SILVA FENNICA

Silva Fennica 41(2) research articles

www.metla.fi/silvafennica · ISSN 0037-5330
The Finnish Society of Forest Science · The Finnish Forest Research Institute

Decomposition of Fine Roots and α-Cellulose in a Short Rotation Willow (*Salix* spp.) Plantation on Abandoned Agricultural Land

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Püttsepp, Ü., Lõhmus, K. & Koppel, A. 2007. Decomposition of fine roots and α-cellulose in a short rotation willow (*Salix* spp.) plantation on abandoned agricultural land. Silva Fennica 41(2): 247–258.

Decomposition of fine roots (<1 mm in diameter) of the clones of *Salix viminalis*, *S. dasyclados* and α-cellulose sheets (50 x 10 x 1 mm) was studied in a 6-years old *Salix* spp. plantation established on abandoned agricultural land in Estonia. The substrates were incubated in litterbags (mesh size 0.14 mm) in 5–10 cm topsoil, in non-fertilised plots for one year. Changes in the ash-free weight of the fine roots were best described by negative exponential models (*S. viminalis* $R^2=0.98$, *S. dasyclados* $R^2=0.96$), and by a linear model for α-cellulose ($R^2=0.63$). The sheets of α-cellulose decomposed roughly twice as rapidly as the fine roots (*S. viminalis* $k=0.325$, *S. dasyclados* $k=0.165$). The remaining (of the initial) ash-free weights of the fine roots were 73.3±0.8% (mean±SE) and 85.8±2.2% respectively, and of the α-cellulose 35.9±8.5%, in the end of the one year of decomposition. The amount of acid detergent (AD) lignin in the fine-roots of *S. viminalis* increased significantly and did not change in *S. dasyclados*, suggesting higher activity of microbial decomposers in the first substrate. Of the studied quality parameters, the AD lignin was the major factor determining the different rate of decomposition of the fine roots of *S. viminalis* and *S. dasyclados*. Nitrogen was recycled in the fine root sub-system in both *Salix* species. This knowledge can be applied in the management of *Salix* plantations, aimed at bioenergy production.

Keywords α-cellulose, decomposition, fine roots, acid detergent lignin, short rotation forest, *Salix* spp.
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Received 15 May 2006 **Revised** 26 February 2007 **Accepted** 1 March 2007
Available at http://www.metla.fi/silvafennica/full/sf41/sf412247.pdf

1 Introduction

Input of organic matter and nitrogen from dead fine roots to the soil often exceeds the amount produced and supplied to the soil from litterfall (Persson 1978, Vogt et al. 1986). In short rotation forestry (SRF) plantations of *Salix*, fine roots can contribute remarkably to carbon and nutrient cycling with turnover rates as high as 4.9–5.8 $year^{-1}$ (Rytter and Rytter 1998) and about 40% of annual net primary production goes belowground, mostly into the fine roots (Rytter 2001). However, nitrogen and carbon cycling in the process of fine-root decomposition in forest ecosystems has been studied less than that of litter shed from the above ground parts of trees (e.g., Persson 1979, Persson 1980, Berg 1984, McClaugherty et al. 1984, Santantonio and Grace 1987, Lõhmus and Ivask 1995, Berg et al. 1998, McEnroe and Helmisaari 2001, Berg and McClaugherty 2003). Currently no publications either about *Salix* fine-root decomposition or fine-root decomposition in SRF are available.

Energy crops constitute the largest biomass potential in Europe in conditions of agricultural overproduction and expanding use of local energy resources (Ericsson and Nilsson 2006). In SRF, *Salix* spp. clones are most widely grown for bioenergy production in Northern Europe. SRF can serve as an alternative crop on former arable lands. In Estonia, the total area abandoned from agricultural use is currently estimated at 400 000 ha (www.stat.ee). The first experimental SRF plantations with *Salix* spp. clones were established in Estonia in 1993 (Koppel et al. 1996). Afforestation of former fields with SRF crops induces changes in the chemical, physical and biological soil parameters, mainly due to less destructive management practices (Jug et al. 1999). The immediate effect after grassland afforestation can lead to losses of organic carbon and total nitrogen in the topsoil due to better aeration after ploughing (Jug et al. 1999). As the leaf and root litter accumulates, humus concentrations increase near the soil surface and the C/N ratios can increase to >20 (Jug et al. 1999). The dynamics and rate of decomposition, as a base for nutrient cycling in the new ecosystems, are yet largely unknown.

Climate, the quality of litter, and the decomposer communities are the main interacting factors determining the rate of decomposition (Swift et al. 1979). Nitrogen content in the decomposing material controls mass loss rate during the first phase of decay, cellulose and particularly lignin content become progressively more important in controlling decomposition rates in later phases (Berg 1984, Melillo et al. 1989, Taylor et al. 1989). Berg and Staaf (1981) distinguished leaching, accumulation, and the final release phases in nitrogen dynamics during decomposition. The release after accumulation starts when nitrogen concentrations have reached a level covering the needs of the microbial decomposers. This critical level of nitrogen varies with the litter, the stage of decomposition, and the ecosystem (Berg and Staaf 1981). Estimated critical N concentration is 1.4 to 1.7% for most litter types (Granhall and Šlapokas 1984, Ågren and Bosatta 1996), but 1.2% has also been reported for fine roots (Pluth et al. 1995).

We used litterbag method to compare the decomposition of fine roots (d<1 mm) of *Salix viminalis* and *S. dasyclados* clones in a SRF plantation, established on abandoned agricultural land in Estonia. The litterbag method is the most widely used technique of measuring decomposition (cf. e.g., Berg 1984, McClaugherty et al. 1984, Lõhmus and Ivask 1995, Gregorich and Janzen 2000, McEnroe and Helmisaari 2001). We also investigated the decomposition of α-cellulose sheets using the same litterbag method, to estimate the 'ability' of a soil to decompose cellulose, a main plant constituent. The dynamics and rate of pure cellulose decomposition is directly affected by the local community of (microbial) cellulose decomposers, and can be considered as an edaphic parameter (Swift et al. 1979).

In this study we aimed to:

1) compare the dynamics of remaining ash-free weight, N, AD (acid detergent) cellulose and AD lignin in decomposing fine-roots of two *Salix* clones;
2) estimate whether N is lost during the first year of fine-root decay, and the critical N levels for the two fine-root substrates;
3) estimate the utilisation rate of the substrate as a measure of functional activity of cellulose decomposers in the same plantation.

2 Materials and Methods

2.1 Site Description

The fine-root decomposition experiment was carried out in a SRF plantation of *Salix viminalis* and *S. dasyclados* in Saare, Central-Eastern Estonia, 58°42´ N and 26°55´ E, over the one-year-period from August 1998 to August 1999. The plantation was established in 1993 on abandoned agricultural land (previously used as a meadow), representing mineral soil of brown Gleyic Podzoluvisol type (FAO-UNESCO, 1988) with a sandy loam texture. The soil was poorly aerated during seasons with heavy rains, due to limited drainage (Koppel et al. 1996). The samples for soil analyses were derived from 20 randomly taken cores (d = 2 cm) collected in October 2000 (pH, org. matter, N) and from 5 to 10 randomly taken cores (d = 4 cm) collected in August 2002 (P, K), from the upper 0–10 cm of the soil. Soil pH was measured in 0.01 M KCl or water, using a 1:2 (w/v) soil:liquid ratio. Content of organic matter was determined on water-free soil by heating at 360 °C. Concentration of total N was measured using Kjeldahl method. Concentrations of extractable P and K were measured using the Egner-Rhiem AL method, based on lactic acid (Mocek et al. 1997). Values for the main characteristics of the soil are shown in Table 1.

Cuttings of *S. viminalis*, clone 78183, and *S. dasyclados*, clone 81090 (obtained in collaboration with the Swedish Energy Forest Programme, Koppel et al. 1996) had been planted with a randomised block design on an area of 0.61 ha. The planting density was an average of 2 cuttings per m^2. The *Salix* project in Estonia is described in details in Koppel et al. (1996). The *Salix* shoots were cut in May 1994 to stimulate denser sprouting, and harvested in March 1998 (Heinsoo et al. 2002). Four months later, when the experiment started, the canopy was already closed after intensive re-sprouting. We ran the experiment in the SRF *Salix* plantation during the 6^{th} year after its establishment. The *Salix* clones are referred to by their species names for simplicity, not inferring that the results apply in general to the species.

The length of the growing season (mean air temperature > 5.0 °C) was normal for the region during the course of the experiment. It lasted for 193 days, from April 18 to October 28, in 1998 (mean air temperature 12.0 °C), and for 190 days, from April 8 to October 15, in 1999 (mean air temperature 13.3 °C). Total precipitation was 800 mm in 1998, which was higher than the average 660 mm for the 1966–1998 period. In 1999 the total precipitation was 665 mm. The average soil temperature was 10.7 °C, measured in the 0–10 cm soil layer during the first stage of the experiment, from August 1 to October 31, 1998, before the cold season. Below zero temperatures prevailed from November 8, 1998 to March 29, 1999, making the duration of the winter period 142 days. The climatic data was gathered at the nearest (Jõgeva) meteorological station, 25 kilometres from the study site. The data for the winter period was gathered at the Tartu station, 50 km from the study site.

Table 1. Soil characteristics in the study site, in the SRF plantation of *Salix viminalis* and *S. dasyclados*. The values represent a composite sample per non-fertilised plot.

	Salix viminalis	*S. dasyclados*
pH (KCl)	5.4	5.2
pH (H_2O)	6.1	6.1
Organic matter %	1.69	2.42
N %, total	0.081	0.104
C : N	12.1	13.5
K (mg kg^{-1}), extractable	79.9	70.4
P (mg kg^{-1}), extractable	65.3	32.7

2.2 Incubation of Fine Roots and α-Cellulose in Litter-Bags

Fine roots (d < 1 mm) of *S. viminalis* and *S. dasyclados* were collected from in-growth cores at the beginning of November 1996 and 1997, after one and two growing seasons, respectively (K. Heinsoo, Estonian University of Life Sciences, unpublished). The roots were washed and bulked into a composite sample for each *Salix* species, and dried at 80 °C for 24 hours. A composite sample of each species was made and approximately 300 (±0.5) mg of fine roots were placed into 5 cm × 5 cm nylon mesh bags. The small volume of the fine root samples enabled better contact with the

soil, minimising the environmental differences in the litterbags in regard to the ambient. Mesh size was 0.140 (±0.003) mm, selected according to the diameter distribution of the fine roots and to enable the participation of microflora, microfauna and smaller mesofauna but to exclude the larger mesofauna and the macrofauna (Swift et al. 1979) in the process of decomposition. According to the estimate made in August 1999 nearly 94% of the root tips of *S. dasyclados* and 75% of those of *S. viminalis* were ectomycorrhizal (Püttsepp et al. 2004). Thirty sample bags were positioned along random transects across the respective plots of each *Salix* species, in the top 5–10 cm soil layer characterized by high rooting density.

To characterise the potential functional activity of the microbial cellulose decomposers, we buried thirty α-cellulose sheets (50 mm × 10 mm × 1 mm) with a mass similar to that of the fine root samples, approximately 300 (±0.5) mg, in mesh bags of 5 cm × 5 cm. The α-cellulose sheets were positioned along a random transect only in the *S. viminalis* clone plot. The advantage of using α-cellulose sheets as test material to distinguish the impact of environmental factors from substrate quality effects on decomposition rates (French 1988, Kurka 2001) is that they are easy to handle, homogeneous, and chemically definable (Brække and Finér 1990).

2.3 Sampling and Analyses

All the bags with fine roots and α-cellulose sheets were placed into the soil on August 6, 1998. Five to six samples of fine roots of each *Salix* clone and of α-cellulose sheets were collected after 78 days (October 24, 1998), 300 days (June 3, 1999), and 358 days (July 31, 1999) of incubation. The first collection was carried out before the cold season, the next one once the new vegetation was fully developed, and the third in the midst of the growing season.

Each sample was cleaned from the adhering soil particles and ingrown roots and then dried at 80 °C for approximately 24 hours and weighed. The material from the 5 to 6 samples collected at the same time at each plot was mixed to form a composite sample. About 300 mg of the material of one composite sample was incinerated in a furnace at 500 °C for 4 hours. The dry weight of the remaining ash was measured. The high ash content of *Salix* fine roots in the initial samples (Table 2) indicates soil contamination, since, due to their small diameter, the roots were difficult to clean.

Table 2. Substrate quality of the fine roots of *Salix viminalis*, *S. dasyclados* and α-cellulose prior to the decomposition experiment. For all the parameters except ash, the values represent their concentrations per ash-free material.

%	*Salix viminalis*	*Salix dasyclados*	α-cellulose
Ash	31.50	31.00	0.00
N	1.51	1.66	-
P	0.31	0.32	-
AD cellulose	24.60	25.06	97.51
AD lignin	25.44	35.13	0.10

- Not measured

Ash, N, AD (acid detergent) cellulose and AD lignin concentrations were estimated as parameters of substrate quality. The concentrations of Kjeldahl-N (Tecator AN 300), AD lignin and AD cellulose were measured in composite samples of fine roots for *S. viminalis* and *S. dasyclados* and in the α-cellulose sheets. All concentrations were recalculated per ash-free weights. For AD cellulose and AD lignin analyses the forage fibre technique was used with the equipment and methods by Tecator (AN 304, ASN 3436, ASN 3430). Fractions of AD fiber and AD lignin were determined successively using sulphuric acid (AOAC method no. 973.18, 1990) and by calculating AD cellulose as AD fiber – AD lignin. The initial quality of fine root substrates and α-cellulose is characterized in Table 2. All chemical analyses were processed at the Laboratory of Plant Biochemistry of the Estonian University of Life Sciences at Tartu.

To estimate initial physical leaching from comparable *Salix* fine roots, we ran an ultrasonic experiment in the laboratory to detect loss of ash-free weight and water extractable nitrogen compounds. For approximate estimation of water-solubles the fine-roots (< 1 mm) of four 1.5 month-old pot-grown cuttings of *Salix viminalis* were used for ultrasonic treatment in laboratory conditions. The initial N concentration of the pot-grown *S. vimi-*

nalis fine roots (1.5%) was similar to that of the incubated fine root material in the field experiment. The fine roots were washed to remove the soil particles, and dried at 85 °C until they reached constant weight. The roots were cut into pieces of ≤5 mm to maximise extraction. Approximately two thirds of the root material was put into ~50 ml of de-ionized water in a beaker and processed in an ultrasonication bath for 3 × 20 minutes at 50 kHz (cf. Granhall and Šlapokas 1984). After each cycle of 20 minutes the de-ionized water with the root pieces was filtered and renewed in the beaker. The treated root material was dried at 85 °C until constant weight. The initial and remaining ash-free weights and the concentrations of ash and N in the samples were measured.

2.4 Statistics

We processed the statistical analysis using the STATISTICA program (STATISTICA 6.0, StatSoft, Inc. 1999). Proportional variables were obtained by dividing the content of nitrogen, AD cellulose or AD lignin in a sample at time *t* by the content of nitrogen, AD cellulose or AD lignin in the initial sample. Normality of variables (proportional nitrogen, AD cellulose, and AD lignin content) was checked using the Lilliefors' test. To analyse the effect of qualitative factors on response variables, one-way ANOVA was applied, since, in all cases, group variances were homogeneous and no significant relationship between group means and standard deviations was found. Tukey HSD (honest significant difference) test for unequal n (Spjotvoll-Stoline test) was used to compare group means for proportional lignin content at different sampling times (days of incubation). Different regression models were used to describe the decomposition dynamics of fine roots (heterogeneous substrate) and α-cellulose (homogeneous substrate). Following symbols were used in these equations:

- t time
- M_0 ash-free dry weight of the initial sample
- M_t ash-free dry weight of the sample at time t
- a, b parameters of an individual model
- k negative exponential function of time, rate of decomposition
- e base of the natural logarithm

Remaining ash-free weight of the fine root samples was described by the negative exponential model (1).

$$\frac{M_t}{M_0} = e^{-k(t)*t} \quad (1)$$

where k is expressed as

$$k(t) = ae^{-bt} \quad (2)$$

Remaining ash-free weight of the α-cellulose sheets was described by the linear model (3).

$$\frac{M_t}{M_0} = a + bt \quad (3)$$

The level of significance $\alpha = 0.05$ was accepted.

3 Results

3.1 Ash-free Weight Dynamics

The decomposition pattern for the fine roots and α-cellulose was different. Fine roots of *Salix viminalis* and *S. dasyclados* decomposed along a negative exponential model (1) during one year (Fig.1, Table 3). Decomposition was faster at the beginning and retarded towards the end of incubation.

In the instance of the ash-free weight dynamics of α-cellulose, a linear model (3) provided a better fit ($R^2 = 0.63$, $p < 0.0003$) than a negative exponential model (1), which was also estimated ($R^2 = 0.40$, k = constant, $p < 0.05$). The variability

Table 3. Regression summaries for the ash-free weight dynamics in decomposition of the fine roots of *Salix viminalis*, *S. dasyclados*, based on model (1). The value for k (Eq. 2) was calculated for one year. The level of probability $p < 0.000001$ in both cases.

Fine root substrate	a	$b_{1\ year}$	R^2	$k_{1\ year}$
S. viminalis	0.00665	2.011	0.98	0.325
S. dasyclados	0.00658	2.695	0.96	0.162

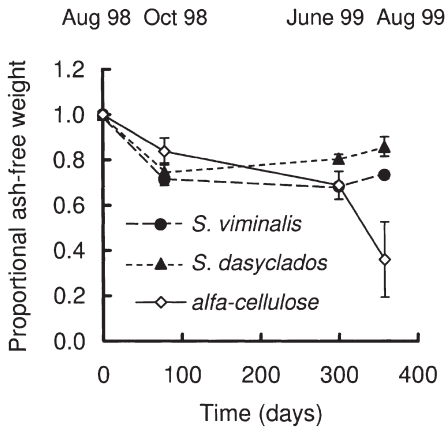

Fig. 1. Changes in proportional ash-free weight (in relation to the initial, M_t / M_0) in the fine roots of *Salix* spp. and α-cellulose during decomposition from August 1998 to August 1999. The substrates were incubated in mesh-bags in the top 5–10 cm of soil, in a non-fertilized *Salix* plantation. Error bars are 95% confidence limits of the means.

between the ash-free weights of the individual samples was high on the last sampling occasion (coefficient of variation = 53%). The remaining ash-free weight of the α-cellulose in the end of the one year of decomposition was 35.9 ± 8.5% (mean ± SE) of the initial. The remaining ash-free weights of the fine roots of *S. viminalis* were 73.3 ± 0.8% (mean ± SE) and of *S. dasyclados* 85.8 ± 2.2% in the end of incubation.

Comparing the fine roots of the two *Salix* species at the first sampling (after 78 days of incubation, October 24), both substrates had lost 25–29% of their initial ash-free weight, but no difference between the species was revealed (Fig. 1). The period between the first and the second sampling (after 300 days of incubation, June 3) covered the winter and the following spring. However, the slight changes registered after 300 days of incubation – insignificant for the individual *Salix* species – gave a significant difference between the species. Consequently, at the second sampling the fine roots of *S. viminalis* had decomposed faster. At the last sampling (after 358 days, July 31), an increase (compared with the previous sampling) in the ash-free weights was found in the litterbags; this was statistically significant for the *S. viminalis* substrate and insignificant for *S. dasyclados*.

When comparing *k* values for the changes in relative ash-free weights calculated for a year (Table 3), the fine roots of *S. viminalis* decomposed approximately twice as rapidly as the fine roots of *S. dasyclados*.

Estimating the share of water-solubles in fine-roots, the pot-grown *Salix viminalis* roots lost 10.1% of their ash-free dry weight after ultrasonic treatment.

3.2 Nitrogen Dynamics

The proportional nitrogen content in the incubated fine roots passed through a stage of decline, accumulation, and decline (Fig. 2a). Unfortunately at the first sampling (after 78 days of incubation), the results of N concentration measurements were unreliable. Instead, we used the results of the ultrasound treatment of the pot-grown *S. viminalis* fine roots as an estimate of physically regulated N loss. The pot-grown fine roots lost 14.8% of their initial N content after ultrasound treatment. At the second sampling in the field, after 300 days of incubation, N accumulation close to the initial values (Table 2), to 1.71% in *S. viminalis* and to 1.65% in *S. dasyclados*, had occurred in the fine root samples. At that time a significant difference appeared between the remaining N contents (Fig. 2), followed by the decrease of the amount of N in the fine roots, significant in *S. viminalis*, by the last sampling.

Nitrogen concentration, initially zero in α-cellulose sheets, increased in a linear pattern during 300 days after incubation to 0.35% of concentration (Fig. 2), and was linearly and negatively correlated to the ash-free weight loss of the sheets ($R = -0.62$, $p = 0.003$). After 358 days of incubation the N content had decreased significantly. The amount of nitrogen in the individual α-cellulose sheets became highly variable at the end of incubation (coefficient of variation = 53%).

3.3 AD Cellulose Dynamics

The decomposition of AD cellulose in α-cellulose sheets was remarkably greater than in the

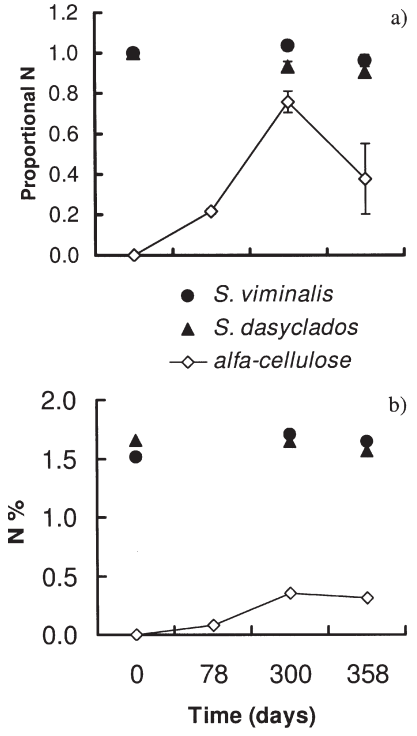

Fig. 2. Changes in a) proportional nitrogen content (in relation to the initial) and b) in nitrogen concentration in the fine roots of *Salix* spp. and α-cellulose during decomposition from August 1998 till August 1999. The substrates were incubated in mesh-bags in the top 5–10 cm soil in a non-fertilised *Salix* plantation. Error bars are 95% confidence limits of the means.

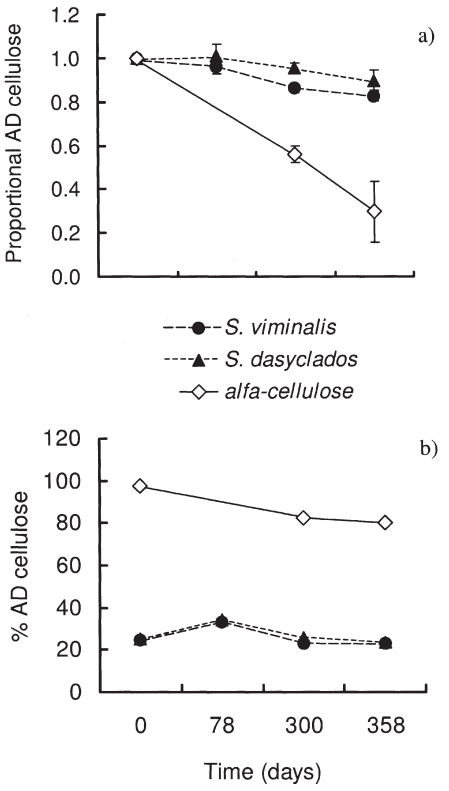

Fig. 3. Changes in a) proportional AD cellulose content (in relation to the initial) and in b) AD cellulose concentration in the fine roots of *Salix* spp. and in α-cellulose during decomposition from August 1998 till August 1999. The substrates were incubated in mesh-bags in the top 5–10 cm soil in a non-fertilised *Salix* plantation. Error bars are 95% confidence limits of the means.

fine roots (Fig. 3a). Linear models best described the changes in the relative AD cellulose content of *Salix* fine roots and of α-cellulose (Fig. 3a, $p<0.005$ for all the substrates). However, the model gave high R^2 values for *S. viminalis* ($R^2=0.81$) and for α-cellulose ($R^2=0.93$), but low for *S. dasyclados* ($R^2=0.41$). A difference in fine-root AD cellulose decomposition between the *Salix* species was revealed after 300 days of incubation. This difference decreased and became non-significant on the last sampling (358 days from the start of incubation) when the mean relative remaining AD cellulose amount in fine roots was 82.6±1.2% (mean±SE) in *S. viminalis* and 89.8±2.5% (mean±SE) in *S. dasyclados*, from the initial. The remaining proportional AD cellulose amount in the α-cellulose sheets was 29.8±7.0% (mean±SE) from the initial on the last sampling. Decreases of AD cellulose content and of ash-free weight in the α-cellulose sheets were highly correlated ($R=0.97$, $p<0.000001$).

3.4 AD Lignin Dynamics

The initial AD lignin concentration was higher in the *Salix dasyclados* fine root material than in *S. viminalis* (Table 2, Fig. 4b). No statistically

significant changes in AD lignin content were revealed in the *S. dasyclados* substrate during the incubation period (Fig. 4a). In the *S. viminalis* fine-root substrate, on the contrary, the AD lignin content increased significantly during the first year of decomposition, up to 40% of initial by the third sampling (300 days after start of incubation).

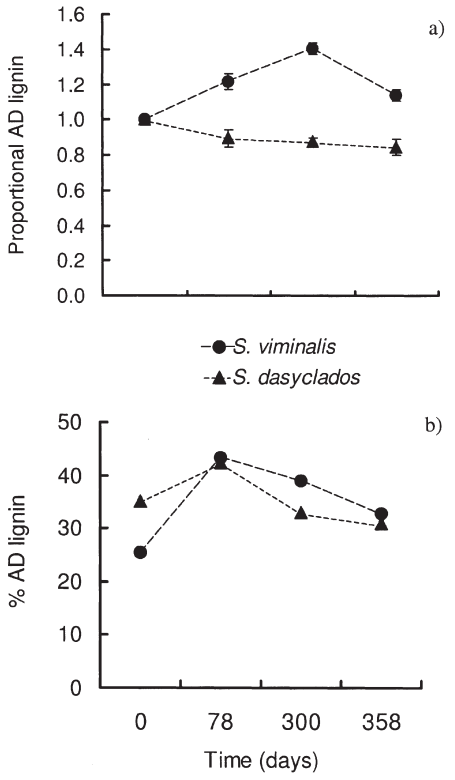

Fig. 4. Changes in a) proportional AD lignin content (in relation to the initial) and b) in AD lignin concentration in fine roots of *Salix* spp. during decomposition from August 1998 till August 1999. The fine roots were incubated in mesh-bags in the top 5–10 cm soil in a non-fertilised *Salix* plantation. Error bars are 95% confidence limits of the means.

4 Discussion

The litterbag method combined with chemical analyses is useful in showing the dynamic processes in substrate quality. As a rule, decomposition rate k is decreasing with time non-linearly (Lõhmus and Ivask 1995, Berg and McClaugherty 2003). Hence, negative exponential model in form (1) providing the best fit, was used. The weight alone may not adequately reflect the decomposition process of organic matter owing to simultaneous import and export of compounds (Cotrufo et al. 2000). Decomposition takes place even if the weight loss is unremarkable (Lõhmus and Ivask 1995).

For equalising the substrates the fine roots were dried at 80 °C in order to kill most of the natural microflora attached to them. Drying probably delayed the beginning of decomposition until the fine roots became re-inhabited by soil microflora. The incubation period until the first sampling was however, sufficiently long (78 days) and unlikely influenced the results. Previous drying of the initially live fine roots of Norway spruce (*Picea abies*) did not cause a severe retarding effect on decomposition in a similar experiment 6 km from our study site, reported by Lõhmus and Ivask (1995). In their study, the fine spruce roots lost 21 to 33% of their dry weight after one year of incubation in different plots. The fine roots of *S. viminalis* had decomposed 27% and those of *S. dasyclados* 14% after one year of incubation. The sheets of α-cellulose had decomposed, on average, roughly twice as rapidly as the fine roots (Fig. 1, Table 3).

Treatment of the fine roots of *S. viminalis* with ultrasound in the lab resulted in a loss of ash-free weight of 10.1%. The ultrasound procedure physically removed the soluble compounds, approximately comparable to the initial 'leaching' phase in the field (cf. Granhall and Šlapokas 1984). *S. dasyclados* and *S. viminalis* in the field lost respectively 25% and 29% of ash-free weight during the first 78 days (Fig. 1). Different origin of the roots can partly explain the difference between the lab and the field results. The initial N concentrations of both field- and pot-grown roots were however, similar. The contribution by microbial decomposition in the early phase in the field cannot be excluded (cf. McClaugherty et al. 1984).

Roots grown into the litterbags from outside were separated out after each sampling. The con-

tent of AD cellulose, the main constituent of the roots, slightly decreased at the last sampling in the fine root substrates. This suggests that the increase of ash-free weight at the last sampling should most likely be attributed to the microbial decomposers (Fig. 1, Fig. 3a).

The different decomposition rate of the fine-root litters of the two *Salix* species can be related to different initial concentrations of AD lignin and to different ectomycorrhizal colonisation.

The initial AD lignin concentration in *S. dasyclados* fine roots was sufficiently high compared to that of *S. viminalis*, to cause completely different pattern of AD lignin dynamics during decomposition (Fig. 4a,b). Lignin undergoes modification during decomposition, and the analytical residue fraction of lignin originates not only from the native lignin but also from other phenolic compounds, carbohydrates, and various nitrogen compounds (Berg and Theander 1984). In plant fibres the cellulose, hemicellulose and lignin are not only combined physically but celluloses are more or less encrusted with lignin (Kerr and Goring 1975, Berg and McClaugherty 2003). The initial AD cellulose concentrations were similar in the two fine root substrates but the decomposition progressed more in the fine roots of *S. viminalis*. We can assume that in the fine roots of *S. viminalis*, at the initially lower AD lignin concentration, the decomposers had better accessibility to celluloses. Hence, there may have been higher microbial activity and more effective modification in the lignin compartment, indicated by increasing AD lignin content. Granhall and Šlapokas (1984) found a similar increase in the analytical lignin fraction during decomposition in the leaf litter of *Alnus incana*, *Betula pendula*, *Salix caprea* and *Salix* sp. clone 082 (in a Swedish SRF plantation). These litters had low initial concentrations of analytical lignin compared with the *Salix* sp. clone 077, in which the lignin concentration decreased during decomposition (Granhall and Šlapokas 1984).

Ryan et al. (1989) were critical concerning the forage fibre method of cellulose and lignin detection. Control estimation of the initial material of α-cellulose in our work showed no essential underestimation of fibre concentration in the substrate (97.5% of cellulose, 0.1% of lignin).

Ectomycorrhizal (EM) colonisation changes root structural and chemical properties and influences root decomposition rates (Langley and Hungate 2003). The EM hyphal sheath provides protection from the heterotrophic decomposers and the relatively high amount of recalcitrant chitin in fungal tissues decreases the decomposition rates of EM roots compared with non-EM roots (Langley and Hungate 2003). Although not studied in this experiment, we know that the EM colonisation in root tips was higher in *S. dasyclados* (94%) than in *S. viminalis* (75%) (Püttsepp et al. 2004), predicting lower decomposition of the first (Langley and Hungate 2003, Langley et al. 2006).

In general, the N-status of *Salix* roots depends on the season and on the developmental stage of the plant or the root (Bollmark et al. 1999, von Fircks et al. 2001). Initial N concentrations in fine-roots of the two *Salix* species remained in the range of the critical values reported for most litters (Pluth et al. 1995, Ågren and Bosatta 1996). Assuming that the loss of N from the fine-roots at the ultrasonic treatment can parallel with the field situation at the first sampling, the N dynamics followed a similar pattern of decrease, accumulation and decrease in both *Salix* substrates, accordingly to the model presented by Berg and Staaf (1981). After N microbial immobilisation to its critical values by the second sampling, 300 days after incubation, the N contents decreased, though significantly only in *S. viminalis* (Fig. 2a).

High utilisation of the homogeneous substrate of α-cellulose during one year indicated the high activity of microbial cellulose decomposers in the former agricultural land. The final drop in the proportional content of N, in ash-free weight and cellulose in α-cellulose (Fig. 1, 2a, 3a) indicated a change in the microbial growth-pattern. When more than half of the substrate carbon is consumed the total microbial carbon decreases (Wagner and Wolf 1998). That turning point occurred between the samplings of 300 and 358 days from incubation and concurred with the release of N.

The quantities of microbial biomass in the 0–10 cm soil in the respective plots of *S. viminalis* and *S. dasyclados* were estimated as substrate-induced respiration (SIR) per soil organic C (data not shown). The substrate-induced respiration was 1.2 times higher in the plot of *S. viminalis* than that in the plot of *S. dasyclados*. This value

supports our suggestion of higher microbial activity in the plot of *S. viminalis*.

The soil in the *Salix* plantation was in transitional stage from agricultural to forest soil, as indicated by the intermediate C:N ratios (Table 1). Agricultural soils and forest soils have different decomposition potentials because of the different inputs of litter, with regard to amount and quality, but also due to varying site management practices, all factors that have impact on the communities of decomposers. Decomposition is more rapid in agricultural soils compared to forest soils (Gregorich and Janzen 2000). The woody crop plantations like SRF, mainly established on previous agricultural land, seem, already within some years, to develop a type of nutrient cycling that more resembles forest ecosystems (Svensson et al. 1994). In this study, the low decomposition rate of fine roots and the N dynamics support that suggestion. Estimated mass losses during the first year of decomposition (27% in *S. viminalis* and 14% in *S. dasyclados*) remain within the limits given for fine roots in a variety of forest ecosystems of boreal and temperate zone (Berg and McClaugherty 2003). In fertilised arable soils N is often leached out (Paustian et al. 1990). Our study on non-fertilised plots showed that N was to a great extent recycled in the fine-root system.

5 Conclusions

The substrate of fine roots can act as alternating sink and source for N, demonstrated by the decrease and immobilisation of N in the fine root substrates of *S. viminalis* and *S. dasyclados*. This knowledge is important for management, since N in decomposing fine roots is retained in the system, which implies lowering risk of N to be leached out in the non-fertilised SRF plantation.

The quality of the substrate was an essential factor in determining the rate of decomposition. The homogeneous α-cellulose sheets decomposed twice as rapidly as the heterogeneous fine-root material during one year, indicating that the cellulose decomposers did not inhibit decomposition. The initial concentration of AD lignin in the fine roots of *S. dasyclados* was high and the rate of decomposition low ($k = 0.162$), whereas the opposite was true for *S. viminalis* ($k = 0.325$). The higher decomposition rate, the increase in the amount of AD lignin, and the lower ectomycorrhizal colonisation in *S. viminalis* fine roots, gave indirect evidence of higher levels of activity in the community of microbial decomposers in the plot of *S. viminalis* than in that of *S. dasyclados*. A higher microbial biomass in the *S. viminalis* plot supported this suggestion.

Acknowledgements

The project was financed by the Estonian Science Foundation, grants no. 4831 and 4895. We wish to thank Prof. Hans Persson and particularly Dr. Ulf Granhall from the Swedish University of Agricultural Sciences for their corrections of the manuscript and for valuable suggestions. Marika Truu from the University of Tartu is thanked for the analyses of microbial biomasses. Jeremy Flower-Ellis helped with language revision.

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