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Effects of forest fertilization on nitrogen leaching and soil microbial properties in the Northern Calcareous Alps of Austria

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Abstract

Several boreal and alpine forests are depleted in nutrients due to acidification. Fertilization may be a remedy, but rapidly-soluble salts (N, P, K, Mg) may pose nitrate problems for the groundwater or decrease microbial activity.

With the aim to investigate potential nitrogen leaching after fertilization we set up an experiment employing intact soil cores (11 cm diameter, 20–40 cm long) from a mixed forest and a *Picea abies* stand (soil type Rendsina) in the Northern Calcareous Alps of Austria. The cores were fertilized with a commercial NPK fertilizer or a methylene-urea-apatite-biotite (MuAB) fertilizer at a rate corresponding to 300 kg N ha^{-1} and incubated for 28 weeks together with unfertilized controls. Both soil water (retrieved 5 cm below the soil surface) and leachate were analyzed for nitrate and ammonium in regular intervals. After the incubation, soil microbial biomass and basal repiration were determined and a nitrogen mineralization assay was performed.

For the control, in the soil water and leachate maximum NH_4^+ and NO_3^- concentrations of 5 and 11 mg N L^{-1}, respectively, were found. Compared to that, MuAB fertilizer resulted in a slow increase of NH_4^+ and NO_3^- in the soil water (up to 11 and 35 mg N L^{-1} respectively) and in the leachate (4 mg NH_4^+-N L^{-1} and 44 mg NO_3^- -N L^{-1}). Highest nitrogen loads were found for the fast release NPK fertilizer, with NH_4^+ and NO_3^- concentrations up to 170 and 270 mg N L^{-1}, respectively, in the soil water. NH_4^+ -N levels in the leachate remained below 5, while NO_3-N levels were up to 190 mg L^{-1}. Fast- release NPK caused a significant decrease of microbial biomass and basal respiration. These parameters were not affected by MuAB fertilizer.

The results suggest that the MuAB fertilizer may be an ecologically appropriate alternative to fast-release mineral fertilizers for improving forest soils.

Introduction

Anthropogenic influences may lead both to an increase or a decrease of soil nutrient contents. On the one hand, atmospheric depositions are considered a possible danger for forest ecosystems through acidification. This may lead to cation losses and N saturation (Bredemeier and Ulrich, 1992). On the other hand,in some alpine forests the site history (litter harvesting) or present-day practices like cattle grazing and full-tree harvest are responsible for nitrogen exports (Glatzel, 1991). Together with increased leaching and erosion this may lead to seasonal N deficits (Liu, pers. commun.).

Fertilization of such stands is considered a possible remedy but some rapid soluble N forms are likely to cause nitrate problems for the groundwater (Katzensteiner et al., 1995). Since many alpine forests are water supply areas this deserves special attention. Fertilization with slow-release N fertilizers has been shown to increase net mineralization and N availability without losses through nitrification and leaching in a number of investigations both in boreal and alpine forests (Katzensteiner et al., 1995; Martikainen et al., 1989). Aarnio et al. (1993) also found long-lasting positive effects on tree growth of slow-release nitrogen fertilizer.

Besides N, other nutrients often are deficient in alpine forests. However, fertilization with rapidly soluble salts of P, K and Mg have been shown to decrease microbial activities (Söderström et al., 1983; Van

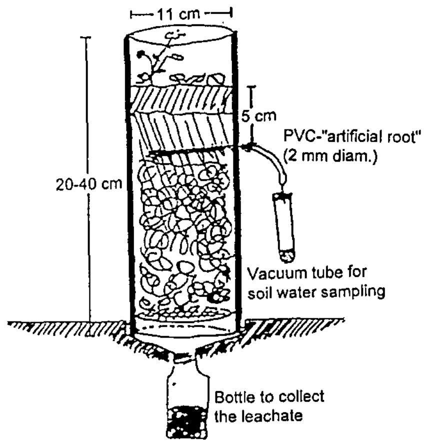

Fig. 1. Schematic picture of the lysimeter setup.

Cleve and Moore, 1978) which in turn may hamper the nutrient supply. Rock powders of apatite and biotite have eliminated the decreasing effect of urea on the microbial activity (measured as CO_2 production) on a poor pine forest site (Martikainen et al., 1989). Thus, apatite and biotite may help to improve the P, K and Mg status of soils, without adverse short term effects.

The aim of this study was to investigate whether a combination of slow-release N fertilizer (methylene-urea) and apatite/biotite would be safe in terms of nitrate leaching without harming microbial properties.

Materials and methods

Sites

Intact soil cores (diameter 11 cm; 20–40 cm long) were sampled at random down to the C horizon from a mixed forest stand and a spruce stand in the province of Tirol, Austria, at 900 m and 1500 m above sea level, respectively. Needle analyses showed deficiencies in nitrogen, phosphorus and magnesium. Some site characteristics are shown in Table 1.

Lysimeter setup

The soil cores were sampled with a steel corer and placed in plexiglass tubes directly in the field (Fig. 1). Ground vegetation, comprised of various grasses, herbs and mosses, was not removed. Four replicate cores from each site were randomly selected and amended with a MuAB (Methylene-urea+Apatite+Biotite) fertilizer or a fast-release NPK fertilizer at application rates equivalent to 300 kg N ha^{-1} (see Table 2). Control cores remained untreated. The lysimeters were incubated in a growth chamber at 15° C. The columns were watered weekly with 100-200 mL deionized water at a rate of 5mL min^{-1} (ca. 0.5 mm min^{-1}) so as to avoid edge effects. The amount of water added during the study period corresponded to the annual precipitation at the study sites. With an Eijkelkamp "artificial root" (2mm diam., PVC) 5 cm below the soil surface the soil water was sampled in two to three week intervals by using 10 mL Vacuum tubes (Greiner, Kremsmünster, Austria). At the bottom of the columns, the leachate was collected in the same frequency.

Analyses

Estimations of plant cover were made at the beginning and at the end of the incubations (Braun-Blanquet, 1964). Ammonium and NO_3^- in the soil water and leachate were analyzed with the diffusion-conductivity (DC) method (Carlson et al., 1990) employing an Ammonia Analyzer (Wescan Model 360, Alltech, San Jose, Cal.). At the end of the incubations, microbial biomass C (C_{mic}) was determined with the substrate-induced respiration technique (Anderson and Domsch, 1978) with a continuous flow setup (Heinemeyer et al., 1989). Basal respiration was determined as CO_2 evolution after 15 h incubation, and the metabolic quotient was calculated according to Anderson and Domsch (1985) and given as mg CO_2-C g^{-1} C_{mic} h^{-1}. For determining N mineralization moist soil samples (30 g fresh weight, 2mm fraction) were incubated in 100 mL Erlenmeyer flasks for three weeks (25°C) according to Beck (1983). NO_3^- and NH_4^+ were extracted with 1 *M* KCl and measured with the DC method.

Statistical analyses

The results were analysed by analysis of variance. Comparison of the means after the analysis was made by Tukey test.

Results and discussion

While the mixed forest stand was dominated by grasses as ground cover, the spruce stand was dominated by herbs and mosses (Table 3). Due to the randomized sampling, there were considerable differences in the initial total plant cover, where the MuAB variant of

Table 1. Site characteristics

Site	Main tree species (stand age, a)	Soil type*	Horizon*** (thickness)	pH (KCl)	P_2O_5 (mg kg^{-1})	K	Mg	N (%)	Organic carbon	Nutrients in needles(%)**
Mixed forest	Spruce (*P. abies*) and beech (*F sylvaticus*) (150 a)	Mull-type rendsina	O(2–10cm) A(15–30cm)	5.9 7.1	120 15	380 30	175 184	1.8 0.7	38% 12%	N : 0.8 P : 0.06–0.08 K : 0.3
Spruce forest	Spruce (*Picea abies* (200 a)	Terra fusca-rendsina	O(8–35cm) A(10-30cm)	5.5 7.1	n.d.	n.d.	n.d.	2.1 0.6	39% 5%	N : 0.9-1.1 P : 0.07–0.11 K : 0.3–0.5

*Soil types according to Kubiena (1953).
**Stöhr, pers. communication.
***Figures for the thickness indicates the variability of the soil structure.
The figures for the O horizon comprise O_l, O_f, and O_h layers. However, for pH and nutrient analyses the O_l layer was omitted.

Table 2. Fertilizers used

Treatment	Form of nitrogen	Application rate g per column	Nutrient contents (mg g^{-1}) N	P_2O_5	K_2O	Mg
Fast-release NPK fertilizer	ammonium-nitrate	1.90	150	50	179	25
Slow-release MuAB fertilizer	methylene urea	1.83	156	53	47	68

the mixed forest site was characterized by a particularly low value (9%). In course of the incubations the average plant cover increased on all fertilized cores.

Nitrate and NH_4^+ concentrations of the control soils leachate were below 15 and 10 mg N L^{-1}, respectively (Fig. 2). For the mixed forest stand, nitrate levels in soil water and leachate were about the same until week 15. Then, NO_3^- values in the soil water decreased. Ammonium levels were negligible. For the spruce site, towards the end of the incubation NH_4^+ levels in the leachate were higher than those in the soil water, and they were considerably higher than the NO_3^- levels. This indicates (i) that mineralization of dead (tree) roots contributes to the soil N_{min} levels which increase with depth and (ii) an inhibition of nitrification at the spruce site which is considerably more acid than the mixed forest site. No significant effect of the length of the columns on leachate NO_3^- and NH_4^+ were found, neither for the control cores nor for the fertilized ones.

On the mixed forest soils, MuAB fertilizer led to an immediate increase of NO_3^- concentrations in the soil water, followed by a subsequent increase of NO_3^- in the leachate (Fig. 3a). Maximum values of 35 and 42 mg NO_3^--N L^{-1} were found for the soil water and leachate, respectively. It has to be noted that the initial plant cover for the mixed forest MuAB treatment was very low (Table 3), which possibly resulted in a comparatively low N retention through plant uptake. Ammonium in the soil water never exceeded 4 mg L^{-1}, NH_4^+ contents in the leachate were negligible. For the spruce site (Fig. 3b), however, a pronounced initial increase of ammonia in the soil water was found (26 mg L^{-1} maximum), while the NO_3^- increase followed later and did not exceed 18 mg L^{-1}. Subsequently, NO_3^- and NH_4^+ in soil water declined. Ammonium in the leachate remained at control levels throughout the incubation time. Nitrate in the leachate, however, slow but steadily tended to increase until the end of the incubation and reached a maximum of 19 mg L^{-1}, about four times the maximum of the control soils. This indicates low nitrification rate at the spruce site. The low inorganic N contents found at the end of the experiment (Fig. 5) showed that most of the nitrogen added was either still present as slow-release methylene-urea

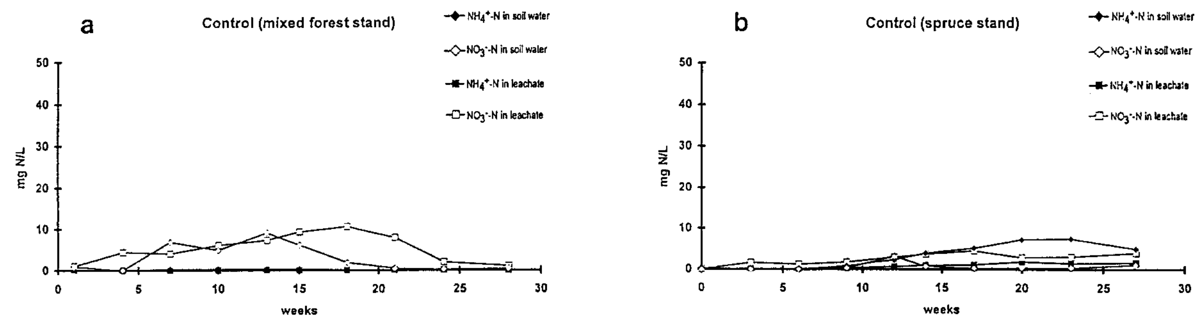

Fig. 2. Nitrate and ammonia in soil water and leachate of unfertilized control cores from (**a**) the mixed forest and (**b**) the spruce forest sites. Coefficient of variation: 87%.

Table 3. Percent plant cover at the beginning and at the end of the incubations

Site		Start of the incubation (1st week)				End of the incubation (28th week)			
		Grasses	Herbs	Mosses	Total	Grasses	Herbs	Mosses	Total
Mixed	Control	28	4	0	**32**	19	23	3	**45**
forest	MuAB	3	3	3	**9**	37	18	0	**55**
	NPK	24	18	3	**45**	23	31	2	**56**
Spruce	Control	3	20	40	**63**	0	13	48	**61**
forest	MuAB	3	21	33	**57**	30	16	15	**61**
	NPK	5	11	6	**22**	16	31	42	**89**

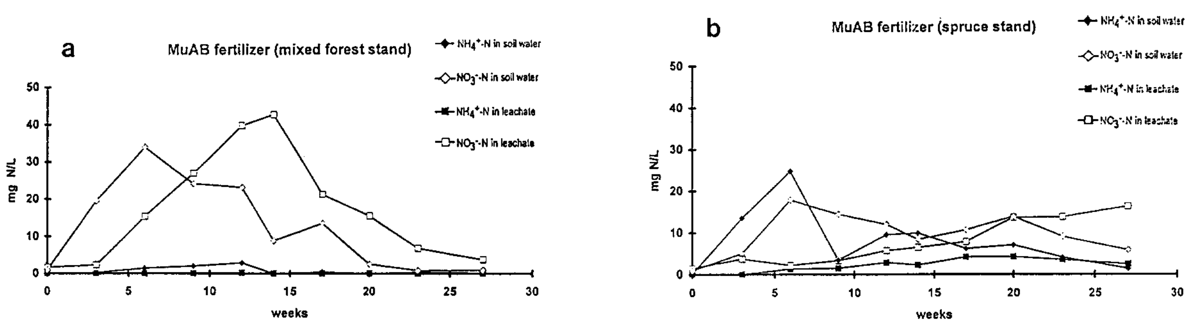

Fig. 3. Nitrate and ammonia in soil water and leachate of MuAb fertilized cores from (**a**) the mixed forest and (**b**) the spruce forest sites. Coefficient of variation: 92%.

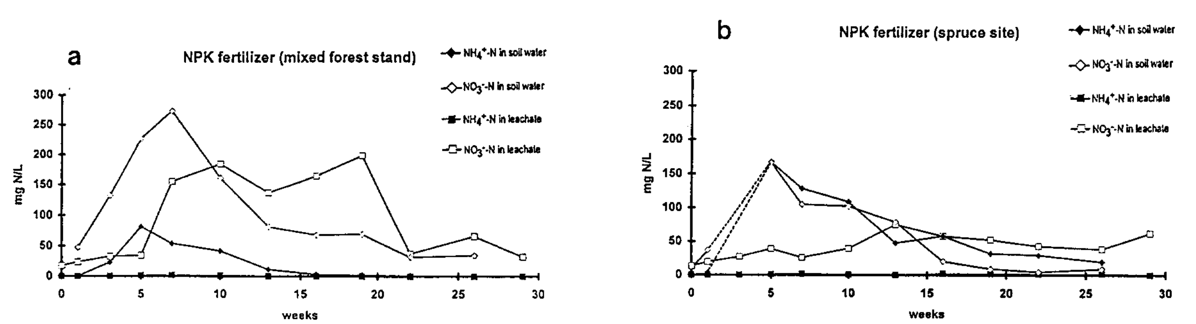

Fig. 4. Nitrate and ammonia in soil water and leachate of cores treated with fast-release NPK fertilizer from (**a**) the mixed forest and (**b**) the spruce forest sites. Coefficient of variation: 76%.

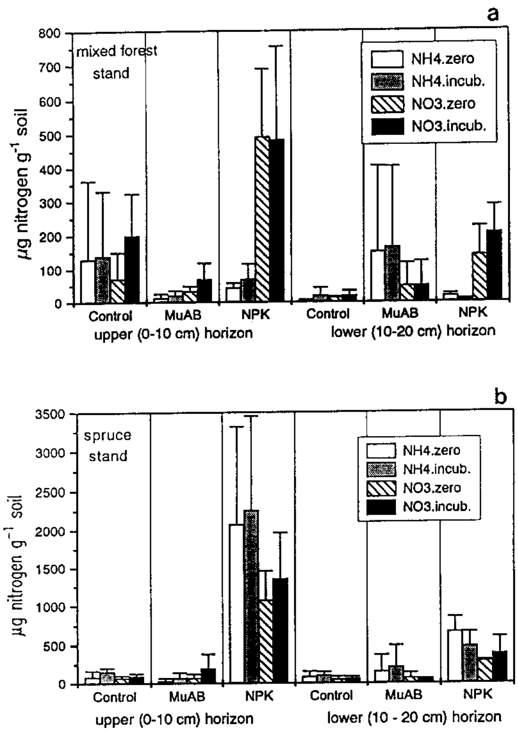

Fig. 5. Nitrogen mineralization assay at the end of the microcosm experiment: NH_4^+ and NO_3^- concentrations in the soil before and after a three-weeks incubation (mean±SE).

granules or immobilized by soil microbes. Despite the high application rates and the absence of trees, less than 10% of the applied N was washed out during the 28 weeks incubation.

As expected, the highest levels of NO_3^- and NH_4^+ were found for the mineral fertilizer (which actually is designed for rapid nutrient release), both in soil water and leachate (Fig.4). After a rapid increase until week 7 (270 mg NO_3^- -N L^{-1} on the mixed forest soils), the nitrate levels in the soil water decreased again. Nitrate concentrations reached maximum values of 200 mg L^{-1} in the leachate. The highest concentrations of NH_4^+ in the soil water of the mixed forest site were around 80 mg L^{-1}, NH_4^+ levels in the leachate were neglectable. On the spruce soils, NO_3^- and NH_4^+ were equally high (170 mg) in the soil water at week 5, both declined slowly towards the end of the incubation (Fig. 4; data for week 3 are missing) to levels around 20 mg L^{-1}. Compared to the mixed forest site nitrification seems to have been far slower. However, only little of the NH_4^+ showed up in the leachate, while NO_3^- levels remained constantly high at about 60 mg L^{-1} for several weeks (note the different scale in Figure 4 as compared to Figures 2-3). N losses through leaching were equivalent to 30% of the applied fertilizer N.

The incubation conditions with the very high application rates were "maximum risk conditions". Indeed, the data suggest potential problems with application of fast-release mineral fertilizer. MuAB fertilization, however, did not increase mineral nitrogen contents of soil water or leachate to critical levels (the current limit for NO_3^- in drinking water is 50 mg L^{-1} in Austria). Although mineral N values in soil water with MuAB fertilizer in this experiment were low, under field conditions in boreal forests Mu (Nitroform, ureaformaldehyde) application has resulted in 1,5-2 times higher stand volume increase compared to fast release mineral N fertilizers (Aarnio et al., 1993). Whether this is so for the sites investigated here should be studied in a long-term field experiment.

After the end of the incubations, soil samples from both the upper (0-10 cm) and the lower (10-20 cm) horizons were taken and sieved. They were then analyzed for their NH_4^+ and NO_3^- contents and incubated to determine mineralization rates. Further, basal respiration and microbial biomass were determined. Due to the variable depth of the O-horizon (see Table 1) the variability of results was high. The results for NH_4^+ and NO_3^- contents are given in Figure 5. Because of the high variability, we refrained from calculating the differences between incubated and not-incubated samples. For the control and MuAB cores, NH_4^+ and NO_3^- contents were in most cases lower than 150 $\mu g\ g^{-1}$ soil. Generally, the NH_4^+ and NO_3^- contents increased during the 3-week incubations. While for the mixed forest samples the nitrate contents were usually higher than the ammonia contents, the reverse was found for the spruce site. This is another indication for a reduced nitrification on the spruce site as has been indicated above. With the mixed forest soils, only for the MuAB cores (10-20 cm) higher NH_4^+ than NO_3^- contents were measured, supporting the view that stimulation of nitrifcation can be avoided by using slow-release fertilizers (Martikainen et al., 1989).

Microbial biomass and respiration were determined (Fig. 6) for the 0-10 and 10-20 cm horizons. Both tended to be lower on the fertilized than on the control cores. The effect, however, was significant (p<0,05) only for the fast-release fertilizer on the mixed forest soils. Observations about depression of microbial biomass after mineral N amendment have been made earlier by Nohrstedt et al. (1989) and Söderström et al. (1983). The metabolic quotient was higher for the spruce forest than for the mixed forest soils which is

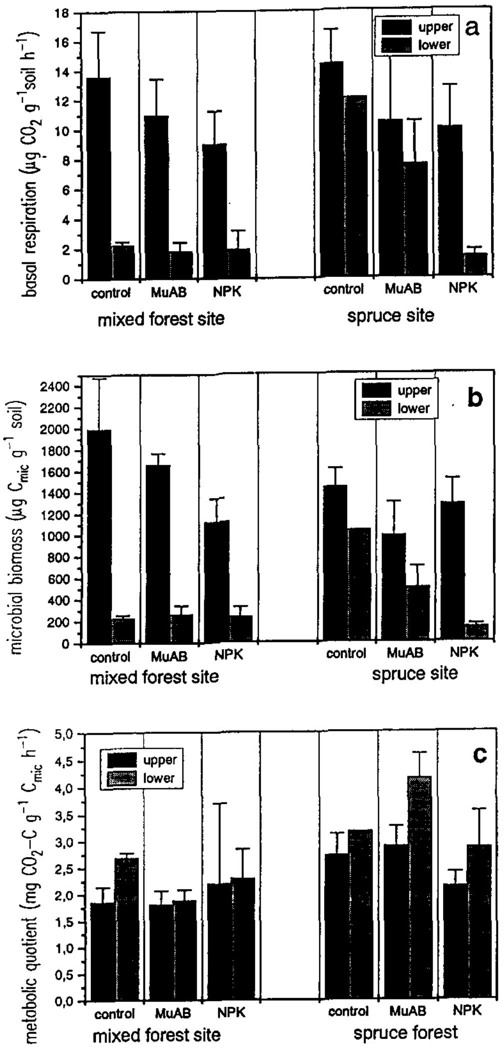

Fig. 6. (**a**) Basal respiration, (**b**) microbial biomass and (**c**) the metabolic quotient in upper (0-10 cm) and lower (10-20 cm) horizons at the end of the microcosm experiment (mean±SE).

in accordance with findings by Ding et al. (1992). The effects of the fertilizers on the qCO_2, however, were not significant. This indicates that none of the fertilizers to any serious degree seems to have affected the ecophysiological status of the microbial community during this short term microcosm experiment.

The very high loads of inorganic nitrogen for the NPK samples, along with a decreased C_{mic} content, indicate a considerable deficit in available C. Several explanations have been proposed to explain why forest fertilization may decrease microbial biomass. Among them are decreased C input through the roots (exudation, productivity, mycorrhizae), and decreased availability of C through condensation of N with humus (Nohrstedt et al., 1989). For the sites investigated here, decreased lignin degradation (which may be rate limiting for decomposition) after NPK treatment through repression of ligninolytic enzymes by ammonium (Berg and Staaf, 1980) is a possible explanation.

Summarized, MuAB fertilization increased nitrate concentrations in the soil water and nitrate leaching. However, compared to a fast-release NPK fertilizer, the rates were moderate. Microbial properties, as well as soil inorganic N content at the end of the experiment were not different from the control. The conditions chosen were such that the risk of N leaching was high (high application rate, incubation temperature higher than usually found in these forests, and the absence of tree roots). These findings thus suggest that MuAB fertilizer may safely be used (in terms of groundwater protection) also under field conditions, while fast-release NPK fertilizer is likely to pose problems. However, for further evaluation a field trial should be considered.

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