

STOCHASTIC SIMULATION OF GENETIC ADVANCEMENT OF MULTIPLE TRAITS USING SEQUENTIAL CULLING

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(Received for publication 5 June 1998; revision 22 February 1999)

ABSTRACT

A software tool was developed for the stochastic simulation of multiple traits and uses sequential culling for the different traits. It selects superior progeny and, at present, can calculate genetic gain from the backward selection of parent clones based on progeny performance, crossing those parent clones, and forwards selection among the progeny. Key information input to the program includes the base population size, number of parents selected, economic weights for different traits, genetic correlations between traits, mating design and number of crosses, number of progeny planted and selected, and proportion culled within the progeny for each trait.

An application of the software, with particular relevance to the *Pinus radiata* D. Don breeding population in New Zealand, deals with the development of breeds for specific products and for specific site types. Gains obtained at age 8 from different selection emphases were compared, for an unspecialised main population of 300 parents and four specialised breeds of 24 parents each. The parents came from a base population of 2000 progeny-tested parents. The simulation covered forwards selection in the progeny obtained by crossing the selected parents. The breeds were designated Structural Timber, Growth and Form Elite, Long Internode, and Fibre, and reflected different end-product objectives. Six traits were involved—namely dbh, stem straightness score, needle retention score, branch cluster frequency score, spiral-grain angle, and wood density. The simulation provided support for differentiating a Structural Timber breed (with a strongly multinodal branching habit, higher wood density, and reduced spiral grain angle). As well, we were able to simulate breeds similar to the existing Long Internode breed and a Growth and Form Elite population. The software can also be used to study the effect of varying population size.

Keywords: elite populations; multiple populations; structural lumber; long internode; spiral grain angle; wood density; straightness; software; tree breeding; *Pinus radiata*.

INTRODUCTION

The genetic improvement of *Pinus radiata* in New Zealand began in the early 1950s (Burdon 1992a). The breeding programme has emphasised selection for fast-growing, well-formed trees with light, wide-angled branches (Thulin 1957; Shelbourne *et al.* 1986). Parallel to the main programme, breeders started in 1970 to develop a “Long Internode”

breed, trees with longer distances between branch clusters (Shelbourne 1970; Jayawickrama, Shelbourne & Carson 1997). There have been similar efforts on “Dothistroma Resistant” and “High Wood Density” breeds (Carson 1989; Carson *et al.* 1990; Burdon 1992a). To some extent other forest tree species also have the potential to differentiate subsets of top genotypes according to the end-product objective. Zobel & Jett (1995) described setting up high-wood-density and low-wood-density *P. taeda* L. (loblolly pine) seed orchards. After the first step of improving growth, form, and adaptability by breeding, forest tree species of commercial interest might be increasingly differentiated into “breeds” similar to agricultural cultivars or the important livestock species.

Further breed development is under way in New Zealand, to maximise the profitability of producing different end-products and use the remarkable phenotypic plasticity of the species. *Pinus radiata* breeders are now improving wood properties related to specific end-product values (Sorensson, Cown, Ridoutt & Xian 1997; Sorensson, Burdon, Cown, Jefferson & Shelbourne 1997). There are also potential gains to be had by using an elite population strategy, with emphasis on the top genotypes (Cotterill 1989; Mahalovich 1990; McKeand & Bridgwater 1992; White *et al.* 1993). Both the Long Internode and High Wood Density breeds involve trade-offs between important traits (Burdon 1992b; Jayawickrama, Shelbourne & Carson 1997); however, this need not be true for all breeds. New Zealand’s largest export earnings for forest products are from three categories (Ministry of Forestry 1996): logs and poles; sawn timber; pulp, paper, and paperboard. We are interested in the option of forming breeds (small, select populations), combining the “elite population” and the “multiple population” (Namkoong 1976; Eriksson *et al.* 1993) concepts, including breeds around these three categories.

As part of the process we wished to test the differentiation of breeds using computer simulation. Simulation approaches have been used to evaluate tree breeding strategies (e.g., Mahalovich 1990; King & Johnson 1993; Bridgwater 1995; Gea 1997a); this is logical given that empirical testing of breeding strategies is costly and slow. Software packages such as NATGEN (Mahalovich 1993), POPSIM (Mullin & Park 1995), and ECOGENE (Degen *et al.* 1996) have been developed to answer forest genetics questions. They add to software developed by animal breeders, such as PEDSIM (Schelling *et al.* 1998) and Gibbon (Szydlowski 1998). However, none of the packages available was completely suited to our task (modelling the simultaneous advancement of multiple traits), so we developed our own programme.

We chose to use sequential culling in the software as an approximation of the way parents are selected in the *P. radiata* breeding programme. Three methods may be used for multi-character selection: index selection, independent culling, and tandem selection (Young 1961). Tandem selection involves selection for one trait at a time over several generations, and independent culling consists of simultaneous but independent culling for different traits. Independent culling was evaluated as an option for tree breeding by Cotterill & James (1981). Sequential culling is a variant of tandem selection, and may be appropriate under certain circumstances (King & Hansen 1997); here the population is culled one trait at a time within the same generation. An example relevant to the New Zealand *P. radiata* programme is the measurement of wood density: because of our cost structures, we first measure the growth, form, and adaptability of all the progenies and then measure wood density on the progenies with good growth and form.

In addition to an appropriate model, a meaningful simulation needs good input parameters. We have progeny-test data for five traits (dbh, straightness, branch cluster frequency, needle retention, and malformation) for over 2000 first-generation *P. radiata* parents. In addition, spiral grain angle and wood density have been measured on the top parents. Fairly good parameter estimates can therefore be constructed for seven important traits.

In this paper, therefore, a software tool is described which was developed for the stochastic simulation of multiple traits and uses sequential culling. An application of the software (with particular relevance to the *P. radiata* breeding population in New Zealand), is also described—namely, the simulation of an unspecialised main population and specialised breeds. The use of the software in simulating the effect of varying population size is also demonstrated.

DESCRIPTION OF THE SOFTWARE

Steps and Procedures

The objective of the software is to generate a parent population and a progeny population with the appropriate means and variances. This is done using a random number generator and an algorithm to convert the random numbers to a standard normal distribution (Press *et al.* 1988). The program is written in Microsoft Visual C++ (Young 1993). An earlier version of the simulation software was described by Jefferson & Weaver (1997). The information input into the program is given in Table 1, and the operations carried out by the program are summarised in Fig. 1 and explained in more detail below:

TABLE 1—Information input into simulation program.

Item	Category	Order of input
Traits to be used	Selection Emphases	1
Economic weight for each trait		2
Number of trees in base population	Characterisation of Parent Population	3
Number of trees selected from base population as candidate parents		4
Inferred population means, additive and environmental variances for each trait		5
Inferred additive, dominance, epistatic and environmental variances for each trait (for the progeny population)	Variances and Correlations	6
Genotypic and environmental correlations		7
Mating design between parent clones	Family Structure	8
Number of individuals per pair-cross (and the number of ramets per individual in the case of clonal replication)		9
Percentage of culling per trait, and which trait is "most preferred" *†	Culling Procedures	10
Maximum number of individuals retained per family		11

* The most preferred trait is used in step O to cull excess individuals within a family.

† Traits are input in the same order as culling will take place.

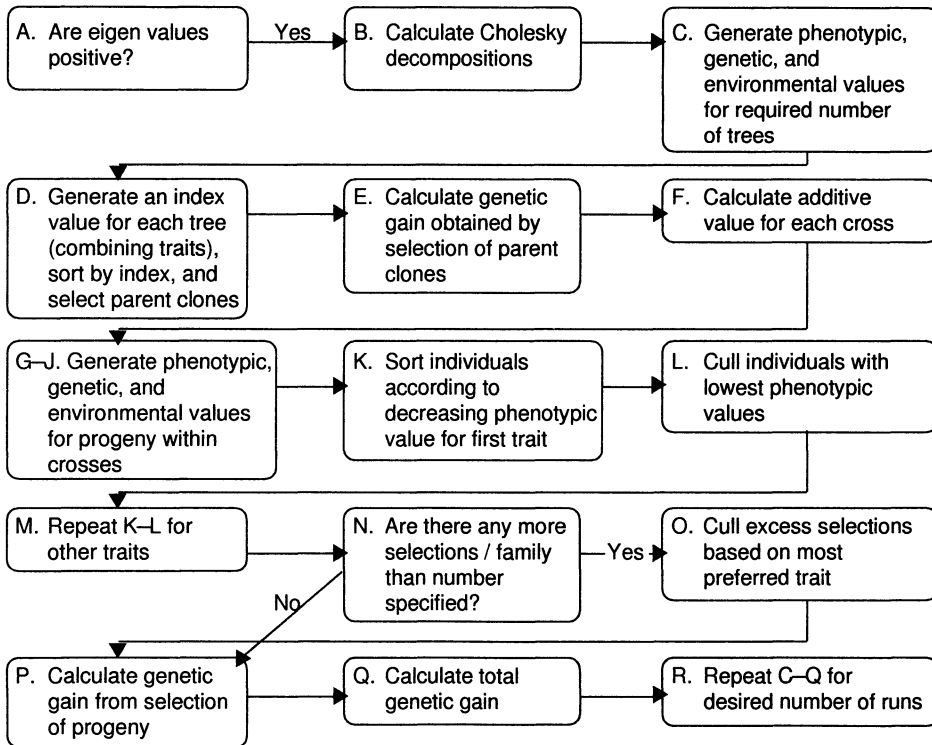


FIG. 1—Flowchart summarising operations carried out by the simulation programme.

- A Test if the eigen values of both the genetic and the environmental correlation matrices are positive, i.e., matrices are positive definite. (*NOTE: if either of these fails the test, the program cannot proceed until the correlation matrices are adjusted to be positive definite.*)
- B If the eigen values are positive, calculate the Cholesky decomposition (equivalent to square roots) of the genetic correlation matrix and environmental correlation matrix. The two resultant matrices will be called C_G and C_E respectively. Details of Cholesky decomposition have been given by Golub & van Loan (1983; Theorem 5.2-3 and Algorithm 5.2-1).
- C Generate phenotypic (P), genetic (G), and environmental (E) values for each tree, up to the number of trees required in Step 3 (Table 1), using the formula

$$p = g + e$$

For the genetic values, the program first generates r , an $n \times 1$ ($n = \text{number of traits}$) vector of random variables with a standard normal distribution. Using C_G , the $n \times n$ matrix calculated above, the genetic values g are calculated from matrix multiplication as

$$g = C_G r$$

This step is repeated for each tree. The environmental values e are also calculated starting with standard normal values, with the difference that they are multiplied by C_E , the $n \times n$ matrix calculated above, giving

$$e = C_E r$$

For a given tree, the environmental values for different traits are independent of each other. Each such set of environmental values would be independent of each other.

- D Using the phenotypic values generated in step C and the economic weights assigned in Step 2 (Table 1), the program calculates an index value, the sum of the (phenotypic value \times economic weight) across traits, for each parent clone, sorts on decreasing index value, and selects the number of parent clones specified in Step 4 (Table 1).
- E For each trait, calculate the realised genetic gain in percentage points (g_1) obtained by selection of parent clones as

$$g_1 = ((\text{additive mean of selected parents} / \text{mean of base population}) - 1) \times 100$$

- F Calculate the additive value (a_C) for each cross (specified in Step 8 of Table 1) as the mean of the additive values of the parents involved, and store the information.
- G For each individual within the cross, use the same approach as in C to generate a vector of additive values a and a vector of environmental values e_1 as shown below. (*Note: We are using different variance-covariance matrices here*).

$$a = a_C + a_1$$

$$a_1 = C_{G1/2} r$$

and $e_1 = C_{E1} r$

with a_1 = additive value from individual deviations

a_C = additive value for the cross

$C_{G1/2}$ = Cholesky decomposition of the additive variance-covariance matrix for the progeny (divided by two).

C_{E1} = Cholesky decomposition of the environmental variance-covariance matrix

r = an $n \times 1$ vector taken from a standard normal distribution (n = number of traits).

- I Generate a phenotypic value (p_1) for each individual as $p_1 = a + e_1$
- J Repeat procedure for all crosses.
- K Sort individuals according to decreasing phenotypic value for the first trait.
- L Cull the individuals with the lowest phenotypic values, to the percentage specified in Step 10 (Table 1), and up to the maximum number to be retained per family (Step 11).
- M Go to the second trait, repeat as in K and L, and continue till culling is completed for all traits.
- N Check whether the number of selections within a family exceeds the number specified
- O When the number of selections within a family is too high, cull excess selections based on their phenotypic value for the most preferred trait. For example, if dbh is the most preferred trait, and only one individual is allowed per family, the individual (within the family) with the highest dbh will be retained.
- P Calculate the realised genetic gain (g_2) obtained by selection of progeny as $g_2 = ((\text{additive mean of selected progeny} / \text{additive mean of all the progeny}) - 1) \times 100$
- Q Calculate the total genetic gain (g_3) as $g_3 = ((\text{additive mean of selected progeny} / \text{mean of base population}) - 1) \times 100$
- R Repeat loop to give specified number of runs (50 or more).

An example of the output file shows the simulated genetic gain from 50 runs (Fig. 2). The six traits are dbh, stem straightness (str), branch cluster frequency (br9), needle retention (nra), spiral grain angle (spg), and juvenile wood density (den).

Comments on the Model

The economic weights used are relative, refer to the genotypic values, and do not need to be changed along with changes in the population means. The final number of selections from the progeny population may be less than the target number, since there is at present no provision for replacement if none of the individuals in a family meets the culling levels. However, the number can be kept very close to the target by appropriate selection of the culling levels. In the simulation described below, for example, the number of selections did not drop below 98% of the target number. It should be noted that the total genetic gain is not the simple addition of g_1 and g_2 but is obtained as described in step Q.

The model is at present a one-generation model, and makes no adjustment for inbreeding. There is no limit to the number of runs (repeat loops), size of the base population, number selected, number of individuals, or the number of replicates per individual (for clonal testing). There is complete flexibility on the mating design used. The model does not warn the user of abnormal values of means, variances, or gain estimates.

The assumptions used are described below in the description of specific simulations. Application of the model is limited chiefly by failure to comply with the assumptions, and by the validity of the genetic parameter estimates used. The variances could be particularly affected with small population sizes.

We envisage the future development of this model to include the option of using a selection index in conjunction with sequential culling, extension to multiple generations with potential changes in variances over generations, and the incorporation of inbreeding.

APPLICATIONS OF THE SOFTWARE

Simulation of Main Population and Breeds

For our simulations we used a base population of 2000 progeny-tested clones, close to the number tested by the New Zealand Radiata Pine Breeding Co-operative (NZRPBC), and selected either 300 parent clones (an unspecialised "main" population) or 24 parent clones (for the specialised "breeds"). We then crossed them according to a double-pair mating design (e.g., $A \times B$, $A \times C$, and $B \times C$) for the main population and a 3×3 factorial mating design for the breeds, and planted 100 seedlings per cross (Table 2). We used genetic parameters for trees at age 8 (means and variances), constructed provisionally from results

TABLE 2—Parameters and structure of simulated main population and breeds.

	Main population	Breeds
Base population from which parent clones were selected	2 000	2000
Number of parent clones selected	300	24
Total number of crosses	300	36
Crossing design	Double-pair mating	3×3 factorial
Total number of progeny	30 000	3600
Number of forwards selections	300	24

Name of Run		Structural Timber Breed - 3/4/98		
Number of Runs		50		
Trait	Init Wgt	Mean	Add Var	Env Var
dbh	3.00	200.00	82.50	20.63
str	2.00	5.20	0.37	0.09
nra	0.50	4.00	0.10	0.03
br9	2.00	5.00	0.55	0.08
spg	1.00	2.00	1.25	0.06
den	3.00	325.00	125.00	8.33
Base Pop	2000	Parents Selected		24
Trait	Add Var	Dom Var	Ep Var	Env Var
dbh	165.00	0.00	0.00	619.00
str	0.73	0.00	0.00	2.75
nra	0.21	0.00	0.00	0.75
br9	1.10	0.00	0.00	2.53
spg	2.50	0.00	0.00	1.67
den	250.00	0.00	0.00	250.00
Families = 36		Individuals/Family = 100		Replications/Individual = 1
Trait	% Culled			
dbh	90 %	str 30 %	nra 5 %	
br9	30 %	spg 40 %	den 60 %	
Maximum per Family: 1		Preferred Trait		dbh
Number Selected:		Mean	S.D.	
		23.9	1.8	
RESULTS:		GAIN -1st Gen	2nd Gen	Combined
Trait: dbh				
Mean		6.2 %	2.7 %	9.0 %
Std Dev		0.8 %	0.9 %	1.1 %
Trait: str				
Mean		14.6 %	3.0 %	18.1 %
Std Dev		2.3 %	2.4 %	3.9 %
Trait: nra				
Mean		3.4 %	1.6 %	5.1 %
Std Dev		1.6 %	1.7 %	2.5 %
Trait: br9				
Mean		17.9 %	4.1 %	22.8 %
Std Dev		2.3 %	2.9 %	4.5 %
Trait: spg				
Mean		19.5 %	18.3 %	40.8 %
Std Dev		10.4 %	10.1 %	12.9 %
Trait: den				
Mean		3.1 %	1.2 %	4.4 %
Std Dev		0.6 %	0.7 %	1.1 %

FIG. 2—Output file showing simulated genetic gain from 50 runs.

of NZRPBC trials and listed in the Appendix (Tables A1 and A2). The traits were dbh, stem straightness score, needle retention score (a measure of resistance to *Cyclaneusma* needle cast), branch cluster frequency score, spiral-grain angle, and juvenile wood density (all routinely used in the evaluation of *P. radiata* genotypes). The trials were established during 1969 to 1988 with wind-pollinated seed from almost 1000 first-generation plus-trees selected in unimproved plantations. These parameters can be compared with published estimates, especially those of King & Burdon (1991) and Burdon (1992b); although the figures are not identical, the trends are similar. The rationale for using juvenile wood density and spiral grain angle in the New Zealand radiata pine breeding programme has been presented by Sorensson, Cown, Ridoutt & Tian (1997) and Sorensson, Burdon, Cown, Jefferson & Shelbourne (1997).

After generating the progeny population, we culled the progeny in the same order of traits as listed above, starting with dbh and ending with wood density. The rationale for this is to cull for growth and form traits (which are less expensive to measure), then for spiral-grain angle and for density (the most expensive) last. The economic weights (*see* General Comments in DISCUSSION section) and culling rates we used are given in Tables 3 and 4. The culling rates refer to the number culled as a percentage of the trees retained at each stage. In the main population, for example, 90% of the 30,000 trees were first culled for dbh, 50% of the remaining 3000 were then culled for straightness, and so on. The final step is re-culling within the 1122 retained after culling for spiral-grain angle (on the preferred trait = dbh) with the target of retaining 300 trees, one from each of the 300 families. This final step is automatic and its culling rates do not have to be specified.

TABLE 3—Relative economic weights used for the selection of parent clones

Trait	Main Population	Structural Timber breed	Growth and Form Elite	Long Internode	Fibre
Dbh	3	3	3	3	3
Stem straightness score	2	2	3	2	0.25
Needle retention score	0.5	0.5	0.5	0.5	0.25
Branch cluster frequency score	0	2	0	3	0.25
Spiral-grain angle	0.5	1	0.25	1	0.5
Wood density	1	3	1	1	3

TABLE 4—Culling rates (%) used for selecting forwards selections within the progeny generation, for the main population and breeds.

Trait	Main Population	Structural Timber breed	Growth and Form Elite	Long Internode	Fibre
Dbh	90	90	96	80	85
Stem straightness score	50	30	45	60	5
Needle retention score	5	5	5	5	5
Branch cluster frequency score	0	30	0	80	2.5
Spiral-grain angle	10	40	15	5	12.5
Wood density	12.5	60	25	20	90
Re-culling on preferred trait (dbh)	73.3	40	50	42.9	42.9

The same procedure was used for the simulation of the four breeds. The breeds were formed with emphasis on different traits (always with attention to growth and form): a “Structural Timber” breed (multinodal habit, high wood density, and low spiral-grain angle), a “Growth and Form Elite” (dbh and straightness), a “Long Internode” breed (uninodal / near-uninodal habit and low spiral-grain angle), and a “Fibre” breed (dbh and high wood density). These four breeds reflect the objectives of producing structural timber, high-quality export logs, clearwood from unpruned trees, and fibre products (pulp, paper, fibreboard, etc.) respectively. Wood density was used as a surrogate for stiffness in the structural timber breed, since we do not yet have good breeding values or genetic parameters for stiffness for *P. radiata* in New Zealand. In addition to achieving good gains for the relevant traits, culling rates were kept high enough to prevent losses in any one trait. This was most important for wood density, since heavy culling for dbh tended to cause losses for density. On the other hand, culling rates had to be balanced across traits since too much culling would result in the programme failing to keep enough progeny.

The following assumptions were made:

- (1) Genetic variance is all additive.
- (2) Variances for the progeny generation are the same as for the parental generation (environmental and genetic segregational variances remain the same, no gametic phase disequilibrium).
- (3) Values for traits are normally distributed.
- (4) Ratios between environmental variances and covariances are the same as the ratios between additive variances and covariances (i.e., phenotypic correlations are equal to the genetic correlations).
- (5) 100% survival.

With respect to the fourth assumption listed above, the model includes the option of specifying a phenotypic correlation different from the genetic correlation. However, we found that specifying the phenotypic correlation matrix (using estimates obtained from the same data as used for the additive correlation matrix) resulted in a non-positive definite environmental correlation matrix, and the operations could not proceed.

Effect of Increasing Population Size

The effect of increasing population size on simulated gain (through the effect on selection intensity) was demonstrated for a hypothetical Structural Timber breed. The same genetic parameters were used as before, and populations of 24, 51, 99, and 300 parent clones were crossed according to a double-pair mating design, with 100 individuals planted per pair-cross. The number of forwards selections in each case was equal to the number of parents crossed. The economic weights were as for Structural Timber in Table 3. The culling rates increased as the populations grew smaller (Table 5).

RESULTS

Main Population and Breeds

Results from the simulation of the main population and breeds are shown in Table 6 and Fig. 3, for trees measured at age 8. Gains in dbh were from 16.4 mm for the main population

TABLE 5—Culling rates (%) used for selecting forwards selections within the progeny generation, for different population sizes of the Structural Timber breed.

Trait	Population size			
	300	99	51	24
Dbh	85	86	88	88
Stem straightness score	20	22	24	25
Needle retention score	5	5	5	5
Branch cluster frequency score	20	22	24	25
Spiral-grain angle	10	12	13	14
Wood density	40	44	48	48
Re-culling on preferred trait (dbh)	79.7	49.6	66.4	50

TABLE 6—Simulated means for a main population and breeds, before and after one generation of selection, crossing, and reselection.

Trait	Population mean before selection	Mean after one generation of breeding				
		Main Population	Structural Timber breed	Growth and Form Elite	Long Internode breed	Fibre breed
Dbh (mm)	200	216.4	218.2	222.8	215.4	216.8
Stem straightness (1–9 scale, 9 = very straight)	5.2	5.88	6.12	6.33	5.78	5.34
Needle retention (1–6 scale, 6 = good retention)	4.0	4.24	4.22	4.35	4.28	4.14
Branch cluster frequency (1–9 scale, 9 = highly multinodal)	5.0	5.50	6.17	5.77	3.84	5.05
Wood density (kg/m ³)	325	326	339	327	332	354
Spiral-grain angle (°)	2.0	1.82	1.35	1.85	1.34	1.88

to 22.8 mm for the Growth and Form Elite (8.2 to 11.4%). The lowest gain for stem straightness score was for the Fibre breed (0.34 units) and the highest was for the Growth and Form Elite (1.33 units). Gains in needle retention score varied little, from 0.14 to 0.35 units. Branch cluster frequency score increased in four of the five populations (with the greatest increase in the Structural Timber breed) and decreased in the Long Internode breed. Wood density was maintained or increased slightly in three populations and increased substantially in the Fibre and Structural Timber breeds. Spiral-grain angle decreased in all five populations, but especially in the Long Internode and Structural Timber breeds.

Increasing Population Size

The effect of increasing population size on simulated gain for the Structural Timber breed is shown in Fig. 4. Simulated gain decreased with increasing population size for all traits. The fall-off in percentage gain was for spiral-grain angle (from 16.9% to 9.2%). These decreases were due largely to decreases in gain from selection of parent clones.

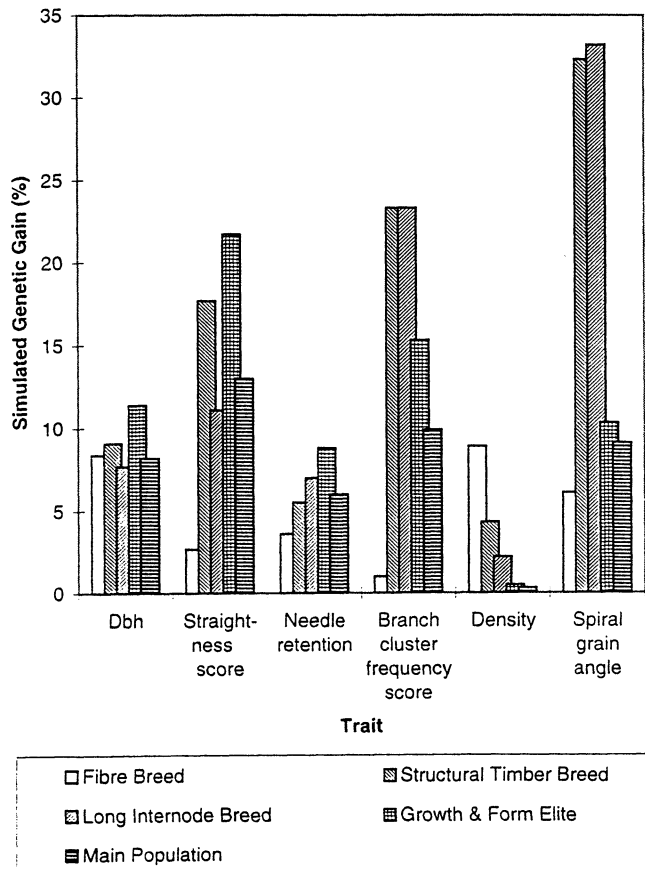


FIG. 3—Simulated gains for a main population and breeds from one generation of selection, crossing, and reselection. (Note: For the Long Internode breed, positive gain reflects a decrease in branch cluster frequency).

DISCUSSION

General Comments

The results show that stochastic simulation of population improvement with six traits and sequential culling is viable. No difficulties were encountered with matrix operations for this set of genetic parameters (the matrices were positive definite) for equal genotypic and phenotypic correlation matrices. At this stage the process of setting economic weights and culling rates was somewhat arbitrary, apart from the considerations listed in the methods and the motivation to obtain clear separation of the different breeds. Although some research has been conducted on the way different traits affect production cost (e.g., Chambers *et al.* 1997) economic weights are not readily available for *P. radiata*. It also recognised that the resources (i.e., number of crosses and progeny) allocated to the main population and breeds in our study are unequal; however, the choice of number of crosses reflects current breeding strategy for *P. radiata* in New Zealand (Jayawickrama, Carson, Jefferson & Firth 1997).

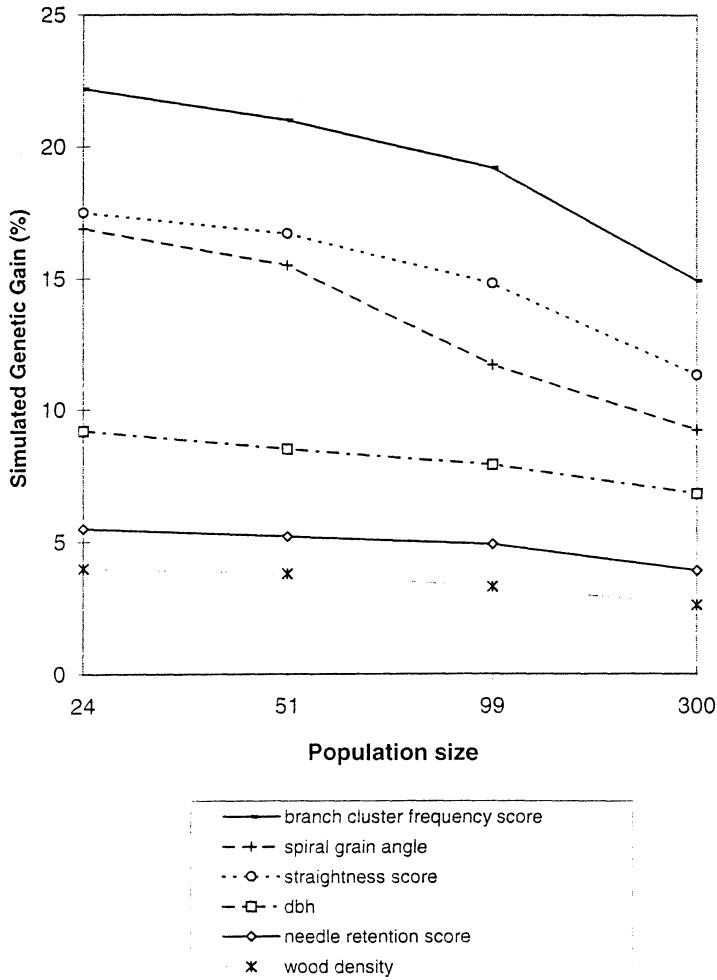


FIG. 4—Effect of increasing population size (decreasing selection intensity) on simulated gain for the Structural Timber breed.

We assume that the value of this simulation is strongly dependent on the accuracy of the estimates of correlations and narrow-sense heritability. While the estimates for dbh, straightness, needle retention, and branch cluster frequency were derived from a number of sites and a very large number of trees, estimates for spiral grain angle were based on fewer families and sites and for wood density on only two sites (*see Appendix*). Some bias may also be introduced into the parameter estimates since wood properties tend to be measured on the progenies with good growth and form. As mentioned previously, our “consensus” phenotypic correlation matrix resulted in a non-positive definite environmental correlation matrix. When faced with this situation, one option (the one we chose) is essentially to place more faith on the “consensus” genetic correlation matrix obtained than on the “consensus” phenotypic correlation matrix. We preferred this to the second option (of tinkering with the matrices until the matrix operations worked) on the basis that the second option is poor

science. The third and best option, naturally, is to get precise, unbiased estimates of correlations; however, at least for wood density, this may not happen for *P. radiata* in New Zealand in the immediate future.

Main Population and Breeds

The levels of gain were comparable across the five populations (main + breeds) for dbh and needle retention score. It became clear that, although little economic weight was placed on needle retention score, gains were made for this trait, probably because of its positive correlation with dbh. If anything, the levels of gain we obtained were conservative, and some of the traits (especially wood density) may have higher heritabilities than were used in the simulation. Estimates of realised gain from the use of improved *P. radiata* seed (compared to unimproved) are as high as 32% for volume, 23% for bole straightness, 57% for branch cluster frequency and 80% for the proportion of acceptable stems (Eldridge 1982; Forest Research Institute 1987; Shelbourne *et al.* 1989). We note, however, the qualification that straightness and branch cluster frequency are subjectively measured on a scale with fixed limits, resulting in a potential for nonlinearity.

Branch cluster frequency is a trait with justification and potential for both increase and decrease. In one genetic gain trial there was a big decrease of branch cluster frequency score (from 5.4 to 2.4) in a Long Internode seedlot and an increase (from 5.4 to 7.3) in crosses between top Growth and Form parent clones (Jayawickrama, Shelbourne & Carson 1997). Thus there is evidence that focussed selection on heritable traits can produce large shifts in population means and that distinct “breeds” can be formed in *P. radiata*.

Positive genetic correlations between dbh, stem straightness, needle retention, and branch cluster frequency facilitate breeding for a large, straight, multinodal tree with good needle retention. It is harder to deliver a large straight tree with long internodes, and this has limited the use of the Long Internode breed (Jayawickrama, Shelbourne & Carson 1997). However the gain estimates in Table 2 (especially comparing the Structural Timber breed with the Long Internode breed) suggest that we could reduce branch cluster frequency without unduly affecting growth rate and straightness. The future may be in a well-formed tree with moderately long internodes, less extreme than the uninodal tree sought in the early years of the programme (Shelbourne 1970). There was a difference of 2.1% between the two breeds in gain in wood density (4.3 v. 2.2), the results suggesting that wood density would not increase as much in the Long Internode breed (and perhaps would not need to).

Comparison of the Structural Timber and the Fibre breeds indicates that the higher wood density of the Fibre breed (15 kg/m³) is gained at the expense of reduced gain in straightness and much lower gain in spiral-grain angle. These results give little support to a separate Fibre breed, given that prices for pulpwood in New Zealand are much lower than for logs suitable for sawing (e.g., Anon. 1998). However, should other traits gain routine emphasis (e.g., stiffness for selecting Structural Timber candidates and pulp yield for Fibre candidates) the situation could change.

Experience with New Zealand’s forest industry suggests few other prospective breeds. Resistance to *Dothistroma* needle cast is highly heritable and there has been good progress in developing a *Dothistroma* Resistant breed, but this breed has not been planted to any great extent. A “Fertile Sites” breed has been proposed, designed for very fertile ex-pasture land

(Jayawickrama, Carson, Jefferson & Firth 1997). *Pinus radiata* trees are more prone to have crooked stems, large branches, malformed tops, poor wood stiffness, and a tendency to internal collapse on such sites. However, rooted cuttings with some physiological ageing (about 2 years) can be used to good effect on these sites, since using such cuttings can lead to a higher proportion of straight, defect-free stems (Menzies & Aimers-Halliday 1997).

Population Size

Increasing gain as the population size of the breed decreased parallels other results, and is one benefit of small (elite) populations of highly select individuals (Mahalovich 1990). The cost of breeding a population of 24 individuals would also be only a fraction of the cost of breeding a population of 300. In contrast, the benefits of keeping a main population include keeping inbreeding low, having a source of material with which to infuse the existing breeds in later generations, having a source of material for new breeds / elite populations, maintaining genetic diversity, and buffering against possible turnarounds between present selection strategies and future needs. Inbreeding can build up quickly in very small populations (Lindgren *et al.* 1996) but a strategy based on several populations of 20–30 unrelated parent clones may be viable for several generations (Gea 1997b). The use of a subline structure (Burdon 1986, 1988) restricting relatedness to within sublines, is a tool to reduce effects of inbreeding on the fitness and vigour of planted stock.

Extensions and Future Applications of the Software

There is potential to extend this work over multiple generations, to evaluate the sensitivity of the results to changes in the estimates of genetic parameters, to evaluate situations with substantial non-additive variance, to optimise culling rates, and to study the effect of different mating designs. A situation where a positive definite matrix could be validly obtained, without requiring the phenotypic correlations to be the same as the genotypic correlations, could also be investigated.

ACKNOWLEDGMENTS

S. Weaver executed the programming aspects of the model described. This work has been supported by funding from the Foundation for Research, Science and Technology and the NZRPBC. The genetic parameter estimates were obtained from trials established by the New Zealand Forest Research Institute and the NZRPBC. F.E. Bridgwater, R.D. Burdon, L.D. Gea, J.N. King, M.F. Mahalovich, S.E. McKeand, and P. Wilcox made useful suggestions on versions of this manuscript.

REFERENCES

- ANON. 1998: Statistics. *New Zealand Pine International* 21: 44.
- BRIDGWATER, F.E. 1995: A locus based model for advanced generation breeding. P.43 in Lavereau, J. (Ed.) Proceedings of the Twenty-Fifth Meeting of the Canadian Tree Improvement Association, Part 2. 8 August – 1 September, Victoria, BC, Canada.
- BURDON, R.D. 1986: Clonal forestry and breeding strategies—a perspective. Pp.645–659 in Proceedings of IUFRO Genetics Group Meeting, October, Williamsburg, VA, USA.
- 1988: Recruitment for breeding populations: objectives, genetics and implementation. Pp. 555–572 in Weir, B.S. *et al.* (Ed.) Proceedings of 2nd International Quantitative Genetics Conference, June 1987, Raleigh, NC, USA.

- 1992a: Introduced forest trees in New Zealand: Recognition, role, and seed source. 12. Radiata Pine (*Pinus radiata* D. Don). (Ed.) J.T. Miller. *New Zealand Forest Research Institute, FRI Bulletin No. 124*.
- 1992b: Genetic survey of *Pinus radiata*. 9: General discussion and implications for genetic management. *New Zealand Journal of Forestry Science* 22: 274–298.
- CARSON, S.D. 1989: Selecting *Pinus radiata* for resistance to Dothistroma needle blight. *New Zealand Journal of Forestry Science* 19: 3–21
- CARSON, M.J.; BURDON, R.D.; CARSON, S.D.; FIRTH, A.; SHELBOURNE, C.J.A.; VINCENT, T.G. 1990: Realising genetic gains in production forests. In Proceedings of IUFRO Working Parties on Douglas fir, Lodgepole pine, Sitka and *Abies* spp. — Breeding and Genetic Resources. Session: Genetic gains in production forests. Olympia, Washington, USA.
- CHAMBERS, P.G.S.; BORRALHO, N.M.G.; BANHAM, P.W.; COX, R.E. 1997: Impact of wood selection traits on a thermo-mechanical pulping system using *Pinus radiata* to produce newsprint. Pp.155–159 in Burdon, R.D.; Moore, J.M. (Ed.) “IUFRO’97 Genetics of Radiata Pine”, Proceedings of NZ FRI-IUFRO Conference 1–4 December and Workshop 5 December, Rotorua, New Zealand. *New Zealand Forest Research Institute, FRI Bulletin No.203*.
- COTTERILL, P.P. 1989: The nucleus breeding system. Pp. 36–42 in Proceedings of 20th Southern Forest Tree Improvement Conference, 26–30 June, Charleston, SC, USA.
- COTTERILL, P.P.; JAMES, J.W. 1981: Optimising two-stage independent culling selection in tree and animal breeding. *Theoretical and Applied Genetics* 59: 67–72.
- DEGEN, B.; GREGORIUS, H.-R.; SCHOLZ, F. 1996: ECO-GENE, a model for simulation studies on the spatial and temporal dynamics of genetic structures of tree populations. *Silvae Genetica* 45: 323–329.
- ELDRIDGE, K.G. 1982: Genetic improvement from a radiata pine seed orchard. *New Zealand Journal of Forestry Science* 12: 404–411.
- ERIKSSON, G.; NAMKOONG, G.; ROBERDS, J.H. 1993: Dynamic gene conservation for uncertain futures. *Forest Ecology and Management* 62: 15–37.
- FOREST RESEARCH INSTITUTE 1987: Which radiata pine seed should you use? *Ministry of Forestry, What’s New in Forest Research No. 157*.
- GEA, L.D. 1997a: Genetic diversity and gain: the concept of a status number. Ph.D. thesis, University of Canterbury, New Zealand.
- 1997b: Predicting population behaviour under a multiple-elite breeding strategy. Pp. 231–236 in Burdon, R.D.; Moore, J.M. (Ed.) “IUFRO’97 Genetics of Radiata Pine”, Proceedings of NZ FRI-IUFRO Conference 1–4 December and Workshop 5 December, Rotorua, New Zealand. *New Zealand Forest Research Institute, FRI Bulletin No. 203*.
- GOLUB, G.H.; van LOAN, C.F. 1983: “Matrix Computations”. John Hopkins University Press, Baltimore, USA. 476 p.
- JAYAWICKRAMA, K.J.S.; SHELBOURNE, C.J.A.; CARSON, M.J. 1997: New Zealand’s long-internode breed of radiata pine. *New Zealand Journal of Forestry Science* 27: 126–141.
- JAYAWICKRAMA, K.J.S.; CARSON, M.J.; JEFFERSON, P.A.; FIRTH, A. 1997: Development of the New Zealand radiata pine breeding population. Pp.217–225 in Burdon, R.D.; Moore, J.M. (Ed.) “IUFRO’97 Genetics of Radiata Pine”, Proceedings of NZ FRI-IUFRO Conference 1–4 December and Workshop 5 December, Rotorua, New Zealand. *New Zealand Forest Research Institute, FRI Bulletin No. 203*.
- JEFFERSON, P.A.; WEAVER, S. 1997: A multiple variable stochastic model for clonal strategies. Pp.58–63 in IUFRO Conference on Silviculture and Improvement of Eucalypts, Vol. 1, 24–29 August, Salvador, Brazil.
- KING, J.N.; BURDON, R.D. 1991: Time trends in inheritance and projected efficiencies of early selection in a large 17-year-old progeny test of *Pinus radiata*. *Canadian Journal of Forest Research* 21: 1200–1207.

- KING, J.N.; HANSEN, J. 1997: Cost-effective selection strategies in continued genetic improvement. Pp. 192–198 in Burdon, R.D.; Moore, J.M. (Ed.) “IUFRO’97 Genetics of Radiata Pine”, Proceedings of NZ FRI-IUFRO Conference 1–4 December and Workshop 5 December, Rotorua, New Zealand. *New Zealand Forest Research Institute, FRI Bulletin No. 203*.
- KING, J.N.; JOHNSON, J. 1993: Monte Carlo simulation models of breeding population advancement. *Silvae Genetica* 42: 68–78.
- LINDGREN, D.; GEA, L.; JEFFERSON, P. 1996: Loss of genetic diversity monitored by status number. *Silvae Genetica* 45: 52–59.
- MAHALOVICH, M.F. 1990: Modeling positive assortative mating and elite populations in recurrent selection programs for general combining ability. Ph.D. thesis, North Carolina State University, Raleigh, North Carolina.
- 1993: NATGEN: An application of genetic principles to natural regeneration. Pp. 214–221 in Proceedings of 22nd Southern Forest Tree Improvement Conference, 14–17 June, Atlanta, GA, USA.
- McKEAND, S.E.; BRIDGWATER, F.E. 1992: Third-generation breeding strategy for the North Carolina - Industry Cooperative Tree Improvement Program. Pp. 234–240 in Proceedings of IUFRO Conference S2.02-08, “Breeding Tropical Trees. Resolving Tropical Forest Resource Concerns Through Tree Improvement, Gene Conservation and Domestication of New Species”. 9–18 October, Cartagena and Cali, Colombia.
- MENZIES, M.I.; AIMERS-HALLIDAY, J. 1997: Propagation options for clonal forestry with *Pinus radiata*. Pp. 256–263 in Burdon, R.D.; Moore, J.M. (Ed.) “IUFRO’97 Genetics of Radiata Pine”, Proceedings of NZ FRI-IUFRO Conference 1–4 December and Workshop 5 December, Rotorua, New Zealand. *New Zealand Forest Research Institute, FRI Bulletin No. 203*.
- MINISTRY OF FORESTRY 1996: New Zealand Forestry Statistics 1995.
- MULLIN, T.J.; PARK, Y.S. 1995: Stochastic simulation of population management strategies for tree breeding: a new decision-support tool for personal computers. *Silvae Genetica* 44: 132–141.
- NAMKOONG, G. 1976: A multiple index selection strategy. *Silvae Genetica* 25: 199–201.
- PRESS, W.H.; FLANNERY, B.P.; TEUKOLSKY, S.A.; VETTERLING, W.T. 1988: “Numerical recipes in C: The Art of Scientific Computing”. Cambridge University Press, New York. 735 p.
- SCHELLING, M.; STRICKER, C.; FERNANDO, R.L.; KÜNZI, N. 1998: PEDSIM—A simulation program for pedigree data. Pp. 475–476 in Proceedings of 6th World Congress on “Genetics Applied to Livestock Production”. January, Armidale, NSW, Australia. Volume 27.
- SCHUTZ, W.M.; COCKERHAM, C.C. 1966: The effects of field blocking on gain from selection. *Biometrics* 22(4): 843–863.
- SHELBOURNE, C.J.A. 1970: Genetic improvement in different tree characters of radiata pine and the consequences for silviculture and utilisation. Pp. 44–58 in Sutton, W.R.J. (Ed.) “Pruning and Thinning Practice”. 16–20 March, Rotorua, New Zealand. *New Zealand Forest Service, FRI Symposium No. 12*.
- SHELBOURNE, C.J.A.; CARSON, M.J.; WILCOX, M.D. 1989: New techniques in the genetic improvement of radiata pine. *Commonwealth Forestry Review* 68: 191–201.
- SHELBOURNE, C.J.A.; BURDON, R.D.; CARSON, S.D.; FIRTH, A.; VINCENT, T.G. 1986: Development plan for radiata pine breeding. Forest Research Institute, Rotorua, New Zealand.
- SORENSEN, C.T.; COWN, D.J.; RIDOUTT, B.G.; TIAN, X. 1997: The significance of wood quality in tree breeding: a case study of radiata pine in New Zealand. In Proceedings of Canadian Tree Improvement Association/IUFRO Wood Quality workshop. “Timber Management Toward Wood Quality and End-Product Value”. 18–22 August, Québec City, Canada.
- SORENSEN, C.T.; BURDON, R.D.; COWN, D.J.; JEFFERSON, P.A.; SHELBOURNE, C.J.A. 1997: Incorporating spiral grain into New Zealand’s radiata pine breeding programme. Pp. 180–191 in Burdon, R.D.; Moore, J.M. (Ed.) “IUFRO’97 Genetics of Radiata Pine”, Proceedings of NZ FRI-IUFRO Conference 1–4 December and Workshop 5 December, Rotorua, New Zealand. *New Zealand Forest Research Institute, FRI Bulletin No. 203*.

- SZYDLOWSKI, M. 1998: Gibbon - C++ program for Monte Carlo estimation of mixed models. Pp. 479–480 in Proceedings of 6th World Congress on “Genetics Applied to Livestock Production”, January, Armidale, NSW, Australia. Volume 27.
- THULIN, I.J. 1957: Application of tree breeding to forestry in New Zealand. *New Zealand Journal of Forestry* 7: 41–46.
- WHITE, T.L.; HODGE, G.R.; POWELL, G.L. 1993: An advanced-generation tree improvement plan for slash pine in the southeastern United States. *Silvae Genetica* 42: 359–371.
- YOUNG, M.J. 1993: “Mastering Microsoft Visual C++ Programming”. SYBEX Inc, San Francisco, USA. 980 p.
- YOUNG, S.S. 1961: A further examination of the relative efficiency of three methods of selection for genetic gains under less restricted conditions. *Genetic Research* 2: 106–121.
- ZOBEL, B.; TALBERT, J. 1984: “Applied Forest Tree Improvement”. John Wiley and Sons, New York.
- ZOBEL, B.J.; JETT, J.B. 1995: “Genetics of Wood Production”. Springer-Verlag, Berlin.

APPENDIX 1

CALCULATION OF MEANS AND VARIANCES

Data from the following open-pollinated progeny trials (of *P. radiata* grown in New Zealand) were used in development of the parameter estimates in Table 1A:

- (1) Progenies of 467 first-generation selections, grown on three sites (total of 34,447 trees, planted with single-tree plots, year of planting 1987).
- (2) A subset (n=58) of the progenies within the trial referred to in (1), on the same three sites (2068 trees total).
- (3) Progenies of 224 first-generation selections, grown on three sites (total of 21,728 trees, planted with single-tree plots, year of planting 1988).
- (4) A subset (n=55) of the progenies within the trial referred to in (3), on two of the three sites (2213 trees total).
- (5) Progenies of 300 first-generation selections, grown on three sites (total of 34,929 trees, planted with 10-tree row plots, year of planting 1971).
- (6) Progenies of 120 first-generation selections, grown on two sites (total of 6000 trees, planted with 10-tree row plots in 1969).

All these trials were planted according to a Sets-in-Replications design, a variant of the Randomised Complete Block Design (Schutz & Cockerham 1966). Dbh, stem straightness score, and branch cluster frequency score data were obtained for all the six groups, spiral-grain angle for Groups (2) and (4), and wood density on Group (6). Spiral-grain angle was measured in Groups (2) and (4) about a year after the respective growth and form assessments. The convention we used here (*see* Sorensson, Burdon, Cown, Jefferson & Shelbourne 1997) for spiral-grain angle was to denote left-handed (S) spirality with a positive sign; the higher the value the more severe the spirality.

Groups (1) to (5) were measured between the ages of 8 and 10 years, and Group (6) was measured at age 6. From these trials "consensus" values for means and variances were obtained by averaging the estimates obtained within each site and by other experience (for *P. radiata* at age 8 years and grown in New Zealand).

TABLE A1—Population means and variances used in the simulation.

	Mean	Variances				
		Addi- tive	Domi- nance	Epi- static	Environ- mental	Pheno- typic
Dbh (mm)	200	165	0	0	619	784
Stem straightness (on 1–9 scale, 9 = very straight)	5.20	0.73	0	0	2.75	3.48
Needle retention (on 1–6 scale, 6=high retention)	4.00	0.210	0	0	0.745	0.955
Branch cluster frequency (on 1–9 scale, 9 = highly multinodal)	5.00	1.10	0	0	2.53	3.63
Spiral-grain angle (°)	2.00	2.50	0	0	1.67	4.17
Wood density (kg/m ³)	325	250	0	0	250	500

CALCULATION OF GENETIC AND PHENOTYPIC CORRELATIONS

The genetic and phenotypic correlations in Table A2 were also calculated from the data from the trials listed above. Dbh, stem straightness score, and branch cluster frequency score data were available for all six groups. Needle retention data were available for Groups (1), (2), (3), and (4), while the spiral grain angle was available from Groups (2) and (4). Wood density information came only from Group (6). Although wood density is routinely measured in progeny trials in New Zealand, the data sets are often too small (being only for the highest-ranked for growth and form) for the reliable estimation of genetic correlations. No genetic correlation estimates could be obtained between spiral grain angle and wood density (because of inadequate data) so this correlation was set to zero. Further, there are no *a priori* reasons to expect appreciable correlations between these traits.

Estimates of additive genetic variances (σ^2_A) and between-trait additive genetic covariances ($\text{cov}_A(\text{trait}_1, \text{trait}_2)$) were used to estimate genetic (r_G) and phenotypic correlations (r_P):

$$r_G = \text{cov}_A(\text{trait}_1, \text{trait}_2) / (\sigma_{A, \text{trait } 1} \times \sigma_{A, \text{trait } 2})$$

$$r_G = r_P$$

After calculation of the estimates for each site, “consensus” estimates were obtained by averaging the estimates. These consensus estimates were therefore an average of between one and 12 point estimates. The resulting correlations are given in the correlation matrix below. Correlations with spiral-grain angle were multiplied by -1 to make a positive correlation with spiral-grain angle favourable. The correlations of branch cluster frequency score with the other five traits were reversed for the long internode breed, so that gain would reflect a decrease in branch cluster frequency.

TABLE A2—Correlation matrix ($r_G = r_P$).

	Dbh	Stem straight- ness score	Needle retention score	Branch cluster frequency score	Spiral- grain angle	Wood density
Dbh	1	0.063	0.371	0.249	-0.178	-0.195
Stem straightness score		1	-0.008	0.352	0.182	-0.050
Needle retention score			1	0.055	-0.087	-0.200
Branch cluster frequency score				1	-0.142	-0.205
Spiral-grain angle					1	0
Wood density						1