                    |
| 5   | 6      | 4.69 abcd              | 5   | 8      | 8.27 abcd                   |
| 6   | 8      | 4.59 abcde             | 6   | 17     | 8.19 abcde                  |
| 7   | 18     | 4.55 abcdef            | 7   | 13     | 8.12 abcdef                 |
| 8   | 19     | 4.53 abcdef            | 8   | 36     | 8.11 abcdef                 |
| 9   | 13     | 4.46 abcddef           | 9   | 16     | 7.84 abcddef                |
| 10  | 24     | 4.41 bcdefgh           | 10  | 19     | 7.51 bcdefgh                |
| 11  | 22     | 4.30 bcdefghi          | 11  | 29     | 7.48 bcdefghi               |
| 12  | 20     | 4.26 cdefghij          | 12  | 25     | 7.29 cdefghij               |
| 13  | 5      | 4.25 cdefghij          | 13  | 24     | 7.22 cdefghij               |
| 14  | 29     | 4.24 cdefghij          | 14  | 22     | 7.19 cdefghij               |
| 15  | 34     | 4.24 cdefghij          | 15  | 34     | 7.07 cdefghijk              |
correlation is a correlation between breeding value

heritability is the proportion of genetic variants to phenotypic variants. Meanwhile, genetic

correlation is a correlation between breeding values for different traits and is ma

At the family level, the diversity of height growth of A. angustiloba between families was separated
into 19 groups, and stem diameter was separated into 18 groups. Early observation at one year old
showed that the height growth of the A. angustiloba was grouped into 16 groups, and the stem diameter
was grouped into seven groups. In comparison, at the age of 3 years, the height growth was clustered in
12 groups, and the stem diameter was clustered in 14 groups [3, 6]. This indicates that drought stress
when the plants are 1.5-2 years old [9] causes a decrease in plant height diversity at three years of age
and increases when the progeny test reaches 4-years old. Furthermore, A. angustiloba with craggy and
stratified branches also produces various internode lengths [26]. The growing diversity in height, stem
diameter, and internode length was also shown in the pulai gading (Alstonia scholaris) in the 4-years-
old progeny test in Gunung Kidul, Yogyakarta [27].

The DMRT (Table 3) shows that the best height comes from 9 families with height intervals between
4.46-5.06 m, while the best stem diameter from 11 families with diameter intervals between 7.48-8.72
cm. Thus, families mostly produced the best height and stem diameter from the Pendopo population,
Muara Enim. This is consistent with the results of the DMRT at the population level (Figure 1) that the
Pendopo population is the best population in terms of height and stem diameter growth.

3.2. Heritability and genetic correlation

Heritability and genetic correlation are very important genetic parameters in breeding activities. [28]

stated that heritability is the proportion of genetic variants to phenotypic variants. Meanwhile, genetic
correlation is a correlation between breeding values for different traits and is mainly caused by genes

| No. | Family | The mean of height (m) | No. | Family | The mean stem diameter (cm) |
|-----|--------|------------------------|-----|--------|-----------------------------|
| 16  | 32     | 4.21 cdefghij          | 16  | 23     | 7.02 cdefghij               |
| 17  | 25     | 4.17 cdefghijk         | 17  | 20     | 6.70 cdefghijk              |
| 18  | 16     | 4.12 defghijk          | 18  | 15     | 6.98 efghijk                |
| 19  | 7      | 4.05 efghijkl          | 19  | 32     | 6.92 efghijkl               |
| 20  | 23     | 4.02 efghijkl          | 20  | 33     | 6.87 fg hjklm               |
| 21  | 33     | 3.96 fghijklm          | 21  | 9      | 6.70 ghijklmn               |
| 22  | 9      | 3.92 fghijklmn         | 22  | 11     | 6.68 ghijklmn               |
| 23  | 15     | 3.85 ghijklmn          | 23  | 7      | 6.67 ghijklmn               |
| 24  | 30     | 3.84 ghijklmn          | 24  | 5      | 6.63 ghijklmn               |
| 25  | 12     | 3.80 hijklmno          | 25  | 10     | 6.47 hijklmno               |
| 26  | 10     | 3.79 hijklmn           | 26  | 30     | 6.40 hijklmn                |
| 27  | 3      | 3.78 hijklmnno         | 27  | 12     | 6.17 ijklmnopq              |
| 28  | 11     | 3.77 iklmnno           | 28  | 31     | 6.16 iklmnopq               |
| 29  | 14     | 3.65 jkmnopq           | 29  | 14     | 6.02 jkmnopq                |
| 30  | 31     | 3.55 kmnopq            | 30  | 26     | 5.94 kmnopq                 |
| 31  | 26     | 3.48 lnmnopq           | 31  | 2      | 5.88 lnmnopq                |
| 32  | 38     | 3.46 lnmnopq           | 32  | 28     | 5.84 lnmnopq                |
| 33  | 2      | 3.36 nmpoq             | 33  | 3      | 5.77 nmpoq                  |
| 34  | 35     | 3.33 mnpq              | 34  | 38     | 5.65 mnpq\textsuperscript{r} |
| 35  | 37     | 3.32 nmpoq             | 35  | 42     | 5.58 nmpoq\textsuperscript{r} |
| 36  | 28     | 3.31 npoq              | 36  | 37     | 5.57 npoqr                  |
| 37  | 4      | 3.19 opq               | 37  | 35     | 5.54 npoqr                  |
| 38  | 1      | 3.10 pqr               | 38  | 1      | 5.33 opq\textsuperscript{r} |
| 39  | 39     | 3.07 pqr               | 39  | 4      | 5.16 pqr\textsuperscript{r} |
| 40  | 42     | 2.96 qrs               | 40  | 39     | 5.06 qr\textsuperscript{r}  |
| 41  | 43     | 2.93 qrs               | 41  | 40     | 4.94 qr\textsuperscript{r}  |
| 42  | 40     | 2.59 rs                | 42  | 43     | 4.89 qr\textsuperscript{r}  |
| 43  | 41     | 2.44 s                 | 43  | 41     | 4.46 r\textsuperscript{r}   |
that affect more than one trait (pleiotrophy) [7]. Estimates of heritability values and genetic correlations for height and stem diameter traits in the 4-years-old plot of *A. angustiloba* are presented in Table 4.

**Table 4.** The estimated value of individual heritability, family heritability, and genetic correlation of height and stem diameter of 4-years-old *A. angustiloba* progeny test in Wonogiri, Central Java.

| Parameter     | Individual Heritability ($h^2_i$) | Family Heritability ($h^2_f$) | Genetic Correlation ($r_G$) |
|---------------|-----------------------------------|-------------------------------|----------------------------|
| Height        | 0.41                              | 0.66                          | 0.97                       |
| Stem Diameter | 0.23                              | 0.50                          |                            |

Estimated heritability values (Table 4) show that height’s individual and family heritability values were 0.41 and 0.66, respectively. Then, the individual and family heritability values for stem diameter were 0.23 and 0.50, respectively. Individual and family heritability for height were included in the high criteria, while stem diameter was included in the moderate criteria [29]. The value of individual and family heritability in this study was higher than when the plants were 3-years-old, i.e., 0.32 and 0.59 on individual and family heritability on height, and 0.20 and 0.49 on height and stem diameter [6]. Thus, the heritability value of the *A. angustiloba* is still changing (increasing) until the progeny test reached four years old. This is probably due to the increasing plant age, allowing genes to change plant growth [30]. The phenomenon of changes in heritability values accordance with increasing age also occurs in several species of plants, including teak (*Tectona grandis*) [31], *Acacia mangium* [32], *Araucaria cunninghamii* [33, 34], and *pulai gading* (*A. scholaris*) [27, 35].

The heritability values of individuals and families in this study were lower than those of the *A. scholaris* of the same age in Gunung Kidul, Yogyakarta [27]. This is because probably the genetic material used to establish the *pulai gading* progeny test came from a natural distribution (Solok, Bali, Lombok, Timor, and Jayapura) that was relatively wider than the genetic material used in this study (Table 1). Therefore, it has a greater genetic diversity and higher heritability values. In addition, differences in the species and locations of the tests are also influencing factors because heritability values only apply to certain species of plants and at certain locations.

The genetic correlation between height and stem diameter of 4-years-old *A. angustiloba* had a strong positive value (0.97). This correlation value illustrates that an increase in plant height is followed by an increase in stem diameter, or vice versa, with a degree of association of 97%. As with the heritability value, the genetic correlation value in this study was greater than the genetic correlation value at the age of 3 years, which was 0.96 [6]. The genetic correlation value in this study was also higher than the genetic correlation value of several other tropical plant species, including 5-years-old teak of 0.84 [31], *araukaria* (*A. cunninghamii*), 1.5 and 5-years-old of 0.8 and 0.83 [33, 34], sandalwood (*Santalum album*) at eight months of 0.87 [36], *sengon* (*Paraserianthes falcatoria*) aged one and 4-years-old of 0.85 and 0.91 [37, 38], and *nyawai* (*Ficus variegata*) 1-year-old of 0.8 [39]. However, the genetic correlation value between height and stem diameter of the 4-years-old *A. angustiloba* was lower than that of the 4-years-old *A. scholaris* in Gunung Kidul, Yogyakarta, which was 0.98 [27]. However, it is assumed that the genetic correlation value can still change with increasing plant age because up to 4 years, the value of genetic correlation still tends to increase. In its application, the value of genetic correlation can be used as information for the efficiency of the selection process. If the correlation value is strong and consistently positive until the plants are ready to be selected, the selection can be based on just one character.

4. Conclusion
The growth in height and stem diameter of the first generation (F-1) progeny test of the 4-years-old *A. angustiloba* in Wonogiri showed significant differences between populations and families. The best population in growth (height and stem diameter) is Pendopo, Muara Enim. The best families in height growth were occupied by nine families, and the stem diameter was occupied by 11 families. Estimates of the heritability value of individuals and families of height and stem diameter were included in the
criteria of moderate to high. The genetic correlation between height and stem diameter was positive and strong (0.97).

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All authors contributed equally to this work as the main contributor.