Discolouration of birch after sapping

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Abstract

Discolouration in the wood of silver birch (*Betula pendula* Roth) was studied in a 60-year-old birch stand in eastern Finland. Altogether 45 trees were analysed two and five years after sapping.

The boring hole made for sapping caused a strongly flattened, conical- shaped discolouration column down- und upwards from the hole. The discolouration spread only very slightly in the radial or the crosswise directions, but increased rapidly in the longitudinal direction. In many trees the discolouration caused by the sapping hole joined with discolouration originating from branches and butt. After five years, the estimated volume of the discolored area was almost four times bigger in these trees.

486 microbial pure cultures were isolated (191 bacteria, 224 fungi, 77 yeasts or yeast-like fungi). The samples from the base of the tree contained a larger proportion of fungal isolates than samples from the highest point of discolouration. The number of pure cultures containing bacteria and yeasts was less after five years than after two years since sapping. Even the samples from sound-looking wood contained microbes, mostly bacteria. Most of the identified fungi belonged to *Phialophora* sp. (especially *Phialophora fastigiata*). *Penicillium* sp.and *Cladospora* sp. were also common. Only three of the isolates contained suspected basidiomycetous decay fungi. Most of the identified bacteria belonged to genera *Serratia*.

Introduction

Sapping of broadleaved trees, like birch species (*Betula* sp.) has been a long tradition. Birch sap can be used for a variety of purposes. The production, composition and properties of the sap, birch syrup, have been rather intensively studied (e.g. Kallio *et al*. 1989). Sap can be collected from a bundle of narrow, cut branches, from one larger branch, or from a hole bored near the base of the trunk. The latter is the most efficient way in terms of sap production. From the forest pathological point of view, however, wounding the tree in this way unavoidably causes wood discolouration and decay later on (Vuokila 1976). Therefore, this method is commonly exploited 5–10 years prior to the felling of the trees. However, the extent or the rate of spread of the discolouration is not well known. The first colour changes in the wood are due to oxidative processes. Micro-organisms appear later, if the environmental conditions are favourable for them (Scheffer 1969, Wilhelmsen 1975). The literature on the microbial flora and its succession at the early stages of injury on birch is relatively scarce. The later stages, decay of birch trees and the microbes from decayed birches are known much better also in Fennoscandia (Björkman 1953, Henningsson 1967).

Material and methods

The study was carried out in a 60- year-old silver birch (*Betula pendula* Roth.) stand in the Koli research forest, eastern Finland (63º 7.3’ N, 29º46,7’ E). The stand was growing in a grove-like, grass-herb mineral site type (Oxalis-Myrtillus site type). The stand was born naturally after prescribed burning, and thus resembles the typical birch stands in the area. A permanent study plot was established in the stand, and three groups of log-sized trees, 20 trees in each, were selected for sapping. The trees in the groups were subjectively selected to resemble each other by their diameter, crown condition and general vigour. Conventional stand and sample tree measurements were carried out. Possible defects such as frost cracks and conks of rot fungi were also recorded.

Sapping was conducted during early summers in two consecutive years. The exact dates were from 6th May until 3rd of June in 1996 and from 12th of May until 3rd of June in1997. 30 trees were tapped in each year. A slightly upwards-slanting hole with a length of 6–7 cm was made near the base of the trunk in each tree with an incremental borer, and sap was tapped through sterilized plastic tubes. The mean height of the hole was 42 cm from the ground. The results such as sap production etc. are reported elsewhere (Salo 2000). After sapping, the holes were either i) left open ii) closed with a plug of birch wood or iii) sealed with beeswax.

Altogether 45 trees were felled two and five years after the sapping year, in 1998, 1999, 2001 and 2002, in the beginning of November. Trees with signs of external injuries or conks were rejected. The average data of the felled trees is presented in Table 1. A disc of about 10 cm containing the sapping hole was first taken. The extend of the discolouration column was then followed down- and upwards. The dimensions of the discoloured area were measured also in radial direction (i.e. the direction of the boring hole) and at right angles to it (in «tangential» direction). A disc containing the highest point of the column was also sawn.

Table 1. Average data of the felled sample trees.

Year of felling	Years from sapping	Dbh, cm	Volume,dm^3	Height,m	Crown base height, m	Crown width, dm	Number of trees
1998	2	25.54	542.98	23.08	10.12	56.00	14
1999	2	24.18	495.43	23.30	10.41	56.90	10
2001	5	20.67	360.60	22.62	10.41	47.50	10
2002	5	22.70	436.66	23.16	11.44	51.91	11

In the laboratory the two discs were aseptically dissected, and small chips of wood were cultured on malt extract agar for the isolation of microbes. The samples were taken from discolored wood just above the hole (sample a), from sound-looking wood at the same height (sample b) and near the highest point of the discolouration (sample c). The microbes were grouped, and some of the groups were identified morphologically using the identifications and descriptions e.g. in Cole & Kendrick 1973, Domsch *et al*. 1983 and Wang & Zabel 1990. Some bacterial cultures were identified by the VTT Technical Research Centre of Finland using the Riboprinter method (DuPont Qualicon, USA).

Fig. 1. A typical discolouration at the height of the boring hole, five years from sapping. The discolored area has spread a little in the radial and tangential directions.

Results

Discolouration

The boring hole made for sapping caused a very narrow, strongly flattened, conical- shaped discolouration column down- and upward from the hole. In most cases, the discolouration widened only a few millimetres in the tangential – or radial dimensions after two and five years (Fig. 1). The dimensions increased greatly, and statistically significantly, in the vertical direction between the dates (Tables 2 and 3). The column was at its widest at the height of the boring hole, narrowing quickly downwards- and also upwards within a distance of 60–70 cm. The typical shapes of the discolouration column caused by the sapping hole after five years are described schematically in Fig. 2.

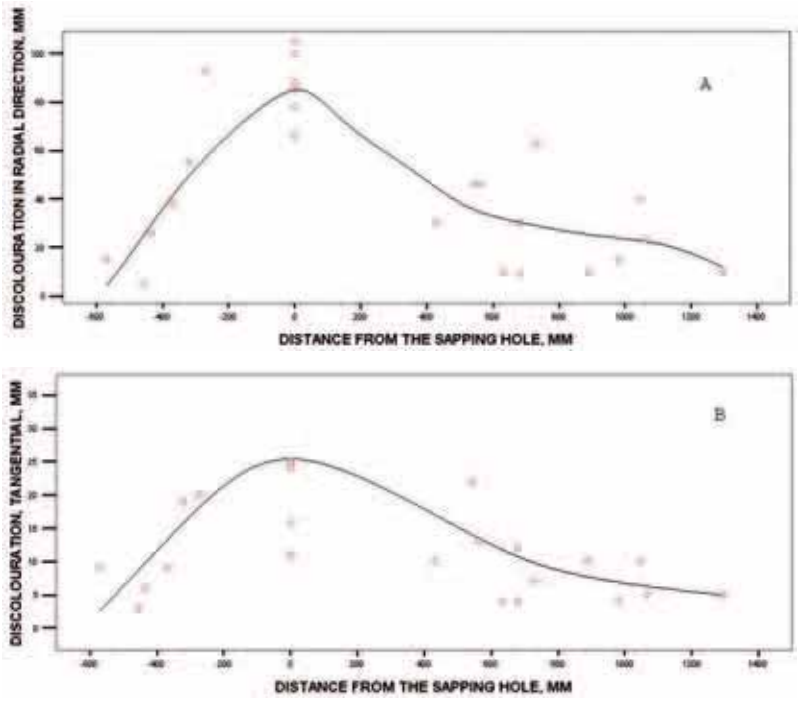

Fig. 2. Schematic presentation of the width of the discolouration column at different heights from the sapping hole. A. Radial direction B. Tangential direction

Table 2. Dimensions of discolouration two and five years after sapping. Data: all felled sample trees.

Time	Dimensions of discolouration, mean ± s.d.			
	Height, cm	Width, radial, cm	Width, tangential, cm	Volume of discolored area, cm^3
2 years after sapping	109.3 ± 93.2	4.7 ± 1.8	1.9 ± 13	
5 years after sapping	245.0 ± 243.5	6.6 ± 2.3	2.3 ± 1.3	7152.1 ± 11672.8
M-W U significance	0.004	0.013	0.059	

In 15 trees (33.3 %) the discolorated area was quite wide, sometimes also near the base of the trunk. Without exception, these were the cases where discolouration originating from branches/ branch stubs or butt of the tree joined the discolouration caused by the sapping hole. This phenomenon complicated the analyses and caused much variation in the dimensions of the discolouration. All the dimensions of the discoloured area were much smaller in the trees in which the discolouration originated from the tapping hole alone. For instance, the estimated volume of the area was 16 x smaller in these trees (Table 3).

Table 3. Dimensions of the discolouration five years after sapping.

Origin of discolouration	Discolouration five years after sapping, mean ± s.d			
	Height, cm	Width, radial,cm	Width, tangential, cm	Volume of discoloured area, cm^3
Sapping + branches and butt	402.9 ± 309.8	8.1 ± 1.8	3.1 ± 1.5	15433 ± 444.4
Only from the sapping wound	126.5 ± 47.9	3.7 ± 0.9	1.3 ± 5.4	941.4 ± 238.4

There were some differences in the dimensions of the discolouration according to the closing method. Due to the difficulties described in the previous chapter, these could not be analysed reliably in all trees. Therefore, the differences between the closing methods were not statistically significant after five years (Table 4).

Table 4. The dimensions of the discolouration by different closing method, five years after sapping

Closing method	Dimensions of the discolouration (in mm) 5 years after sapping		
	Mean height	Width, radial direction	Width, tangential direction
Control	1178	36	14
Wood	1637	63	22
Wood + wax	1095	74	33
Kruskal- Wallis Chi-Square	.831	3.568	.695
K-W significance	.660	.168	.707

Microbes

486 microbial pure cultures were obtained (191 bacteria, 224 fungi, 77 yeasts or yeast-like fungi). The greatest change between the two dates of sampling (two and five years after sapping) was the reduction in the number of cultures containing bacteria (from 183 to 65 cultures). The number of cultures containing fungi also reduced slightly, from 122 to 102. The numbers containing yeasts or yeast-like fungi were 44 and 33, respectively. After five years, 90 % the a- samples (samples from the discoloured wood just above the boring hole) contained fungi. Even the b-samples (from sound-looking wood) contained microbes, mostly bacteria, although over 40 % of them were sterile (Table 5). After five years, only 3 % of the cultures contained fungi, which were suspected to be decay fungi. These were found in trees with discolouration originating from branches.

Table 5. Proportion of microbial groups in different sampling points (a, b,c)*.

	2 years after sapping			5 years after sapping		
	a	b	c	a	b	c
	Proportion of samples containing…					
Bacteria	.88	.46	.63	.38	.29	.57
Fungi	.63	.00	.21	.90	.00	.33
Yeasts	.54	.04	.29	.38	.19	.48
Sterile	.08	.42	.33	.05	.43	.24

*The samples were taken a) from discolored wood just above the hole, b)from sound-looking wood at the same height, c) and near the highest point of the discoloration

Phialophora sp. was the most common of the fungal genera (65 isolations). Some of these resembled morphologically *Phialophora fastigiata* (Lagerberg & Melin) Conant (Fig. 3). 51 (80 %) of the *Phialophora* sp. samples were obtained in sampling point a. *Penicillium* sp. (in 21 cultures) and *Cladosporium* sp. (in 8 cultures) were also common fungal genera. Yeasts and yeast-like fungi were also common, but it was not possible to identify them at this stage. Moreover, it was very difficult to separate bacteria/fungi/ yeasts in some samples with conventional culturing- subculturing methods (e.g. dilution plates etc.).

Fig. 3. The most common fungal isolate, morphologically identified as Phialophora fastigiata*, with funnel-shaped collaret's (1000 x).*

Of the samples taken 2 years after sapping, 18 bacterial pure cultures were selected for identification with the Riboprinter method. 10 of these were identified as *Serratia proteamaculans* subsp. *quinovora*. The proper name should now be *Serratia quinivorans* (Ashelford *et al.* 2002). Five of the isolates remained unidentified, and the remaining three were *Serratia proteamaculans* subsp. *proteamacula, Rahnella aquatilis* and *Hafnia alvei*.

Discussion

The present study gives support to the hypothesis that bacteria, yeasts, and other nonhymenomycetes are the primary colonists of discolored tissues. Most likely the early colonizers such as non-decay fungi (*Phialophora*) alter cell wall components, and degrade wound-initiated vessel plugs The may also modify phenolic substances in the reaction zone. All these primary degradations may modify wood xylem sufficiently for the decay fungi to break down the main part of the cell walls (lignin and cellulose). Mutualistic associations of bacteria and yeasts with wood-destroying hymenomycetes are also possible, since Basidiomycetous hyphae have been observed only in tissues where amorphous vessel deposits had been degraded by pioneer microorganisms (Shortle & Cowling 1978, Blanchette & Shaw 1978, Blanchette 1979). *Phialophora*- species have been found to be the predominant non-decay fungal species in wood a long time ago (Shigo 1967, Stewart *et al.* 1979).

Serratia appears to be a ubiquitous bacterial genus in nature, and ten species are currently recognized. Serratia species have been isolated from water, soil, animals (including man), and from plant surfaces (Grimont & Grimont, 1992). Their role in the discolouration process of wood is however unknown to the author.

There was no indication that the wounds made for sapping are infected by typical decay fungi of birch in this study. Hallaksela and Niemelä (1998) did not find typical birch decayers in their study on planted silver birch either, although some decay fungi were isolated from discolored wood. Lilja and Heikkilä (1995) found decay fungi, esp. *Chondostereum purpureum* in older defects in young birch trees broken by moose. *Phialophora fastigiata* was a common isolate in their material, and it also grew together with bacteria.

The results of this small-scaled study showed that the boring hole made for sapping caused only a minimal risk to the technical quality of the birch trees after five years, assuming that there are no other pathways for the infection of decay fungi.

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