

1 Does nitrogen deposition increase forest production? The role of phosphorus

2
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6 7 Abstract

8 Effects of elevated N deposition on forest aboveground biomass were evaluated using long-
9 term data from N addition experiments and from forest observation plots in Switzerland. N
10 addition experiments with saplings were established both on calcareous and on acidic soils, in
11 3 plots with *Fagus sylvatica* and in 4 plots with *Picea abies*. The treatments were conducted
12 during 15 years and consisted of additions of dry NH_4NO_3 at rates of 0, 10, 20, 40, 80, and
13 $160 \text{ kg N ha}^{-1} \text{ yr}^{-1}$. The same tree species were observed in permanent forest observation plots
14 covering the timespan between 1984 and 2007, at modelled N deposition rates of 12-46
15 $\text{kg N ha}^{-1} \text{ yr}^{-1}$. Experimental N addition resulted in either no change or in a decreased shoot
16 growth and in a reduced phosphorus concentration in the foliage in all experimental plots. In
17 the forest, a decrease of foliar P concentration was observed between 1984 and 2007,
18 resulting in insufficient concentrations in 71% and 67% of the *Fagus* and *Picea* plots,
19 respectively, and in an increasing N:P ratio in *Fagus*. Stem increment decreased during the
20 observation period even if corrected for age. Forest observations suggest an increasing P
21 limitation in Swiss forests especially in *Fagus* which is accompanied by a growth decrease
22 whereas the N addition experiments support the hypothesis that elevated N deposition is an
23 important cause for this development.

24
25 **Capsule:** Low P levels limit growth in Swiss forests

26 **Keywords:** *Fagus sylvatica*, *Picea abies*, growth, nitrogen deposition, phosphorus, carbon
27 sequestration

28

29 **Introduction**

30 Anthropogenic nitrogen emission from combustion and agricultural activities has led to
31 eutrophication of forest ecosystems in large parts of the industrialized world (Nihlgård, 1985;
32 Aber et al., 1989; Galloway, 1995). As a consequence, previously N limited ecosystems are
33 getting saturated with nitrogen (Aber et al., 1998) and leaching nitrate (Dise and Wright,
34 1985; Gundersen et al., 1998). The release of N limitation stimulates growth at least initially;
35 an increase in foliar N is expected to increase photosynthesis (Reich et al., 1995). It has
36 therefore been postulated that nitrogen will increase carbon sink strength of forests by a
37 substantial amount (Townsend et al., 1996; IPCC, 2001; Reich et al., 2006). A larger forest
38 growth has indeed been observed in the second half of the last century which was attributed to
39 higher N availability (Spiecker et al., 1996). There are, however, observations that this growth
40 increase after N addition is not sustainable as other limitations will come into effect. The
41 newest IPCC report acknowledges these limitations (Nabuurs et al., 2007). Aber et al. (1989)
42 suggest that phosphorus is getting limiting to plant growth when N limitation is removed.
43 This concept of nutrient limitation would, however, explain the absence of further growth
44 stimulation but not a decrease below pristine situation as observed e.g. by Nellemann and
45 Thomsen (2001) and by Boxman et al. (1998) in Norway spruce, by Magill et al. (2004) in
46 pine and by McNulty et al. (2005) in red spruce.

47

48 A network of forest observation plots in Switzerland, with a gradient of modelled N
49 deposition ranging from 12 to 46 kg N ha⁻¹ yr⁻¹, allows to analyze N effects on growth and
50 nutrition by epidemiological methods. The mechanistic background for such statistical

51 analyses is given by complementary N addition experiments with saplings of *Fagus sylvatica*
52 and *Picea abies* in different regions. Both datasets will be presented here and the conclusions
53 compared.

54

55 **Materials and Methods**

56 *Permanent observation plots*

57 135 permanent observation plots are maintained in different regions of Switzerland, with a
58 wide range of altitudes, soil types, and pollution climates (Flückiger and Braun, 1998; Braun
59 et al., 1999). Each plot consists of 60-70 beech (*Fagus sylvatica* L.) and/or Norway spruce
60 trees (*Picea abies* Karst.) in pure or mixed stands. Nitrogen deposition for each plot was
61 modelled by Rihm (Meteotest) after the method described in Kurz et al. (1998).

62

63 Stem diameter was measured every 4th year at marked points at breast height. Surface area of
64 the plots was determined using a Leica GPS GS50 (Leica Geosystems, Heerbrugg,
65 Switzerland). The increment of base area of individual trees was either used for regression
66 analysis with foliar nutrient concentrations (eight trees per plot) or summed up over the whole
67 plot and divided by the plot area, if necessary corrected for the base area of other tree species
68 present in the plot.

69

70 Shoots for nutrient analysis were harvested every 4th year in July by helicopter from always
71 the same eight trees per plot, starting in 1984. The current shoot length was used for the
72 regression analysis. Beech leaves and one year old needles of Norway spruce were dried,
73 ground, and analyzed for nitrogen and phosphorus according to Walinga et al. (1995). A
74 subset of samples from previous harvests were reanalyzed each time to avoid systematic shifts
75 in the analytical results. Quality control was achieved by analysis of certified samples (NIST

76 apple leaves, National Institute of Standards and Technology, Gaithersburg, USA) and by
77 taking part in a sample exchange program (WEPAL, University of Wageningen).

78

79 For amino acid analysis, beech leaves and one year old spruce needles were frozen in liquid
80 nitrogen within one day of harvest and freeze dried. The amino acids were extracted
81 according to Vance and Zaerr (1990) and analyzed by HPLC (Waters 600E/715) using the
82 Waters AccQ TagTM kit. This includes a pre-column derivatisation with 6-aminoquinolyl-N-
83 hydroxysuccinimidylcarbamate, a reversed-phase gradient separation on a Nova-Pak C18
84 column (4 µm) and fluorescence detection (Waters 450, emission at 250, excitation at 395
85 nm).

86

87 Soil solution was collected in 18 plots below the rooting zone (usually 80 cm depth) at
88 monthly intervals, with 5 replicates per plot. Nitrate concentrations were determined by ion
89 chromatography (Dionex GP50, detection ED50 with suppressed conductivity) after filtration
90 through a 0.45 µm filter (Whatman OE67, cellulose acetate). A hydrological model was used
91 to calculate runoff amounts per collection period (Schulla and Jasper, 2007) which were
92 multiplied with the nitrate concentrations to obtain estimates of N leaching.

93

94 ***Nitrogen addition experiments***

95 N addition experiments were established in 7 plots, with varying altitudes and geology (Table
96 1). 3-year old saplings of beech (*Fagus sylvatica* L.) or Norway spruce (*Picea abies* Karst.)
97 from a local provenance were planted in 1992. They were arranged in a fully randomized
98 block system with 12 replicates per treatment. Dry NH₄NO₃ was applied at annual rates of 0,
99 10, 20, 40, 80 and 160 kg N ha⁻¹ between 1992 and 2006, split into 3 portions over the season.

100 Foliage for nutrient analysis was collected every 2-3 years in August and analyzed as

101 described above for the forest observation plots except that in Norway spruce current year
102 needles were used. The length of the leader shoot was measured each year in August.

103

104 Fine roots of beech for phosphatase measurements were collected in May and June
105 (Hochwald) and in July 2005 (Zugerberg), after 13 years of treatment. Phosphatase activity in
106 tips of about 1 cm length was measured within 4 days after collection according to Johnson et
107 al. (1999) at a pH of 5.0.

108

109 In the plot Hochwald, mycorrhiza were assessed using the in-growth bag method described by
110 Wallander et al. (2001). The bags with quartz sand (acid washed sand with 0.36-2 mm grain
111 size, nylon mesh size 50 μm) were exposed after 13 years of treatment and left in the field for
112 two years at a depth of 5 cm. After this time, mycelial growth was visually assessed. The
113 following score classes were assigned: 1= no mycelium, 2= little mycelium present, 3=
114 mycelium present, but no aggregation of sand particles, 4= high mycelium density and little
115 aggregation of sand particles, 5= high mycelium density and strong aggregation of sand
116 particles.

117

118 Soil solution was collected during 11 years on Zugerberg and 9 years in Lurengo. Six ceramic
119 suction cups per treatment were installed at a depth of 50 cm (Zugerberg) and 20 cm
120 (Lurengo), respectively. Leaching was calculated as described above for the forest
121 observation plots except that in Lurengo measurements could only be made during the
122 summer months (May-September) and annual loads had to be extrapolated.

123

124 **Statistics**

125 All statistical analyses were conducted using the software packages SYSTAT (SYSTAT
126 Richmond, USA, version 11) and S-PLUS (Insightful, Seattle, USA, version 7).

127

128 Data analysis was performed using a multivariate mixed regression (S-PLUS function lme;
129 Pinheiro and Bates, 2000) with plot and year as groups, including the confounding variables
130 altitude and age. Age was included as a constant for each observation plot (i.e. not changing
131 with time) as recommended for cohort studies (Glenn, 2007). For the graphs the confounding
132 factors were accounted for by correction of the data according to the regression results. The
133 relationship between foliar nutrient concentration and stem increment or shoot growth was
134 analyzed on an individual tree basis, with the six growth assessments included
135 simultaneously. This model included a separate random intercept for each combination of site
136 and year (example given below for stem increment):

$$137 \quad y = b_0 + b_{11} * \text{altitude} + b_{12} * \text{altitude}^2 + b_2 * \text{ndep} + b_{31} * \text{pconc} + b_{32} * \text{pconc}^2 +$$
$$138 \quad b_4 * \log(\text{age}) + c(\text{plot}, \text{year}) + e$$

139 where $c(\text{site}, \text{year})$ denotes the random intercept for the respective site in the respective year,
140 f_1 - f_4 are polynomial functions and e denotes the Gaussian error term for the respective tree.

141

142 To facilitate the graphical representation of the large dataset, the x-axis was divided into
143 classes and the mixed model repeated with the classified variables to obtain mean and
144 confidence intervals for each class. The residuals of the regression analyses were checked for
145 normality. Growth data were root transformed to ensure a normal distribution. In the case of
146 the amino acid concentrations, a log transformation was necessary.

147

148 Data evaluated in different years were also pooled the N addition experiments. For nutrients,
149 data were available from the 4th to the 10th, in some cases up to the 12th year of the experiment

150 (beech) and from the 4th up to the 9th experimental year, respectively (Norway spruce). In the
151 case of growth, measurements from the 6th up to the 13th-16th experimental year were
152 combined. The error probabilities for the regression with N addition and the confidence
153 intervals were calculated using a mixed regression with year as grouping variable.

154

155 The differences of the mycelium density score classes were assessed using an exact linear-by-
156 linear association test (StatXact for SYSTAT, version 1, Cytel Inc., Cambridge, USA). This
157 test is especially suited for small cell counts of score classes within ordered treatments.

158

159 **Results**

160 *Permanent observation plots*

161 *Relation between growth and nutrition*

162 Stem increment of *Fagus* and *Picea* was significantly related with foliar P concentrations and
163 N deposition (Figure 1, Table 2). In the case of stem increment of *Picea*, the N relations were
164 weak, the regression with N deposition being only significant in the multivariate analysis, not
165 in a univariate regression. This result may, however, be affected by an intercorrelation
166 between N deposition and altitude which was more serious in the spruce dataset than in beech.
167 Whereas stem increment was more related to N deposition, shoot growth responded more to
168 foliar N (Figure 2, Table 2). The relation with P was significant for both stem increment and
169 shoot growth in both species but linear only in the case of stem growth of Norway spruce. In
170 all other cases, a quadratic term was significant indicating a nonlinear response, suggesting a
171 saturation at higher concentrations.

172

173

174 *Amino acid concentrations*

175 From the amino acids analyzed, arginine concentrations showed the strongest regression with
176 nutrient concentrations and N deposition. Arginine in the foliage of both beech and Norway
177 spruce was positively correlated with N concentration and negatively with P (Table 3). In the
178 case of beech, the relationship with P was stronger than with N as indicated by the larger
179 standardized coefficient and the smaller p-values. N deposition was a significant predictor to
180 arginine in Norway spruce, not in beech. Neither altitude nor age were significant covariates.

181

182 *Nitrogen leaching*

183 The observation plots leached substantial amounts of nitrate from the soil as shown in Figure
184 3.

185

186 *Developments*

187 Between 1984 and 2007, a significant decrease in N and P foliage concentrations of mature
188 *Fagus sylvatica* was observed (Figure 4, Table 4). There was a confounding effect of age
189 which was removed by correlation analysis with stand age. The data in Figure 4 are corrected
190 for this. P decreases in beech accounted for 28.0% in total and for 25.0% after correction for
191 age. In *Picea abies*, the total P decrease was by 18.7%, of which 15.1% were attributed to
192 time. In the majority of both beech and Norway spruce plots, the average foliar P
193 concentration in 2007 was below 1 mg g⁻¹ d.m., the lower threshold for normal nutrition
194 (Stefan et al., 1997). The P decrease was paralleled by a weaker decrease of N. In beech, the
195 N:P ratio increased significantly as well whereas in Norway spruce it remained constant.
196 These changes in nutrition showed no correlation to soil factors and were similar on
197 calcareous soils and on soils with a low base saturation (data not shown).

198

199

200 Stem increment on a hectare basis decreased significantly between 1987 and 2002. Between

201 2002 and 2006 no further decrease was observed (Figure 5).

202

203

204 *Nitrogen addition experiments*

205 N addition caused a P decrease in all plots (Figure 6) whereas N concentrations increased

206 moderately (Figure 7). The P decrease was not a result of a dilution effect as no significant

207 growth stimulation was observed (Figure 8). In 3 out of 7 plots, there were even significant

208 growth inhibitions by N addition, with the strongest response in the beech plot Hochwald

209 where the highest treatment ($160 \text{ kg N ha}^{-1} \text{ yr}^{-1}$) caused a growth decrease by up to 58%. This

210 plot had the lowest P concentration in the leaves, with average values below the range for

211 normal nutrition according to Stefan et al. (1997) even in the controls. In some of the N

212 addition plots also other nutrients limited growth, such as K (Hochwald) and Mg (Zugerberg,

213 Lurengo) (data not shown).

214

215

216 *Phosphatase activity in fine roots*

217 Phosphatase activity in fine roots increased with increasing N addition both in Hochwald and

218 on Zugerberg although significant differences to the control were only observed at relatively

219 high N addition rates (Figure 9).

220

221 *Mycorrhiza*

222 Mycorrhizal density in in-growth bags decreased between 0 and 80 kg N ha⁻¹ yr⁻¹ (Figure 10),
223 with significant differences to the control at 40 and 80 kg N ha⁻¹ yr⁻¹.

224

225

226 *Nitrogen leaching*

227 A substantial amount of the added N was recovered in leaching water of the plots Zugerberg
228 and Lurengo (Figure 11). On an average of all examined treatments the estimated leaching
229 rates were 81% on Zugerberg and 78% on Lurengo. It has, however, to be mentioned that the
230 estimates in Lurengo are based on values for the summer months only whereas the Zugerberg
231 data base on whole year measurements.

232

233 **Discussion**234 *N status*

235 Both the observation plots and the experimental plots have to be characterized as saturated
236 with N. This is suggested by N leaching data which exceeds the 4-5 kg N ha⁻¹ yr⁻¹ set as limits
237 by the working group on Critical Loads for Nitrogen (UNECE, 1992) in more than half of the
238 18 examined forest observation plots and by the high proportion of added N leaching from the
239 experimental plots. Foliar N concentration in the forest was unrelated with N deposition in
240 both beech and Norway spruce (data not shown). This is in contradiction to the results of the
241 experiments where a slight increase of foliar N concentration was observed and to a number
242 of field observations (Aber et al., 1998; ICP Forests, 2003), but it adds also evidence to the
243 suggestion that the examined forest plots are saturated with N.

244

245 The missing positive growth response to N addition is also a sign of N saturation and is in
246 accordance with several long term studies on N response. Nellemann and Thomsen (2001
247 analyzed 31'606 increment cores of Norway spruce in different regions of Norway and found
248 that the initial growth stimulation in the high nitrogen areas was followed by a drop even
249 below the growth from the low nitrogen area. N addition resulted in a transient growth
250 stimulation of Norway spruce in the study by Högberg et al. (2006) or even in a depression as
251 shown for pine by Magill et al. (2004) and for red spruce by McNulty et al. (2005). In the
252 NITREX roof experiment in the Netherlands, where ambient nitrogen deposition was reduced
253 from 56 to 4 kg N ha⁻¹ yr⁻¹, trees in the roof clean condition grew better than in the control
254 with ambient deposition (Boxman et al., 1998).

255

256 *P status*

257 In view of the high N status of the plots, the lacking growth stimulation in the experiments is
258 not surprising. A negative growth response to N addition needs, however, additional
259 explanation, with the P status probably playing a crucial role. The experimental plot with the
260 lowest foliar P concentrations was the one with the strongest negative growth response to N
261 addition. P limitation seems to be also an issue in the forest observation plots. This is
262 suggested by the correlations between growth and foliar P concentrations as well as by the
263 weakness of the regressions between N deposition and stem increment of *Picea*. In *Fagus*
264 both stem increment and shoot-growth seem to be co-limited by N and by P.

265

266 The P concentrations in the foliage of mature trees in the observation plots, both beech and
267 Norway spruce, have to be considered as low in 2003 and 2007. Stefan et al. (1997) regard a
268 P concentration in the foliage of >1 mg g⁻¹ d.w. as minimum value for sufficient nutrition.
269 The relations between growth and P in the foliage support this limit as in 3 out of 4 cases the
270 regressions start to level off around 1 mg g⁻¹ d.w.. The base area increment of Norway spruce

271 increases more linearly with increasing P concentrations. In 2007, only 29% of the beech and
272 33% of the Norway spruce showed average P concentrations $>1 \text{ mg g}^{-1} \text{ d.w.}$ and can be
273 classified as well supplied with P. For the ratio between N and P different interpretations are
274 available. The ranges recommended by Stefan et al. (1997) (11-25 for beech, 6-17 for Norway
275 spruce) are very wide but just based on dividing the lower end of the normal nutrition range
276 for N by the higher end of the range for P and vice versa. Güsewell (2004) regards plants with
277 a N:P ratio of <10 generally as N limited, with ratios of >20 as P limited. Reich and Schoettle
278 (1988) suggest N:P ratios to be at optimum at values $<7-10$. Boyce et al. (2006) found a
279 colimitation of the photosynthesis of *Pinus aristata* and *Pinus longaeva* by N and P and
280 conclude that the critical N:P ratio is approximately 8.3 (P:N 0.12). In a compilation from
281 various sources, Flückiger and Braun (2003) deduced recommendations for normal ranges for
282 N:P ratios between 10 and 17 for beech and between 7 and 12 for Norway spruce. Thus, the
283 N:P ratios observed in mature beech in observation plots are beyond normal levels
284 irrespective which reference the interpretation is based on, whereas in Norway spruce the
285 evaluation is less clear but certainly not in the N limited region. The strong relationship
286 between arginine and P concentration in the foliage of both beech and Norway spruce adds
287 further evidence to the view that P limitation is widespread in the observation plots. In beech,
288 the foliar P concentrations were even better correlated with arginine than the N
289 concentrations. This is in accordance with the high N:P ratios suggesting strong P limitation.

290

291 ***Trend of nutrient concentrations***

292 A decline of foliar P concentration in mature forest trees was observed also in other European
293 regions (Bavaria: Prietzel et al., 2008, Baden-Württemberg: Hildebrand, pers comm.). The N
294 addition experiments suggest that increased nitrogen deposition may be one important reason
295 for this decline. This is in agreement with quite a number of studies (Mohren et al., 1986;
296 Aber et al., 1989) although Högberg et al. (2006) observed such a reduction only in the first

297 years of their fertilisation experiment of Norway spruce in Sweden. The foliar P
298 concentrations in the latter study were, however, much higher (1.7-1.8 mg g⁻¹ d.w.) than in the
299 data presented here. Apart from growth dilution effects which can be excluded in the present
300 study, the most plausible explanation for this phenomenon is an inhibition of mycorrhiza by
301 nitrogen (Wallander and Nylund, 1992; Erisman et al., 1998; Brandrud and Timmermann,
302 1998; Nilsson and Wallander, 2003). Mycorrhiza play a very important role in the P uptake
303 by forest trees (Andersson et al., 1996; Colpaert et al., 1999; Wallander, 2000). The decrease
304 in the extramatrical mycelium observed in the plot Hochwald is in accordance to this
305 hypothesis although the nonlinear response (re-increase above 80 kg N ha⁻¹ yr⁻¹) is difficult to
306 explain and does not fit with the P uptake response. The functional group of the mycelia in the
307 bags was not examined here but Wallander et al. (2001) came to the conclusion that the
308 invasion of saprophytic fungi to the mesh bags can be neglected. A possible explanation
309 would be a shift in the community of mycorrhizal species by increasing N as it has been
310 reported e.g. for arbuscular mycorrhizal fungi by Egerton-Warburton and Allen (2000). A
311 chemical fixation of P with Al in the roots (McCormick and Borden, 1972) as a result of soil
312 acidification is also possible in acidic soils, but not in the calcareous plot Hochwald. Bääth
313 and Söderström (1979) propose a microbial fixation of P as a result of N load. Citrate
314 extractable P in the soil of Hochwald and Lurengo was not affected by the N treatment (data
315 not shown), suggesting that rather uptake than availability in the soil is impaired by N. The
316 slight increase in phosphatase activity of fine roots in Hochwald may be interpreted as a
317 compensatory response for P deficiency and is in accordance to the results of Johnson et al.
318 (1999) who found that the activity of root-surface phosphomonoesterase (PME) in *Plantago*
319 *lanceolata* and *Agrostis capillaris* was increased after transplantation into soil with previous
320 long-term N addition (7 yr).

321

322 Foliar N concentrations in the forest observation plots decreased almost in parallel to the P
323 concentrations. Possible reasons could be (a) a reduction in N deposition, (b) a decreased N
324 uptake as a consequence of P deficiency or (c) a reduced uptake of P and N in parallel.
325 Indeed, NO₂ in air has declined during the observation period by about 50% in rural regions,
326 but the concentrations of NO₃⁻ and NH₄⁺ in rainwater have not (EKL, 2004), and NH₃
327 concentrations in the air have not changed since 2000 (monitoring programme according to
328 Thöni et al., 2004). Overall, a slight reduction in N deposition can be assumed during the
329 observation time. But in view of the weak response of foliar N concentrations to N addition
330 and of the missing correlation between N deposition and foliar N concentrations it is
331 suggested that hypothesis (a) is unlikely. Hypothesis (b) is supported by the findings of
332 Stevens et al. (1993) who were able to improve foliar N concentrations and reduce nitrate
333 leaching in a Welsh Sitka spruce stand with P and K addition. It is, however, in contrast to the
334 results from a fertilization experiment with mature Norway spruce in the Swiss Alps where
335 addition of a slow release fertilizer containing all macroelements increased only the P
336 concentrations in the needles, not N (Flückiger and Braun, 1995). While hypothesis (b) cannot
337 be excluded, the present data suggest rather an impaired uptake for both P and N in parallel
338 (hypothesis c) which may be a consequence of decreased mycorrhizal uptake efficiency
339 (Hatch, 1937). Mycorrhizal species differ quite substantially in their activity of enzymes such
340 as phosphatase (Courty et al., 2005)), and a shift in the species composition by N as
341 suggested by Wallenda and Kottke (1998) may therefore result also in a changed activity.

342

343 ***Growth trend***

344 The decrease in P concentration in the foliage may be one cause for the observed decrease in
345 stem increment. From the average P concentrations in 1984 and 2007 and the calculated
346 regressions between base area increment and P concentration, the expected growth decrease in

347 beech amounts to 6.8% and in Norway spruce to 6.5%. The observed growth decrease on an
348 individual tree basis in the same time period was 7.4% in beech. In Norway spruce, the
349 interannual variation of P and of growth was higher and the time series in growth was shorter,
350 not allowing a direct comparison, but the observed average growth reduction (10.2% between
351 1987 and 2006) is compatible with this hypothesis. Because of the lacking relationship
352 between stem increment and foliar N concentration it is unlikely that the N decrease has
353 played a role. Prietzel et al. (2008) come to a similar conclusion in two Scots pine stands in
354 Central Europe: foliar P concentrations explained a large proportion of the annual increment
355 of merchantable wood, in contrast to foliar N concentrations.

356

357 **Conclusions**

358 The observation plots leach nitrogen in considerable amounts, suggesting saturation with N.
359 The low P concentrations ($<1 \text{ mg g}^{-1} \text{ d.w.}$) in the foliage of mature forest trees, the high N:P
360 ratios especially in beech, the relation between P and arginine concentration and the observed
361 relationship between foliar P concentration and stem increment suggest P limitation in the
362 forest trees. The decrease of P concentrations after N addition in the experiments indicates
363 that N deposition may have played an important role for inducing P limitation, with altered
364 uptake being the more likely reason than P deficiency in the soil. The results suggest that
365 phosphorus limitation is an important issue in Swiss forests and is affecting today's forest
366 growth. This has to be considered in growth models aiming at predicting carbon uptake rate,
367 biofuel production or soil acidification.

368

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379

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530

531 **Figure legends**

532 Figure 1: Relationship between base area increment of *Fagus* (left) and *Picea* (right) with N
533 deposition and P concentration in the foliage (d.m.=dry matter). Bars=confidence intervals for
534 the classified independent variable. Black line: fit to the whole dataset (all variables
535 continuous). For statistics see Table 2.

536 Figure 2: Relationship between shoot growth of *Fagus* (left) and *Picea* (right) with N and P
537 concentration in the foliage. Bars=confidence intervals for the classified independent variable.
538 Black line: fit to the whole dataset (all variables continuous). For statistics see Table 2.

539 Figure 3: N leaching in 18 observation plots. Average of 3-5 annual rates between 1998 and
540 2002.

541 Figure 4: Development of the foliar concentrations of N (left), P (middle), and of N:P ratio
542 (right) of mature beech and Norway spruce in forest observation plots. The data analysis was
543 restricted to plots which had data back to at least 1987. Dashed lines: lower limit of normal
544 range (upper line in the graphs of N and N:P for beech, lower line for Norway spruce; for P
545 the limits are the same for both species). Values are corrected for age trend. Bars = 95%
546 confidence intervals of plot medians. For statistics refer to Table 4.

547 Figure 5: Development of base area increment in 51 forest observation plots. The data are
548 corrected for age trend. Data from beech and Norway spruce stands were combined. Bars =
549 95% confidence interval.

550 Figure 6: P concentration in the foliage of beech and Norway spruce in relation to N addition.
551 Bars: 95% confidence intervals. Significant differences to the control are indicated with filled
552 symbols, significant linear relations to N treatment with p values in the upper right corner.
553 Dashed lines: lower and upper limit for the range of normal nutrition according to Stefan et al.
554 (1997).

555 Figure 7: N concentration in the foliage of beech and Norway spruce in relation to N addition.
556 Bars: 95% confidence intervals. For explanation of symbols refer to Figure 6.

557 Figure 8: Response of shoot growth (relative to control) in beech and Norway spruce to
558 increasing N addition. Bars: 95% confidence intervals. Dashed line: 100% (control).

559 Figure 9: Phosphatase activity in fine roots of beech in the N addition plots Hochwald (May
560 and June) and Zugerberg (July). SYSTAT boxplots, with median as horizontal line, the box
561 extending from the 25% to 75% percentile (for definition of whiskers refer to SYSTAT
562 manual). An overall significant linear trend is indicated with the p-values in the upper right
563 corner. Significant differences to the control are indicated by * $p < 0.05$, ** $p < 0.01$.

564 Figure 10: Score classes of mycelium density in mycorrhizal in-growth bags after two years
565 of exposure in the N experimental plot Hochwald extending from 1= no mycelium to 5= high
566 mycelium density. Differences to the control are significant at * $p < 0.05$, ** $p < 0.01$.

567 Figure 11: Annual N leaching rates in two of the N addition experimental plots. Bars:
568 standard error between different years (Lurengo 9 years, Zugerberg 11 years of data).

569

570 **Tables**

571 Table 1: Description of the sites used in the N fertilization experiments in afforestation plots.

Plot	Species	Altitude (m asl)	Soil type (FAO, 1988)	Geology	N deposition on	pH (CaCl ₂) ²⁾	Base saturation (%) ²⁾	C/N ³⁾	Annual rainfall (mm) ⁴⁾
Axalp	spruce	1700	Rendzic Leptosol	limestone	13.8	6.8	100	18.5	1710
Hochwald	beech	670	Rendzic Leptosol	limestone	15.3	6.9	100	21.1	1260
Lurengo	spruce	1600	Ferralic Cambisol	gneiss	11.5	4.1	13	23.3	2020
Möhlin	beech	290	Haplic Acrisol	gravel	13.7	3.8	12	20.1	1060
Rötiboden	spruce	1580	Cambic Podzol	granite	12.1	3.3	26	25.4	1850
Wengernalp	spruce	1880	Cambic Podzol	limestone	9.2	3.7	23	21.3	1970
Zugerberg	beech	1000	Dystric Cambisol	till	19.6	4	12	18.5	2010

572 ¹⁾ Modelled N deposition, calculated for open field (kg N ha⁻¹ yr⁻¹)573 ²⁾ Calculated as average in 0-40 cm soil depth574 ³⁾ In uppermost organic layer (Oh or Ah)575 ⁴⁾ Calculated from data published by Landeshydrologie (1992)

576

577 Table 2: p-values for the regressions in Figure 1 and Figure 2 (multivariate analysis, including polynomial

578 functions for altitude and P concentration).

	<i>Fagus sylvatica</i>		<i>Picea abies</i>	
	base area increment	shoot growth	base area increment	shoot growth
altitude	0.001	n.s.	0.002	0.001
(altitude) ²	<0.001.	n.s.	n.s.	n.s.
log(age)	n.s.	<.001	n.s.	<.001
N deposition	<.001	n.s.	0.029	n.s.
N concentration	n.s.	<.001	n.s.	<.001
P concentration	0.008	0.004	<.001	<.001
(P concentration) ²	0.027	0.002	n.s.	<.001
number of observations	2296	3464	1253	1835

579

580 Table 3: Arginine concentration (\log_{10} mmol g^{-1} d.m.) in beech leaves and one-year old needles of Norway
 581 spruce in relation to nutrient concentrations. Altitude and age were not significant. Standardized coefficient: all
 582 variables standardized to mean 0 and standard deviation 1 prior to regression analysis.

	<i>Fagus sylvatica</i>				<i>Picea abies</i>			
	coefficient	std. error	p-value	standardized coefficient	coefficient	std. error	p-value	standardized coefficient
(Intercept)	3.321	0.551	<.0001	0	2.320	0.372	<.0001	-0.006
N deposition	n.s.				0.018	0.006	0.0059	0.298
foliar N concentration	0.0709	0.0297	0.0288	0.379	0.146	0.032	0.0001	0.471
foliar P concentration	-2.0472	0.3936	0.0001	-0.826	-1.0043	0.1858	<.0001	-0.562

583

584

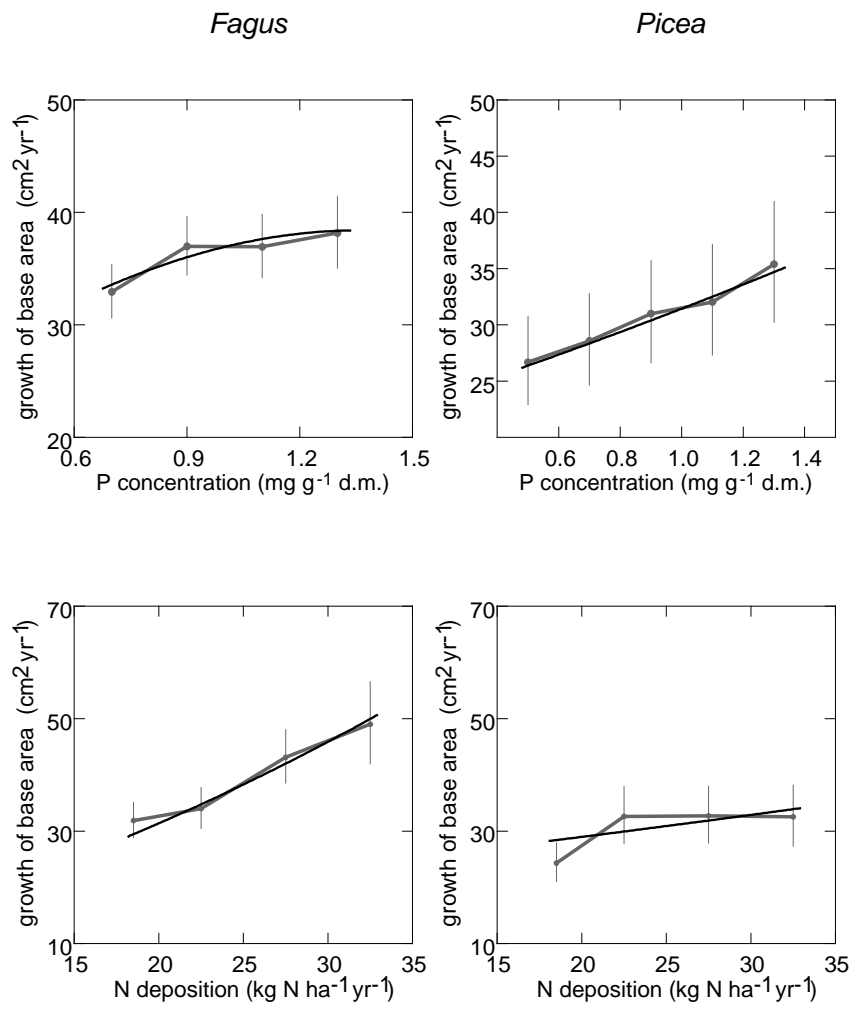
585 Table 4: p-values for the regressions in Figure 4 (multivariate analysis with age as covariate).

	<i>Fagus sylvatica</i>			<i>Picea abies</i>		
	nitrogen	phosphorus	N:P	nitrogen	phosphorus	N:P
year	<0.001	<0.001	<0.001	<0.001	0.024	0.054.
age	<0.001	<0.001	n.s.	<0.001	0.003	n.s.
number of plots						
1984	51			15		
1987-2007	61-64			27-33		

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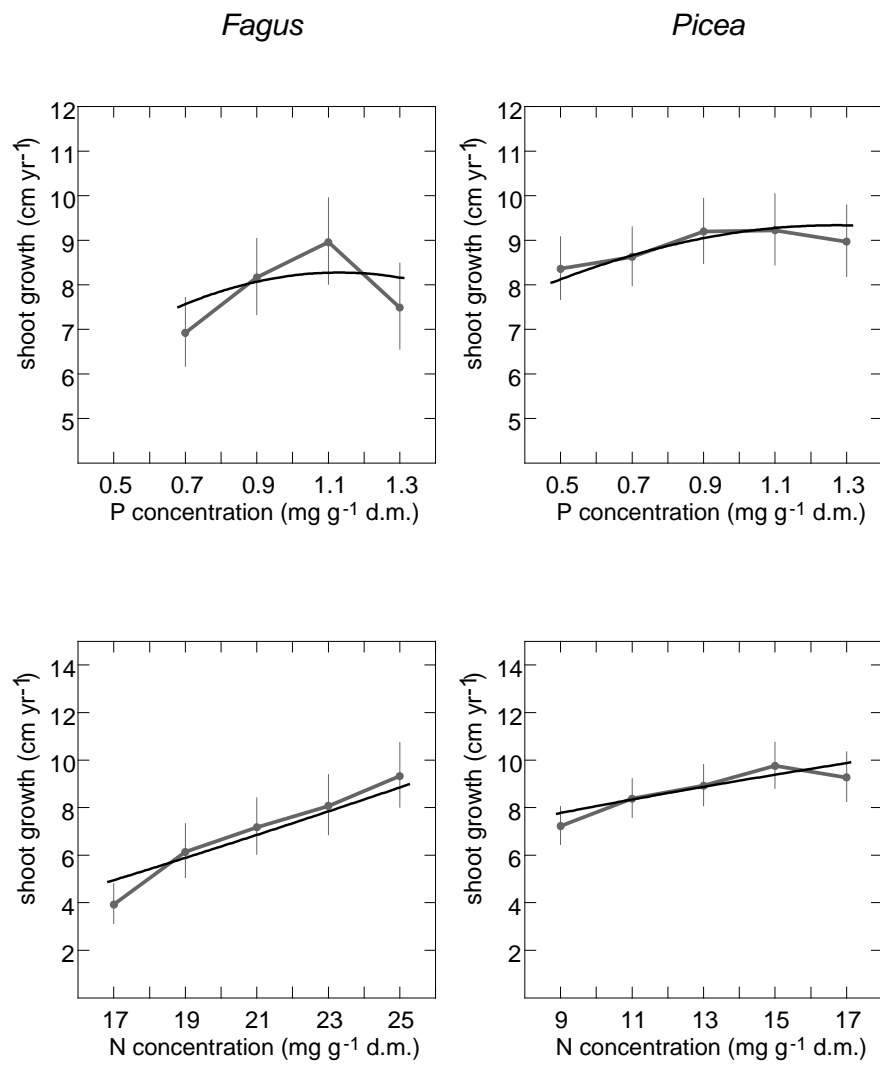
588 **Figures**



589

590 Figure 1

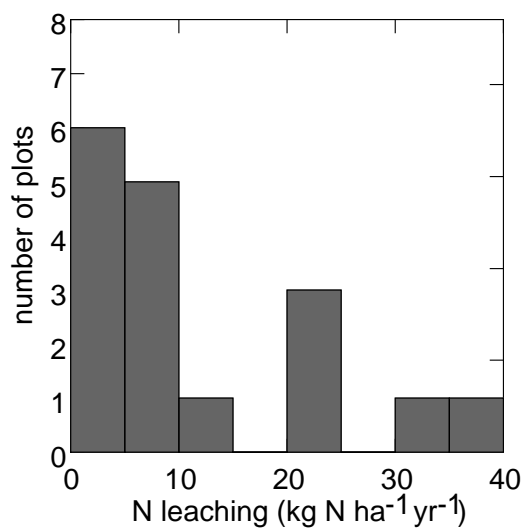
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593 Figure 2

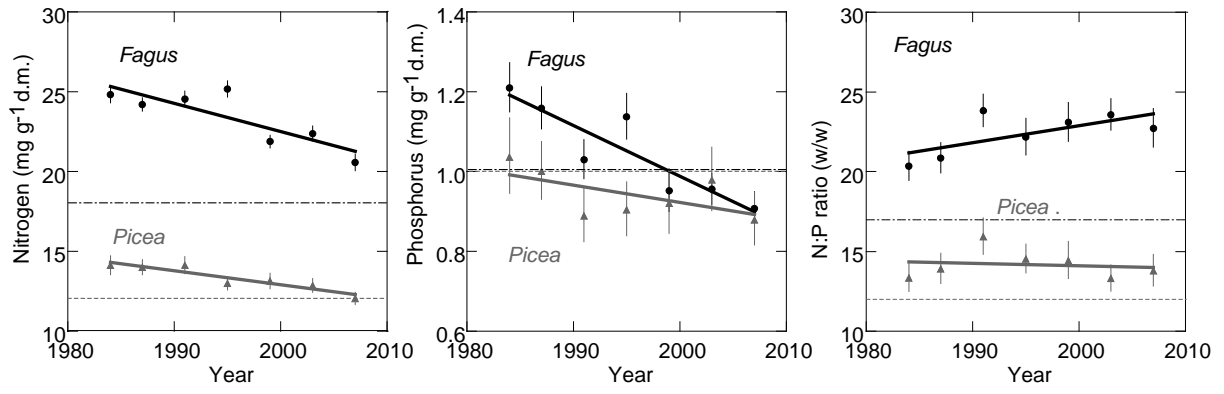
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596 Figure 3

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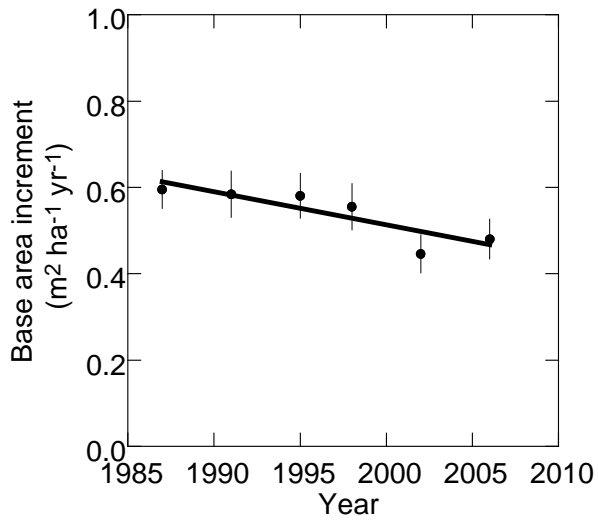


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599 Figure 4

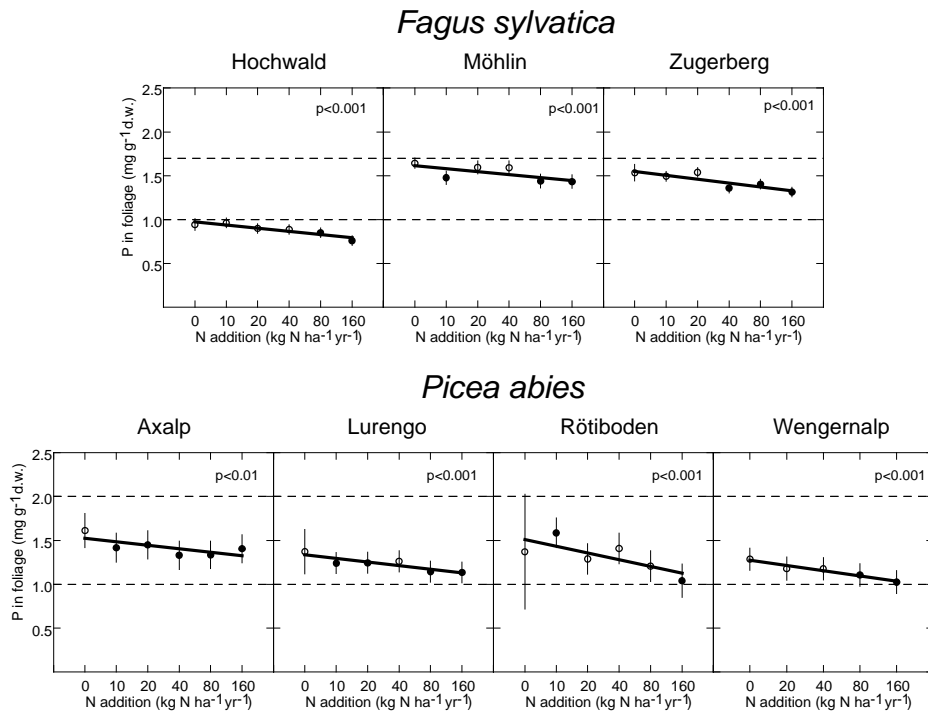
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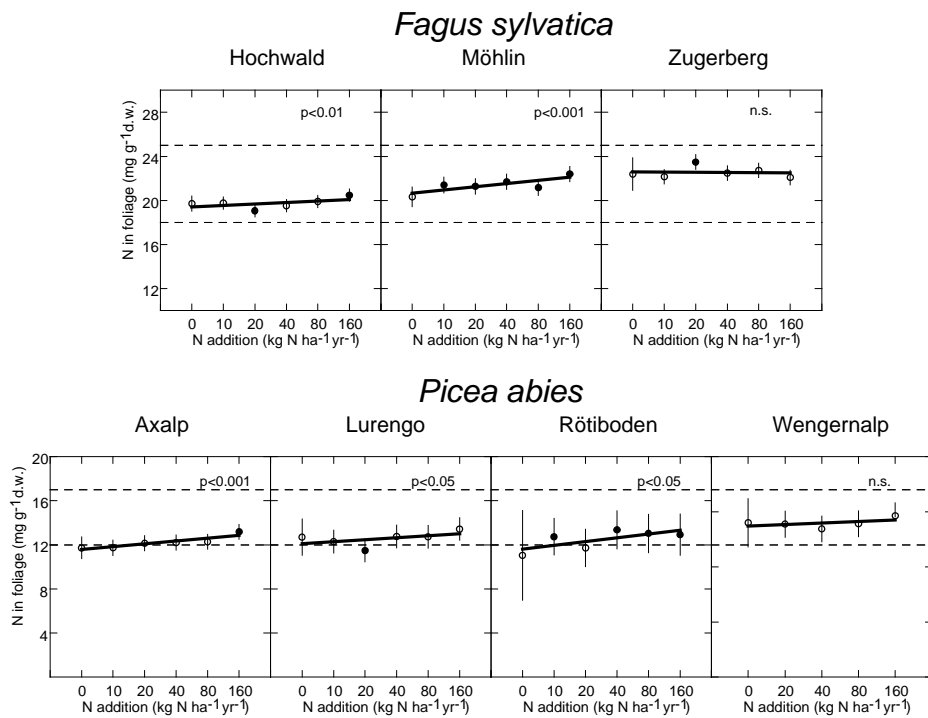
603 Figure 5



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605 Figure 6

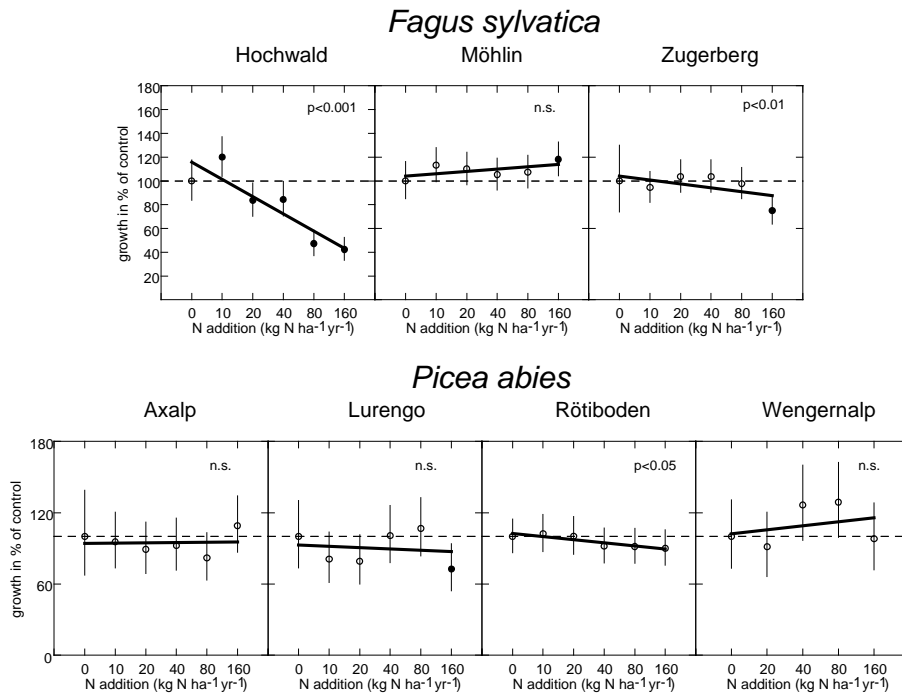
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608 Figure 7

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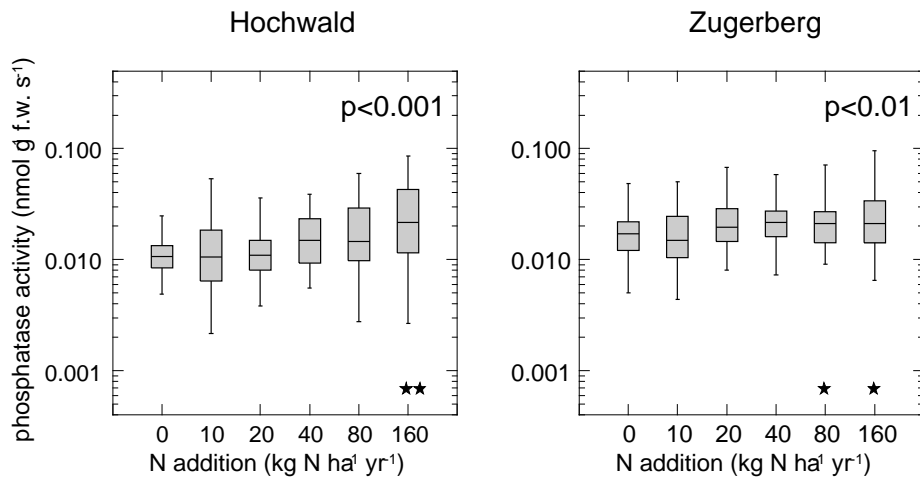


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611 Figure 8

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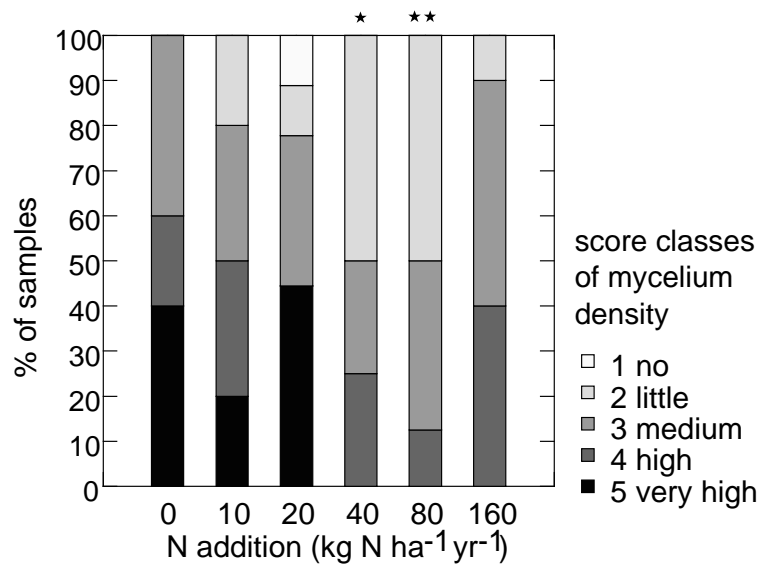
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615 Figure 9

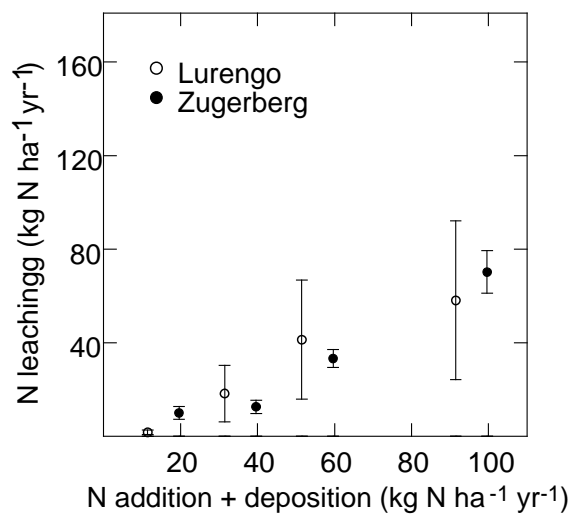
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618 Figure 10

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621 Figure 11

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