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## Clonal Variation and Genotype by Environment Interactions in Growth and Wood Density in *Eucalyptus camaldulensis* at Three Contrasting Sites in Vietnam

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### Abstract

A total of 172 clones of *Eucalyptus camaldulensis* were tested in three clonal tests in northern, north-central and southern Vietnam, with 32 of them planted across all three sites. At age 3–5 years, the clonal repeatabilities were 0.18–0.42 for growth traits, 0.71–0.78 for wood basic density and 0.56–0.66 for pilodyn penetration. Genotypic correlations between growth and density at the three sites were from –0.24 to 0.17, and did not differ significantly from zero. Genotypic correlations between sites were 0.32–0.56 for growth traits at age 3 years, and 0.72–0.88 for density and pilodyn penetration. Selection gains for breast height diameter at individual sites at a selection proportion of 5% were 22–32%, with minor effects on density. Selection for diameter at one site gave indirect responses in diameter at the other two sites that were only 40–60% of the gains obtainable from direct selection at those sites. This study shows that fast-growing *E. camaldulensis* clones can be selected in Vietnam with only minor effects on density. Selection for growth should be regionally based to maximize selection gain whereas clonal rankings for density will change little across regions.

**Key words:** clonal repeatability, correlated response, *Eucalyptus camaldulensis*, genotype by environment interaction, selection gain.

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### Introduction

*Eucalyptus camaldulensis* Dehnh is the most widely distributed eucalypt species, occurring primarily in riverine environments through most of the drier regions of Australia. The species displays major provenance variation (ELDRIDGE *et al.*, 1993). Recent taxonomic studies (BROOKER and KLEINIG, 2004; McDONALD *et al.*, 2009) identified several sub-species, distinguished by their differing leaf, bud and fruit morphology, with distributions that are usually geographically distinct, but overlap in some areas. In a recent study of population genetics using molecular markers, this sub-specific variation was shown to align closely with genetic differentiation (BUTCHER *et al.*, 2009). Physiological studies have revealed that provenances of *E. camaldulensis* differ in their mechanisms for adapting to drought conditions and in their water-use efficiency, and this has been related to the differing environmental conditions in their regions of origin (GIBSON *et al.*, 1994, 1995).

Provenances occurring in northern Australia have proved adaptable to a wide range of sub-humid tropical climates and *E. camaldulensis* has emerged as one of the most widely used plantation species in the seasonally dry tropics of many countries (MIDGLEY *et al.*, 1989; ELDRIDGE *et al.*, 1993) including India, Thailand and Vietnam, mainly for pulpwood, poles and less often small sawlogs, with rotation times from 4 to 10 years.

*Eucalyptus camaldulensis* was introduced into Vietnam in the 1930s (KHA *et al.*, 2003) and has become an important planting species both for large scale plantations and for small plantings on farms, along canals and roadsides. The total areas of eucalypt plantations in Vietnam was estimated to be 348,000 ha in 2001 (MARD, 2002), with *E. camaldulensis* and *E. urophylla*

the two most important species. Commercial plantations of *E. camaldulensis* in Vietnam are mainly in southern and central Vietnam. However, in parts of south-east and central Vietnam which experience a long rainy season, plantations of this species have been severely damaged by defoliation and shoot blight mainly caused by the fungi *Cryptosporiopsis eucalypti* and *Cylindrocladum quinqueseptatum* (THU *et al.*, 2000; OLD *et al.*, 2002).

Clonal plantations of *Eucalyptus* have been successful in many countries (COSSALTER and PYE-SMITH, 2003). Clonal forestry is practiced with *E. camaldulensis* as it is easily mass-propagated by stem cuttings. *Eucalyptus camaldulensis* is also important for hybrid breeding. Selected clones of inter-specific hybrid combinations between *E. camaldulensis* and other eucalypt species such as *E. urophylla* (KHA *et al.*, 2003) and *E. grandis* (GWAZE *et al.*, 2000) are commercially important in some countries.

Clonal forestry provides the opportunity to capture both additive and non-additive genetic effects, which should result in larger gains from selection than from sexually produced offspring (LIBBY and RAUTER, 1984). A successful clonal deployment program requires (i) knowledge of genetic parameters in order to determine best strategy for clonal testing and predicting selection gains from deployment of the best clones (WHITE, 1987; NAMKOONG *et al.*, 1988); and (ii) knowledge of genotype by environment interaction which is critical for testing design, selection and deployment in any tree improvement program (BURDON, 1977; NAMKOONG, 1981). Existence of genotype by environment interaction affects testing and selection in tree improvement programs, and may result in reduced overall genetic gains. Strategies to manage genotype by environment interactions include selecting environmentally stable genotypes that perform well in different environments or using different sets of clones in different environments.

Because of its importance in plantations worldwide, genetic improvement of *E. camaldulensis* has been undertaken in many countries. Provenance variation and provenance by site interaction in growth have been reported to be significant (OTEGBEYE, 1985; EMERY and LEDIG, 1987; CHUONG, 1992; PINYOPUSARERK *et al.*, 1996; KHA *et al.*, 2003; MAHMOOD *et al.*, 2003; VARGHESE *et al.*, 2008). In Vietnam, the best performing provenances of *E. camaldulensis* are Kennedy River, Morehead River and Laura River from northern Queensland and Katherine from Northern Territory (CHUONG, 1992; KHA *et al.*, 2003). Heritabilities of growth traits have been reported to be low to moderate and with significant genotype by environment interaction in Thailand (PINYOPUSARERK *et al.*, 1996), Pakistan (MAHMOOD *et al.*, 2003) and southern India (VARGHESE *et al.*, 2008).

The overall objective of our study was to develop a clonal forestry strategy for *E. camaldulensis* in Vietnam. This study was based on evaluation of growth and wood density in three clonal tests of *E. camaldulensis* at contrasting sites in southern, north-central and northern Vietnam. The clones tested originated from ortets from a provenance-progeny trial in southern Vietnam, and from plantations in southern and northern Vietnam. We esti-

mated genotypic parameters and their age trends, magnitude of genotype by environment interactions, and predicted selection gain of growth and the correlated responses for wood density and for growth at other sites. Furthermore, we tested whether clones originating from different provenance regions differ in their performance.

## Materials and Methods

### Genetic materials

A total of 172 clones were used to establish three clonal tests at three sites in Vietnam: Ham Thuan Nam in the south; Dong Hoi in north-central; and Ba Vi in the north. The planting material was 4-month old clonal ramets derived from rooted stem cuttings, raised in polyethylene bags in a soil-based potting mixture. Each trial tested 90–120 clones. Among these clones, 131 clones were selected from a progeny test of *E. camaldulensis* at Chon Thanh (latitude: 11°15'N, longitude: 106°38'E, mean annual rainfall 2000 mm) in the south, 30 phenotypically superior trees were selected from plantations at Chon Thanh and a further 11 superior trees from plantations at Ba Vi in the north (latitude: 21°08'N; longitude: 105°28'E; mean annual rainfall: 1700 mm). Numbers of commonly tested clones in different pairs of these trials are provided in Table 2. Assignment of clones to sites was based on the number of cuttings per clone available at the time of planting. Only 32 clones were represented at all three sites, because at the time of planting there were not enough ramets of all the clones for planting at all three sites.

The selection of clones from the provenance/progeny trial of 154 families at Chon Thanh was based on family and individual tree performance for growth and stem straightness, favouring families and trees without visible disease symptoms. The progeny trial at Chon Thanh was heavily affected by leaf spot and shoot blight diseases, mainly caused by *Cryptosporiopsis eucalypti* (OLD *et al.*, 2002). The clones were selected from 53 superior families from 12 natural provenances of *E. camaldulensis* from northern Queensland and Northern Territory (Table 1). Recent taxonomic study (MCDONALD *et al.*, 2009) indicated these provenances represent different sub-species (Table 1, Figure 1). The sub-species represented by provenance Laura River Crossing PDR (CSIRO seedlot 15825) was uncertain because of overlapping distributions of the different sub-species in that area (M. MCDONALD, personal communication). Provenance identity of the clones selected in commercial plantations was unavailable.

The provenances known to be represented can be grouped into three distinct geographic regions (Figure 1) according to latitude and longitude: (i) the Petford region in northern Queensland; (ii) the far north Queensland region; and (iii) the Katherine region in Northern Territory (Figure 1). The Petford region is slightly cooler and drier than the other two regions (Bureau of Meteorology, 2009).

### Location and trial description

Site conditions, silvicultural treatments and experimental design in the clonal tests are described in Table

3. The trial site at Ham Thuan Nam in the south is more suitable for good growth of tropical provenances of *E. camaldulensis*, with a hot and seasonally dry climate, sandy and deeper soil whereas the other two sites are colder in the winter, and with shallower rocky soils. The site conditions at Ham Thuan Nam, Dong Hoi and Ba Vi

are typical for the south-east coast, north-central and northern Vietnam, respectively. These trials used randomized row-column incomplete block designs generated by the computer program CycDesignN (WILLIAMS *et al.*, 2002) providing two-dimensional incomplete blocking (rows and columns) within replicates.

Table 1. – Provenance origin and sub-species information of *Eucalyptus camaldulensis* clones, number of clones and families (in parentheses) for each provenance and sub-species at different trials.

CSIRO seedlot number	Provenance location	Sub-species	Latitude	Longitude (E)	Trial		
					Ba Vi	Dong Hoi	Ham Thuan Nam
14346	Emu Ck Petford	<i>acuta</i>	17°21'S	144°57'	1(1)	2(1)	2(1)
14348	Emu Ck Petford	<i>acuta</i>	17°21'S	144°57'	1(1)	1(1)	0
14351	Emu Ck Petford	<i>acuta</i>	17°24'S	144°59'	3(1)	1(1)	2(1)
16720	Petford area	<i>acuta</i>	17°24'S	145°02'	1(1)	3(2)	4(2)
18242	Kennedy River	<i>acuta</i>	15°30'S	144°07'	3(1)	5(2)	5(2)
18275	Kennedy River	<i>acuta</i>	15°26'S	144°11'	19(9)	16(8)	17(7)
19010	Morehead River	<i>acuta</i>	15°02'S	144°00'	4(2)	8(5)	6(3)
16622	Fergusson R, Katherine	<i>obtusa</i>	14°05'S	131°58'	2(1)	4(2)	3(2)
18987	20 km W-SW Katherine	<i>obtusa</i>	14°33'S	132°04'	7(5)	12(6)	12(6)
15827	Kennedy Ck PDR	<i>simulata</i>	15°39'S	144°33'	3(3)	11(5)	10(5)
18276	Laura River	<i>simulata</i>	15°38'S	144°30'	17(10)	24(11)	23(12)
15825	Laura R Crossing PDR	<i>unknown</i>	15°44'S	144°41'	6(3)	8(6)	7(4)
	Chon Thanh plantation	<i>unknown</i>	11°23'N	106°36'	13	25	9
	Ba Vi plantation	<i>unknown</i>	21°08'N	105°28'	10	0	0
Total					90	120	100

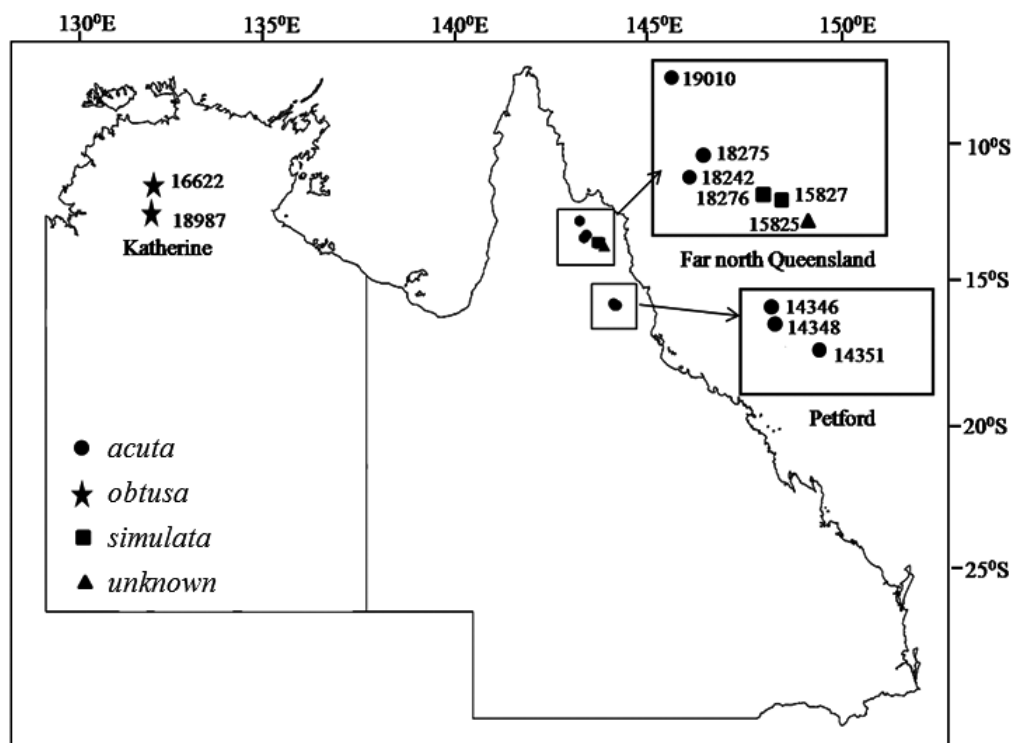


Figure 1. – Map of locations of original seedlots of clones of *Eucalyptus camaldulensis* from progeny trial at Chon Thanh, southern Vietnam.

**Table 2.** – Number of common clones for each region of *Eucalyptus camaldulensis* at different pairs of clonal trials and across all three trials.

Number of common clones in each pair of site	Number of clones in common				
	Far north Queensland	Petford	Katherine	Unknown	Total
Ham Thuan Nam – Dong Hoi	52	5	12	8	77
Ham Thuan Nam – Ba Vi	31	4	6	2	43
Dong Hoi – Ba Vi	36	3	8	10	57
Across 3 sites	22	3	5	2	32

**Table 3.** – Site conditions, silvicultural treatments and experimental design of the clonal trials.

	Ham Thuan Nam	Dong Hoi	Ba Vi
Latitude (N)	11°40'	17°28'	21°08'
Longitude (E)	108°54'	106°59'	105°28'
Altitude (m)	22	40	60
Soil type	Sandy alluvium	Ferralitic clay loam	Ferralitic clay loam
Soil depth (cm)	> 100	50 – 60	80 – 100
Annual rainfall (mm)	1400	2300	1700
Rainy season	June - October	August - February	April - October
Mean annual temp. (°C)	26.2	24.8	23.2
Mean daily maximum temp. of hottest month (°C)	33.7	34.3	31.8
Mean daily minimum temp. of coldest month (°C)	20.4	17.3	14.3
Planting time	August 2001	November 2002	July 2004
Site preparation	Herbicide treatment and manual preparation	Slash burned and ripped	Slash burned and ripped
Fertiliser (kg/ha)	2000 kg micro- organic fertilizer + 200 kg NPK	2200 kg cattle manure + 220 kg NPK	2200 kg cattle manure + 220 kg NPK
Design			
Replicates	8	8	8
Rows/replicate	10	10	10
Columns/replicate	10	12	9
Tree per plot	2	2	2
Spacing (m)	4 x 2.5	3 x 3	3 x 3

### Data collection

Height and DBH of all trees were measured at ages one, two and three years at all three sites and five years at Ham Thuan Nam and Dong Hoi. Foliar disease based on visible symptoms and irrespective to pathogen was evaluated at Dong Hoi at age three years using a four-class crown health score as follows: class 1 – major defoliation, class 2 – heavy leaf spots and/or defoliation of the lower crown, class 3 – good crown retention with few leaf spots and class 4 – little or no visible disease symptoms. The crown health evaluations were not performed at Ham Thuan Nam and Ba Vi because most of the trees in these two sites were healthy with little or no visible disease symptoms (i.e. class 4).

Wood basic density (DEN) and pilodyn penetration (PP) were evaluated after the final growth measurement at each site (age five years at Ham Thuan Nam and Dong Hoi, three years at Ba Vi). PP was measured on all trees having DBH larger than 5 cm using a Pilodyn 6J Forest, by removing a small section of bark at stem height of 1.3 m from ground and taking two Pilodyn shots on each tree, according to the method described by HANSEN (2000). The directions for Pilodyn shots were the same for all trees in all trials, one on the southern and one on the eastern side of the stem.

After measurement of DBH, HT and PP, one tree in each plot was systematically felled in the first 6 replicates at all sites to provide samples for DEN measure-



ment. On each felled tree, a five cm disk was sampled at a height of 1.3 m from ground (just above the pilodyn shot points). The disks were immediately stored in plastic bags and later debarked and water soaked. DEN was determined by the water displacement method (OLESEN, 1971), with two weights for each sample: weight of water displaced by immersion of disk, which indicates fresh volume of the sample ( $w_1$ ); and oven dry weight ( $w_2$ ). DEN was then calculated as:

$$DEN = \frac{w_2}{w_1} \times 1000 \text{ (kg m}^{-3}\text{)}$$

#### Data analysis

Statistical data analyses were implemented using ASReml 2.0 software (GILMOUR *et al.*, 2006) and the following mixed linear model equation was used in the analyses based on individual tree observations:

$$\mathbf{y} = \mathbf{X}_B \mathbf{b} + \mathbf{X}_S \mathbf{s} + \mathbf{Z}_W \mathbf{w} + \mathbf{Z}_N \mathbf{n} + \mathbf{Z}_P \mathbf{p} + \mathbf{Z}_C \mathbf{c} + \mathbf{e} \quad (1)$$

where  $\mathbf{y}$  is the vector of individual tree observations,  $\mathbf{b}$  is the vector of fixed replicate effect,  $\mathbf{s}$  is the vector of provenance region effect,  $\mathbf{w}$  is the vector of random row within replicate effect,  $\mathbf{n}$  is the vector of random column within replicate effect,  $\mathbf{p}$  is the vector of random plot (row by column) within replicate effects,  $\mathbf{c}$  is the vector of random clone effects, and  $\mathbf{e}$  is the vector of random residuals.  $\mathbf{X}_B$ ,  $\mathbf{X}_S$ ,  $\mathbf{Z}_W$ ,  $\mathbf{Z}_N$ ,  $\mathbf{Z}_P$ , and  $\mathbf{Z}_C$  are incidence matrices relating  $\mathbf{b}$ ,  $\mathbf{s}$ ,  $\mathbf{w}$ ,  $\mathbf{n}$ ,  $\mathbf{p}$ , and  $\mathbf{c}$  to  $\mathbf{y}$ . Family effects were not modeled because there were only one to four clones per family and 41 clones were selected in plantations without any family identity. Furthermore, because the purpose of our study was to consider implications for deployment of clonal forestry, we were mainly interested in total clonal effects.

The random effects were assumed to follow an independent multivariate normal distribution with zero means and (co)variances as follows:

$$VAR \begin{bmatrix} \mathbf{w} \\ \mathbf{n} \\ \mathbf{p} \\ \mathbf{c} \\ \mathbf{e} \end{bmatrix} \sim \begin{bmatrix} \mathbf{W} \otimes \mathbf{I} & \mathbf{0} & \mathbf{0} & \mathbf{0} & \mathbf{0} \\ \mathbf{0} & \mathbf{N} \otimes \mathbf{I} & \mathbf{0} & \mathbf{0} & \mathbf{0} \\ \mathbf{0} & \mathbf{0} & \mathbf{P} \otimes \mathbf{I} & \mathbf{0} & \mathbf{0} \\ \mathbf{0} & \mathbf{0} & \mathbf{0} & \mathbf{C} \otimes \mathbf{I} & \mathbf{0} \\ \mathbf{0} & \mathbf{0} & \mathbf{0} & \mathbf{0} & \mathbf{R} \otimes \mathbf{I} \end{bmatrix}$$

where  $\mathbf{0}$  is a null matrix,  $\mathbf{W} = \{\sigma_{w_i w_j}\}$ ,  $\mathbf{N} = \{\sigma_{n_i n_j}\}$ ,  $\mathbf{P} = \{\sigma_{p_i p_j}\}$ ,  $\mathbf{C} = \{\sigma_{c_i c_j}\}$  and  $\mathbf{R} = \{\sigma_{e_i e_j}\}$  are the row, column, plot, clone and residual variance-covariance matrices between trait  $i$  and  $j$ , denoting variance when  $i = j$ . For density, where only one tree per plot was measured, the plot effect was omitted. To ensure that the variance-covariance matrix was positive definite, restrictions were in some cases applied to the parameters. The significance of fixed provenance region effects was assessed by F-tests based on univariate analysis using model 1.

Genotypic correlations between sites were estimated based on REML analysis, by treating measurements from different sites as different traits based on model 1. In  $\mathbf{R}$ , all off-diagonal elements were assumed to be zero for a combination of traits measured in different trials. The aims of this analysis were to estimate magnitude and significance of genotypic correlations between sites.

Log-likelihood ratio tests were used to test whether the correlations were different from one and whether genotypic correlations between different pair of sites differed significantly.

#### Genotypic parameters

Clonal repeatability at single site ( $H_C^2$ ), coefficient of genotypic variation ( $CV_G$ ) and genotypic correlation ( $r_G$ ) were calculated as:

$$H_C^2 = \frac{\sigma_c^2}{\sigma_c^2 + \sigma_p^2 + \sigma_e^2}$$

$$CV_G = \frac{\sigma_c}{\bar{X}} \times 100 \text{ (\%)}$$

$$r_G = \frac{\sigma_{c_1 c_2}}{\sigma_{c_1} \sigma_{c_2}}$$

where  $\sigma_c^2$  is the clone variance,  $\sigma_p^2$  is the plot within replicate within site variance (row by column), which is equal to zero in case of single tree-plot,  $\sigma_e^2$  is the residual variance,  $\bar{X}$  is the phenotypic mean value of trait,  $\sigma_{c_1 c_2}$  is the genotypic covariance between two traits, respectively;  $\sigma_{c_1}$ ,  $\sigma_{c_2}$  are the genotypic standard deviations of trait 1 and trait 2. Standard errors of the estimates of repeatabilities and genotypic correlations were calculated using a standard Taylor series approximation (GILMOUR *et al.*, 2006).

Predicted selection gain was calculated according to MULLIN and PARK (1992) as:

$$R = i_{n,N} H_C^2 \sigma_{\bar{P}}$$

Where  $R$  is the predicted selection gain,  $i_{n,N}$  is the selection intensity based on selection of  $n$  from  $N$  tested. Values for  $i_{n,N}$  were derived from BECKER (1992).  $\sigma_{\bar{P}}$  is the phenotypic standard deviation of clone mean and  $H_C^2$  is the clone-mean repeatability calculated as:

$$H_C^2 = \frac{\sigma_c^2}{\sigma_p^2} = \frac{\sigma_c^2}{\sigma_c^2 + \frac{\sigma_p^2}{r} + \frac{\sigma_e^2}{nr}}$$

Where  $r$  is number of replications and  $n$  is the number of ramets per plot.

The correlated response (trait Y resulting from selection for trait X at single site or the same trait at one site resulting from selection at the another site) was estimated according to FALCONER and MACKAY (1996) as:

$$CR_Y = i_{n,N} H_{\bar{C}_X} H_{\bar{C}_Y} r_{G_{XY}} \sigma_{\bar{P}_Y}$$

where  $CR_Y$  is the correlated response,  $H_{\bar{C}}$  is the square root of clone-mean repeatability,  $r_G$  is the genotypic correlation, and X and Y are selection trait and response trait, respectively.

#### Stability analyses of clones

Analysis of stability was based on a reduced data set including 32 clones common for all sites. Ecovalence proposed by WRICKE (1962) estimates the contribution of individual clones to the total interaction sum of square. Clones with low ecovalence values are considered relatively stable among a given set of clones. The ecovalence

values were calculated using clone means at each site following the formula proposed by WRICKE (1962):

$$W_i = \sum_j (\bar{X}_{ij} - \bar{X}_{i.} - \bar{X}_{.j} + \bar{X})^2$$

where  $W_i$  is the ecovalence of clone  $i$ ,  $\bar{X}_{ij}$  is the mean value of clone  $i$  at site  $j$ ,  $\bar{X}_{i.}$  and  $\bar{X}_{.j}$  are the marginal means of clone  $i$  and site  $j$ , respectively, and  $\bar{X}$  is the overall mean.

SHUKLA's stability variance (SHUKLA, 1972) is linearly related to the ecovalence (KANG *et al.*, 1987) and ranking of clones by the two parameters were identical. Therefore, the ecovalence values were tested for statistical significance using the formula described by SHUKLA (1972):

$$F^* = \hat{\sigma}_i^2 / \hat{\sigma}_0^2$$

where:  $\hat{\sigma}_i^2 = [t(t-1)W_i - \sum_i W_i] / (s-1)(t-1)(t-2)$  (after KANG *et al.*, 1987),  $\hat{\sigma}_0^2 = \sum_i \sum_j \sum_k (\bar{X}_{ijk} - \bar{X}_{ij})^2 / str(r-1)$  and  $\bar{X}_{ijk}$  is the mean value of clone  $i$  in the site  $j$  at replicate  $k$ ,  $s$  is the number of sites,  $t$  is the number of clones and  $r$  is the number of replicates at each site.  $F^*$  have an approximate  $F$  distribution on  $(s-1)$  and  $st(r-1)$  degrees of freedom.

## Results

### Growth performance and provenance region variation

In all clonal tests in southern, north-central and northern Vietnam, survival through to age three or five years ranged from 71.9 to 86.1% (Table 4). The lower survival in the trial at Ham Thuan Nam was due to grazing damage by wild rabbits and livestock shortly after planting. The best DBH was recorded at Ham Thuan Nam where DBH at age three years was 9.3 cm, followed by Ba Vi (7.2 cm) and Dong Hoi (6.4 cm) (Table 4). DEN was relatively similar at age five years at Ham Thuan Nam and Dong Hoi, and ranged from 528 to 540 kg m<sup>-3</sup>. Average DEN at age three years at Ba Vi was 502 kg m<sup>-3</sup>.

Provenance regions did not differ statistically for either growth (age 3 years) or DEN at Ham Thuan Nam and Dong Hoi. At Ba Vi, significant difference between provenance regions were recorded for all traits (Table 5), clones from Katherine region were the best in growth whereas clones from Petford region were the worst. Clones from far north Queensland region had highest DEN while the other regions had similar DEN (Table 5).

Table 4. – Survival, mean value, clonal repeatabilities ( $H_C^2$ ), clone mean repeatabilities ( $H_C^2$ ) and coefficient of genotypic variation ( $CV_G$ ) for traits at different ages in each trial.

Site	Age	Trait	Survival (%)	Mean $\pm$ se	$H_C^2 \pm$ se	$H_C^2 \pm$ se	$CV_G$ (%)
Ham Thuan Nam	1	HT1 (m)	74.3	3.4 $\pm$ 0.1	0.30 $\pm$ 0.04	0.78 $\pm$ 0.04	13.5
	2	HT2 (m)	73.8	6.9 $\pm$ 0.2	0.33 $\pm$ 0.04	0.78 $\pm$ 0.04	10.1
	3	HT3 (m)	73.3	9.3 $\pm$ 0.2	0.38 $\pm$ 0.04	0.83 $\pm$ 0.03	9.7
	5	HT5 (m)	71.9	14.1 $\pm$ 0.3	0.42 $\pm$ 0.05	0.85 $\pm$ 0.03	9.9
	1	DBH1 (cm)		2.8 $\pm$ 0.2	0.27 $\pm$ 0.04	0.75 $\pm$ 0.04	18.5
	2	DBH2 (cm)		6.9 $\pm$ 0.3	0.31 $\pm$ 0.04	0.78 $\pm$ 0.04	14.9
	3	DBH3 (cm)		9.3 $\pm$ 0.3	0.36 $\pm$ 0.04	0.82 $\pm$ 0.03	13.7
	5	DBH5 (cm)		13.4 $\pm$ 0.4	0.42 $\pm$ 0.04	0.86 $\pm$ 0.03	13.6
	5	DENSITY (kg m <sup>-3</sup> )		539.7 $\pm$ 4.7	0.78 $\pm$ 0.03	0.95 $\pm$ 0.01	6.7
	5	PILODYN (mm)		14.6 $\pm$ 0.1	0.65 $\pm$ 0.04	0.94 $\pm$ 0.01	8.4
Dong Hoi	1	HT1 (m)	94.2	3.8 $\pm$ 0.1	0.09 $\pm$ 0.03	0.46 $\pm$ 0.08	7.0
	2	HT2 (m)	91.7	6.3 $\pm$ 0.1	0.16 $\pm$ 0.03	0.60 $\pm$ 0.06	6.8
	3	HT3 (m)	90.9	8.1 $\pm$ 0.2	0.18 $\pm$ 0.03	0.63 $\pm$ 0.05	6.3
	5	HT5 (m)	86.1	10.5 $\pm$ 0.3	0.22 $\pm$ 0.04	0.69 $\pm$ 0.05	7.6
	1	DBH1 (cm)		2.8 $\pm$ 0.1	0.06 $\pm$ 0.02	0.38 $\pm$ 0.10	7.2
	2	DBH2 (cm)		4.7 $\pm$ 0.2	0.14 $\pm$ 0.03	0.58 $\pm$ 0.06	9.0
	3	DBH3 (cm)		6.4 $\pm$ 0.2	0.20 $\pm$ 0.03	0.68 $\pm$ 0.05	9.8
	5	DBH5 (cm)		8.8 $\pm$ 0.2	0.26 $\pm$ 0.03	0.74 $\pm$ 0.04	12.5
	5	DENSITY (kg m <sup>-3</sup> )		528.0 $\pm$ 3.9	0.71 $\pm$ 0.03	0.93 $\pm$ 0.01	5.1
	5	PILODYN (mm)		14.8 $\pm$ 0.1	0.56 $\pm$ 0.04	0.91 $\pm$ 0.01	6.6
Ba Vi	1	HT1 (m)	93.1	3.8 $\pm$ 0.1	0.26 $\pm$ 0.04	0.80 $\pm$ 0.03	10.4
	2	HT2 (m)	90.7	6.8 $\pm$ 0.1	0.32 $\pm$ 0.04	0.82 $\pm$ 0.03	12.1
	3	HT3 (m)	86.2	9.5 $\pm$ 0.2	0.39 $\pm$ 0.04	0.87 $\pm$ 0.02	13.3
	1	DBH1 (cm)		2.9 $\pm$ 0.1	0.23 $\pm$ 0.03	0.78 $\pm$ 0.03	14.4
	2	DBH2 (cm)		5.2 $\pm$ 0.1	0.39 $\pm$ 0.04	0.82 $\pm$ 0.03	16.4
	3	DBH3 (cm)		7.2 $\pm$ 0.2	0.41 $\pm$ 0.04	0.89 $\pm$ 0.02	17.9
	3	DENSITY (kg m <sup>-3</sup> )		502.3 $\pm$ 4.9	0.77 $\pm$ 0.03	0.95 $\pm$ 0.01	6.5
	3	PILODYN (mm)		15.5 $\pm$ 0.1	0.66 $\pm$ 0.04	0.94 $\pm$ 0.01	8.0

**Table 5.** – Diameter at breast height (DBH), height (HT) at age 3 years and wood basic density (DEN) of different provenance regions of *Eucalyptus camaldulensis* at three trials and crown health at Dong Hoi.

Provenance regions	Ham Thuan Nam			Dong Hoi				Ba Vi		
	DBH (cm)	HT (m)	DEN (kg m <sup>-3</sup> )	DBH (cm)	HT (m)	DEN (kg m <sup>-3</sup> )	Crown health (1-5)	DBH (cm)	HT (m)	DEN (kg m <sup>-3</sup> )
Petford	8.8	9.1	523.0	6.2	8.0	521.3	3.2	6.3	8.8	476.4
Far north Queensland	9.3	9.3	539.4	6.5	8.2	531.3	3.5	7.2	9.5	502.3
Katherine	9.5	9.1	543.7	6.4	8.2	518.4	3.2	8.4	10.6	477.3
p-value for difference between provenance regions	0.50	0.75	0.58	0.23	0.79	0.22	<0.01	<0.01	0.02	0.01

At Dong Hoi, 85% of trees were healthy (crown health class 3–4) and significant differences in crown health were recorded at region ( $p < 0.01$ ) (Table 5) level. Clones from the far north Queensland region had the best crown health while those from the Petford and Katherine regions were similar.

#### Genotypic parameters and correlations

Clonal repeatabilities for DBH and HT increased with ages at all three sites (Table 4). At the same age, clonal repeatabilities estimated at Ham Thuan Nam and Ba Vi were relatively similar and higher than those at Dong Hoi. The trend of increasing repeatabilities with ages for growth traits was clearer at Dong Hoi compared to the other two sites. At ages three and five years, clonal repeatabilities for growth traits were from 0.18 to 0.42; for directly measured DEN from 0.71 to 0.78, while those for PP ranged from 0.56 to 0.66 (Table 4). Clone mean repeatabilities at age three and five years were in the range from 0.68 to 0.89 for growth traits, and from 0.91 to 0.95 for both DEN and PP at all sites (Table 4).

Coefficients of genotypic variation were generally high for growth traits at all ages, ranging from 7.2 to 18.5% for DBH, and from 6.3 to 13.5% for HT. Coefficients of genotypic variation for DEN and PP were lower than those observed for growth traits, ranging from 5.1 to 6.7% for DEN, and from 6.6 to 8.4% for PP (Table 4).

Genotypic correlations between measurements of growth traits at earlier ages and final measurement at age 5 years at Ham Thuan Nam and Dong Hoi were

generally high, particularly measurements between ages from two years ( $r_G = 0.84\text{--}0.89$ ) (Table 6). The genotypic correlations between DBH and DEN, and HT and DEN at the three sites ranged from  $-0.24$  to  $0.21$  (Table 7) and did not differ significantly from zero. The genotypic correlations between DEN and PP were from  $-0.89$  to  $-0.95$  (Table 7) and were highly significant at  $p < 0.001$ .

#### Genotype by environment interaction and stability of clones

Genotypic correlations for growth traits between each pair of sites at age three years ranged from 0.32 to 0.56, while those between Ham Thuan Nam and Dong Hoi at age 5 years were from 0.45 to 0.48, with large standard errors (Table 8). Genotypic correlations between pairs of sites for DEN and PP (5 years at Ham Thuan Nam and Dong Hoi, 3 years at Ba Vi) ranged from 0.72 to 0.88 with small standard errors (Table 8). Log-likelihood ratio tests indicated all the genotypic correlations were significantly different from one, but the genotypic correlations between pairs of sites were not statistically different from one another, for any trait.

The above results indicated that genotype by environment interaction was important for growth traits but not for DEN. Therefore, stability of the 32 clones tested at all three sites was calculated for DBH only, at age 3 years. The mean ecovalence value for DBH was 3.1%, ranging from 0.1% to 9.2%. SHUKLA's test indicated that clones did not contribute equally to total interac-

**Table 6.** – Age-age genotypic correlations for diameter at breast height (DBH) and height (HT) at each trial.

Traits	Age	Ham Thuan Nam			Dong Hoi			Ba Vi	
		2	3	5	2	3	5	2	3
DBH	1	0.94 ± 0.02	0.85 ± 0.04	0.73 ± 0.06	0.95 ± 0.02	0.86 ± 0.11	0.63 ± 0.13	0.93 ± 0.02	0.86 ± 0.03
	2		0.95 ± 0.01	0.89 ± 0.03		0.97 ± 0.02	0.88 ± 0.04		0.92 ± 0.01
	3			0.96 ± 0.01			0.96 ± 0.01		
HT	1	0.87 ± 0.03	0.79 ± 0.04	0.68 ± 0.07	0.92 ± 0.04	0.74 ± 0.11	0.62 ± 0.11	0.93 ± 0.02	0.82 ± 0.02
	2		0.95 ± 0.01	0.88 ± 0.03		0.94 ± 0.03	0.84 ± 0.06		0.93 ± 0.01
	3			0.92 ± 0.02			0.90 ± 0.03		

Table 7. – Genotypic correlations between diameter at breast height (DBH), height (HT), wood basic density (DEN) and pilodyn penetration (PP) at Ham Thuan Nam, Dong Hoi and Ba Vi.

Site	Trait	HT	DEN	PP
Ham Thuan Nam (5 years)	DBH	0.85 ± 0.03	0.17 ± 0.07	0.09 ± 0.11
	HT		0.21 ± 0.11	-0.04 ± 0.11
	DEN			-0.89 ± 0.03
Dong Hoi (5 years)	DBH	0.87 ± 0.04	-0.16 ± 0.11	0.18 ± 0.11
	HT		0.07 ± 0.11	-0.05 ± 0.12
	DEN			-0.95 ± 0.01
Ba Vi (3years)	DBH	0.90 ± 0.03	-0.24 ± 0.11	0.33 ± 0.11
	HT		0.01 ± 0.12	0.18 ± 0.12
	DEN			-0.90 ± 0.03

Table 8. – Genotypic correlations between sites and standard errors for diameter (DBH), height (HT), wood basic density (DEN) and pilodyn penetration (PP) measured at age 3 and 5 years (DEN and PP at Ba Vi were measured at age 3 years).

Trait	Site	Age 3 years		Age 5 years	
		Ham Thuan Nam	Dong Hoi	Ham Thuan Nam	Dong Hoi
DBH	Dong Hoi	0.47 ± 0.13		0.48 ± 0.12	
	Ba Vi	0.43 ± 0.14	0.52 ± 0.14		
HT	Dong Hoi	0.56 ± 0.13		0.45 ± 0.13	
	Ba Vi	0.32 ± 0.16	0.42 ± 0.17		
DEN	Dong Hoi			0.76 ± 0.06	
	Ba Vi			0.72 ± 0.08	0.88 ± 0.04
PP	Dong Hoi			0.81 ± 0.05	
	Ba Vi			0.74 ± 0.07	0.88 ± 0.04

tion sum of squares; 11 clones contributed significantly ( $p < 0.05$ ) and they accounted for 70% of the total interaction (Figure 2).

#### Selection gains and correlated responses

Selection gains for DBH were from 18.4% to 27.1% at a selection proportion of 10%, and from 21.6% to 31.8% at selection proportion of 5% at individual sites (Table 9). Correlated responses in DEN from selection for DBH were from -3.1% to 2.2%. The average indirect responses in DBH at two sites resulting from selection at the third site were from 10.1% to 13.2% at 5% or 10% selection proportion, respectively; equivalent to only 40–60% of the potential gains obtained from direct selection at individual sites.

## Discussion

#### Growth performance and genotype by environment interaction

Mean height annual increments to age 3 years at the three trial sites in our study were 2.5–3 m (Table 4). This is relatively similar to height increments recorded

in genetic trials of *E. camaldulensis* in southern India on sites receiving lower rainfall, ranging from 950 mm to 1150 mm (VARGHESE *et al.*, 2008). However, the growth rate in our trials was slower than those reported in Thailand on the sites of fertile sandy soils with rainfall of 1100–1200 mm where annual height increment to age 2 years was about 4 m (PINYOPUSARERK *et al.*, 1996).

Although height growth increments to age 3 years did not differ greatly between our three trials, trees at Ham Thuan Nam in the south maintained much faster growth over the following 2 years, reaching a mean height of 14 m, compared to 10.5 m at Dong Hoi in the north-central Vietnam (Table 4). The significant differences in growth between sites and strong genotype by environment interaction in growth of individual clones of *E. camaldulensis* can be explained mainly by the differences in soil conditions. The experimental sites at Ba Vi and Dong Hoi are typical of most hill-sites in northern and north-central Vietnam with shallow, somewhat stony, yellow ferralitic clay loam soils with pH 3.5–4.5, low in phosphorus and potassium (CHIEU and THUAN, 1996), while the soil at Ham Thuan Nam is sandy alluvium, light in texture, less acid, deeper and more fertile



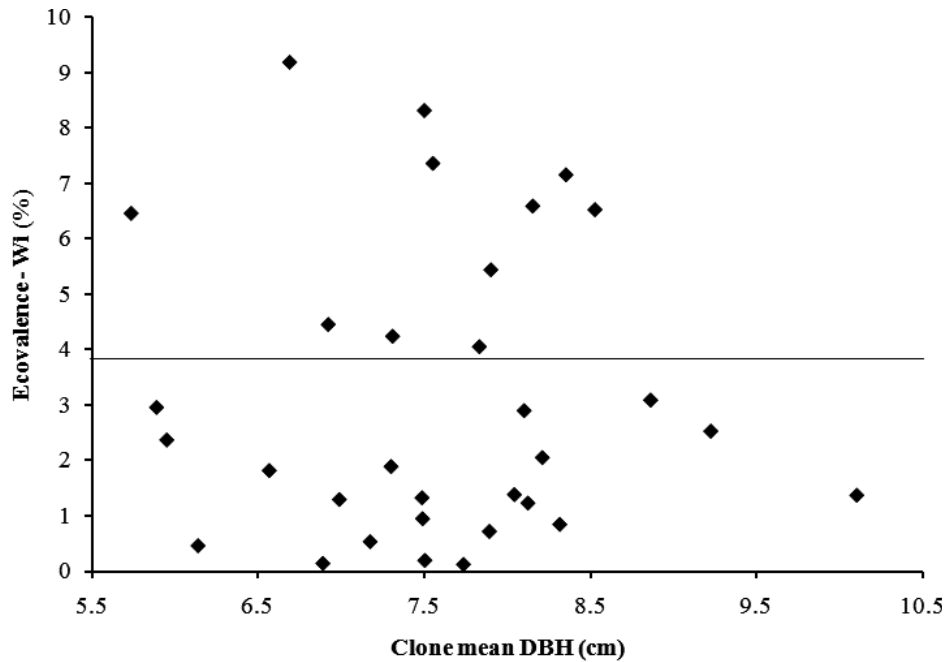


Figure 2. – Ecovalence against clone mean of 32 clones of *Eucalyptus camaldulensis* across three sites in Vietnam (clones above horizontal line showed significant interaction).

Table 9. – Selection gains for diameter at breast height (DBH) at different selection proportions in each trial, correlated responses in wood basic density (DEN) at the same site and DBH at other trials (selection ages were 5 years at Ham Thuan Nam and Dong Hoi and 3 years at Ba Vi).

Site	Selection proportion (%)	Direct response (%)	Correlated response in DEN (%)	Average indirect response in DBH at other sites (%)
Ham Thuan Nam	5	25.9	2.2	12.2 (10.9-13.4)
	10	22.1	1.9	10.4 (9.3-11.5)
Dong Hoi	5	21.6	-1.4	13.2 (11.3-15.1)
	10	18.4	-1.2	11.3 (9.7-12.7)
Ba Vi	5	31.8	-3.1	11.8 (11.3-12.3)
	10	27.1	-2.7	10.1 (9.7-10.5)

than the other two locations. Additionally, the lower temperatures as well as lower light intensities in winter months in northern and north-central Vietnam (DAC and TOAN, 1993) might further reduce growth at these sites.

The level of disease in our trials appeared not to have had a major impact on growth in any of the trials. Therefore, it would not be a major cause of strong genotype by environment interaction in growth traits. The trial site at Dong Hoi has high rainfall (2300 mm) and mean daily minimum temperature of the coldest month is about 17°C, indicating high risk of leaf spots and shoot blight disease caused by fungal pathogens *C. eucalypti* and *C. quinqueseptatum* in this area according to BOOTH *et al.* (2000). The low level of disease at Dong Hoi may be explained as clones were selected from progeny trial and plantation at Chon Thanh in the south, a high-rainfall, disease prone location (BOOTH *et al.*, 2000; OLD

*et al.*, 2002), so they had been pre-screened for disease resistance. The relatively low annual rainfalls and long dry season at the southern site, and the cool and dry winter at the northern site probably explain the low levels of disease symptoms in these sites. Clones selected at Ba Vi in the north were not tested in the southern and north-central trials, where they might have proved more susceptible to disease.

Adaptability of clones from different provenance regions to local conditions may also contribute to genotype by environment interaction. In a growth chamber experiment, GIBSON *et al.* (1995) reported that under cool and water-limited conditions, seedlings from dry tropic region in Katherine had better root development and better water-use efficiency compared to seedlings from more humid tropic regions. This may explain the better growth performance of clones from the Katherine

region in the trial in northern Vietnam, which experiences a cool and dry winter. Superior growth of the Katherine provenance in northern Vietnam was also reported by CHUONG (1992).

In contrast to growth traits, the genotypic correlations between sites for wood density were strong, either measured directly (DEN) or indirectly (PP). The results indicate that clonal rankings for density are more stable across sites than are rankings for growth traits. The mean basic density at Ham Thuan Nam ( $538.7 \text{ kg m}^{-3}$ ) was not much different from that obtained at Dong Hoi ( $528.0 \text{ kg m}^{-3}$ ) at the same age while large differences in growth were recorded. The results agreed well with previous studies in other eucalypt species, which found little genotype by environment interaction in wood density (LIMA *et al.*, 2000; MUNERI and RAYMOND, 2000; OSORIO *et al.*, 2001).

Significant genotype by environment interaction for growth traits in *E. camaldulensis* have also been reported at the provenance, family and/or clonal level in India and Thailand (PINYOPUSARERK *et al.*, 1996; VARGHESE *et al.*, 2008). It appears advisable that different sets of tested clones should be used in different regions in Vietnam to maximize productivity of plantations. In northern and north-central Vietnam, growth of *E. camaldulensis* is poorer than that of *E. urophylla* (KHA, 2003; KHA *et al.*, 2003). Commercial plantations of *E. camaldulensis* in these regions are not common, although the species is widely planted along roadsides, canal banks and farm boundaries. However, the best clones of *E. camaldulensis* selected and tested in these areas can be used in hybrid breeding programs with *E. urophylla* to produce inter-specific hybrid combinations targeted for these regions (KHA *et al.*, 2003).

#### Genotypic parameters

Because clones used in our study were selected within best-performing families in the progeny trial and phenotypically selected in the commercial plantation, the genotypic parameters estimated may not be applicable to the original provenances and regions. However, results from our study are comparable to values reported by VARGHESE *et al.* (2008) where individual clonal repeatability for growth traits ranged from 0.24 to 0.62 from three clonal trials in southern India at age 3 years, and was 0.67 for DEN in one of the trial. Narrow-sense within provenance heritability for growth traits in *E. camaldulensis* estimated from provenance-progeny trials was in the range from 0.06 to 0.20 (PINYOPUSARERK *et al.*, 1996; MAHMOOD *et al.*, 2003; VARGHESE *et al.*, 2008). The high and stable clonal repeatabilities for growth traits at Ham Thuan Nam suggest clones are highly differentiated by age one year under favorable conditions, while slower growth at Dong Hoi delayed the differentiation between clones until later ages. The genotypic correlations between measurements at earlier ages and those at age five years for growth traits were strong, especially from age two years, suggesting that selection at age two years should give reliable clonal rankings, enabling rapid deployment of the best clones from future clonal testing programs.

#### Selection gains and correlated responses

The substantial gains from selection for DBH indicate high potential for improving productivity of *E. camaldulensis* plantations through selection of best clones and vegetative propagation for planting at certain sites in Vietnam. This was further confirmed by result from a second clonal test at Ham Thuan Nam which showed that stem volume of selected clones was significantly ( $p < 0.001$ ) greater than that of a local commercial seedling control. Stem volume of the 5 best clones was 170% greater than the seedling control at age 4.5 years (KIEN, unpublished data). This trial tested 28 clones that were selected based on 1-year HT rankings from the clonal test at the same site reported in this study. The genotypic correlation for DBH for these 28 clones between the two trials at final measurement (5 years for the first trial and 4.5 years for the second trial) was 0.86. This suggests that selection for growth can be made as early as at age 1 year in southern Vietnam, although further screening for disease resistance seems advisable, especially before deploying clones to wetter and disease-prone regions.

Because of strong clone by site interaction for growth, in order to obtain maximum potential selection gains in growth traits, different sets of the best tested clones should be used in different regions. Using clones selected from one region for planting in the other regions is not recommended because only 40–60% of the maximum potential gains in DBH could be obtained compared to selection locally. Another alternative is to use environmentally stable and fast-growing clones in all regions. As an example, taking the best 3 clones (clones located on the lower right hand side of Figure 2) that were stable and had good growth across sites out of 32 common clones (a selection proportion of 10%), the gain from selection of these three clones was from 15% to 26%, which is about 80–95% of the potential gains obtained from selection locally at the same selection proportion.

Because of weak correlations between DBH and DEN, it is possible to select clones with both rapid growth and reasonably high DEN. Wood basic density of *E. camaldulensis* at age five years in our study was in the range  $530\text{--}540 \text{ kg m}^{-3}$  and within the optimum DEN range for pulpwood which is between  $500\text{--}550 \text{ kg m}^{-3}$  (DEAN, 1995). Kraft pulp yield of *E. camaldulensis* is about 43–46% at age 4–8 years (PISUTTIPICHED *et al.*, 2003), lower than many other eucalypt species. Therefore, breeding of *E. camaldulensis* in Vietnam for pulpwood plantations should emphasize improving wood volume, pulp yield and disease resistance while maintaining optimum DEN for pulp production.

#### Conclusions and implications for tree improvement

*Eucalyptus camaldulensis* performed well in southern Vietnam at a trial site with relatively low rainfall and more fertile soil, while growth was slower in north and north-central Vietnam on shallower and less fertile soils.

Clonal repeatability of growth traits for *E. camaldulensis* was moderate and increased with age from one to

five years. Repeatabilities for wood density were high either based on direct (DEN) or indirect measurement (PP). Clone mean repeatabilities were high for all traits suggesting that substantial gain can be achieved for all traits. Age-age genotypic correlations in growth traits were strong, suggesting that early selection for growth at age one or two years could reduce time requirements for identifying *E. camaldulensis* clones for deployment. However further testing of selected clones for disease resistance in disease-prone environments, before commercial deployment, appears warranted.

Genotype by environment interaction was strong for growth traits, indicating it is practically important for growth, and therefore different clones should be used on different sites to maximize selection gains for growth traits. Clones did not contribute equally to total interaction variance in DBH. The clone by site interaction was low for DEN and PP, suggesting clonal ranking in DEN will remain nearly constant across sites and the interaction effects can be ignored.

Selection gain for DBH was high at age three or five years with minor effects on DEN. Selection for growth traits should emphasize local selection, or selection of fast-growing, stable clones, in order to obtain maximum potential gain. Use of all selected clones from one region for planting in other regions is not recommended as only 40–60% of potential gains in growth were achieved.

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