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Research paper

Xylogenesis in stems and roots after thinning in the boreal forest of Quebec, Canada

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The reduction of competition through thinning increases radial growth in the stem and roots of many conifer species. However, not much is known about the effect of thinning on the dynamics of wood formation and intra-annual development of the growth ring, especially in the roots, which are an essential part of the tree for stability and resource acquisition. The aim of this study was to evaluate the effect of an experimental thinning on the dynamics and phenology of xylogenesis in the stem and roots of black spruce and balsam fir. Experimental and control trees were selected in two mature even-aged stands, one black spruce (*Picea mariana* (Mill.) BSP) and one balsam fir (*Abies balsamea* (L.) Mill.). Wood microcores were collected weekly in the stem and roots from May to October for a period of 4 years. The onset and ending of each cell differentiation phase were computed, as well as growth rate and total cell production. Results show that thinning increased the cell production rate of stem and roots of black spruce and balsam fir. This higher daily growth rate caused an increase in the total number of cells produced by the cambium. The intensity of the treatment was sufficient to significantly increase light availability for residual trees, but insufficient to modify soil temperature and water content to a point at which a significant change in the timing or duration of xylogenesis would be induced. Thus, thinning increased cell production rate and total number of cells produced in both stem and roots, but did not result in a change in the phenology of wood formation that could lead to increased risks of frost damage in the spring or autumn.

Keywords: *Abies balsamea*, boreal forest, cell differentiation, growth, intra-annual ring development, phenology, *Picea mariana*.

Introduction

Traditionally, clearcuts have been the most common silvicultural treatments in Quebec's boreal forest. However, the transition to ecosystem-based forest management requires silvicultural options that maintain or enhance the long-term health and functions of forest ecosystems. Thinning, which involves removing only a selected part of the trees in a stand (Côté 2000), can be used to either maintain or modify stand structure and composition, or increase the resistance of some stands to spruce budworm (Bauce 1996, Ministère des Ressources Naturelles 2003).

Conifers are very important for the wood industry in eastern Canada, particularly black spruce (*Picea mariana* (Mill.) B.S.P.) and balsam fir (*Abies balsamea* (L.) Mill.), two species of great commercial interest. Black spruce has the capacity to grow in a broad range of conditions but is slow-growing (Viereck and Johnston 1990), while balsam fir is more shade-tolerant (Frank 1990) and can react more vigorously than black spruce to a canopy opening (Lemay et al. 2016). The effect of thinning on the growth of residual stems of both species typically results in increased radial growth, usually within 2–4 years following the intervention (Ruel et al. 2003, Vincent et al. 2009, Goudiaby

et al. 2012, Pamerleau-Couture et al. 2015, Lemay et al. 2016). However, the effect of such treatment on root growth is less documented. Growth increases in roots have been observed to occur 1–2 years earlier and be more marked than those in the stem (Kneeshaw et al. 2002, Ruel et al. 2003, Vincent et al. 2009, Krause et al. 2014). This has been interpreted as a response to the increased availability of resources in the soil and to the mechanically induced forces resulting from increased wind penetration in the stand (Ruel 1995, Thibodeau et al. 2000).

Xylogenesis, or wood formation, is a complex process of cambium division and differentiation, where cambial cells divide and generate derivatives that differentiate physiologically and morphologically until their final maturity (Rossi et al. 2006*b*). Cell morphology is the result of processes occurring in two successive differentiation phases: duration and rate of cell enlargement determine the final cell radial diameter, while the duration and rate of wall deposition determine the amount of secondary cell wall (Cuny et al. 2014). Environmental conditions have an influence on the dynamics of xylogenesis, timing of production and differentiation of xylem cells and, thus, on radial growth (Linares et al. 2009, Cuny and Rathgeber 2016, Rossi et al. 2016). Lupi et al. (2012*b*) found that an increase in soil temperature resulted in earlier onsets of xylogenesis and that the effect of warming was especially marked in the phenology of the roots. It has also been shown that a reduction of competition through thinning prolongs xylem cell production (Linares et al. 2009), and that dominant trees, which are subject to low competition for resources, have longer periods of cambial activity than suppressed trees (Rathgeber et al. 2011). Thus, the changes in environmental conditions following thinning, such as increased soil temperature and light availability, could influence xylogenesis, especially in the roots through soil warming. However, an earlier or longer growth period might expose trees to unfavourable conditions, for instance a late frost in spring, at the onset of cell division, or in autumn, before the beginning of winter dormancy. Despite this, little attention has been dedicated to understanding the effect of a partial canopy removal on the phenology and dynamics of wood formation. Most previous studies on xylogenesis have yielded information on the dynamics of cambial activity and the effects of climate, and the majority of those studies have concentrated on the stem. Very little information is available on the roots, an essential component for resource acquisition and tree stability. Better knowledge of the dynamics of wood formation after thinning could help identify how critical factors affect development and production of wood.

The aim of the study was to evaluate the effect of an experimental thinning on the intra-annual growth dynamics of the stem and roots of black spruce and balsam fir in the boreal forest. Our hypotheses were that: (i) more cells are produced in residual trees following treatment; (ii) thinning induces an earlier onset and longer duration of wood formation; and (iii) both of these effects should be more marked in the roots.

Materials and methods

Study sites and experimental design

The study site is located within the Simoncouche research station (48°12′N, 71°14′W, 350 m above sea level), in the Laurentides Wildlife Reserve of Quebec, Canada. Two mature, even-aged and homogenous stands originating from a forest fire in the 1920s (Gagnon 1989) were selected. One was dominated by black spruce and the other by balsam fir, and they were located about 400 m apart. The area is included in the balsam fir-white birch bioclimatic domain of Quebec (Saucier et al. 1998). The mean annual temperature and total precipitation recorded at the sites during the 4 years of the study were 3.0 °C and 760 mm, respectively. Because of the cold climate and long snow cover, the period of cell differentiation at the sites generally occurs between the end of May and end of September (Thibeault-Martel et al. 2008).

At each site, three experimental trees and three control trees were selected randomly. The study trees were dominant or codominant individuals, healthy-looking and free of visible injury or defect, with a diameter at breast height (DBH) between 14 and 22 cm (Table 1). Trees had at least four main lateral roots with a horizontal diameter larger than 10 cm. An experimental thinning was conducted in early spring 2012, which consisted of removing all neighbouring trees with a DBH greater than 10 cm within a 4 m radius around the experimental trees. To quantify thinning intensity, Hegyi's competition index (Hegyi 1974) was calculated before and after treatment according to the following formula:

$$CI = \sum_{j=1}^{n} \left(\frac{DBH_j}{DBH_i} \times \frac{1}{Dist_{ij}} \right)$$

Table 1. Mean experimental trees characteristics (±standard deviation), measured at the beginning of the study in 2011, and number of competitors removed during the thinning in 2012.

	Black spruce				Balsam fir			
Trees	DBH (cm)	Height (m)	Number of competitors	Competitors removed	DBH (cm)	Height (m)	Number of competitors	Competitors removed
Treated	18.6 ± 2.0	17.1 ± 2.7	9.3 ± 1.1	4.0 ± 1.0	17.5 ± 1.2	16.4 ± 0.7	12.7 ± 4.7	3.7 ± 1.1
Control	20.8 ± 2.0	19.0 ± 1.9	4.0 ± 1.7	–	20.0 ± 1.1	18.6 ± 2.5	8.7 ± 3.0	–

where DBH_i and DBH_j are the diameter at breast height of the subject tree *i* and competitor tree *j*, respectively, $Dist_{ij}$ is the distance between subject tree *i* and competitor *j*, and *n* is the number of competitors within a 4 m radius around the subject tree *i* (Mailly et al. 2003). The treatment reduced the Hegyi's competition index by 60 and 45% for black spruce and balsam fir, respectively.

Microclimatic measurements

Soil temperature sensors were installed in November 2011 near one of the main lateral roots of each tree, in the northwest direction about 50 cm from the stem, between the organic and mineral layers. Measurements were taken every 15 min and data were stored as hourly averages in two CR10X dataloggers (Campbell Scientific Corporation, Edmonton, Canada). The soil volumetric water content was measured weekly with a Field Scout TDR 200 soil moisture metre (Spectrum Technologies Inc., Plainfield, IL, USA) at a distance of 1 m from the stem of each tree, in four orthogonal directions and at a depth of 20 cm. Understory light conditions were measured once in July 2014, 2 years after thinning, using measurements of the percentage of above-canopy photosynthetic photon flux density (PPFD) acquired under overcast sky conditions (Gendron et al. 1998). Measurements were taken at 30 cm above ground, in four orthogonal directions and every 50 cm from the stem up to 4 m using a quantum sensor for photosynthetically active radiation (PAR), connected to a LI-1400 datalogger (sensor LI-190SA, Li-Cor Inc., Lincoln, NE, USA). To obtain the percentage of above-canopy PPFD, each measurement was divided by the total available light, recorded at the same time by another PAR sensor installed in a nearby open area and connected to a datalogger.

Sample collection and preparation

From 2011 to 2014, wood microcores (2 mm in diameter, 25 mm long) were sampled weekly with a Trephor (Rossi et al. 2006*a*) from May to October. The microcores were collected on the stem and on one main lateral root of each tree. On the stem, samples were taken every 10 cm following a counter-clockwise rising spiral, starting 1 m from soil level. On the roots, samples were taken every 3 cm following a zigzag pattern on the upper part of the root, starting 25 cm from the collar (Thibeault-Martel et al. 2008) and ending when root diameter became less than 4 cm, which in all cases remained within a distance of 60 cm from the stem base. This sampling method avoids the eccentric growth pattern often observed in roots (Fayle 1968, Krause and Eckstein 1993). The repetitive sampling can generate a stress leading to the formation of traumatic resin ducts (Deslauriers et al. 2003). Root sampling was thus limited to a single year, and different roots of the same tree were sampled for each year of the study. Microcores contained phloem tissues, the cambium and xylem tissues consisting of the developing annual ring and about five rings from the previous years. After collection, samples were stored in a water:ethanol solution (1:1) to avoid tissue deterioration during transport.

The microcores were dehydrated using successive immersions in ethanol and D-limonene, and embedded in paraffin. Samples were cut into 7 µm thick sections with a rotary microtome, fixed on slides and stained with cresyl violet acetate (0.16% in water) (Thibeault-Martel et al. 2008). The sections were observed under visible and polarized light at magnifications of 400–500×. Xylem formation was assessed by counting the number of developing cells in the cambial zone, in radial enlargement, in wall thickening and lignification, and the number of mature cells, each along three radial files (Deslauriers et al. 2003, Rossi et al. 2006*c*). Cells in the cambial zone had thin walls and a small radial diameter, while enlarging cells had a radial diameter at least twice that of cambial cells, with thin walls that did not shine under polarized light. Cells in the wall thickening and lignification phase had walls that changed from light violet (at the start of lignification) to blue (when maturation was complete) and shone under polarized light. Xylem tracheids were considered mature when walls were completely blue (Rossi et al. 2006*c*).

Xylem formation was considered to have begun when the average of number of cells in enlargement was greater than one. At the end of the growing season, xylem formation was considered complete when the average of number of cells in wall thickening and lignification was less than one. Onset and ending of enlargement, and wall thickening and lignification and first mature cell were computed in days of the year (DOY).

Data analyses and statistics

In 2012, one thinned black spruce root did not produce a tree ring so that year was eliminated from analyses. This is not unusual; it is well known that missing rings are found more frequently in roots than stem (Fayle 1968, Krause and Eckstein 1993, Schweingruber 1996) and that a root can temporarily stop secondary growth for some years.

To analyse the differences in xylem phenology, a multifactor analysis of variance (ANOVA) with repeated measures was performed using PROC MIXED of the SAS 9.1 statistical package (SAS Institute, Cary, NC, USA). The tree was designated as a random factor and year was the repeated measure on each tree. The dates of onset and ending of the different phenological phases were modelled using DOY as dependent variable.

A multifactor ANOVA was also used to assess the treatment effect on soil temperature and volumetric water content. For these two variables, an average per month was calculated and used as the dependent variable. Once again, the tree around which measurements were taken was considered a random factor, while month was the repeated measure. The PPFD was analysed in a similar way, but in this case using the distance from the stem as the repeated measurement.

The total number of tracheids produced over time was described with the Gompertz function defined as:

$$y = A \exp[-e^{(\beta - \kappa t)}] \quad (1)$$

where y is the weekly cumulative sum of tracheids, t is DOY, A is the function asymptote (maximum number of tracheids) and β and κ are the x-axis placement parameter and rate of change (Rossi et al. 2003). Using proc NLIN (SAS Institute), the parameters of the function were estimated by minimizing the sum of squared differences between the total number of tracheids observed and number of tracheids predicted by the model. A multifactor ANOVA with repeated measures was performed to compare the Gompertz parameters between species, treatments, tree parts and years. Before the analyses, data were examined to verify the normality of the distributions and homogeneity of the variances (Quinn and Keough 2002).

Results

Xylem cell production

The amount of tracheids produced by the cambium, summarized by the Gompertz function asymptote (Figure 1), was significantly different over time between control and thinned trees and this effect differed between species (parameter A, Table 2). Indeed, there was an increase in cell production after thinning in treated trees compared with the controls. This change was noticeable in the stem of both species, and in the roots of balsam fir. Black spruce roots did not respond the same way in the year 2013. In general, stem and roots of thinned trees produced more cells than controls in the third year after thinning, except for balsam fir stems in which this difference occurred 1 year earlier. The average number of cells produced by the stem cambium in a growing season was higher in balsam fir than black spruce, with a mean difference of about 20 cells. Root cambium produced fewer cells than stem, with the difference averaging 14 fewer cells for black spruce and 37 for balsam fir. The β parameter of the Gompertz function differed by species and tree part, but was unaffected by the treatment (Table 2). The κ parameter was different between species, between tree parts and the treatment effect differed over time between the tree parts (Table 2).

Cambial activity, cell differentiation and xylem phenology

The date of onset of cell differentiation was significantly different between control and thinned trees, and between parts, species, years, and the interaction between parts and years (Figure 2, Table 3). Differences between years were not due to thinning. In balsam fir, cell enlargement in both stem and roots was observed to occur about 10 days earlier than in black spruce. On average, differentiation in stems also started 10 days before that of roots, in both species. The start of xylem differentiation occurred at the earliest in the stem on DOY 122 (2 May) and at the latest on DOY 169 (18 June) in roots, with averages of DOY 144 for black spruce stems and DOY 136 for balsam fir stems, and delays of 10 days in roots for both species. Results varied substantially among the 4 years studied for both thinned and control trees. In the stem, the 2011 onset occurred later than in the

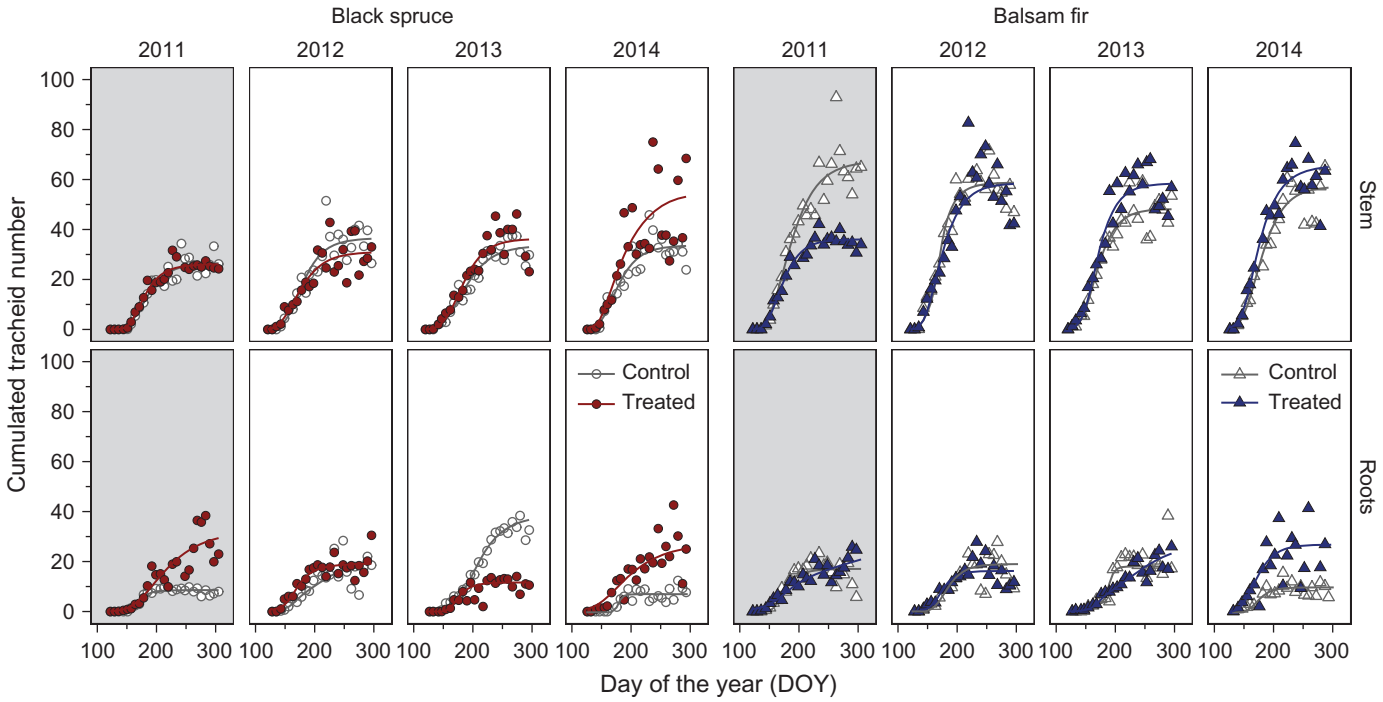

Figure 1. Number of xylem cells over time (circles and triangles) and Gompertz function (lines) for stem and roots of black spruce and balsam fir during the 4 years of the study. Shaded areas mark the year before treatment.

Table 2. ANOVA results of the Gompertz parameters tested in the stem and roots of black spruce and balsam fir during the 4 years of the study (2011–14). Part refers to the tree part where the measurements were taken, i.e., stem or roots. Significant effects ($P < 0.05$) are highlighted in bold.

	A		β		κ	
Source	F	P	F	P	F	P
Treatment	0.00	0.9556	2.74	0.1031	3.41	0.0695
Part	75.47	**<0.0001**	14.33	**0.0004**	13.46	**0.0005**
Treatment × Part	0.50	0.4842	3.13	0.0819	4.27	**0.0430**
Species	0.56	0.4583	4.58	**0.0364**	5.47	**0.0226**
Species × Treatment	0.02	0.8955	0.33	0.5665	0.79	0.3768
Species × Part	15.54	**0.0002**	3.06	0.0855	2.95	0.0910
Species × Treatment × Part	1.12	0.2937	0.06	0.8003	0.28	0.5988
Year	0.97	0.4112	0.20	0.8985	0.11	0.9529
Year × Treatment	2.83	**0.0458**	1.15	0.3366	1.25	0.2991
Year × Part	1.40	0.2507	0.40	0.7556	0.31	0.8159
Year × Treatment × Part	2.66	0.0562	1.73	0.1710	2.78	**0.0486**
Year × Species	0.08	0.9714	0.80	0.5008	0.90	0.4478
Year × Species × Treatment	3.08	**0.0340**	2.08	0.1115	2.09	0.1110
Year × Species × Part	0.29	0.8349	0.84	0.4757	0.98	0.4097
Year × Species × Treatment × Part	0.38	0.7657	1.98	0.1266	1.95	0.1303

other 3 years. For roots, the onset of cell enlargement was highly variable between years in black spruce, while a more regular pattern was observed in balsam fir.

The end of wall thickening and lignification occurred in the roots about 8 days prior to the stem. On average, xylem differentiation was completed on DOY 261 (18 September) in the roots, and on DOY 269 (26 September) in the stem. There was no significant treatment effect on these values.

For both species, the duration of xylogenesis was 18 days shorter in roots than in the stem, but this difference varied significantly between species and years. Xylogenesis was about 7 days shorter in spruce than fir. No influence of the treatment was detected.

The cell production rate was significantly affected by the interaction between treatment, year and tree parts (Figure 2, Table 3). The cell production rate was generally lower in treated trees prior to treatment (i.e., 2011), except in the roots of black spruce. By the second or third year after thinning, rates became significantly higher in treated trees. In the third year after treatment, more cells per day were produced in the stem (0.52 for black spruce, 0.64 for balsam fir) than in the roots (0.27 for both species).

Effect of thinning on microclimate

A significant treatment effect was observed in the percentage of above-canopy PPFD. The canopy opening created by the thinning was favourable for treated trees, which benefited from significantly more light after treatment than control trees (Figure 3, Table 4). On average, about 30% of the total PPFD reached the soil around the treated spruces, compared with 16% in the control. In balsam fir, most of the total available light was intercepted by the canopy, as only 8% of the PPFD reached the ground in treated stands compared with 4.5% for control. Close to the stem, black spruce and balsam fir trees had up to 22% and 6.5% more light available, respectively, than the control.

Soil temperature varied from −6 to 17 °C, being mostly around 0 °C between December and April, and close to 15 °C in July and August. After the removal of competitors around the treated trees, soil temperature was significantly affected over time, but also differed according to the species that dominated the forest cover (Table 4, Figure 3). The biggest differences in the monthly averages of soil temperature between treated and control trees occurred in the months of May and June for balsam fir (up to 2 °C warmer for treated trees), and later for black spruce, in July and August (up to 1 °C warmer for treated trees). From May to October, soil volumetric water content varied considerably (Figure 3). This variable showed differences between species, which were growing in two different sites, but was not significantly affected by the thinning (Table 4).

Discussion

Xylem cell production

Several authors working with different species have studied the growth response of residual trees to thinning, with the most common result being a radial growth increase in the stem (Peltola et al. 2007, Olivar et al. 2014, Pamerleau-Couture et al. 2015, Montoro Girona et al. 2016) and roots (Ruel et al. 2003, Vincent et al. 2009, Krause et al. 2014). The results in this study confirmed these patterns, with thinning inducing a significant increase over time on the number of cells produced by the cambium. Stem and roots of trees subjected to thinning produced more cells by the third year after treatment, or the second year in the stem of balsam fir. This faster reaction of balsam fir has also

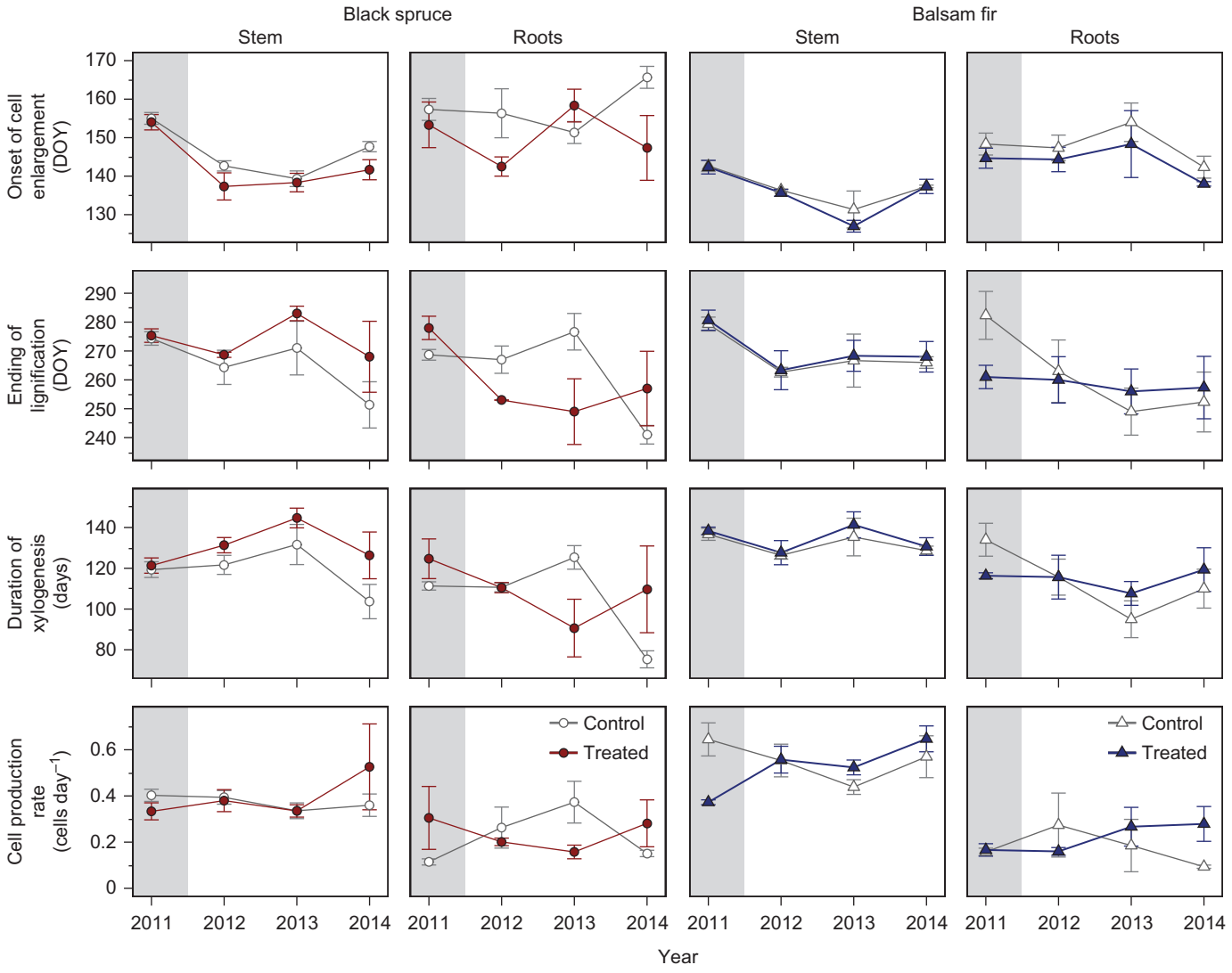

Figure 2. Xylem phenology and cell production rate in stem and roots of black spruce and balsam fir during the 4 years of the study. Shaded areas mark the year before thinning.

been observed in other studies (Doucet and Blais 2000, Lemay et al. 2016). However, contrarily to what was previously shown by Vincent et al. (2009) for black spruce and by Ruel et al. (2003) for balsam fir, roots did not react more rapidly or more strongly than stems. One of the reasons for this difference may be that the root sampling in our study was conducted on only one root per tree. The variation in the response of roots from the same tree to thinning could therefore not be distinguished from the tree-to-tree variation, which probably affected the sensitivity of our analysis. Indeed, black spruce and balsam fir stems produced more cells in general, with all trees presenting a similar response, whereas in the roots, more variability in the growth response was observed between the different trees. This large variation might explain the higher number of cells observed in control roots of black spruce in 2013. A high variability in root growth is common (Fayle 1968, Krause and Eckstein 1993, Drexhage et al. 1999), as root radial growth is more irregular than in the stem, mainly because of the differences in soil conditions and the many functions of the different root parts (Fayle 1968). Furthermore, the experimental thinning applied in this study was of relatively low intensity. Unlike thinnings conducted in commercial conditions, our treatment created only small gaps in the forest cover due to the removal of competitors only around the sample trees. Carlson and Groot (1997) showed that small 9-m diameter circular openings and intact forest have the same microclimate conditions. Our observations are similar with slightly smaller canopy openings. This suggests that our experimental trees were subjected to conditions comparable to those of the control trees, and were probably not exposed to as much wind stress as in a high intensity thinning. Thus, it seems that the thinned trees may not have needed to invest more in root growth to improve their ground anchorage, as Krause et al. (2014) observed after commercial thinning.

Table 3. ANOVA results of the onset, ending, duration and rate of the wood formation process tested in the stem and roots of black spruce and balsam fir during the 4 years of the study (2011–14). Significant effects ($P < 0.05$) are highlighted in bold.

	Onset of cell enlargement		Ending of wall thickening and lignification		Duration of xylogenesis		Cell production rate	
Source	*F*	*P*	*F*	*P*	*F*	*P*	*F*	*P*
Treatment	9.84	**0.0026**	0.07	0.7856	2.79	0.0998	0.61	0.4371
Part	55.70	**<0.0001**	12.03	**0.0010**	42.09	**<0.0001**	95.48	**<0.0001**
Treatment × Part	1.74	0.1914	2.88	0.0947	0.83	0.3658	0.31	0.5785
Species	40.47	**<0.0001**	0.07	0.7983	7.11	**0.0098**	1.58	0.2128
Species × Treatment	0.99	0.3228	0.37	0.5478	0.98	0.3250	0.01	0.9373
Species × Part	0.00	0.9549	0.04	0.8367	0.04	0.8342	13.82	**0.0004**
Species × Treatment × Part	0.05	0.8275	0.64	0.4250	0.38	0.5402	0.58	0.4474
Year	5.91	**0.0013**	8.46	**<0.0001**	3.36	**0.0242**	0.81	0.4933
Year × Treatment	1.27	0.2917	1.52	0.2188	2.23	0.0932	2.99	**0.0377**
Year × Part	7.43	**0.0002**	1.03	0.3856	3.97	**0.0118**	1.75	0.1664
Year × Treatment × Part	0.94	0.4259	0.59	0.6246	1.20	0.3179	2.97	**0.0387**
Year × Species	1.98	0.1255	1.93	0.1347	2.94	**0.0399**	0.08	0.9727
Year × Species × Treatment	2.52	0.0664	1.73	0.1709	2.79	0.0997	2.45	0.0717
Year × Species × Part	1.63	0.1920	0.23	0.8768	0.57	0.6344	0.00	0.9997
Year × Species × Treatment × Part	0.58	0.6272	2.46	0.0706	2.51	0.0666	0.37	0.7714

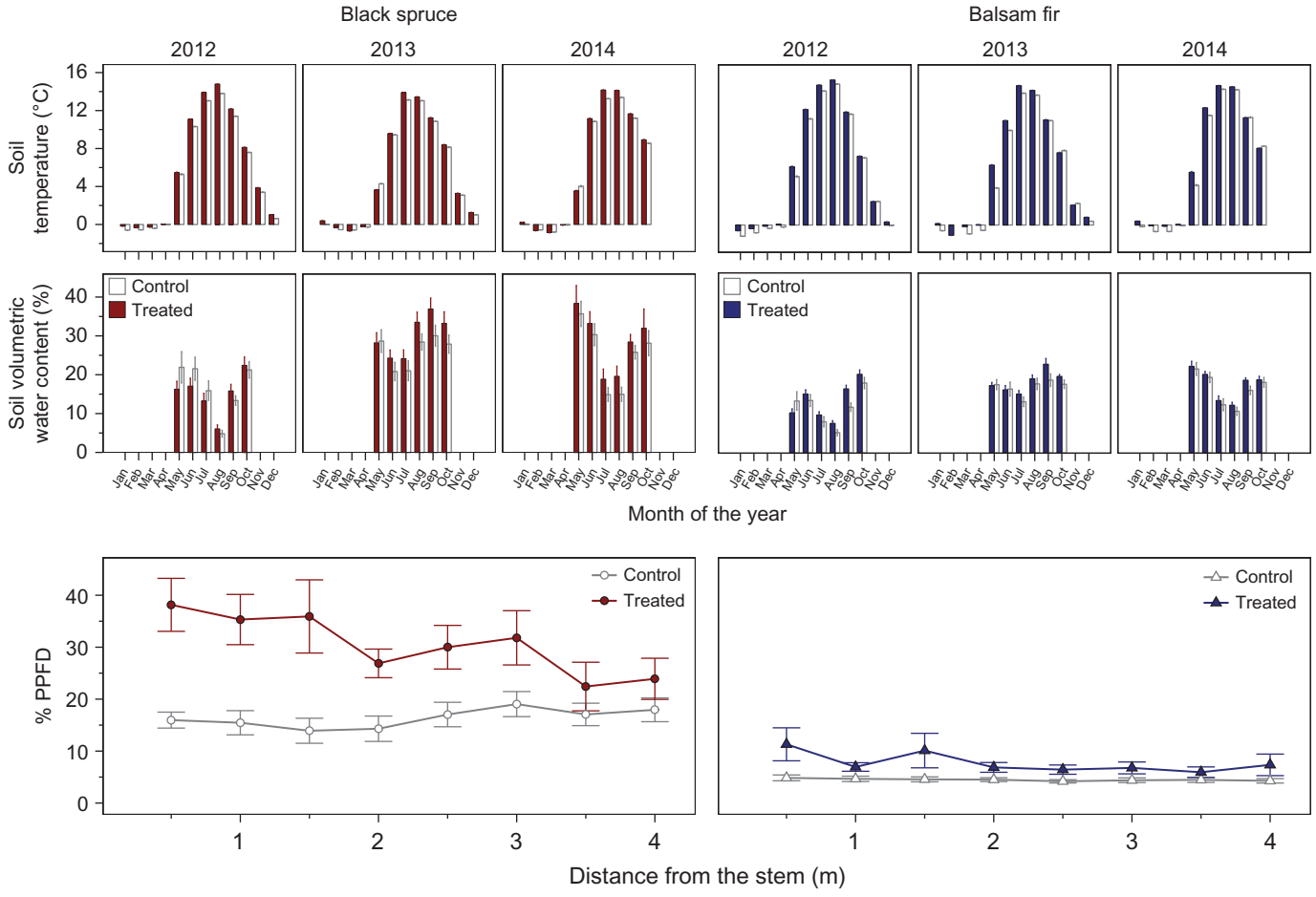

Figure 3. Soil temperature and volumetric water content monthly averages in the 3 years following the experimental thinning, and percentage of above-canopy PPFD measured once in 2014.

Table 4. ANOVA results of the environmental variables tested in black spruce and balsam fir during the 4 years of the study (2011–14). Distance refers to the distance from the stem where the measurements of the percentage of above-canopy PPFD were taken. Significant effects ($P < 0.05$) are highlighted in bold.

	Soil temperature (°C)		Soil volumetric water content (%)		% PPFD	
Source	*F*	*P*	*F*	*P*	*F*	*P*
Treatment	67.33	**<0.0001**	3.59	0.0596	51.54	**<0.0001**
Species	33.71	**<0.0001**	15.46	**0.0001**	203.72	**<0.0001**
Treatment × Species	0.03	0.8731	0.19	0.6610	20.49	**<0.0001**
Month	1404.64	**<0.0001**	8.19	**<0.0001**		
Month × Treatment	1.44	0.2125	0.45	0.8108		
Month × Species	10.58	**<0.0001**	0.57	0.7210		
Month × Species × Treatment	5.96	**<0.0001**	0.04	0.9992		
Distance					1.26	0.2847
Distance × Treatment					1.62	0.1464
Distance × Species					0.53	0.8110
Distance × Species × Treatment					0.79	0.5957

Cambial activity, cell differentiation and xylem phenology

Conflicting results emerge when looking at the effects of thinning on xylogenesis. Some studies observed a prolonged growing season after thinning (Linares et al. 2009, van der Maaten 2013), while another found that the beginning, ending and duration of cambial activity were little affected (Wodzicki 2001). In our study, the dates of onset and ending and the duration of wood formation remained unaffected by thinning. There was also no change observed in the timing of cell enlargement or wall thickening and lignification. Instead, a significant treatment effect was observed over time on the cell production rate. Studying the effects of thinning in *Pinus sylvestris*, Wodzicki (2001) also observed that the daily rate of tracheid production had increased after treatment. Similarly, the maximum radial increment rates and the final tree-ring width were affected after thinning treatments in *Abies pinsapo* (Linares et al. 2009). Boivin-Dompierre et al. (2017) showed that after thinning in spruce-fir stands in eastern Canada, trees increased their wood production per unit leaf area in response to higher light availability. Our findings suggest that thinning in this forest type affects wood formation mostly by acting on the efficiency of cambial cell division, as treatment affected the number of cells produced mainly by an acceleration in the accumulation rate of new cells, and not by a change in the duration of cell production. As a result, this increase of the cell production rate in treated trees was responsible for the increase in the total number of tracheids produced by the cambium at the end of the growing season.

Effect of thinning on microclimate

In this part of the eastern boreal forest of Canada, precipitation events are frequent, and the low temperatures limit evaporation, which implies that water availability is not an important limiting factor for growth. We can deduce that the thinning in our study did not lead to a sufficient change in the environmental conditions around the sample trees to modify the soil water availability. Goudiaby et al. (2011) also noticed that soil water content was not affected by thinning, and that light availability increased according to the basal area removed. The significantly higher light available after treatment in our study supports the hypothesis that an increase in light intensity as a consequence of thinning is more important for plant growth than a potential increase in air or soil temperature (MacDonald 2000). In our study, soil temperature was statistically higher for treated trees between June and September, but the differences remained marginal. Previous studies on mature trees showed no change in xylem phenology and cell production after a 4 °C soil warming for 6 years (Lupi et al. 2012*a*, Dao et al. 2015) and no change in bud burst after a 5 °C soil warming (Bergh and Linder 1999).

Comparison between species and tree parts

Wood formation was initiated in black spruce about a week later than balsam fir while the end of cell differentiation occurred at about the same time. The fact that bud break occurs earlier in fir than spruce could explain why balsam fir had an earlier xylem differentiation onset because these events have been found to be synchronous in both black spruce and balsam fir (Antonucci et al. 2015).

In the 4 years of the study, the onset of cell enlargement in the roots was synchronized with the stem or occurred a few days later. This result is similar to what Thibeault-Martel et al. (2008) observed with the same species. The onset of wall thickening also occurred earlier in the stem than in roots, and the ending of xylem differentiation happened earlier in the roots or was similar. Accordingly, total duration of xylogenesis and duration of wall thickening and lignification were shorter in the roots, with no difference between treatments. The warming of the soil in spring, which occurs later than air temperature warming, could explain the different dynamics between the stem and roots (Thibeault-Martel et al. 2008). Our experimental thinning did induce

changes in soil temperatures but the magnitude of the effect was likely insufficient to lead to a measurable effect on xylogenesis in roots during the short monitoring period of the study.

Lupi et al. (2010) showed that a higher number of cells produced by the cambium was related to an earlier onset of cell differentiation, which in turn could have influenced the ending of cell differentiation. The synchronized or delayed onset that we observed in the roots can thus explain the fewer cells produced in the roots compared with the stem. This result contradicts other studies that observed larger ring widths in roots than stem after thinning (Ruel et al. 2003, Krause et al. 2014). However, these previous studies did not measure the number of cells constituting the rings. It is possible that the growth rings in our study were in fact similar to or wider than in the stem, as observed in other studies. We observed that fewer cells were produced by the root cambium after thinning, but the duration of the cell enlargement and wall thickening and lignification phases were the same as in the stem. Therefore, individual root cells may have on average spent a little more time in these phases compared with a cell produced in the stem. A longer differentiation period for root cells could in turn result in larger cross-sectional areas, wider cell walls and higher lignin content after thinning. Indeed, cell wall thickness of a mature tracheid was observed to be correlated with the duration of its maturation phase (Wodzicki 2001). Larger lumen and thicker walls in the root cells produced after thinning would allow a more efficient water transport to counterbalance the higher transpiration rate of the trees (Gebhardt et al. 2014, Boczoń et al. 2016). Further analyses should be conducted, especially on the roots, to understand the effect of thinning on water transport, and thus improve our understanding of the below ground part of the trees.

Conclusions

The experimental thinning applied in this study had an effect on the cell production rate of the stem and roots, which was significantly increased over time after the treatment. This higher growth rate triggered an increase in the total number of cells produced by the cambium, confirming our first hypothesis. However, no influence of thinning was observed on the timing or duration of xylogenesis, which rejects our second hypothesis. And contrary to our third hypothesis, roots were not more affected by the treatment than the stem as roots produced fewer cells than the stem. Thinning applied in such conditions can be considered a useful treatment to increase wood production in stem and roots, without exposing trees to drastic changes in environmental conditions. It is therefore unlikely that thinning would lead to increased risks of frost damage in the spring or autumn with a longer or shifted growing season. To our knowledge, this is the first study in which xylogenesis was monitored weekly after thinning. It is also one of the rare studies on root xylogenesis. Our results provide a better understanding of the effect of this silvicultural treatment on the process of xylem formation, and thus a better understanding of tree physiology and its potential effects on wood properties.

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Conflict of interest

None declared.

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