



Article

**The role of starch in the formation of annual radial growth increments in silver birch
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Abstract

The major environmental factors determining annual, radial growth in trees are temperature, water supply and light. These factors influence the physiological processes determining the magnitude of radial growth. According to the currently prevailing concept annual, radial growth is determined mostly by photosynthetic activity. One of the most important functions of the xylem is the transport of sap, and the total volume of the water transport system depends on the rate of transpiration at plant surfaces. There is a correlation between the width of an annual ring and the phytomass that transpires water. Therefore, transpiration is the major factor affecting the annual, radial growth. The entire volume of the water supply system is used by birches for the flow of sap in spring but during the summer it is only in the newly formed annual ring that minimal resistance to water flow is observed. We assume that this can be attributed to the fact that the sugars contained in the spring sap are not completely used for tissue formation, the surplus being transformed into starch, which temporarily clogs part of the xylem vessels. The formation of new conduits is stimulated by obstruction of the existing water vessels and is dependent upon the degree of this obstruction. Comparative microscopic observations of the presence of starch in vessels prior to and during the vegetative period confirmed our hypothesis.

Keywords: *Betula pendula*, radial growth, starch, water supply system, xylem**Introduction**

The main function of the xylem system is to move water and nutrients from the root to the tree top. Data concerning the number of the annual rings that participate in lifting the sap are rather variable. In the most popular pipe model, a plant is considered as a set of elementary pipes (xylem vessels), each of which supports and supplies a certain mass of leaves (Usoltsev, 1996). According to this model, a correlation must exist between the transpiring phytomass and radial growth. However, due to the variability of forest habitat conditions and differences between tree species, numerous and significant deviations from this model have been observed.

In this connection, the general opinion in the literature is that the volumetric growth of a tree trunk is a measure of assimilate surplus, that is, of the remainder after the assimilates have been used for root, branch, and photosynthesizing phytomass growth. The annual, radial growth

of a trunk is directly proportional to its volumetric growth, though the actual amount naturally depends on its diameter (Usoltsev, 1996). The major environmental factors determining the annual, radial growth are temperature, water supply, and light (Kramer and Kozlovski, 1960). These factors influence the physiological processes in a tree that determine the amount of annual, radial growth although, here again, there are substantial contradictory data.

During a study of the dynamics of annual, radial growth in connection with the tolerance of birches toward defoliation by herbivorous insects (Tolkach *et al.*, 2007), cases of annual, radial growth increase during a year of tree desiccation caused by defoliation were observed. In this connection it is suggested that reserve nutrients such as starch and lipids should be considered as factors influencing the magnitude of annual, radial growth.

Materials and methods

The study was undertaken, in the southern Sverdlovsk Region of the Central Urals, in birch forest stands composed mainly of silver birch (*Betula pendula* Roth) with some pubescent birch (*B. pubescens* Ehrh.).

For the determination of the amount of starch and its localization within the xylem, 10 trees were selected and felled at Sverdlovsk Silvicultural Farm and 10-13 wood samples sawn out from the upper, middle, and lower part of each trunk. The presence of starch was determined visually under a microscope after treatment of the wood plates with Lugol's solution (0.3% iodine in 1.5% aqueous potassium iodide). The wood plates were selected and analyzed during the winters of 2002/2003, 2006/2007, and in spring and summer of 2007.

For determination of the effect of the intensity of spring sap flow on annual, radial growth, 30 trees with similar diameters (24-28 cm at a height of 1.3 m) were selected and numbered in a birch stand on the outskirts of Yekaterinburg in the spring of 2007. During the period of active sap flow, immediately before the start of foliage development, notches sized 10 x 10 x 5-6 cm were made on the southern side of the trunks of 10 of these trees at a height of 1.3 m. Another 10 trees were cut in the same manner a week later, that is, immediately after the end of spring sap flow and completion of foliage development. Another 10 trees were left undamaged as controls. Cores were taken from all 30 trees in the autumn of the same year. Radial growth increments, determined using a microscope (MBS-10), are presented in standard units (1 unit = 0.2 mm).

Statistical processing of the data was carried out with the program STATISTICA.

Results and discussion

Storage of reserve nutrients is believed to be the basic function of starch in live cells, bundle sheath cells of vessels, and pith rays in the parenchyma of xylem (Goodwin and Mercer, 1983) but the functions of starch in the xylem might not be limited to just that. Changes in starch content within the xylem in the course of a vegetative season are well known (Sudachkova *et al.*, 1997). Maximal amounts of starch are detected at the end of the growing season, decreasing considerably in the autumn, then increasing again by the end of the winter, and drastically diminishing after foliage development. These facts, though well known, appear insufficiently understood. The dynamics can be explained quite elegantly by assuming that, in addition to the nutrient-reserving function, starch also carries out water management and anti-freeze functions. Upon leaf abscission in the autumn, with the onset of frosty weather, the xylem vessels need to be spared of water or they could be damaged by ice crystals. The amount of water in the bundle sheath cells in the vessels is reduced when the starch is hydrolyzed into low molecular weight carbohydrates and this explains the disappearance of starch from xylem in the autumn. The smaller water-soluble carbohydrates take on the anti-freeze function. The gradual loss of water in the frozen trunk over the winter leads to the dehydration of low-molecular carbohydrates, which results in the reformation of starch and water. That is why the content of starch in xylem is highest at the beginning of the sap flow in spring. However, the quantity of starch at this time is directly dependent upon the amount of available carbohydrates in the xylem, the total volume of the trunk and branches, and the temperatures during the preceding winter period.

In March 2003, the determination of starch in the xylem of *B. pendula* was carried out while taking into account the overall recovery of the crown following defoliation by insects. We assigned all tree crowns to three categories: fully developed, moderate (weaker but not desiccated) and weak (desiccated). Analysis of the differences in starch distribution across xylem annual rings in *B. pendula* in conjunction with the extent of crown restoration after defoliation by insects demonstrated that a moderate or high degree of damage to the crown correlated with an increase in the number of annual rings containing starch within the section of the trunk between ground level and the lower crown. In the damaged trees there were 10-35 starch-containing rings compared to only 2-4 in those birches that had completely restored their crowns after defoliation (Fig. 1).

In March 2007, observations of starch content in the birch wood were repeated. This time trees with variable diameter (30-55 cm at base) were selected from a forest site adjacent to the

one used in 2003. Starch was not detected anywhere along the trunk. The dramatic qualitative difference is attributed to reactions to the differences in weather conditions during the winter seasons. The mean temperatures ($^{\circ}\text{C}$) and the sums of negative temperatures in 2003 and 2007 were, respectively, -10.5 and -7.1; -1629 and -1115. Thus strong frosts in 2002/2003 stimulated accumulation of starch in the xylem and a corresponding reduction of the sugar content by the end of the winter.

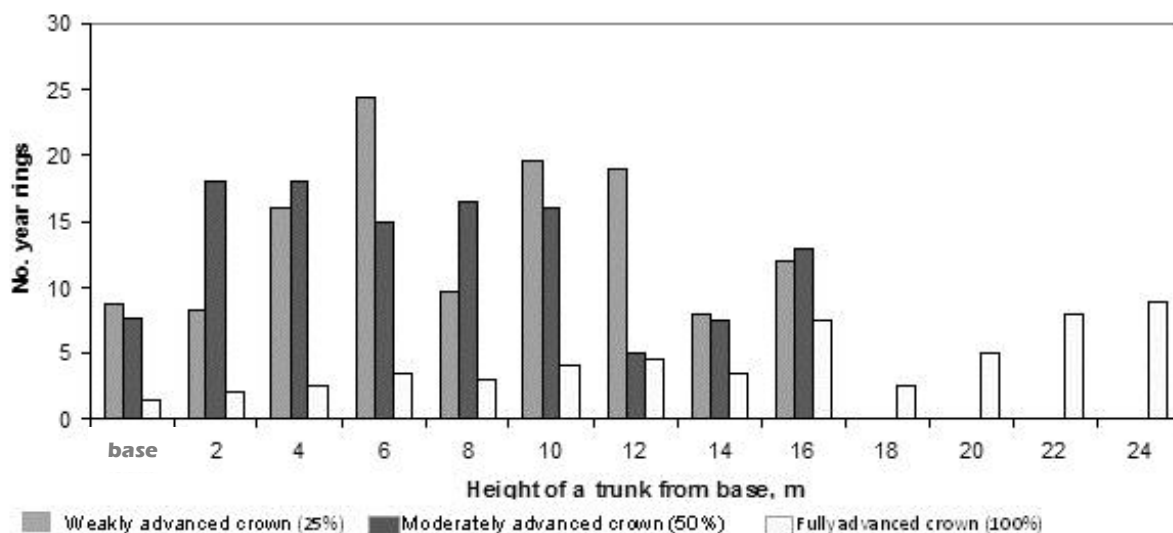


Fig. 1. The quantity of starch in birch wood, as measured by the number of annual growth rings, with respect to trunk height and crown status.

The authors propose that there is another function of starch in the xylem. Active sap flow in the spring occurs within the entire xylem or the major part of it (Ivanov, 1931; Kramer and Kozlovski, 1960; Usoltsev, 1996), and the maximum amount of water in the trunk is observed in spring, before the start of bud breaking. The question arises as to how the water is carried up the vessels in the absence of leaf suction. Due to root pressure alone, water can advance to as high as about 2 meters. Any further upward movement of water and nutrients from the root is believed to be due to narrowing and widening of vessels through the action of the parenchyma cells (Zholkevich, 2001). Further lifting of water up to the crown could also be connected with the hydrolysis of starch, whose maximum content in the trunk wood is observed at the beginning of spring. Sugars are formed during starch hydrolysis, fructose and glucose prevailing in birches (Kramer and Kozlovski, 1960). The involvement of water in the hydrolysis results in water molecules being dragged additional distances up the trunk. Thus, water transport up the trunk could be the result of a combination of processes that are not only physical and metabolic (Zholkevich, 2001) but also chemical. It is only in the spring that the

xylem sap of birch has high sugar content. Upon leaf development, translocation in the xylem is facilitated by the suction effect of transpiration. Not all of the carbohydrates contained in the sap are used for tissue construction, and the surplus goes through non-enzymatic transformations, whose products block some vessels of the xylem. The simultaneous formation, of a new annual ring compensates for vessels temporarily excluded from the water-supply system thus providing additional transportation routes. Histochemical analyses of *B. pendula* wood samples obtained during the winter and summer and treated with Lugol's iodine have shown that during these seasons starch is only present in live cells, but not in the xylem vessels. When these analyses are undertaken in spring, immediately after the leaves have fully expanded, coloration on the inside of vessel walls is observed. Starch grains can be found inside some vessels, mostly in the small ones, those formed early in the season. Wider vessels exhibit coloration less often. However, in those trees whose most recent annual rings are wide, these wide rings tend to be colored entirely, whereas in narrow rings it is mostly the late wood that reveals the presence of starch. In both cases, the annual ring of the previous year usually appears entirely colored.

Finally, the experiment, during which annual, radial growth analyses were undertaken after damage to the vessels had been inflicted during the spring sap flow, will be described (Table 1). Firstly, statistically significant, substantial (two-fold) increases in annual, radial growth in trees cut during sap flow were observed. Significant increases of radial tree growth were measured within the trunk segments ranging from 0.15 m lower than the notched area to up to 1.0 m above, around the entire circumference of the trunk. Secondly, significant changes in annual, radial growth were only observed when notches were cut during intensive sap flow. Identical treatments 7 days later, immediately after cessation of sap flow, when active transpiration and vessel flushing had started, did not result in any significant increase of radial growth, though an increase of growth directly at the notch was noticed. Thus an increase in radial growth was not merely a reaction to xylem damage but rather a reaction that was specific to the time period when the vessels contained large amounts of sugars.

Finally, the reaction extended further along the trunk above the notch than beneath it, that is, it was more significant in areas where water transport up the trunk could be attributed to the suction force of transpiration rather than the root pressure. At the same time, larger growth increases were observed below the notch, that is, in areas where trees were unable to flush vessels because of the injury to the transport system.

Table 1. Changes in annual, radial growth (in standard units) observed upon trunk notching at 1.3 meters above ground. Significant differences ($P < 0.05$) within one variant are distinguished by letter qualifiers.

Core height, orientation	Time of notching					
	During sap flow		After sap flow		Control	
	2006	2007	2006	2007	2006	2007
0.5 m, South	11.8±2.0 a	15.3±2.9 a	14.1±2.5 a	13.1±1.7 a	10.2±2.7 a	13.9±2.8 a
1.15 m, South	14.0±1.4 a	31.5±4.5 b	14.8±2.2 a	16.4±3.6 a	12.1±2.4 a	15.5±3.0 a
1.3 m, East	16.7±2.0 a	20.7±2.1 a	12.4±2.1 a	18.9±2.8 a	11.8±3.7 a	15.6±3.5 a
1.3 m, North	13.6±0.9 a	22.0±2.1 b	12.4±2.3 a	17.6±2.8 ab	11.8±3.7 a	15.6±3.5ab
1.45 m, South	15.1±3.1 a	25.0±3.8 b	10.8±1.8 a	13.0±1.7 a	11.3±2.7 a	12.3±2.3 a
2.3 m, South	14.2±1.1 a	20.5±1.9 b	-	-	-	-
4.5 m, South	13.6±1.9 a	17.8±2.0 a	11.8±2.0 a	14.7±2.3 a	12.1±2.9 a	14.5±2.5 a

In general, our data confirm that the magnitude of the radial growth is to a large extent defined by success in vessel cleaning after the spring sap flow. This is how the phenomenon of radial growth increase in trees drying out upon defoliation by herbivorous insects can be explained (Ierusalimov, 2004). Those trees that for any reason have not had enough time to flush their vessels prior to defoliation or those in which a second sap flow has begun but could not be completed because of defoliation at the end of summer are compelled to build additional vessels and thus use their remaining reserves, which results in their inability to form a good crown with an adequate photosynthetic system. As a result, tree drying occurs despite the fact that on the whole there is plenty of carbohydrates in the xylem (Ierusalimov 2004). This can probably also explain a significant correlation (0.56 – 0.75) between widths in adjoining annual rings in *B. pendula* observed in pine-birch stands within the forest zone and northern forest-steppe (Tolkach *et al.*, 2007).

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References

- Goodwin, T.W. and Mercer, E.L.** 1983. Introduction to plant biochemistry. Oxford: Pergamon Press.
- Ivanov, L.A.** 1931. Anatomy of plants. Moscow: Goslestekhizdat. (In Russian)
- Ierusalimov, E.N.** 2004. Zoogenic defoliation and forest community. Moscow: KMK Scientific Press Ltd. (In Russian)
- Kramer, P. and Kozlowski, T.** 1960. Physiology of trees. New York: McGraw-Hill Book Company.
- Sudachkova, N.E., Shein, I.V., Romanova, L.I.** 1997. Biochemical indicators of stressful condition in woody plants. Novosibirsk: Nauka. (In Russian)
- Tolkach, O.V., Sokolov, S.L., Schneider, A.** 2007. Reaction of annual, radial growth to external conditions depending on a degree of tolerance of birch forest stands. Wood Magazine 3: 14–20. (In Russian)
- Usoltsev, V.A.** 1996. Biological and ecological aspects of tree biomass estimation. Yekaterinburg: UB RAS, Yekaterinburg. (In Russian)
- Zholkevich, V.N.** 2001. Transport in plants and its endogenous regulation. Moscow: Nauka. (In Russian)