Biogeochemistry **47:** 297–317, 1999.

Influence of dolomitic lime on DOC and DON leaching in a forest soil

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Received 10 December 1998; accepted 22 December 1998

Key words: dissolved organic carbon, dissolved organic nitrogen, Haplic podzol, hydrophobic and hydrophilic compounds, liming

Abstract. The influence of liming on leaching and distribution of dissolved organic carbon (DOC) and nitrogen (DON) in mineral soil was investigated in a leaching experiment with soil columns. Soil samples from separate horizons (O, A and B horizons) were collected from control and limed plots in a field liming experiment in a spruce forest in southern Sweden. The field liming (0.88 kg m^{-2}) had been carried out 8 years before sampling. To minimize the variation among replicates, soil profiles were reconstructed in the laboratory so that the dry weight was the same for each individual soil horizon regardless of treatment. Two soil column types were used with either the O+A horizons or the O+A+B horizons. One Norway spruce seedling (*Picea abies* (L.) Karst) was planted in each soil column. Average pH in the leachate water was greater in the limed treatment than in the control treatment (5.0 versus 4.0 for O+A columns and 4.3 versus 3.8 for O+A+B columns). After reaching an approximate steady state, the leaching of DOC was 3–4 times greater from the limed O+A and O+A+B columns than from the corresponding control columns but the leaching of DON increased (3.5 times) only in the limed O+A columns. There was a significant correlation between DOC and DON in the leachates from all columns except for the control O+A+B columns, which indicated a decoupling of DOC and DON retention in the B horizon in the control treatment. This might be explained by a selective adsorption of nitrogen poor hydrophobic compounds (C/N ratio: 32–77) while there was a lower retention of nitrogen rich hydrophilic compounds (C/N ratio: 14–20). Proportionally more hydrophobic compounds were leached from the limed soil compared to the unlimed soil. These hydrophobic compounds also became more enriched in nitrogen after liming so in the limed treatment nitrogen might be adsorbed at nearly the same proportion as carbon, which might explain the fact that there was no decoupling of leached DOC and DON from the B horizon after liming.

Introduction

In temperate and boreal forests most of the nitrogen is stored in soil organic matter, both in forest floor and mineral soil (David et al. 1995). In soil water, organic nitrogen is often the major form of nitrogen (Yavitt & Fahey 1984;

Göttlein et al. 1991). The solubility of organic matter varies depending on molecular size, aromaticity, acid strength and charge density. An increase in pH and/or decrease in ionic strength leads to an increase in the leaching of dissolved organic carbon (DOC) (Evans et al. 1988; Jardine et al. 1989; Vance & David 1989; Andersson et al. 1994), which may also increase microbial activity (Brunner & Blaser 1989; Andersson et al. 1994). Conversely, microbial activity may also affect the solubility of organic matter (Guggenberger & Zech 1993).

Liming, i.e., the application of calcite or dolomite, is one way to counteract the acidification of soils. Curtin and Smillie (1983) found that liming increased the amount of organic matter in the soil solution in some Inceptisols and Alfisols. In a soil incubation experiment with field limed mor humus (O horizon), the plots receiving the highest lime applications also had the highest concentration of DOC (Andersson et al. 1994). Göttlein et al. (1991) determined that DOC and dissolved organic nitrogen (DON) increased after liming with dolomite, particularly in the O horizon. However, in some other liming experiments no differences in DOC leaching from the O horizon were detected (Cronan et al. 1992; Smolander et al. 1995)

The composition of DOC and the DOC adsorption properties of B horizons have been investigated in several studies (Vance & David 1991; Guggenberger & Zech 1992; Kaiser et al. 1996). However, the influence of the B horizon on the adsorption of DON is less known and few studies have been conducted which show the composition of DON (Qualls & Haines 1991). The adsorption of DOC and DON in the B horizon is important for the long-term storage of C and N in the soil (David et al. 1995). Estimations of the total carbon and nitrogen storage in the whole soil profile (forest floor + mineral soil) in some Swedish podsols showed that 74% of total carbon was accounted for in the upper 80 cm of the mineral soil (Eriksson et al. 1996) and >80% of total nitrogen in the upper 60 cm of the mineral soil (Bringmark & Kvarnäs 1995). The increase in pH caused by liming may change the soil flux of DOC and DON as well as the chemical composition of the dissolved organic matter. These changes may affect the adsorption properties and thereby the storage and microbial availability of C and N.

The objective of the present experiment was to investigate the influence of the B horizon on the fluxes and transformations of DOC and DON in a limed and unlimed forest soil in a laboratory experiment. A further objective was to determine the extent to which charge characteristics of the dissolved organic matter could explain the leaching pattern of DOC and DON through a podzol and possible storage differences in the soil.

One problem in connection with the incubation of soil in laboratory experiments where growing plants are excluded is that unnatural conditions could be created in the soil. There might be an accumulation of mineralized inorganic nitrogen, which would affect the ionic strength and pH in the soil water and thereby also affect the leaching of DOC (Evans et al. 1988; Andersson et al. 1994). A more natural system would be created if the soil had growing plants in a plant-soil microcosm. Under such conditions, the uptake of NH_4^+ and NO_3^- by plants might reduce the pool of soil mineral N and the NH_4^+ uptake might also decrease nitrification, even though a column study could never recreate a natural system. Norway spruce seedlings (*Picea abies* (L.) Karst) were selected for planting in the microcosms because this is the main tree species at the field site where the soil was collected. The set-up of the experiment was designed in order to investigate the treatment effect only (liming) and not the specific influence of the plants on the nitrogen mineralisation.

Materials and methods

Soil material and sampling

Soil samples from ten soil pits per plot were collected in September 1992 from four separate control plots and four limed plots in a field-liming experiment located at Hasslöv in southern Sweden (56°24′ N, 13°00′ E, 190 m above M.S.L.). The annual mean temperature and precipitation of the site are 6.5 °C and 1100 mm, respectively. The Hasslöv site is a former *Calluna* heathland which was planted in 1949 with Norway spruce (*Picea abies* (L.) Karst) and which has almost no ground vegetation, apart from various mosses and the grass *Deschampsia flexuosa* (Trin.) where the canopy is more open. In 1984, the limed plots were treated with dolomite corresponding to 880 g m^{-2}. The field experiment was set up as a completely randomized block design with four blocks (one replicate per block), and this design was used in the laboratory experiment.

The soil profile, classified as Typic Haplorthod (USDA 1990) and Haplic Podzol (FAO 1988), was divided into O, A and B horizons. Due to a high organic matter content in the upper 0–5 cm mineral soil, a typical E horizon could not be clearly distinguished. The O horizon was further divided, from top to bottom, into a litter layer (Oi), a layer with intermediately decomposed organic material (Oe) and a layer with highly decomposed organic material (Oa). The Oe and Oa horizons were sieved through a 5-mm mesh sieve, and the mineral soil through a 3-mm mesh sieve. Coarse material (twigs, cones and coarse roots) was removed by hand from the Oi horizon. The soil was then stored at 4 °C under field-moist conditions for 14 days until used in the experiment.

Experimental design

Incubation columns were constructed from PVC cylinders with an inner diameter of 10 cm (eight 15-cm-high cylinders [O+A] and eight 21-cm-high cylinders [O+A+B], i.e., four replicates for each treatment). They were equipped with a bottom plate furnished with a 3-mm drainpipe connected to a vacuum-system. A plastic filter plate (approx. pore diam. 15 μm) was placed 2 cm above the bottom plate. The ten soil samples from each plot were pooled by horizon and plot. There was a high variation among the plots at the field site concerning the dry weight and the water content in the different horizons. To reduce this variation, we reconstructed the soil horizons as follows: The amount of dry matter per horizon used in the experiment was the same for all columns corresponding to the average density and thickness for each horizon in the field (Oi: 6.9 g dw, Oe: 20.8 g dw, Oa: 6.9 g dw, A: 304.0 g dw and B: 369.5 g dw). Before placing the soil into the incubation columns, the Oi, Oe and Oa horizons were moistened with distilled water to reach the same water content for all columns (300% of dry weight). The A and B horizons were moistened to 23–29% of dry weight and 19–29% of dry weight, respectively, corresponding to the field capacity in the A and B horizon materials. The total amount of carbon and nitrogen corresponded to 4083 g C m^{-2} and 185 g N m^{-2} in the control O+A columns, 3819 g C m^{-2} and 186 g N m^{-2} in the limed O+A columns, 5425 g C m^{-2} and 252 g N m^{-2} in the control O+A+B columns, and finally 5289 g C m^{-2} and 264 g N m^{-2} in the limed O+A+B columns.

Norway spruce (*Picea abies* (L.) Karst) seedlings were cultivated from seeds for 2 weeks in a mixture of sand:perlite:vermiculite, and then placed in a nutrient culture solution (Ingestad & Lund 1986) for 3 weeks before being transferred to the columns. Seedlings selected for the experiment all had nearly the same size and fresh weight. The average dry weight was 21 mg per seedling. One spruce seedling per column was planted in the Oe+Oa horizon, whereupon the litter was applied. Columns were then placed in a climate chamber with a constant air temperature (15 °C, average mean summer temperature in southern Sweden) and humidity (80%) and a 20 h per day light regime with a light intensity of approx. 350 mE.

At the beginning of the experiment, the water content in the columns was adjusted with distilled water every other day to keep it at 93% of the total dry weight in the O+A columns, and 70% in the O+A+B columns, which approximately corresponded to the water holding capacity of the whole soil column. Later on, these adjustments were made at longer time intervals. The water content was gradually lowered during the experiment so that by the end of the experiment the water content was 80% in the O+A columns and 62% in the O+A+B columns. Between water-adjusting occasions the columns were irrigated with a solution having a concentration of neutral salts (ion concentrations: 3.01 mg K^+ L^{-1}, 4.71 mg Na^+ L^{-1}, 1.56 mg Ca^{2+} L^{-1}, 0.71 mg Mg^{2+} L^{-1}, 1.72 mg NH_4^+ L^{-1}, 6.26 mg NO_3^- L^{-1} and 15.00 mg Cl^- L^{-1}) whose composition and ionic strength corresponded approximately to the throughfall precipitation at the field site. However, sulphate was excluded from the solution because sulphur transformations were studied in a concurrent experiment (Valeur et al., unpublished). The amount of irrigation water was adjusted to compensate for the average evapotranspiration from the columns. Immediately before leachate collection, the columns were irrigated with an additional amount of 50–250 mL of the same solution.

Sampling and chemical analyses

Soil leachates were collected under tension approximately every four weeks for a total of 9 percolation events. The samples were immediately passed through a 0.45-μm filter (Millipore HA 0.45 HAWP04700), which was rinsed beforehand with 50 mL distilled water to remove contaminating DOC originating from the filter. After filtering, subsamples were taken for measurements of DOC, pH, NO_3^--N, NH_4^+-N, and total dissolved N (TDN). DON was calculated by subtracting NO_3^--N and NH_4^+-N from TDN. pH in the leachates were measured when the leachates had reached room temperature (within one day). Because of measurement errors, DON from the third percolation event and the pH from the last percolation event were omitted.

In connection with the fifth percolation event in the middle of the experiment and the last percolation event, DOC and DON were fractionated according to Leenheer (1981) and Qualls and Haines (1991), respectively. Before fractionation, the solutions were acidified with conc. HCl to pH 2 and then passed through a column with Amberlite XAD-8 resin. Before use, the XAD-8 resin was cleaned according to Leenheer (1981). The hydrophilic DOC and DON fractions were eluted, while the hydrophobic fraction was adsorbed to the XAD-8 resin (Leenheer 1981). The hydrophilic fraction and the solution before fractionation were analysed for DOC, TDN, NO_3^--N and NH_4^+-N. The amounts of DOC and DON in the hydrophobic fraction were calculated by subtracting the amounts of DOC and DON in the hydrophilic fraction from the amounts of DOC and DON in the solution before fractionation. The hydrophobic fraction consists of a mixture of fulvic acids, humic acids, polyphenols, hydrocarbons, carbonyl compounds, and a minor content of polynuclear amines, whereas the hydrophilic fraction is a mixture of simple aliphatic acids, amphoteric proteinaceous compounds, carbohydrates, and probably, low-molecular-weight fulvic acids (Leenheer 1981; Qualls & Haines 1991; Vance & David 1991).

In order to detect losses of C and N during the experiment, measurements were made of total C, total N, KCl extractable (1 M) NO_3^--N and NH_4^+-N and the loss on ignition (LOI), in samples taken from the separate horizons at the beginning and end of the experiment. Seedling shoots were dried at 85 °C, and the roots were freeze-dried. Dry weights of needles, twigs and roots were measured, and subsamples were taken for analyses of total C and N.

Soil dry weight was determined at 105 °C and LOI was determined at 550 °C. In order to estimate the amount of CO_3-C originating from the lime treatment, total C in ignited residues was determined by a dry combustion procedure at 1000 °C (Carlo Erba). This value was then subtracted from the total C content in the dried soil sample. About 10% of the ignited carbon at 550 °C could originated from inorganic carbon (T. Persson, pers. comm.).

Total C and total N in the dried soil and in the seedlings were determined by a similar dry combustion procedure on a Leco analyser at 1000 °C. DOC was analysed on a Shimatdzu TOC-500 TOC-analyser (TC–IC method). pH in the leachates was measured with a combination glass electrode. NO_3^--N in the percolates was determined by ion chromatography (Dionex column), while NH_4^+-N was analysed by flow injection. The total dissolved N samples were digested with potassium persulphate (10 g $K_2S_2O_8$ in 1 L of 0.15 M NaOH), which converted all dissolved nitrogen to nitrate. The persulphate reagent was mixed with an equal volume of the sample, then boiled under pressure (14 kPa) for 25 minutes, after which 0.25 mL of 1.44 M H_2SO_4 was added before analyses. The nitrate was then analysed by flow injection. NO_3^--N and NH_4^+-N in KCl extracts were analysed directly by flow injection.

Statistics

All data were treated statistically by analysis of variance (ANOVA). When significant differences were detected, pair-wise tests were made using Tukey's studentized range test (HSD) on all main-effects means. Association between two variables was estimated by the Pearson product-moment correlation coefficient. Statistical significance was set at $p < 0.05$. In the statistical calculations the O+A columns and the O+A+B columns were treated as two separate experiments, except for some comparisons between column types to indicate adsorption processes in the B horizon (see the Results and Discussion sections).

Table 1. Dry weight (g), total C (g), total N (g) and C/N ratio of the spruce seedlings at the end of the experiment. No significant differences were found between treatments ($n = 4$). OA = columns with control O+A horizons. OAB = columns with control O+A+B horizons. OA-D = columns with limed O+A horizons. OAB-D = columns with limed O+A+B horizons.

Treatment	Dry weight	Total C	Total N	C/N
OA	3.98	1.92	0.04	51.5
OA-D	4.74	2.28	0.05	48.0
OAB	4.85	2.33	0.05	48.1
OAB-D	4.56	2.15	0.05	47.1

Results

Seedlings

There was a high variability in seedling growth among the columns, and seedlings were not significantly affected by the lime treatment either with respect to dry weight, amount of nitrogen or C/N ratio (Table 1). However, the plant roots in the limed soil had a white 'fluffy' type of mycorrhiza, whereas those in the control soil had a brown, smooth mycorrhiza.

Leaching of DOC

Leachate pH was significantly greater in the limed treatment than in the control treatment in both the O+A and O+A+B experiments (Figure 1), except for the first three percolation events in the O+A+B columns. The concentration of DOC was significantly greater in the limed treatment except for the O+A columns during the first 75 days (Figure 2), owing to an initially high variation among individual O+A columns. There was also a high leaching of both DOC and DON in the O+A columns during the first three percolation events (Figures 2 and 3), which may have been due to disturbance effects at the beginning of experiment. Because of these initial effects, the main data analyses were carried out for the latter part of the experiment when the leaching of DOC and DON approached a steady-state condition (percolation events 4–9).

The total flux of leached DOC between percolation events 4–9 was significantly greater in the limed treatment of the O+A experiment compared to the control treatment and there was the same trend in the O+A+B experiment although it was not significant (Figure 4). The adsorption of DOC in the

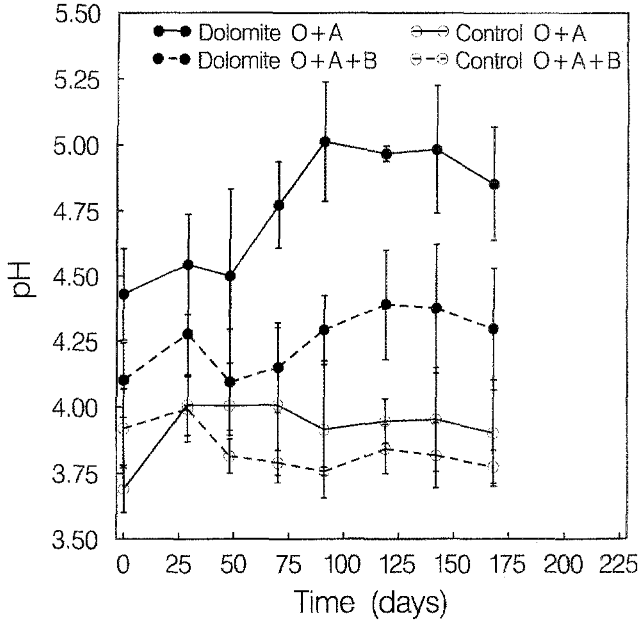

Figure 1. Temporal variation in leachate pH during the course of the O+A and O+A+B experiments. Because of measurement error, values for percolation event 9 are excluded from the graph. Error bars represent ± SD ($n = 4$).

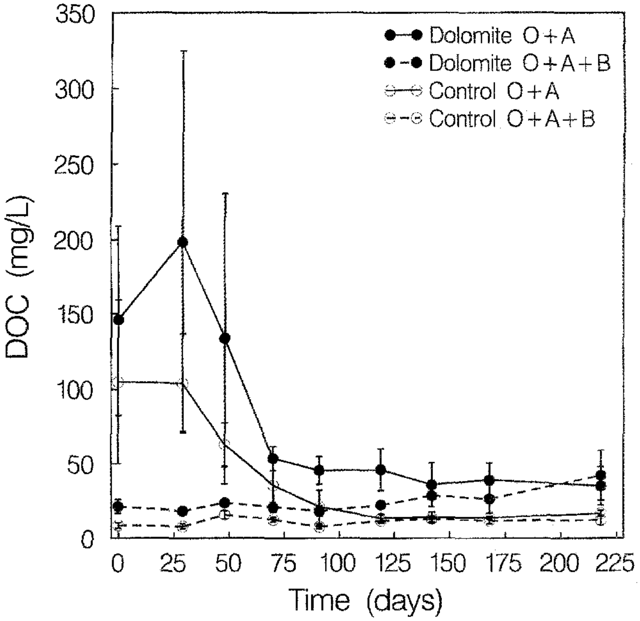

Figure 2. Temporal variation in leachate DOC concentration during the course of the O+A and O+A+B experiments. Error bars represent ± SD ($n = 4$).

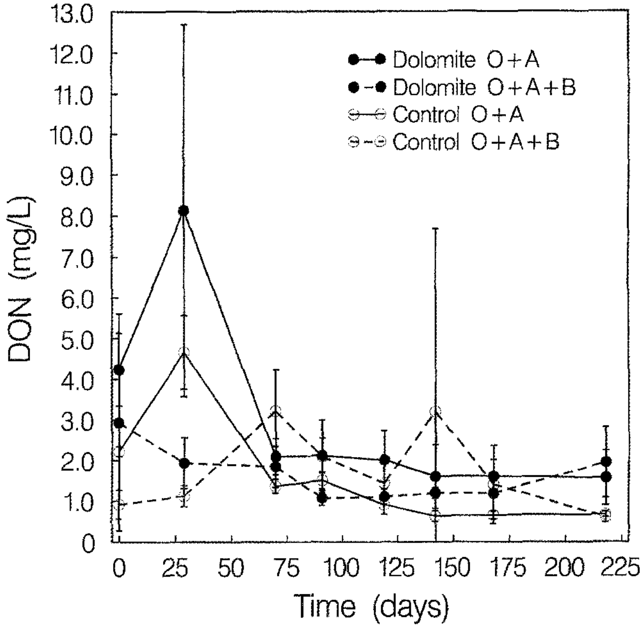

Figure 3. Temporal variation in leachate DON concentration during the course of the O+A and O+A+B experiments. Because of measurement error, values for percolation event 3 are excluded from the graph. Error bars represent ± SD ($n = 4$).

B horizon compared to the input of DOC from the A horizon tended to be proportionally higher in the limed treatment than in the control treatment, since the ratio between total leached DOC from the O+A+B columns and total leached DOC from the O+A columns was 0.34 in the limed treatment and 0.45 in the control treatment (Figure 4).

Leaching of DON

The concentration of DON had the same general pattern as DOC in the O+A columns, but variability in concentration was greater (Figure 3). In the middle of the experiment DON concentrations tended to be greater in the control O+A+B columns than in the limed O+A+B columns (Figure 3). However, DON in the control O+A+B columns was very low compared with the TDN values, so these values became more erratic. The total DON flux between percolation events 4–9 was significantly greater in the limed treatment of the O+A experiment, but there was no corresponding difference in the O+A+B experiment (Figure 5).

Carbon and nitrogen contents in the dissolved organic matter in the percolates (percolation events 4–9) were strongly correlated in the O+A experiment with a correlation coefficient (r) of +0.65 ($p < 0.001$) for the

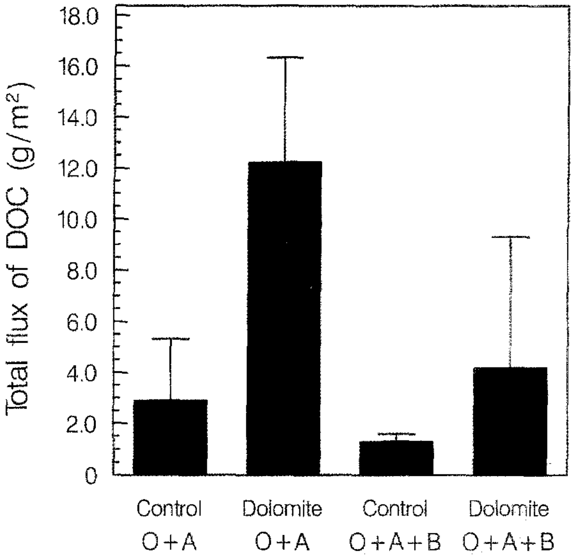

Figure 4. Total flux of DOC between percolation events 4–9 from the O+A and O+A+B experiments. Error bars indicate 95% confidence limits ($n = 4$).

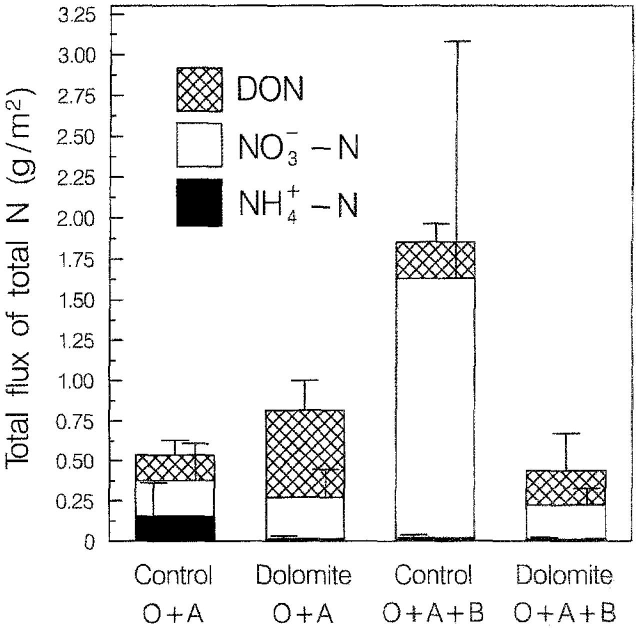

Figure 5. Total flux of DON, NH_4^+-N and NO_3^--N between percolation events 4–9 from the O+A and O+A+B experiments. Error bars indicate 95% confidence limits ($n = 4$).

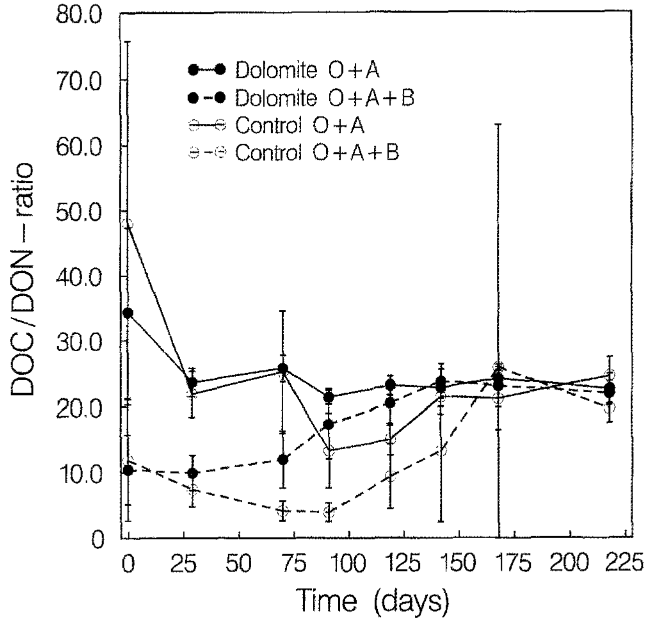

Figure 6. Temporal variation in the DOC/DON (w/w) ratio during the course of the O+A and O+A+B experiments. Because of measurement error, values for percolation event 3 are excluded from the graph. Error bars represent ± SD ($n = 4$).

control and $r = +0.95$ ($p < 0.001$) for the limed treatment, but for the O+A+B experiment only the limed treatment showed a significant correlation ($r = +0.67$, $p < 0.001$), and not the control ($r = +0.14$).

The ratio between DOC and DON in the percolates was initially high (34–48) in the O+A columns and low (10–12) in the O+A+B columns (Figure 6), but later (percolation events 4–9) the DOC/DON ratio stabilized at a level of 22–23 for the limed treatment in the O+A and O+A+B columns. The DOC/DON ratio in the percolates of the control O+A was significantly lower than in the limed O+A columns at percolation event 6, but by the end of the experiment the ratio was the same for both treatments. Very low DOC/DON ratios were detected in the percolates of the control O+A+B columns during the whole experiment except for the two last percolation events (Figure 6), but as for the DON values the uncertainty of the DOC/DON ratios was higher compared with the limed O+A+B columns and the O+A columns.

Leaching and change in storage of inorganic nitrogen

During the first 100 days of the experiment, the concentration of NH_4^+-N in the leachates was higher in the control O+A columns than in the limed columns (Figure 7(a)), and the total leached amount of NH_4^+-N for the period between percolation events 4 and 9 was also significantly higher (Figure 5).

The nitrate concentration initially increased in the O+A+B columns, especially in the control treatment, but it decreased in both treatments towards the end of the experiment (Figure 7(b)). The total flux of NO_3^--N from the control O+A+B columns between percolation events 4 and 9 was significantly higher than in the limed treatment (Figure 5). The high concentration of nitrate in the O+A+B columns coincided with a decrease in pH after 50 days, but pH increased again in the limed O+A+B columns when the nitrate concentration decreased (Figures 1 and 7(b)). The higher concentration of ammonium in the control O+A columns probably contributed to an increase in pH in these columns (Figures 1 and 7(a)). The increased concentration of NO_3^- in the control O+A+B columns and NH_4^+ in the control O+A columns may have influenced the DOC leaching since there was a negative correlation between the leaching of DOC and NO_3^- in control O+A+B columns ($r = -0.41$, $p < 0.05$) and a positive correlation between the leaching of DOC and NH_4^+ in control O+A columns ($r = +0.71$, $p < 0.001$) for the period between percolation events 4 and 9.

By the end of the experiment the KCl extractable NH_4^+ and NO_3^- had decreased in the Oe+Oa, A, and B horizons, except for the Oe+Oa horizon in the control O+A+B columns, where the amounts of both ammonium and nitrate increased. In the A and B horizons in the same columns the amounts of nitrate increased (Table 2). The root biomass of the seedlings in the control O+A+B columns was negatively correlated with the net change of accumulated KCl extractable NO_3^--N in the whole profile by the end of the experiment ($r = -0.986$, $p < 0.05$). The same trend was also detected with NH_4^+-N but it was not significant ($r = -0.931$).

DOC and DON fractionation

The higher pH in the liming treatment was connected with a relative increase in hydrophobic substances and a corresponding decrease in hydrophilic substances in the percolates of both the O+A and O+A+B experiment (Figure 8(a, b)). These differences were statistically significant at percolation event 5 (Figure 8(a)), but not at percolation event 9 (Figure 8(b)). Liming also significantly decreased the ratio between DOC and DON in the hydrophobic fraction at percolation event 9 while there was the same DOC/DON-ratio in the nitrogen-rich hydrophilic fraction independently of treatment and horizon (Figure 8(d)). In the limed treatment at percolation event 5 and the control treatment at percolation event 9, the hydrophobic/hydrophilic ratio for DOC and DON decreased significantly in the percolates in the O+A+B experiment compared to those in the O+A experiment (Figure 9(a, b)). There was a

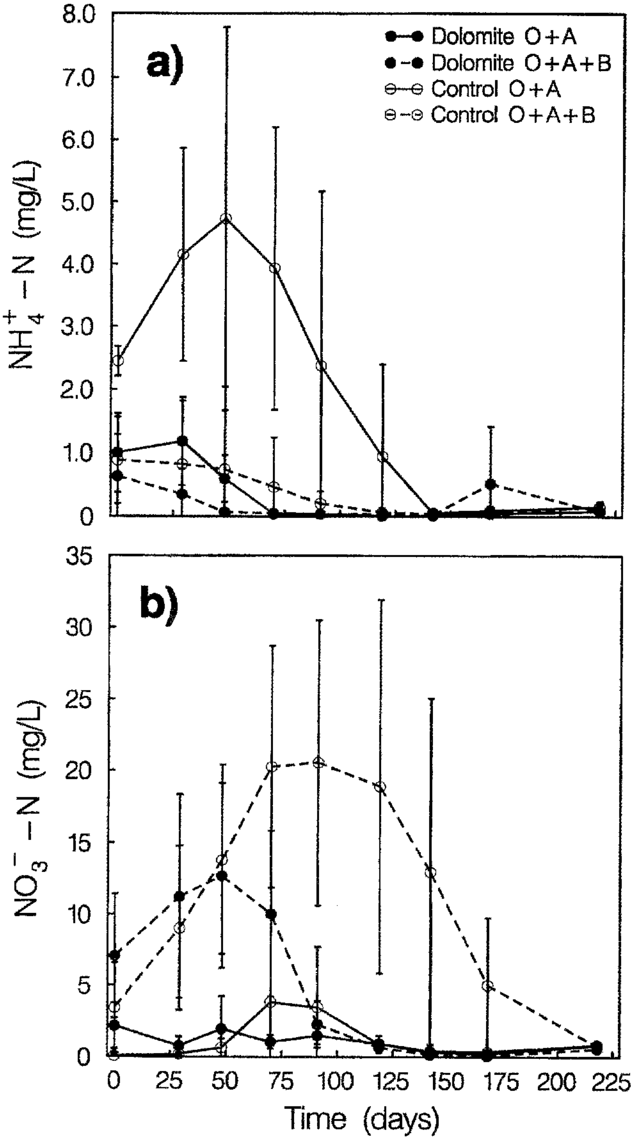

Figure 7. Temporal variation in (a) leachate NH_4^+-N concentration and (b) leachate NO_3^--N concentration during the course of the O+A and O+A+B experiments. Error bars represent ± SD ($n = 4$).

Table 2. KCl (1 M) extractable inorganic N in the separate horizons at the start and end of the experiment. Diff = end value – start value. Values with different superscript letters within each column are significantly different with respect to treatment for each pair of OA and OA-D or OAB and OAB-D ($p < 0.05$) ($n = 4$). All values in g m^{-2}. See Table 1 for treatment abbreviations.

Horizon	Treatment	NH_4^+-N start	NO_3^--N start	NH_4^+-N end	NO_3^--N end	NH_4^+-N diff	NO_3^--N diff
Oe+Oa	OA	0.149^a	0.020^a	0.111^a	0.009^a	-0.038^a	-0.011^a
	OA-D	0.210^a	0.205^b	0.035^a	0.003^a	-0.175^a	-0.202^b
	OAB	0.149^a	0.020^a	0.237^a	0.054^a	$+0.088^a$	$+0.035^a$
	OAB-D	0.210^a	0.205^b	0.053^a	0.104^a	-0.157^a	-0.101^a
A	OA	0.079^a	0.026^a	0.042^a	0.003^a	-0.037^a	-0.023^a
	OA-D	0.087^a	0.042^a	0.054^a	0.006^a	-0.033^a	-0.037^a
	OAB	0.079^a	0.026^a	0.041^a	0.042^a	-0.037^a	$+0.015^a$
	OAB-D	0.087^a	0.042^a	0.041^a	0.001^a	-0.046^a	-0.042^a
B	OAB	0.097^a	0.031^a	0.078^a	0.037^a	-0.019^a	$+0.006^a$
	OAB-D	0.090^a	0.071^a	0.026^a	0.008^a	-0.064^a	-0.063^a

similar trend in the lime treatment at percolation event 9 although it was not statistically significant. These changes in hydrophobic/hydrophilic ratio indicate that the hydrophobic fraction may be selectively adsorbed in the B horizon compared to the hydrophilic fraction.

Discussion

Liming increased the concentration of DOC and DON. The initially high leaching of DOC and DON was somewhat unexpected (Figures 2 and 3). It might be explained by the excessively high water content of the O horizon at the beginning of the experiment (corresponding to 300% by dry weight) which could have increased the production of DOC. This relationship between a high water content and an increased production of DOC has been shown by Christ and David (1996), and the effect could be explained by a higher biological activity in the soil. A functional relationship between biological activity and DOC release has been suggested by Zech et al. (1994) and McDowell and Likens (1988) among others. The initially high leaching of DOC and DON (percolation events 1–3) may have resulted from a build-up of soluble C and N which were soon leached out from the O+A columns. The high water content seemed to conceal the treatment effects, but later on

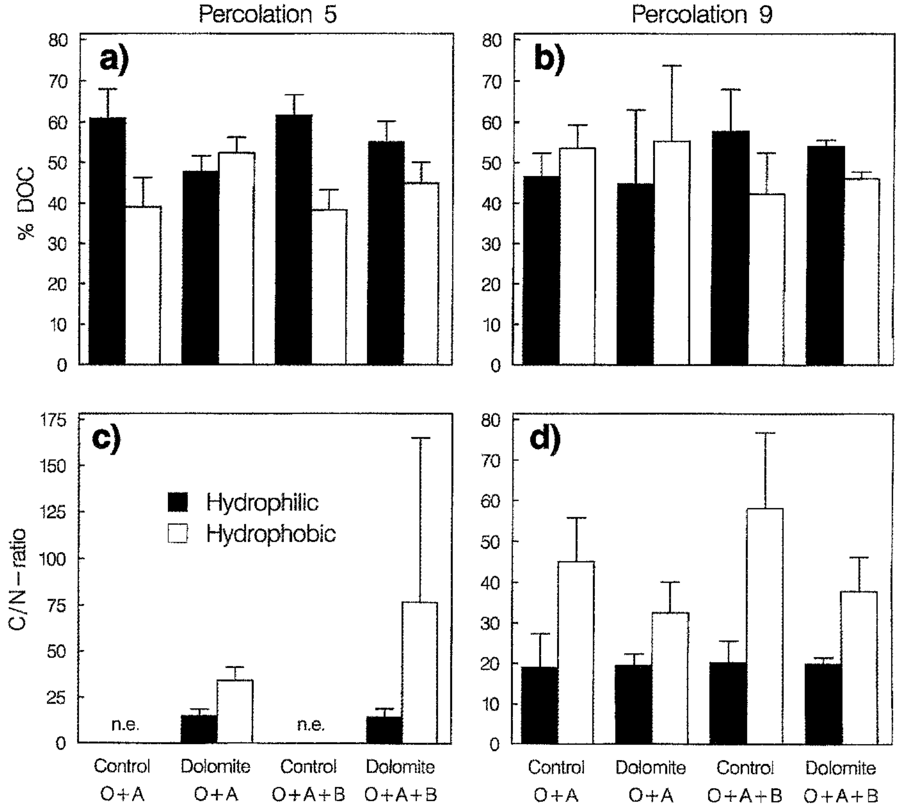

Figure 8. Percentage of DOC in the hydrophobic and hydrophilic fractions at (a) percolation event 5 and (b) percolation event 9 and corresponding C/N ratio at (c) percolation event 5 and (d) percolation event 9. n.e. = not estimated because of obvious analytical errors. Error bars indicate 95% confidence limits (*n* = 4). Note: different scale on the *y*-axis in (c).

(percolation events 4–9) the liming effect on DOC and DON leaching was clearly visible (Figures 2 and 3). Furthermore, the concentrations became very similar to those found in lysimeter water collected from the same horizons at the Hasslöv field site (Nilsson et al., unpublished). In the O+A+B experiment, there was no such increase in DOC in the percolates at the beginning of the experiment, probably owing to an effective retention in the B horizon (Figure 2). The retention mechanisms in the B horizon have been investigated in many laboratory studies (Jardine et al. 1989; David & Zech 1990; Guggenberger & Zech 1992; Kaiser et al. 1996). These studies have all shown that the decrease in DOC concentration in percolates after passing the B horizon depended primarily on abiotic adsorptive processes and only to a minor extent on carbon mineralization.

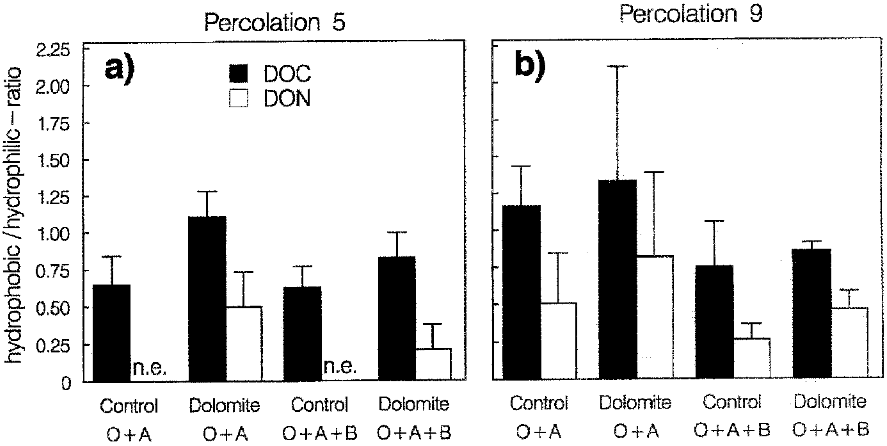

Figure 9. Ratios between the hydrophobic and hydrophilic fractions at (a) percolation event 5 and (b) percolation event 9. n.e. = not estimated because of obvious analytical errors. Error bars indicate 95% confidence limits ($n = 4$).

There is always a risk of artefacts in this type of laboratory experiment. The use of sieved soil instead of intact soil cores might have changed the mobilization and leaching of DOC. However, in a leaching experiment where intact forest floor cores and sieved forest floor samples from the same field site were used, the pattern of DOC leaching was similar in both intact and sieved soil (Gödde et al. 1996). The disturbance of the soil may also cause increases in N mineralization and nitrification. The results obtained could therefore differ from those observed in the field (Johnson et al. 1995). The soil from the Hasslöv site used in this experiment seemed to have a high nitrification potential in the field after liming (Nilsson et al. unpublished). Other laboratory incubation experiments with the Hasslöv soil have shown that even the control humus and mineral soil had a nitrification potential (T. Persson pers. comm.). The root uptake of mineral N by the spruce seedlings in the present experiment should have had a moderating effect on NO_3^- leaching. However the degree of moderation was insufficient in the control O+A+B experiment, which had a net increase of both NH_4^+ and NO_3^- in the Oe+Oa horizon by the end of the experiment (Table 2) and the highest NO_3^- leaching of all treatments (Figure 5). The negative correlation between the total root biomass in the control O+A+B columns and the NO_3^- content in the whole profile indicated a casual connection between poor root development and increase in NO_3^- storage which should have affected the NO_3^- leaching. It is not clear, however, why this root development-nitrate pattern did not appear in the other columns.

The increased concentration of NO_3^- in the control O+A+B columns seemed to have an impact on DOC leaching. High nitrification decreased the pH (Figure 1 and Figure 7(b)), which might decrease the solubility of DOC (Andersson et al. 1994). There may also be an impact by NH_4^+ on the solubility of DOC. The positive correlation between NH_4^+ and DOC in the control O+A columns may partly be explained by increased pH, but monovalent cations such as NH_4^+ could also form "soluble salts" with humic and fulvic acids (Norman et al. 1987). A similar connection between high amounts of NH_4^+ and DOC was observed in other experiments (Andersson et al., submitted).

The pattern of the DOC/DON ratio in the O+A+B experiment differed from the O+A columns (Figure 6). Nitrogen-rich compounds in the B horizon were initially leached out (percolation events 1–7), but subsequently the soil water in the B horizon was probably more affected by the leachates from the A horizon, which had a higher DOC/DON ratio. Due to this successive mixing of water from the two horizons, there was a high temporal variability in the DOC/DON ratio. In the later part of the experiment (percolation events 7–9) the O+A columns and the limed O+A+B columns had nearly the same DOC/DON ratio (21–25). This was somewhat higher than the measured DOC/DON ratios (15–20) in the lysimeter water from the A and B horizons in the control and limed plots at the Hasslöv site during the same period of the sampling (Nilsson et al. unpublished).

The fractionation data indicated that the hydrophobic fraction had a low N content compared to the hydrophilic fraction which is in accordance with results presented by Qualls and Haines (1991). A decrease was found in the DOC/DON-ratio in the hydrophobic fraction after liming, which indicated that more nitrogen may have been incorporated and immobilized in this fraction with increasing pH. A significant portion of N-containing organic substances and ammonia can be complexed with humic-like substances and polyphenols that resist degradation (Nömmik & Vahtras 1982; Fog 1988). An increase in pH may enhance the nonbiological incorporation of ammonia (Nömmik & Vahtras 1982) and might be one explanation for the lower DOC/DON-ratio in the hydrophobic fraction of the limed treatment.

A certain decrease in the hydrophobic fraction of the dissolved organic matter was observed after the soil solution had passed through the B horizon (Figure 8(a, b)). Similar observations were made by Cronan and Aiken (1985), David et al. (1995), Easthouse et al. (1992), Guggenberger and Zech (1993) and Vance and David (1989). The hydrophobic fraction has a high affinity to hydrous oxides in the B horizon, and in soils with a limited number of binding sites, the strong competition between hydrophobic and hydrophilic DOM causes the hydrophobic DOM to suppress the binding of hydrophilic substances (Kaiser et al. 1996). There was only a slight difference in hydrophobic/hydrophilic ratio between control O+A (0.64) and control O+A+B (0.62) for the DOC at percolation event 5, but the difference became pronounced and statistically significant at percolation event 9 (1.16 in control O+A columns versus 0.75 in control O+A+B columns) (Figure 9(a, b)). We estimated the total hydrophobic/hydrophilic ratio from two other comparable investigations. The estimated values were 0.69 for the E horizon and 0.62 for the B horizon in lysimeter water from a Norwegian podzol (Easthouse et al. 1992), while David et al. (1992) found a ratio of 1.33 in the upper B horizon and 0.79 in the lower B horizon in lysimeter water collected from a Spodosol at the Bear Brook Watershed site in Maine, U.S.A.

Liming increased the hydrophobic fraction of the dissolved organic matter at percolation event 5 (Figure 8(a)), but the treatment difference was levelled out at percolation event 9. Göttlein et al. (1991) found that DOC in water extracts from limed field plots had a higher content of 'less polar' compounds in the O horizon and in the upper mineral soil compared with DOC in water extracts from unlimed plots. These 'less polar' compounds had a higher molecular weight than the 'polar' compounds. Compounds with a high molecular weight could be hydrophobic acids and neutrals (Qualls & Haines 1991).

Calculations based on the total amount of leached DOC during the whole experiment (percolation events 1–9), showed that about 70–75% of the DOC leached from the A-horizon was adsorbed in the B horizon in both treatments (Figure 3). The increased concentration of DOC caused by liming enabled more DOC to be adsorbed in the B horizon (cf. Vance & David 1989). On the other hand, the calculated amount of DOC adsorption was less than 1% of the total amount of carbon in the B-horizon (data not shown), so it would be very difficult to detect as a difference between the initial and final C storage. The duration of the experiment was probably too short for detecting significant changes in the storage of carbon in the B horizon. A study with soil samples from 40-year-old limed and unlimed plots in one beech stand and three spruce stands in southern Sweden showed, however, that the concentration of total carbon in the limed plots was often increased in the upper mineral soil in comparison with unlimed plots (Persson et al. 1995).

The transport and leaching of DON was more complicated than the leaching of DOC owing to the effects of counteracting mechanisms. The non-existing correlation between DOC and DON in the control O+A+B columns indicated a decoupling between DOC and DON after passing the B horizon. The main part of the DON was in the hydrophilic fraction which tended to be adsorbed less efficiently in the B horizon compared to the hydrophobic fraction. After passing the B horizon the percolate was enriched with DON, which was indicated by the low DOC/DON ratios of the control O+A+B columns during the main part of the experiment (percolation events 1–7) (Figure 6). The lower adsorption of DON compared to DOC in the B horizon may partly explain the almost equal amounts of totally leached DON in control O+A and O+A+B columns during percolation events 4–9 (Figure 5). However, the hydrophobic compounds became proportionally more nitrogen-rich after liming, so although more DON could be leached in the hydrophobic fraction, more organic nitrogen was also likely to be adsorbed in the B horizon of the limed treatment, which may explain the comparatively small amount of totally leached DON from the limed O+A+B columns compared with the limed O+A columns (Figure 5).

Conclusions

Under conditions approaching steady state, liming increased the leaching of DOC from soil columns consisting of either the O+A horizons or the O+A+B horizons of a Spodosol. The leaching from the two column types indicated that adsorption of DOC in the B horizon was nearly proportional to the input from the A horizon. Consequently, the carbon storage in the B horizon of a Spodosol would eventually increase after liming.

In the control columns DOC and DON were leached from the O+A columns at a nearly constant ratio, i.e., there was a significant positive correlation between DOC and DON. In the control O+A+B columns (after having passed the B horizon) there was a relative increase of the DON leaching (no measurable adsorption, because the main part of DON was in the hydrophilic fraction which tended to be less adsorbed than the nitrogen poor hydrophobic fraction) and DOC and DON seemed to be uncoupled, i.e., they were no longer correlated.

Liming increased the DON leaching from the O+A columns, and the DOC and DON were positively correlated (coupled) as in the control O+A columns. In the limed O+A+B columns, DOC and DON were still positively correlated, probably due to a certain increase in the N content of the hydrophobic fraction which tended to be preferentially adsorbed in the B horizon, combined with an increase in the hydrophobic/hydrophilic ratio of the dissolved organic matter. For this reason the nitrogen storage in the B horizon of a Spodosol would possibly increase after liming.

Acknowledgements

This research was financed by grants from the National Swedish Environmental Protection Agency and the Swedish Council for Forestry and Agricultural Research. We are grateful to Dr T. Ericsson for providing the spruce seedlings, A. Hovberg for assistance with culturing the seedlings in the nutrient solution and Dr H. Wallander for characterisation of the mycorrhiza types. We thank Prof. T. Persson and Dr J. Mulder for reviewing the manuscript. We also thank Dr D. Tilles and Dr M. McAfee for correcting the language.

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