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## Effects of long-term fertilisation on soil properties in Scots pine and Norway spruce stands

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### Highlights

- N fertilisation increased the amount of carbon in the organic layer.
- N fertilisation decreased the C/N ratio in the surface soil.
- N addition increased the amount of most nutrients in the organic layer.
- N fertilisation tended to lower pH, although only slightly.

### Abstract

The response of surface soil after 45- to 52-years to repeated nitrogen and phosphorus fertilisation was studied. This study included 30 factorial experiments established in young (5- to 30-year-old) stands using plots of 900 m<sup>2</sup>, on average, and by randomising treatments within each experiment. Total amount of N added varied from 534 to 1908 kg ha<sup>-1</sup> and that of P from 69 to 193 kg ha<sup>-1</sup>, repeated at every second N fertilisation. Liming was performed twice; in total, 6000 kg ha<sup>-1</sup> of dolomite was applied. Nitrogen fertilisation increased the mass of the organic layer and the amount of carbon and consequently the amounts of most of the elements in the organic layer. In both the organic layer and the 0–10 cm layer of mineral soil, nitrogen fertilisation decreased the C/N ratio and tended to lower pH, although only slightly. Phosphorus fertilisation increased the amounts of P and Ca. Liming increased the total amounts of most elements in the organic layer, except for C and N. We were able to derive models to describe how changes in the chemical properties of the surface soil depended on doses of elements and on site and stand properties.

**Keywords** boreal forest; forest soils; nitrogen; phosphorus; *Pinus sylvestris* L.; *Picea abies* (L.) Karst.; liming

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## 1 Introduction

In the 1950's it was not clear which nutrients were most limiting for tree growth on upland soils in Finland, what would be suitable dosages of each nutrient and what would be the duration of a possible growth response. It was also unclear how site fertility, weather conditions, tree species and stand age would correlate with a possible growth response. Therefore, to explore these questions, fertilisation experiments were established in Finland (Viro 1967).

Since the 1950's many types of fertilisation experiments in the Nordic countries have revealed that nitrogen is the only nutrient which given alone could significantly increase tree growth on upland sites i.e. on sites with mor or moder layer on the top of mineral soil (Viro 1967; Nilsen 2001; Nohrstedt 2001; Saarsalmi and Mälkönen 2001; Jacobson and Pettersson 2010). It follows that nitrogen fertilisation will increase the amounts of soil organic matter, carbon and nitrogen, and in some cases also calcium, magnesium and phosphorus, and decrease the C/N ratio in the organic layer (Mälkönen et al. 1990; Högberg et al. 2006; Ring et al. 2011). Long-term nitrogen fertilisation in reasonable doses (150 to 200 kg ha<sup>-1</sup> per 5–10 years) seems to have no or very small effects on soil acidity, at least in terms of pH (Mälkönen et al. 1990; Högberg et al. 2006; Derome et al. 2009).

Additional nitrogen by deposition or fertilisation has been suspected to cause in addition to an increase in tree growth, enrichment of nitrogen in forest ecosystems with harmful consequences such as leaching of nitrate and base cations to ground and surface waters (Smolander et al. 1995; Ring et al. 2006), imbalance between nitrogen and other nutrients, e.g. boron deficiency (Möller 1983), and changes in ground vegetation (Ellenberg 1988; Mäkipää 1995; Strengbom and Nordin 2008; Hedwall et al. 2011). According to the reviews of Binkley and Högberg (1997) and Saarsalmi and Mälkönen (2001), however, there is no general risk for nitrogen saturation or even milder harmful effects in Finnish forests due to repeated nitrogen fertilisation. The relative effects of nitrogen fertilisation on tree stand and soil seem to be greater on less fertile sites with a high C/N ratio and in pine-dominated stands than on more fertile sites with a low C/N ratio and in spruce-dominated stands (Mälkönen et al. 1990; Nohrstedt 2001; Saarsalmi and Mälkönen 2001). If the C/N ratio of the organic layer is very low, around 20, intensive nitrogen fertilisation may, instead of increasing stand growth, cause or worsen growth disturbances due to boron deficiency (Aronsson 1983; Möller 1983; Saarsalmi and Tamminen 2005) and lead to leakage of nitrate from surface soil (Matzner and Grosholz 1997; Tietema et al. 1998).

In Finland, fertilisation in practical silviculture reached its maximum in 1975, decreasing sharply in the late 1970's and again in the late 1980's (Kukkola and Nöjd 2000). During the last 15 years fertilisation areas have increased again, and at present on uplands the annual area fertilised is c. 20 000 ha<sup>-1</sup>. Nitrogen deposition has decreased as much as 50% during the last 40 years: from maximum 8 kg ha<sup>-1</sup> a<sup>-1</sup> (1971–1988) to maximum 4 kg ha<sup>-1</sup> a<sup>-1</sup> (2009) (Järvinen and Vänni 1990) (<http://www.metla.fi/metinfo/kestavyys/c2-deposition-of-air.htm>).

Liming was one of the treatments used in Finnish fertilisation experiments because in agricultural soils, liming was a basic method used to increase plant production; on the other hand, Scots pine productivity was found to correlate with calcium in the soil (Viro 1951). However, broadcast liming on forested upland sites decreased or had no effect on tree growth (Derome et al. 1986; Nilsen 2001; Nohrstedt 2001). Later on, in the 1980's and 1990's, liming was used to counteract acidification of soils and waters, which was efficient in terms of acidity (pH) or acid buffering capacity, but hardly useful in Nordic boreal forests (Derome et al. 1986; Binkley and Högberg 1997; Derome et al. 2000; Sikström et al. 2001). However, liming raised soil pH, increased concentrations of Ca and Mg (Derome et al. 1986; Saarsalmi and Levula 2007; Saarsalmi et al. 2011) and also increased the mass and N content of the organic layer (Derome et al. 1986).

Phosphorus was included in Finnish fertilisation experiments as a heritage from agricultural practice, similarly to liming, and possibly due to some positive results from regeneration experiments (Nilsen 2001). Phosphorus and potassium have been shown to limit tree growth on many peat soils (Moilanen 2005), but not on upland sites (Nilsen 2001; Nohrstedt 2001; Saarsalmi and Mälkönen 2001). In some cases, when given together with nitrogen, phosphorus may also increase tree growth on upland sites, mostly in productive spruce stands (Viro 1972; Tamm 1991; Kukkola and Saramäki 1983). Phosphorus fertilisation increases the concentration and amount of phosphorus in surface soil (Kaunisto and Paavilainen 1988; Braekke and Finér 1991; Fransson and Bergkvist 2000) due to effective chemical (e.g. Ca, Al and Fe compounds) and biological fixation (plant structures), the level being higher years after the last fertilisation (Fransson and Bergkvist 2000).

Forest fertilisation has seldom been monitored for 50 years as it has in our Finnish fertiliser trials. Monitoring the characteristics of a tree stand is much easier than monitoring soil properties, because the dimensions of tree stems are systematically larger after every growing season, unlike most soil properties. Soil parameters may have trends, but may also show unsystematic annual, seasonal and human-induced (cuttings, sampling) variation (Kilian 1981; Lockman and Molloy 1984; Lundell 1987; Ilvesniemi 1991; Roberts et al. 1998; Tamminen and Derome 2005). During the past 50 years, soil sampling, pretreatment and analyses have varied considerably. In addition, in our Finnish case, there are no samples from before the first fertilisation, sampled soil layers have changed and storing of samples has not always been the same. To study the effects of long-term fertilisation on soil, temporal cross-section, i.e. sampling of all experiments at the same moment, is often the only option (Derome et al. 1986; Mälkönen et al. 1990; Högberg et al. 2006; Ring et al. 2011).

Our aim in this study was to ascertain what has happened in surface soil, when nitrogen, lime or phosphorus has been spread in the forest over a period of 45 to 52 years. We expect that nitrogen fertilisation has, on average, increased the amounts of carbon and nitrogen, decreased the C/N ratio but not changed the pH in the organic layer. We also expected that liming has increased pH and the amounts of calcium, magnesium and organic matter in the surface soil. In addition, we expected that phosphorus fertilisation would have increased the amount of phosphorus both in the organic layer and in the mineral soil. A further aim was to determine how soil changes due to fertilisation are correlated with the doses of fertilised elements and with site and stand properties.

## 2 Material and methods

### 2.1 Experiments

Professor P. J. Viro and his colleagues established about 200 factorial fertilisation experiments throughout the country during the years 1958–1965. Of these we were able to use 30 experiments which had been monitored until the year 2009 (Table 1, Fig. 1). The experimental treatments contained nutrients N, P and liming (Ca+Mg), arranged in a factorial design ( $2^3=8$  plots). The eight treatment combinations were: unfertilised control, N, P, lime, N+P, N+lime, P+lime and N+P+lime.

Experiments were established in young (5- to 30-year-old) Scots pine (*Pinus sylvestris* L.) and Norway spruce [*Picea abies* (L.) Karst] stands using square plots with an average area of 900 m<sup>2</sup> (625–1000 m<sup>2</sup>) and by randomising treatments within each experiment. The distance between plots was at least 10 m. When thinning was needed, the trees to be removed were chosen in connection with tree stand measurements in order to control the removal. After that, thinning was carried out before the next growing season. Branches and tops were always left in the stand. Natural removal is included in the growth results.

**Table 1.** Site and stand characteristics of the experiments and total amount of the applied fertilisers.

Exp.	Lat. °N	Long. °E	Tempsum d.d. (>5 °C)	Tree species	Started at	Stand age at 2009	SI <sup>a)</sup> , m	Prod <sup>a)</sup> , m <sup>3</sup> ha <sup>-1</sup> a <sup>-1</sup>	N, kg ha <sup>-1</sup>	P, kg ha <sup>-1</sup>	Lime, kg ha <sup>-1</sup>	Soil texture <sup>b)</sup>
25	61.817	29.329	1239	Pine	1958	84	27.9	7.4	1676	149	6000	SL
33	61.875	29.343	1234	Pine	1958	70	29.3	5.9	1908	120	6000	LS
35	62.409	28.707	1168	Spruce	1958	77	18.3	8.3	714	144	6000	SL
36	62.409	28.710	1168	Spruce	1958	77	19.9	8.5	714	144	6000	SL
37	61.415	28.545	1240	Pine	1958	75	20.0	7.4	714	109	6000	LS
38	61.416	28.542	1233	Pine	1958	75	21.5	8.2	714	109	6000	S
52	62.024	24.811	1154	Pine	1959	58	25.9	8.1	1496	149	6000	LS
55	61.662	29.304	1170	Pine	1959	65	26.1	9.8	534	69	6000	SL
56	62.927	25.607	1025	Pine	1959	76	22.8	4.5	1496	149	6000	SL
57	62.935	25.678	1020	Spruce	1959	77	19.4	5.9	1496	149	6000	SL
58	62.935	25.677	1024	Spruce	1959	77	21.5	6.9	1496	149	6000	SL
60	62.930	25.666	1020	Spruce	1959	77	23.0	7.7	1136	109	6000	SL
64	61.493	29.066	1215	Pine	1959	90	22.0	5.3	714	109	6000	LS
67	61.537	29.062	1160	Pine	1959	70	21.1	9.3	714	109	6000	SL
68	61.956	27.575	1147	Pine	1959	84	21.5	7.6	1404	149	6000	S
73	62.759	24.747	1034	Pine	1959	55	25.9	8.0	1254	160	6000	SL
75	62.913	24.571	1028	Pine	1959	54	25.3	4.9	742	109	6000	SL
76	62.912	24.570	1028	Pine	1959	54	26.3	5.2	742	109	6000	SL
77	62.911	24.568	1028	Pine	1959	54	23.3	4.1	742	109	6000	SL
82	63.300	25.340	999	Pine	1959	54	17.6	3.7	965	193	6000	SL
97	62.574	24.119	1074	Pine	1960	61	24.8	8.5	742	193	6000	LS
98	62.579	24.125	1074	Pine	1959	60	24.5	6.9	1356	69	6000	SL
103	63.215	24.624	1006	Pine	1960	58	22.2	4.2	742	109	6000	LS
106	63.389	24.300	1013	Pine	1960	73	15.0	3.0	1136	120	6000	LS
107	63.095	24.294	1013	Pine	1960	63	18.0	2.6	742	153	6000	LS
113	61.172	26.050	1251	Spruce	1961	58	28.4	12.8	1486	193	6000	SL
135	67.250	23.869	791	Pine	1961	87	17.6	4.6	1194	120	6000	SL
155	61.169	26.048	1250	Spruce	1962	59	28.2	13.4	1074	160	6000	SL
157	61.111	26.026	1229	Pine	1962	62	24.2	7.1	1317	160	5000	LS
194	66.855	27.133	754	Spruce	1964	74	19.5	1.5	1110	160	6000	SL

a) SI is H100 and Prod is the mean annual stem volume production for plots not fertilised with N (4 per experiment).

b) Soil texture classes: S=sand, LS=loamy sand and SL=sandy loam.

## 2.2 Fertilisation

At the beginning the nitrogen fertiliser was ammonium sulphate (N 26%) (1958–1963). Later urea was used (N 46%). From 1972 on ammonium nitrate with lime (N 27%, Ca 6%, Mg 3%) (1972–1996) was used. From 1997 its composition was a bit different (N 26%, Ca 3%, Mg 1%, B 0.02%). Nitrogen doses were at first 82 kg ha<sup>-1</sup> (1958–1962), ending up with 180 kg ha<sup>-1</sup> (1970–). In Lapland smaller doses were used (120–150 kg ha<sup>-1</sup>). Fertilisation intervals were usually 5 or 10 years, in some cases 4 or 6 years. The total amount of N added varied from 534 to 1908 kg ha<sup>-1</sup>. The time period from the most recent N fertilisation to soil sampling varied from 1 to 20 years.

Liming was performed twice, first with 2000 kg ha<sup>-1</sup> of dolomite powder when the experiment was established and later with 4000 kg ha<sup>-1</sup> of dolomite after 17–23 years had elapsed. The time period from the most recent liming to soil sampling varied from 23 to 31 years. Unfortunately, the chemical composition of the lime material used is unknown.

The phosphorus fertiliser was superphosphate (P 9%, Ca 20%); and the P dose was, on average, 40 kg ha<sup>-1</sup>, a Ca dose being c. 90 kg ha<sup>-1</sup>. Phosphorus fertilisation was repeated not as often

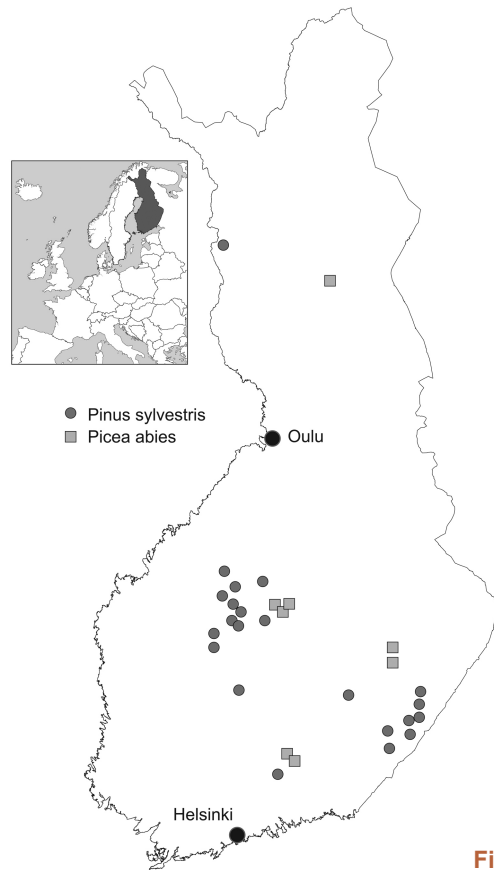


Fig. 1. Fertilisation experiments.

as nitrogen fertiliser. The total amount of P added varied from 69 to 193 kg ha<sup>-1</sup>. The time period from the most recent P fertilisation to soil sampling varied from 2 to 30 years.

All fertilisers were spread manually and thus evenly. Fertilisation and thinning were carried out also on a 5 m wide buffer zone surrounding the plot.

### 2.3 Soil sampling

Soil samples were taken in spring 2009, i.e. 45 to 52 years after the first treatments. A composite sample from the organic layer (forest floor) consisted of 25 subsamples (d=60 mm) taken systematically. A composite sample from the 0–10 cm mineral soil layer was taken with a volumetric cylinder (d=39 mm, h=100 mm) or on stony sites with spades at five spots, i.e. the plot center and corners. Thickness of the organic layer and average penetration of a steel rod (d=10 mm) into the mineral soil to estimate stoniness (max. to 30 cm) was measured at every spot where the organic layer was sampled. The volumetric proportion of stones and boulders (d>20 mm) was estimated based on mean penetration of the steel rod: percentage of stones in the 0–30 cm layer = 82.5 – 2.75\*penetration<sub>cm</sub> and percentage of stones in the 0–10 cm layer = 82.5 – 8.25\*penetration<sub>cm</sub> (Viro 1952; Tamminen 1991).

Bulk density for mineral soils sampled with spades was predicted with a regression equation (1).

$$BD_2 \left( \text{kg dm}^{-3} \right) = 1.492 - 0.236 * \ln(OM + 1) - 0.0076 * \text{Gravel} - 0.00222 * \text{Fines}; n = 176; r^2 = 0.892 \quad (1)$$

where OM is concentration of organic matter (%), Gravel is the mass percentage (%) of the 2–20 mm fraction, Fines is the mass percentage (%) of the under 63  $\mu\text{m}$  fraction,  $n$  is the number of volumetric samples and  $r^2$  is a squared correlation coefficient between the measured and predicted bulk densities.

## 2.4 Pretreatment of soil samples

Samples were delivered to the laboratory within 2 to 3 days after collection, where they were first weighed fresh and then dried at c. 40 °C. After drying, the samples were weighed as air-dry. Organic samples were ground in a mill with a 2 mm bottom sieve, and mineral soil samples were sieved with a 2 mm sieve to obtain the mass of fine earth and the proportion of gravel.

## 2.5 Laboratory analysis

All samples were analysed at the laboratory of the Finnish Forest Research Institute in Vantaa. Moisture and concentration of organic matter of samples were determined with a Leco TGA 601 oven and total C and N concentrations with a Leco CHN 1000 instrument. pH was determined in a 0.01 M  $\text{CaCl}_2$  suspension, where 10 ml of sample was wetted with 25 ml of liquid, stirred and measured after 2 hours with a calibrated (buffer solutions of pH=4.0 and 7.0) pH meter. All samples were extracted with 1 M acid ammonium acetate (pH=4.65) (abbreviation AAA) (Halonen et al. 1983). Then 10 ml of sample and 100 ml of extractant were shaken for 2 hours and filtered. For organic samples, dry combustion was used. Ashed samples (Leco TGA 601 oven) were extracted with HCl (Halonen et al. 1983). Elemental concentrations in AAA and dry combustion analyses were determined with the ICP device Thermo Jarrell Ash Iris Advantage.

## 2.6 Tree stand measurements

Tree stands were measured at the time the experiments were established and after that at intervals of 4 to 10 years. The breast-height diameter ( $d_{1.3}$ ) of all trees was measured. On each plot permanent sample trees representing different size categories were chosen for tree height measurements using a hypsometer. The sample trees were used for estimating the height and stem volume.

Stand characteristics were calculated using the KPL calculation programme (Heinonen 1994). A function based on breast height diameter and tree height was used for calculating the volume of the sample trees (Laasasenaho 1982). Plotwise volume equations based on breast height diameter were then calculated using the data from the sample trees in order to estimate the volume of the other trees. Mean annual production was obtained by dividing total stem volume production, including removals, by the number of years since first fertilisation.

## 2.7 Statistical analyses

Preliminary checking of data revealed that only N fertilisation had increased the thickness of the organic layer; and therefore a new variable, mean thickness of the organic layer on plots not fertilised with N, was introduced to be used as a covariate in ANOVA and as an independent variable in regression analyses. The aim was to remove the effect of the originally variable thickness of the organic layer between experiments on soil analysis variables.

Skewed distributions of some variables were normalised by using logarithm transformation,  $\ln(x)$  or  $\ln(x+1)$ . In the logarithmic models, i.e. in the models where a dependent variable was in the form  $\ln(y_i)$ , the constant was adjusted to yield the original arithmetic mean by adding a residual variance of the regression model to the constant, i.e.  $\text{constant} + s_e^2/2$ .



Analyses of variance or covariance and regression analyses were performed using a mixed model technique. The experiment was treated as a random effect, and fertilisation treatment and possible covariates or independent variables as fixed effects. In all statistical tests the level of significance was set at  $p < 0.05$ . For statistical calculations a PASW Statistics 17 package was used.

### 3 Results

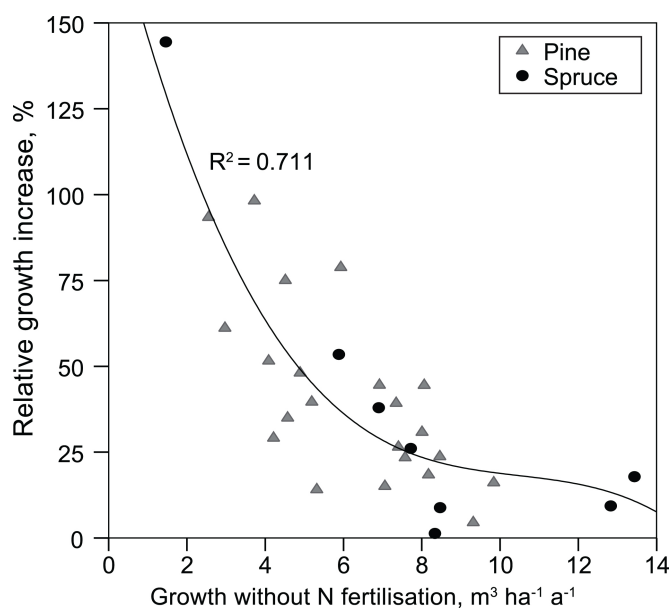
#### 3.1 Tree growth

Mean annual production of the plots not fertilised with N was  $6.2$  ( $2.6$ – $9.8$ )  $\text{m}^3 \text{ha}^{-1} \text{a}^{-1}$  in pine stands and  $8.1$  ( $1.5$ – $13.4$ )  $\text{m}^3 \text{ha}^{-1} \text{a}^{-1}$  in spruce stands. Only N fertilisation increased stand production. In the pine stands, growth increased, on average,  $2.2$  ( $0.4$ – $4.7$ )  $\text{m}^3 \text{ha}^{-1} \text{a}^{-1}$  and in spruce stands  $1.8$  ( $0.1$ – $3.2$ )  $\text{m}^3 \text{ha}^{-1} \text{a}^{-1}$ . Relative growth response to N fertilisation correlated negatively with annual production of the site (Fig. 2), and positively with the C/N ratio of the organic layer ( $r_{\text{Spearman}} = 0.61$ ).

#### 3.2 Effects of fertilisation treatments on soil

N and P fertilisation and liming affected soil characteristics independently, regardless of treatment combination (Tables 2 and 3), i.e. there was no interaction between treatments. Hence, the effects of N, P and lime treatments on soil characteristics could be analysed using dummy variables N (0/1), P (0/1) and lime (0/1) or cumulative N (kg/ha), P (kg/ha) and lime (Mg/ha) doses as independent variables.

According to the mixed model ANOVA, the main treatments N, P and liming had statistically significant effects on soil concentrations of the respective elements N, P and Ca, both in the organic layer and in the mineral soil (Table 2). Liming had the strongest effect on soil as the variables pH and Ca concentrations differed most between the treatments (Table 2). About  $6000 \text{ kg ha}^{-1}$  of lime increased soil  $\text{pH}_{\text{CaCl}_2}$ , on average,  $0.4$  to  $1$  units. In the organic layer, soil pH was



**Fig. 2.** Relative increase in stand production caused by nitrogen fertilisation as a function of mean annual production on unfertilised (no N) plots.

**Table 2.** Concentrations of elements by fertilisation treatment. Covariates in the ANOVA were effective temperature sum, tree species, i.e. pine (0/1), and mean thickness of the organic layer on plots not fertilised with N.

Variable <sup>b)</sup>	Treatments <sup>a)</sup>								F value <sup>c)</sup>
	Cntrl	N	P	L	NP	NL	PL	NPL	
Organic layer									
C <sub>tot</sub> , g kg <sup>-1</sup>	442a	443a	428ab	362e	444a	403bc	370de	396cd	28.4
N <sub>tot</sub> , g kg <sup>-1</sup>	12.6b	15.2a	12.1b	10.5c	15.1a	14.4a	10.9c	14.3a	60.0
C/N	35.7a	29.6b	38.1a	35.2a	29.8b	28.7b	34.9a	28.4b	66.7
Ca <sub>tot</sub> , mg kg <sup>-1</sup>	2823c	2960c	4261c	8246b	4378c	9420ab	10471a	10525a	87.2
K <sub>tot</sub> , mg kg <sup>-1</sup>	769a	622cd	734ab	693bc	597d	620d	738ab	641cd	13.6
Mg <sub>tot</sub> , mg kg <sup>-1</sup>	396b	502b	386b	1841a	491b	2019a	2104a	2231a	30.5
P <sub>tot</sub> , mg kg <sup>-1</sup>	796b	757bc	946a	706c	912a	714bc	951a	966a	33.4
pH <sub>CaCl2</sub>	3.22d	3.21d	3.38bc	4.06a	3.35cd	4.07a	4.19a	4.18a	191
Ca <sub>AAA</sub> , mg kg <sup>-1</sup>	1964d	2014d	2902cd	5157b	2963c	5922ab	6212a	6255a	118
K <sub>AAA</sub> , mg kg <sup>-1</sup>	784a	581c	752ab	687b	583c	583c	704b	570c	28.1
Mg <sub>AAA</sub> , mg kg <sup>-1</sup>	273b	347b	252b	773a	337b	904a	782a	912a	45.3
P <sub>AAA</sub> , mg kg <sup>-1</sup>	230bc	179de	270a	184de	232bc	157e	246ab	212bcd	25.9
Mineral soil									
C, g kg <sup>-1</sup>	25.9bc	30.5ab	25.3c	29.2abc	29.6abc	28.4abc	27.5abc	31.8a	4.39
N <sub>tot</sub> g kg <sup>-1</sup>	1.03cd	1.25ab	1.02d	1.13bcd	1.20abcd	1.21abc	1.05cd	1.34a	7.80
C/N	25.9ab	24.6bc	25.1abc	26.1ab	24.8bc	23.7c	27.0a	24.4bc	5.51
pH <sub>CaCl2</sub>	3.99b	3.80b	3.92b	4.68a	3.81b	4.37a	4.71a	4.55a	74.1
Ca <sub>AAA</sub> , mg kg <sup>-1</sup>	63c	78c	120c	680a	105c	474b	694a	683a	52.2
K <sub>AAA</sub> , mg kg <sup>-1</sup>	31.3a	30.2a	32.7a	33.2a	29.6a	27.8a	32.4a	30.1a	2.17
Mg <sub>AAA</sub> , mg kg <sup>-1</sup>	11.1b	15.3b	12.9b	83.5a	16.3b	80.7a	81.1a	86.0a	42.5
P <sub>AAA</sub> , mg kg <sup>-1</sup>	9.6c	8.8cd	12.4a	7.1de	11.8ab	6.1e	10.0bc	9.2cd	20.3

a) Cntrl=control, N=nitrogen fertilisation, P=phosphorus fertilisation, L=liming.

b) AAA=acid ammonium acetate extraction, tot=dry combustion concentration.

c) F value from mixed model ANOVA with covariates. If  $F_{7,200} > 3.64$ , then  $p < 0.001$ .**Table 3.** Amounts of elements by fertilisation treatment. Potential covariates in the ANOVA were effective temperature sum, tree species, i.e. pine (0/1), and mean thickness of the organic layer on plots not fertilised with N.

Variable <sup>b)</sup>	Treatments <sup>a)</sup>								F value <sup>c)</sup>
	Cntrl	N	P	L	NP	NL	PL	NPL	
Organic layer									
C <sub>tot</sub> , Mg ha <sup>-1</sup>	16.8b	22.8a	17.5b	16.0b	22.5a	21.0a	16.4b	20.5a	25.2
N <sub>tot</sub> , kg ha <sup>-1</sup>	488b	778a	496b	468b	764a	741a	485b	734a	58.9
Ca <sub>tot</sub> , kg ha <sup>-1</sup>	112d	150cd	177cd	373b	221c	488a	485a	545a	64.9
P <sub>tot</sub> , kg ha <sup>-1</sup>	30.5e	38.2cd	38.8cd	31.7e	46.1ab	36.8d	42.2bc	49.8a	35.6
Mineral soil									
C <sub>tot</sub> , Mg ha <sup>-1</sup>	20.8bc	24.3ab	20.3c	22.6abc	23.7ab	22.7abc	22.6abc	24.5a	5.28
N <sub>tot</sub> , kg ha <sup>-1</sup>	842d	998a	822d	883bcd	961abc	977ab	860c	1023a	9.38
Org. + min. soil									
Ca <sub>AAA</sub> , kg ha <sup>-1</sup>	127d	162d	214d	771bc	233d	693c	862ab	870a	88.8
P <sub>AAA</sub> , kg ha <sup>-1</sup>	16.4cd	16.1cd	21.2a	14.0de	21.5a	13.2e	19.4ab	18.5bc	33.3

a) Cntrl=control, N=nitrogen fertilisation, P=phosphorus fertilisation, L=liming.

b) AAA=acid ammonium acetate extraction, tot=dry combustion concentration.

c) F value from mixed model ANOVA with covariates. If  $F_{7,200} > 3.64$ , then  $p < 0.001$ .



higher in the P and NP treatment than in the control and N treatment probably due to extra Ca added in P fertilizer (Table 2).

According to the F values from ANOVA, liming had the most striking effect on amounts of soil Ca; but N and P fertilisation also increased the amounts of the respective elements (Table 3). The amount of total Ca seemed to be higher in the organic layer in the NL (L=lime), PL and NPL treatments compared to the L-alone treatment, perhaps due to extra Ca in the N and P fertilisers; in ammonium nitrate-based N fertilisers the amount of Ca was 3–6% and in superphosphate 20%. In addition, the amount of total P in the organic layer tended to be higher in the combined NP and NPL treatments than in the P treatment, perhaps due to the higher mass of the organic layer in the N-fertilised plots.

Nitrogen fertilisation increased the concentrations of N, and decreased the C/N ratio and pH both in the organic layer and in the 0–10 cm layer of mineral soil (Tables 4 and 5). According to the N equation in Table 4, the N concentration of the organic layer would have increased from 11.7 g kg<sup>-1</sup> (no N) to 14.8 or to 17.7 g kg<sup>-1</sup> with the N dose 1000 or, respectively, with the N dose 2000 kg ha<sup>-1</sup>. The corresponding C/N ratios would have been 37, 32 and 26. According to the pH equations, the N dose 2000 kg ha<sup>-1</sup> would have lowered the pH<sub>CaCl2</sub> in the organic layer from 3.1 (no N) to 3.0, and in the 0–10 cm mineral soil layer from 4.0 to 3.5. Similarly, N fertilisation tended to decrease the concentrations of AAA-extractable P and K and total K. For instance, with the N dose 2000 kg ha<sup>-1</sup> the AAA- extractable concentration of P in the organic layer would have decreased from 222 to 154 mg kg<sup>-1</sup> (Table 5). In both soil layers, P fertilisation increased total and AAA- extractable concentrations of P (Tables 4 and 5). Liming increased both pH and the concentrations of Ca, but decreased the AAA- extractable concentrations of P in both soil layers. In the organic layer, liming also decreased the concentrations of N and K<sub>AAA</sub> (Tables 4 and 5).

Nitrogen fertilisation increased the mass of the organic layer and the amount of C and consequently the amounts of most elements in the organic layer (Tables 6 and 7) because the mass of

**Table 4.** Regression equations for total concentrations of elements as a function of cumulative amounts of fertilisers, and site and tree-stand properties. All coefficients are statistically significant ( $p < 0.05$ ).

Independent variables <sup>a)</sup>	Dependent variables					
	Organic layer					Mineral soil 0–10 cm
	N <sub>tot</sub> g kg <sup>-1</sup>	C/N	lnCa <sub>tot</sub> mg kg <sup>-1</sup>	K <sub>tot</sub> mg kg <sup>-1</sup>	P <sub>tot</sub> mg kg <sup>-1</sup>	ln(N <sub>tot</sub> +1) g kg <sup>-1</sup>
Constant <sup>b)</sup>	–4.61	50.9	7.16	237	316	–.278
N, Mg ha <sup>-1</sup>	3.07	–5.76		–116		.0820
P, kg ha <sup>-1</sup>			.000849		1.47	
Lime, Mg ha <sup>-1</sup>	–.208		.167			
Ol. thickn., cm	1.22					
Stand age, a	.0978		.00701	5.77	5.56	
Pine (0/1)	–2.24	8.09			–129	–.278
Tempsum, d.d.	.00703	–.0197				.000896
Fines, %			.0107	3.47	4.72	.00680
Stones, %						.0531
R <sup>2</sup> (obs./pred.)	.667	.738	.688	.441	.619	.667
						.256

a) N=cumulative amount of fertilised nitrogen, P=cumulative amount of fertilised phosphorus, Lime=cumulative amount of spread lime, Ol. thickn.=mean organic layer thickness on plots not fertilised with nitrogen, Stand age=stand age at year 2009, Pine=1, if pine, 0 if spruce, Tempsum=average effective temperature sum, Fines=sum of clay+silt, i.e. under 63 µm fraction, Stones=Volumetric percentage of stones.

b) In the logarithmic models the constant has been adjusted to yield the original arithmetic mean by adding a residual variance of the regression model, constant+s<sub>r</sub><sup>2</sup>/2.

**Table 5.** Regression equations for pH and concentrations of elements extracted with acid ammonium acetate as a function of cumulative amounts of fertilisers and site and tree-stand properties. All coefficients are statistically significant.

Independent variables <sup>a)</sup>	Dependent variables							
	Organic layer				Mineral soil 0–10 cm			
	pH <sub>CaCl2</sub>	Ca <sub>AAA</sub> mg kg <sup>-1</sup>	K <sub>AAA</sub> mg kg <sup>-1</sup>	P <sub>AAA</sub> mg kg <sup>-1</sup>	pH <sub>CaCl2</sub>	lnCa <sub>AAA</sub> mg kg <sup>-1</sup>	lnK <sub>AAA</sub> mg kg <sup>-1</sup>	lnP <sub>AAA</sub> mg kg <sup>-1</sup>
Constant <sup>b)</sup>	3.88	2879	932	163	3.31	4.00	2.29	1.97
N, Mg ha <sup>-1</sup>	-.0874		-135	-33.7	-.248	-.207		
P, kg ha <sup>-1</sup>				.412				.00295
Lime, Mg ha <sup>-1</sup>	.139	.562	-6.46	-4.75	.114	.360		-.0464
Ol. thickn., cm	-.120					-.240	-.163	-.150
Stand age, a			5.27					
Pine (0/1)	-.342	-1222			.421			
Tempsum, d.d.			-.495				.000855	
Fines, %				1.96	.0101	.0292	.0189	.0231
Stones, %							.00565	
R <sup>2</sup> (obs./pred.)	.811	.756	.442	.346	.646	.575	.583	.540

a) N=cumulative amount of fertilised nitrogen, P=cumulative amount of fertilised phosphorus, Lime=cumulative amount of spread lime, Ol. thickn. = mean organic layer thickness on plots not fertilised with nitrogen, Stand age=stand age at year 2009, Pine = 1, if pine, 0 if spruce, Tempsum=average effective temperature sum, Fines=sum of clay+silt, i.e. under 63 µm fraction, Stones= Volumetric percentage of stones.

b) In the logarithmic models the constant has been adjusted to yield the original arithmetic mean by adding a residual variance of the regression model, constant +  $s^2/2$ .

**Table 6.** Regression equations for total amounts of elements as a function of cumulative amounts of fertilisers, and site and tree-stand properties. All coefficients are statistically significant.

Independent variables <sup>a)</sup>	Dependent variables					Mineral soil 0–10 cm	
	Organic layer						
	C <sub>tot</sub> kg ha <sup>-1</sup>	N <sub>tot</sub> kg ha <sup>-1</sup>	lnCa <sub>tot</sub> kg ha <sup>-1</sup>	K <sub>tot</sub> kg ha <sup>-1</sup>	P <sub>tot</sub> kg ha <sup>-1</sup>	lnC <sub>tot</sub> kg ha <sup>-1</sup>	lnN <sub>tot</sub> kg ha <sup>-1</sup>
Constant <sup>b)</sup>	8898	579	5.01	35.7	39.7	8.21	5.28
N, Mg ha <sup>-1</sup>	4476	245	.139		6.31	.104	.158
P, kg ha <sup>-1</sup>			.000829		.0727		
Lime, Mg ha <sup>-1</sup>	-.229	-4.10	.181	.325		.00769	
Tempsum, d.d.						.00148	.00134
Ol. thickn., cm	2562						
Fines, %						.00957	.00845
Pine (0/1)		-125	-.338	-8.01		-.278	-.405
R <sup>2</sup> (obs./pred.)	.400	.544	.593	.192	.468	.578	.641

a) N=cumulative amount of fertilised nitrogen, P=cumulative amount of fertilised phosphorus, Lime=cumulative amount of spread lime, Ol. thickn. = mean organic layer thickness on plots not fertilised with nitrogen, Pine = 1, if pine, 0 if spruce, Tempsum=average effective temperature sum, Fines=sum of clay+silt, i.e. under 63 µm fraction.

b) In the logarithmic models the constant has been adjusted to yield the original arithmetic mean by adding a residual variance of the regression model, constant +  $s^2/2$ .

**Table 7.** Regression equations for elemental amounts extracted with acid ammonium acetate as a function of cumulative amounts of fertilisers and site and tree-stand properties. All coefficients are statistically significant.

Independent variables <sup>a)</sup>	Dependent variables							
	Organic layer				Mineral soil 0–10 cm			
	Ca <sub>AAA</sub> kg ha <sup>-1</sup>	K <sub>AAA</sub> kg ha <sup>-1</sup>	P <sub>AAA</sub> kg ha <sup>-1</sup>	S <sub>AAA</sub> kg ha <sup>-1</sup>	lnCa <sub>AAA</sub> kg ha <sup>-1</sup>	lnK <sub>AAA</sub> kg ha <sup>-1</sup>	lnP <sub>AAA</sub> kg ha <sup>-1</sup>	S <sub>AAA</sub> kg ha <sup>-1</sup>
Constant <sup>b)</sup>	121	23.9	-5.65	-2.34	5.78	2.43	1.649	2.823
N, Mg ha <sup>-1</sup>	19.2	1.14		.756	-.206	.0854		.201
P, kg ha <sup>-1</sup>			.0193	.00283			.00302	
Lime, Mg ha <sup>-1</sup>	29.4			-.113	.36		-.0477	-.0983
Tempsum, d.d.						.000781		
Ol. thickn., cm		3.78	2.29	.786	-.305	-.118		
Fines, %					.0246	.012	.0165	
Stones, %							-.0065	-.00832
Pine (0/1)	-59.8	-8.28	-2.86					
Stand age, a			.123	.065	-.024			.0114
R <sup>2</sup> (obs./pred.)	.67	.235	.469	.348	.624	.505	.433	.380

a) N=cumulative amount of fertilised nitrogen, P=cumulative amount of fertilised phosphorus, Lime=cumulative amount of spread lime, Ol. thickn. = mean organic layer thickness on plots not fertilised with nitrogen, Stand age=stand age at year 2009, Pine=1, if pine, 0 if spruce, Tempsum=average effective temperature sum, Fines=sum of clay+silt, i.e. under 63 µm fraction, Stones= Volumetric percentage of stones.

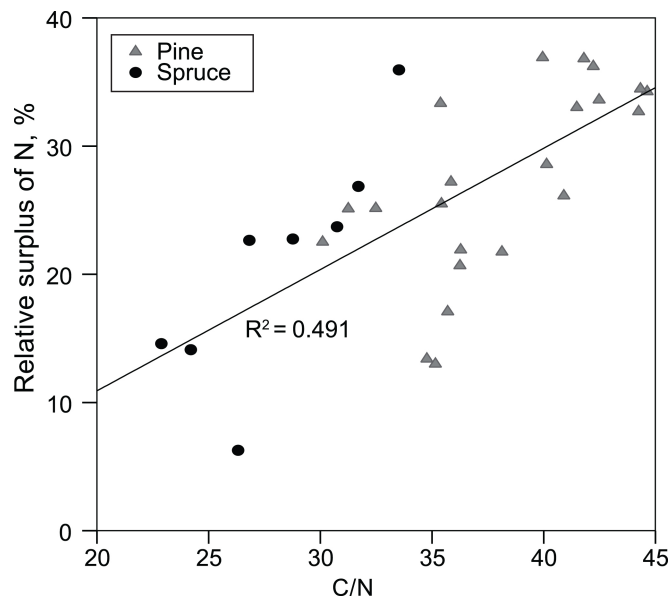
b) In the logarithmic models the constant has been adjusted to yield the original arithmetic mean by adding a residual variance of the regression model, constant + s<sup>2</sup>/2.

the organic layer systematically correlated with the amounts of all elements from K ( $r=0.53$ ) to C ( $r=0.81$ ). According to the C equation in Table 6, the N dose 2000 kg ha<sup>-1</sup> would have increased the amount of C in the organic layer from 18 (no N fertiliser) to 27 Mg ha<sup>-1</sup>. Correspondingly, the N dose 1000 or 2000 kg ha<sup>-1</sup> would have increased the amount of N in the organic layer from 454 (no N) to 699 or 944 kg ha<sup>-1</sup>, respectively.

Phosphorus fertilisation increased the amounts of P in both the organic and the mineral soil layers, and the amount of Ca in the organic layer (Table 6 and 7). In the organic layer, liming increased the amounts of total and AAA- extractable Ca and total K but decreased the amounts of C and N (Tables 6 and 7). In the mineral soil, liming increased the amounts of C and Ca<sub>AAA</sub> and decreased the amounts of P<sub>AAA</sub> (Tables 6 and 7).

There was a significant negative correlation between the time period from most recent fertilisation and the total amount of nutrient added in fertiliser. Consequently, possible effect of the time period from most recent N or P fertilization on the results was surpassed by the total amount of corresponding nutrient added during the study.

We estimated that, on average, 7% of P, 21% of Ca and 25% of N added in fertilisers was found in the organic layer, and 39% of added N was found in the organic + 0–10 cm mineral soil layer. The fertiliser N found, i.e. the relative surplus of N in the organic layer, correlated positively with the C/N ratio in the organic layer (Fig. 3).



**Fig. 3.** Relative surplus of N in the organic layer, i.e. (amount of N on the fertilised plots–amount of N on the non-fertilised plots)/amount of N in fertilisers, as a function of the C/N ratio in the organic layer on the unfertilized plots.

## 4 Discussion

Trees, other plants and microbes use fertiliser N quickly, because N is a growth-limiting nutrient that circulates tightly within boreal forest ecosystem (Nömmik and Möller 1981; Nömmik and Larsson 1989; Preston and Mead 1994; Weetman et al. 1997; Tamm et al. 1999). Fertiliser N increases production and annual above- and below-ground litter deposition 5 to 10 years after each fertilisation (Kukkola and Saramäki 1983; Mälkönen et al. 1980; Lipas 1988; Mäkipää 1994; Helmisaari and Hallbäcken 1998; Högberg et al. 2006; Jacobson and Pettersson 2010). In carefully selected Finnish coniferous stands, N application ( $150 \text{ kg ha}^{-1}$ ) usually gives a growth increase of  $12\text{--}20 \text{ m}^3 \text{ ha}^{-1}$  (Saarsalmi and Mälkönen 2001; Nohrstedt 2001).

Long-term N fertilisation favours more demanding understorey plants, i.e. herbs and grasses, rather than dwarf shrubs and mosses (Mälkönen et al. 1980; Mäkipää 1994), producing faster-decomposable litter with a lower C/N ratio. On the other hand, N fertilisation seems to decrease microbial biomass and activity, e.g. soil respiration and hence decomposition of litter (Söderström et al. 1983; Nohrstedt et al. 1989; Smolander et al. 1994; Olsson et al. 2005). The signs of long-term N fertilisation detected in the organic layer in our study, i.e. greater amounts of organic carbon and nitrogen and a lower C/N ratio, are probably due to greater and N-richer production of plant litter and retarded decomposition (Smolander et al. 2000). Because the C/N ratio in the organic layer decreased due to N fertilisation and because the C/N ratio correlates closely with tree-stand production (Urvas and Erviö 1974; Tamminen 1993; Chen et al. 1998; Yan et al. 2012), repeatedly N-fertilised sites have been temporarily moved to a higher level of productivity. In addition to N, the amounts of Ca, P and S in the organic layer were also somewhat higher on the N-fertilised plots, where the mass of the organic layer was increased due to fertilisation. This is in accordance with the results reported by Mälkönen et al. (1990). Decreased concentrations and amounts of K in the organic layer may indicate a dilution of K in tree and other plant tissues and litter due to N fertilisation, or it may be explained also by increased leaching of K due to liming.

In accordance with this study, Nohrstedt (1992) reported decreased concentrations of acid ammonium acetate-extractable P in the organic layer after long-term N fertilisation. In a Swedish experiment with Scots pine, however, repeated N addition, a total of 1800 kg ha<sup>-1</sup>, increased concentrations of ammonium acetate-extractable P in the organic layer (Ring et al. 2011). Repeated N fertilisation has not usually changed P concentrations in needles (Mälkönen et al. 1990; Jacobson and Nohrstedt 1993; Nilsen and Abrahamsen 2003; Jacobson and Pettersson 2010), although in some cases P concentrations have been increased (Jacobson and Pettersson 2010) or decreased (Mälkönen et al. 1990; Clarholm 1993).

The most common harmful side effects of N fertilisation are possible deficiency of B and growth disturbances due to the deficiency (Möller 1983; Jalkanen 1990; Jalkanen and Aalto 1993) and leaching of nitrate to ground water (Smolander et al. 1995; Johannisson et al. 1999; Ring et al. 2006; Bergh et al. 2008). A single normal dose of N fertilisation, 120 to 180 kg/ha will seldom cause significant leaching of nitrate or B deficiency; but repeated N fertilisation may, on the other hand, more likely cause leaching of nitrate and shortage of B and may also be seen in soil parameters, such as increased amounts of N and C and decreased C/N ratio.

In spite of the rather large addition of N, on average, 1100 kg ha<sup>-1</sup> or 22 kg ha<sup>-1</sup> a<sup>-1</sup> in about 50 years, N fertilisation did not lead to signs of N saturation, in which case the C/N ratio in the organic layer is low, nitrate leaches from the surface to the deeper soil layers and tree stands do not respond positively to N fertilisation (Matzner and Grosholz 1997; Aber et al. 1998; Tietema et al. 1998). Retention of deposition N or fertiliser N seems to be related to the N status of the forest ecosystem, which can be measured, e.g. by determining the C/N ratio of the organic layer (Tietema et al. 1998). This means that when there is a shortage of N and the C/N ratio is high, extra N coming from outside the ecosystem will be retained in the system and not leach out of it.

Our estimate of the relative surplus of N in the surface soil, 39%, corresponds to the N fertiliser recovery reported by Preston and Mead (1994) from a lodgepole pine stand. In their material, eight growing seasons after single fertilisation (100 kg ha<sup>-1</sup> of N with labelled isotopes) c. 40% of N from the fertiliser remained within the 0–30 cm soil layer.

In the present study the surplus P in the organic layer was small. Most of the fertilised P had probably been retained by the mineral soil (Cade-Menun et al. 2000) as well as by the tree stand. The surplus of Ca on the limed plots related to the added amounts of Ca, 21%, corresponded moderately to the figures of Derome et al. (1986), who estimated the Ca surpluses in the organic layer of pine and spruce stands to be 16 and 36%, respectively.

Long-term N fertilisation tended to acidify soil, although only weakly. The N fertiliser used most often, Finnish ammonium nitrate, contained some lime, on average, 0.7 Mg ha<sup>-1</sup> (0.33 to 1.18 Mg ha<sup>-1</sup>), which according to the pH equations in Table 5 would raise the pH<sub>CaCl2</sub> in the organic layer by 0.09 pH units (+0.05 to +0.16 units) and in the mineral soil by 0.08 units (0.04 to 0.13 units). However, according to these pH equations, containing independent variables N and lime doses, the N doses used would lower the pH<sub>CaCl2</sub> in the organic layer from 0.05 (N<sub>minimum</sub> = 534 kg ha<sup>-1</sup>) to 0.17 (N<sub>maximum</sub> = 1908 kg ha<sup>-1</sup>) and, on average, 0.10 pH units (N<sub>median</sub> = 1092 kg ha<sup>-1</sup>), and in the 0–10 cm mineral soil layer from 0.13 to 0.47 and, on average, 0.27 pH units. Average changes in pH<sub>CaCl2</sub> corresponded to increases in proton concentrations of 1.33\*10<sup>-4</sup> mol dm<sup>-3</sup> in the organic layer and 1.13\*10<sup>-4</sup> mol dm<sup>-3</sup> in the mineral soil. Our findings of increasing acidity in the mineral soil due to N fertilisation correspond to those of Mälkönen et al. (1990) and Högberg et al. (2006).

## 5 Conclusions

Nitrogen fertilisation over a period of 42 to 50 years led to larger amounts of organic matter, carbon and most nutrients and to a decrease in the C/N ratio in the surface soil, as was expected on the basis of earlier observations. The amounts of C and N were, on average, 30% and 56%, respectively, higher in the organic layer and 10% and 16%, respectively, higher in the 0–10 cm mineral soil layer on the N fertilised than on the unfertilised plots. A decrease in soil concentrations of K indicates a dilution of K in tree and plant tissues due to N fertilisation. But K has certainly not become a growth-limiting factor on upland sites even after the highest doses of N, i.e. 1908 kg/ha in c. 50 years or 37 kg ha<sup>-1</sup> a<sup>-1</sup>. Nitrogen fertilisation may decrease pH, but only slightly.

Phosphorus fertilisation increased the concentrations and amounts of total and AAA-extractable P in the organic layer and in the 0–10 cm mineral soil layer. However, very little P was retained in the surface soil. As expected, liming increased the pH and the concentrations and amounts of Ca. Liming seemed to decrease concentrations and/or amounts of AAA-extractable K, P and S.

Long-term N fertilisation led to more or less useful changes in the chemistry of the surface soil, i.e. improved fertility and accumulation of carbon; but changes due to P fertilisation and liming are more or less useless from the standpoint of forest production.

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