

DEZZOTTI, A. and SANCHOLUZ, L.: Los bosques de *Austrocedrus chilensis* en Argentina: ubicación, estructura y crecimiento. *Bosque* **12**(2): 43–52 (1991). — FERREYRA, L. I., LATINO, A., CALDÉRON, A. and GARDENAL, C. N.: Allozyme polymorphism in *Austrocedrus chilensis* (D. DON) FLORIN et BOUTELJE from Patagonia, Argentina. *Silvae Genetica* **45** (2–3): 61–64 (1996). — GALLO, L. A.: Nutzung und Konservierung patagonischer forstlicher Genressourcen. Research Project No. 9348. Society for Technical Cooperation of Germany (GTZ). 5 pp. (1993). — GALLO, L. A. and GEBUREK, TH.: A short note: genetics of enzyme variants in *Austrocedrus chilensis* (ENGL.) FLORIN et BOUTELJE. *Phyton*. **34**(1): 103–107 (1994). — GREGORIUS, H.-R. and BERGMANN, F.: Analysis of isoenzyme genetic profiles observed in forest tree populations. In: Population genetics and genetic conservation of forest trees. pp. 79–96. PH. BARADAT, W. T. ADAMS and MÜLLER-STARK (eds.). 1995. SPB Academic Publishing, Amsterdam, The Netherlands (1995). — HARRY, D. E.: Identification of a locus modifying the electrophoretic mobility of malate dehydrogenase isozymes in "Incense Cedar" (*Calocedrus decurrens*), and its implications for population studies. *Biochem. Genet.* **21**: 417–434 (1983). — HARRY, D. E.: Inheritance and linkage of isozyme variants in "Incense Cedar". *Journal of Heredity* **77**, 261–266 (1986). — HATTEMER, H. H., BERGMANN, F. and ZIEHE, M.: Einführung in die Genetik. J. D. Sauerländer's Verlag, Frankfurt a. M.. 492 pp. (1993). — HAVRYLENKO, M., ROSSO, P. and FONTENLA, S.: *Austrocedrus chilensis*: contribución al estudio de su mortalidad en Argentina. *Bosque* **10**(1), 29–36 (1989). — MILLAR, C. I. and MARSHALL, K. A.: Allozyme variation of "Port-Oxford Cedar" (*Chamaecyparis lawsoniana*): implications for genetic conservation. *For. Sci.* **37**, 1060–1077 (1991). — PAPAGEORGIOU, A. C., BERGMANN, F., GILLET, E.

and HATTEMER, H. H.: Genetic analysis of isoenzyme variation in mediterranean cypress (*Cupressus sempervirens* L.). *Silvae Genetica* **42** (2–3): 109–111 (1993). — PAPAGEORGIOU, A. C., PANETSOS, K. P. and HATTEMER, H. H.: Genetic differentiation of natural Mediterranean Cypress (*Cupressus sempervirens* L.) populations in Greece. *Forest Genetics* **1**(1): 1–12 (1994). — PASTORINO, M. and GALLO, L.: Isozyme variants in *Austrocedrus chilensis* and their relation to moisture gradients. IUFRO XX World Congress, Tampere, 6–12/VIII/1995. Poster Abstracts No. 129. p. 74 (1995a). — PASTORINO, M. and GALLO, L.: Variación genética en Ciprés de la Cordillera (*Austrocedrus chilensis* (D. DON) FLORIN et BOUTELJE) a través de marcadores genéticos isoenzimáticos. IV Jornadas Forestales Patagónicas, San Martín de los Andes, (Argentina) 24–27/X/1995. (1995b). — PERRY, D. J. and KNOWLES, P.: Inheritance and linkage relationships of allozymes of eastern white cedar (*Thuja occidentalis*) in northwestern Ontario. *Genome* **32**: 245–250 (1989). — SCHMALTZ, J.: La reconquista de la estepa por el bosque de Ciprés. Descripción de un caso estudiado, Primer Informe. CIEFAP, Publicación Técnica 11, octubre 1992, 7 pp. (1992). — SOKAL, R. and ROHLF, F.: Biometría, principios y métodos estadísticos en la investigación biológica. 1ª edición española, Blume Ediciones, Madrid. 832 pp. (1979). — XIE, C. Y., DANCİK, B. P. and YEH, F. C.: Inheritance and linkage of isozymes in *Thuja orientalis*. *Journal of Heredity* **82**(4), 329–334 (1991). — XIE, C. Y., DANCİK, B. P. and YEH, F. C.: Genetic structure of *Thuja orientalis*. *Biochemical Systematics and Ecology* **20**(5), 433–441 (1992). — YEH, F. C.: Isozyme variation of *Thuja plicata* (Cupressaceae) in British Columbia. *Biochem. Syst. and Ecol.* **16**, 373–377 (1988).

Clonal Variation in Wood Quality and Growth in Young Sitka Spruce (*Picea sitchensis* (BONG.) CARR.): Estimation of Quantitative Genetic Parameters and Index Selection for Improved Pulpwood

By J. COSTA E SILVA¹), H. WELLENDORF¹) and H. PEREIRA²)

(Received 22nd May 1997)

Summary

Repeatabilities and clonal correlations were estimated for wood properties (pilodyn and lignin content), stem straightness and growth characters (height, diameter and volume index) measured in young Sitka spruce clones, under fast growing conditions. The estimates of the quantitative genetic parameters were based on a random sample of 30 clones taken from a population of 253 clones at age 9 years from field planting. The magnitude of the clone-mean repeatability estimate indicated that the examined characters are able to respond well to selection based on clone means and subsequent vegetative propagation. However, for lignin content, the phenotypic variation was rather low (i.e. coefficients of variation of 3.7% and 2.9% on the individual ramet and clone mean bases, respectively), which may limit the magnitude of the genetic gain. The estimated clonal correlations between the wood properties and growth pointed out that clonal selection for reducing the population level in lignin content and/or increasing the whole-ring density is expected to result in an indirect genetic decline in growth.

Multiple trait selection indices were constructed to explore the possibilities of improving the yield and/or the quality of young Sitka spruce wood for pulp production. In the absence of quantitative economic data – and to ascertain the possible consequences of clone selection and clonal propagation for the proposed end use – contrasting multiple trait index selection options were compared on the basis of predicted genetic responses in wood properties, growth and stem straightness.

Key words: *Picea sitchensis*, lignin content, pilodyn, wood density, growth traits, stem straightness, repeatability, clonal correlation, index selection, pulp production.

FDC: 165.3; 165.441; 165.6; 181.64/65; 232.13; 812.31; 813.11; 861.0; 174.7 *Picea sitchensis*; (489).

Introduction

In a tree breeding programme that aims at producing raw material with better qualities for a given wood processing industry, it is essential to include wood properties as potential selection criteria. The present work places particular emphasis on whole-ring density and lignin content in wood, and it is intended to provide information that can be especially relevant to the utilization of Sitka spruce (*Picea sitchensis* (BONG.) CARR.) wood for pulping purposes.

Sitka spruce wood has pale colour, good fiber characteristics and low resin and other extractive content, which make this

¹) Department of Botany, Dendrology and Forest Genetics, Royal Veterinary and Agricultural University, Arboretum, Kirkegårdsvej 3A, DK-2970 Hørsholm, Denmark

²) Department of Forestry, Institute of Agriculture, Technical University of Lisbon, Tapada da Ajuda, P-1399 Lisboa Codex, Portugal

species suitable for pulping purposes (HARDING, 1988). Comparing thermomechanical pulps based on Sitka spruce and radiata pine woods, CORSON *et al.* (1989) reported that the energy requirement to refine the former to a specified freeness was lower than that of the latter. CORSON *et al.* (1989) also found that, for pulp quality at 100 freeness level, the pulp produced from both core and slab wood of Sitka spruce had somewhat better strength properties (tensile, burst and tear index), sheet density and optical properties (scattering coefficient and opacity) than radiata pine.

Lignin is an unfavourable wood component for most pulp uses. Taking an average figure of 27% (on an extractive-free basis) for the content of lignin in young Sitka spruce wood, a reduction of this level in the population by selection and breeding may lead to the production of raw material with more desirable properties for pulping.

For a given pulping process and conditions, reducing the content of lignin in wood may contribute to improve the bleaching operations and the final results obtained on bleaching. Particularly for chemical pulping, decreasing the content of lignin in wood may increase pulp yield per unit weight of dry wood charged, and it may result in better conditions (i.e. time, temperature and chemical consumption) on pulping to a given delignification degree (RYDHOLM, 1965; NYLINDER, 1973).

Depending on the growing conditions, the overall mean wood density is generally moderate to low at both juvenile and mature growth stages of Sitka spruce, although it can achieve high values in the first annual rings from the pith. Furthermore, information concerning the environmental influence on density of Sitka spruce wood indicates that, at a given ring age and sampling height, there is a tendency for this wood property to decrease in conditions promoting vigour of growth, at both juvenile and mature growth periods (BRAZIER, 1970; HARVALD and OLESEN, 1987; PETTY *et al.*, 1990; COSTA E SILVA *et al.*, 1994). Therefore, increasing (or, at least, keeping constant) the population level in whole-ring density may improve the quality of Sitka spruce wood, especially when it is produced under conditions of rapid growth.

Due to the relationship between fiber density and the quality variables of pulp prepared from conifers, increasing wood density may lead to the production of pulp with properties suitable for some paper types, but less desirable for other end uses of the paper (DINWOODIE, 1966; ZOBEL and VAN BUIJTENEN, 1989). However, pulp yield per unit volume of wood will increase as wood density increases, and this will result in less consumption of wood per ton of pulp produced. As wood consumption can be the dominating component in the total manufacturing costs per ton of pulp produced (RYDHOLM, 1965), higher levels of wood density will have an important contribution to reduce the latter costs, particularly if pulpwood is bought on a volume basis. In conifer studies, it has been indicated that increasing wood density will reduce production costs on both mechanical (KELLOGG, 1982) and chemical (KIRK *et al.*, 1972; KELLOGG, 1982) pulping processes.

The present work provides information on clonal variation in the wood properties concerned and on their correlations with other characters of interest in a young plantation-grown Sitka spruce population, cultivated under fast growing conditions. In a subsequent phase, the objective is focused on the application of the index selection methodology to explore the possibilities of clonal selection and propagation for improving the yield and/or the quality of young Sitka spruce wood for pulp production.

Material and Methods

Plant material and field layout

The clonal trial – denoted locally as F205A – is situated in the Frijsenborg forest estate, Jylland, Denmark. The area is flat, and it has a clay soil with a 15 cm layer of humus.

The ortets were selected for height within a 4-year-old bulk offspring of the Rye Nørskov reference seed stand (F299), presumably of Washington origin. A total number of 253 clones were field planted during the spring of 1984, when the ramets were 3 years old from cutting propagation. The experimental layout is a randomized block design, with 10 replications and single-tree plots. The ramets were planted at a spacing of 1.5 m x 1.5 m.

The experiment is representative of *fast growing conditions* for Sitka spruce plantations in Denmark. Early results from this trial have been reported by NIELSEN (1994).

The characters evaluated and experimental procedures

All the characters under consideration in this study were evaluated in the F205A experiment, by using data pertaining to a *random sample of 30 clones* taken from the population of 253 clones. The measurements were carried out when the clone material was 9 years old from field planting (i.e. 12 years old from cutting propagation). Further details concerning the examined characters and the applied experimental procedures are described as follows.

Growth traits

Height (HT) was measured in dm, until the tip of the leading shoot. Breast-height *diameter* (DBH) measures (in mm) were carried out over bark, by using a standard calliper. *Volume* (VI) was approximated by an index, calculated as DBH^2 . HT (BENTZER *et al.*, 1989). The observations were made in all 10 replications.

Wood properties

The average whole-ring density at breast-height was assessed indirectly by using the *pilodyn* wood tester (PL). After removing the bark, a single pilodyn reading was taken systematically on the west side of each ramet, at an internodal position close to breast-height. The pilodyn measures were carried out in 9 replications (i.e. from 1 to 9), and the applied instrumental conditions were a spring strength of 6 joules and a 2.0 mm pin diameter.

For *lignin* (LG) determinations, the collection of wood samples included only the *current annual ring*. At the end of the growing season (i.e. October), an increment core was taken through the south side of each tree, at an internodal position close to breast-height. The sampling was carried out in 3 replications (i.e. rep. 1, 3 and 5), following the major soil-gradient at the site. The content of lignin in the fully developed current annual ring was predicted by using a calibration model, relating FTIR (Fourier transform infrared spectroscopy) data to lignin values obtained by chemical analysis. The applied calibration and prediction procedures are reported by COSTA E SILVA (1996) and COSTA E SILVA *et al.* (1998).

For both wood properties, and in order to avoid compression wood, the sampling took place on side or opposite wood in more leaning stems.

Stem straightness

Each ramet was scored for *stem straightness* (ST), according to a subjective scale with values ranging from 1 to 9 (1 = very crooked stem; 9 = perfect stem straightness). The scoring was carried out in all 10 replications.

Data analysis

Analysis of variance

A two-way analysis of variance without interaction was performed for each of the studied traits, by using the following linear mixed effects model:

$$y_{ij} = \mu + B_i + c_j + \varepsilon_{ij}$$

where y_{ij} is the observation made on the ramet of the clone j in the replication i , μ is the overall mean of the population, B_i is the fixed effect of the i^{th} replication, c_j is the random effect of the j^{th} clone and ε_{ij} is the random error for single-tree plots. The c_j are assumed to be normally distributed with mean zero and a variance σ_c^2 , and the residuals of the model (i.e. ε_{ij}) are supposed to be independent and normally distributed with mean zero and a constant variance σ_e^2 .

For the trait in question, the population value of the clonal component of variance (i.e. σ_c^2) was estimated from data concerning the applied random sample, and its significance was inferred through the use of the F -test (SNEDECOR and COCHRAN, 1976). Independence, normal distribution and variance homogeneity of the errors, as well as the additivity of the model effects, are assumptions required for the validity of the statistical inference. For the distribution of the ε_{ij} of the model described above, the departure from normality was evaluated by calculating measures of skewness and kurtosis, and by comparing these observed values with those given by SNEDECOR and COCHRAN (1976, page 552) in tables for testing these distribution parameters. The SHAPIRO and WILK (1965) test of normality was also taken into account as an additional information. All of these measures were obtained from the SAS[®] PROC UNIVARIATE (SAS[®], 1990a). Residual analysis (i.e. the plot of the ε_{ij} against the predicted values) was carried out for each trait, as an indication of heterogeneity of the error variance. From the exploratory analysis of the data, there was no evidence of serious violation of the normality and variance homogeneity assumptions for any of the characters studied here (data not shown).

Due to the existence of missing plots, a least-squares analysis was used to accommodate the unbalanced nature of the data when calculating the sums of squares. These correspond to the Type III sums of squares computed by the SAS[®] PROC GLM (SAS[®], 1990b), which are the same as the Type II and IV sums of squares in the case of a two-way model without interaction (LITTELL *et al.*, 1991).

Variance components

Estimates of the components of variance were obtained by equating the observed mean squares to the corresponding Type III expectations of mean squares and solving the equations as follows:

$$\hat{\sigma}_c^2 = \frac{(\text{Mean square for clone} - \text{Mean square for error})}{K}$$

$$\hat{\sigma}_e^2 = \text{Mean square for error}$$

where $\hat{\sigma}_c^2$ and $\hat{\sigma}_e^2$ represent, respectively, the estimates of the clonal variance and of the variance due to random error for single-tree plots; the K value is the coefficient of the clonal component of variance in the expected mean square for clone, and it was computed by the SAS[®] PROC GLM (statement RANDOM) to accommodate the unequal number of trees per genotype. The calculated K coefficients were: 8.241 for height, diameter, volume and stem straightness; 7.310 for pilodyn and 2.724 for lignin content.

Repeatabilities

The estimated variance components were applied to calculate the repeatabilities of the characters under consideration. The calculations were carried out on a *individual ramet* basis as,

$$\text{clonal repeatability} = \frac{\hat{\sigma}_c^2}{\hat{\sigma}_p^2} = \frac{\hat{\sigma}_c^2}{\hat{\sigma}_c^2 + \hat{\sigma}_e^2}$$

and on a *clone-mean* basis as,

$$\text{repeatability of clone means} = \frac{\hat{\sigma}_c^2}{\hat{\sigma}_{\bar{p}}^2} = \frac{\hat{\sigma}_c^2}{\hat{\sigma}_c^2 + \hat{\sigma}_e^2 / K}$$

where $\hat{\sigma}_c^2$ and $\hat{\sigma}_e^2$ are defined as above, $\hat{\sigma}_p^2$ is the estimated phenotypic variance of individual ramets, $\hat{\sigma}_{\bar{p}}^2$ is the estimated phenotypic variance of clone means and $\hat{\sigma}_e^2 / K$ estimates the average error variance of a clone mean.

The standard errors of the repeatability values were obtained by adapting the present situation to the general approximation described by NAMKOONG (1979) and BECKER (1984), for determining the variance of a heritability ratio that is derived from the analysis of variance approach.

Clonal correlations

Estimates of genetic correlations with low accuracy may be expected when the heritability of one (or both) of the characters involved is calculated with poor precision, as the result of a less effective control of the variation due to experimental error (ROBERTSON, 1959). Therefore, and owing to the unequal information available for the traits measured, the estimation of the clonal correlations was preceded by analysing the significance of the clonal component of variance for the following sampling groups or subsets of the replications (data not shown):

- sampling group 1, including the replications where LG was measured (i.e. rep. 1, 3 and 5) and, thus, containing information for all of the examined traits – the analysis of variance results indicated no statistical significance (at the 5% level) of the clonal component of variance for DBH and VI; for HT, PL and ST, the clonal variance was statistically significant at the 1% level;
- sampling group 2, including the replications where information is missing for LG (i.e. rep. 2, 4, and 6 to 10) – for the growth traits, as well as for PL and ST, the clonal component of variance was statistically significant at the 0.1% level.

According to the process of estimation, the genetic correlation can be classified as (BURDON, 1977): *Type A* correlation, when the estimates are based on observations on the *same individuals* within genetic groups; *Type B* correlation, when the estimates are based on observations on *different individuals* within genetic groups. The *Type B* genetic correlation approach has been applied to study genotype-environment interactions (i.e. the *same trait* measured in two different sites), but its scope of application can be extended to *two different traits* measured in different groups of individuals within a given site.

In the present report, the *Type A* clonal correlations pertain to the relationships involving the growth traits, PL and ST. The *Type B* clonal correlations refer to the relationships concerning LG – evaluated from observations made in the sampling group 1 – and the growth traits, PL and ST measured in the sampling group 2 of the same trial.

When compared with the sampling group 1, more precise measurements with higher repeatabilities were obtained from the larger sampling group 2 for the growth traits, PL and ST. Therefore, and for studying the relationships involving these characters and LG, it is expected that the clonal correlation calculated as *Type B* provides a more reliable estimate of the population value of the quantitative genetic parameter under consideration.

The *Type A* clonal correlations were calculated by,

$$\hat{r}_a(x, y) = \frac{\hat{\sigma}_c(x, y)}{\sqrt{\hat{\sigma}_c^2(x) \cdot \hat{\sigma}_c^2(y)}}$$

where $\hat{r}_a(x, y)$ is the *Type A* clonal correlation between the traits x and y , $\hat{\sigma}_{c(x,y)}$ is the estimated clonal covariance between x and y , $\hat{\sigma}_{c(x)}^2$ and $\hat{\sigma}_{c(y)}^2$ are the clonal components of variance estimated for x and y (respectively). The estimate of the clonal covariance $\hat{\sigma}_{c(x,y)}$ was obtained by using all the ramets with information on both characters x and y . The corresponding clonal components of variance were estimated from the common individual tree data, following the analysis of variance method described before.

The procedure adopted to derive the clonal covariances was the same as that applied for estimating the clonal variances: this means equating the observed mean cross products to their *Type III* expectations of mean cross products, and solving the equations as explained before. The MANOVA statement of the SAS[®] PROC GLM provided the sums of cross products. The options HTYPE = 3 and ETYPE = 3 were used to give the matrices with the *Type III* sums of cross products for clone and error (respectively).

The *Type B* clonal correlations were computed by,

$$\hat{r}_b(x, y) = \frac{\hat{r}_{\bar{p}}(x_1, y_2)}{\sqrt{\hat{R}_{\bar{c}(x_1)} \cdot \hat{R}_{\bar{c}(y_2)}}}$$

where $\hat{r}_b(x, y)$ is the *Type B* clonal correlation between the traits x and y , $\hat{r}_{\bar{p}}(x_1, y_2)$ is the phenotypic correlation of clone means estimated between x measured in "environment" 1 and y measured in "environment" 2, $\hat{R}_{\bar{c}(x_1)}$ and $\hat{R}_{\bar{c}(y_2)}$ are the square roots of the repeatabilities of clone means estimated for x and y at the "environments" (or sampling groups) 1 and 2 (respectively).

For each trait measured in the respective sampling group, least-squares means were obtained for each clone. The LSMEANS statement of the SAS[®] PROC GLM computes the least-squares means which, for an unbalanced design, are estimators of the class marginal means that are expected for balanced data (LITTELL *et al.*, 1991). The SAS[®] PROC CORR (SAS[®], 1990a) was applied to calculate the phenotypic correlations of least-squares clone means for each pair of traits, measured on common clones at two different sampling groups.

In order to compare the *Type A* clonal correlations with respect to their accuracy, the approximation described by MODE and ROBINSON (1959) and BECKER (1984) was adapted to the present situation for calculating the standard errors of the estimates. Due to the absence of an approximate procedure available for the current situation, no attempt was made for calculating the standard errors of the *Type B* estimates.

Selection strategy (index selection)

The index selection approach was employed to explore the possibilities of improving an aggregate genotype function (H) integrating – *either simultaneously or in alternative combinations* – growth, wood properties and stem straightness. *Table 1* lists the multiple trait indices applied to optimize selection for several definitions of the aggregate genotype function.

The computations for constructing the selection indices were carried out by using the SAS[®]/IML software (SAS[®], 1990c).

H combining all traits

To optimize selection for a breeding objective combining all traits, an aggregate genotype function was specified as:

$$H = w_{VI}G_{VI} + w_{PL}G_{PL} + w_{LG}G_{LG} + w_{ST}G_{ST}$$

where VI, PL, LG and ST are defined as before; $G_{VI} \dots G_{ST}$ are the candidate's unknown genetic values for the traits VI...ST, and $w_{VI} \dots w_{ST}$ are the respective weighting coefficients. $G_{VI} \dots G_{ST}$ concern the candidate's *total genetic worth* (i.e. *clonal value*) to be predicted for young (i.e. at age 9 years from field planting) vegetatively propagated material.

The *unequal* information, available for the traits measured on the examined sample of clones, was taken into consideration when constructing the selection indices. An index value I was estimated for each clone by using the following expression:

$$I = b_{VI_1}VI_1 + b_{PL_1}PL_1 + b_{LG_1}LG_1 + b_{ST_1}ST_1 + b_{VI_2}VI_2 + b_{PL_2}PL_2 + b_{ST_2}ST_2$$

where $b_{VI_1} \dots b_{ST_1}$ are estimates of index coefficients, weighting the phenotypic measures $VI_1 \dots ST_1$ obtained in group 1; $b_{VI_2} \dots b_{ST_2}$ are the index coefficients estimated for $VI_2 \dots ST_2$ measured in group 2. At the sampling group in question, the phenotypic measures pertain to least-squares clone means estimated for traits VI...ST from multiple observations on the given selection candidate.

The index coefficients are partial regression coefficients of H on I and, in the least-squares sense, they are estimated to maximize the correlation between predicted (i.e. I) and actual (i.e. H) aggregate genotype values. The least-squares approach leads to a set of simultaneous index equations with solution $b = P^{-1}Cw$, where: b is a (7 x 1) vector of index coefficients, weighting the phenotypic measures included in I; P is a (7 x 7) matrix of variances and covariances of the phenotypic measures included in I; C is a (7 x 4) matrix of clonal covariances between the phenotypic measures included in I and the clonal values of the target traits combined in H; w is a (4 x 1) vector of weighting coefficients of the target traits integrated in H. The elements in the P and C matrices were specified taking into consideration the distinct variance-covariance structures of the observations in sampling groups 1 and 2.

The vector $\Delta \hat{g}$ of genetic responses expected in the characters integrated in H, under selection on I, was obtained by applying the following expression (CUNNINGHAM, 1970; WHITE and HODGE, 1989):

$$\Delta \hat{g} = \frac{b'C}{\sqrt{b'Pb}} \cdot i$$

where b , P and C are defined as above, b' is the transpose of b , and i is the intensity of selection (i.e. the standardized selection differential) on the index.

The definition of H assumed that Sitka spruce wood with favourable combinations *simultaneously* in growth, wood properties and stem quality will contribute to improve the raw

material for pulp production, implying that genetic deterioration in any of the traits involved will have an adverse impact on pulpwood value. Given this, unit changes in the composite characters may, however, have different relative economic importance, depending on their effects on the quality of the final product and total pulping costs for a pulp mill based on given pulping process and conditions.

Due to the lack of quantitative economic data, it was not possible to estimate the relative economic value of a unit change in each of the examined traits, for pulp and paper manufacture using young Sitka spruce wood. Under these circumstances, the traits combined in the definition of H were weighted according to the "equal emphasis" approach, described by COTTERILL and JACKSON (1985) and COTTERILL and DEAN (1990). The weighting coefficients calculated by this method give equal emphasis to a phenotypic standard deviation change in each of the composite characters or, in other words, it is assumed that changes of one phenotypic standard deviation in each of these traits have equal economic value. Similar procedures have been reported in the index selection literature (WILCOX and SMITH, 1973; SHELBOURNE and LOW, 1980; COTTERILL, 1985; DEAN *et al.*, 1986; FALCONER, 1989; BORRALHO *et al.*, 1992).

"Equal emphasis" weights were placed on the traits in H under selection on the index I1 (Table 1), and they were deter-

mined as the reciprocal of the phenotypic standard deviation of clone means, by using all the information available for the respective character in the examined sample of clones. Negative weighting coefficients were given to PL and LG, as selection will be directed towards decreasing the average level of these characters for young Sitka spruce pulpwood produced under fast growing conditions.

The iterative approach suggested by COTTERILL and DEAN (1990, page 37) was then applied to examine the sensitivity of the selection index to different combinations of empirical weights. Alternative combinations of weighting coefficients were developed by multiplying the "equal emphasis" weights by factors of 3, 5 and 10, which gradually increase the importance placed on the given trait.

The effects of inputting different combinations of weighting factors were analysed in terms of genetic responses expected in VI, wood properties and ST, following index selection. The predicted responses were sensitive to variations in weighting factors across the examined range, reflecting particularly the effect of the adverse clonal correlations between growth and the wood properties. The selection alternatives allowed for further examination were those which did not lead to genetic decline in any of the target traits. In addition it was assumed that, relatively to the other characters, ST has less importance for pulp production. Given this, the options that were then

Table 1. – Multiple trait indices (I) applied to optimize selection for several definitions of the aggregate genotype function (H): $G_{VI} \dots G_{ST}$ are the candidate's unknown genetic values for the traits VI...ST, and $w_{VI} \dots w_{ST}$ are the respective weighting coefficients. The weighting coefficients indicated in the table represent the factors by which the "equal emphasis" weights were multiplied, according to the iterative approach suggested by COTTERILL and DEAN (1990).

selection index		definition of H	weighting coefficients				
			VI	PL	LG	ST	
H combining all traits	I1	$H = w_{VI}G_{VI} + w_{PL}G_{PL} + w_{LG}G_{LG} + w_{ST}G_{ST}$	1	-1	-1	1	
	unrestricted indices		I2	5	-3	-3	1
			I3	10	-10	-1	5
			I4	10	-3	-10	1
	restricted indices		R1	3	0	0	1
			R2	0	-5	-5	1
H excluding some traits	unrestricted indices	I5	1.7	-1	-1.3	0	
		I6	2	-2	0	1	
		I7	1	-1	0	0	
		I8	2	0	0	1	
		I9	0	-1	-1	0	

taken into account to optimize selection for H refer to combinations of weighting coefficients attempting to (Table 1):

- achieve a simultaneous genetic progress in growth, wood properties and stem straightness (i.e. I2);
- keep at (or close to) zero the genetic response expected in lignin content (i.e. I3) or in whole-ring density (i.e. I4).

The imposition of restrictions on the conventional index selection methodology was introduced by KEMPTHORNE and NORDSKOG (1959). The idea is to hold at zero change the expected genetic response(s) in the restricted character(s), while maximizing the improvement in the other traits in H.

The restricted index selection approach described by CUNNINGHAM (1970) was applied to impose a restriction on both wood properties or, on the other hand, on growth. Taking the “equal emphasis” weights as a base, several restricted selection indices were derived under alternative combinations of weighting coefficients. With selection based on R1 (Table 1), a set of weights was chosen to maximize the genetic response in growth and to improve somewhat ST, while preventing a genetic deterioration in the two wood property traits. The restricted index R2 (Table 1) attempts to optimize selection for an aggregate genotype function keeping at zero change the genetic response in growth, while maximizing the genetic responses in *both* wood properties and improving somewhat ST.

H excluding some traits

In the context of a breeding programme for improved Sitka spruce pulpwood, measuring and selecting simultaneously on growth, whole-ring density, lignin content and stem quality may not be justified and/or feasible for a given pulp production system. Nevertheless, once the information on genetic correlations is available, it is possible to predict correlated responses in secondary characters for evaluating the consequences of not including them in the breeding programme.

Table 1 presents multiple trait indices (I5 to I9) that aim at optimizing selection for alternative aggregate genotype functions, defined by the following subsets of target characters:

- growth, whole-ring density and lignin content (i.e. I5);
- growth, whole-ring density and stem straightness (i.e. I6);

- growth and whole-ring density (i.e. I7);
- growth and stem straightness (i.e. I8);
- whole-ring density and lignin content (i.e. I9).

When the definition of H *did not include* LG, the specification of the elements in the P matrix was simplified by pooling the experimental data, obtained for the measured characters in both of the sampling groups considered. Therefore, the phenotypic measures included in the linear function I are least-squares clone means estimated with all the observations made on the examined character. For these cases, the C matrix is identical to the G matrix of clonal variances and covariances of the target traits combined in H.

The *direct* genetic responses expected in the target traits, under selection on the multiple trait indices, were computed by using the expression given above for $\Delta\hat{g}$. *Correlated* genetic changes were also predicted for secondary characters (i.e. traits not combined in H) as the result of selection based on the multiple trait indices. For each correlated response being predicted, an extra column was added to the matrix C, referring to the clonal covariances between the phenotypic measures included in I and the secondary character in question; an extra row was also added to the column vector w, corresponding to a zero weighting coefficient placed on the secondary character.

The combination of weighting coefficients placed on the target traits in I5, I6, I8 and I9 aims at achieving a genetic progress in these characters that will not be much altered when compared with that under selection on, respectively, I2, I3, R1 and R2.

Conversion of the predicted genetic responses in pilodyn into corresponding wood density changes

As no direct evaluations of wood density were obtained from the F205A clonal trial, an approximation (described in detail by COSTA E SILVA, 1996) was taken to express the predicted genetic responses in PL into corresponding wood density changes.

A regression model was constructed to obtain an estimate of the effect on wood density of a unit change in PL for young Sitka spruce trees – the slope of the regression equation relating wood density to PL indicated that basic density is

Table 2. – Test means, range of clone means and analysis of variance for growth and quality characters measured at age 9 years from field planting in 30 clones of Sitka spruce.

characters	test mean	range of clone means	analysis of variance	
			F_{VALUE}	probability of $F_{\text{CALC.}} > F_{\text{TAB.}}$
height (dm)	60.0	47.5 - 71.6	5.64	< 0.001
diameter (mm)	78.9	62.7 - 95.7	2.86	< 0.001
volume (dm ³ /tree)	41.1	21.8 - 66.6	3.21	< 0.001
pilodyn (mm)	14.4	11.4 - 17.5	4.40	< 0.001
lignin content (%)	26.6	24.4 - 28.2	2.99	< 0.001
stem straightness (point)	5.2	4.0 - 6.9	6.57	< 0.001

increased by 8.6 kg/m³ for each unit (i.e. mm) decrease in PL penetration. When these results were expressed relatively to the means estimated for the traits in question, 1% decrease in the PL measurement contributed to an increase of 0.6% in basic density. These figures were used to convert the predicted genetic responses in PL into expected changes in wood density, taking 360 kg/m³ as an approximate estimate of the average value of breast-height density in the examined clone material at age 9 years from field planting.

Results

Clonal variation and repeatabilities

For the characters evaluated in this study, the estimated test means, the range of clone means and the results of the analysis of variance are presented in *table 2*. Under the linear model defined for the observed data, the results in *table 2* indicated highly significant ($P < 0.001$) clonal variance for all the traits under examination.

The estimates of the components of variance between clones and due to random error are given in *table 3*, together with the phenotypic standard deviations and the repeatabilities calculated on the individual ramet and clone-mean bases. The clonal repeatability is relevant for the “mass selection and cloning” breeding option, whereas the clone-mean repeatability is applicable to the “clone selection and cloning” strategy (following the terminology given by MULLIN and PARK, 1992).

For the growth traits, the clonal repeatability estimates ranged from 0.18 to 0.36, with the latter value calculated for HT. For the wood properties, the clonal repeatabilities were 0.32 and 0.42 for PL and LG, respectively, and for ST the

estimate obtained was 0.40. These results suggest that moderate genetic responses could be expected for HT, wood properties and ST, following mass selection and subsequent vegetative propagation of the selected phenotypes for planting.

The magnitudes of the clone-mean repeatabilities were considerably higher than those calculated on the individual ramet basis, reflecting the reliability of multiple measures on each selection candidate as a better guide to the respective genetic (i.e. clonal) value. The estimates obtained, ranging from 0.65 to 0.85, indicate that the characters studied here are expected to respond well under selection based on clone means, and subsequent vegetative propagation of the best tested clones for operational plantation.

Despite the moderate to high repeatabilities for LG, the phenotypic variation was rather low (i.e. phenotypic coefficients of variation of 3.7% and 2.9% on the individual ramet and clone mean bases, respectively), which will limit the magnitude of the genetic progress expected in this trait from selection and breeding.

The accuracy of the heritability values obtained under given experimental conditions can be expressed in terms of the sampling coefficient of variation of this genetic parameter (ROBERTSON, 1959). The calculated sampling coefficient of variation (i.e. standard error/estimate) ranged from about 19% to 35% for the repeatability based on individual tree values, and from about 5% to 16% for the repeatability determined on a clone-mean basis. This suggests that, for the traits examined at the given age and site, the population value of the genetic parameter of interest can be inferred with reasonable accuracy from the estimates derived under the applied sampling procedures and experimental conditions.

Table 3. – Estimated variance components, phenotypic standard deviations and repeatabilities for growth and quality characters measured at age 9 years from field planting in 30 clones of Sitka spruce. For the variance components and repeatability values, the approximate estimates of the standard errors are given in parentheses.

characters	variance components		phenotypic standard deviations		repeatabilities	
	clonal	error	individ. ramet	clone means	clonal	clone means
height (dm)	34.047 (10.536)	60.486 (5.875)	9.723	6.433	0.36 (0.076)	0.82 (0.048)
diameter (mm)	53.599 (21.103)	236.966 (23.016)	17.046	9.075	0.18 (0.063)	0.65 (0.095)
volume (dm ³ /tree)	99.149 (36.854)	370.247 (35.961)	21.666	12.003	0.21 (0.066)	0.69 (0.085)
pilodyn (mm)	1.796 (0.593)	3.855 (0.401)	2.377	1.524	0.32 (0.077)	0.77 (0.062)
lignin content (%)	0.401 (0.158)	0.551 (0.108)	0.976	0.777	0.42 (0.118)	0.67 (0.108)
stem straightness (point)	0.516 (0.155)	0.764 (0.074)	1.132	0.780	0.40 (0.077)	0.85 (0.042)

Table 4. – Clonal correlations for growth and quality characters measured at age 9 years from field planting in 30 clones of Sitka spruce. The clonal correlations involving LG and the other traits were calculated as *Type B* estimates. The remaining values refer to *Type A* clonal correlations. The approximate standard errors of the correlation coefficients are given in parentheses for the *Type A* estimates.

	height	diameter	volume	pilodyn	lignin content	stem straightness
height		0.62 (0.143)	0.84 (0.072)	0.24 (0.208)	0.34	0.16 (0.216)
diameter			0.96 (0.021)	0.55 (0.173)	0.47	0.04 (0.269)
volume				0.48 (0.187)	0.42	0.05 (0.244)
pilodyn					0.15	0.12 (0.218)
lignin content						-0.34

LEE (1993) reported individual heritability estimates for height and diameter of 0.24 and 0.13, respectively, from Sitka spruce half-sib progeny tests at age 10 years. LEE (1993) also indicated an individual heritability of 0.41 for pilodyn at age 15. Working with 15-year-old open-pollinated families of interior spruce in British Columbia, YANCHUK and KISS (1993) obtained an individual heritability of 0.22 for pilodyn. Clonal repeatabilities for lignin content in slash pine wood at age 5 (EINSPAHR *et al.*, 1964) and in loblolly pine wood at age 10 years (VAN BULJTENEN *et al.*, 1968) were 0.72 and 0.42, respectively.

Clonal correlations

The estimated clonal correlations are shown in *table 4*, and they concern:

Clonal correlations between growth traits

The clonal correlation between HT and DBH was 0.62, indicating that clonal selection for one of these characters is expected to produce a considerable indirect improvement in the other. A phenotypic clone-mean correlation of 0.65 was estimated for these traits, from observations made in a Sitka spruce clonal trial at age 18 years from field planting (COSTA E SILVA *et al.*, 1994).

High values of the genetic parameter in question were also obtained for VI and the other growth traits, the clonal correlation between VI and DBH being stronger than that pertaining to the former character and HT. A similar tendency was observed for the phenotypic clone-mean correlations estimated in the experiment mentioned above (COSTA E SILVA *et al.*, 1994), where total stem volume was evaluated by using the Danish standard volume functions described by MADSEN (1987).

Clonal correlations between quality traits

The clonal correlation between PL and LG was low but favourable (i.e. 0.15), and it suggests that a simultaneous

genetic progress may be achieved in both traits, under multiple trait index selection that aims at both reducing the population level in lignin content and increasing the whole-ring density (i.e. decreasing the pilodyn measurement).

For ST and LG, the clonal correlation was -0.34 , pointing out that clonal selection for more straight stems may be accompanied by a somewhat reduction in lignin content. ST was poorly positive correlated with PL. In Sitka spruce, LEE (1993) reported a null genetic correlation between stem straightness at age 7 and pilodyn at age 17 years from planting.

In conifers, poor stem straightness tends to be associated with higher frequency of compression wood, which is characterized for having higher density and lignin content relative to those found in normal wood (ZOBEL and VAN BULJTENEN, 1989). When sampling the ramets for PL and LG measurements, care was taken to avoid compression wood, by sampling side or opposite wood in more leaning stems. However, it is not possible to assure that the sampling was not affected by the presence of compression wood in the cell tissues and, to a certain extent, this may be reflected on the magnitude and sign of the clonal correlations estimated between ST and either PL or LG.

Clonal correlations between growth and quality traits

LG had consistent positive clonal correlations with the growth traits – the estimates obtained were 0.47 and 0.42 with DBH and VI (respectively), the correlation with HT being somewhat lower (i.e. 0.34). In spruce, NYLINDER (1973) reported that lignin content increases with vigour of growth (expressed as increasing ring width).

Positive clonal correlations were also estimated for PL and the growth traits – the calculated values were 0.55 and 0.48 with DBH and VI (respectively), the estimate with HT being lower (i.e. 0.24). In Sitka spruce, and estimating genetic correlations between pilodyn at age 17 years and growth traits

evaluated at several ages from planting, LEE (1993) indicated values ranging from 0.55 to 0.64 with height, and from 0.66 to 0.72 with diameter. For interior spruce in British Columbia, YANCHUK and KISS (1993) reported values of 0.20 and 0.35 for the genetic correlations involving height and diameter (respectively) and PL observations made at age 15 years from planting.

At the phenotypic and/or genetic levels, negative correlations between growth rate and wood density have been pointed out for *Picea* spp., in the juvenile and mature periods of wood formation (BRAZIER, 1970; NYLINDER, 1973; OLESEN, 1976; HARVALD and OLESEN, 1987; PETTY *et al.*, 1990; CORRIVEAU *et al.*, 1991; COSTA E SILVA *et al.*, 1994; ZHANG, 1995).

The estimated clonal correlations between the wood properties and the growth traits were unfavourable, in the sense that clonal selection which aims at reducing the population level in lignin content and/or increasing the whole-ring density is expected to produce an indirect genetic decline in growth. However, the magnitudes of the estimates were moderate, indicating that it may be possible to find genotypes with desirable characteristics in various trait combinations.

Weak positive clonal correlations (ranging from 0.04 to 0.16) were obtained for ST and the growth traits. In Sitka spruce, and calculating genetic correlations between stem straightness

at age 7 years and growth characters measured at several ages in the field, LEE (1993) reported values ranging from 0.00 to 0.11 for height, and from 0.00 to 0.04 for diameter.

Index selection

H combining all traits

Table 5 shows the selection options that were put forward for examination to represent the possibilities of improving H with different sets of weights placed on simultaneous combinations of growth, wood properties and stem straightness. The predicted genetic changes in the individual characters are given in the measurement units for each trait and in percentage of the trait mean values (in parentheses). They pertain to responses expected at age 9 years when the best clone in every 100 ($i = 2.607$) is selected on the basis of its index value, considering the population size equal to 300 clones (BECKER, 1984). This assumes that the quantitative genetic parameters – derived from the random sample of 30 clones, selected among the 253 clones represented in the whole F205A experiment – are valid for a 9-year-old Sitka spruce population of 300 clones, growing in environmental conditions similar to those of the present clonal trial.

With selection based on the index I1, where “equal emphasis” weights were placed on the traits in H, ST is the character most favoured and no changes are expected in VI.

Table 5. – Genetic responses expected from one generation of truncation selection based on *unrestricted* (I1 to I4) and *restricted* (R1 and R2) multiple trait indices, that aim at improving an aggregate genotype *combining simultaneously* growth, wood properties and stem quality. The restricted index approach was employed to construct two types of selection indices, which attempt to prevent from a genetic decline in both whole-ring density and lignin content (i.e. R1) or in growth (i.e. R2). The weighting coefficients represent the factors by which the “equal emphasis” weights were multiplied. The expected genetic changes are expressed in the measurement units of the trait, as well as in percentage of the trait mean values (in parentheses), and assuming a selection intensity on the index equal to 2.607.

index ref.	weighting coefficients				expected genetic responses				
	VI	PL	LG	ST	VI dm ³ /tree	PL mm	WD ^a kg/m ³	LG %	ST point
I1	1	-1	-1	1	0.1 (0.2)	-1.1 (-7.6)	16.5 (4.6)	-0.8 (-3.0)	1.3 (25.0)
I2	5	-3	-3	1	9.4 (22.9)	-1.0 (-6.9)	15.0 (4.2)	-0.4 (-1.5)	0.8 (15.4)
I3	10	-10	-1	5	8.4 (20.4)	-1.6 (-11.1)	24.0 (6.7)	0.0 (0.0)	0.7 (13.5)
I4	10	-3	-10	1	7.5 (18.3)	-0.13 (-0.9)	1.95 (0.5)	-0.8 (-3.0)	0.9 (17.3)
R1	3	0	0	1	15.6 (38.0)	0.0 (0.0)	0.0 (0.0)	0.0 (0.0)	0.9 (17.3)
R2	0	-5	-5	1	0.00 (0.0)	-1.9 (-13.2)	28.5 (7.9)	-0.8 (-3.0)	0.7 (13.5)

^a) The correspondence between the predicted genetic responses in PL and expected changes in wood density (WD) was made as described by COSTA E SILVA (1996).

This is apparent when the predicted genetic responses are expressed in percentage of the overall mean values (Table 5), as well as when they are normalized using the phenotypic clone-mean standard deviations as references (data not shown). ST was the most heritable character and, besides the weak association with PL, was not adversely correlated with the other traits at the clonal level. Both of these factors contributed to the results obtained under selection on I1. The direct impact of ST on pulp production is less known, although BLAIR *et al.* (1974) reported that wood from straight loblolly pine trees improved significantly pulp yield (on a percentage basis) and tear factor, relative to wood from crooked trees. Nevertheless, selection based on I1, hence favouring ST, may be difficult to accept when the aim is to improve pulpwood value.

Compared with I1, selection index I2 – with greater weighting given to VI (5 times) and wood properties (3 times) than to ST – resulted in a large increase in gain for VI at the expense of less marked changes in the wood traits and ST. With selection based on I2, the effect of the unfavourable clonal correlations is prevented under a combination of weighting coefficients leading to a certain minimum improvement in each of the traits involved. In other words, for a given selection intensity, the responses in the individual characters following selection on I2 ranged from around 30% to 45% relatively to those expected under *single trait simple selection*. The selection option I2 approximates the conservative solution suggested by several authors (NAMKOONG, 1979; KING *et al.*, 1988; NAMKOONG *et al.*, 1988), in conditions of uncertainty of value functions involving traits for which reliable information of their economic importance is lacking.

Under the unrestricted indices I3 and I4, the combinations of weighting coefficients lead to outcomes of selection that are similar to those from imposing a restriction on changes in lignin content and whole-ring density, respectively. When compared with I1, I3 raises markedly the predicted response in VI and improves wood density to some extent. In comparison to I1, selection based on I4 does not change LG and increases considerably the gain for VI.

Using the restricted index R1, the expected genetic gain in growth with restrictions imposed on both wood properties is substantially maximized relatively to that in I1. Selection based on R2, with a restriction imposed on growth, improves somewhat the genetic progress in whole-ring density and does not change LG when compared with I1. For a given selection intensity, the response in growth following selection on R1 was 72% relatively to that predicted under *single trait simple selection*. The expected genetic progress in PL and LG to selection based on R2 was only 62% and 56% as great as selection on either trait alone.

H excluding some traits

The genetic changes expected at age 9 years, following selection ($i = 2.607$) on the multiple trait indices presented in table 6, concern both the *direct* responses in the target traits and the *correlated* (or indirect) responses in the characters not combined in the definition of H.

A marginal indirect improvement can be achieved in ST following selection on I5 (Table 6), for predicted genetic responses in growth and in the wood properties that are not appreciably altered when compared with those expected in the

Table 6. – Genetic responses expected from one generation of truncation selection based on multiple trait indices that aim at improving alternative aggregate genotype functions, *not combining simultaneously* growth, wood properties and stem quality. The predicted genetic changes refer to both the *direct* responses in the target traits and *correlated* responses in secondary traits not integrated in the definition of the aggregate genotype function. The weighting coefficients placed on the target traits represent the factors by which the "equal emphasis" weights were multiplied. The expected genetic changes are expressed in the measurement units of the trait, as well as in percentage of the trait mean values (in parentheses), and assuming a selection intensity on the index equal to 2.607.

index ref.	weighting coefficients				expected genetic responses				
	VI	PL	LG	ST	VI dm ³ /tree	PL mm	WD ^a kg/m ³	LG %	ST point
I5	1.7	-1	-1.3	0	7.6 (18.5)	-1.2 (-8.3)	18.0 (5.0)	-0.5 (-1.9)	0.3 (5.8)
I6	2	-2	0	1	9.4 (22.9)	-1.5 (-10.4)	22.5 (6.3)	0.1 (0.4)	0.7 (13.5)
I7	1	-1	0	0	9.7 (23.6)	-1.9 (-13.2)	28.5 (7.9)	0.4 (1.5)	-0.2 (-3.9)
I8	2	0	0	1	18.5 (45.0)	1.4 (9.7)	-21.0 (-5.8)	0.2 (0.8)	1.0 (19.2)
I9	0	-1	-1	0	-13.9 (-33.8)	-2.5 (-17.4)	37.5 (10.4)	-1.0 (-3.8)	0.2 (3.9)

^a) The correspondence between the predicted genetic responses in PL and expected changes in wood density (WD) was made as described by COSTA E SILVA (1996).

Table 7. – SPEARMAN rank correlation coefficients between the index values estimated for each clone, by solving the index equations referring to the examined selection options.

	I2	I3	I4	R1	R2	I5	I6	I7	I8	I9
I1	0.85	0.74	0.81	0.70	0.88	0.69	0.66	0.42	0.36	0.56
I2		0.87	0.83	0.89	0.88	0.90	0.81	0.70	0.50	0.40
I3			0.52	0.81	0.78	0.72	0.96	0.86	0.42	0.34
I4				0.71	0.78	0.81	0.43	0.31	0.52	0.36
R1					0.62	0.70	0.78	0.66	0.77	0.01
R2						0.87	0.71	0.63	0.16	0.75
I5							0.67	0.70	0.28	0.52
I6								0.92	0.42	0.29
I7									0.29	0.26
I8										- 0.47

same characters under selection on I2 (Table 5). Minor adverse correlated responses are expected in LG following selection on I6 (Table 6), for a genetic progress in the target traits that is not substantially changed when compared with the corresponding outcomes predicted under selection on I3 (Table 5).

Selection based on I7 may lead particularly to an unfavourable correlated response in LG. Under selection on I7, the average content of LG can be *increased* up to 0.4% in one generation, while following selection on I5 the corresponding response is a *decrease* of 0.5%. When comparing these two options for improved *chemical* pulp production, a difference of 0.9% in LG may result in changes in pulp yield per unit weight of dry wood. If the result is a decrease in this variable, pulp yield per unit volume of wood will also be reduced, unless it can be compensated by the improvement expected in wood density (in the present example, it means a difference of 10.5 kg/m³ estimated between selection under I5 and I7).

The two most extreme options, I8 and I9, will lead to substantial correlated losses in wood density and growth, respectively. On the other hand, under selection based on I8 and I9, the direct genetic progress in growth and wood properties is considerable when compared with the conservative option I2. Particularly in the case of I9, a simultaneous improvement of 37.5 kg/m³ and – 1.0% was estimated for wood density and lignin content.

Correlations between selection indices

The candidate rankings based on the index values were computed for all the selection alternatives presented above. The total agreement between candidate rankings from the various selection indices was evaluated by calculating Spearman rank correlation coefficients (Table 7).

Selection based on I2 leads to clone rankings that correlate well with most of the other indices. The exceptions refer to the lower Spearman rank correlation coefficients of 0.7, 0.5 and 0.4, estimated between I2 and the indices I7, I8 and I9, respectively.

The clone rankings generated from I7 agree well with those from I3 and I6, with estimated Spearman rank correlations of 0.86 and 0.92, respectively. The indices I8 and I9 are in general poor correlated with the other selection options. Furthermore, they lead to clone rankings that are very different from each other: no common clones were found among the best five candidates identified by I8 and I9 (data not shown) and the sign of the respective correlation (Table 7) points out an inverse relationship among the index values, suggesting a divergent direction in terms of selection decisions.

Discussion

Inclusion of lignin as a target trait in a breeding programme

The estimated genetic changes in LG were small in magnitude, even with high selection intensity of 1 in 100. In this sense, the question will be if the predicted genetic progress in LG justifies its inclusion in an intensive breeding programme for improved Sitka spruce pulpwood.

The average content of LG (say a mean level of 27% at age 9 years) is expected to be reduced by an amount varying from 0.4% to 1% in one generation, depending on the index used. If changes of this magnitude have any impact on pulp production, then many candidates for selection have to be screened (as, for example, in different series of clonal trials), in order to get a reasonable number of clones for both operational plantation and as parents for the next-generation of breeding.

Although lignin may not currently be considered as a target trait owing to, for example, difficulties with large scale screening in breeding programmes, it is still a wood property with potential importance for pulp production. Therefore, the correlated response in LG should be considered when deciding on a selection strategy. The results in *table 6* indicated that the most noticeable genetic decline is expected to occur when selection aims at improving simultaneously growth and wood density (i.e. I7), and it reflects the moderate but adverse clonal correlation between VI and LG (*Table 4*). This tendency was reduced when ST was included in H, as a result of the favourable clonal correlation between ST and LG.

The relationship between the lignin content in wood and pulp yield and quality variables would provide the basis to evaluate the changes in this character, in terms of maximization of economic returns for a given pulp production system. In pulp industry, a *slight* increase in cellulose yield can be very valuable: reducing lignin content in wood would be advantageous in decreasing energy and chemical consumptions, while increasing pulp yield per unit of dry wood charged. Giving as an example the study made by BORRALHO *et al.* (1993) for *Eucalyptus globulus* pulp industry, the estimated cost savings of 1% increase in pulp yield (per unit weight of dry wood) at harvest were considerable, and the resulting profit would repay the extra costs of measuring and selecting on pulp yield in the breeding programme. In this context, the FTIR technique used in the present study may offer some advantages when compared with conventional wet-lab chemical methods. It is a *rapid* assessment technique, which is able to estimate with reasonable *precision* the content of lignin in *small* wood samples, hence contributing to a reduction in the measurement costs.

Single versus multiple populations

Increasing whole-ring density and reducing lignin content (or keeping constant their population levels) may be important goals to improve the wood quality of *young* Sitka spruce in *fast growing conditions*. Although quantitative economic data are absent to validate and make decisions on the most appropriate strategy, selection based on I2 – with more weighting given to VI (5x) relatively to PL (3x), LG (3x) and ST (1x) – is a reasonable compromise solution to improve a *single broad population* for all characters. Alternatively, selection index I6, placing two times more weight on both VI and PL than on ST, is a good compromise option to improve simultaneously these traits, while resulting in minor indirect changes in LG.

Instead of a single index solution, contrasting selection options – as, for example, I2, I7, I8 and I9 – could be considered in the context of a *multiple population* strategy (NAMKOONG, 1976; BURDON and NAMKOONG, 1983; NAMKOONG *et al.*, 1988; WELLENDORF, 1995).

Breeding objectives intended to improve pulp production can be defined in terms of increasing dry weight yield per unit area of plantation and/or the digester yield (BORRALHO *et al.*, 1992). The options I8 and I9, attempting to optimize selection for H placing emphasis either on growth or wood properties, could constitute a system of two populations defined, respectively, to increase stem dry weight per unit area of plantation and pulp yield (per unit of both volume of wood and weight of dry wood). In the former case, it must be mentioned that the option I8 assumes that tree growth has the major impact on the intended objective and, therefore, it is supposed that the decrease of wood density in the population will be compensated by the large expected improvement in growth. This has been suggested for several reports with conifers (NAMKOONG *et al.*, 1969;

BRIDGWATER *et al.*, 1983; ZOBEL and VAN BUIJTENEN, 1989) and hardwoods (BORRALHO *et al.*, 1992, 1993).

In a multiple population context, more than one of the illustrated selection options could be followed for the production of pulp suitable for the manufacture of different paper grades, with diverse specifications in strength and optical properties. Taking wood density as an example, the level of this character can have the most important influence on the strength properties of the pulp produced, as reported in DINWOODIE'S (1966) work with Sitka spruce and in other pulping studies with conifers (ZOBEL and VAN BUIJTENEN, 1989). For a certain mean level (say an actual average estimate of 360 kg/m³), the value of improving versus decreasing (or keeping constant) wood density may be determined by the quality requirements of a given paper type and, if economically viable, separate populations could be taken into account for specific end-product uses. In a similar manner, WILCOX and SMITH (1973) formulated several multiple trait selection indices for identifying groups of clones with desired combinations of wood characteristics for diverse end uses in loblolly pine pulp production.

The assessment of the wood properties

The effect of the measurement age

The wood properties examined here were measured at an early age of the clonal experiment and, strictly speaking, the results reported apply only to wood with juvenile characteristics. The information will be of especial interest with short-rotation forestry, as the amount of juvenile wood will dominate the final merchantable volume (ZOBEL and VAN BUIJTENEN, 1989).

The effect of the measurement methods

The wood density and lignin assessments carried out in this work constitute indirect evaluations of the whole-tree values, at the given measurement age and growing conditions. The pilodyn wood tester provides an estimate of the average wood density of the last few annual rings at breast-height. Lignin content was measured in the current annual ring, taken at breast-height. The efficiency of these indirect assessments will depend on the consistency of longitudinal and radial patterns of within-tree variation for the wood properties concerned.

In Sitka spruce (FARR, 1973; BRAZIER and HOWELL, 1979), as well as in other conifers (ZOBEL and VAN BUIJTENEN, 1989), the determination of the average wood density at breast-height has been found to be suitable for evaluating the whole-stem density of *adult* trees. For adult radiata pine trees, ZOBEL and VAN BUIJTENEN (1989, cit. WARREN and MIRAMS, 1963, and HARRIS, 1965) reported that the use of only few outer rings at breast-height can give a satisfactory measure of the whole-stem density values.

Although core wood observations at breast-height can be less efficient for predicting the entire density of older coniferous trees, they can give a good estimate of the *juvenile* wood trait values throughout the stem (ZOBEL and VAN BUIJTENEN, 1989, cit. HARRIS, 1965). Using Sitka spruce clonal material from a 18-year-old experiment, a correlation of 0.85 was calculated on the individual ramet basis ($n = 65$) between the average density of the breast-height rings 6 to 9 counted from the pith and the whole-disc (data not shown). This result suggested that, for *young* Sitka spruce clones, wood density taken from the examined annual rings should give a reasonable indication of the whole-disc density at breast-height.

Several studies with *young* coniferous plant material have been demonstrating the effectiveness of the pilodyn tester for

assessing wood density at a given sampling height. In Douglas-fir (KING *et al.*, 1988) and in interior spruce in British Columbia (YANCHUK and KISS, 1993), high correlations between the pilodyn measure and wood density were found at the phenotypic and genetic levels. On a phenotypic clone-mean basis, the same tendency was pointed out for radiata pine (COWN, 1978) and Sitka spruce (COSTA E SILVA *et al.*, 1994).

Conclusion

The estimated clonal correlations between growth and the wood properties limited the simultaneous improvements in growth rate, wood density and lignin content.

Losses in growth are expected when the selection goal aims at improving the wood properties only. On the other hand, a substantial correlated drop in wood density is expected when selection is based on growth and stem straightness. The most noticeable indirect genetic decline in lignin content occurred when the objective was to increase simultaneously growth rate and wood density. This tendency was prevented when stem straightness was also included as a selection goal, reflecting the favourable clonal correlation between stem straightness and lignin.

Placing more weight on growth would be a reasonable recommendation to achieve genetic progress in all characters following index selection, as growth was the trait which had the greatest sensitivity to changes in the weighting coefficients. The unrestricted index selection approach could overcome the problems of the unfavourable clonal correlations, under a compromise solution giving more weight to growth (5x) relatively to wood properties (3x) and stem straightness (1x). However, when compared with more divergent selection options, targeting either growth or the wood properties, the compromise solution led to substantial reductions in gain for growth rate, wood density and lignin content.

In all cases, the predicted genetic responses in lignin content were small in magnitude, due to a low phenotypic variation of clone means. Nevertheless, keeping constant the population level for lignin content can be important to improve the quality of young pulpwood in fast growing conditions. In this context, placing two times more weight on both growth and wood density than on stem straightness would be a reasonable option to improve simultaneously these characters following index selection, while resulting in minor correlated changes in lignin content.

Acknowledgements

We would like to thank *Fundação para a Ciência e a Tecnologia* (Lisboa, Portugal) and the EU-project AIR (contract nr. CT 920143) for financial support. Thanks to Dr. KNUD CHRISTENSEN (Dep. of Animal Breeding and Animal Health, KVL, Copenhagen), Dr. BJARNE H. NIELSEN (Dep. of Organic Chemistry, DTU, Lyngby), ENG. JOSÉ RODRIGUES (Centro de Estudos de Tecnologia Florestal, Instituto de Investigação Científica Tropical, Lisboa) and Dr. HANS ROULUND (Arboretum, KVL, Hørsholm) for their assistance and helpful criticisms during this work. The technical support given by Dr. HANS C. B. HANSEN (Dep. of Chemistry, KVL, Copenhagen), VIGGO JENSEN and KIRSTEN SØRENSEN (Arboretum, KVL, Hørsholm) is gratefully acknowledged. We are also very grateful for the critical comments and helpful suggestions of Dr. NUNO BORRALHO.

References

BECKER, W. A.: Manual of Quantitative Genetics. Academic Enterprises, Pullman, WA. 196 pp. (1984). — BENTZER, B. G., FOSTER, G. S., HELMBERG, A. R. and PODZORSKI, A. C.: Trends in genetic and environmental parameters, genetic correlations, and response to indirect selection for 10-year volume in a Norway spruce clonal experiment. *Can. J. For. Res.* **19**: 897–903 (1989). — BLAIR, R. L., ZOBEL, B. J., FRANKLIN, E. C., DJERF, A. C. and MENDEL, J. M.: The effect of tree form and rust infec-

tion on wood and pulp properties of loblolly pine. *Tappi* **57**: 46–50 (1974). — BORRALHO, N. M. G., COTTERILL, P. P. and KANOWSKI, P. J.: Genetic parameters and gains expected from selection for dry weight in *Eucalyptus globulus* ssp. *globulus* in Portugal. *For. Sci.* **38**: 80–94 (1992). — BORRALHO, N. M. G., COTTERILL, P. P. and KANOWSKI, P. J.: Breeding objectives for pulp production of *Eucalyptus globulus* under different industrial cost structures. *Can. J. For. Res.* **23**: 648–656 (1993). — BRAZIER, J. D.: Timber improvement. II. The effect of vigour on young-growth Sitka spruce. *Forestry* **43**: 135–150 (1970). — BRAZIER, J. D. and HOWELL, R. S.: The use of a breast-height core for estimating selected whole-tree properties of Sitka spruce. *Forestry* **52**: 177–185 (1979). — BRIDGWATER, F. E., TALBERT, J. T. and JAHROMI, S.: Index selection for increased dry weight in a young loblolly pine population. *Silvae Genet.* **32**: 157–161 (1983). — BURDON, R. D.: Genetic correlation as a concept for studying genotype-environment interaction in forest tree breeding. *Silvae Genet.* **26**: 168–175 (1977). — BURDON, R. D. and NAMKOONG, G.: Short note: multiple populations and sublines. *Silvae Genet.* **32**: 221–222 (1983). — CORRIVEAU, A., BEAULIEU, J. and DAoust, G.: Heritability and genetic correlations of wood characters of Upper Ottawa Valley white spruce populations grown in Quebec. *For. Chron.* **67**: 698–705 (1991). — CORSON, S. R., FOSTER, R. S. and RICHARDSON, J. D.: New Zealand grown spruce and radiata pine can have similar TMP properties. *Appita* **42**: 345–349 (1989). — COSTA E SILVA, J.: Clonal variation in wood quality, growth and growth rhythm in young Sitka spruce (*Picea sitchensis* (BONG.) CARR.): indirect assessment of wood density and lignin amount, estimation of quantitative genetic parameters and index selection for improved pulpwood. Ph. D. thesis, The Royal Veterinary and Agricultural University, Arboretum, Hørsholm. 157 pp. (1996). — COSTA E SILVA, J., NIELSEN, B. H., RODRIGUES, J., PEREIRA, H. and WELLENDORF, H.: Rapid determination of lignin content in Sitka spruce (*Picea sitchensis* (BONG.) CARR.) wood by Fourier transform infrared spectrometry. (Submitted to *Can. J. For. Res.*, 1998). — COSTA E SILVA, J., NIELSEN, U. B. and ROULUND, H.: Sitka spruce clonal performance with special reference to basic density. 23 years' results of a clonal trial. *Silvae Genet.* **43**: 82–91 (1994). — COTTERILL, P. P.: On index selection. II. Simple indices which require no genetic parameters of special expertise to construct. *Silvae Genet.* **34**: 64–69 (1985). — COTTERILL, P. P. and DEAN, C. A.: Successful Tree Breeding with Index Selection. CSIRO, Australia. 79 pp. (1990). — COTTERILL, P. P. and JACKSON, N.: On index selection. I. Methods of determining economic weight. *Silvae Genet.* **34**: 56–63 (1985). — COWN, D. J.: Comparison of the pilodyn and torsionmeter methods for the rapid assessment of wood density in living trees. *N. Z. J. For. Sci.* **8**: 384–391 (1978). — CUNNINGHAM, E. P.: Animal Breeding Theory. Lanbrugs Boghandelen, Universitets Forlag, Oslo (1970). — DEAN, C. A., COTTERILL, P. P. and EISEMANN, R. L.: Genetic parameters and gains expected from selection in *Pinus caribaea* var. *hondurensis* in northern Queensland, Australia. *Silvae Genet.* **35**: 229–236 (1986). — DINWOODIE, J. M.: The influence of anatomical and chemical characteristics of softwood fibers on the properties of sulfate pulp. *Tappi* **49**: 57–67 (1966). — EINSPAHR, D. W., GODDARD, R. E. and GARDNER, H. S.: Slash pine, wood and fiber property heritability study. *Silvae Genet.* **13**: 103–109 (1964). — FALCONER, D. S.: Introduction to Quantitative Genetics. Longman, Harlow, UK. 438 pp. (1989). — FARR, W. A.: Specific gravity of western hemlock and Sitka spruce in southeast Alaska. *Wood Sci.* **6**: 9–13 (1973). — HARDING, T.: British Softwoods: Properties and Uses. Forestry Commission Bull. 77, HMSO, London. 41 pp. (1988). — HARVALD, C. and OLESEN, P. O.: The variation of the basic density within the juvenile wood of Sitka spruce (*Picea sitchensis*). *Scand. J. For. Res.* **2**: 525–537 (1987). — KELLOGG, R. M.: Coming to grips with wood quality. *For. Chron.* **58**: 254–257 (1982). — KEMPTHORNE, O. and NORDSKOG, A. W.: Restricted selection indices. *Biometrics* **15**: 10–19 (1959). — KING, J. N., YEH, F. C., HEAMAN, J. C. and DANCIC, B. P.: Selection of wood density and diameter in controlled crosses of coastal Douglas-fir. *Silvae Genet.* **37**: 152–157 (1988). — KIRK, D. G., BREEMAN, L. G. and ZOBEL, B. J.: A pulping evaluation of juvenile loblolly pine. *Tappi* **55**: 1600–1604 (1972). — LEE, S. J.: Breeding strategy for Sitka spruce in Britain. In: Proc. Nord. Group for Tree Breeding, Edinburgh, Scotland, Oct. 1993. Forestry Commission, pp. 95–109 (1993). — LITTELL, R. C., FREUND, R. J. and SPECTOR, P. C.: SAS® System for Linear Models. SAS® Institute Inc., Cary, NC (1991). — MADSEN, S. F.: Vedmassefunktioner ved Forskellige Aflægningsgrænser og Nøjagtighedskrav for Nogle Vigtige Danske Skovtræarter. Det Forstlige Forsøgsvæsen i Danmark XLI, 350, pp. 41–242 (1987). — MODE, C. J. and ROBINSON, H. F.: Pleiotropism and the genetic variance and covariance. *Biometrics* **15**: 518–537 (1959). — MULLIN, T. J. and PARK, Y. S.: Estimating genetic gains from alternative breeding strategies for clonal forestry. *Can. J. For. Res.* **22**: 14–23 (1992). — NAMKOONG, G.: A multiple-index selection strategy. *Silvae Genet.* **25**: 199–201 (1976). — NAMKOONG, G.: Introduction to Quantitative Genetics in Forestry. US Dept. Agric. Tech. Bull. 1588, 342 pp. (1979). — NAMKOONG, G., BAREFOOT, A. C. and HITCHINGS, R. G.: Evaluating control of wood quality through breeding. *Tappi* **52**:

1935–1938 (1969). — NAMKOONG, G., KANG, H. C. and BROUARD, J. S.: Tree Breeding: Principles and Strategies. Springer-Verlag, Berlin. 180 pp. (1988). — NIELSEN, U. B.: Genetisk variation i Sitkagran (*Picea sitchensis* (BONG.) CARR.) i højdevækst, stammeform og frosthærdighed – vurderet ud fra danske proveniens-, afkoms-, og klonforsøg. Forskningscentret for Skov & Landskab, Forskningsserien 9–94, 332 pp. (1994). — NYLINDER, P.: Wood quality and fibre product. In: Proc. IUFRO-5 Meet., Vol. 2, Republic of South Africa, pp. 832–844 (1973). — OLESEN, P. O.: The Interrelation Between Basic Density and Ring Width of Norway Spruce. Det Forstlige Forsøgsvæsen i Danmark **34**, 340–359 (1976). — PETTY, J. A., MACMILLAN, D. C. and STEWARD, C. M.: Variation of density and growth ring width in stems of Sitka and Norway spruce. *Forestry* **63**: 39–49 (1990). — ROBERTSON, A.: The sampling variance of the genetic correlation coefficient. *Biometrics* **15**: 469–485 (1959). — RYDHOLM, S. A.: Pulping Processes. Interscience Publishers, New York. 1269 pp. (1965). — SAS[®] Institute Inc.: SAS Procedures Guide, Version 6. SAS[®] Institute Inc., Cary, NC (1990a). — SAS[®] Institute Inc.: SAS/STAT User's Guide, Version 6, Vol. 1 and 2. SAS[®] Institute Inc., Cary, NC (1990b). — SAS[®] Institute Inc.: SAS/IML Software: Usage and Reference, Version 6. SAS[®] Institute Inc., Cary, NC (1990c). — SHAPIRO, S. S. and WILK, M. B.: An analysis of variance test for normality (complete samples). *Biometrika* **52**: 591–611 (1965). — SHELBORNE, C. J. A. and LOW, C. B.: Multi-trait index selection and associated genetic gains of *Pinus radiata* progenies at five sites. *N. Z. J. For. Sci.* **10**: 307–324 (1980). — SNEDECOR, G. W. and COCHRAN, W. G.: *Statistical Methods*. The Iowa State University Press, Ames, U.S.A.. 593 pp. (1976). — VAN BULJTENEN, J. P., EINSPAHR, D. W. and PECKHAM, J. R.: Micropulping loblolly pine grafts selected for extreme wood specific gravity. *Silvae Genet.* **17**: 15–19 (1968). — WELLENDORF, H.: Divers nucleus breeding: selection in different directions from one central multiple-trait selected breeding population. In: *Evolution and Tree Breeding*, CTIA/WFGA Conf., Victoria, BC., pp. 87 (1995). — WHITE, T. L. and HODGE, G. R.: Predicting Breeding Values with Applications in Forest Tree Improvement. Kluwer Academic Publishers, Dordrecht. 367 pp. (1989). — WILCOX, M. D. and SMITH, H. D.: Selection indices for wood quality in loblolly pine. In: Proc. 12th South. For. Tree Improve. Conf., Louisiana. pp. 322–342 (1973). — YANCHUK, A. D. and KISS, G. K.: Genetic variation in growth and wood specific gravity and its utility in the improvement of interior spruce in British Columbia. *Silvae Genet.* **42**: 141–148 (1993). — ZHANG, S. Y.: Wood density considerations in black spruce (*Picea mariana*) tree improvement program. In: *Evolution and Tree Breeding*, CTIA/WFGA Conf., Victoria, BC.. pp. 91 (1995). — ZOBEL, B. J. and VAN BULJTENEN, J. P.: *Wood Variation. Its Causes and Control*. Springer-Verlag, Berlin. 363 pp. (1989).

Inheritance and Linkage Relationships of Allozymes, and Estimation of Outcrossing Rates in a Seed Orchard of *Cunninghamia konishii* HAY.

By CH.-T. WANG and T.-P. LIN¹⁾

Taiwan Forestry Research Institute, 53 Nan-Hai Road, Taipei, Taiwan

(Received 16th July 1997)

Summary

Inheritance and linkage relationships of 11 polymorphic loci from 9 enzyme systems in *Cunninghamia konishii* were analyzed by horizontal starch gel electrophoresis using megagametophyte haploid tissues collected from Chyunshan seed orchard, Taiwan. The outcrossing rate was also estimated

based on multilocus and single-locus models. The observation of segregated female gametophytes of heterozygous trees revealed simple MENDELIAN inheritance for most of the allozyme loci. Linkage relationships were examined for 39 pairs of polymorphic allozyme loci. Three pairs with significant joint segregation were detected: *Mdh-1/6Pgd-2* with a recombination value (R) 0.098, *Fest-1/Fest-3* with R = 0.168, and *Fest-2/Fest-3* with R = 0.038. Single-locus estimates (t_s) of outcrossing rate varied between 0.640 and 0.991 with an average of 0.847. Compared with the single-locus outcrossing rate estimates, the multilocus estimate ($t_m = 0.902$) indicated that a part of the inbreeding may be biparental.

¹⁾ Correspond: Dr. Tsan-Piao Lin, Taiwan Forestry Research Institute, 53 Nan-Hai Road, Taipei, Taiwan.
Contribution no. 98 of Taiwan Forestry Research Institute.
FAX: 886 (2) 2389-5531
E-mail: tpl@serv.tfri.gov.tw