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Genetic parameters and performance stability of white spruce somatic seedlings in clonal tests

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ABSTRACT

The commercial use of white spruce varieties produced by somatic embryogenesis (SE) permits increased forest productivity compared to other reproductive technologies. However, the use of SE in clonal forestry requires an accurate assessment of genetic parameters and the performance stability of clones in plantations. For these reasons, two clonal tests were established of 52 white spruce somatic clones. In each clonal tests, we measured survival, bud dormancy, stem form, growth and branching characteristics of clones 4 years after outplanting. There was a large variability among clones for characteristics related to growth and branching. At this juvenile stage, the clonal heritability estimates for all characteristics remained low. Of all the characteristics studied, height had the highest heritability. The selection of the top 20 clones (38% of the clones) provided a genotypic gain in height of about 4% for the two planting sites, which is reasonable for such a low selection intensity. High genotypic correlations were observed between growth and branching characteristics. Although a significant site effect was observed for most characteristics, the genotype × site (G × E) interaction was low and consequently the correlation between the two sites for the same characteristic was high. The performance stability of the somatic clones at both sites indicates that opportunities exist for selection of clones that adapt and perform well over different ecological regions, permitting a tangible increase in forest productivity.

1. Introduction

Plantation forestry using genetically improved material is an efficient way of maximizing fiber production on small areas of land (Sedjo, 1999, 2003; FAO, 2002). White spruce (*Picea glauca* (Moench) Voss) is an important species for lumber, pulp, and paper (Zhang et al., 2004) and is one of the most widely used tree species in reforestation programs in Canada. For example, 27 million large white spruce seedlings (height ⩾35 cm) were produced in Quebec in 2008, 18% of the total annual seedling production (Beaulieu et al., 2009). White spruce has a transcontinental range in North America and is adapted to a wide range of soil and climatic conditions (Nienstadt and Zasada, 1990).

Provenances and progenies representative of the entire natural range of white spruce have been evaluated over the last few decades. Results showed the presence of geographical variation in growth characteristics as well as a large intra-provenance variation (Corriveau and Boudoux, 1971). Moreover, a high genetic variation both among and within white spruce families for growth and biomass characteristics have been reported in several studies (Yanchuk and Kiss, 1993; Li et al., 1997; Carles et al., 2011). Breeders have used this variation to establish seed orchards, which are now producing most of the plants used in reforestation programs (Rainville and Beaulieu, 2007; Mullin et al., 2011). Over the past decade, cutting propagation of high-value families obtained through controlled crossing of superior genotypes has also been introduced into the production system to capture more genetic gains (Beaulieu and Bernier-Cardou, 2006; Gravel-Grenier et al., 2011). More recently, somatic embryogenesis has been used as an advanced propagation method, and large differences in morpho-physiological variables were observed among containerized white spruce somatic clones under nursery conditions (Lamhamedi et al., 2000) and in clonal field tests (Rainville et al., 2011; Weng et al., 2008). Hence, there is a trend in eastern Canada towards the implementation of multi-varietal or clonal forestry of white spruce (Weng et al., 2008), with the aim of increasing forest productivity and achieving greater uniformity of wood quality attributes.

The first critical step in the successful implementation of such a program is the identification of genetically superior clones. Genetic characterization of clones at a young age makes it possible to select clones that show fast initial growth, thus reducing the costs

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associated with plantation maintenance. However, for early selection to be efficient, age–age correlations must be sufficiently high, since this would indicate that there is a good likelihood that clones which perform well at a young age will also perform well at a later age. Fortunately, Li et al. (1993) demonstrated that age–age correlations were indeed quite high for white spruce height growth. For wood quality traits, on the other hand, longer testing time is needed because wood quality traits change as trees mature (Zobel and van Buijtenen, 1989).

Well established and designed clonal tests are critical for the success of any selection program using somatic clones. These tests should permit an accurate estimate of genetic parameters and thus increase the reliability with which clonal performance can be predicted. Both of these are essential to determining optimum strategies for clonal forestry and in calculating genetic gains. Replicated clonal tests also make it possible to estimate genotype × environment (G × E) interactions. In tree improvement, it is important to take account of the G × E interaction as both its magnitude and pattern have profound implications for breeding, testing, and seed management. The presence of a significant G × E interaction implies that clone performance depends on the environment where it is planted, and that specific clones must be matched to specific environments to maximize plantation yield. The G × E interaction may be attributable to various causes, including the heterogeneity of genotypic variance and/or the lack of perfect genetic correlation among environments (Xie, 2003). G × E interaction in clonal tests has been reviewed by several authors (Paul et al., 1997; Baltunis and Brawner, 2010).

In clonal forestry, mass propagation of coniferous species at the operational scale most commonly done by cuttings (Baltunis et al., 2007, 2008; Baltunis and Brawner, 2010; Isik et al., 2003). However, success of clonal propagation by cuttings is limited by different factors such as the age of the stock plant and the limited number of cuttings that can be harvested from each stock plant. Somatic embryogenesis (SE) has been shown to be the most promising technology to mitigate these disadvantages (Baltunis et al., 2009). Few studies have been conducted to estimate genetic parameters of somatic clones in field trials (Dean, 2008; Baltunis et al., 2009), and even fewer studies have involved white spruce (O'Neill et al., 2005; Weng et al., 2008). The literature on G × E interactions is extensive for forest trees in general (Xie, 2003), but is basically non-existent for white spruce somatic clones. Consequently, the objectives of this study are (i) to evaluate genetic parameters such as variances, heritabilities, genotypic correlations and genetic gains using white spruce clonal material produced through somatic embryogenesis (SE), and (ii) to examine the genotype × environment interactions to characterize the genetic stability of SE clones.

2. Materials and methods

2.1. Plant material and growth conditions

White spruce seeds were obtained from controlled crosses conducted with selected genotypes. Fifty-two clones were propagated from 14 full-sib families for the establishment of clonal tests (Table 1). The parent trees are part of a large breeding population maintained by the MRNF, and they were chosen for their superior performance and expected genetic gain. These tests are the first of a series of trials that are currently installed to identify the best of over 1000 somatic clones that have been produced and planted under different ecological site conditions. Embryos were excised from immature seed of the full-sib families and consecutively placed in induction and maintenance media as described in Tremblay (1990). Tissues were then transferred to a maturation medium containing abscisic acid for 6 weeks to advance embryo development. After the maturation phase, somatic embryos were placed directly onto Sorbarod™ plugs saturated with liquid Campbell and Durzan salts at half strength, followed by germination in Corning containers (250 ml) and placed in *ex vitro* nursery conditions for acclimation, as described in Lamhamedi et al. (2003). Germinant somatic clones were transplanted into containers (IPL 25-310, IPL®, Saint-Damien-de-Buckland, Quebec, Canada) filled with a peat-vermiculite growing medium (3/1, v/v; bulk density of 0.084 g/cm^3). After acclimation, somatic seedlings (ramets) were transferred in April 2005 to an unheated polyethylene tunnel for their first growing season (1 + 0), and cultivated outside for their second growing season (2006). Seedling production was conducted over two consecutive growing seasons following standard cultural practices for white spruce, under nursery conditions at the Pépinière forestière de Saint-Modeste (Québec, Canada, 47°50′N, 69°30′W) (Lamhamedi et al., 2006) .

Table 1
Controlled crosses, families (14) and somatic clones (52) of white spruce.

Controlled crosses (♀ × ♂)	Family codes	Clonal codes
WES-1 × BEL-2	1	1
PTH-1 × PTH-8	2	2, 3, 4, 10, 11, 12, 16, 20, 21, 24, 25
ALG-10 × ALG-8	3	5, 7, 8, 9, 14, 22
PFS-10 × PFS-2	4	6
BRO-1 × PFS-3	5	26, 28
BEL-2 × ZEN-2	6	43, 44, 98
MIT-2 × PTH-1	7	46, 50, 55, 76, 78, 79, 80, 81, 94, 95
BRO-3 × PFS-6	8	48
PTH-3 × PTH-11	9	49, 57, 71
WES-2 × AEC-2	10	60, 61, 63, 82, 83, 86
SUN-1 × ALG-8	11	72, 84, 85, 96
PTH-3 × ALG-7	12	88, 90
FRA-3 × CAR-3	13	89
PFS-12 × PFS-4	14	18

2.2. Clonal tests and experimental design

Two-year-old (2 + 0) containerized somatic seedlings were planted in two clonal tests in abandoned bare-root fields at Grandes-Piles and Saint-Modeste provincial forest nurseries in Québec, Canada, on 22–23 May 2007. The Grandes-Piles and Saint-Modeste sites correspond to 2B-T (sugar maple) and 4F-T (eastern balsam fir) bioclimatic subdomains, respectively (Saucier et al., 1998). Information on site locations as well as climatic and growing conditions associated with these subdomains are provided in Table 2. Somatic seedlings were laid out following a randomized complete block design with 52 clones replicated in 10 blocks, each clone being represented by a single-tree plot, corresponding to one ramet/clone/block/site. Clones were planted at a 2.5 × 2.5 m spacing

Table 2
Climate data and growing conditions in the sugar maple and balsam fir subdomains in which the white spruce clonal field tests are being conducted.

Planting sites	Latitude (N)	Longitude (W)	Altitude (m)	Pma[a](mm)	Tma[a] (°C)	DD[a] (°C)	AI[a]
Grandes-Piles (sugar maple subdomain)	46°41′	72°43′	55	900–1000	2.5–5	3000–3400	175–225
Saint-Modeste (balsam fir subdomain)	47°50′	69°30′	268	900–1000	2.5	2200–2600	150–200

[a] Pma: mean annual precipitation, Tma: mean annual temperature, DD: Degree Days, AI: Aridity Index (Saucier et al. (1998)).