Trends of genetic parameters and stand volume productivity of selected clones of *Eucalyptus pellita* observed in clonal trials in Wonogiri, Central Java

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Abstract. Breeding program of *Eucalyptus pellita* has proceeded into advanced generations with clonal development as best way to deliver greater genetic gain. Clonal test is a prerequisite for selecting and verifying the clones productivity before deployment. Following the breeding strategy, clonal trials of *E. pellita* were established in Wonogiri, Central Java. The purposes of this study are to observe trends of genetic parameters and stand volume productivity of clones tested in clonal trials of *E. pellita*. The trials were laid out in two experimental designs: 1) nine clones, 16 replications, single tree-plot, spacing 3 x 2m; 2) 4 clones, 2-4 replications, 16 squares tree-plot, spacing 3 x 2m. Each trial also contains control (i.e. seedling material). Measurements were conducted five times from six months to 46 months age. Results showed that trends of growth increased along the measurements with nine tested clones performed better than control seedlings. The growths were significantly different among clones along the ages with clone repetibility ranging from 0.49 to 0.98. Three selected clones namely clone 1, 2 and 3 on the basis of height growth and multiplication rates showed higher stand volume productivity compared to control and consistently increased on average of 0.37 m$^3$/ha at 6 months to 73.93 m$^3$/ha at 46 months age.

1. Introduction

*Eucalyptus pellita* F. Muell is a fast growing species developed in Industrial Forest Plantation as raw material for pulp and paper industries. To support the increasing of raw material, some silviculture practices to improve the stand productivity of *E. pellita* are applied. However, it has often not achieved the expected results because of the high variation in stands [1]. Breeding program would be the alternative effort that could be practiced through developing genetically improved seed. One of the advanced breeding strategy in *E. pellita* is the development of superior clones. This is because the development of clones is proven to able to increase genetic parameters, relatively uniform stands growth and more economical compared to that of seedlings [2].
development of *Eucalyptus* clones in several countries has been carried out to increase the supply of raw material, especially pulp and paper, such as in China, Brazil and Congo [3] [4] [5].

In order to improve the productivity of *E. pellitae* in Indonesia through advanced breeding strategy, the Center for Forest Biotechnology and Tree Improvement, Yogyakarta has established two clonal trials of *E. pellita* in 2011 [6]. Some progress on the early growth of the trials has been reported up to 24 months age after planting [7] [8]. However, progress on the subsequent advanced ages has not been reported as yet. Therefore the purpose of this study is to observe the trends of genetic parameters and stand volume productivity of clones tested in the two clonal trials of *E. pellitae* up to advanced ages.

2. Materials and Methods

2.1. Clonal trial of *E. pellita*

The genetic materials tested in the trials are some clones of *E. pellitae* derived from nine selected plus trees in first-generation of progeny trial. The both trials were established in Wonogiri, Central Java. The original provenances of genetic material were from Papua New Guinea. The clone trials were established in 2011 [6], while the progeny trial was in 1995 [9]. The clonal propagation were practiced by shoot cuttings collected from rejuvenated sproutsof selected plus trees following the cutting technique that had been developed [10]. As genetic control, some seedlings raised using the improved seed from first-generation seedling seed orchard were planted together with the clones in the trial. Site descriptions of the clonal trials are represented in Table. 1.

| Item                        | Information                  |
|-----------------------------|------------------------------|
| Geographic position         | 7°80' S and 110°93'E         |
| Climate                     | Type D (Schmidt-Fergusson)   |
| Rain fall (mm/year)         | 1,878                        |
| Temperature (min-max)       | 21 - 32°C                    |
| Soil order                  | Vertisol                     |
| Elevation (m asl)           | 141                          |
| Slope (%)                   | 10                           |

Source : [6]
The clonal trials were laid out in two experimental designs, i.e. single tree-plot and multiple tree-plot models[6]. For the single tree-plot, nine clones were tested in 16 replications with 3 x 2m spacing, while for the multiple tree-plot, 4 selected clones in term of rooting ability were tested in 2-4 replications, 16 squares tree-plot, and spacing of 3 x 2m. Measurements were conducted five times from six months to 46 months of age after planting on height (m), diameter at breast height (dbh, cm) and individual stem volume (m³).

2.2. Measurement and data Analysis

2.2.1. Analysis of variance

Analysis of variance was done for individual data with linear model as follow:

\[ Y_{ij} = \mu + R_i + C_j + E_{ij} \] ................................................................. (1)

where: \( Y_{ij} \) is individual observation of the \( j \)-th clone and \( i \)-th replication; \( \mu \) is mean of population; \( R_i \) is an effect of the \( i \)-th replication; \( C_j \) is an effect of the \( j \)-th clone and \( E_{ij} \) is an error associated with \( Y_{ij} \).

2.2.2. Repeatability values

The estimation of repeatability was calculated on the basis of clone means (clone mean heritability) using equation [11] as follow:

\[ R^2_c = \frac{\sigma^2_c}{\sigma^2_c + (\sigma^2_e/b)} \] ................................................................. (2)

Where:

\( H^2_c \) = clone heritability; \( \sigma^2_c \) = clone component of variance; \( \sigma^2_e \) = error component of variance, and \( b \) = replication

2.2.3. Stand productivity

As a function of height and diameter, the individual stem volume (V) was calculated for each individual tree using the equation [12] as follow:

\[ V = \frac{dbh^2h_t}{23163.87 + 149.03 \, dbh} \] ................................................................. (3)

Where: \( V \) = stem volume; \( dbh \) = diameter at breast height and \( h_t \) = height

The stand volume in each plot was then calculated by summing up the individual stem volume in each plot. Furthermore, the stand volume per hectare was calculated by
converting the total stand volume in each plot into the stand volume per hectare (m$^3$/ha) using the equation [13] as follow:

\[
\text{Stand volume per Hectare} = \frac{\text{stand volume on plot}}{\text{area of plot in square meter}} \times 10000
\]

3. Result and Discussion

3.1. Growth

Growth of \textit{E. pellita} clones were observed from 6 to 46 months of ages after planting for tree height and diameter. Analysis of variance showed that there were significant differences between tested clones for growth traits at all measurement times as shown in Table 2 and 3. It should be noted here that the results of analysis for 12 and 24 months used in this study are adopted from two previous studies that reported significant differences in growth traits in the \textit{E. pellita}clonal trials [7] [8]. These results showed that the trend of large variation among clones tested in the clonal trial was consistent from the early growth up to advanced growth ages. Another study in clonal trial of \textit{E. pellita}using different genetic materials also resulted in a significant difference among clones in East Kalimantan at four years of age [14]. These indicated that \textit{E. pellita}could provide some potential superior genotypes for selection through clonal development.

\begin{table}
\centering
\begin{tabular}{llllllllll}
\hline
\textbf{Source of variation} & \textbf{df} & \textbf{Height (m)} & & & & & \textbf{Diameter (cm)} & & & \\
 & & 6 & 12 & 24 & 36 & 46 & 6 & 12 & 24 & 36 & 46 \\
\hline
Rep & 15 & 0.486* & 0.963** & 4.216** & 3.788* & 4.888ns & 0.353** & 0.917** & 3.342** & 5.059** & 6.629** \\
Clone & 9 & 1.541** & 4.301** & 11.440** & 9.153** & 6.230* & 0.806** & 1.480** & 5.028** & 8.170** & 5.373** \\
Error & 125 & 0.259 & 0.316 & 1.007 & 2.062 & 3.092 & 0.110 & 0.248 & 0.888 & 1.917 & 2.399 \\
\hline
\end{tabular}
\caption{Analysis of variance for growth traits at 6 to 46 months of age in clonal trial of \textit{E. pellita} in Wonogiri, Central Java}
\end{table}

Remark:
* : Significantly different at 5% level
** : Significantly different at 1% level
ns : non significant
Table 3. Analysis of variance for stem volume at 6 to 46 months of age in clonal trial of *E. pellita* in Wonogiri, Central Java

| Source of variation | df  | Mean square          |
|---------------------|-----|----------------------|
|                     | 6   | 12                   | 24   | 36   | 46   |
| Rep                 | 15  | 0.015**              | 0.136** | 39.362** | 209.348** | 562.894** |
| Clone               | 9   | 0.041**              | 0.280** | 61.193** | 360.478** | 550.420** |
| Error               | 125 | 0.005                | 0.038  | 8.389  | 80.445  | 234.210   |

Remark
* : Significantly different at 5% level
** : Significantly different at 1% level
ns : non significant

The average of growth traits for height, diameter and stem volume increased along the ages as shown in Table 4. The growth increments varied among the clones and the traits. At final observation (46 months of age), Clones 2 and 14 showed the highest stem volume at around 0.0564 m³ and 0.0586 m³, respectively. While Clones 1, 2, 3, and 14 showed a constant increase in growth from six until 46 months of age. Setyaji [8] and Sunarti [7] also reported that the significant difference in growth in the clonal trial is the effect of genetic factors and indicated the importance of clone selection for increasing the productivity of *E. pellita*. Results from another study on *Eucalyptus* showed that the increase in growth traits of each clones of *E. urophylla* × *E. grandis* quite varies [4].

Table 4. Mean of growth traits at ages six to 46 months in clonal trial of *E. pellita* in Wonogiri, Central Java

| Clone | Height (m) | Traits | Stem volume (m³) |
|-------|------------|--------|------------------|
|       | 6 12 24 36 46 | 6 12 24 36 46 | 6 12 24 36 46 |
| 1     | 2.53 3.73 8.49 11.49 13.63 | 1.56 1.91 6.28 8.18 8.81 | 0.0004 0.0007 0.0149 0.0343 0.0465 |
| 2     | 2.73 3.72 8.23 11.80 13.65 | 1.46 2.04 6.33 8.59 9.92 | 0.0003 0.0007 0.0140 0.0361 0.0564 |
| 3     | 2.72 3.98 7.70 11.42 12.67 | 1.45 2.13 5.74 7.84 8.58 | 0.0003 0.0008 0.0112 0.0310 0.0417 |
| 4     | 2.12 2.92 6.86 11.11 13.18 | 1.24 1.73 6.04 8.48 9.21 | 0.0002 0.0004 0.0110 0.0343 0.0519 |
| 5     | 1.99 2.75 6.15 10.71 12.67 | 0.93 1.41 5.39 7.66 8.44 | 0.0001 0.0003 0.0079 0.0267 0.0391 |
| 6     | 2.35 3.12 6.60 10.33 11.95 | 1.27 1.62 5.21 6.60 8.18 | 0.0002 0.0004 0.0090 0.0228 0.0404 |
| 7     | 2.02 2.87 6.13 10.18 12.19 | 1.26 1.89 6.04 8.16 9.49 | 0.0002 0.0005 0.0099 0.0293 0.0475 |
| 8     | 1.96 2.84 6.31 10.37 12.12 | 1.10 1.59 5.13 7.05 8.62 | 0.0001 0.0004 0.0077 0.0228 0.0391 |
| 9     | 2.39 3.35 7.39 12.02 13.59 | 1.49 2.01 6.29 8.73 9.99 | 0.0003 0.0007 0.0128 0.0388 0.0586 |
| Control| 1.83 2.32 5.64 9.55 11.90 | 0.92 1.20 4.66 6.64 7.88 | 0.0001 0.0002 0.0063 0.0201 0.0350 |
| Mean  | 2.26 3.16 6.95 10.90 12.75 | 1.27 1.75 5.71 7.79 8.91 | 0.0002 0.0005 0.0105 0.0296 0.0456 |

The results also showed that the growth of tested clones along the ages were generally better than the control (seedlings) derived from improved seed collected...
from first-generation seedling seed orchard. This indicates the superiority consistency of selected clones over the seedlings in *E. pellita*. The superiority of clones was also reported in other studies, such as *E. grandis*×*E. camadulensis* clones in Africa [15] and *E. urophylla*×*E. grandis* clones in Kalimantan [16]. The average of height and diameter of tested clones (without control) at the age of 46 months in this study were 12.85 m and 9.03 cm, respectively. These are lower as compared to the results of other studies in East Kalimantan at 48 months of age, in which the mean of height and diameter were 18.6-18.8 m and 11.8-12.5 cm, respectively [14]. Another study in *E. hybrid* at 44 months in China reported the average height and diameter of 14.4 m and 11.0 cm, respectively [5].

Beside the differences in genetic material, this discrepancy might also be due to the differences in site planting environments. This is because clone development is very sensitive to site condition, and the clones tend to vary in growth depending on the environment [14].

### 3.2. Genetic parameter

The estimation of variance component and clone repeatability is presented in Table 5. In general the clone repeatability was categorised as high (>0.8) for all growth traits [17], except for 46 month age that showed the decrease of value at a range of 0.49-0.77. Clone repeatability tended to decrease as tree getting older. This is in line with the observation reported by Wu [18] for clones of *E. urophylla* that showed a decreasing trend of the repeatability from 21 to 96 months of age in China. Yang [19] also reported that based on the observation from 1.5 to 8 years of age in Southern China, there repeatability of *E. urophylla*×*E. tereticornis* started to decrease at 4.4 years of age.

| Ages (month) | Height (m) | Dbh (cm) | Stem volume (m³) |
|--------------|------------|----------|------------------|
| Variance component clone (σ²_c) | 0.10 0.27 0.84 0.50 0.27 | 0.05 0.08 0.33 0.52 0.25 | 0.01 0.04 7.74 35.94 38.46 |
| Repeatability clone (H²_c) | 0.82 0.94 0.94 0.96 0.49 | 1.00 0.94 0.88 0.96 0.76 | 1.00 1.00 0.97 0.98 0.77 |

The clone repeatabilities at the final age of observation (46 months) in this study are higher than those reported by Ramadan [14], that are 0.55-0.66 (height), 0.38-0.57
(diameter), and 0.43-0.63 (stem volume) on 48 months old *E. pellita* clones in Kalimantan. But, they were lower than study results reported by Wu [5], with 0.86 (height), 0.8 (diameter), and 0.8 (stem volume) on 44 months old of *Eucalyptus* clones in China.

Among the growth traits observed in this study, stem volume showed the highest value of repeatability in 46 months at around 0.77 (Table 3). Concerning the traits for selection criteria, the high repeatability in stem volume would provide a simple selection in the clonal trial. This result implies that selection of the tested clone based on the stem volume will comprehensively improve the growth productivity of *E. pellita*.

### 3.3. Stand productivity

Stand productivity were observed in the clonal trials designed in multiple tree-plot. This trial was established using 3 of 9 clones used in single tree-plot. Those 3 clones were selected based on the best rooting ability during clone multiplication. The 3 selected clones were Clones 1, 2 and 3. Clones 1 and 3 were collected from plus trees originated from provenances of Papua New Guinea, i.e. Serisa Village and Keru To Nata, while Clone 2 was collected from plus trees in bulkseed plantation with unidentified provenance. Referring to the stem volume productivity (Table 4), the 3 best clones in rooting ability also performed the best stem volume in single tree-plot of clonal trial. Therefore the clonal trial under multiple tree-plot could also be used as validating trial to verify the performance of selected clones into larger plot in order to obtain the information on stand productivity (m$^3$/ha).

The stand productivity of the three selected clones observed in multiple tree plot of clonal trial is presented in Table 4. The stand productivity increased with the increase of ages. The mean of clone stand productivity ranged from 0.37 m$^3$/ha (at 6 months) to 73.93 m$^3$/ha (at 46 months). A very remarkable leap of stand productivity was observed at 24 months age in which the mean of productivity increased almost 20 times over 12 months age productivity. Among the three clones, the highest stand volume productivity was shown by Clone 3, followed by Clone 2 and Clone 1. All the three clones showed higher stand productivity compared to the control (seedlings). The superiority of the three clones over the control ranged from 91% to 249%.
Figure 1. Stand volume productivity of the best three clones in clonal trial of *E. pellita* in Wonogiri, Central Java

The stand productivity of *E. pellita* clones represented by individual stem volume on this study is lower compared to study reported by Ramadan [14]. This discrepancy might be due to the differences in site conditions and silviculture practices. The soil of Wonogiri site is marginal with low annual rainfall (Table 1) and the study by Ramadan [14] was done in East Kalimantan with A climate type according to Schmidt–Ferguson and annual rainfall 1,400 – 2,500 mm/year. The *E. pellita* commonly shows good growth in favor of sufficient fertilizer, humidity and water [20]. Although showing lower stand productivity due to the poorer site condition, these results provide an opportunity to increasing stand productivity of *E. pellita* through further development of selected clones in operational plantation. The escalation of stand productivity along the age of *E. pellita* clones in this study also provides an important information for the management of clonal plantation, particularly in determining harvesting time in order to minimize the loss of potential gain in productivity from the selected clones and to obtain maximum wood production.

4. Conclusion

The observation up to three fourth of rotation age in clonal trials showed that the growth traits were significantly different among the tested clones of *E. pellita*, accompanied by the
consistent higclone repeatability. This indicates that the growth traits of the tested clones were highly controlled by genes along the growth periods and provides a good opportunity for selecting superior clones. Three top superior clones of *E. pellita* have been selected and they were further validated in larger plot providing the rates of superiority in stand productivity at range of 91%-249% over the seedlings derived from genetically improved seeds collected from first generation seedling seed orchard.

**Acknowledgment**

We would like to thank Acacia and Eucalyptus’s research team in CFBTI for all the fieldwork and data collections.

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