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Effectiveness of spatial analysis in *Cryptomeria japonica* D. Don (sugi) forward selection revealed by validation using progeny and clonal tests

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Abstract

· *Key message* **Accurate evaluation of genetic performances of trees is crucial in order to improve the efficiency of forest tree breeding. We revealed that spatial analysis is effective for predicting individual tree breeding values at the forward selection stage of *Cryptomeria japonica* D. Don (sugi) breeding program by using a novel validation approach.**

· *Context* In the process of selecting genetically superior trees for breeding, appropriate handling of environmental effects is important in order to precisely evaluate candidate trees. Spatial analysis has been an effective statistical approach for genetic evaluation at sites with heterogeneous microenvironments. However, the efficiency of spatial analysis on forward selection has not been validated on a practical scale to date.

· *Aims* This study aimed to reveal the effectiveness of spatial analysis, which incorporates spatially autocorrelated residuals into mixed models, for the prediction of breeding values at the forward selection stage by validation using progeny or clonal tests of forward-selected individuals.

· *Methods* Tree height was analyzed by ordinary randomized complete block design models and spatial models incorporating spatially autocorrelated residuals in a linear mixed model framework, and model selection was conducted at thirty *Cryptomeria japonica* D. Don breeding population sites having various topographical ruggedness. For validation, three clonal tests and one progeny test of individuals selected from three and four breeding populations, respectively, were used. The effectiveness of forward selection using the two models was evaluated based on the correlation between individual breeding values at the stage of forward selection and genotypic and breeding values that were estimated by clonal and progeny tests.

· *Results* Spatial models were more predictive than ordinary models in all cases. Spatial correlation parameters tend to increase with the topographical ruggedness index of each site. The correlation coefficients between breeding values at the time of forward selection and genotypic or breeding values evaluated in succeeding clonal and progeny tests were significantly higher in spatial models than in ordinary models in six out of nine cases.

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• **Conclusion** Validation using progeny and clonal tests of forward-selected individual trees revealed that spatial analysis is more effective for the evaluation of genetic performance of individuals at the stage of forward selection in *Cryptomeria japonica*.

Keywords Individual selection · AR model · Spatially correlated residuals · Clonal test · Progeny test

1 Introduction

Trees in forest stands experience environmental factors that are spatially heterogeneous (White et al. 2007). The growth of forest trees is regulated by genetic potential and the extent that expression of the genetic potential is altered by environmental factors (Pallardy 2008). Forest tree breeding programs involve selecting trees that have higher genetic potential from environmentally heterogeneous forest lands. Therefore, appropriate handling of environmental heterogeneity is essential for the success of the breeding program.

Environmental heterogeneity is present even within a forest site, exerting different effects on individual trees growing there. Such micro-environmental effects are caused by various factors, including soil fertility, soil moisture, water availability, light availability determined by topography or competition with other plants, and others. Genetic trials, which are established in order to estimate genetic parameters and to gain data for predicting genetic performance of breeding materials (Fins et al. 1992), are generally conducted on sites in forest lands with heterogeneous environments and are affected by different micro-environments.

Micro-environmental effects sometimes show random patterns within a site but often exhibit spatially structured patterns (Fu et al. 1999). To handle such micro-environmental effects, multiple field test designs such as randomized complete block design and incomplete block design have been adopted (Fins et al. 1992). However, appropriate blocking is difficult to achieve in some cases due to the lack of information available in advance about spatially structured pattern of micro-environmental factors at the forestry study site (e.g., the extent, range and direction of similarities). As a result, even within a block or replication in a field test design, there is often great environmental variation, causing trees in the same replication to be affected to different extents by environmental effects (Fu et al. 1999, Costa e Silva et al. 2001). Further, it is often difficult for forest tree breeders to design field trials because data available for analysis was collected in field trials conducted several decades ago by their predecessors using methods and knowledge contemporaneous to the study (Fins et al. 1992). These situations produce bias in estimating genetic parameters (Magnussen 1999) and also decreased prediction accuracy of genetic performance of the trees because trees in the same replication or block are assumed to be under the same environmental conditions in ordinary analysis regardless of the environmental heterogeneity (White and Hodge 1989). Consequently, failing to take spatially structured patterns of the microenvironment into account would reduce the efficiency of forest tree breeding programs.

To handle spatial patterns in micro-environments, a posteriori data adjustment methods that utilize spatial information have been considered (Fins et al. 1992). White et al. (2007) reviewed several statistical methods that utilize spatial information. Among these methods, incorporating spatial autocorrelation in residual covariance matrices in the mixed model framework have recently become popular (Costa e Silva et al. 2001, Dutkowski et al. 2006; Ye and Jayawickrama 2008). The mixed model framework is typically used for evaluation of tree breeding materials and, therefore, spatial analysis is easy to incorporate into the tree breeding framework.

In previous reports, spatial analysis has been applied to progeny trials or clonal trials, and the effect of spatial analysis by comparison of models using statistics obtained from the data collected in one generation was reported (Costa e Silva et al. 2001; Dutkowski et al. 2006; Ye and Jayawickrama 2008). There are two main objectives for estimating the genetic value of materials in tree breeding (White et al. 2007): backward selection and forward selection. In backward selection, the data from progenies or clones are analyzed in order to evaluate the parents or ortets. For this selection, previous reports have already revealed high efficiency of spatial analysis. For forward selection, data of the individual itself and its relatives, including siblings and ancestors, are analyzed in order to evaluate the focal individual. However, there are no direct progeny or clones of forward-selected individuals at the time of forward selection; therefore, it is difficult to practically evaluate the accuracy of the genetic evaluation at the time of forward selection, or in other words, using data collected in one generation. Practical validation of the efficiency of spatial analysis on forward selection requires the data of succeeding generation; i.e., progeny or clonal trials in which the next generation of trees from forward-selected individuals are planted. Such validation of the efficiency of spatial analysis on forward selection is definitely important but has not yet been examined.

The aim of this paper was to demonstrate the effectiveness of spatial analysis in forward selection for *Cryptomeria japonica* in Japan. *C. japonica* is the most planted forestry species in Japan, representing approximately 30.3% of the reforested area (Forest Tree Breeding Center, Forestry and Forest Products Research Institute 2016). We assume that

spatial analysis is effective in forward selection as well as in backward selection, and to confirm this hypothesis, we compared the results of ordinary randomized complete block design model with spatial analysis model using two approaches, a model selection at the stage of forward selection and validation using the tree height data collected from clonal and progeny tests in *C. japonica*. For model selection approach, we compared the two models at 30 breeding population sites and showed differences in results of individual tree selections between the models. For the validation approach, we compared the individual tree breeding values at the stage of forward selection with genotypic or breeding values by clonal or progeny tests of forward-selected individual trees.

2 Materials and methods

We compared spatial analysis using a spatial autoregressive model (AR model) to that using ordinary analysis with a randomized complete block design model (RCB model) by two approaches: (1) model selection and comparison of statistics based on the dataset available at the time of forward selection (model selection approach) and (2) validation using a progeny trial or clonal trial of forward-selected individuals (validation approach).

2.1 Breeding population sites for forward selection

We analyzed 30 breeding population sites planted for the forward selection of second-generation plus trees of *C. japonica* in the Kyushu breeding region, Japan (Supplementary Table 1). The sites were planted from 1969 to 2007, and the trees at most sites are mainly artificially crossed families of first-generation plus trees, but some sites include open-pollinated families, and the number of families at each site ranges from 19 to 185 (average 49.1) with the number of trees ranging from 990 to 5633 (average 2311.4). These sites were settled based on randomized complete block designs with two to six replications (average 4.83), and multiple trees per family were planted for each replication with duplicated single-tree plots or rectangular multi-tree plots at spacing of 1.8 m.

2.2 Clonal test sites and progeny test site for validation

For the validation approach, clonal test sites and a progeny test for forward-selected individuals were used. Clonal tests were conducted for the individuals forward-selected from three sites (Supplementary Table 1). The original three breeding population sites were established from 1969 to 1970 and had 66 to 185 families. Based on growth traits (height, diameter) and tree form, 50 to 98 individuals were selected as second-generation plus trees from each site, propagated by grafting,

and planted in a clonal archive on the campus of the Kyushu Breeding Regional Office, Forest Tree Breeding Center, Japan (Kumamoto, Koshi, Japan) from 1998 to 1999. Four clonal test sites for selected individuals were established as single-tree plots or in row-plot designs of three replications by cuttings from 2001 to 2004. Each test site consisted of the clones of selected individuals from one corresponding original site (Table 1). The number of individuals at each site ranged from 432 to 1422. The number of ramets differed depending on the clone and site (average, 12.52). A progeny test was conducted for individuals forward-selected from 1998 to 2005 and from four breeding population sites established from 1969 to 1975, consisting of 32 to 185 families (Supplementary Table 1). The three of the four breeding population sites were common with the sites where individuals dedicated to clonal testing were selected. Twenty-eight individuals (3 to 11 individuals from each site) were selected as parents and were artificially crossed to each other or were open pollinated. Then, 840 individuals obtained from 34 families were planted at the progeny test site in 2011.

2.3 Measurement of tree height

For the model selection approach, data on tree height measured in 5-year intervals at each breeding population site were used. Measurement years differed depending on site, with the number of sites measured at 5, 10, 15 and 20 years being 30, 28, 23, and 12, respectively (Supplementary Table 1). For validation by clonal tests, the age of measurement of tree height in the original breeding population site differed among sites, ranging from 10 to 28 years. The height of each ramet in clonal tests was measured at stand ages of 5 and 10 years. For validation by progeny tests, data on tree height measured at age 10 at four breeding population sites were used because 10 years was the maximum common measurement age for the four breeding population sites. The measurement age for the progeny test was 5 years. Tree height was measured using a measuring pole or trigonometric methods.

2.4 Statistical models

To evaluate the efficiency of incorporating spatially autocorrelated residuals in the evaluation model to predict individual tree breeding values for forward selection, the RCB model and the AR model were compared in a linear mixed model framework:

$$y = Xb + Zu + e$$

where y is the vector of data, X is the design matrix for fixed effects, b is the vector of fixed effects, Z is the design matrix for random effects, u is the vector of random effects, and e is the vector of residuals.

Table 1 The correlation coefficients between individual tree breeding values at forward selection and genotypic values by clonal test (GV_t) or breeding value by progeny test (BV_t)

Test Type	Site Code		Tree Age		Number of FS Individuals[3]	CC[4] with GV_t or BV_t			p-value for Difference of CC
	Original[1]	C/P Test[2]	Original	C/P Test		Phenotype	BV_{fs} by RCB[5]	BV_{fs} by AR[6]	
Clonal	1108	7031	28	5	62	-0.174 ns	-0.1225 ns	0.4670 ***	<0.001 ***
				10	62	-0.137 ns	-0.0810 ns	0.4849 ***	<0.001 ***
	1107	7034	20	5	55	-0.233 ns	-0.1661 ns	0.2069 ns	0.003 **
				10	55	-0.177 ns	-0.1409 ns	0.1021 ns	0.058 ns
		7035	20	5	38	0.008 ns	-0.0387 ns	0.3982 *	0.008 **
				10	38	-0.231 ns	-0.1256 ns	0.2907 ns	0.013 *
	1098	7028	10	5	97	-0.143 ns	0.2323 *	0.2713 **	0.307 ns
				10	97	-0.175 ns	0.3287 **	0.3292 ***	0.990 ns
Progeny	4 Sites	7044	10	5	28	-	-	0.3861 ns	0.6282 ***	0.006 **

* *ns* not significant

*Significant at the 0.05 probability level; **Significant at the 0.01 probability level; ***Significant at the 0.001 probability level

[a] Breeding population site where forward selection was conducted

[b] Clonal or progeny test

[c] The number of forward selected individuals used for validation

[d] Correlation coefficient

[e] Breeding values predicted at the stage of forward selection by random complete block design model

[f] Breeding values predicted at the stage of forward selection by spatial model

The RCB model, an ordinary randomized complete block design model, has been used for the analysis of test sites in the evaluation of genetic materials in Japan. The variance-covariance matrix of e in the RCB model is

$$R = \sigma_e^2 I$$

where σ_e^2 is random residual variance and I is the identity matrix with a size the same as the number of data points.

In the AR model, an autoregressive model for spatial analysis (Dutkowski et al. 2002), the variance-covariance matrix of e is

$$R = \sigma_\eta^2 [AR1(\rho_c) \otimes AR1(\rho_r)]$$

where σ_η^2 is the variance of spatially autocorrelated residuals; $AR1(\rho_c)$ and $AR1(\rho_r)$ are first-order autoregressive correlation matrices for column and for row directions, respectively; and ρ_c and ρ_r are autocorrelation parameters for column and row directions, respectively. $\otimes$ is the Kronecker product. $AR1(\rho_c)$ or $AR1(\rho_r)$ is

$$AR1(\rho) = \begin{bmatrix} 1 & \rho & \rho^2 & \cdots & \rho^{n-1} \\ \rho & 1 & \rho & \cdots & \rho^{n-2} \\ \rho^2 & \rho & 1 & \cdots & \rho^{n-3} \\ \vdots & \vdots & \vdots & \ddots & \vdots \\ \rho^{n-1} & \rho^{n-2} & \rho^{n-3} & \cdots & 1 \end{bmatrix}$$

where ρ is the autocorrelation parameter for row or column, and n is the number of individuals for column

direction (n_c) or row direction (n_r). In this model, the data were sorted by column and row direction, and dummy data were added for missing trait data or individuals in the $n_c \times n_r$ field rectangle. In this model, a spatially independent random factor having variance σ_e^2 is added for random effect. In both the RCB and AR models, fixed effects include an effect of the block and random effects include the genetic effect of individual trees and their parents, the genetic effect of family, and the effect of interaction between block and family. The genetic relationship among individuals and their parents was provided by an additive relationship matrix (Lynch and Walsh 1998). The above-mentioned parameters (variances and autocorrelation parameters) were estimated by restricted maximum likelihood (REML) method. When parameter estimation using the REML method did not converge for either model, random factors describing the interaction between block and family and random factors describing family were removed in the presented sequential order from both models, and parameters were estimated at each removal. In the case where parameter estimation did not converge for either model even with the two random factors removed, subsequent analysis was not conducted. The predictions for random factors, including best linear unbiased predictor (BLUP) of individual tree breeding values, were obtained as the solution of Henderson's mixed-model equations (Robinson 1991).

2.5 Model selection at the stage of forward selection

For the model selection approach, the Akaike information criterion (AIC, Akaike 1974) was used to select a more predictive model. The AIC was calculated as

$$\text{AIC} = -2\ln(L) + 2m$$

where $\ln(L)$ is the log-likelihood of the model and m is the number of estimated parameters.

Comparison of the adopted model based on AIC and the alternate model was conducted from several perspectives. The correlation of breeding values of selected individuals between the models, the number of individuals selected in common for both models, and the ratio of relative genetic gain of selected individuals by the alternate model to the adopted model was used as indicators of the impact of incorporating spatially autocorrelated random factors at forward selection. The top 1% of individuals in the breeding value of height was treated as selected individuals for each site for both the adopted and alternate model. The ratio of relative genetic gain was calculated as the ratio of the average breeding values of individuals selected in the alternate model to those for the adopted (better) model, based on the breeding values calculated in the adopted model. As the number of replications differs among test sites, the influence of different replication numbers was tested by the Kruskal-Wallis test for each stand age.

To estimate the relationship between land forms of sites and the autocorrelation parameters, we calculated the topographic ruggedness index (Riley 1995) for each site. The calculation was executed using the r.tri command of GRASS GIS 7 software (GRASS Development Team 2015) based on 10 m digital elevation models provided by the Geospatial Information Authority of Japan. The topographic ruggedness index for the range of each site was averaged and compared with the autocorrelation parameter for each site.

2.6 Validation using clonal and progeny tests

For validation using clonal tests, the breeding value at forward selection (BV_{fs}) of tree height of selected individuals was calculated for each original site based on both the RCB model and the AR model. The age at measurement differed by site (Table 1). The genotypic value by clonal test (GV_t) of selected individuals was obtained from each clonal test by a linear mixed model framework with REML/BLUP methods. Four models, including the RCB model with an additive relationship matrix, the RCB model without an additive relationship matrix, the AR model with an additive relationship matrix, and the AR model without an additive relationship matrix, were tested, and the most predictive (the lowest AIC) model was adopted for each clonal site to predict GV_t.

For validation using the progeny test, BV_{fs} was calculated by multisite analysis. In the linear mixed model, fixed effects include site effect and block-in-site effects, and random effects include individual and parental effects and their interaction with the site. In the multisite RCB model, the variance-covariance matrix of residuals is

$$R = \begin{bmatrix} \sigma_{e_1}^2 I_{n_1} & 0 & 0 & 0 \\ 0 & \sigma_{e_2}^2 I_{n_2} & 0 & 0 \\ 0 & 0 & \sigma_{e_3}^2 I_{n_3} & 0 \\ 0 & 0 & 0 & \sigma_{e_4}^2 I_{n_4} \end{bmatrix}$$

where $\sigma_{e_x}^2$ is the residual variance of site x, I_{n_x} is the identity matrix with size n_x, and n_x is the data size of site x. In the multisite AR model, single-site analysis is first executed for each site by the AR model as described above, and a spatially autocorrelated residual for each tree is estimated. Then, the obtained spatially autocorrelated residual is subtracted from each measured tree height. Finally, the adjusted tree heights are used as a dependent variable for the multisite analysis conducted with the same residual variance-covariance matrix structure used in the multisite RCB model. The breeding value by progeny test (BV_t) of selected individuals was obtained from a single progeny test by a linear mixed model framework with REML/BLUP methods. Two models, the RCB model with an additive relationship matrix and the AR model with an additive relationship matrix, were tested and the more predictive (the low AIC) model was adopted to predict BV_t.

We assume that GV_t and BV_t are more precise than the individual tree breeding values at forward selection (BV_{fs}), because the information from multiple direct ancestors or multiple ramets of target individuals were used to obtain BV_t and GV_t. The Pearson's correlation coefficients between BV_{fs} and GV_t or between BV_{fs} and BV_t were calculated for both the RCB and AR models at forward selection as a measure of the accuracy of the prediction of breeding values at forward selection. Higher correlation coefficients mean that the relative magnitude of BV_{fs} is more analogous to the relative magnitude of BV_t or GV_t, and this implies that ranking of breeding materials at the stage of forward selection is more accurate. Statistical testing for the difference between the correlation coefficients of the RCB model and the AR model was conducted with William's test (Williams 1959) by the psych package in R (R Foundation for Statistical Computing, Vienna, Austria).

2.7 Calculations

The estimation of variances and correlation parameters, and the prediction of random factors, were conducted using ASReml software (VSN International, Hemel Hempstead, UK).

3 Results

3.1 Model selection at the stage of forward selection

In the model selection approach, 93 cases, which are the combination of test sites and measurement age, were analyzed. In six cases, the AR model did not converge in the REML procedure, and those cases were excluded from the subsequent analyses. In 87 converging cases, AICs were compared between the AR and RCB models, and the AR model showed smaller AIC in all cases, implying that the AR model is better than the RCB model for all sites and measurement ages in this study.

The average of the autocorrelation parameters (the average of ρ_c and ρ_r) ranged from 0.575 to 0.966 (average 0.853), and the ratio of σ_η^2 to σ_e^2 ranged from 0.239 to 4.08 (average 0.898) for the AR model. One case of estimated autocorrelated residuals is depicted in Fig. 1. The relationship between the average topographical roughness index of site and autocorrelation parameters is shown in Fig. 2. Pearson's correlation coefficients between topographic ruggedness index and autocorrelation parameter were increased with test age, i.e., 0.01 (ns), 0.398 ($p < 0.05$), 0.470 ($p < 0.05$), and 0.542 (ns) for measurement ages 5, 10, 15, and 20 years, respectively, while the correlation coefficients at age 20 were not significant.

When selection of the top 1% individuals based on height breeding value is simulated, the percentage of commonly selected individuals between the two models was 13.3 to 92.8% (average 58.4%) at stand age 5, 0.0 to 85.5% (average 53.7%) at stand age 10, 0.0 to 89.2% (average 53.2%) at stand age 15,

and 6.5 to 85.5% (average 59.7%) at stand age 20 (Fig. 3a). The correlation coefficients of breeding values of selected individuals between the RCB and AR models for each site were −0.095 to 0.936 (average 0.569) at stand age 5, −0.467 to 0.958 (average 0.463) at stand age 10, −0.107 to 0.910 (average 0.624) at stand age 15, and 0.038 to 0.936 (average 0.440) at stand age 20 (Fig. 3b). The relative genetic gains were calculated as the ratio of the average breeding values of individuals selected in RCB model to those in AR model, based on the breeding values calculated in the AR model, because AR model was selected for all cases. The relative genetic gains ranged from 0.514 to 0.995 (average 0.904) at stand age 5, 0.472 to 0.996 (average 0.887) at stand age 10, 0.562 to 0.996 (average 0.907) at stage 15, and 0.421 to 0.990 (average 0.900) at stand age 20 (Fig. 3c). The effect of the number of replications was not significant for any measurement age when comparing percentages of commonly selected individuals, correlation coefficients of breeding values, or relative genetic gains.

3.2 Validation using clonal and progeny tests

In the validation approach, we compared the performance of forward selection between the AR and RCB models by examining the relationship between BV_{fs} and GV_t or between BV_{fs} and BV_t. The correlation between evaluation value (BV_{fs}) at the selection and validated values by clonal (GV_t) or progeny test (BV_t) is shown in Table 1. For clonal tests, the correlation between phenotypic value (raw measured height value) at the selection and validated genotypic values were not significant
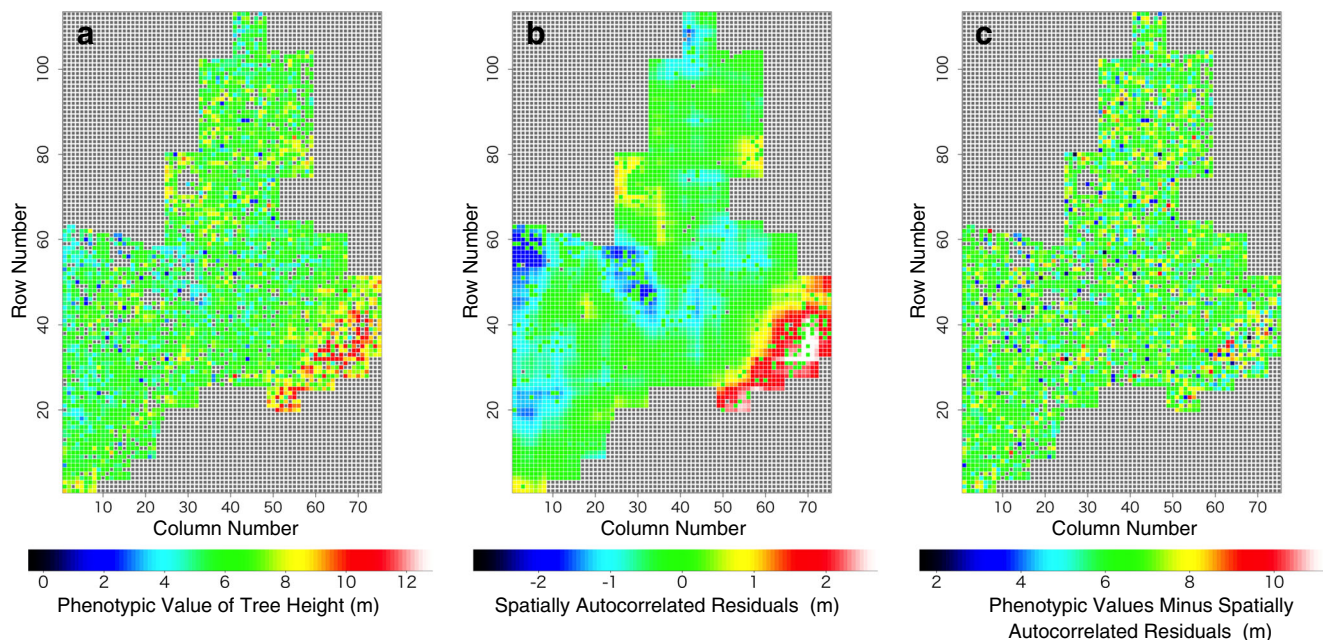

Fig. 1 Tree height and estimated spatially autocorrelated residuals for site No. 1180 at stand age 10. **a** Measured phenotypic value of tree height (m). **b** Estimated spatially autocorrelated residuals. **c** Calculated difference between phenotypic value and spatially autocorrelated residuals. Each cell shows one individual. The average value of the autocorrelation parameters for row and column directions is 0.912
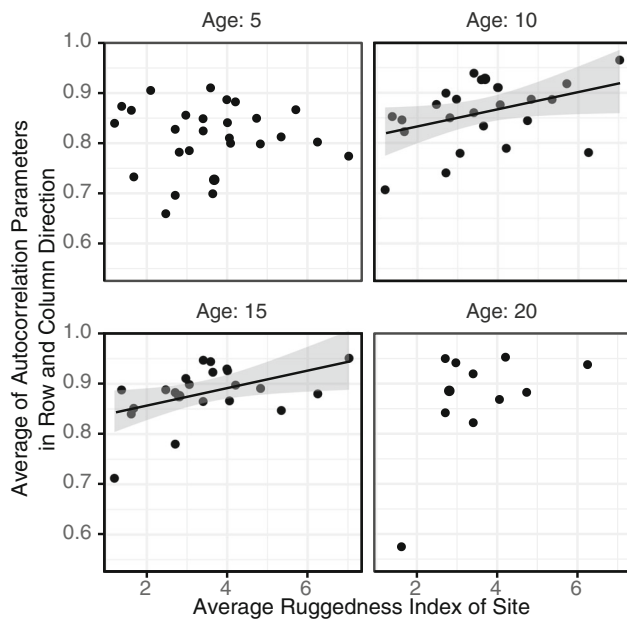
Fig. 2 The relationship between average ruggedness index of a site and average autocorrelation parameters of row and column directions at various stand ages. The number of sites included in the analysis was 28, 26, 22, and 11, and Pearson's correlation coefficients between the average ruggedness index and average autocorrelation parameters were 0.01 (ns), 0.398 ($p < 0.05$), 0.470 ($p < 0.05$), and 0.542 (ns) for stand ages 5, 10, 15, and 20 years, respectively. Lines are regression lines and the gray area indicates the 95% confidence interval

for all cases. With the RCB model, only two out of eight cases showed a significant correlation coefficient between BV_{fs} and GV_t. In contrast, five out of eight cases in the AR model showed significant correlation coefficients. In all cases, correlation coefficients were higher for the AR model than for the RCB model, and the differences of correlation coefficients were significant for five cases (Table 1). In validation using the progeny test, the correlation coefficient for the RCB model was 0.386 (ns), whereas the value for the AR model increased to 0.628 ($p < 0.001$; Table 1), and the difference of correlation coefficients was significant ($p < 0.005$; Table 1). The relationship between BV_{fs} and BV_t for trees selected at four sites is depicted in Fig. 4. From the results, we found that the breeding values predicted by the AR model are more accurate than those predicted by the RCB model.

4 Discussions

In our research, a novel validation approach using clonal or progeny tests of forward-selected individual trees revealed that spatial analysis is effective for the evaluation of genetic performance of individuals at the stage of forward selection. Previous reports (Costa e Silva et al. 2001; Dutkowski et al. 2006; Ye and Jayawickrama 2008) illustrated the effectiveness of spatial models based on better model fitting using datasets collected from trees in one generation. This study added

practical validation results showing the superiority of spatial analysis for forward selection using a dataset spanning two generation.

As conducted in previous reports, we analyzed model selection approach in *C. japonica*. The AR model was more predictive than the RCB model in all the 87 cases we tested at the stage of forward selection by model selection approach. This means that incorporating spatially autocorrelated residuals is an effective way to explain tree height in the genetic experiments in *C. japonica*, as in other species (Costa e Silva et al. 2001; Dutkowski et al. 2006; Ye and Jayawickrama 2008). The extent of effectiveness depended on individual cases in our research. For the case of using relative genetic gain during indirect selection by RCB to AR as a measure, the values were almost the same for both models at several sites (0.996 in maximum), while some sites showed lower relative genetic gain by the RCB than the AR model (< 0.75) even at sites with six replications (Fig. 3c). Incorporating spatially autocorrelated random factors changed the composition of the selected population about 50% on average, and completely in some cases, as shown by the numbers of individuals commonly selected by both models (Fig. 3a). Thus, the adoption of spatial analysis would have large impacts on the population composition of the selected individuals depending on the circumstances.

In addition to the model selection approach, we further revealed the superiority of the AR model using a validation approach. As shown in the model selection approach results, the correlation of breeding values of selected individuals between the RCB model and the AR model was not so high (Fig. 3b). The question of which model more accurately predicted the breeding values was practically approached by validation using successive clonal or progeny tests in this research. This is the first study to practically demonstrate the superiority of the AR model for forward selection. Integration of results from the model selection approach with the results of the validation approach suggests that the spatial model is effective for the evaluation of genetic performance of tree height for forward selection in *C. japonica*. In the validation approach, the correlation coefficients between breeding values at the stage of forward selection and breeding values by clonal or progeny tests are not so high (0.628 for progeny tests and a maximum 0.485 by clonal test) whereas the AR model showed higher values than RCB model, and the RCB model showed very low values in many cases. This partly shows the difficulty of predicting breeding values at the stage of forward selection. This low correlation coefficient was also caused by a decrease in genetic variation in the group of selected individuals. In this research, not all individual trees but phenotypically superior individual trees from each site were collected and dedicated to use in clonal and progeny tests. Therefore, genetic variation would decrease compared to the genetic variation of the group of all individual trees at a site. The residual
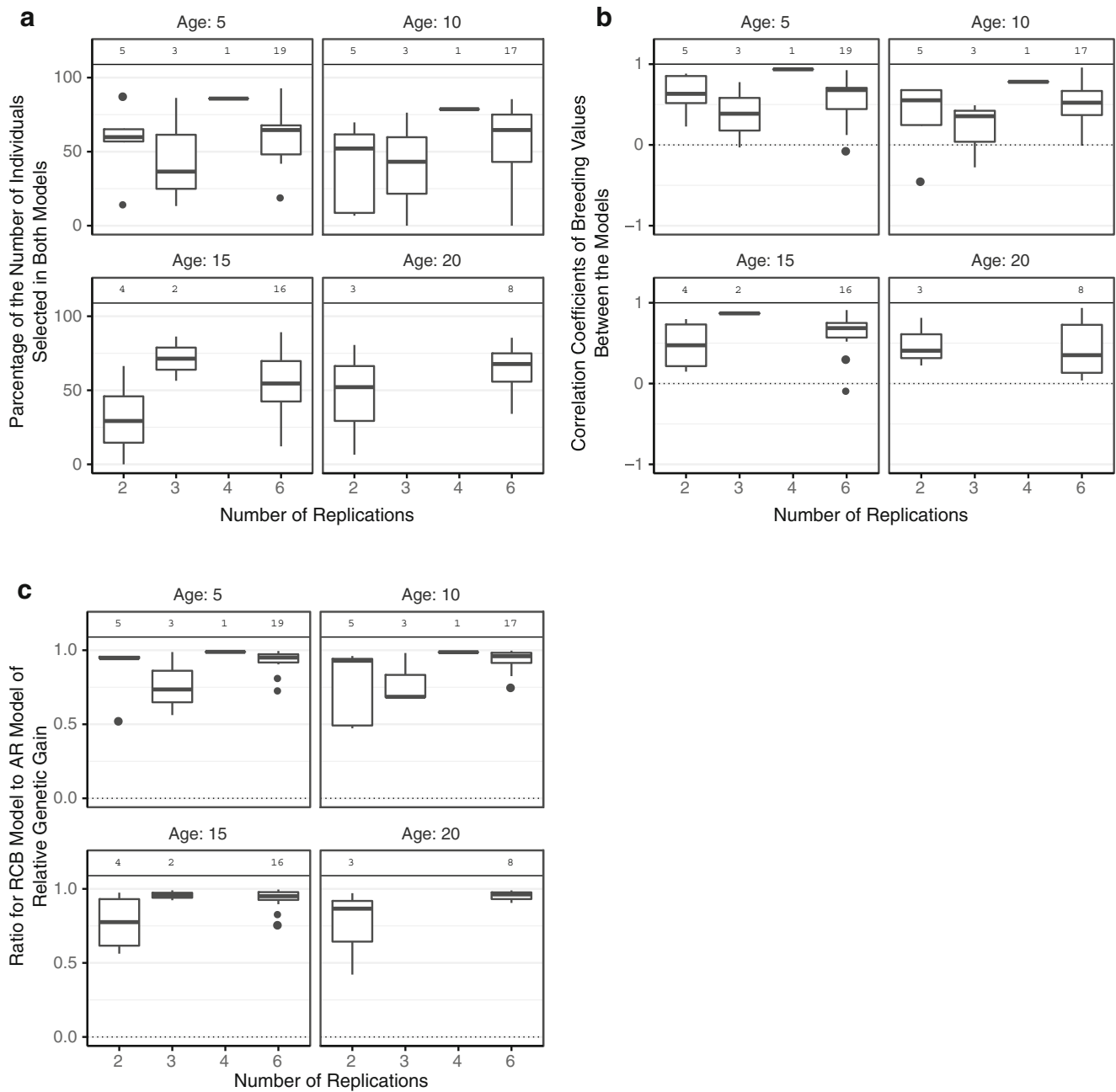
Fig. 3 Comparison of spatial model and randomized complete block design model. **a** The percentage of number of individuals selected for both models. **b** The correlation coefficients of breeding values between models. **c** The ratio of relative genetic gain of the RCB model to the AR model. The top 1% of individuals for each site was assumed to be selected. The number above each box indicates the number of the test site

variation would not change with selection; as a result, the correlation coefficients for both models would be biased downward compared to correlation analysis targeting all individual trees at a site.

The validation by clonal tests showed lower correlation than validation by the progeny test. It is likely that the clonal test is affected by the rooting performance for each individual since there is clonal variation for rooting performance (Ohba 1993), even though *C. japonica* is generally known for having high rooting performance. The research concerning the

comparison of genetic growth performance between clonal propagation and seedlings is limited. Gaspar et al. (2005) reports that there is no significant effect of interaction between the propagation method (cuttings and seedlings) and families in *Eucalyptus globulus* Labill. whereas their report did not examine rooting performance at the individual level. Matsunaga et al. (2008) reported that the effect of propagation method on growth performance depends on genotypes by comparing cuttings and open pollinated seedlings of plus trees in *C. japonica*. For the published data in Matsunaga et al.
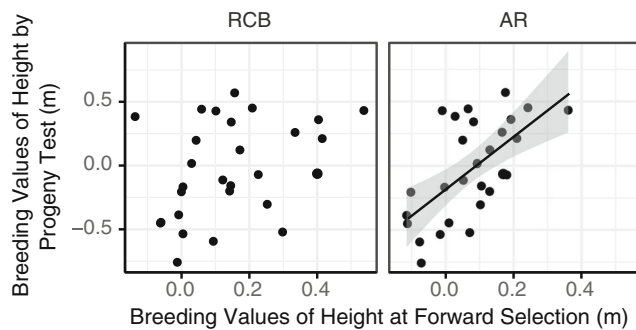
Fig. 4 The relationship between individual tree breeding values of height (in meters) at selection stage and breeding values of height (in meters) for forward-selected individual trees by the progeny test site for the RCB and the AR. $n = 28$. The correlation coefficients were 0.386 (ns) and 0.628 ($p < 0.001$) for RCB and AR, respectively. Line is a regression line and the gray area indicates the 95% confidence interval

(2008), the cutting average of growth performance had no significant correlation with seedling average among plus trees in *C. japonica*. A difference in rooting performance, which is difficult to predict at the stage of forward selection, would likely affect subsequent growth of clonal trees and make the prediction of clonal growth performance difficult. Even though the rooting effect exists, the AR model should be adopted for forward selection targeted toward clonal forestry.

In this research, we used tree height as a target trait. Dutkowski et al. (2006) analyzed many traits important to forestry by the spatial model and showed that such a model is effective for selection of growth traits but is not effective for some other traits, especially those related to wood properties. Several traits related to wood properties are related to the growth state of individuals (Zobel and Jett 1995). For example, Kijidani et al. (2010) showed that the height-diameter ratio affects the wood stiffness in *C. japonica*, and Fukatsu et al. (2015) showed residual correlation between growth traits and several traits related to wood properties in *Larix kaempferi* (Lamb.) Carrière. For such traits related to growth traits, there is a possibility that spatial analysis is effective for forward selection.

There was a trend for the spatial parameters to increase with ruggedness of the site in this research. Our research showed that a randomized complete block design is inadequate for rugged sites like typical forest locations in Japan. Attempts for adjusting the micro-environmental effects on growth have been conducted (Ohba et al. 1975; Matsuzaki and Akashi 1987) in Japan, although the methods have not been adopted due to the complexity of procedures and the uncertainty of their utility. On the other hand, methods incorporating spatially autocorrelated residuals into a mixed model (Cullis and Gleeson 1991) are concise and easily applicable to a practical tree-breeding strategy, and the efficiency of these methods has been demonstrated by previous and this research. Software for this analysis, like ASReml (Gilmour et al. 2009) or the BreedR package in R statistical package (Muñoz and

Sanchez 2017), is available. Spatial analysis is recommended as a default for the evaluation of individuals for the forward selection in the practice of tree breeding.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflicts of interest.

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