Forest Ecology and Management 328 (2014) 292–299

Contents lists available at ScienceDirect

Forest Ecology and Management

journal homepage: www.elsevier.com/locate/foreco

Non-additive effects of mixing hybrid poplar and white spruce on aboveground and soil carbon storage in boreal plantations

M. Chomel [a,b,*], A. DesRochers [a], V. Baldy [b], M. Larchevêque [c], T. Gauquelin [b]

[a] Université du Québec en Abitibi-Temiscamingue, Institut de Recherche sur les forêts, 341 rue principale Nord, Amos, Québec J9T 2L8, Canada
[b] Institut Méditerranéen de Biodiversité et d'Ecologie, Aix Marseille Université, CNRS, IRD, Avignon Université, IMBE UMR 7263, 13331 Marseille Cedex 03, France
[c] Université du Québec en Abitibi-Temiscamingue – Institut de Recherche en mines et en environnement, 341 rue principale Nord, Amos, Québec J9T 2L8, Canada

ARTICLE INFO

Article history:
Received 14 March 2014
Received in revised form 25 May 2014
Accepted 28 May 2014

Keywords:
Boreal forest
Carbon storage
Mixed-species plantations
Productivity
Soil horizon
Synergistic effect

ABSTRACT

The use of trees under intensive management is particularly important for rapid fiber production in boreal regions. Mixed-species plantations using species that have complementary ecological niches, such as hybrid poplar and white spruce, potentially can maximize the use of resources and, consequently, increase productivity. In the context of climate change, vegetation and soil carbon sequestration is of a particular interest as part of a possible means of compensating for CO_2 emissions. Since higher productivity leads to higher CO_2 sequestration, the use of mixed-species plantations could improve the ecological service of carbon storage compared to mono-specific plantations. We compared above-ground and soil C storage of nine-year-old mono-specific plantations of white spruce and hybrid poplar with mixed plantations of these two species. Soil carbon was evaluated by separately sampling four soil horizons, while aboveground carbon was assessed from tree biomass estimates using allometric relationships. Mixing white spruce and hybrid poplar exerted a substantial synergistic effect on above-ground and soil carbon storage. This positive effect was due to greater productivity of poplar (47% of biomass increase) and great accumulation of litter in soil surface horizons (52% L-horizon carbon increase) of mixed-species compared to mono-specific plantations. These results imply that in addition to wood production gains by poplar trees, mixed-species plantations of hybrid poplar and white spruce promotes greater carbon sequestration than mono-specific plantations of either hybrid poplar or white spruce, an important aspect of forest ecosystem services.

1. Introduction

In 2010, the total area of planted forest was estimated to cover 264 million hectares worldwide (FAO, 2010). Although they constituted only 7% of global forest cover (FAO, 2010), these plantations were estimated to supplied about 35% of global roundwood needs (Shvidenko et al., 2005). The use of trees under intensive management is particularly important for rapid fiber production in boreal regions of Canada, where growth rates of natural forests are relatively low (Pothier and Savard, 1998). Within this biome, short-rotation forestry has great potential for supporting ecosystem services in (1) valuing abandoned agricultural lands and degraded forests, (2) reducing harvesting pressure on natural forests (FAO, 2010), (3) becoming sustainable sources of wood supplies, and (4) promoting carbon storage (Kelty, 2006). Many researchers have focused upon vegetation and soil carbon sequestration in natural or planted ecosystems, as a possible means of compensating for CO_2 emissions, which is particularly important in the context of climate change (IPCC, 2007). Soil carbon storage could represent from 50% of total carbon storage in tropical forests to 98% in cropland systems; boreal forests have an intermediate level, with soil organic carbon concentrations corresponding to 84% of total carbon storage at the ecosystem level (Bolin et al., 2000). In this context, maximizing the potential for carbon storage by tree plantations becomes an interesting proposition for increasing compensation for or offsetting increasing CO_2 emissions. For example, afforestation of crop fields and pastures of central Saskatchewan with trembling aspen (*Populus tremuloides* Michaux) was shown to have the potential to sequester 30–75 Mg ha^{-1} of carbon over the next 50–100 years (Fitzsimmons et al., 2004). Forest management has traditionally relied upon mono-specific plantations, which are easier to establish, tend and harvest compared to mixed-species plantations. The former have been criticized for having poor ecological characteristics (Lamb et al., 2005;

---

* Corresponding author. Present address: Institut Méditerranéen de Biodiversité et d'Ecologie marine et continentale (IMBE), Aix Marseille Université, CNRS, IRD, Avignon Université, 13331 Marseille Cedex 03, France. Tel.: +33 413551233; fax: +33 413551151.

E-mail address: mathilde.chomel@imbe.fr (M. Chomel).

http://dx.doi.org/10.1016/j.foreco.2014.05.048
0378-1127/

Erskine et al., 2006) and greater risks for the spread of diseases that are incurred by fungal pathogens (Burdon, 2001). In contrast, mixedwoods may have many advantages over pure stands such as higher productivity (Man and Lieffers, 1999; Johansson, 2003) and greater resistance to abiotic and biotic stresses, including damage caused by pests or fungal pathogens (McCracken and Dawson, 1997; Burdon, 2001). However, the productivity benefits that are derived from mixed stands depend upon species composition, because such benefits are not consistently observed in studies of mixture effects (Rothe and Binkley, 2001; Piotto, 2008). Mixed-species stands can be more productive than mono-specific stands through two mechanisms: facilitation between species, i.e., one species improves environmental conditions and, thereafter, the growth of another; or niche segregation, where there is divergence in the use of resources between species with different functional traits, which leads to decreased competition and a better efficiency in using local resources (Vandermeer, 1989).

Mixedwood forests of trembling aspen and white spruce (*Picea glauca* [Moench] Voss) are common across boreal Canada. These two species have complementary ecological niches (i.e., they exhibit niche segregation) resulting in maximal use of resources (Kelty, 1992; Man and Lieffers, 1999; Kelty, 2006): white spruce is a slow-growing, superficially rooted and moderate shade-tolerant species, while aspen (like hybrid poplar) is a fast-growing, more deeply rooted and shade-intolerant species. Due to this complementarity, boreal mixedwoods could be more productive than single-species forest ecosystems (Chen and Popadiouk, 2002). Yet this hypothesis has not always been confirmed. In natural forests, some studies have found greater productivity of mixed compared to pure stands (Martin et al., 2005), with a positive effect of aspen (if less than 41% of total stand basal area) on spruce growth in mixtures (Légaré et al., 2004). Others have found negative effects of spruce on aspen productivity (MacPherson et al., 2001), negative effects of aspen on spruce productivity (Kabzems et al., 2007), or no effect of mixed compared to mono-specific stands (Cavard et al., 2010). In plantations, at least one previous study found positive effects of mixing hybrid poplar (*P. maximowiczii* × *balsamifera clone*) and white spruce in intimate mixtures on the growth of the two species (Benomar et al., 2013). Since greater tree productivity leads to greater CO_2 sequestration, the use of mixed species plantations could improve the ecological service of aboveground carbon storage compared to mono-specific plantations. Furthermore, increases in forest productivity can also increase litter production and litterfall (Rothe and Binkley, 2001; Liu et al., 2005), leading to greater accumulation of organic matter on the forest floor (Sayer et al., 2011; Leff et al., 2012), which may result in an increase in soil carbon storage. Soil carbon storage depends upon the balance between C input rates, i.e., senescent organic matter (branches, leaves, and roots), and output rates, i.e., the decomposition of this organic matter. Some studies found a positive effect of mixing litters on decomposition rates; however, like the effects of mixtures on tree productivity, mixture effects on litter decomposition are also largely dependent upon the particular species that are present in the mixture (Gartner and Cardon, 2004; Hättenschwiler et al., 2005). Needle litter of conifers is often acidic, complex in terms of its chemistry, and generally less palatable for soil decomposers compared with the leaf litter shed by broadleaf deciduous species. In boreal mixedwoods, aspen improved litter decomposition relative to spruce through an increase of soil organism abundance, together with an improvement in litter quality and soil physical and chemical properties (Légaré et al., 2005; Laganière et al., 2009). Consequently, aspen forests store less soil carbon than black spruce forests (*Picea mariana* [BSP] Miller), given the faster rates of decomposition processes in the former compared to the latter (Gower et al., 2000; Vance and Chapin, 2001; Laganière et al., 2011).

In tree plantations, studies have generally focused on tree growth and productivity to determine the best management practices that promote higher timber yield, whereas soil carbon storage is largely less thoroughly investigated. This paucity of information contrasts with studies that have been conducted in natural forest environments (Johnson, 1992). In this paper, we compared above-ground and soil carbon storage in nine-year-old mono-specific plantations of white spruce and hybrid poplar *versus* mixed plantations of these two species. For this purpose, we examined hybrid poplar and white spruce growth, together with humus morphology, in the different planted plots. Quantities of soil carbon were estimated by separately sampling four soil horizons, whereas the quantity of aboveground carbon was assessed from tree biomass, which was calculated using allometric relationships.

We hypothesized the following: (i) Based on the low resource quality of spruce needles and slow decomposition rates in natural spruce forests, we expected that carbon storage would be greater in surface soil horizons of mono-specific spruce plantations compared to mono-specific hybrid poplar and mixed plantations. (ii) Due to an increase in productivity, we expected carbon storage in aerial biomass to be higher in mixed plantations compared to mono-specific plantations. (iii) Given a potentially positive effect of mixing species on organic matter decomposition rates compared to mono-specific plots, we expected lower carbon storage within the soil surface horizons (non-additive effect).

2. Materials and methods

2.1. Study area

The study was located in the boreal region of Abitibi-Témiscamingue, Quebec, Canada. Three sites were selected for study: Amos (48°36′N, 78°04′W), Rivière Héva (48°11′N, 78°16′W), and Nédelec (47°45′N, 79°22′W). The Amos site was abandoned farmland with a heavy clay soil that was dominated by grasses and sparse patches of speckled alder (*Alnus incana* [L.] Moench ssp. *rugosa* [Du Roi] R.T. Clausen), willow (*Salix* spp.), and trembling aspen. Rivière Héva was an abandoned farmland site with heavy clay soil, which was also dominated by shrubs, including patches of alder, willow, and aspen. Nédelec had been previously dominated by trembling aspen forest, which was commercially harvested in 2000. In addition to aspen, the main species that were present included white or paper birch (*Betula papyrifera* Marshall) and pin cherry (*Prunus pensylvanica* L.f.), which were growing on soil with a sandy loam texture. Soil type of the three sites ranged from a Brunisol with a Bm-layer to a grey Luvisol with a Bt-layer or Gleysol (Soil Classification Working Group, 1998). Based on a 30-year running climate average (1970–2000), Amos and Rivière Héva annually receive an average of 918 mm $year^{-1}$ (Amos station) and have a mean temperature of 1.2 °C, while Nédelec has mean precipitation of 916 mm $year^{-1}$ and a mean temperature of 1.9 °C (Remigny station, Environment Canada 2014). Site preparation before planting was conducted in 2002. A bulldozer was used to remove tree stumps at Nédelec, while shrubby vegetation at Rivière Héva was removed using a brush shredder mounted on a farm tractor. At Amos, scattered tree stumps and shrub clumps were removed using chains and a farm tractor. Sites were then ploughed to a depth of about 30 cm, followed by disking in spring 2003 to level the soil surface and remove most woody debris (Benomar et al., 2011). The plantations were established in 2003, using one hybrid poplar clone (*Populus maximowiczii* A. Henry × *P. balsamifera* L., clone MB915319), and an improved white spruce family from a provincial seed orchard. These two species were planted in mono-specific plots of 36 trees (6 × 6 trees) with 1 × 1 m spacing, and in mixed species plots, where rows of spruce alternated with rows of poplar, which was also planted under a spacing of 1 × 1 m. Spacing corresponded to a tree density of 10,000 stems ha^{-1} (Fig. 1). Each experimental unit thus contained 36 trees, of which only the 16 interior trees were considered for the study. This left a 1 row-wide buffer along each plot edge. The experiment was designed as a split-split-plot layout, with replicate sites as the whole-plot factor, and plantation type as the subplot factor. Soil horizon or tree parts were a sub-sub-plot factor nested in plantation type. Weed management was done during the first five years after planting, using a manual rototiller between rows and trees.

2.2. Aboveground tree biomass and carbon storage

Height, stem basal diameter (at 10 cm above the soil surface) and diameter at breast height (DBH, 1.3 m) were measured on the 16 interior trees at the end of the ninth growing season (mid-October 2011). Above-ground biomass of stems, branches and needles of the spruces were estimated from allometric equations that related biomass to basal diameter (D10), according to Pitt and Bell (2004). Above-ground biomass of stems, branches and leaves of the poplars were estimated from allometric equations that related biomass to DBH, based on Benomar et al. (2012). According to these allometric equations, the relationship between DBH and the biomass of stems, branches or leaves/needles was a power function model, and data were fitted to the following equation:

$$W = a\,\mathrm{DBH}^b$$

where W is the biomass of stems, or branches, or leaves (kg dry mass), DBH is the diameter at breast height (cm), and a and b are parameters that are estimated from the model. At the plot level, aboveground biomass (Mg ha^{-1} of dry mass) was estimated by multiplying aboveground tree biomass by tree density at planting.

For aboveground carbon storage (Mg ha^{-1}), we assumed that the organic matter contains 50% carbon (Lieth, 1975). The quantity of carbon per tree that had been obtained from biomass measurements was multiplied by tree density (stems per hectare). For mixed plantations, we performed the same calculation, but spruce and poplar densities were each 5000 stems ha^{-1}.

2.3. Litterfall

Annual litterfall was assessed using litter traps. Spruce branches were about 20 cm above the ground, while poplar branches were more than 1 m off the ground surface; the littertraps were adapted to the structure and height of each tree species. Each trap consisted of a wooden frame measuring 40 × 60 cm (corresponding to 0.24 m^2) for the poplar leaf harvest, or 30 × 50 cm (corresponding to 0.15 m^2) for the spruce needle harvest. The traps were each supported by four legs that were 40 and 20 cm high, respectively. Steel 2-cm mesh screening was placed on the sides and bottom of the frames for poplars. Nylon screening (2-mm mesh), which was covered with a permeable fabric to prevent needle loss, was attached to the sides and bottom of the wooden frames for spruce. Four traps were placed within each mono-specific plot, and eight traps were placed within the mixed-species plots (4 traps under poplar, and 4 under spruce trees), for a total of 48 traps. The traps were installed in October 2011. Litterfall was collected weekly during poplar leaf fall, and once a month otherwise, from October 2011 to October 2012. After collection of litterfall, the litter was oven-dried at 65 °C to constant mass and weighed.

2.4. Field procedures and carbon concentrations

The soil samples were divided into four layers according to the Canadian system of soil classification (Agriculture Canada Expert Committee on Soil Survey, 1987): two organic layers, i.e., L (fresh litter) and F (fermentation layer); one organo-mineral layer that had been disturbed by plowing (Ap); and one mineral layer (Bm or Bt). Sampling took place within the center space located among 4 trees, and with 5 replicates being taken in each plot (spruce, poplar, and mixed). Soil sampling from L, F and Ap horizons was performed using a wooden frame (20 cm × 20 cm). For the B horizon, sampling was done with a steel cylinder (6 cm deep, 170 cm^3). Only the first 6 cm were sampled because we considered that possible changes in carbon concentration in this horizon mainly involved the uppermost few centimeters of soil, due to the young age of the plantations. For this reason, we further assumed that for this study, the B horizon was only 6 cm thick. Ap-layer materials were sieved to pass a 2-mm mesh, and sieve residues (i.e. leaves, needles, twigs, bark, seeds, and cones) were added to the F-layer, while live roots were removed and discarded. In total, 180 samples were oven-dried at 60 °C to constant mass, and weighed to determine their dry mass.

Soil samples were finely ground with a ball mill (MM301, Retsch Inc., Newtown, PA), and carbon concentration were determined with a C/N elemental analyser (Flash EA 1112 series, ThermoScientific, Rodano, Italy). To determine the bulk density of the B-horizon in each plantation, B-layer mass was divided by the volume of the steel cylinder (170 cm^3). To determine soil carbon storage (Mg ha^{-1}), the mass of each soil horizon was multiplied by its carbon concentration, and the values were scaled to one hectare from the surface area of the wooden sampling frame.

2.5. Data analyses

Mean values (soil carbon concentrations, litterfall, tree aboveground biomass and C storage) were compared among plantation types and soil layers, or tree aerial parts for aboveground biomass, with hierarchical linear mixed-effects models using the *lme* function in the *nlme* package (Pinheiro et al., 2014) of R (Version 2.15.1, R Development Core Team 2008). Site replicates were treated as random effects, and plantation type was nested in site

Fig. 1. Photographs of the three plantation types at the Nédelec site. Spruce, mixed-species and poplar plantations are depicted from left to right.

replicates to reflect the structure of our data set (split-split-plot design).

To better meet the assumptions of normality and homoskedasticity, the data for carbon storage were ln-transformed. Means were separated using Tukey's multiple comparison tests (differences are noted thereafter as, for example, $a < b < c < d$). The significance threshold was set at $\alpha = 0.05$. For further evaluation of mixed plantation effects on carbon storage, we calculated the relative effects of mixing species by comparing the observed values with the predicted values of carbon storage, based on the respective mono-specific plantation treatments. Predicted values for the mixed plantation were estimated by averaging carbon storage of the component species that had been planted in mono-specific plots in the site-specific replicates. According to Wardle et al. (1997), the relative mixture effect can be calculated as the ratio: [(observed – predicted)/predicted]* 100. If this ratio differs from zero, it would indicate non-additive effects of mixing species on carbon storage. Negative and positive deviations from zero are referred to as antagonistic and synergistic effects, respectively. To test if the observed vs predicted ratios of carbon storage in mixed plantations differed significantly from zero, we used one-sample Student's *t*-tests with 95% confidence intervals.

3. Results

3.1. Tree growth and litter productivity

Basal stem diameter, DBH and total height of poplar trees were greater in mixed compared to mono-specific plantations ($P < 0.01$), but litterfall was similar among plantation types ($P = 0.17$) (Table 1). In contrast, basal diameter and DBH of spruce trees were smaller in mixed compared to mono-specific plantations ($P < 0.001$ and $P = 0.024$, respectively). However, total height ($P = 0.30$) and litterfall ($P = 0.70$) of spruce trees was not affected by plantation type (Table 1).

Aboveground biomass of spruce was 38% lower in mixed plots than pure plots. In contrast, poplar biomass was 47% greater in mixed plots than in pure plots ($P < 0.001$; Table 1).

3.2. Carbon concentration of soil horizons and bulk density

Carbon concentration of each horizon is reported in Table 2 (3 plantation types combined, as there were no differences among plantation type), and differed significantly among soil layers from the superficial horizons to the mineral soil (results of the linear mixed model are reported in Table 3). Bulk density (mean ± standard error) of the B-horizon was 0.86 g cm^{3} ± 0.04 g cm^{3}, 0.91 g cm^{3} ± 0.04 g cm^{3}, and 0.87 g cm^{3} ± 0.02 g cm^{3} for spruce, poplar and mixed plantations respectively, but did not significantly differ among plantation types (linear mixed model, numDF = 2, denDF = 4, F = 0.43, $P = 0.68$).

Table 2
Mean (±standard error) values of C (%) in the soil horizons ($N = 144$).

Horizons	C (%)		
	Spruce	Poplar	Mixed
L	43.2 ± 0.8 (d)	43.2 ± 1.3 (d)	43.6 ± 1.3 (d)
F	27.5 ± 2.6 (c)	25.6 ± 3.3 (c)	29.5 ± 2.6 (c)
Ap	9.8 ± 0.8 (b)	7.9 ± 0.7 (ab)	9.7 ± 0.7 (b)
B	4.0 ± 0.3 (a)	4.0 ± 0.3 (a)	4.5 ± 0.2 (a)

Note: Different letters within %C across horizon represent a significant difference between means according to Tukey test.

3.3. Carbon storage

3.3.1. Soil carbon storage

Soil C storage across the whole soil profile was similar among plantation types. However, when we decomposed carbon storage into each of the four soil layers, differences between plantations emerged for the three uppermost horizons (Table 3). In the L horizon, carbon storage was greater in mixed (2.67 Mg ha^{-1} of carbon) compared to mono-specific (1.40 Mg ha^{-1} of C for spruce and 1.30 Mg ha^{-1} of C for poplar) plots. In the F horizon, carbon storage was greater in spruce (2.46 Mg ha^{-1} of carbon) than in poplar and mixed (1.17 and 1.45 Mg ha^{-1} of carbon, respectively) plots. Finally, for the Ap horizon, we observed greater carbon storage in mono-specific poplar and mixed-species (3.44 and 3.01 Mg ha^{-1} of carbon, respectively) than in mono-specific spruce (1.98 Mg ha^{-1} of carbon) plots (Table 3 and Fig. 2).

3.3.2. Aboveground carbon storage

The effect of plantation type on aboveground carbon storage depended upon the tree part that was being considered. When we examined stems or branches, carbon storage was greater in mono-specific poplar (15.25 and 14.66 Mg ha^{-1} of carbon, respectively) and mixed-species (17.40 and 15 Mg ha^{-1} of carbon, respectively) plots, compared with mono-specific spruce plots (4.40 and 4.19 Mg ha^{-1} of carbon, respectively). However, carbon storage did not significantly differ among plantation types for leaves and needles (Fig. 2 and Table 3). With respect to carbon storage distribution, carbon storage was greater in leaves in spruce plantations, whereas it was greater in stems and branches in poplar and mixed plantations (Fig. 2).

3.3.3. Sum of above and soil carbon storage

Total carbon storage (above-ground + soil carbon storage) was lower in spruce plantations (42 Mg ha^{-1} of carbon) compared to poplar and mixed-species plantations (63 and 69 Mg ha^{-1} of carbon respectively; linear mixed model: poplar plantation, $P = 0.02$; mixed plantation, $P = 0.01$). With respect to the comparison between above-ground *versus* soil carbon storage, spruce stands stored less carbon in the aboveground compartment (on average only 37%), compared to poplar and mixed-species stands

Table 1
Mean (±standard error) height, basal stem diameter, diameter at breast height (DBH) and litterfall of hybrid poplar and spruce growing in mono-specific and mixed plantations.

Species	N	Spruce		Poplar	
		Pure	Mixed	Pure	Mixed
Tree height (m)	118	2.66 ± 0.09 (a)	2.49 ± 0.14 (a)	7.73 ± 0.29 (a)	9.33 ± 0.26 (b)
Basal stem diameter (cm)	118	5.12 ± 0.17 (b)	4.19 ± 0.20 (a)	7.38 ± 0.27 (a)	10.18 ± 0.36 (b)
DBH (cm)	118	2.45 ± 0.14 (b)	1.92 ± 0.17 (a)	5.33 ± 0.26 (a)	7.75 ± 0.35 (b)
Biomass (kg tree^{-1})	118	3.22 ± 0.26 (b)	2.00 ± 0.22 (a)	7.02 ± 0.56 (a)	13.19 ± 0.97 (b)
Litterfall (Mg ha^{-1} year^{-1}·)	48	0.50 ± 0.10 (a)	0.37 ± 0.09 (a)	3.79 ± 0.30 (a)	4.02 ± 0.23 (a)

Note: Different letters for each species within a row represent a significant difference between means according to Tukey test.

Table 3
Results of mixed-effects model analysis of plantation types and soil horizons on mean soil C and soil C storage, and effects of plantation type and tree aerial parts on mean aboveground C storage. $N = 180$ for soil characteristics, $N = 282$ for aboveground characteristics.

Fixed effects	df	C concentration (%)		C storage			
				Soil		Aboveground	
		F-statistic	*P*-value	*F* statistic	*P*-value	*F* statistic	*P*-value
Plantation type	2	1.98	0.14	1.01	0.37	111.99	<0.001
Horizon / Tree part	3	727.97	<0.001	353.13	<0.001	9.84	<0.001
Interaction	6	0.43	0.86	4.56	<0.001	40.35	<0.001

Note: df = Degrees of freedom.

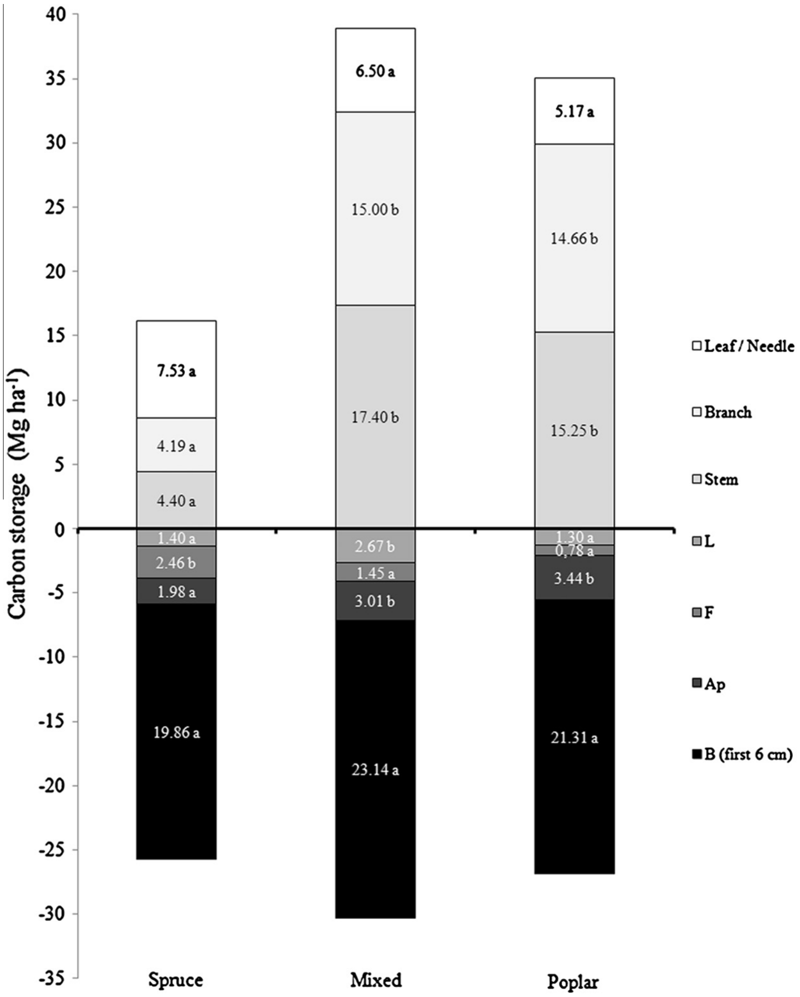

Fig. 2. Aboveground (above the *X*-axis) and soil (below the *X*-axis) carbon storage in the different plantation types. Across plantation types, different letters within each compartment represent a significant difference between means according to Tukey test.

(57% and 56% respectively; linear mixed-model, poplar plantation: $P = 0.04$; mixed plantation, $P = 0.05$).

3.4. Net effect of mixed species plantation on carbon storage

Non-additive effects (NAE) of species mixing on total aboveground and soil carbon storage were recorded, but only synergistic effects were significant. NAE on aboveground carbon storage was +68% and differed significantly from zero (One-sample *t*-test: $t = 5.84$, $DF = 22$, $P < 0.001$). The mean net effect of species mixing on soil carbon storage represented an increase of 15% and differed significantly from zero (One-sample *t*-test: $t = 3.36$, $DF = 14$, $P = 0.005$) (Fig. 3). When decomposed into various above-ground and soil compartments (Fig. 3), significant positive NAE of species mixing were observed for stems (One-sample *t*-test: $t = 5.86$, $DF = 22$, $P < 0.001$), branches (One-sample *t*-test: $t = 5.78$, $DF = 22$, $P < 0.001$), and for the L (One-sample *t*-test: $t = 3.45$, $DF = 14$, $P = 0.004$) and B (One-sample *t*-test: $t = 3.33$, $DF = 14$, $P = 0.005$) horizons.

4. Discussion

4.1. Effect of plantation types on soil horizons and carbon storage

Stands containing hybrid poplars (monocultures and mixed plots) stored lower quantities of carbon in the F-layer compared

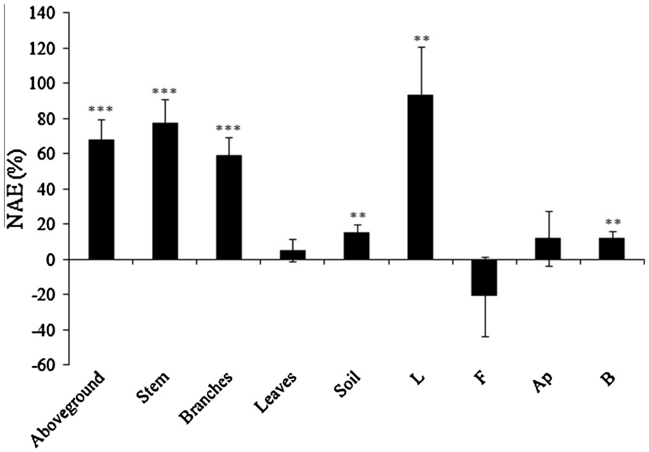

Fig. 3. Net effect of mixed plantation on carbon storage (Mean ± SE) in various soil and vegetation compartments. Non-additive effects (NAE) were calculated as 100 × (observed − predicted)/predicted. NAE that significantly different from zero, according to one-sample Student's *t*-tests, are indicated by *($P < 0.05$), **($P < 0.01$), or *** for ($P < 0.001$).

to mono-specific spruce plots, and conversely, they stored greater amounts of carbon in the Ap-layer. Among the factors that are known to influence organic layer characteristics, the activity of soil biota and the composition of the biota that are present profoundly affect litter degradation and incorporation of organic materials into mineral soil horizons (e.g. Wolters, 2000; Chauvat et al., 2007). The F-horizon is the site of major soil faunal activity, where the organic matter is partially fragmented and degraded until it is eventually incorporated into the Ap-layer. In spruce plots, the activity of decomposers seemed to be less efficient than in the other plantation types, in that organic matter accumulated in the F-horizon. In mixed plots, poplar litter can promote the abundance and activity of organisms (Saetre et al., 1999; Laganière et al., 2009) and seems to counteract the negative effects that were imposed by spruce litter on decomposers in the F-horizon. Carbon content of the deeper mineral soil layer (B-layer) did not significantly differ among plantation types, demonstrating that trees influenced only the uppermost layers during 9 years that had elapsed since planting.

4.2. Effect of mixing species on tree growth and productivity

The productivity of mixed-species plots was the greatest (4322 kg ha^{-1} year^{-1} of carbon) when compared to mono-specific plots (spruce and poplar, 1791 and 3897 kg ha^{-1} year^{-1} of carbon, respectively), but the difference was significant only compared to spruce plantations. Our estimates for the mono-specific plots are similar to those reported in the literature for older natural forests of spruce or trembling aspen (Gower et al., 1997; Alexander et al., 2012). Hybrid poplars attained greater heights and diameters in mixed plots, while spruce had reduced diameter growth in mixed-species compared to mono-specific plots. Hybrid poplars averaged 21% greater heights and 45% greater DBH in mixed than in mono-specific plots, while spruce had 25% lower DBH in mixed compared to mono-specific plots. These results confirm preliminary findings that were obtained by Benomar et al (2013) for these same plantations after six growing seasons, except for the spruce, which had greater height growth in mixed compared to mono-specific plots. The canopy was more open after six growing seasons, and competition for light induced greater allocation of carbon to height rather than to diameter growth (Grams and Andersen, 2007). After nine growing seasons, hybrid poplars were closing the canopy in mixed-species plots, while the canopy was still open in the spruce monocultures, which could retard spruce height growth in the mixed plots. In natural forests, Légaré et al. (2004) found a positive effect of aspen on black spruce (*Picea mariana* [Miller] BSP) growth, but only when aspen represented <40% of stand basal area. In our study, hybrid poplars represented a stand basal area above this threshold, which could explain the negative effects of mixing spruce with poplar on spruce growth. These results suggest that mixed plantings would benefit poplar harvesting, at least after 10 years of growth; they would be negative for spruce unless thinning of the poplars was preformed soon after canopy closure. However, the spacing that was used in this study (1 × 1 m) is not representative of what is normally practiced in forestry for wood production (i.e. 3–6 m spacing between hybrid poplars). Greater spacing between trees would probably delay canopy closure and allow hybrid poplars to reach maturity (*ca.* 20 years, Dickmann et al., 2001) before growth of spruce was excessively and negatively affected. As suggested by Kelty (2006), managers could also reduce the proportion of the taller species in the mixed plantations to increase productivity of the lower canopy species.

4.3. Non-additive effects of mixing species on carbon storage

The comparison between predicted carbon storage from mono-specific plantations and observed carbon storage in mixed-species plantations showed that mixing hybrid poplar and white spruce trees affected both above-ground and soil carbon storage through synergistic effects. Examination of the different ecosystem compartments revealed that this positive effect of species mixing was mainly due to carbon storage gains aboveground for stems and branches, and in L-horizon for the soil. Calculation of non-additive effects is commonly employed in litter decomposition studies (Wardle et al., 1997; Bonanomi et al., 2010; Barantal et al., 2011; Coq et al., 2011), but this technique is rarely used in forest productivity and carbon storage analyses. Compared to manipulative experiments with different litter mixtures in litter bags for decomposition studies, observational experiments on natural or planted forests is more constraining. One explanation is the difficulty involved in finding strictly mono-specific forests and mixtures of two species under natural conditions, that would allow non-additive effects of species mixing to be evaluated. Simple comparisons between productivity of mixed stands of aspen and spruce compared to pure aspen stands showed positive effects of mixing species on productivity (MacPherson et al., 2001; Martin et al., 2005). In the present study, spruce growth was not enhanced by planting this species in mixtures with poplar, but poplar productivity was sufficiently increased so that greater productivity in mixed-species plantations was attained than would be expected (i.e., mean of the mono-specific poplar and spruce plot productivities). Thus, spruce did not affect poplar carbon storage and, indeed, adding spruce to poplar benefitted poplar wood production, at least over the short-term. We can attribute this positive finding for poplar trees to the favorable microclimatic conditions that were provided by mixing, with greater canopy space available and less competition for light and nutrients in these tightly spaced plots (Benomar et al., 2013), and more efficient biogeochemical cycling (Chen and Popadiouk, 2002). This finding is consistent with the theory that mixed stands are more productive than monocultures (Vandermeer, 1989), and with other reports showing that stands with shade-intolerant hardwoods growing over shade-tolerant conifers were more productive than shade-intolerant hardwoods growing alone (Kelty, 1989; Man and Lieffers, 1999).

In the present study, we observed that mixed plots had greater carbon accumulation in the L-horizon. Considering that litter production of spruce and poplar in mixed plots was similar to mono-specific plots, these higher accumulations of litter in mixed plots could have resulted from lower decomposition rates. Moreover, capture of light resources can increase in mixtures through canopy stratification, where less shade-tolerant species over-top more shade-tolerant species and light interception is increased (Richards et al., 2010). Increased light interception may have induced colder microclimatic conditions at the soil surface, which could have retarded decomposition processes (Chapin et al., 2002). Further, spruce induced the formation of a denser understory habitat within mixed plots because of their size and shape, possibly limiting the dispersal of poplar leaves by the wind, which could have contributed to a greater accumulation of poplar litter in the mixed plots compared to poplar monocultures.

5. Conclusion

This study showed that aboveground poplar growth was enhanced in mixed-species compared to mono-specific poplar plantations. Conversely mixed-species planting was detrimental to spruce growth. These results suggest that mixed plantations would be positive only for the growth of poplar (at least under this tight spacing), which should reach merchantable sizes before the poplars in that were established mono-specific plantations. Mixed plantations negatively affected spruce growth after 9 years; however, since the conifers reach maturity much later than poplars, their growth could be enhanced by selective harvesting of the poplars before being severely hindered by interspecific competition. A greater accumulation of carbon was observed in the L-horizon in mixed-species plantations, probably due to colder microclimatic conditions that were brought on by greater light interception of the mixed canopy and a denser understory layer that limited the export of litter.

These differences in the aboveground and soil compartments led to greater carbon storage in mixed-species plots than was expected, demonstrating a synergistic effect of mixing (i.e., mean of the mono-specific poplar and spruce plot carbon storage values).

In addition to an increase in poplar timber yield (a desirable aspect for the forest industry), this study showed that mixed-species plantations of white spruce and hybrid poplar also promoted carbon sequestration, which is an important strategy for compensating CO_2 emissions, and for plantations management within boreal regions. Finally, these synergistic effects were found only 9 years following afforestation and should be subjected to further study to confirm sustained positive trends in the longer-term.

Acknowledgments

We thank Line Blackburn for field assistance and tree measurements, and W.F.J. Parsons of the CEF for English language correction, and for his comments on the manuscript. This study was supported by the Natural Sciences and Engineering Research Council of Canada (NSERC-CRSNG) through a Collaborative and Research Development grant awarded to AD. We also thank the CRSNG-UQAT-UQAM Industrial Chair in Sustainable Forest Management, the Quebec Intensive Silviculture Network and the Centre for Forest Research (CFR) for additional support.

References

Agriculture Canada Expert Committee on Soil Survey, 1987. The canadian system of soil classification. 2nd. Agriculture Canada Publication. 164 pp.

Alexander, H.D., Mack, M.C., Goetz, S., Beck, P.S.A., Belshe, E.F., 2012. Implications of increased deciduous cover on stand structure and aboveground carbon pools of Alaskan boreal forests. Ecosphere 3 (5), 45. http://dx.doi.org/10.1890/es11-00364.1.

Barantal, S., Roy, J., Fromin, N., Schimann, H., Hättenschwiler, S., 2011. Long-term presence of tree species but not chemical diversity affect litter mixture effects on decomposition in a neotropical rainforest. Oecologia 167 (1), 241–252. http://dx.doi.org/10.1007/s00442-011-1966-4.

Benomar, L., DesRochers, A., Larocque, G.R., 2011. Changes in specific leaf area and photosynthetic nitrogen-use efficiency associated with physiological acclimation of two hybrid poplar clones to intraclonal competition. Can. J. For. Res. 41 (7), 1465–1476. http://dx.doi.org/10.1139/x11-073.

Benomar, L., DesRochers, A., Larocque, G.R., 2012. The effects of spacing on growth, morphology and biomass production and allocation in two hybrid poplar clones growing in the boreal region of Canada. Trees-Struct. Funct. 26 (3), 939–949. http://dx.doi.org/10.1007/s00468-011-0671-6.

Benomar, L., DesRochers, A., Larocque, G.R., 2013. Comparing growth and fine root distribution in monocultures and mixed plantations of hybrid poplar and spruce. J. For. Res. 24 (2), 247–254. http://dx.doi.org/10.1007/s11676-013-0348-7.

Bolin, B., Sukumar, R., Ciais, P., Cramer, W., Jarvis, P., Kheshgi, H., Nobre, C., Semenov, S., Steffen, W. 2000. Global perspective. In: Watson, R.T., Noble, I.R., Bolin, B., Ravindranath, N.H., Verardo, D.J., Dokken, D.J. (Eds.), Land Use, Land Use Change and Forestry, A Special Report of the IPCC, pp. 23-51.

Bonanomi, G., Incerti, G., Antignani, V., Capodilupo, M., Mazzoleni, S., 2010. Decomposition and nutrient dynamics in mixed litter of Mediterranean species. Plant Soil 331 (1–2), 481–496. http://dx.doi.org/10.1007/s11104-009-0269-6.

Burdon, R.D., 2001. Genetic diversity and disease resistance: some considerations for research, breeding, and deployment. Can. J. For. Res. 31 (4), 596–606. http://dx.doi.org/10.1139/cjfr-31-4-596.

Cavard, X., Bergeron, Y., Chen, H.Y.H., Pare, D., 2010. Mixed-species effect on tree aboveground carbon pools in the east-central boreal forests. Can. J. For. Res. 40 (1), 37–47. http://dx.doi.org/10.1139/x09-171.

Chapin, F.S., Matson, P.A., Mooney, H.A., 2002. Principles of Terrestrial Ecosystem Ecology, Springer.

Chauvat, M., Ponge, J.F., Wolters, V., 2007. Humus structure during a spruce forest rotation: quantitative changes and relationship to soil biota. Eur. J. Soil Sci. 58 (3), 625–631. http://dx.doi.org/10.1111/j.1365-2389.2006.00847.x.

Chen, H.Y.H., Popadiouk, R.V., 2002. Dynamics of North American boreal mixedwoods. Environ. Rev. 10 (3), 137–166. http://dx.doi.org/10.1139/a02-007.

Coq, S., Weigel, J., Butenschoen, O., Bonal, D., Hättenschwiler, S., 2011. Litter composition rather than plant presence affects decomposition of tropical litter mixtures. Plant Soil 343 (1–2), 273–286. http://dx.doi.org/10.1007/s11104-011-0717-y.

Dickmann, D.I., Isebrands, J.G., Eckenwalder, J.E., Richardson, J., 2001. Poplar Culture in North America. NRC Research Press, Ottawa, Canada.

Erskine, P.D., Lamb, D., Bristow, M., 2006. Tree species diversity and ecosystem function: can tropical multi-species plantations generate greater productivity? For. Ecol. Manage. 233 (2–3), 205–210. http://dx.doi.org/10.1016/j.foreco.2006.05.013.

FAO, 2010. Global Forest Resources Assessment, Main Report, Rome, Italy, 90 pp.

Fitzsimmons, M.J., Pennock, D.J., Thorpe, J., 2004. Effects of deforestation on ecosystem carbon densities in central Saskatchewan, Canada. For. Ecol. Manage. 188 (1–3), 349–361. http://dx.doi.org/10.1016/j.foreco.2003.08.005.

Gartner, T.B., Cardon, Z.G., 2004. Decomposition dynamics in mixed-species leaf litter. Oikos 104 (2), 230–246.

Gower, S.T., Hunter, A., Campbell, J., Vogel, J., Veldhuis, H., Harden, J., Trumbore, S., Norman, J.M., Kucharik, C.J., 2000. Nutrient dynamics of the southern and northern BOREAS boreal forests. Ecoscience 7 (4), 481–490.

Gower, S.T., Vogel, J.G., Norman, J.M., Kucharik, C.J., Steele, S.J., Stow, T.K., 1997. Carbon distribution and aboveground net primary production in aspen, jack pine, and black spruce stands in Saskatchewan and Manitoba, Canada. J. Geophys. Res. Atmosp. 102 (D24), 29029–29041. http://dx.doi.org/10.1029/97jd02317.

Grams, T.E., Andersen, C., 2007. Competition for resources in trees: physiological versus morphological plasticity. In: Esser, K., Löttge, U., Beyschlag, W., Murata, J. (Eds.), Progress in Botany. Springer, Berlin Heidelberg, pp. 356–381.

Hättenschwiler, S., Tiunov, A., Scheu, S., 2005. Biodiversity and litter decomposition in terrestrial ecosystems. Annu. Rev. Ecol. Evol. Syst. 36, 191–218.

IPCC, 2007. Climate Change 2007: The Physical Science Basis. Contribution of Working Group I to the Fourth Assessment Report of the Intergovernmental Panel on Climate Change. In: Solomon, S., Qin, D., Manning, M., Chen, Z., Marquis, M., Averyt, K.B., Tignor, M., Miller, H.L. (Eds.). Cambridge University Press, Cambridge, United Kingdom and New York, NY, USA.

Johansson, T., 2003. Mixed stands in Nordic countries – a challenge for the future. Biomass Bioenergy 24 (4–5), 365–372. http://dx.doi.org/10.1016/s0961-9534(02)00165-4.

Johnson, D.W., 1992. Effects of forest management on soil carbon storage. Water Air Soil Pollut. 64 (1–2), 83–120. http://dx.doi.org/10.1007/bf00477097.

Kabzems, R., Nemec, A.L., Farnden, C., 2007. Growing trembling aspen and white spruce intimate mixtures: Early results (13–17 years) and future projections. BC J. Ecosyst. Manage. 8 (1), 1–14.

Kelty, M.J., 1989. Productivity of New-England hemlock hardwood stands as affected by species composition ans canopy structure. For. Ecol. Manage. 28 (3–4), 237–257. http://dx.doi.org/10.1016/0378-1127(89)90006-6.

Kelty, M.J., 1992. Comparative productivity of monocultures and mixed-species stands. In: Kelty, M.J. (Ed.), The Ecology and Silviculture of Mixed-Species Forests. Kluwer Academic, Dordrecht, The Netherlands, pp. 125–141.

Kelty, M.J., 2006. The role of species mixtures in plantation forestry. For. Ecol. Manage. 233 (2–3), 195–204. http://dx.doi.org/10.1016/j.foreco.2006.05.011.

Laganière, J., Angers, D.A., Paré, D., Bergeron, Y., Chen, H.Y.H., 2011. Black spruce soils accumulate more uncomplexed organic matter than aspen soils. Soil Sci. Soc. Am. J. 75 (3), 1125–1132. http://dx.doi.org/10.2136/sssaj2010.0275.

Laganière, J., Paré, D., Bradley, R.L., 2009. Linking the abundance of aspen with soil faunal communities and rates of belowground processes within single stands of mixed aspen-black spruce. Appl. Soil Ecol. 41 (1), 19–28. http://dx.doi.org/10.1016/j.apsoil.2008.08.005.

Lamb, D., Erskine, P.D., Parrotta, J.A., 2005. Restoration of degraded tropical forest landscapes. Science 310 (5754), 1628–1632. http://dx.doi.org/10.1126/science.1111773.

Leff, J.W., Wieder, W.R., Taylor, P.G., Townsend, A.R., Nemergut, D.R., Grandy, A.S., Cleveland, C.C., 2012. Experimental litterfall manipulation drives large and rapid changes in soil carbon cycling in a wet tropical forest. Glob. Change Biol. 18 (9), 2969–2979. http://dx.doi.org/10.1111/j.1365-2486.2012.02749.x.

Légaré, S., Paré, D., Bergeron, Y., 2004. The responses of black spruce growth to an increased proportion of aspen in mixed stands. Can. J. For. Res. 34 (2), 405–416. http://dx.doi.org/10.1139/x03-251.

Légaré, S., Paré, D., Bergeron, Y., 2005. Influence of aspen on forest floor properties in black spruce-dominated stands. Plant Soil 275 (1–2), 207–220. http://dx.doi.org/10.1007/s11104-005-1482-6.

Lieth, H., 1975. Modeling the primary productivity of the world. In: Lieth, H., Whittaker, R.H. (Eds.), Primary Productivity of the Biosphere. Ecological Studies 14, Springer-Verlag, Berlin, pp. 237–263.

Liu, L.L., King, J.S., Giardina, C.P., 2005. Effects of elevated concentrations of atmospheric CO_2 and tropospheric O_3 on leaf litter production and chemistry in trembling aspen and paper birch communities. Tree Physiol. 25 (12), 1511–1522.

MacPherson, D.M., Lieffers, V.J., Blenis, P.V., 2001. Productivity of aspen stands with and without a spruce understory in Alberta's boreal mixedwood forests. For. Chronicle 77 (2), 351–356.

Man, R.Z., Lieffers, V.J., 1999. Are mixtures of aspen and white spruce more productive than single species stands? For. Chronicle 75 (3), 505–513.

Martin, J.L., Gower, S.T., Plaut, J., Holmes, B., 2005. Carbon pools in a boreal mixedwood logging chronosequence. Glob. Change Biol. 11 (11), 1883–1894. http://dx.doi.org/10.1111/j.1365-2486.2005.01019.x.

McCracken, A.R., Dawson, W.M., 1997. Growing clonal mixtures of willow to reduce effect of Melampsora epitea var. epitea. Eur. J. For. Pathol. 27 (5), 319–329.

Pinheiro, J., Bates, D., DebRoy, S., Sarkar, D., The R Core team 2014. nlme: Linear and Nonlinear Mixed Effects Models. R package version 3.1-115 <http://CRAN.R-project.org/package=nlme>.

Piotto, D., 2008. A meta-analysis comparing tree growth in monocultures and mixed plantations. For. Ecol. Manage. 255 (3–4), 781–786. http://dx.doi.org/10.1016/j.foreco.2007.09.065.

Pitt, D.G., Bell, F.W., 2004. Effects of stand tending on the estimation of aboveground biomass of planted juvenile white spruce. Can. J. For. Res. 34 (3), 649–658. http://dx.doi.org/10.1139/x03-234.

Pothier, D., Savard, F., 1998. Actualisation des tables de production pour les principales espèces forestières du QuébecMinistère des Ressources naturelles, Direction des inventaires forestiers, Québec. RN98-3054. 184 pp.

Richards, A.E., Forrester, D.I., Bauhus, J., Scherer-Lorenzen, M., 2010. The influence of mixed tree plantations on the nutrition of individual species: a review. Tree Physiol. 30 (9), 1192–1208. http://dx.doi.org/10.1093/treephys/tpq035.

Rothe, A., Binkley, D., 2001. Nutritional interactions in mixed species forests: a synthesis. Can. J. For. Res. 31 (11), 1855–1870. http://dx.doi.org/10.1139/cjfr-31-11-1855.

Saetre, P., Brandtberg, P.O., Lundkvist, H., Bengtsson, J., 1999. Soil organisms and carbon, nitrogen and phosphorus mineralisation in Norway spruce and mixed Norway spruce-birch stands. Biol. Fertil. Soils 28 (4), 382–388. http://dx.doi.org/10.1007/s003740050508.

Sayer, E.J., Heard, M.S., Grant, H.K., Marthews, T.R., Tanner, E.V.J., 2011. Soil carbon release enhanced by increased tropical forest litterfall. Nature Climate Change 1 (6), 304–307. http://dx.doi.org/10.1038/nclimate1190.

Shvidenko, A., Barber, C.V., Persson, R., Gonzalez, P., Hassan, R., Lakyda, P., McCallum, I., Nilsson, S., Pulhin, J., Van Rosenburg, B., Scholes, B., 2005. Chapter 21: Forest and Woodland Systems. In: Hassan, R., Scholes, R., Neville, A. (Eds.), Ecosystems and Human Well-Being: Current State and Trends. Millenium Ecosystem Assessment Series 1, Island Press, Washington, pp. 587–621.

Soil Classification Working Group, 1998. The canadian system of soil classification, Agriculture Canada Publication, 187 pp (3rd.).

Vance, E.D., Chapin, F.S., 2001. Substrate limitations to microbial activity in taiga forest floors. Soil Biol. Biochem. 33 (2), 173–188. http://dx.doi.org/10.1016/s0038-0717(00)00127-9.

Vandermeer, J., 1989. The Ecology of Intercropping. Cambridge University Press, Cambridge, UK.

Wardle, D.A., Bonner, K.I., Nicholson, K.S., 1997. Biodiversity and plant litter: experimental evidence which does not support the view that enhanced species richness improves ecosystem function. Oikos 79 (2), 247–258.

Wolters, V., 2000. Invertebrate control of soil organic matter stability. Biol. Fertil. Soils 31 (1), 1–19. http://dx.doi.org/10.1007/s003740050618.