Effects of adventitious roots on age determination in Balsam fir (*Abies balsamea*) regeneration

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Abstract: The age structure of balsam fir (*Abies balsamea* (L.) Mill.) regeneration is frequently used to investigate boreal forest dynamics of North America. Tree ages are usually estimated by counting annual growth rings at the shoot–root interface located above or close to the root system. Inaccurately locating the shoot–root interface could lead to imprecise age determination. In this study, balsam fir seedlings (<2 m height) were harvested in whole from closed forest stands located in the province of Quebec, Canada. Seedling age was determined by (*i*) counting the number of annual growth rings at the presumed shoot–root interface, and (*ii*) counting all terminal bud scars from the apex to the base of the hypocotyl (true collar). For all seedlings with adventitious roots, the number of terminal bud scars on the entire trunk was higher than the number of growth rings at the shoot–root interface. The formation of adventitious roots on the belowground trunk was accompanied by a reverse taper phenomenon, i.e., the number of annual growth rings decreased from the presumed shoot–root interface to the true collar. Counting annual growth rings at any level on the trunk of balsam fir seedlings that form adventitious root systems would not be reliable, underestimation's exceeding 20 years are possible and the resulting age structures could lead to erroneous interpretations of regeneration dynamics.

Résumé : La structure d'âge de la régénération de sapin baumier (*Abies balsamea* (L.) Mill.) est souvent utilisée pour étudier la dynamique de la forêt boréale en Amérique du Nord. L'âge des arbres est habituellement déterminé en comptant les cernes annuels à la jonction entre la tige et les racines qui est située au-dessus ou près du système racinaire. La détermination de l'âge peut manquer de précision si la jonction entre la tige et les racines n'est pas localisée avec exactitude. Dans cette étude, des semis de sapin baumier (<2 m de haut) ont été récoltés en entier dans des peuplements forestiers fermés situés dans la province de Québec, au Canada. L'âge des semis a été déterminé (*i*) en comptant le nombre de cernes annuels à la jonction présumée entre la tige et les racines et (*ii*) en comptant les cicatrices des écailles de tous les bourgeons terminaux de l'apex à la base de l'hypocotyle (collet véritable). Chez tous les semis avec des racines adventives, le nombre de cicatrices des écailles des bourgeons terminaux sur toute la tige était plus élevé que le nombre de cernes annuels à la jonction entre la tige et les racines. La formation de racines adventives sur la portion hypogée de la tige était accompagnée d'un phénomène de défilement inversé, c'est-à-dire que le nombre de cernes annuels diminuait de la jonction présumée entre la tige et les racines jusqu'au collet véritable. Le décompte des cernes annuels à n'importe quel niveau sur la tige des semis de sapin baumier qui forment un système de racines adventives n'est pas fiable; on peut sous-estimer l'âge par plus de 20 ans et les structures d'âge qui en résultent peuvent conduire à des interprétations erronées de la dynamique de la régénération.

[Traduit par la Rédaction]

Introduction

Balsam fir (*Abies balsamea* (L.) Mill.) is a *Pinaceae* broadly distributed across sub-boreal and sub-alpine forests of northeastern North America. The prolific regeneration of this species under various closed canopies forms, with time, an abundant seedling bank (Frank 1990). Frequency histograms of balsam fir age are commonly used to study forest dynamics (Ghent 1958; Batzer and Popp 1985; Côté and Bélanger 1991; Bélanger et al. 1993; Bergeron and Charron 1994; Osawa 1994; Galipeau et al. 1997; Morin and Laprise 1997; Sirois 1997; Kneeshaw 1998). In nearly all of these studies, tree age is determined by counting the number of annual growth rings present at the presumed collar, i.e., at the shoot–root interface located above or close to the first roots and (or) by counting terminal bud scars from the presumed collar to the apex.

Ecologists and foresters acknowledge precise age determination is a difficult task when balsam fir has grown under a closed canopy (Zarnovican 1981; Strauch 1991; Morin and Laprise 1997). Morin and Laprise (1997) suggested that understory balsam firs could experience frequent radial and height growth interruptions that would have resulted in a systematic underestimation of the true age of the seedling bank. However, growth cessation leading to undetected growth rings or bud scars has never been clearly observed. Alternatively, several studies indicate that the age of seedlings

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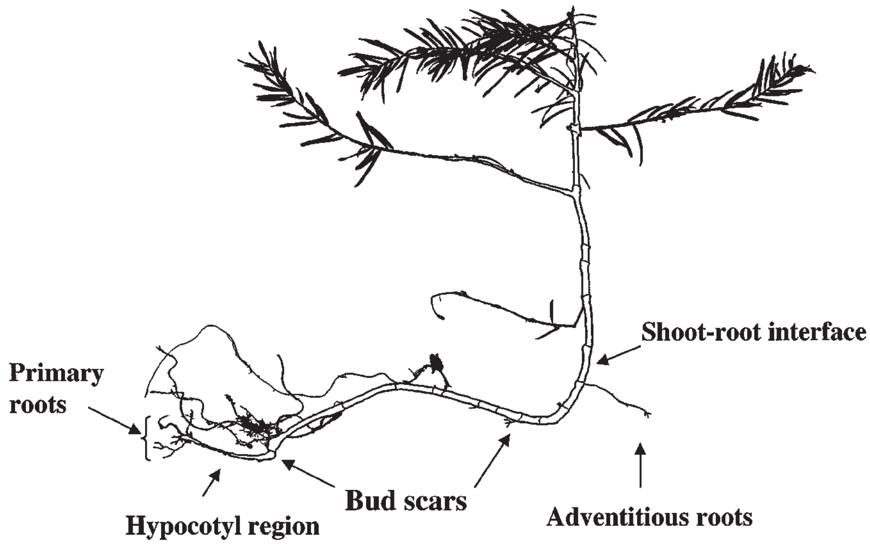
Fig. 1. Typical balsam fir seedling harvested showing belowground portion with adventitious roots, terminal bud scars, hypocotyl region, and primary roots.



could be underestimated if the true collar is inaccurately located (LeBarron 1945; Kohyama 1983; Telewski and Lynch 1991; Telewski 1993). During understory growth, the trunk base may gradually become embedded, promoting the formation of adventitious roots. With time, these adventitious roots could cause a reduction or a cessation of radial growth at the hypocotyl level resulting in a reverse taper phenomenon where the number of growth ring decrease from the shoot–root interface towards the hypocotyl, i.e., the true collar (DesRochers and Gagnon 1997). The possibility that the presence of adventitious roots could lead to imprecise age determination in balsam fir seedlings has been acknowledged (McLaren and Janke 1996) but never tested. In this study, we compared seedling ages determined by conventional ring count with those obtained by counting all terminal bud scars on the entire trunk when the true collar is precisely located.

Main study site

A balsam fir –*Hylocomium* stand was selected 100 km north of Lake Saint-Jean, Que. (49°46′N, 72°42′W). The stand is located within the black spruce (*Picea mariana* (Mill.) BSP) – feather moss (*Pleurozium schreberi* (Brid.) Mitt.) ecoclimatic domain, region 12B (Thibault 1987). The selected stand was initiated around 1841, and two spruce budworm outbreaks that occurred around 1880 and 1909 (Morin 1994) only altered its structure. The canopy is closed and dominated by balsam fir averaging 15 m in height. Ten percent of the total basal area is composed of black spruce, white spruce (*Picea glauca* (Moench) Voss), and white birch (*Betula papyrifera* Marsh.). The density of balsam regeneration (<2 m height) was evaluated at 87 000 seedlings/ha with a mean height of 20 cm. The age structure of the balsam fir regeneration was reported to have a unimodal distribution centered on 20 years (Morin and Laprise 1997). The bottom layer is dominated by *Hylocomium splendens* (Hedw.) B.S.G. in association with *Pleurozium schreberi* and *Ptilium crista-castrensis* (Hedw.). The humus depth varies between 10 and 20 cm. The stand is located on a moderately drained sandy till.

Methods

At the end of September 1998, 120 fir seedlings (<30 cm height) were harvested whole (shoots and roots). The seedlings had to be sound looking, without trunk damage, and established on hypnaceous moss. Seedlings with no hypocotyl and suspected to be from a layer origin, connected to a parental stem or emerging from a long belowground stem were excluded.

In the laboratory, the seedlings were completely immersed in water to remove small soil and humus particles with tweezers from the fine root system under a binocular lens (63×). Afterwards, the seedlings were cut into aboveground and belowground portions at 1 cm above the first root, i.e., the shoot–root interface (Fig. 1). A small disk section was sampled at the cut for a subsequent ring count. The belowground section was carefully examined to identify its morphological components. In general, the hypocotyl region was present and could be identified by its smooth bark devoid of leaf scars and adjacent primary roots (Fig. 1). The diameter of the hypocotyl and of the trunk at the shoot–root interface were measured with a precision of 0.01 mm. The underground portion was dried 24 h at 70°C and weighed with a precision of 0.0001 g (Mettler AE160). Adventitious and primary roots (<0.7 mm diameter) were weighed separately.

Seedling ages were first determined by the usual method: counting the number of annual rings on the disk section sampled at the shoot–root interface and, as a check, the number of terminal bud scars on the aboveground trunk. Rings were carefully counted under the microscope (40–100×). Secondly, all terminal bud scars observed on the belowground trunk from the shoot–root interface to the hypocotyl were counted, and this number was added to the number of terminal bud scars counted on the aboveground trunk. Ages obtained by the different techniques were correlated using Pearson's r (DesRochers and Gagnon 1997), and means were compared using paired t tests (JMP-SAS statistical package; SAS Institute Inc. 1996).

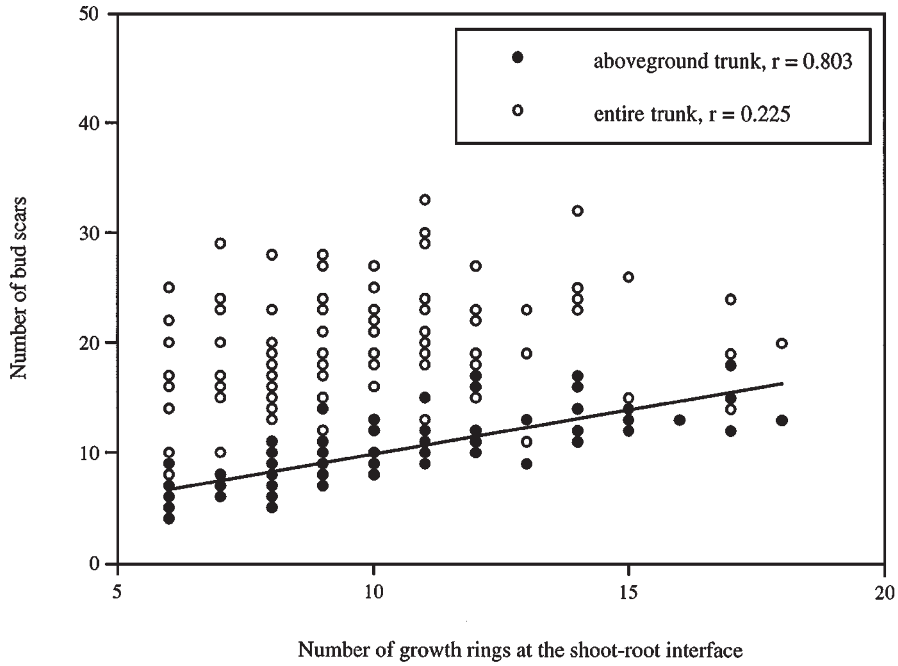
Other sampling

To increase the representativeness of our sampling, balsam fir seedlings (<2.0 m) were randomly chosen and harvested in whole (shoots and roots) in four other stands located in two other Quebec regions. The first region is located near Lake Duparquet, Abitibi at about 650 km from the main study site, region 8C1 (Thibault

Table 1. Characteristics of the seedlings sampled in the main study site, Lake Liberal, Lac St-Jean.

	Minimum	Maximum	Mean	SD
Height growth in 98 (mm)	0.07	44.6	13.1	7.55
Total height (mm)	52.8	265	131	41.27
Trunk diameter (mm) at the presumed shoot–root interface	0.82	4.6	2.09	0.74
Hypocotyl diameter (mm)	0.07	3.53	1.23	0.57

Fig. 2. Scatterplot of the number of terminal bud scars counted on the aboveground trunk (solid circles) and the number of terminal bud scars counted on the entire trunk from the apex to the belowground hypocotyl when present (open circles) versus the number of annual growth rings counted at the shoot–root interface. A fitted regression line is also shown.



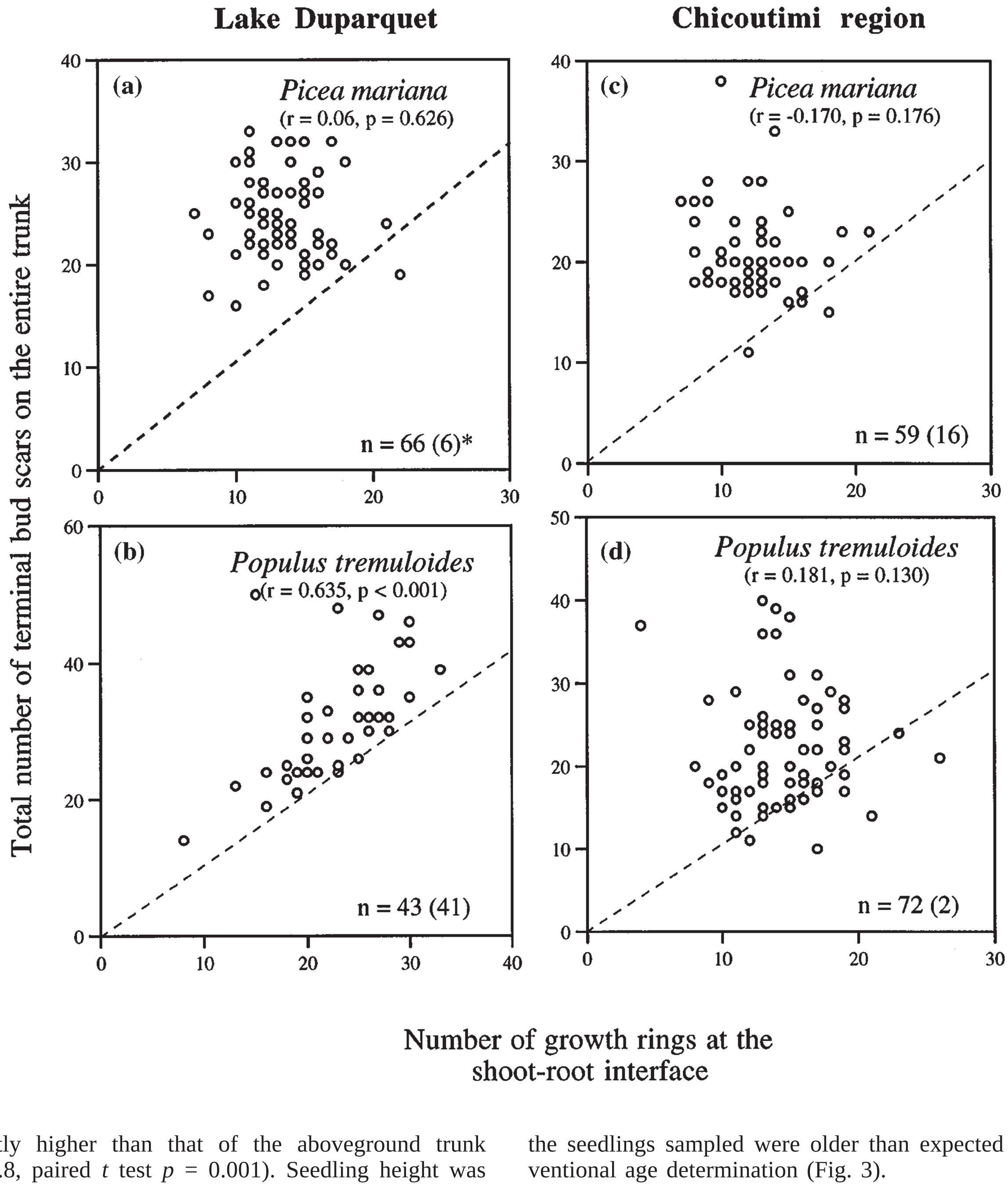
1987). The second region is located 300 km south of the main study site, near Chicoutimi, region 8E (Thibault 1987), at the northern limit of the Réserve du Parc des Laurentides on the Laurentidian plateau. In both regions, a stand dominated by trembling aspen (*Populus tremuloides* Michx.) and a stand dominated by *Picea mariana* were selected. These two stand compositions were selected to sample seedlings that grew under distinct stand types (deciduous and coniferous). We determined the age of all the seedlings sampled using the techniques described above.

Results

Table 1 shows some general characteristics of the seedlings sampled in the main study site (Lake Liberal area). Of the 120 seedlings harvested, 108 had adventitious roots and 20 of these had no primary roots. The belowground section was composed of a principal axis in continuity with the trunk. This axis often curved to become almost horizontal to the forest floor (Fig. 1). The presence of terminal bud scars and leaf scars on this axis indicated that this belowground axis was formerly part of the aboveground trunk. On this axis, fine adventitious lateral roots (<0.7 mm diameter) were observed at regular intervals. The percentage of fine adventitious lateral roots on total fine root mass increased with the number of terminal bud scars on the belowground trunk (r = 0.611, $p < 0.001$). The diameter of the hypocotyl region was almost always smaller than the diameter of the aboveground trunk measured at the shoot–root interface (paired t test, $p < 0.001$; Table 1).

The number of annual growth rings counted at the shoot–root interface was weakly correlated with the number of terminal bud scars counted on the entire trunk ($r = 0.225$, $p = 0.025$) and strongly correlated with, and often equaled, the number of terminal bud scars counted on the aboveground trunk ($r = 0.803$, $p < 0.001$; Fig. 2). According to our observations, the remaining variation of 0.197 can be attributed to inaccuracy in detecting narrow and sometimes incomplete annual rings, missing rings, and in detecting all terminal bud scars on the trunk. We found no correlation between the number of terminal bud scars counted on the aboveground trunk and the number of terminal bud scars counted on the belowground trunk ($p = 0.108$). The average number of terminal bud scars counted on the entire trunk (19.3 ± 5.3) was

Fig. 3. Scatterplots of age determined by counting the number of terminal bud scars present on the entire trunk versus age determined by counting the number of growth rings on a cross section of the trunk at the presumed shoot–root interface. Seedlings were sampled in *Picea mariana* (*a* and *c*) and *Populus tremuloides* (*b* and *d*) dominated stands located in the Lake Duparquet (*a* and *b*) and Chicoutimi (*c* and *d*) regions. The broken line indicates the 1:1 relationship. Circles located above the broken line represent seedlings whose ages are unequivocally underestimated by ring count. The values in parenthesis next to the sample size indicates the number of seedlings on which the hypocotyl was still present and precise age determination could be done.



significantly higher than that of the aboveground trunk (10.0 ± 2.8, paired *t* test $p = 0.001$). Seedling height was correlated with the number of terminal bud scars on the aboveground trunk ($r = 0.306$, $p < 0.001$) but was not correlated with the number of terminal bud scars counted on the entire trunk ($r = 0.167$; $p = 0.09$)

For the balsam fir sampled in the four extra stands located near Lake Duparquet and Chicoutimi, the number of terminal bud scars counted on the entire trunk is only an indication of the minimal age of many seedlings, since the hypocotyl was often absent. On all sites, more than 80% of the seedlings sampled were older than expected from conventional age determination (Fig. 3).

Discussion

Ring count at the presumed shoot–root interface nearly equals the number of terminal bud scars on the aboveground trunk. However, the number of terminal bud scars on the entire trunk was much higher than the number of rings counted at the presumed shoot–root interface. Although the number of growth rings was highest close to the presumed shoot–root

Fig. 4. An example of the variation of both the number of annual growth rings (solid circles) and trunk diameter (open circles) on the entire trunk.

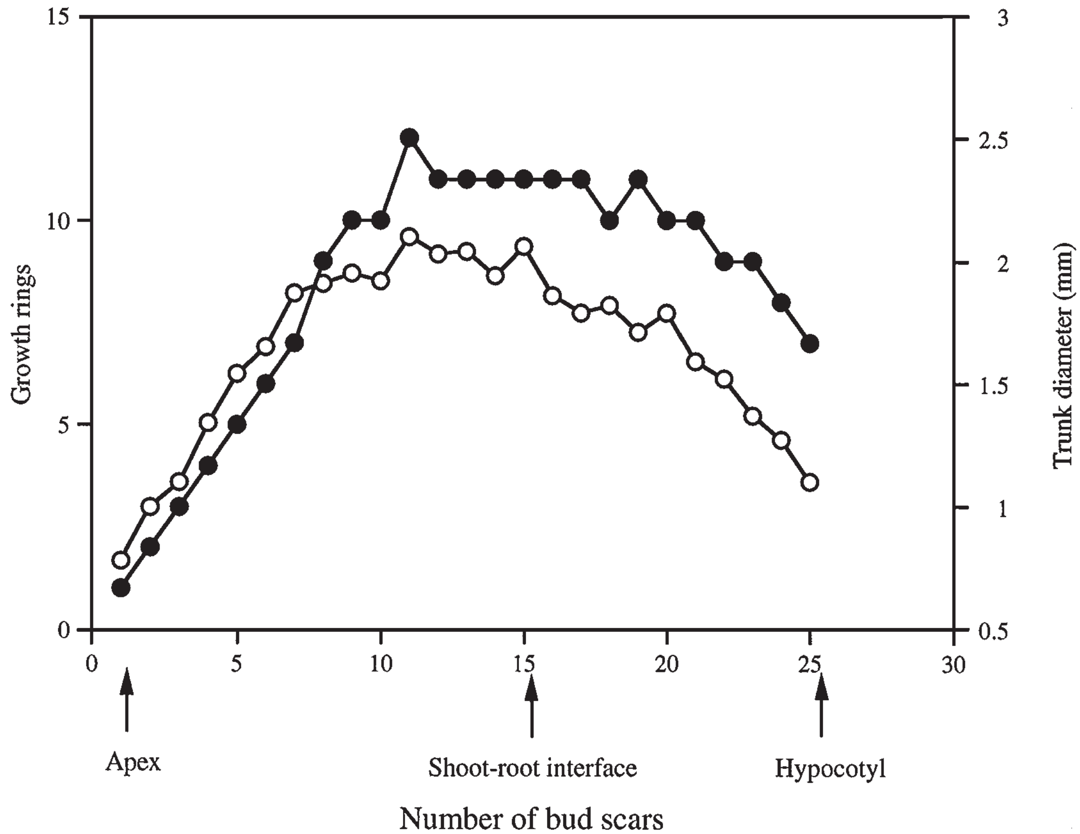


Table 2. General characteristics of stands visited in the two regions.

	Age (years)	Canopy closed (%)	Deposit and soil texture	Drainage	Forest floor dominated by
Chicoutimi region					
Picea mariana	90	75–80	Till sandy loam	Xeric	Moss (*P. scheberi*)
Populus tremuloides	70	60	Till sandy loam	Mesic	Broadleaf
Lake Duparquet					
Picea mariana	75	15–75	Glacio-lacustrine clay	Hydric	Moss (*Spagnum* sp.)
Populus tremuloides	75	70	Glacio-lacustrine clay	Mesic	Broadleaf

interface, both the number of annual growth rings and trunk diameter decreased from this interface to the hypocotyl region (Table 1, Fig. 4). This progressive loss of growth rings (reverse taper) has been described in detail by DesRochers and Gagnon (1997) for black spruce trees (mean height 9.3 m). These authors also noticed that the juvenile period was often characterized by prostrate growth, which possibly promoted the formation of adventitious roots. The presence of adventitious roots was not an exceptional phenomenon in balsam fir seedlings, including those growing in deciduous stands (Table 2). In the main study site, a seedling measuring 7 cm aboveground and believed to be 7 years old based on the number of annual growth rings at the shoot–root interface or the number of terminal bud scars on the aboveground trunk could be in fact 10, 20, or even 30 years old (Fig. 2). Consequently, counting growth rings at the presumed shoot–root interface of balsam fir seedlings with adventitious roots can underestimate seedling age by more than 20 years. Conventional ring count on these seedlings can also produce inaccurate age–height relationships and very different age structures (Fig. 5). The age of these seedlings cannot be determined without completely extracting the belowground portion. When the hypocotyl region is absent, precise age determination becomes almost impossible (Telewski 1993; DesRochers and Gagnon 1997). A reexamination of the ecological interpretation of most studies dealing with age of balsam fir seedlings is required at the light of the results presented here.

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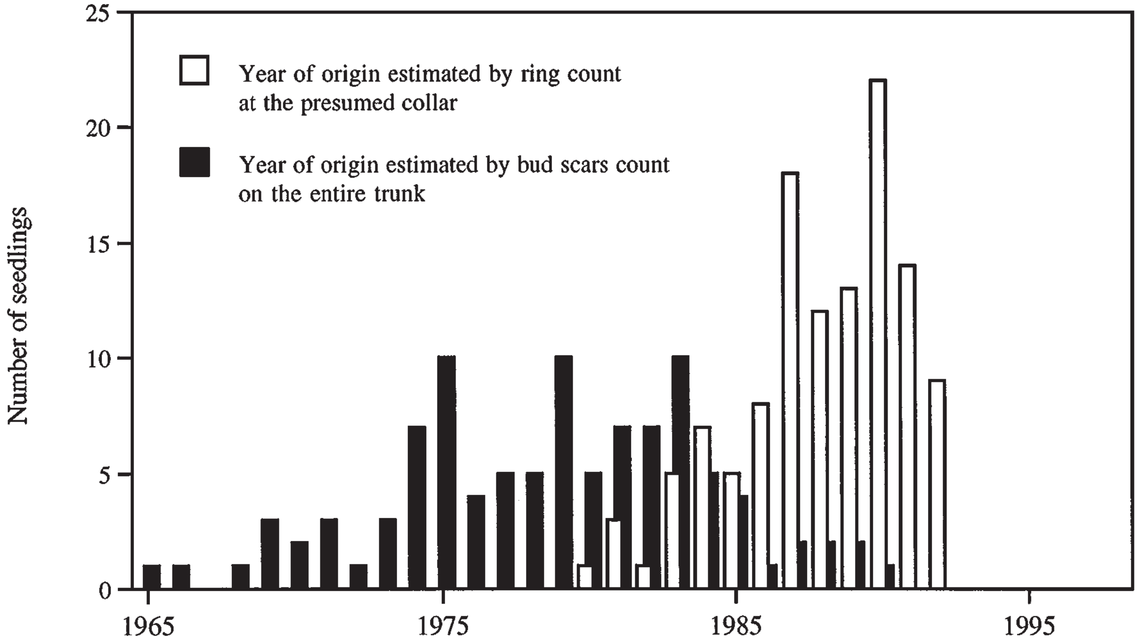
Fig. 5. Age structures of seedlings sampled in the main study site (Lake Liberal) evaluated using the conventional ring count method and our method.



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References

Batzer, H.O., and Popp, M.P. 1985. Forest succession following a spruce budworm outbreak in Minnesota. For. Chron. **61**: 75–80.

Bélanger, L., Allard, D., and Meek, P. 1993. Dynamique d'établissement d'un peuplement bi-étagé de bouleau blanc et de sapin baumier en zone boréale. For. Chron. **69**: 173–176.

Bergeron, Y., and Charron, D. 1994. Postfire stand dynamic in a southern boreal forest (Québec): a dendrochronological approach. Écoscience, **1** :173–184.

Côté, S., and Bélanger, L. 1991. Variations de la régénération préétablie dans les sapinières boréales en fonction de leurs caractéristiques écologiques. Can. J. For. Res. **21**: 1779–1795.

DesRochers, A., and Gagnon, R. 1997. Is ring count at ground level a good estimation of black spruce age? Can. J. For. Res. **27**: 1263–1267.

Frank, R.M. 1990. *Abies balsamea* (L.) Mill. Balsam fir. *In* Silvics of North America. Vol. 1. Conifers. U.S. Dep. Agric. Agric. Handb. No. 654.

Galipeau, C., Kneeshaw, D., and Bergeron, Y. 1997. White spruce and balsam fir colonization of a site in the southeastern boreal forests as observed 68 years after fire. Can. J. For. Res. **27**: 139–147.

Ghent, A.W. 1958. Studies of regeneration in forest stands devasted by the spruce budworm II. Age, height growth, and related studies of balsam fir seedlings. For. Sci. **4**: 135–146.

Kneeshaw, D.D. 1998. Effets des épidémies de la tordeuse des bourgeons de l'épinette sur la dynamique de la régénération dans la forêt boréale du nord-ouest du Québec. Ph.D. dissertation, Université du Québec à Montréal, Montréal.

Kohyama, T. 1983. Seedling stage of two subalpine *Abies* species in distinct form sapling stage: a matter-economic analysis. Bot Mag. Tokyo, **96**: 49–65.

LeBarron, R.K. 1945. Adjustment of black spruce root system to increasing depth of peat. Ecology, **26**: 309–311.

McLaren, B.E., and Janke, R.A. 1996. Seedbed and canopy cover effects on balsam fir seedling establishment in Isle Royale National Park. Can. J. For. Res. **26**: 782–793.

Morin, H. 1994. Dynamics of balsam fir forests in relation to spruce budworm outbreaks in the boreal zone of Quebec. Can. J. For. Res. **24**: 730–741.

Morin, H., and Laprise, D. 1997. Seedling bank dynamics in boreal balsam fir forests. Can. J. For. Res. **27**: 1442–1451.

Osawa, A. 1994. Seedling responses to forest canopy disturbance following a spruce budworm outbreak in Maine. Can. J. For. Res. **24** : 850–859.

Sirois, L. 1997. Distribution and dynamics of balsam fir (*Abies balsamea* (L.) Mill.) at its northern limit in the James Bay area. Écoscience, **4**: 340–352.

Strauch, P.J. 1991. The establishment and early growth of red spruce and balsam fir regeneration. *In* Proceedings of the Conference on Natural Regeneration Management. *Edited by* C.M. Simpson. Forestry Canada, Maritimes Region, Fredericton, N.B. pp. 147–156.

Telewski, F.W. 1993. Determining the germination date of woody plants: a proposed method for locating the root/shoot interface. Tree-Ring Bull. **53**: 13–16.

Telewski, F.W., and Lynch, A.M. 1991. Measuring growth and development of stems. *In* Techniques and approaches in forest tree ecophysiology. *Edited by* J.P. Lassoie and T.M. Hinkley. CRC Press. Inc., Boston, Mass. pp. 503–555.

Thibault, M. 1987. Les régions écologiques du Québec méridional. Deuxième approximation. Carte. Service de la recherche, ministère de l'Énergie et des Ressources du Québec, Québec.

Zarnovican, R. 1981. À propos de l'âge du sapin baumier et de sa détermination. Can. J. For. Res. **11**: 805–811.