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Across-Sites Genetic Parameters of Internode-Length Variables in *Pinus radiata* assessed by Laser Measurements

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Abstract

Branching habit is extremely variable in *Pinus radiata* (D.Don), and figures prominently in breeding goals. Branch cluster frequency (BR) scores are useful for ranking families, but do not give absolute mean internode length or internode index values. Also, comparing BR scores across different sites is complicated by large differences among site means and variances. It is desirable for the breeder to characterise families for mean internode length. Using a laser to measure internode length is potentially more precise and faster than other methods (e.g. climbing, height pole, and PhotoMARVL). We address the potential of a new laser technique to: (1) estimate genetic parameters of mean internode length and internode index; (2) quantify the relationship between BR and mean internode length; and (3) determine the optimum zone of stem for characterising whole-tree mean internode length.

Fifty open-pollinated families, *ca* 20 trees/family, were assessed at two sites, one on the Kaingaroa Plateau, and the other on Northland coastal dunes (Pouto), 38 families being common to both sites. Mean internode length of the first log (MIL1) and second log (MIL2), mean internode length in the 4-7 m height zone (MIL4-7 m), and internode index of the first log (IIX1) and second log (IIX2) were assessed at age 17-18 years using a laser. These measurements were compared with previously collected data (diameter at breast height over bark and BR scores) from an age 8 assessment of the trees.

On average, MIL2 exceeded MIL1 (0.51 cm and 0.40 cm respectively at Kaingaroa; 0.38 cm and 0.35 cm respectively at Pouto). Across-sites narrow-sense heritability (h^2) estimates of BR, MIL1, MIL2, IIX1 and IIX2 were 0.18, 0.25, 0.23, 0.26 and 0.27 respectively, approximately half the within-site estimates. Across-sites genetic correlation estimates for these traits were ca 0.45, further indicating considerable rank-change genotype-environment interaction. Mean internode length in the 4-7 m height zone had higher estimated h^2 than BR, and a high genetic correlation (about 0.98) with BR, indicating that laser assessment could be made more cost-efficient by assessing MIL4-7 m, which might be used to compare the families for mean internode length at different sites. To convert BR scores to obtain a reliable site mean for mean internode length, 30 random trees from each BR score class could suffice at a given site.

Keywords: branching pattern; internode index; heritability; genetic correlation; genotype × site interaction; *Pinus radiata*

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Introduction

Radiata pine (*Pinus radiata* D.Don) has an extremely variable branching pattern (e.g. Bannister, 1962; M. Carson & Inglis, 1988). Depending largely upon both genetic and environmental factors, a tree will typically produce from one to five branch clusters on the leader in a growing season. Since the spacing of these clusters is irregular, this leads to great variability in the lengths of 'internodes', the distances between successive branch clusters. The variation in internode length is of major interest for tree breeding and for wood processing and utilisation (M. Carson, 1988). In choosing a branching ideotype for tree breeding, various trade-offs are involved. On the one hand, a regular, short-internode ('multinodal') habit tends to be associated genetically with some control of branch size, and good growth and form, which favour production of structural timber. On the other hand, internodes of sufficient length favour production of recutting grades that provide clearcuttings between successive clusters of knots.

Assessing the potential of selection candidates in a breeding programme to produce such clearcuttings is not straightforward. For direct measurement, various techniques including climbing, use of height pole, and more recently stereoscopic photography (PhotoMARVL) (Firth et al., 2000) have been used for measuring internode lengths. All of these techniques, however, are costly and time-consuming, especially for assessing large numbers of trees in progeny trials. Consequently, branch cluster frequency scores (BR) are currently used as an indirect subjective measure of internode length in all progeny trials. In the interests of achieving resolution of progeny differences, the general tendency is to use, as far as possible, the entire range of the 1-9 scale for assessing individual trees. Thus, these scores do not give a good indication of absolute values of mean internode length, nor of internode index, which is the proportion of log length providing clearcuttings above a threshold length. Indeed, the interpretation of a given BR score across different sites, among which mean internode length and internode index can vary widely, could be quite complex (Grace & M. Carson, 1983; Turner et al., 1997).

Recent trends suggest that structural grades and appearance grades represent a large proportion of the radiata pine sawn timber produced in New Zealand (Beauregard et al., 2002). For appearance-grade timber, a long-internode branching habit in unpruned logs is needed to give high yields of better shop (clearcuttings) grades; and minimising various appearance defects, such as resin pockets and internal checking, is also important (Shelbourne et al., 1997). It is quite difficult to predict the amounts of recutting (factory) grade and clearcuttings recovery achievable from a seedlot with a given breeding value rating (White et al., 2007, p 117ff) for BR. Thus, understanding the genetic control of internode length and rating and ranking families for mean internode length is becoming increasingly important. There are a number of published reports (reviewed by Wu et al., 2008) on genetic control of BR, but very few on genetic parameter estimates of internode length or internode index as such.

A new technique for measuring internode length, using an Impulse 200 Laser, was developed in 1996 (Anonymous, 1996; Carr, 1996). Because it directly provides digital data on location of branch clusters, using this instrument has the advantage of being more precisely quantitative than the various earlier methods mentioned previously. This technique also has the potential to establish a stronger cross-reference between BR scores and mean internode length and/or internode index. The present study, based on Impulse 200 Laser data and earlier BR scores, was conducted at two sites between which mean internode length and internode index were known to differ and which were expected to show marked rank-change genotype x site interaction for the branching-pattern variables. The main objectives of this study were to:

(1) estimate across-sites genetic parameters of mean internode length (MIL) and internode index (IIX) along with BR;

(2) quantify the interrelationships between BR, MIL, and IIX, for a new sample population;

(3) determine the optimum zone (or length) within the visible portion of the stem for characterising the mean internode length of the whole tree, as a basis for future assessments; and

(4) determine a sampling strategy (by using bootstrap sampling) for obtaining precise ranking of families in order to help reduce the cost of ranking families for internode length at other sites.

Compared with previous work (Grace & M. Carson, 1993; Shelbourne et al. 1997; Turner et al., 1997), this study involved a larger genetic sample, an additional internode-length variable (mean internode length, 4-7 m height), and focused on the refinement of sampling protocols.

Materials and Methods

Genetic material

An open-pollinated (OP) progeny trial of 468 plus trees ("885"-series, selected in unimproved land-race stands) was established in 1987, involving plus trees selected in 1985 (R.D. Burdon et al., unpubl.). The trial was established at four sites. Of these, data from only two sites (Kaingaroa Forest and Pouto Forest) are reported here. Kaingaroa Forest Cpt 324 (Lat.: 38° 42' S; Long.: 176° 23' E) is at an altitude of 550 m asl, has an annual rainfall of ca 1500 mm, and a rhyolitic pumice type soil. In contrast, Pouto Forest (Lat.: 36° 20' S; Long.: 174° 05' E) is at an altitude of ca

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30 m asl, has an annual rainfall of ca 1300 mm, and a coastal dune sand soil. Branch cluster frequency (BR), although rated, was not a criterion for selecting the seed parents. Consequently, around 10% of the resulting selections had a long-internode branching habit (BR scores 1-2). In addition to “885”-series plus-tree progenies, OP progenies of 74 long-internode parents of the “883”-series were also tested at the Kaingaroa site.

Seed was sown in 1986 to raise 1/0 bare-root planting stock in the Forest Research Institute Nursery, Rotorua, using a sets-in-replicates layout (cf Schutz & Cockerham, 1966). Each set of 25 entries (including controls) of 30-seed row-plots formed a contiguous block within a nursery replicate. The field planting was done in 1987. The layout at each site combined the sets within 25 replicates with use of randomised single-tree plots. The 883-series progenies were planted as separate sets.

Data were obtained from these trials when the trees were aged 8-years (from planting), and some were used in our study. These data were summarised and used to assess and rank of progenies, and thence parents, for growth and form traits at age 8 from planting (K.J.S. Jayawickrama et al., unpubl.). Importantly, this assessment included BR scores for individual trees and diameter at breast height over bark.

Detailed assessment of internode-length variables was done at the Kaingaroa and Pouto sites at ages 17 and 18 respectively from planting. At Pouto, 50 “885”-series families were assessed. Of these, only 38 families were assessed at Kaingaroa. Instead, a further 12 families of the “883”-series were assessed at Kaingaroa. Twenty-three of the common 38 families of “885”-series were ranked (based on index selection involving age-8 performance for diameter, stem straightness and needle retention across three sites) in the top 10%, but the remaining 15 families of “885”-series were among the above-average families only at Kaingaroa (K.J.S. Jayawickrama et al., unpubl.). Approximately 20 trees were assessed per family at each site. The trials were planted at 600 stems/ha, and no thinning took place prior to this study. However, differential mortality (although very minor) could contribute to discrepancies between results for traits assessed at an early age (e.g. BR) and a later age (e.g. mean internode length).

Assessment

An Impulse 200 Laser™ (http://www.opticsplanet.com/s/impulse+200/) was used to measure the distance between branch clusters. For each tree, a horizontal distance was established between the instrument position and the stem. Then, from the same position, the heights of the top and bottom of each branch cluster (Grace & M. Carson 1993; Figure 1) from stump height (0.2 m) upwards were recorded to height 12 m. The internode length was calculated as the distance from the top of one cluster to the bottom of the next. Mean internode length of the first log (0.2–5.7 m height), and the second log (5.7–11.2 m height) was calculated for each tree by averaging all internode length measurements. Internode index was also calculated for the first two logs, as the percentage of log length represented in internodes ≥60 cm long. The parts of internodes at the log ends were treated as terminating at that point, instead of assigning that full internode length to either log. The algorithm calculating mean internode length for tree height between 4-7 m was set to start from a cluster in the range of 3.5 to 4.5 metres and finish on a cluster between 6.5 and 7.5 metres. This range was chosen to encompass a length of stem approximately (but not exactly) 3 metres. Internode index is the sum of internodes of 0.6 m or longer, expressed as a fraction of log length (Whiteside & Manley, 1986; Grace & M. Carson, 1993). The BR scores obtained at age 8 years were used to establish a relationship with mean internode length and internode index. The following branching-related variables (calculated for each tree) were considered for analyses in this study:

- MIL1: mean internode length of first log (0.2–5.7 m);
- MIL2: mean internode length of second log (5.7–11.2 m);
- MIL4-7m: mean internode length of a portion, 4-7 m height, of a tree;
- IIX1: Internode index of first log;
- IIX2: Internode index of second log; and
- BR: Branch cluster frequency score (1-9 scale, where: 1 = uninodal; 9 = strongly multinodal) assessed at age 8 years.

The same field crew operated at both sites.

Genetic analysis

At each site, the significance of family differences for the above variables was tested using the following model:

$$\text{Phenotype} = \mu + R + F + \text{error} \qquad [1]$$

where μ, R and F represent the general mean, replicate, and family effects, respectively. Replicate was considered as a fixed effect, while family and error effects were treated as random effects. Data transformation was considered to be unnecessary based on experience with data from similar trials.

Individual-tree heritability (h^2) and genetic correlation (r_g) estimates were obtained from:

$$\hat{h}^2 = 4 \times \hat{\sigma}^2_f \,/\, (\hat{\sigma}^2_f + \hat{\sigma}^2_e) \qquad [2]$$

$$r_g = \hat{\sigma}_{xy} \,/\, \sqrt{\hat{\sigma}^2_x \times \hat{\sigma}^2_y} \qquad [3]$$

where σ^2_f ($\approx 0.25\ \sigma^2_A$) and σ^2_e are the among-family (or general combining ability, i.e. GCA) variance and within-family variance components respectively, σ^2_A being the additive genetic variance; σ_{xy} represents the additive genetic covariance between two traits; and σ^2_x and σ^2_y are additive genetic variances for the two traits. REML (Restricted Maximum Likelihood) estimates of genetic and phenotypic variances were obtained through an iterative process (Gilmour et al., 1997). The methods of Burdon (1977) and Hodge and White (1992) were used for calculating Type-B (between-sites) genetic correlation estimates (r_B) and across-sites estimates of h^2, respectively, as follows:

$$r_B = \hat{\sigma}_{gca12} \,/\, \sqrt{\hat{\sigma}^2_{gca1} \times \hat{\sigma}^2_{gca2}} \qquad [4]$$

$$\hat{h}^2 = 4 \times \hat{\sigma}_{gca12} \,/\, \sqrt{(\hat{\sigma}^2_{P1} \times \hat{\sigma}^2_{P2})} \qquad [5]$$

The covariance (σ_{gca12}) is estimated directly as the mean cross-product of between group (i.e. half-sib family) means at a pair of sites. Estimates of GCA variance components at the two sites were represented by σ^2_{gca1} and σ^2_{gca2} while the total phenotypic [random-effect] variances at the two sites were represented by σ^2_{P1} and σ^2_{P2}. When the same trait is being measured at the two sites, the covariance (σ_{gca12}) provides an estimate of GCA variance. Multivariate analyses were conducted using open-access ASREML software (Gilmour et al., 1997). Approximate standard errors of estimated genetic parameters were also obtained using ASREML software. Phenotypic correlations, at the individual-tree level and at the family-mean level, were obtained using SAS PROC CORR software (SAS Institute Inc., 1988).

Sample size for genetic parameter estimation

The effect of sub-sampling on the estimation of both narrow-sense heritability and family ranking was investigated, by using bootstrap sampling. Random sub-sampling of different numbers of offspring per family (5, 10 and 15 trees per family) was repeated 50 times using a SAS Macro programme, and the results reviewed. Minimum, maximum and average estimates are reported.

Sample size for quantifying visually assessed BR scores

To quantify BR scores in terms of mean internode length and internode index, mean mean internode length and internode index values were obtained for predetermined numbers of sample trees from each BR score class at each site. The results could then be used to convert the existing BR scores into estimates of mean internode length and internode index on a site-by-site basis. The number of trees at a given site required for reliable conversion of the existing BR scores to mean internode length and internode index was investigated using sub-sampling. Random sub-sampling of different numbers of trees (30, 40 and 50 from each of the eight BR score classes) was repeated 50 times. Minimum, maximum and average estimates are reported.

Results and Discussion

Basic statistics

The average BR scores at Kaingaroa and Pouto were 3.71 and 6.69, respectively, for all 50 families at the respective sites (Table 1). Reducing the sample studied to the 38 families common to both sites had little impact on site averages for BR scores, internode lengths and internode indices at Pouto (Table 1), since all 12 of the excluded families came from the same parental population with respect to BR scores. In contrast, reducing the sample studied to the 38 families common to both sites increased average BR score and generally reduced the averages for internode lengths and internode indices at Kaingaroa (although not greatly), because the disregarded families were from the “883” (long-internode) series (Table 1).

For any group of families, means for all empirical internode-length variables were higher at Kaingaroa than at Pouto (Table 1), this contrast being most marked for IIX1 and IIX2. It has been suggested that this could be due, at least in part, to the Kaingaroa site being cooler than Pouto (e.g. Grace & M. Carson, 1993). Another possible effect is that of observer variation in assessing BR scores. While such an effect may have contributed to the overall low average BR score at Kaingaroa, it is more likely to have dampened down any difference in BR scores between the two sites. This is because of a tendency for observers to centre their scores around a mid-range. The relatively high average BR score noted for Pouto is typical of trials established on coastal sand sites, where internodes generally tend to be shorter than on inland sites. Average MIL2 was higher than for MIL1 at both sites, but the difference was smaller at Pouto than at Kaingaroa (Table 1). M. Carson and Inglis (1988) reported higher mean values for MIL2 (0.40–0.64 m) compared with MIL1 (0.36–0.49 m).

TABLE 1: Average values and ranges for all (approximately 20) trees per family, obtained using individual-tree data, of different variables at each site. Diameter (DBHOB) and branch cluster frequency (BR) were measured at age 8.

Variable	**Units**	**Kaingaroa (age 17)**			**Pouto (age 18)**		
		Mean	**Range**	**CV(%)**	**Mean**	**Range**	**CV(%)**
A. All families							
DBHOB	mm	199	116 – 282	12.5	209	125 – 304	13.9
BR	1 – 9 score	3.71	1 – 9	49.6[1]	6.69	1 – 9	53.0[1]
MIL1	m	0.40	0.18 – 0.83	23.7	0.35	0.08 – 0.87	25.3
MIL2	m	0.51	0.13 – 1.60	38.8	0.38	0.09 – 1.07	30.9
MIL 4-7 m	m	0.63	0.09 – 2.40	44.8	0.45	0.06 – 2.02	39.8
IIX1	See text	0.37	0.00 – 0.81	47.1	0.21	0.00 – 0.78	82.8
IIX2	See text	0.38	0.00 – 0.94	66.1	0.20	0.00 – 0.86	95.5
B. Families common to both sites							
DBHOB	mm	198	123 – 282	12.7	210	125 – 286	13.5
BR	1-9 score	4.00	1 – 9	48.5[1]	6.73	1 – 9	51.6[1]
MIL1	m	0.39	0.18 – 0.78	43.3	0.35	0.13 – 0.87	24.8
MIL2	m	0.47	0.13 – 1.29	34.0	0.38	0.09 – 1.07	30.5
MIL 4-7 m	m	0.58	0.09 – 1.95	43.0	0.46	0.12 – 1.96	38.6
IIX1	See text	0.34	0.00 – 0.81	50.1	0.21	0.00 – 0.78	85.7
IIX2	See text	0.33	0.00 – 0.94	71.1	0.20	0.00 – 0.86	95.0

[1] *Using the formula: Standard deviation/[(mean – 1)(9 – mean)]*$^{0.5}$ *(Burdon, 2008)*

Using full-sib families from a diallel experiment, Turner et al., (1997) reported a significant influence of site on MIL2 (i.e. Woodhill, 0.32 m; Eyrewell. 0.38 m; Kaingaroa Cpt 327, 0.52 m; Golden Downs, 0.53 m; Taringatura, 0.53 m). Our figures (Pouto, 0.38 m and Kaingaroa, 0.51 m) agree closely with theirs, which is reassuring given that Pouto is near to Woodhill, and that our other study site adjoins Kaingaroa Cpt 327. Earlier studies (e.g. M. Carson & Inglis, 1988; Grace & M. Carson, 1993) suggested a weak association between mean internode length and latitude (i.e. longer mean internode length at higher latitudes). The markedly lower internode index values for both IIX1 and IIX2 at Pouto compared with Kaingaroa were associated with high coefficients of variation (CVs) (Table 1), whereas the CVs for BR scores, when corrected for position of means within the range (Burdon, 2008), were essentially the same between the two sites.

Surprisingly, the differences in mean internode length between logs were not matched by appreciable differences in internode index at either site (Table 1). The protocol of including part internodes at the ends of logs in calculating internode index would tend to dampen down differences in internode index compared to mean internode length. However, there is probably also an effect due to smaller variability in length among internodes in the second log. This may reflect a difference in mean internode length between the two halves of the first log (cf Grace & M. Carson, 1993). The average mean internode length in the 4-7 m height portion of the stem (MIL4-7m) was greater than for either MIL1 or MIL2 at both sites (Table 1). These results suggest that the expression of internode length in the 4-7 m height zone could be a better selection trait than internode length in either log, if shown to be under strong genetic control.

Jayawickrama et al. (1997) reported that the average mean internode length and internode index of families in the Long Internode breed (which includes the “883” series) were much higher than those of families selected mainly for growth and form. However, families with desirable long-internode habit also had less desirable charactistics such as slower growth, and more malformation than families selected for their growth and form habits (Jayawickrama et al., 1997).

TABLE 2: Estimates of narrow-sense heritability (h^2) at Kaingaroa (K) and Pouto (P) respectively for all (approximately 20) trees per family. Across-site h^2 estimates and between-sites genetic correlation estimates (r_B) are also shown.

Variable	**All 50 families**		**38 families in common**		**Across-sites h^2**	r_B	$\hat{\sigma}^2_f/(\hat{\sigma}^2_f + \hat{\sigma}^2_{fs})$
	K	**P**	**K**	**P**			
DBHOB	0.16	0.13	0.17	0.13	0.09	0.45	0.67
BR	0.48	0.46	0.22	0.57	0.18	0.39	0.48
IIX1	0.46	0.50	0.27	0.65	0.26	0.54	0.64
IIX2	0.72	0.54	0.30	0.64	0.27	0.43	0.64
MIL1	0.54	0.56	0.27	0.66	0.25	0.45	0.83
MIL2	0.74	0.62	0.28	0.71	0.23	0.34	0.61
MIL 4-7 m	0.58	0.50	0.33	0.65	0.29	0.55	0.66

Genetic control and Genotype x Environment interaction (G x E)

Estimated heritabilities of different variables are shown for both individual sites and across sites in Table 2. With all 50 families considered at each site, the within-site heritability estimates were almost all high (≥ 0.5; standard error $\approx 0.12 - 0.15$) at both sites.

The estimated heritabilities for BR in all 50 families at Kaingaroa and Pouto were 0.48 and 0.46, respectively (Table 2). These estimates are slightly higher than generally reported values (Shelbourne & Low 1980; S. Carson, 1991; Burdon et al., 1992; Jayawickrama et al. 1997; Jayawickrama, 2001). Estimated heritabilities for BR obtained from all 467 OP families of the "885"-series were 0.33 and 0.37 at Kaingaroa and Pouto respectively (details not shown). A review (Wu et al., 2008) of inheritance of BR reported narrow-sense heritability estimates varying from 0.06 to 0.64, with an average of 0.35. In contrast, the corresponding estimates for BR at our sites were approximately 50% lower at Kaingaroa than Pouto (0.22 cf 0.57) for the subset of 38 families represented at both sites. This showed that resolution of family differences was much sharper at Pouto. The approximate standard errors of these estimated heritabilities were about 0.10 and 0.16 at Kaingaroa and Pouto, respectively.

Interestingly, the quantitative measures (mean internode length and internode index) of branching habit had generally higher estimated heritabilities than the visual measure (BR) both within and across sites. The comparison between genetic controls of these traits could be complicated by different ages of assessment since BR was measured at age 8-years and mean internode length at age 20-years. However, the generally high heritability for MIL 4-7 m, instead of BR, indicated that it could be incorporated in the breeding programme in order to provide more effective selection for the target trait (e.g. clear-cuttings at rotation age). Strength of genetic correlation between the selection trait and the target trait would influence the efficiency of such selection. Since growth and form traits are generally assessed at age 7-8 years, it should be possible to obtain mean internode length measurements at 4-7 m height on all trees at most sites with good tree growth.

Across-sites heritability estimates were considerably lower than the corresponding averages of within-site estimates (Table 2). The presence of any G x E interaction could depress the overall heritability. Results from this study indicate an average between-sites genetic correlation estimate (r_B) of 0.45 for the various measures of internode length, and, therefore, considerable rank-change G x E interaction across these two sites. Estimates of rank correlation between family means at the two sites were very similar to the r_B values (results not shown). Previous reports (S. Carson, 1991; Jayawickrama, 2001) of generally very high (>0.80) between-site genetic correlations for BR, have indicated minimal rank change across sites. On the other hand, Shelbourne and Low (1980) reported low between-site genetic correlations (r_B = 0.39) for scores of "Branch Quality" (similar but not identical to BR scores) for similar sites to ours, particularly between a northern coastal-dune site (Woodhill, near Pouto) and a central North Island site (Kaingaroa). This correlation is very similar to that observed for BR in our study. In the study by Shelbourne & Low, intensive selection of parents for branching pattern would have tended to depress the observed between-site genetic correlations. The use of a small number (38) of families in our study would affect the reliability of r_B values. Estimates of r_B values, obtained from all 467 common families, were 0.51 and 0.65 for diameter at breast height over bark (DBHOB) and BR respectively. The ratio $\hat{\sigma}^2_f/(\hat{\sigma}^2_f + \hat{\sigma}^2_{fs})$ was consistently higher than r_B (Table 2). The variance components, $\hat{\sigma}^2_f$ and $\hat{\sigma}^2_{fs}$

represent across-site between families, and family-by-site interaction variances, respectively. An expectation that the two should be equivalent would depend on the variance structures being constant between sites, a condition that clearly did not apply.

The combination of low heritabilities at Kaingaroa and the relatively low between-sites genetic correlation estimates may reflect two factors: (1) that the parents/ortets had been strongly selected for phenotypic performance in the same region as the Kaingaroa study site (i.e. on the volcanic plateau); and (2) that nearly 40% of the 38 common families had been chosen solely on performance at Kaingaroa. In turn, that selection would tend to truncate (due to correlated response) among-families variance and, therefore, depress apparent heritabilities of internode-length variables and various genetic correlations involving these variables. With G x E interaction, the expression of among-families variance would tend to be less truncated, with less depression of apparent heritability.

Using data from a diallel experiment, Turner et al. (1997) observed that correlations of family-mean mean internode length between sites were such that G x E variance for mean internode length was largely due to differences in the variance among families on different sites. M. Carson and Inglis (1988) reported a statistically significant G x E interaction for MIL2, but this did not lead to important changes of rank for different breeds. Johnson and Burdon (1990) and S. Carson (1991) showed that selections can be made that are near-optimal over all sites despite appreciable G x E interaction. If, however, the material is not evaluated using a site category that is interactive with the remaining sites, gain on that category may be quite low.

Interestingly, G x E interaction for BR has been observed in New Zealand at the level of the native Californian provenances Año Nuevo and Monterey, from which New Zealand land-race material has been derived (Burdon et al., 1997a). Towards the south of the New Zealand (in Otago and Southland), and at higher altitudes, BR scores of Año Nuevo provenance trees are lower in comparison with those of Monterey provenance (Burdon et al., 1997b, 1998).

Genetic correlations between traits

Estimates of genetic correlations between combinations of BR, mean internode length and internode index are shown in Table 3. Again, it should be noted that BR was assessed at age 8 (when only a part of the second log is visible) while mean internode length and internode index were assessed at age 20. A negative correlation of BR with other traits was expected, and is due to the fact that high BR scores reflect a larger proportion of multinodal trees. The estimated genetic correlations between combinations of BR, internode index and mean internode length were all very high (generally >>0.8) at both sites (Table 3), especially when the long-internode "883"-series families were included at Kaingaroa.

These very high estimates of genetic correlations between variables, and cheap assessment and

TABLE 3: Estimated genetic correlations (r_g) for all (approximately 20) trees per family at Kaingaroa (above diagonal) and at Pouto (below diagonal).

Trait	DBHOB	BR	MIL1	MIL2	MIL4-7 m	IIX1	IIX2
A. All families at each site							
DBHOB	-	-0.26	0.39	0.41	0.38	0.38	0.50
BR	-0.45	-	-0.99	-0.95	-0.97	-0.97	-0.95
MIL1	0.48	-0.89	-	0.95	0.96	0.98	0.94
MIL2	0.46	-0.92	0.68	-	0.99	0.98	0.99
MIL4-7 m	0.53	-0.99	0.95	0.89	-	0.98	0.98
IIX1	0.47	-0.94	0.98	0.75	0.97	-	0.97
IIX2	0.47	-0.94	0.66	0.99	0.88	0.73	-
B. Families common to both sites							
DBHOB	-	-0.31	0.45	0.77	0.59	0.50	0.64
BR	-0.42	-	-0.99	-0.86	-0.94	-0.89	-0,80
MIL1	0.33	-0.87	-	0.89	0.87	0.95	0.81
MIL2	0.41	-0.91	0.62	-	0.99	0.99	0.98
MIL4-7 m	0.53	-0.98	0.94	0.87	-	0.93	0.91
IIX1	0.42	-0.93	0.99	0.73	0.97	-	0.90
IIX2	0.36	-0.92	0.61	0.99	0.85	0.71	-

generally good heritability for BR score, show that BR score has been an excellent criterion for the selection of families for internode length (cf M. Carson, 1988; M. Carson & Inglis, 1988). Using data from four sites, M. Carson and Inglis (1988) reported that the phenotypic correlation between BR and internode index, at the individual-tree level, varied between -0.45 and -0.72 for the first log, and between -0.58 and -0.78 for the second log when assessed at the same age (i.e. 13–15). Similarly, the phenotypic correlation between mean internode length and internode index ranged from 0.75 to 0.84 and from 0.76 to 0.88 for the first and the second log respectively (M. Carson & Inglis, 1988). The individual-tree phenotypic correlations in our study were very similar to those reported by M. Carson and Inglis (results not shown).

Burdon et al. (2004) have presented a case for separate treatment of the lowest 3 m of a butt log. Estimation of mean internode length and internode index in this zone would be of great interest. Our results (Table 1) showed that MIL1 was lower than MIL2, suggesting a decreasing mean internode length towards the base of stem. Grace and M. Carson (1993) also showed that the mean internode length of the lowest 3 m of the first log would be smaller than that of the whole first log.

This study was based on the Whiteside and Manley (1986) method for obtaining internode index. Alternative measures of internode length and internode index could also be used (e.g. Todoroki et al., 2002), however. It is expected that genetic parameters of alternative measures of mean internode length and internode index would not be very different but further work is needed to test this hypothesis.

Estimated genetic correlations between DBHOB and BR (Table 3) were consistently negative in our study, in contrast to the mostly positive correlations recorded between stem diameter and branch cluster counts (Wu et al., 2008). Approximate standard errors of these estimated genetic correlations (Table 3) were high (ranging from 0.26 to 0.35), indicating that the precision was very low. In our study, selection of parents on the basis of growth and form would tend to bias correlation estimates, and inclusion of the “883” families reduced the positive correlation estimates. Consequently, we attach little weight to the observed sign of the estimated genetic correlations involving DBHOB in this study. The phenotypic correlations between DBHOB and the BR were 0.13 and 0.27 at Kaingaroa and Pouto respectively (details not shown).

Effect of sample size on estimation

Family means

For the variables studied, family means obtained using five or 10 offspring were often considerably different to those obtained using all 20 available trees. Within sites, the average correlations for different traits were generally in the range of 0.70 to 0.80 or the range of 0.80 to 0.90 for a sample size of five and 10 offspring per family respectively, when compared with estimates for 20 trees/family (Appendix 1). These results suggest that a sample of 10 offspring per family could still give imprecise estimates of family means, so it is proposed that at least 15 offspring per OP family should be sampled to obtain a reliable estimate of family means (or breeding values) for mean internode length and internode index at a given site.

Heritability estimates

Estimates of h^2 obtained from 50 different samples of five trees per family at both sites varied widely, and the range was still considerable for a sample size of 10 trees per family (Appendix 2). There was a general tendency towards positive skewness. Overall, a sample of 15 trees per family usually gave estimates very similar to those (Table 2) from the all available trees (20) per family.

Quantifying internode-length variables from BR scores

Sub-sampling of 30, 40 and 50 trees was carried out in all BR score classes that had at least 50 trees available. The average mean internode length and internode index obtained from sub-sampling of 30 trees per BR score at each site were very similar to those obtained from all available trees. For example, the average MIL2 obtained from 50 sub-samples of 30 trees each was compared with the average based on all available trees (Table 4). The results suggest that it should be possible to quantify BR scores into mean internode length or internode index values with good precision by laser-scanning 30 trees from each BR class at any given site. This should result in a major cost saving.

Inferring Breeding Values for mean internode length

Ranking family performance for either mean internode length or internode index instead of BR scores is desirable, so that achievable recoveries for both recutting (factory) grade and clear-cuttings can easily be predicted for a seedlot. Using linear regression, Turner et al. (1997) evaluated the relationship between observed second-log mean internode length and predicted parental breeding value for BR. Their results suggested that these relationships varied among different selection series and among sites within series. They suggested that in order to obtain accurate prediction of mean internode length in commercial plantations, a ‘genetic effect’ (which needs to be derived from the observed family differences in mean internode length) should be incorporated in planning (or growth) models. In this study, we have attempted to convert the observed individual-tree BR

TABLE 4: The average MIL2 obtained from all available trees (approximately 20 trees per family), and from sub-sampling of 30 trees from each BR score class. Note: Mid-range scores not assigned in interest of maximising resolution of family differences. When the available number of trees were <30, sub-sampling could not be undertaken.

	All Trees			Sub-sampling 30 trees		
Site	**BR score**	**No. of trees**	**Average MIL2**	**Min. MIL2**	**Max. MIL2**	**Average MIL2**
Kaingaroa	1	95	0.72	0.64	0.79	0.72
	2	178	0.58	0.50	0.66	0.58
	3	226	0.51	0.45	0.57	0.51
	4	183	0.47	0.42	0.56	0.47
	5	-	-	-	-	-
	6	162	0.42	0.37	0.45	0.41
	7	67	0.40	0.37	0.43	0.40
	8	16	0.36	-	-	-
	9	2	0.34	-	-	-
Pouto	1	8	0.67	-	-	-
	2	28	0.50	-	-	-
	3	74	0.44	0.41	0.47	0.44
	4	67	0.43	0.39	0.47	0.43
	5	-	-	-	-	-
	6	140	0.42	0.36	0.46	0.42
	7	313	0.37	0.33	0.41	0.38
	8	259	0.34	0.31	0.38	0.34
	9	125	0.32	0.29	0.35	0.32

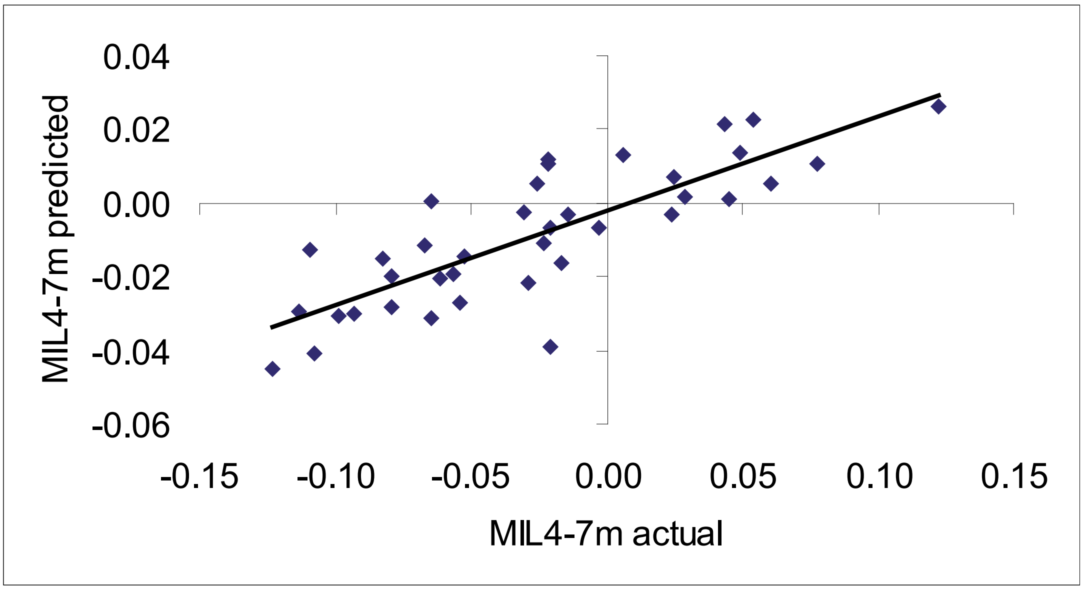

FIGURE 1: Correspondence (R = 0.81) between across-sites breeding values for MIL4-7 m based on two options. Option 1 uses values obtained from actual Impulse 200 Laser measurements (MIL4-7 m actual) on full sample of 38 families (*ca* 20 trees per family per site); Option 2 assigns to each tree, for its BR score, the mean MIL4-7m value for the sample of 30 trees at its site for that BR score (MIL4-7 m predicted).

scores into mean internode length in order to predict parental breeding value for mean internode length. An indication of the reliability of measuring mean internode length on a sample of trees (n = 30) for each BR score at each site in order to infer parental breeding values for mean internode length is given in Figure 1. The diagram shows the relationship between across-sites parental breeding values for MIL4-7 m, (inferred for individual trees on the above basis) and values calculated from direct Impulse 200 Laser measurements on all trees. With R = 0.81, the conversion of BR scores into mean internode length values should be very cost-efficient if large numbers of selection candidates need to be evaluated.

The historical breeding goal for radiata pine has emphasised selection for fast-growing, well-formed trees with light, wide-angled branches (Shelbourne et al., 1986). Selection for these characteristics has driven average internode length below that in plantations of unimproved trees (M. Carson & Inglis, 1988), resulting in decreased yield of clearwod in unpruned logs. A separate breed with long internodes was initiated to maximise the production of clear-cuttings (Jayawickrama et al., 1997). M. Carson (1988) compared profitability and timber grade recovery from long-internode and multinodal radiata pine trees grown in a direct sawlog regime. His results showed that increases in yields of clear-cuttings from the improved Long Internode breed will have some associated opportunity cost, due to slower growth rate, poorer stem form, and decreased yield of structural timber compared with multinodal trees of the Growth and Form breed. Mean internode length is directly associated with the amount of unpruned clearwood (or clearcuttings). Increasing mean internode length is expected to increase financial returns to growers and processors since knot-free timber usually commands a price premium, assuming all other stand characteristics are held constant.

Conclusions

From the results of this study, we conclude that:

- visual branch cluster frequency scores (BR) obtained at age 8 years were almost perfectly correlated genetically with both mean internode length and internode index assessed at age 20 years by means of a laser technique;
- genetic expression (h^2) of branching habit was markedly stronger in the second log within, but not necessarily across sites;
- estimates of genetic parameters were limited in their precision by the number of families sampled;
- mean internode length, assessed in the 4-7 m height zone, could be used to make laser assessment more cost-efficient in future;
- a sample of about 15 trees per OP family would provide acceptable estimates of family means and narrow-sense heritability for mean internode length and internode index;
- a sample of 30 trees from each BR score class should be used to quantify BR scores in terms of mean internode length and internode index at a given site; and
- previous observations that both site and genotype x site interaction between site types have a considerable effect on internode-length variables have been confirmed.

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APPENDIX 1: Correlations between ‘family means obtained using all (approximately 20) trees’ and ‘family means obtained using sub-samples of 5, 10 or 15 trees’ for different traits at two sites, namely Kaingaroa Compartment 324 and Pouto. The minimum, maximum and mean values over 50 sub-samples are presented.

Trait	**Sub-samples (number of trees/family)**								
	(5)			**(10)**			**(15)**		
	Min.	**Max.**	**Mean**	**Min.**	**Max.**	**Mean**	**Min.**	**Max.**	**Mean**
A. Kaingaroa Cpt 324									
BR	0.60	0.83	0.76	0.85	0.94	0.90	0.96	0.98	0.97
IIX1	0.64	0.86	0.76	0.84	0.93	0.89	0.95	0.98	0.97
IIX2	0.72	0.89	0.81	0.89	0.96	0.93	0.97	0.99	0.98
MIL1	0.55	0.87	0.71	0.64	0.93	0.82	0.76	0.98	0.89
MIL2	0.72	0.89	0.79	0.89	0.96	0.89	0.96	0.99	0.94
MIL4-7 m	0.67	0.88	0.78	0.85	0.96	0.91	0.95	0.98	0.97
B. Pouto									
BR	0.59	0.83	0.71	0.82	0.91	0.88	0.92	0.97	0.95
IIX1	0.60	0.85	0.74	0.83	0.93	0.89	0.94	0.97	0.96
IIX2	0.61	0.86	0.76	0.84	0.95	0.90	0.94	0.98	0.96
MIL1	0.24	0.86	0.69	0.38	0.94	0.83	0.51	0.98	0.90
MIL2	0.63	0.87	0.74	0.86	0.95	0.87	0.94	0.98	0.93
MIL4-7 m	0.62	0.85	0.74	0.81	0.93	0.89	0.93	0.98	0.96

APPENDIX 2: Narrow-sense heritability (h^2) estimates for different numbers of trees per family at two sites, namely Kaingaroa Compartment 324 and Pouto. The minimum, maximum and mean values over 50 sub-samples are presented.

Trait	**All trees**	**Sub-samples (number of trees/family)**								
	(ca. 20/family)	**(5)**			**(10)**			**(15)**		
		Min.	**Max.**	**Mean**	**Min.**	**Max.**	**Mean**	**Min.**	**Max.**	**Mean**
Kaingaroa Cpt 324										
BR	0.48	0.10	0.85	0.45	0.32	0.70	0.50	0.35	0.57	0.47
IIX1	0.46	0.11	0.97	0.44	0.26	0.64	0.46	0.31	0.53	0.43
IIX2	0.72	0.16	1.18	0.73	0.41	0.97	0.74	0.63	0.82	0.72
MIL1	0.54	0.18	0.84	0.51	0.34	0.76	0.54	0.40	0.63	0.50
MIL2	0.74	0.16	1.36	0.79	0.45	0.97	0.77	0.66	0.85	0.76
Pouto										
BR	0.46	-0.01	0.78	0.39	0.21	0.64	0.39	0.30	0.56	0.41
IIX1	0.50	-0.03	0.83	0.45	0.31	0.72	0.47	0.38	0.62	0.48
IIX2	0.54	0.18	1.12	0.52	0.30	0.83	0.52	0.36	0.70	0.53
MIL1	0.56	0.05	0.93	0.50	0.34	0.74	0.52	0.35	0.65	0.51
MIL2	0.62	0.20	1.02	0.57	0.31	0.80	0.59	0.48	0.71	0.58