

A comparison of deterministically predicted genetic gains with those realised in a South African *E. grandis* breeding program

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Synopsis

Tree breeders attempt to predict the genetic gains which are likely to be achieved through selection and breeding of new generations, using stochastic or deterministic modelling. There are many factors which may cause a discrepancy between the predicted and realised genetic gains. Often the predictions for genetic gains are based on single trait selection, whereas in reality, the breeding tends to be multi-trait in nature. The violation of Hardy-Weinberg conditions, assumptions regarding out crossing and relatedness, assumptions regarding the effect of the interaction between the environment and the genotype and numerous possible errors in the process of breeding, all could result in unexpected discrepancies between the realised and predicted genetic gains.

A series of genetic gains trials containing representatives of three generations of *Eucalyptus grandis* selections, are compared with the view to verifying the effectiveness of the *E. grandis* breeding program. Genetic gains of the F₃ (third generation of pedigreed progeny) over the F₂ generation (second generation of pedigreed progeny) were 15% for tree growth (volume). A comparison between F₂ and P₀, revealed an improvement of between 20 and 33% for growth. This exercise highlighted complexities of modelling the predicted genetic gains of assimilated genetic breeding trials.

The predictions of genetic gains did deviate (in both directions) from those realised, although these deviations may be explained as functions of imperfect modelling. On average, however, the predicted genetic gains for tree volume over three generations were 13% between generations, whereas the average realised genetic gain in the genetic gains trial, was 14%. It is therefore assumed that the *E. grandis* breeding population is indeed performing as expected, following classical tree breeding assumptions.

Introduction

The *Eucalyptus grandis* breeding program of the CSIR has been based on classical quantitative genetic breeding assumptions. These assumptions include those of the Hardy-Weinberg population (Hardy 1908; Weinberg 1908), i.e.:

- Applies to a large population
- Absence of selection
- Random mating, i.e. no inbreeding/ selfing

The CSIR *E.grandis* F₃ breeding population was in part derived from selections out of plantations (approximately 44%), and in part derived from other breeding programs (20%), and the remainder derived from provenance imports directly from Australia (36%) (**Figure 1**).

Thirty nine percent of the “F₂” breeding population was derived from the “South African Plantation series”. The P₀, F₁, F₂ generations of the South African population provides us with the opportunity to verify our genetic progress, through genetic gains trials, and thereby determine whether the breeding is performing as would be expected, given the Hardy-Weinberg and other assumptions.

Confidence in our ability to genetically improve *E.grandis*, and in the accuracy of deterministic models predicting the consequences of our breeding endeavours, are essential for modelling and predicting the economic impact of further genetic improvement.

Materials and Methods

The “South African Population” (plantation origin) breeding lines with the F₁ generation (‘SSO’-series), F₂ (‘A’-series) and F₃ (‘B’ -series) were studied as the two breeding cycles over three generations.

The selection process of the South African population and details from P₀ to F₃, are presented in **Table 1**. These details and parameters (as presented in **Figures 2, 3 and 4**) were used as input variables in the deterministic modelling algorithm, G-Assist Version 4 (Verryn & Snedden 1998; Verryn & Snedden 2000), in order to predict the genetic gains which should be achieved in the F₃ from selection in the F₂,

as opposed to those predicted in the F_2 over that of the F_1 . The G-Assist algorithm is an algorithm which has been used in various deterministic studies of breeding strategies (Shelbourne *et al.* 2007; Snedden *et al.* 2000; Snedden & Verry 2004; Verry *et al.* 2000). It was assumed that the coefficient of relationship was 0.3, on the basis of a study (Verry 1993) that estimated that there is approximately 20% inbreeding in the open pollinated population (see also Griffin *et al.* 1987; Griffin & Cotterill 1988; Hodgson 1976a; Hodgson 1976b). The gains were predicted for one trait, namely that of individual tree volume (Volume) in cubic meters at the selection age. Volume was calculated using the *E.grandis* equation of Bredekamp & Loveday (1984).

The “realised” genetic gains were estimated using two genetic gains trials, namely trial EA62/27 planted at Dukuduku and Boschoek plantations. The trial details are presented in **Table 1**.

The mean realised genetic gains observed for volume in the two trials of EA62/27, were then compared with those predicted in the deterministic models using the breeding population trials’ parameters.

In the prediction of genetic gains derived from the F_1 to F_2 , the genetic gains were predicted for selections from trials SSO1 and SSO4, as representatives of the improvement. It was assumed that selective thinning of the ‘male families’ took place at 50%. (Male families are trees which contribute towards the pollen cloud. These families may be selectively thinned or rogued.) In reality, the thinnings were 75% per family, however selections were on a plot (replication) basis- i.e. the best tree in each plot remained, not the best trees overall per family over the trial. This implies a lower selection intensity of one in four for each of the nine replications, as opposed to the nine in 36 on a per family basis. It was assumed that 18 in 36 would be a fair

estimate of the net male thinning effect. There is the possibility that pollen contamination from neighbouring plantations would reduce the gains further. In addition, the independent culling of trees with unacceptable stem form, resulted in further dilution of the within family selection for volume.

The genetic and other parameters of SSO1 and SSO4 (for predicting the F_1 to F_2 genetic gains) were calculated on thinned, rogued trials. This may bias the genetic gain predictions, although it is difficult to predict the direction of the bias, given a combination of a thinning plus rouguing regime. Different effects of thinning have been reported. Danjon (1994) reported depressed heritabilities before systematic thinning of trials of *Pinus pinaster*. Matheson & Lindgren (1985) reported simulated increases in heritabilities through strict computer-selection of trees at a plot level, on *P.radiata* data. Selective roguing of families is, however, a more effective reduction of the genetic variability, and it is possible that this exercise may therefore reduce the heritability estimates.

Due to the very uneven thinning of the genetic gain trials at a later age, the realised genetic gains are calculated for trial series EA62/27 using 33 and 34 months data (prior to thinning), however the predicted gains are modelled using older age parameters (81 and 68 months), i.e. approximately our normal selection age. The younger age 'realised gains' would be biased downwards should the generic correlation between the two ages be below one.

The "F₃ select bulk" material is a result of combined, multitrait, selection indexes, where the economic weight for volume ranged over the trials from 40% to 70% (i.e. selection took place separately in the trials, at different economic emphasis on tree stem volume), with an average weighting of 60% over trials A1 to A4. (Other selection traits were stem form, wood density, log end splitting and disease

tolerance) These selections were pooled in the F₃ select bulk. The predicted gains for volume were therefore moderated by 40% (i.e. 100% minus 60%) to accommodate the multi-trait selection which occurred. This is based on the assumption that the predicted gains for a single trait would be factored by the economic weight of that trait in a multi-trait model. No genetic correlation was assumed between the selection traits for the purposes of the genetic gains modelling. The F₃ select bulk seed was also collected after thinning of the four trials, to nine trees per family (one tree per replication). The genetic gains prediction from the F₂ trials is presented in **Figure 4**. As trial A3 had a heritability which represented the mean of the heritabilities for volume in trials A1 to A4, the standard deviation and heritabilities of this trial was used for the modelling of the predicted genetic gains. As variable amounts of selections for the “F₃ select bulk” were derived from trials A1-A4, the total number of families, and number of selected families for these four trials, was used.

The within-family selection strategy for female parents of the F₃ (from the F₂) sometimes included up to four trees per family (for a few of the very best families), as a stratified within family selection strategy was used. In addition, trees were thinned to one tree per replication (as with the F₁ to F₂ selection process). It was not possible to model the gains as a result of this strategy with the software, but it is assumed that the gains prediction was biased downwards in this instance, although only a 50% male thinning was assumed.

Results

Predicted Genetic Gains from F1 to F2

The genetic gains in tree stem volume from the F₁ to F₂, as predicted using G-Assist version 4 were 23.4%, or 0.07 m³ per tree for SSO1 (**Figure 2**) and 23.9% or 0.027m³ per tree for SSO4 (**Figure 3**), at their respective measurement ages of 81 and 64 months. The predicted gains can be factored downwards by 15% to account for independent culling for stem form and other properties (based on an estimate of

the impact of the independent culling, by considering the percentage of independent culling which took place and therefore a weakened selection intensity), resulting in a predicted gain of approximately 19.5%.

Realised Genetic Gains from P₀ to F₂

No proper comparison of these two sources exists in the genetic gains trials. The F₁ rogued Clonal Seed Orchard (38047) is intensely selected, and not an appropriate benchmark of the F₁ breeding population. The comparison between P₀ and F₂ gives some indication. The mean measured improvement between 23197 (P₀) and 38046 (F₂) is 20% and that between 23198 (P₀) and the same F₂ is 33%; therefore, the average improvement of 26.5% was estimated of the F₂ over the P₀.

Predicted Genetic Gains from F₂ to F₃

The single-trait predicted genetic gains for the selection from F₂ to F₃ (38046 F₂ bulk and F₃ select bulk) is estimated at 12.8 % (**Figure 4**). The predicted gains for volume between these two sources, after correction for multi-trait selection, was 7.7%.

Realised Genetic Gains from F₂ to F₃

The mean realised genetic gains from the 38046 (F₂ bulk) to the F₃ select bulk were 15% for volume in the EA62/27 trials at 33 and 34 months of age. Older measurements were not reliable indicators of realised genetic gain, due to a very uneven thinning which took place over the trial.

Discussion and Conclusions

The mean realised genetic gains of 26.5% for volume from the P₀ to F₂ are noteworthy, although given that the predicted genetic gains from F₁ to F₂ is 19.5%, we may have even expected more genetic improvement. There are a number of possible explanations for this. A plausible explanation is that the P₀ to F₁ selection may well have placed considerable emphasis on other key traits, such as stem straightness and wood properties. This would have reduced the genetic gains

expected for tree volume. The comparison is also not direct, as the P_0 test material is assumed to be equivalent to P_0 population.

The mean realised genetic gains from the F_2 to F_3 of 15% for volume in the EA62/27 trials, is also encouraging. This realised genetic gain is, however, higher than the predicted genetic gains of approximately 7.7%. Possible explanations for the difference are numerous, however the following should be considered:

1. There may be positive genetic correlations with the other selection traits, causing the increased improvement in volume to be greater than predicted using G-Assist, and factored down to account for the impact of multi-trait selection.
2. The explanation likely to contribute substantially to this difference is the stratified selection strategy used between these generations. Up to four trees were selected from the best families, and only one tree per family from the lower-ranking selected families. On average, 1.92 trees were selected per family. It was not possible to model this staggered selection strategy in G-Assist.

There are many variables which are difficult to account for, estimate or measure in these comparisons. This study attempts to use the two most feasible measures or scenarios which are available. The afore mentioned indications are that the realised genetic gains for the main selection trait are in the order of 14% per generation, on average over two generations of improvement. This is against predicted genetic gains of 7.7 to 19% per generation, giving a mean of 13% predicted gains. The variability of the predicted gains, and deviations from the realised gains is probably a function of the simplification of the complex selection processes, for the purpose of modelling.

The above realised genetic gains of 14% are also in line with the reported reduction in rotation length of *E. grandis* sawtimber plantations of between 10 and 15% (Verryn 2002). In the latter study, it was reported that the eucalypt sawtimber industry has reduced the forest rotation length by 10 to 15%, due to trees reaching the same harvestable size earlier, i.e. more rapid growth.

Given the success of the breeding program in terms of genetic gains, and the rough alignment with predicted genetic gains, it seems reasonable to assume that the Hardy-Weinberg assumptions (together with adjustments for a 20% inbreeding or selfing) are fair assumptions in this breeding population. In addition:

- a. The population can be considered large- each generation has involved between 20 000 and 30 000 trees. (This study considered a sub-sample of two of the trials to predict the gains.) It is estimated that the P_0 was drawn from approximately 1.25 million plantation trees which were originally visually screened in composing the South African sub-population studied here.
- b. It should be noted that there is selection in the population. This could result in changes in gene frequency and distributions.
- c. We assume that the pollinators assisted in random mating. Previous studies do indicate a degree of inbreeding . As a result, a coefficient of relationship of 0.3 was used. There could be a need to increase this constant coefficient, should it be shown that the inbreeding is of a higher magnitude in more advance populations.

Overall, the basic classical selection and prediction theory appears to be appropriate, although care should be exercised in the comparison of predicted and realised gains by considering all the potential factors which may influence the predicted genetic progress.

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Table 1 Details of the trials used to estimate the genetic parameters and genetic gains.

Trial Identity (name)	Site	Genetic material	Experimental design	Assessment age
EA6206 (SSO1)	JDM Keet, RSA	99 F ₁ families, thinned to 1 tree per plot, and rogued to 60 families	RCB 9 replicates 1x4 tree plots 2.1x2.1m spacing	81 months
EA6215 (SSO4)	JDM Keet, RSA	99 F ₁ families, thinned to 1 tree per plot, and rogued to 59 families	RCB 9 replicates 1x4 tree plots 2.1x2.1m spacing	68 months
EA62/A3 (A3)	JDM Keet	72 F ₁ families, thinned to 1 tree per plot	RCB 9 replicates 1x4 tree plots 2.7x2.7m spacing	76 months
EA62/27 (Gains trial)	Dukuduku, RSA	23198: P ₀ unimproved commercial plantation seed from Venus plantation. 23197: P ₀ unimproved commercial plantation seed from HL&H plantation in the Northern Province. 38047: F ₁ (Rogued Clonal Seed Orchard seed, selected from P ₀ plantations). 38046: F ₂ bulk from SSO2,4,5 (F ₁) after thinning and roguing F ₃ - Selections from thinned A1-A4 at JDM Keet plantation (F ₂)	RCB 3 replicates 27 plots 6x6 tree plots 3x3m spacing	33 months
EA62/27 (Gains trial)	Boschoek, RSA	38047: P ₀ 38046: "F ₂ bulk" from F ₁ SSO2,4,5 after thinning and 30% rogued for SSO1 & 4. "F ₃ select bulk"- Selections from thinned F ₂ trials A1-A4 at JDM Keet plantation, not rogued.	RCB 3 replicates 27 plots 6x6 tree plots 3x3m spacing	34 months

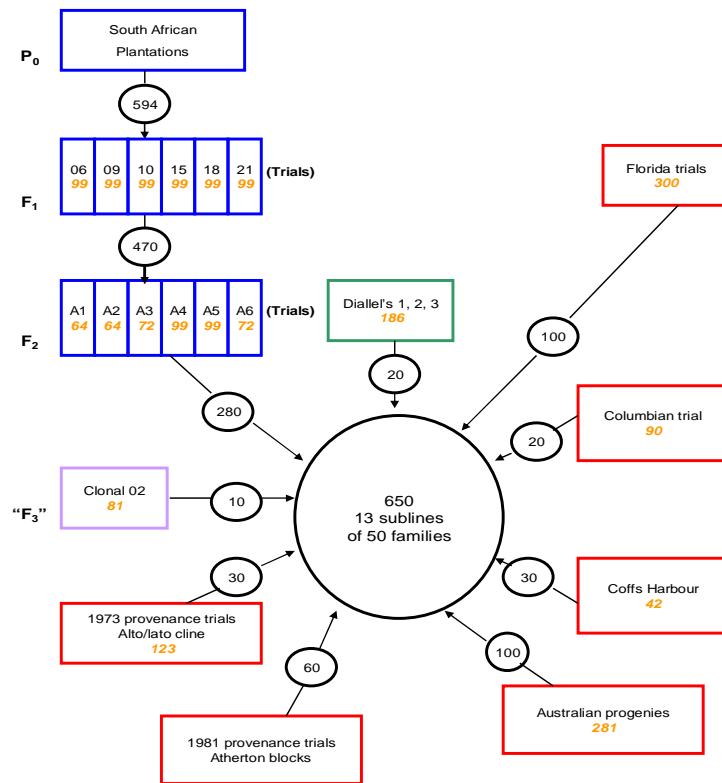


Figure 1 The numbers of selections (in small circles) and families (orange italicised text) in the various trials (square boxes) which composed the CSIR F₁, F₂, (blue boxes) and F₃ (large circle) breeding populations.

***E. grandis* genetic gains prediction of selections from SSO1 for EA62/27 gains trial F2 lot at 81 months**

BREEDING POPULATION SCENARIO: Breeding population of OP families

TRAIT: Volume

DELTA GAIN FEMALE: 0.046140326 m³

DELTA GAIN MALE : 0.023848850 m³

DELTA GAIN TOTAL : 0.069989176 m³ on the mean of 0.2990 m³

PERCENTAGE GAIN : 23.408% , giving improved mean: 0.368 m³

PERCENTAGE GAIN PER YEAR: 23.408% [Breeding cycle of 1 years]

For h**2= 0.4470 Coefficient of relationship = 0.3000

Phenotypic Std dev of BP = 0.1020

Selection age = 1.00 years

Number of families = 99

Effective population size after selection (Ne) = 120.0000

Coefficient of inbreeding Ft = 0.00416667

Ne & Ft assume random family sizes, are approx. & are on the last iteration.

Trees per family = 36

Selection intensity, among families, female = 0.6296000 (60.00,99)

Selection intensity, within families, female = 2.118 (1.00,36)

Selection intensity, among families, male= 0.6296000 (60,99)

Selection intensity, within families, male = 0.781 (18,36)

Figure 2 The predicted genetic gains predicted for selection from SSO1 with parameters and variables as listed by the G-Assist program output.

***E. grandis* prediction of genetic gains for selections for volume from SSO4 for EA62/27 gains trial F2 lot at 68 months**

BREEDING POPULATION SCENARIO: Breeding population of OP families

TRAIT: Volume

DELTA GAIN FEMALE: 0.017622718 m³

DELTA GAIN MALE : 0.009500374 m³

DELTA GAIN TOTAL : 0.027123092 m³ on the mean of 0.1135 m³

PERCENTAGE GAIN : 23.897% , giving improved mean: 0.141 m³

PERCENTAGE GAIN PER YEAR: 23.897% [Breeding cycle of 1 years]

For h²= 0.4300 Coefficient of relationship = 0.3000

Phenotypic Std dev of BP = 0.0420

Selection age = 1.00 years

Number of families = 99

Effective population size after selection (Ne) = 144.0000

Coefficient of inbreeding Ft = 0.00347222

Ne & Ft assume random family sizes, are approx. & are on the last iteration.

Trees per family = 36

Selection intensity, among families, female = 0.6296000 (60.00,99)

Selection intensity, within families, female = 2.01450 (1.50,36)

Selection intensity, among families, male= 0.6296000 (60,99)

Selection intensity, within families, male = 0.781 (18,36)

Figure 3 The predicted genetic gains predicted for selection from SSO4 with parameters and variables as listed by the G-Assist program output.

BREEDING POPULATION SCENARIO:	Breeding population of OP families
TRAIT:	Volume
DELTA GAIN FEMALE:	0.025617066 m ³
DELTA GAIN MALE :	0.006902846 m ³
DELTA GAIN TOTAL :	0.032519912 m ³ on the mean of 0.2550 m ³
PERCENTAGE GAIN :	12.753% , giving improved mean: 0.288 m ³
PERCENTAGE GAIN PER YEAR:	12.753% [Breeding cycle of 1 years]
For h**2= 0.2790 Coefficient of relationship = 0.3000	
Phenotypic Std dev of BP = 0.0800	
Selection age = 1.00 years	
Number of families = 296	
Effective population size after selection (Ne) = 347.1781	
Coefficient of inbreeding Ft = 0.00144018	
Ne & Ft assume random family sizes, are approx. & are on the last iteration.	
Trees per family = 36	
Selection intensity, among families, female = 0.8825600 (132.00,296)	
Selection intensity, within families, female = 2.10144 (1.92,36)	
Selection intensity, among families, male= 0 (296,296)	
Selection intensity, within families, male = 0.870 (16,36)	

Figure 4 Prediction of genetic gains from selections from thinned A1 to A4, using the heritability, mean and standard deviation from trial A3.