Application of spatial statistics in two genetic tests of eastern white pine

Peng Li, Jean Beaulieu, Steen Magnussen,
Gaétan Daoust and Ariane Plourde

Natural Resources
Canada

Ressources naturelles
Canada

Canadian Forest
Service

Service canadien
des forêts

Canadä

Application of spatial statistics in two genetic tests of eastern white pine

Peng Li, Jean Beaulieu, Steen Magnussen[1], Gaétan Daoust,
and Ariane Plourde

[1] Canadian Forest Service, Pacific Forestry Centre, 506 West Burnside Road, Victoria, British Columbia
V8Z 1M5.

TABLE OF CONTENTS

LIST OF TABLES

ABSTRACT

Sharing common microenvironments among neighbour trees and plots introduces positively correlated errors, which violates the assumption of independence among errors in analysis of variance, introduces bias in predicting random effects and overestimates individual narrow-sense heritabilities. The objectives of this study were: 1) to estimate the extent of spatial autocorrelation in two eastern white pine provenance/progeny tests, 2) to examine the efficiency of analysis of covariance in increasing the precision of family and seed source comparisons, 3) to assess the efficiency of two spatial models for improving precision in estimating family and seed source means, and 4) to test a new spatial method for estimating individual narrow-sense heritability. Heights of 7- to 11-year-old trees from 285 families belonging to 71 provenances were measured in two eastern white pine provenance/progeny tests planted in two rows per clearcut strip between hardwood bands in Quebec. Spatial autocorrelation was moderate (range 0.15 to 0.19) at one site and weak (range 0.09-0.12) at the other site. Relative light intensity and drainage classes were tried as covariates in analysis of covariance, but they reduced the error mean squares by less than 1%. At the site with moderate spatial autocorrelation, the two spatial models improved precision in estimating family means, but spatially adjusted and unadjusted family means were very strongly correlated, suggesting no changes in family ranks. At the other site, which had weak spatial autocorrelation, the spatial models did not increase the precision of family comparisons. A new procedure for estimating the additive genetic variance based on the combined effect of genetics and spatial positions of trees was proposed and tested for the two sites. The new method gave acceptable estimates of individual narrow-sense heritabilities, but the statistical properties of the estimators must be examined.

RÉSUMÉ

Le fait que les parcelles et les arbres voisins partagent des microsites communs entraîne des erreurs à corrélation positive, ce qui viole l'hypothèse d'indépendance des erreurs exigée par l'analyse de variance, introduit un biais dans la prévision des effets aléatoires et entraîne une surestimation des héritabilités individuelles au sens strict. Dans le cadre de la présente étude, nos objectifs étaient les suivants : (1) déterminer l'importance de l'autocorrélation spatiale dans deux essais de provenances - descendances de pin blanc; (2) augmenter la précision des comparaisons entre les familles et entre les provenances en utilisant l'intensité lumineuse relative et les classes de drainage comme covariables pour l'analyse de covariance; (3) évaluer deux modèles spatiaux, à l'échelle des parcelles, quant à la capacité de ces modèles à augmenter la précision des comparaisons des familles et des provenances; (4) proposer pour l'estimation de l'héritabilité individuelle au sens strict une nouvelle méthode fondée sur les relations spatiales et génétiques entre arbres survivants. Nous avons mesuré les hauteurs à 7, 8, 9, 10 et 11 ans sur des arbres issus de 285 familles appartenant à 71 provenances, dans deux essais de provenances - descendances de pin blanc réalisés au Québec. Les arbres ont été plantés sur deux rangs dans chaque bande coupée à blanc séparant deux bandes de feuillus. Le coefficient d'autocorrélation était modéré (0,15 à 0,19) dans un des sites et faible (0,09 à 0,12) dans l'autre. Nous avons tenté d'utiliser l'intensité lumineuse relative et les classes de drainage comme covariables pour l'analyse de covariance, mais l'emploi de ces paramètres réduisait de moins de 1 % les carrés moyens des résidus. Dans le cas de la station présentant une autocorrélation spatiale modérée, les deux modèles spatiaux ont augmenté la précision de l'estimation des moyennes de famille, mais les moyennes ainsi ajustées présentaient une corrélation très forte avec les moyennes non ajustées, ce qui semble indiquer que les modèles ne modifiaient pas le classement des familles par ordre de rang. Dans le cas de l'autre station, où l'autocorrélation spatiale était faible, les modèles d'analyse spatiale n'ont pas augmenté la précision des comparaisons entre familles. Enfin, nous proposons une nouvelle méthode d'estimation de la variance génétique additive, fondée sur les effets combinés de la parenté génétique et de la position relative des arbres. Cette nouvelle méthode, que nous avons mise à l'essai dans les deux stations, produit une estimation raisonnable des héritabilités individuelles au sens strict, mais il faudrait encore examiner les propriétés statistiques des estimateurs utilisés.

INTRODUCTION

A key assumption in statistical analysis of genetic tests is the independence of error terms (plot or within-plot) for different observations (Snedecor and Cochran 1967; Searle *et al.* 1992). However, spatial autocorrelation and/or competition may introduce a covariance among neighbouring observations, which could violate the assumption. Spatial autocorrelation refers to the phenomenon that neighbouring trees and plots tend to be more alike than experimental units drawn at random because neighbours share a common microenvironment due to patchy patterns of environmental variability or environmental gradients with the patch size exceeding that of the experimental unit (an individual tree or a plot). Spatial autocorrelation reduces the precision of treatment comparisons by inflating the among-plot variance and deflating the within-plot variance (Besag and Kempton 1986; Magnussen 1989a). It also inflates the estimate of the family variance component, and thus additive genetic variance and individual narrow-sense heritabilities, although family heritability is weakly affected (Magnussen 1993a). Competition among neighbouring trees or plots leads to negatively correlated errors, which inflates the power of treatment comparisons by reducing the among-plot variance and increasing the within-plot variance (Magnussen 1989a, 1993b) and results in biased estimates of genetic parameters (Magnussen 1989b, 1993b, 1994). In forest genetic tests, spatial autocorrelation is prevalent until competition for resources commences, beyond which the two effects are inseparable and only their net effect can be quantified. Given the often patchy heterogeneous site conditions, the large number of genetic entries, and the long duration of forest genetic tests, spatially correlated observations are the norm rather than the exceptions (Tuskan and van Buijtenen 1986; Magnussen 1990, 1994; Loo-Dinkins 1992).

Three approaches appear to reduce the effects of spatial autocorrelation. First, one could choose experimental designs that reduce or eliminate the effects of the environmental covariance caused by spatial autocorrelation. Contiguous multiple-tree plot designs are affected most by the positive environmental covariance due to spatial auto-correlation (Loo-Dinkins 1992; Magnussen 1993a). Designs using single-tree plots or noncontiguous mutiple-tree plots allow unbiased comparisons among genetic entries and unbiased estimation of variance components and individual narrow-sense heritabilities before the onset of competition (Libby and Cockerham 1980; Loo-Dinkins 1992; Magnussen 1993a). These

designs, however, are the most prone to the effects of negative covariance caused by competition (Magnussen 1989b). Neighbour-balanced designs have been proposed by certain authors to correct for the effect of spatial autocorrelation (e.g., Martin 1986; Kunert 1987; Wild and Williams 1987). The bias from competition could be reduced by competition-balanced designs that could accommodate only a limited number of genetic entries (Federer and Basford 1991). These designs are difficult to construct and may work well only under certain correlated structures, whence their limited use.

Second, the effect of spatial autocorrelation could be reduced by knowledge of the environmental variability in the test field. Maps to the plot or individual tree level could then be drawn up, for example, for light availability, soil properties, topography (contours), or surface vegetation. Maps of these environmental variables are helpful for improving our understanding of the spatial processes leading to spatial autocorrelation (Samra *et al.* 1992; Ball *et al.* 1993). These variables can also serve to delineate environmental response surfaces (Trangmar *et al.* 1987), or as covariates in analysis of covariance to reduce the effects of uncontrolled variation (nuisance parameters) (Mulla *et al.* 1990). The lack of complete understanding on how environmental factors affect tree growth hampers the usefulness of the analysis of covariance as a tool to reduce the effect of a heterogeneous environment. Analysis of covariance assumes that the regression of dependent variables on covariates is linear and independent of treatments and blocks. If the linear regression relationship varies among treatments, analysis of covariance cannot be used to increase the precision of treatment comparisons (Snedecor and Cochran 1967). As environmental conditions are difficult and time-consuming to quantify, such data are rarely available and in the best cases, only a very few attributes could be characterized at the plot level.

As the use of classical statistical analysis in field trials with significant spatial autocorrelation leads to less precision in treatment comparisons and biased estimates of variance components, many spatial models have been formulated to alleviate these problems. Most models split the spatially correlated error term into a trend or a residual where the trend is a model-based description of the spatial relationships of the observations (Kirk *et al.* 1980; Besag and Kempton 1986; Binns 1987; Gleeson and Cullis 1987; Cullis and Gleeson 1991; Zimmerman and Harville

1991). They were originally proposed for analysis at the plot level and were subsequently extended to individual trees in forest genetic tests (Magnussen 1993a, 1994). Model choice is a major problem as the patterns of microsite variability are largely unknown and often derived from the residuals of a data analysis model, thus it depends on the specific position of genotypes at the test site. The trend component based on information from neighbours has been shown to be adequate in many circumstances (Correll and Anderson 1983; Besag and Kempton 1986; Magnussen 1990, 1993a; Federer and Basford 1991). The use of these spatial models does not eliminate the spatial covariances, and does not necessarily lead to unbiased estimates of treatment means and variance components because the extent to which true genetic effects of families may be adjusted out is not known. This results from the inseparability of the within-plot genetic and environmental effects. Despite this, the use of spatial models could increase the precision of treatment comparisons (Williams 1986; Cullis and Gleeson 1989; Magnussen 1990; Kempton *et al.* 1994), and could potentially reduce the bias in estimating variance components (Magnussen 1993a, 1994).

Eastern white pine (*Pinus strobus* L.) grows naturally in southeastern Canada and the northeastern United States, and is a high-value timber species (Wendel and Smith 1990). The reforestation effort for this species has encountered two major obstacles: white pine blister rust (*Cronartium ribicola* J.C. Fisch.) and white pine weevil (*Pissodes strobi* [Peck]). To decrease the risk of blister rust, white pine is planted in areas with low rust susceptibility in Quebec (Lavallée 1986). Damage from the white pine weevil is mainly controlled by silvicultural methods such as close spacing (Alfaro and Omule 1990), planting white pine under hardwood cover and between hardwood bands, or in association with hardwood species (Stiell and Berry 1985; USDA 1985; Alfaro *et al.* 1995). In accordance with these silvicultural treatments, genetic tests of white pine in Quebec were established within clear-up strips between hardwood bands or planted in association with European speckled alder (*Alnus incana* Moench) in order to obtain realistic estimates of genetic parameters in a plantation setting.

White pine genetic tests established between hardwood bands may have greater environmental heterogeneity as hardwoods in the uncut stand compete with white pine trees for light and nutrients and may create microenvironmental patches that may not follow boundaries of the patches caused by soil heterogeneity. The spatial autocorrelation in such trials may be weaker than in genetic tests established in an open field. In this study, our objectives were: 1) to determine the extent of spatial autocorrelation in two provenance/progeny tests of eastern white pine, 2) to increase the precision of comparisons between families or seed sources by using relative light intensity and drainage classes as covariates in analysis of covariance, 3) to test the efficiency of two spatial models at the plot level for improving the precision of comparisons among families and seed sources, and 4) to propose a procedure for estimating individual narrow-sense heritability based on spatial and genetic relationships of surviving trees.

MATERIALS AND METHODS

Experimental materials

A range-wide sample of 133 seed sources of eastern white pine was used in this study. Of these 133 seed sources, 62 were bulked seed collections and for the remaining 71 seed sources, identities of families were retained. Each of these 71 provenances was composed of one to ten open-pollinated families, with a total of 285 families.

Seeds were sown in March 1984 in a greenhouse, and 1-year-old seedlings were transplanted into a nursery. Two genetic tests were established with 4-year-old seedlings. Seedlings were planted as two rows within each 3-m clear-cut strip between two adjacent hardwood bands, with each row 0.5 m away from the hardwood band. The two seedling rows were separated by 2 m, and seedlings within a row by 2 m. The clear-cut strips run north-south to provide maximum light for tree growth. At one site, Grand-Mère (lat. 46°36', long. 72°39', elev. 110 m), main hardwood species in the 5-m-wide uncut hardwood band are trembling aspen (*Populus tremuloides* Michx.), largetooth aspen (*P. grandidentata* Michx.) and red maple (*Acer rubrum* L.) with heights between 15 and 20 m and an age of about 50 years. At the other site, Notre-Dame-du-Rosaire (lat. 46°00', long. 75°33', elev. 210 m), the 4-m-wide hardwood band is composed of white birch (*Betula papyrifera* Marsh.), trembling aspen and mountain maple (*A. spicatum* Lamb.) with heights between 9 and 15 m and an age of about 30 years. A randomized complete block design was used, with seven blocks at the Grand-Mère site and six blocks at the Notre-Dame-du-Rosaire one. Each site was split into two sections (north and south), with the Grand-Mère site having four blocks in the northern section and three in the southern one and the Notre-Dame-du-Rosaire site split into a northern and a southern

section, each with three blocks. Within blocks, each family was randomly allocated to a 4-tree row plot (family plot), while each bulked seedlot was replicated twice in randomly arranged 4-tree row plots.

Microsite characterization

Microsites in the clear-cut strip (down to the plot level) were characterized for light, drainage, soil, and indicator species in order to better understand spatial patterns of environmental variation. The intent was to test the potential of these environmental variables to adjust out some environmental variance.

Relative light intensity was determined for each plot in six of the seven blocks at the Grand-Mère site in the summer of 1994. Photosynthetic photon flux density (PPFD) was measured simultaneously by two point quantum sensors (LI-COR Inc., Lincoln, Nebraska): one at a height as close as possible to the terminal shoots of white pine trees within a plot (Q_0), and the other at a point free of shade above the canopy of the hardwood stand (Q_i). The relative light intensity was calculated as (Q_0/Q_i) x 100 (Parent and Messier 1995, 1996). Light measurements were taken on completely overcast days as the relative light intensity measured under these conditions is nearly constant during the day, and more importantly, is strongly correlated with the mean relative light intensity for both overcast and clear sky conditions (Messier and Puttonen 1995: Parent and Messier 1996).

Drainage conditions were mapped at the Grand-Mère site. Three drainage classes were defined: rapid, good and imperfect. The objective here was to characterize the level of water availability, and to assess its effect on tree height.

The soil was surveyed in 1990. Soils were sampled at 92 and 90 points at the Grand-Mère and Notre-Dame-du-Rosaire sites respectively to determine soil types. A subsample of size 7 to 12 was used for detailed analyses of physical and chemical properties (Lamontagne 1990a, 1990b). At the Grand-Mère site, soil was Gleyified Humo-Ferric Podzol, classified as Ivry series, with five variants found. At the Notre-Dame-du-Rosaire site, soils belonged to three series: two were Humo-Ferric Podzol and the third was Orthic Humo-Ferric Podzol. Given that the soil analysis was based on about 10 samples, it was not possible to map physical and chemical properties of the soil to the plot level and consequently no attempts were made to use soil data as covariates in further analyses. However, soils in the test sites were relatively homogeneous.

In the spring of 1993, an ecological survey was conducted in the clear-cut strip and uncut hardwood bands at the Grand-Mère site to identify indicator species (herbs or shrubs) to characterize microsites. A random sample of 50 quadrats (1 m x 2 m) was surveyed, with 25 in each of the two sections of this site. This survey, however, failed to find indicator species associated with soil types, drainage and light conditions. Thus, results from this ecological survey will not be discussed further in this report.

Height measurements

At the Grand-Mère site, height was measured in 1990, 1991, 1992 and 1993 when trees were 7 to 10 years old from seed. At the Notre-Dame-du-Rosaire site, height measurements were taken in 1992, 1993 and 1994 when trees were 9 to 11 years old from seed.

Statistical analyses

Statistical analyses were conducted using SAS Interactive Matrix Language, SAS Macro processing, SAS GLM and SAS VARCOMP procedures. Plot means were used except for estimating heritabilities where heights of individual trees were used. Unless otherwise stated, bulked seedlots were excluded from the analyses.

Spatial autocorrelation analysis

Spatial autocorrelation coefficient (ρ) among first-order neighbouring plots was calculated to examine the extent of sharing of microsite environments in these two tests (Cliff and Ord 1981). It was calculated for each section at each site from residuals resulting from analysis of variance with a model specifying seed source and families within seed source as fixed effects. The model used was:

$$Y_{ij} = \mu + P_i + F_{ij} + e_{ij} \quad [1]$$

where Y_{ij} is the plot mean height for the jth family in the ith seed source, μ is the grand mean, P_i is the ith seed source effect, F_{ij} is the jth family within the ith seed source effect, and e_{ij} is the residual. The bulked seedlots were treated here as though they contained only one family. Due to the presence of hardwood bands and to the layout of the plantation, the spatial autocorrelation coefficients were computed only for neighbouring plots between the two adjacent hardwood bands.

Effect of relative light intensity and drainage on 10-year height

As relative light intensity and drainage classes were determined for each plot, we used them separately as covariates in the analysis of covariance. We compared the error mean square obtained from this analysis with that from analysis of variance to assess the reduction in error mean square due to the use of covariates. If the effect due to the covariate was significant, we tested for interactions between the covariate and the seed sources and/or families within seed sources. In these analyses, bulked seedlots were excluded. The model of the analysis of variance was:

$$Y_{ijk} = \mu + B_i + P_j + F_{jk} + e_{ijk} \qquad [2]$$

where Y_{ijk} is the plot mean 10-year height for the kth family (F) from the jth provenance (P) in the ith block (B) and e_{ijk} is the plot error. The analysis of covariance took the same form as [2] and was conducted in two stages: in the first stage by adding a covariate term as an effect in the model, and in the second stage by adding the covariate effect and the interactions between the covariate and seed sources or families within seed sources.

The mapping of plots into three drainage classes allowed us to use drainage as another blocking factor in the analysis of variance. Given that a subset of families was represented in each drainage class, we used the same set of common families in the analysis of variance. This was done by using the Type IV sum of squares in the SAS GLM procedure. The model used was:

$$Y_{ijkl} = \mu + B_i + D_j + P_k + F_{kl} + e_{ijkl} \qquad [3]$$

where D_j is the fixed effect of drainage, e_{ijkl} the plot error, and the other terms are defined as in [2].

Application of two spatial models

We conducted analysis of variance for the randomized complete-block design as defined in [2] and used the results as a basis for comparisons with two spatial adjustment methods adopted for this study. The two spatial models are Wright's moving plot average method (Wright 1978) and an iterative method (Magnussen 1993a). These two methods were chosen because they have been proven to reduce the effects of spatial autocorrelation in previous studies (Magnussen 1990, 1993a). Wright's method assumes that residuals from analysis of variance provide reliable estimates of microsite variability, and when used as an

adjustment, they may remove the environmental covariances due to the spatial autocorrelation from the error mean square (Wright 1978). Given that our tests are composed of pairs of rows of contiguous plots, we used the average residual of five adjacent neighbours as the adjustment in our analysis, i.e., the adjusted plot mean was obtained by the subtraction of the average residual of the five neighbouring plots from the original plot mean. As Wright's method calculates the adjusted plot mean as a deviation from the perfect regression line expressing microsite variability (Wright 1978), Magnussen (1993a) modified this approach by taking into account the spatial autocorrelation coefficient (ρ). The 5-plot average residual was multiplied by a factor $\rho/(1-\rho)$ before its use as an adjustment. This may be more realistic in expressing the relationship between neighbour plots because the one between microsite variability and residuals is not perfect. This procedure was iterated four times until no changes were observed for the error mean squares. The initial plot residuals for this method were obtained from model [1].

Once adjusted by the above-described methods, the plot means were then used as input for analysis of variance with the following model:

$$Y'_{ij} = \mu + P_i + F_{ij} + e_{ij} \qquad [4]$$

where Y'_{ij} is the adjusted plot mean height for the jth family in the ith seed source and other terms as defined in [1]. In this model, block effects were ignored since boundaries between contiguous blocks after adjusting for spatial effects are not likely to produce any meaningful effect.

The adequacy of the two spatial models was judged by comparing the error mean square obtained from model [4] with that from the conventional analysis (model [2]). The error mean square was used because it describes the precision in estimating seed source and family means. For breeding purposes, it is important to determine whether the same set of families would be selected from the spatially adjusted and original data. Thus, rank correlations for seed sources and families were calculated between ranks estimated from the least-squares method in model [2] and the ranks from model [4] with the adjusted plot means.

Estimating individual heritability using spatial positions

Even a small degree of spatial autocorrelation in genetic tests can lead to a significant overestimation of the family variance component and consequently individual narrow-sense heritability (Magnussen

1993a). Given that it is a norm rather than an exception that spatial autocorrelation exists in genetic tests, estimates of individual heritabilities might be inflated in most cases. One approach to overcome this problem is to use spatial models to adjust for these inflated estimates (Magnussen 1993a, 1994), which is computationally intensive and the choice of the spatial models is difficult. Inflated estimates of heritabilities could be avoided by using the spatial relationship of surviving trees in a genetic test. It is based on the fact that covariances between different kinds of neighbouring trees have different expected covariances in a genetic test with contiguous multiple-tree plots. Covariance between unrelated neighbouring trees from two adjacent plots contains only the environmental covariance component, while covariance between genetically related trees within a plot has two components: environmental covariance and genetic covariance. Genetic covariance represents 1/4 of the additive genetic variance if trees are related as half-sibs, 1/2 of the additive genetic variance and 1/4 of dominant genetic variance for full-sib families (Falconer 1981). The difference between covariance for half-sibs and for unrelated trees then estimates 1/4 of the additive genetic variance for trees in comparable spatial positions. Total phenotypic variance could be obtained by classical analysis of variance since the spatial autocorrelation does not change the total variance, but only affects the allocation of the total variance among blocks, families, plots and within plots (Magnussen 1994).

To estimate additive genetic variance from the spatial positions of surviving trees, we calculated covariances between different kinds of neighbouring trees for 7- to 11-year heights at the two sites: covariances among progenies of open-pollinated families, among trees within bulked seedlots, and among unrelated trees. The covariance for bulked seedlots was estimated and compared with the covariance for open-pollinated families (we assume that open-pollinated progenies within a family are related as half-sibs). As a bulked seedlot contains both half-sibs and unrelated trees, the average genetic relationship among trees within a bulked seedlot is expected to be greater than zero (for unrelated trees) and less than 0.25 (for half-sibs). Thus covariance among trees of a bulked seedlot will be smaller than that among true half-sibs, and larger than that among unrelated trees. Covariance among unrelated trees, which represents the environmental covariance, was estimated in two ways: one for the covariance among unrelated trees within a row and the other for the covariance among unrelated trees across the two rows

within the clear-cut strip. To ensure that a tree was used only once in calculating covariances, we used only trees 2 and 3 in the same 4-tree plot to estimate covariances for open-pollinated families and for bulked seedlots. Tree 4 in a plot and tree 1 in the neighbouring plot within the same row were used to calculate the covariance of unrelated trees within a row. The covariances among progenies of open-pollinated families, among trees within bulked seedlots, and among unrelated trees within or across rows were estimated for each block. The covariances for different kinds of neighbours in each of the two tests were obtained as the weighted average of covariances from seven blocks at the Grand-Mère site and six blocks at the Notre-Dame-du-Rosaire site, respectively, with the weight being the number of pairs of trees for each kind of neighbour in a block.

The genetic covariance for open-pollinated families was estimated as the difference between covariances among open-pollinated progenies and unrelated trees. As separate families were nested within seed sources, the genetic covariance for open-pollinated families contains the seed source variance component. This seed source variance component was estimated (see model [5] below), and was subtracted from the genetic covariance for open-pollinated families. This adjusted genetic covariance for open-pollinated families estimates 1/4 of the additive genetic variance (assuming that open-pollinated progenies are related as half-sibs). The individual narrow-sense heritability for the spatial method was calculated as the ratio of the additive genetic variance to the total phenotypic variance (σ^2_T). The total phenotypic variance was estimated as $\sigma^2_T = \sigma^2_{F(P)} + \sigma^2_E + \sigma^2_W$ from the following model:

$$Y_{ijkl} = \mu + B_i + P_j + F_{jk} + e_{ijk} + w_{ijkl} \quad [5]$$

where Y_{ijkl} is the height for the lth individual of kth family (F) from the jth provenance (P) in the ith block (B), e_{ijk} is the plot error and w_{ijkl} is the within-plot error. In this analysis, we used only those trees included in calculating the covariances for open-pollinated families and for unrelated trees within a row.

We also calculated the individual narrow-sense heritability from the classical method and compared it with the estimated individual narrow-sense heritability from the spatial method. In the classical method, the additive genetic variance was estimated as four times the family variance component ($\sigma^2_{F(P)}$), and the total phenotypic variance was calculated as above. The heritability was computed as $4\sigma^2_{F(P)} / \sigma^2_T$. To ensure that the two heritability estimates are comparable, in the classical analysis we also used the same trees

included in estimating the heritability from the spatial method.

RESULTS AND DISCUSSION

Survival was 93.5% for 10-year-old trees at the Grand-Mère site and 85.3% for 11-year-old trees at the Notre-Dame-du-Rosaire site. Mortality was randomly distributed and no significant differences in survival rate were detected among seed sources or families. White pine blister rust infected 1.5% and 6.6% of the trees at the Grand-Mère and Notre-Dame-du-Rosaire sites, respectively, with incidence of white pine weevil attack less than 0.6% at both sites. Ten-year height averaged 2.03 m at the Grand-Mère site and the average 11-year height was 1.54 m at the Notre-Dame-du-Rosaire site. Significant differences in heights at both sites were found among seed sources and among families within seed sources (p-value $\leq$ 0.001).

Spatial autocorrelation analysis

The spatial autocorrelation coefficients were comparable in the northern and southern sections at the two sites, and increased with age from 0.15 for 7-year height to 0.19 for 10-year height at the Grand-Mère site, and from 0.09 for 8-year height to 0.12 for 11-year height at the Notre-Dame-du-Rosaire site (Table 1). A similar trend in the increase in spatial autocorrelation with age was also observed in two young jack pine tests: from 0.24 to 0.38 in one test and from 0.25 to 0.30 in the other test for 6- and 8-year height (Magnussen 1990). However, the spatial autocorrelation decreased from 0.39 for 12-year height to 0.13 for 17-year height in an older jack pine test (Magnussen 1994). This initial increase in spatial autocorrelation presumably reflects the gradual response of trees to their shared microsite variability among adjacent plots. The decrease in autocorrelation for older jack pine trees probably shows that the effect of competition has at least partially negated the positive environmental covariance due to shared microsites, but has not yet led to the negatively correlated plot errors.

Table 1. Spatial autocorrelation coefficients calculated from plot residuals.

AGE (YEARS)	NORTHERN SECTION	SOUTHERN SECTION	MEAN
Grand-Mère site			
7	0.152	0.155	0.153
8	0.154	0.155	0.155
9	0.164	0.182	0.172
10	0.189	0.192	0.190
Notre-Dame-du-Rosaire site			
9	0.084	0.103	0.093
10	0.121	0.088	0.105
11	0.120	0.126	0.123

Apparently, spatial autocorrelation varies with tests where patterns of shared microenvironments differ. The spatial autocorrelation was much lower (0.10) for one jack pine site than the two other sites cited above (Magnussen 1990). Our white pine tests have lower spatial autocorrelations than the three jack pine tests (Magnussen 1990, 1994), which could result from the competition for light between hardwoods and white pine saplings as opposed to the mere sharing of microsite environments in the jack pine tests where seedlings were planted in an open field. The weaker spatial autocorrelation observed at the Notre-Dame-du-Rosaire site is likely due to further competition from ferns as each year this vegetation had to be controlled manually by weeding to prevent damage caused by ferns being pressed down by heavy snow.

Effect of relative light intensity and drainage on 10-year height at the Grand-Mère site

In the six blocks where relative light intensity was measured, plots varied greatly in relative light intensity, ranging from 0.02 to 1.00 with a mean relative light intensity of 0.44. Although relative light intensity was a significant covariate (p-value = 0.006), the error mean square for 10-year height (1027.1) was only

0.6% smaller than that from the analysis of variance (1032.8). Thus, the use of relative light intensity did not improve the precision of comparisons of seed sources and families to the extent of practical relevance. Further analyses revealed weak effects of seed source-by-light and family-by-light interactions (p-value > 0.07), suggesting that the relationship between relative light intensity and 10-year height is similar among seed sources and families within seed sources. The use of drainage class as a covariate also did not improve the precision of the experiment as the error mean square from analysis of variance (1062.8) was the same as that from analysis of covariance (1062.5). In addition, in the analysis of variance with drainage as another blocking factor, drainage classes did not differ significantly in 10-year height (p-value = 0.358). Thus, we can conclude that the use of relative light intensity and drainage classes as covariates did not improve the precision of comparisons among families or seed sources for 10-year height at the Grand-Mère site.

Several reasons could account for the failure of relative light intensity and drainage as covariates. Drainage was classified into three subjective classes: rapid, good and imperfect, and thus only represents an approximation of water availability at the plot level. Relative light intensity was measured only at one spot within a plot (1 m x 8 m) due to the large number of measurements required, and thus may not well represent the average relative light intensity at the plot level, given that light conditions within a plot could vary greatly depending on the distribution of hardwood trees in the adjacent uncut strip of hardwoods. Another reason why relative light intensity did not correlate well with height growth of trees is that eastern white pine does not respond to light intensity in a linear fashion, as it is a moderately shade-tolerant species (Logan 1966; Wendel and Smith 1990).

In agricultural trials, covariates such as soil ion concentrations and pH are shown to be effective in reducing the error mean squares (e.g., Mulla et al. 1990). This probably reflects the tendency that agricultural plots are more homogenous than forest plots given that greater control could be exerted over environmental conditions in agricultural trials than in forestry ones. As tree plots are larger and more heterogenous, one must characterize a plot at several spots. This can be very time-consuming. Thus, the only feasible way to increase the precision of comparisons among treatments in forest trials may be the use of spatial models in the analytical stage if the experiments have already been installed.

Application of two spatial models

At the Grand-Mère site, the two spatial models tested were more effective for the southern section than for the northern one although both had comparable spatial autocorrelation coefficients (Table 2). In the southern section, the error mean square (MSE) for 10-year height was reduced by 11% and 17% for Magnussen's and Wright's methods, respectively, while the reductions for the northern section were 4% and 5%. This difference may result from greater heterogeneity in soils in the southern section than in the northern one (Lamontagne 1990a). Interestingly, analysis of variance showed that the block effect was significant in the northern section (p-value < 0.0001), but not in the southern one (p-value = 0.452). This suggests that the spatial adjustment procedure works well in tests where blocking is ineffective (Magnussen 1990).

At the Grand-Mère site, the efficiency of the spatial models increased with the age of the trees: the reduction in MSE in the combined analysis increased from 1.5% for 7-year height to 8% for 10-year height for Magnussen's method, and from 1% to 11% for Wright's method (Table 2). At the Notre-Dame-du-Rosaire site, the use of the two spatial models resulted in little reduction in MSE (with a maximum reduction of 1.5% for 11-year height), and Wright's method actually increased the MSE on two occasions (Table 3). When comparing the results from spatially adjusted data with those from original data, the statistical significance of seed source effects increased slightly, while the statistical significance of family effects decreased slightly (results not shown). The small reduction in MSE for younger trees at the Grand-Mère site and for all ages at the Notre-Dame-du-Rosaire site may be related to the small spatial autocorrelation coefficients (p-value < 0.15). Spatial models seem to increase the precision of treatment comparisons only when spatial autocorrelation is greater than 0.15 (Magnussen 1990; Loo-Dinkins 1992).

Although the spatial models reduced the MSE noticeably for 10-year height at the Grand-Mère site, the ranking of families and seed sources changed little for the spatially adjusted and unadjusted data (Table 4). We did not calculate the rank correlation between spatially adjusted and unadjusted means for Notre-Dame-du-Rosaire as the two spatial models tested did not improve the precision of comparisons among seed sources and families (Table 3). At the Grand-Mère site, the least-squares family means from

the block analysis were very strongly correlated with those from Wright's analysis (correlation coefficient = 0.96 or 0.97 for 7- to 10-year heights) and with those from Magnussen's analysis (correlation coefficient between 0.99 and 1.00). Our results differed from those in two jack pine tests where spatially adjusted and unadjusted family means were strongly correlated, but with a decrease in the correlation coefficients with age (from 6 to 8 years) (Magnussen 1990). This difference may be due to a greater spatial autocorrelation in these jack pine tests than in our white pine tests. Thus, the use of the two spatial models led to very similar rankings of families and seed sources with the conventional analysis, and did not result in the selection of different families or seed sources for breeding programs for the two white pine tests. However, if the goal is to predict genetic gains, the use of spatial models is worthwhile because spatial autocorrelation causes inflation in genetic variances and individual narrow-sense heritabilities, thus inflating predicted genetic gains from mass selection (Magnussen 1993a) and from family selection as well (Magnussen 1993b).

Table 2. Comparisons of error mean squares for analysis of variance from block (model [2]) and two spatial models (model [4]) at the Grand-Mère site.

ANALYSIS[a]	NORTHERN SECTION	SOUTHERN SECTION	COMBINED
7-year height			
Block	230.31	228.82	265.53
Wright	279.03 (99.5%)[b]	220.65 (96.4%)	262.89 (99.0%)
Magnussen	276.28 (98.6%)	216.89 (95.8%)	261.59 (98.5%)
8-year height			
Block	420.87	366.44	406.59
Wright	417.54 (99.2%)	330.88 (90.3%)	389.37 (95.8%)
Magnussen	413.17 (98.2%)	338.27 (92.3 %)	392.10 (96.4%)
9-year height			
Block	730.36	696.68	718.31
Wright	711.28 (97.4%)	646.40 (92.8%)	659.08 (91.8%)
Magnussen	711.39 (97.4%)	622.33 (89.3%)	673.51 (93.8%)
10-year height			
Block	1 045.03	1 094.59	1 061.34
Wright	989.03 (94.6%)	904.85 (82.6%)	945.54 (89.1%)
Magnussen	1 000.08 (95.7%)	969.98 (88.6%)	978.66 (92.2%)

[a] Block analysis was that defined by model [2]. Wright analysis is that defined by model [4] with spatial adjustment from Wright (1978). Magnussen analysis is that defined by model [4] with spatial adjustment from Magnussen (1993a).

[b] Percentage of the error mean square from the block analysis.

Table 3. Comparisons of error mean squares for analysis of variance from block (model [2]) and two spatial models (model [4]) at the Notre-Dame-du-Rosaire site.

ANALYSIS[a]	NORTHERN SECTION	SOUTHERN SECTION	COMBINED
	9-year height		
Block	393.89	452.75	440.01
Wright	384.56 (97.6%)[b]	443.80 (98.0%)	436.26 (99.1%)
Magnussen	401.69 (102.0%)	439.96 (97.2%)	436.18 (99.1%)
	10-year height		
Block	514.60	588.03	558.24
Wright	519.55 (101.0%)	614.80 (104.6%)	572.03 (102.5%)
Magnussen	526.31 (102.3%)	576.16 (98.0%)	555.47 (99.5%)
	11-year height		
Block	684.07	931.08	826.45
Wright	688.55 (100.7%)	935.83 (100.5%)	831.62 (100.6%)
Magnussen	693.12 (101.3%)	904.98 (97.2%)	813.98 (98.5%)

[a] Block analysis was that defined by model [2]. Wright analysis is that defined by model [4] with spatial adjustment from Wright (1978). Magnussen analysis is that defined by model [4] with spatial adjustment from Magnussen (1993a).

[b] Percentage of the error mean square from the block analysis.

Table 4. Spearman rank correlation for provenance and family means between least-squares means from block analysis and the two spatial models at the Grand-Mère site.

PROVENANCE		FAMILY	
Wright	Magnussen	Wright	Magnussen
	7-year height		
0.964	0.996	0.967	0.996
	8-year height		
0.960	0.993	0.967	0.995
	9-year height		
0.949	0.993	0.961	0.994
	10-year height		
0.952	0.992	0.958	0.993

Estimating individual narrow-sense heritability using spatial positions

Covariances and individual narrow-sense heritabilities

As expected, the covariance among trees from open-pollinated families was greater than that among trees of bulked seedlots, which in turn was greater than the covariance of unrelated trees at both sites (Table 5). At the Grand-Mère site, the within-row covariance among unrelated trees did not differ much from the across-row covariance among unrelated trees. At the Notre-Dame-du-Rosaire site, the covariance among unrelated trees across rows was much greater than that within rows (Table 5), suggesting that trees across rows share more common microsites than trees within rows. Given that at both sites two rows of eastern white pine were planted

within the north-south clear-cut strip between the two hardwood bands, it is difficult to understand why the within- and across-row covariances showed this different trend. It could be explained by an environmental gradient perpendicular to the cut strip within which trees were planted at the Notre-Dame-du-Rosaire site. This gradient could result from the layout of the clear-cut strips perpendicular to the different soil series revealed by the soil survey, and from the drainage conditions (Lamontagne 1990b). For estimating genetic covariance, we used within-row covariance at the Grand-Mère site and across-row covariance at the Notre-Dame-du-Rosaire site as the environmental covariance.

The classical estimates of heritabilities varied little among different years at both sites (Table 6). At the Grand-Mère site, the heritability estimates from the spatial method were greater than the classical estimates, and the estimated heritability was smaller for 10-year height than for 7- to 9-year height (Table 6). As two blocks had very small covariances among unrelated trees within a row compared with five other blocks (results not shown), we excluded these two blocks from both spatial and classical analyses. The heritabilities for these five blocks estimated from the spatial method decreased from 0.28 for 7-year height to 0.18 for 10-year height, with the heritability for 10-year height being smaller than that from the classical method (Table 6). At the Notre-Dame-du-Rosaire site, the heritabilities estimated from the spatial method decreased from 0.31 for 8-year height to 0.14 for 11-year height, with the heritability for 11-year height being smaller than that from the classical method (Table 6).

Table 5. Estimates of covariances among different kinds of neighbouring trees.

			UNRELATED TREES	
Trait	Half-sibs	Bulked seeds	Within a row	Across rows
Grand-Mère site				
7-yr height	227.1	198.8 (0.19)[a]	98.7	76.7
8-yr height	367.5	322.2 (0.22)	170.9	135.9
9-yr height	661.9	546.4 (0.20)	313.9	261.7
10-yr height	1 019.5	769.5 (0.10)	563.9	419.3
No. of pairs	1 562	635	1 287	3 680
Notre-Dame-du-Rosaire site				
9-yr height	237.4	229.6 (0.23)	25.0	108.3
10-yr height	310.0	300.0 (0.23)	7.4	167.4
11-yr height	462.7	435.0 (0.23)	-26.7	263.4
No. of pairs	2 839	697	571	1 908

[a] The numbers in parentheses are apparent average genetic relationships among trees of bulked seedlots, estimated by multiplying 0.25 with the ratio of the genetic variance for trees of bulked seedlots to the genetic variance for open-pollinated families (assuming that open-pollinated progenies are related as half-sibs with a genetic relationship of 0.25), with the genetic variance calculated by subtracting the environmental variance (within-row covariance at Grand-Mère and across-row covariance at Notre-Dame-du-Rosaire) from the covariance for open-pollinated families and that for bulked seedlots, respectively.

Table 6. Estimates of individual narrow-sense heritabilities by classical and spatial methods.

TRAIT	CLASSICAL METHOD	SPATIAL METHOD
Grand-Mère site		
7-yr height	0.265[a] (0.206)[b]	0.387[a] (0.280)[b]
8-yr height	0.275 (0.218)	0.387 (0.279)
9-yr height	0.259 (0.196)	0.411 (0.238)
10-yr height	0.260 (0.191)	0.312 (0.177)
Notre-Dame-du-Rosaire site		
9-yr height	0.146	0.308
10-yr height	0.132	0.153
11-yr height	0.168	0.139

[a] Heritability estimate based on data from all seven blocks.
[b] Heritability estimate based on data from five blocks excluding blocks 1 and 7.

The lower estimates from the new spatial method for 10- or 11-year height were expected because the additive genetic variance estimated by the new method may not be inflated due to spatial autocorrelation (Magnussen 1993a). However, the larger estimates from the new spatial method when trees were less than 10 years old were unexpected. This could be due to the inseparability of the common nursery environmental covariance from the estimate of the genetic covariance due to field planting method. The trees within a 4-tree row plot in the field shared a common nursery microsite given that the 4-seedling plot in the nursery was planted as a plot in the field. The covariance among unrelated trees from different plots within or across rows did not include this common nursery environmental covariance because the rerandomization of the nursery plots in field planting made it extremely unlikely that two neighbouring plots in the nursery would also be neighbours in the field. Thus, this common nursery environmental covariance was included in the estimate of the genetic covariance for the spatial method. The contribution of the nursery environmental covariance to the estimate of genetic covariance is expected to diminish with tree age because as trees age, genetic variance for height increases (Balocchi *et al.* 1993 and references therein).

This new spatial procedure to estimate additive genetic variance has several advantages over the classical approach. First, it would give putative unbiased estimates of individual narrow-sense heritabilities, as the effects of spatial autocorrelation are reduced. Second, it would allow the estimation of heritabilities for single-block experiments with contiguous plots. Third, the spatial procedure makes no assumptions regarding the independence of errors and the normal distribution of residuals. Fourth, the new spatial procedure would likely give unbiased estimates for additive genetic variance and heritability even in the presence of strong competition as long as the effect of competition among trees from the same family is similar to that among trees from different families. However, the spatial method also has its limitations and must be studied further. First, the new proposed spatial procedure could not be used in genetic tests where single-tree plots were used, since in the single-tree plot design few relatives will be neighbours. For the single-tree plot design, the classical estimates are unbiased and most efficient for estimating heritabilities before the onset of competition among trees (Magnussen 1993a). Second, a procedure such as bootstrapping must be developed to estimate standard deviation of the spatial estimates of additive genetic variance or heritability. Third, the heritability estimated from the spatial method may be biased upward because of the potential common nursery environmental covariance if trees in a plot in the nursery are planted as a contiguous plot in the field tests. More studies are needed to compare heritability estimates from the spatial and classical methods. Fourth, the statistical properties of the spatial estimators for the additive genetic variance and heritabilities must be examined. A simulation study such as Magnussen's (1993a) would likely give some answers.

Estimating genetic relatedness for bulked seedlots

The covariance among trees from bulked seedlots was smaller than the covariance among trees from open-pollinated families, as expected. The genetic covariance for bulked seedlots was estimated by subtracting the environmental covariance from the covariances among trees from bulked seedlots, and it was smaller than that among open-pollinated families (results not shown). Assuming that open-pollinated progenies are related as half-sibs (genetic relationship $\phi = 0.25$), we estimated the apparent average genetic relatedness among trees within bulked seedlots by multiplying the genetic relatedness among half-sibs ($\phi = 0.25$) by the ratio of the estimated genetic covariance among trees of bulked seedlots to the estimated genetic covariance for open-pollinated families (Table 5). At the Grand-Mère site, the apparent genetic relatedness decreased from 0.22 for 8-year height to 0.10 for 10-year height, while at the Notre-Dame-du-Rosaire site it was stable (0.23) for 8- to 10-year height. The changes with age in the genetic relatedness at the Grand-Mère site is perplexing as it should be the same given that the same trees were used for calculating the covariances. This lack of stability could be related to inaccurate estimates of environmental variances for heights of different ages. The genetic relatedness among trees for the bulked seedlots at the Notre-Dame-du-Rosaire site ($\phi = 0.23$) approaches that for half-sibs, which seems unlikely as each bulked seedlot was composed of seeds from a few to many mother trees. This could result from an upward bias in estimating the genetic covariance among trees for bulked seedlots from the spatial method. Or it could be due to the unequal representation of seeds from different mother trees, to differential germination of seeds from different mother trees, or to differential survival of trees from different mother trees in the field.

Thus, the spatial method could be used to estimate the apparent genetic relatedness of trees within bulked seedlots if a test includes trees from both bulked seedlots and open-pollinated families. Further studies must be conducted to evaluate the value of such an estimate and the conditions required to obtain a good estimate.

CONCLUSIONS

This and previous studies (Magnussen 1990, 1994) showed that spatial autocorrelation varies across test sites. The use of relative light intensity and drainage classes as covariates did not increase the precision of comparisons among seed sources and families. This illustrates the fact that indexing test environments would be nearly impossible with limited resources. Instead, the use of two spatial models increased precision of seed source and family comparisons by reducing error mean squares when spatial autocorrelation was greater than 0.15. Yet, such models would rarely change families to be selected because unadjusted family means were very highly correlated with adjusted ones.

Given that classical estimates of individual narrow-sense heritabilities are potentially upwardly biased (Magnussen 1993a), a spatial method to estimate heritability was proposed. This procedure models the covariances between trees as a function of the spatial and genetic relationship. The method is free of assumptions otherwise made in the classical analysis of variance and could therefore be used in unreplicated experiments. The application of this new procedure in two genetic tests gave acceptable heritability estimates, and thus presumably alleviated the problem of inflated estimation of the additive genetic variance in the classical method. However, the statistical properties of the estimates from the new spatial procedure should be examined before this new procedure is used routinely.

ACKNOWLEDGEMENTS

REFERENCES

Alfaro, R.I.; Omule, S.A.Y. 1990. The effect of spacing on Sitka spruce weevil damage to Sitka spruce. Can. J. For. Res. 20: 179-184.

Alfaro, R.I.; Borden, J.H.; Fraser, R.G.; Yanchuk, A. 1995. The white pine weevil in British Columbia: basis for an integrated pest management system. For. Chron. 71: 66-73.

Ball, S.T.; Mulla, D.J.; Konzak, C.F. 1993. Spatial heterogeneity affects variety trial interpretation. Crop Sci. 33: 931-935.

Balocchi, C.E.; Bridgwater, F.E.; Zobel, B.J.; Jahromi, S. 1993. Age trends in genetic parameters for tree height in a nonselected population of loblolly pine. For. Sci. 39: 231-251.

Besag, J.; Kempton, R. 1986. Statistical analysis of field experiments using neighbouring plots. Biometrics 42: 231-251.

Binns, M.R. 1987. Practical use of neighbor methods and alternatives in the analysis of field trials. Can. J. Plant Sci. 67: 477-489.

Cliff, A.D.; Ord, J.K. 1981. Spatial processes. Pion, London, England.

Correll, R.L.; Anderson, R.B. 1983. Removal of intervarietal competition effects in forestry varietal trials. Silvae Genet. 32: 162-165.

Cullis, B.R.; Gleeson, A.C. 1989. Efficiency of neighbour analysis for replicated variety trials in Australia. J. Agric. Sci., Cambridge 113: 233-239.

Cullis, B.R.; Gleeson, A.C. 1991. Spatial analysis of field experiments - an extension to two dimensions. Biometrics 47: 1449-1460.

Falconer, D.S. 1981. Introduction to quantitative genetics. 2nd ed. Longman Inc., New York.

Federer, W.T.; Basford, K.E. 1991. Competing effects designs and models for two-dimensional field arrangements. Biometrics 47: 1461-1472.

Gleeson, A.C., Cullis, B.R. 1987. Residual maximum likelihood (REML) estimation of a neighbour model for field experiments. Biometrics 43: 277-288.

Kempton, R.A., Seraphin, J.C.; Sword, A.M. 1994. Statistical analysis of two dimensional variation in variety yield trials. J. Agric. Sci., Cambridge 122: 335-342.

Kirk, H.J.; Hayes, F.L.; Monroe, R.J. 1980. Application of trend analysis to horticultural field trials. J. Am. Soc. Hortic. Sci. 105: 183-193.

Kunert, J. 1987. Neighbour balanced block designs for correlated errors. Biometrika 74: 717-724.

Lamontagne, L. 1990a. Étude pédologique d'une plantation de pins blancs (*Pinus strobus* L.) dans la région de Grand-Mère. Équipe pédologique du Québec, Agriculture Canada, C.R.T. Ste-Foy.

Lamontagne, L. 1990b. Étude pédologique d'une plantation de pins blancs (*Pinus strobus* L.) dans la région de Notre-Dame-du-Rosaire. Équipe pédologique du Québec, Agriculture Canada, C.R.T. Ste-Foy.

Lavallée, A. 1986. Zones de vulnérabilité du pin blanc à la rouille vésiculeuse au Québec. For. Chron. 62: 24-28.

Libby, W.J., Cockerham. C.C. 1980. Random non-contiguous plots in interlocking field layouts. Silvae Genet. 29: 183-190.

Logan, K.T. 1966. Growth of tree seedlings as affected by light intensity. II. Real pine, white pine, jack pine and eastern larch. Canada Dept. of Forestry. Publ. no. 1160.

Loo-Dinkins, J. 1992. Field Test Design. *Edited by* L. Fins, S. T. Friedman and J. V. Brotschol. Kluwer Academic Publishers, The Netherlands. pp. 96-137.

Magnussen, S. 1989a. Inter-plant interactions and their influence on within and among plot variances. Scand. J. For. Res. 4: 369-377.

Magnussen, S. 1989b. Effects and adjustments of competition bias in progeny trials with single-tree plots. For. Sci. 35: 532-547.

Magnussen, S. 1990. Application and comparison of spatial models in analyzing tree-genetics field trials. Can. J. For. Res. 20: 536-546.

Magnussen, S. 1993a. Bias in genetic variance estimates due to spatial autocorrelation. Theor. Appl. Genet. 86: 349-355.

Magnussen, S. 1993b. Design and analysis of tree genetic trials. Can. J. For. Res. 23: 1144-1149.

Magnussen, S. 1994. A method to adjust simultaneously for spatial microsite and competition effects. Can. J. For. Res. 24: 985-995.

Martin, R.J. 1986. On the design of experiments under spatial correlation. Biometrika 73: 247-277.

Messier, C.; Puttonen, P. 1995. Spatial and temporal variation in the light environment of developing Scots pine stands: the basis for a quick and efficient method of characterizing light. Can. J. For. Res. 25: 343-354.

Mulla, D.J.; Bhatti, A.U.; Kunkel, R. 1990. Methods for removing spatial variability from field research trials. Adv. Soil Sci. 13: 201-213.

Parent, S.; Messier, C. 1995. Effets d'un gradient de lumière sur la croissance en hauteur et la morphologie de la cime du sapin baumier régénéré naturellement. Can. J. For. Res. 25: 878-885.

Parent, S.; Messier, C. 1996. A simple and efficient method to estimate microsite light availability under a forest canopy. Can. J. For. Res. 26: 151-154.

Samra, J.S., Rajput, R.K.; Katyal, V. 1992. Structured heterogeneity of soil pH and grain yield of rice and wheat grown in a sodic soil. Agron. J. 84: 877-881.

Searle, S.R.; Casella, G.; McCulloch, C.E. 1992. Variance components. John Wiley & Sons, Inc., New York.

Snedecor, G.W.; Cochran, W.G. 1967. Statistical Methods. 6th edition. The Iowa State University Press, Ames, Iowa.

Stiell, W.M.; Berry, A.B. 1985. Limiting white pine weevil attacks by side shade. For. Chron. 61: 5-9.

Trangmar, B.B., Yost, R.S.; Wade, M.K.; Uehara, G.; Sudjadi, M. 1987. Spatial variation of soil properties and rice yield on recently cleared land. Soil Sci. Soc. Am. J. 51: 668-674.

Tuskan, G.A.; van Buijtenen, J.P. 1986. Inherent differences in family response to inter-family competition in loblolly pine. Silvae Genet. 35: 112-118.

USDA For. Serv. 1985. Insects of eastern forests. USDA For. Serv., Misc. Publ. No. 1426.

Wendel, G.W.; Smith, H.C. 1990. *Pinus strobus* L., Eastern White Pine. *In* Silvics of North America. Vol. 1, Conifers. *Technical Coordinators*: R. M. Burns and B. H. Honkala. Agriculture Handbook 654. For. Serv., USDA, Washington, DC. pp. 476-488

Wild, P.R., Williams, E.R. 1987. The construction of neighbour designs. Biometrika 74: 871-876.

Williams, E.R. 1986. A neighbour model for field experiments. Biometrika 73: 279-287.

Wright, J.W. 1978. An analysis method to improve statistical efficiency of a randomized complete block design. Silvae Genet. 27: 12-14.

Zimmerman, D.L.; Harville, D.A. 1991. A random field approach to the analysis of field-plot experiments and other spatial experiments. Biometrics 47: 223-239.