DOI	https://doi.org/10.1007/s10533-023-01082-3
CITATION	Verstraeten, A., Bruffaerts, N., Cristofolini, F. et al. Effects of tree pollen on throughfall element fluxes in European forests. Biogeochemistry 165, 311–325 (2023). https://doi.org/10.1007/s10533-023-01082-3

Effects of tree pollen on throughfall element fluxes in European forests

Arne Verstraeten[1], Nicolas Bruffaerts[2], Fabiana Cristofolini[3], Elena Vanguelova[4], Johan Neirynck[1], Gerrit Genouw[1], Bruno De Vos[1], Peter Waldner[5], Anne Thimonier[5], Anita Nussbaumer[5], Mathias Neumann[6], Sue Benham[4], Pasi Rautio[7], Liisa Ukonmaanaho[8], Päivi Merilä[8], Antti-Jussi Lindroos[8], Annika Saarto[9], Jukka Reiniharju[9], Nicholas Clarke[10], Volkmar Timmermann[10], Manuel Nicolas[11], Maria Schmitt[5], Katrin Meusburger[5], Anna Kowalska[12], Idalia Kasprzyk[13], Katarzyna Kluska[13], Łukasz Grewling[14], Małgorzata Malkiewicz[15], Lars Vesterdal[16], Morten Ingerslev[16], Miklós Manninger[17], Donát Magyar[18], Hugues Titeux[19], Gunilla Pihl Karlsson[20], Regula Gehrig[21], Sandy Adriaenssens[22], Agneta Ekebom[23], Åslög Dahl[24], Marco Ferretti[5], Elena Gottardini[3]

[1]Research Institute for Nature and Forest (INBO), Geraardsbergen, Belgium

[2]Mycology and Aerobiology, Sciensano, Brussels, Belgium

[3]Research and Innovation Centre, Fondazione Edmund Mach (FEM), San Michele all'Adige, Italy

[4]Forest Research, Alice Holt Lodge, Farnham, Surrey, UK

[5]WSL, Swiss Federal Institute for Forest, Snow and Landscape Research, Birmensdorf, Switzerland

[6]Institute of Silviculture, Department of Forest and Soil Sciences, University of Natural Resources and Life Sciences, Vienna, Austria

[7]Natural Resources Institute Finland (LUKE), Rovaniemi, Finland

[8]Natural Resources Institute Finland (LUKE), Helsinki, Finland

[9]The Biodiversity Unit of the University of Turku, Turku, Finland

[10]Norwegian Institute of Bioeconomy Research (NIBIO), Ås, Norway

[11]Office national des forêts, Fontainebleau, France

[12]Forest Research Institute, Sękocin Stary, Poland

[13]University of Rzeszów, Rzeszów, Poland

[14]Laboratory of Aerobiology, Department of Systematic and Environmental Botany, Faculty of Biology, Adam Mickiewicz University, Poznan, Poland

[15]Institute of Geological Sciences, University of Wroclaw, Wroclaw, Poland

[16]*University of Copenhagen, Frederiksberg, Denmark*
[17]*University of Sopron, Forest Research Institute, Sárvár, Hungary*
[18]*National Public Health Center, Budapest, Hungary*
[19]*Université catholique de Louvain, Louvain-la-Neuve, Belgium*
[20]*Swedish Environmental Research Institute IVL, Stockholm, Sweden*
[21]*Federal Office of Meteorology and Climatology MeteoSwiss, Switzerland*
[22]*PCS Ornamental Plant Research, Belgium*
[23]*Swedish Museum of Natural History, Stockholm, Sweden*
[24]*Department of Biological and Environmental Sciences, University of Gothenburg, Sweden*

Corresponding author: Arne Verstraeten, ORCID: 0000-0002-8813-1261, tel: +32 479 99 01 52, arne.verstraeten@inbo.be

Abstract

The effects of tree pollen on precipitation chemistry are not fully understood and this can lead to misinterpretations of element deposition in European forests. We investigated the relationship between forest throughfall (TF) element fluxes and the Seasonal Pollen Integral (SPIn) using linear mixed-effects modelling. TF was measured in 1990–2018 during the main pollen season (MPS, arbitrary two months) in 61 managed, mostly pure, even-aged *Fagus*, *Quercus*, *Pinus*, and *Picea* stands which are part of the ICP Forests Level II network. The SPIn for the dominant tree genus was observed at 56 aerobiological monitoring stations in nearby cities. The net contribution of pollen was estimated as the TF flux in the MPS minus the fluxes in the preceding and succeeding months. In stands of *Fagus* and *Picea*, two genera that do not form large amounts of flowers every year, TF fluxes of potassium (K^+), ammonium-nitrogen (NH_4^+-N), dissolved organic carbon (DOC), and dissolved organic nitrogen (DON) showed a positive relationship with SPIn. However – for *Fagus* – a negative relationship was found between TF nitrate-nitrogen (NO_3^--N) fluxes and SPIn. For *Quercus* and *Pinus*, two genera producing many flowers each year, SPIn displayed limited variability and no clear association with TF element fluxes. Overall, pollen contributed on average 4.1–10.6 % of the annual TF fluxes of K^+ >

DOC > DON > NH_4^+-N with the highest contribution in *Quercus* > *Fagus* > *Pinus* > *Picea* stands. Tree pollen appears to affect TF inorganic nitrogen fluxes both qualitatively and quantitatively, acting as a source of NH_4^+-N and a sink of NO_3^--N. Pollen appears to play a more complex role in nutrient cycling than previously thought.

Keywords

Throughfall
Airborne pollen concentrations
Nitrogen
Potassium
Dissolved organic carbon
ICP Forests

Introduction

The current understanding of canopy exchange processes, and particularly microbiological interactions affecting the chemical composition of precipitation, is still limited as for example indicated by recent evidence for nitrifying micro-organisms in the phyllosphere affecting throughfall (TF) inorganic nitrogen (N) fluxes (Ulrich 1983; Lindberg et al. 1986; Guerrieri et al. 2015, 2021). Temperate and boreal forests are dominated by anemophilous tree species that flourish and release pollen in the air mainly during spring (Faegri and van der Pijl 1979; Tormo-Molina et al. 1996). Some tree genera with anemophilous pollination strategy, including *Pinus* and *Quercus*, produce vast amounts of flowers and pollen each year, while *Fagus*, *Betula*, and *Picea* switch between years with few and abundant inflorescences (Kelly and Sork 2002; Geburek et al. 2012; Grewling et al. 2012; Lebourgeois et al. 2018; Nussbaumer et al. 2020). Only a tiny fraction of the emitted pollen is effectively involved in pollination, and the majority is washed out from the air by precipitation and dry deposition (Kluska et al. 2020). When pollen rehydrates, either on the pistil of a flower or in the TF, it becomes biochemically active and releases a mixture of organic substances, including lipids and functional proteins from the so-called pollen coat (Rejón et al. 2016). During *in vitro* leaching experiments, rehydrating pollen from temperate tree species discharged mainly the elements carbon (C), nitrogen (N), potassium (K), and phosphorus (P) up to one fifth of the initial pollen biomass (Doskey and Ugoagwu 1989; Rösel et al. 2012). Studies purely based on TF monitoring data reported peaks in the fluxes of the aforementioned elements during spring but didn't separate the effects induced by pollen from those controlled by other factors, such as passive leaching, insect infestations, and microbial activities on forest canopies (Carlisle et al. 1966; Draaijers et al. 1996; Ferm 1997; Le Mellec et al. 2010; Pitman et al. 2010; Verstraeten et al. 2016).

A possible way to disentangle the effects of pollen on TF chemistry is to evaluate TF element fluxes in combination with local pollen concentrations in the air. Both types of data are available from national/regional or international monitoring networks. TF deposition is monitored at the pan-European scale by the International Co-operative Programme on Assessment and Monitoring of Air Pollution Effects on Forests (ICP Forests, see http://icp-forests.net) under the UNECE Air Convention, and in parallel by several national networks since the early 1990s. TF is sampled continuously in permanent intensive monitoring (Level II) plots and analysis is carried out by the individual regions/countries on a weekly to monthly basis, according to harmonized methods (Clarke et al. 2022). TF sampling is traditionally performed with bulk collectors, which are open-funnel systems equipped with mesh filters, typically sized 0.25–1 mm^2 to keep out larger particles such as litter and insects, without obstructing the water flow. Pollen grains, however, are smaller (generally between 20 and 80 µm, but up to 150 µm for *Picea*), allowing them to pass through these filters and enter the TF collectors. Airborne pollen concentrations from various plants, including several common and widely distributed tree genera (e.g., *Betula*, *Alnus*, *Quercus*), are monitored daily in densely populated areas across the globe because of their allergenicity (Buters et al. 2018; Biedermann et al. 2019). The Seasonal Pollen Integral (SPIn), defined as the sum of the observed daily mean airborne pollen concentration during the main pollen season, is a standard measure for quantifying and comparing the yearly airborne pollen abundances (Galán et al. 2017).

In the present study, we used TF monitoring data from the past three decades collected in managed, mostly pure, even-aged stands of *Fagus sylvatica*, *Quercus* sp., *Pinus* sp. and *Picea abies* across

Europe, to assess the impact of tree pollen on TF element fluxes. We focused on K^+, NH_4^+-N, NO_3^--N, dissolved organic carbon (DOC), and dissolved organic nitrogen (DON) because these compounds are known or suspected to be influenced by pollen amount and activity (as stated above), and their concentrations in TF are available for most regions and countries. We also examined the relationship between TF element fluxes and airborne pollen concentrations (SPIn) of the dominant tree species observed at aerobiological monitoring stations located in the surroundings of Level II plots using linear mixed-effects modelling (LME). We hypothesize that there is a significant relationship between airborne pollen concentrations and TF element fluxes of the aforementioned compounds.

Materials and Methods

Intensive forest monitoring (Level II) plots

A total of 61 forest monitoring plots located in 12 European countries were selected for this study based on the dominant tree species at the site, tree age and the availability of data from nearby aerobiological monitoring stations (as stated below). The permanent monitoring plots are part of the UNECE ICP Forests Level II network and the Swedish Throughfall Monitoring Network (SWETHRO). Sites are geographically distributed from 43.2° N to 69.6° N and 0.9° W to 29.3° E with an altitudinal range of 0 to 1900 m above sea level, thus covering temperate, boreal, continental, Alpine, and Mediterranean climate zones (Fig. 1, Table S1). Plots were originally installed in mature, managed, mostly pure, even-aged forest stands of European beech (*Fagus sylvatica* L.), oak (*Quercus robur* L., *Q. petraea* (Matt.) Liebl., *Q. cerris* L.), pine (*Pinus sylve*stris L., *P. mugo* Turra, *P. cembra* L., *P. nigra* Arnold subsp. *laricio* Maire) and Norway spruce (*Picea abies* (L.) Karst.). The terms pure and even-aged refer to the dominant tree layer, disregarding any scattered dominant individuals and suppressed ingrowth of secondary tree species. For statistical analyses, plots were aggregated into four groups at tree genus level, i.e. *Fagus* (n = 22), *Quercus* (n = 12), *Pinus* (n = 10) and *Picea* (n = 17) (Fig. S1).

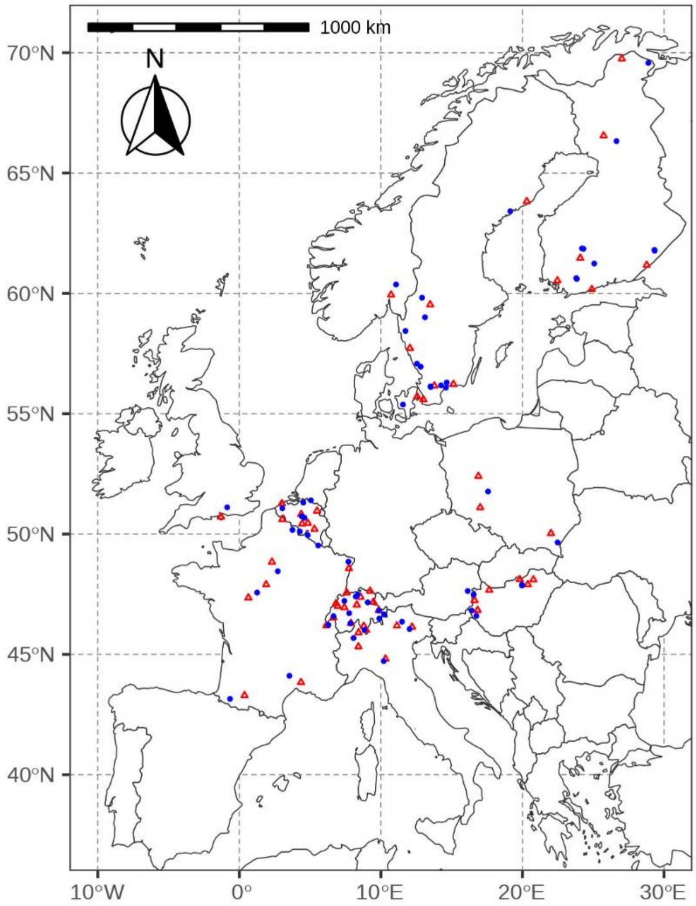


Fig. 1 Locations of the 61 selected Level II plots (blue dots) based on ICP Forests data and 56 aerobiological monitoring stations (red triangles)

Throughfall fluxes

The temporal range of available TF deposition fluxes measured in selected Level II plots varied in extent from 6 to 29 years, covering the period 1990–2018 (Table S1). For each plot, the longest possible time series was used in the calculations and statistical analyses, including only those years for which corresponding data on airborne pollen concentrations from nearby stations for the dominant tree genus were available (see below). After selection, a total of 1091 plot-year combinations (annual TF fluxes) was included in the analysis. TF sampling and chemical analyses were carried out by the countries/regions according to harmonized methods following the ICP Forests Manual Part XIV (Clarke et al. 2022). Quality assurance in the programme is ascertained through regular revision of the procedures for sampling and analysis by the Expert Panels, ring tests for laboratories and data validation procedures as described in the ICP Forests Manual Part III (Ferretti et al. 2021).

Airborne pollen concentrations

A total of 56 aerobiological monitoring stations from regional and national monitoring networks were selected for this study using their distance to the Level II plots as the main criterion. Trees of the same species show synchronicity of masting, i.e. high pollen and seed production, across relatively large areas (Ranta et al. 2008; Nussbaumer et al. 2016). This may explain why in rather flat areas like the Swiss plateau temporal patterns of the main pollen season can be very similar at distances of 50 km and still well comparable at distances of 150–200 km, while in mountainous areas the correlation often is much lower at these distances (Gehrig 2019). To ensure a certain level of representativeness, we applied an upper limit of 100 km distance in flat areas and 75 km in mountainous areas (the mean distance being 54.2 km) regardless of the prevailing wind direction (Fig. 1, Table S2). We used data on the SPIn, being the sum of all average daily pollen air concentrations over a single season (pollen grains * day * m^{-3}) for the respective tree genera (Galán et al. 2017). For each Level II plot, mean annual values of SPIn for the dominant genus were calculated using data from the nearest aerobiological monitoring stations, with a maximum of four stations. For each Level II plot also the main pollen season (MPS) for the dominant genus was determined. In the context of this study, MPS was defined as the average period covering the annual peak of airborne pollen concentrations (arbitrary two months, i.e. from one month before to one month after the pollen peak day; semi-monthly time step starting on the 1st or the 15th day of the month), based on data with daily resolution available for a limited number of aerobiological monitoring stations (Table 1). If data on a daily level were not available for a site, the MPS was determined as the best possible estimate considering its location (latitude, longitude, altitude) and the MPS for nearby plots with the same dominant tree genus.

Table 1. Number of Level II plots (n) with different timing of the MPS as derived from the days when maximum airborne pollen concentrations were registered.

Tree genus	April	May	June	July

<table>
<tr><td rowspan="2">*Fagus*
(n = 22)</td><td colspan="4">17</td><td></td><td></td><td></td><td></td></tr>
<tr><td></td><td colspan="4">5</td><td></td><td></td><td></td></tr>
<tr><td rowspan="2">*Quercus*
(n = 12)</td><td colspan="4">10</td><td></td><td></td><td></td><td></td></tr>
<tr><td></td><td colspan="4">2</td><td></td><td></td><td></td></tr>
<tr><td rowspan="4">*Pinus*
(n = 10)</td><td></td><td colspan="4">1</td><td></td><td></td><td></td></tr>
<tr><td></td><td></td><td colspan="4">5</td><td></td><td></td></tr>
<tr><td></td><td></td><td></td><td colspan="4">3</td><td></td></tr>
<tr><td></td><td></td><td></td><td></td><td colspan="4">1</td></tr>
<tr><td rowspan="4">*Picea*
(n = 17)</td><td></td><td colspan="4">2</td><td></td><td></td><td></td></tr>
<tr><td></td><td></td><td colspan="4">8</td><td></td><td></td></tr>
<tr><td></td><td></td><td></td><td colspan="4">5</td><td></td></tr>
<tr><td></td><td></td><td></td><td></td><td colspan="4">2</td></tr>
</table>

Calculations and statistical analysis

The production of figures and statistical analysis were carried out in RStudio v2022.07.2 with the R software version 4.2.1 (RStudio Team 2022, R Core Team 2022). Linear mixed-effects models (LME) for the TF fluxes of K^+, nitrate-nitrogen (NO_3^--N), ammonium-nitrogen (NH_4^+-N), DOC, and DON during the MPS (Table 1) as a function of SPIn were evaluated for the four genera using the R package ‘lme4’ (Bates et al. 2015). A random intercept term for plot was included in the model to account for variance heterogeneity among sites (e.g., related to differences in local environmental conditions and in sampling and chemical analyses carried out by the individual countries/regions). This resulted in the following modelling function:

$$TF(MPS)_{ij} = \alpha + \beta_1 \times Plot_i + \beta_2 \times SPIn_{ij} + \varepsilon_{ij} \qquad (1)$$

$Plot_i$ is a factor with n levels (i = 1, …, n) and j is the number of years. Bootstrapped (n = 1000 simulations) 95 %-confidence intervals for predicted values were calculated using the add_ci function included in the R package ‘ciTools’ (Haman and Avery 2020). Model validity was judged based on the homogeneity of residual variance across levels of the predictor variable (SPIn) and the fitted values (Zuur et al. 2009). All variables had a skewed distribution and were therefore $\log_e$-transformed prior to modelling. For SPIn, TF DON, TF NH_4^+-N and TF NO_3^--N the value of 0.5 had to be added before transforming because the data contained zeros (McDonald 2014).

The contribution of pollen to TF (in kg ha^{-1} yr^{-1}) for each compound was calculated as the difference between the mean annual TF flux during the MPS (Table 1) and the sum of the mean TF flux during the previous and the following month for each Level II plot:

$$TF(pollen) = TF(MPS) - TF(previous + following\ month) \qquad (2)$$

The contribution of pollen to TF fluxes was also expressed in terms of a percentage (%) of annual mean TF deposition. Values were compared among the four genera and among the five compounds using the non-parametric multiple comparison test after Kruskal-Wallis (R function ‘Kruskalmc’) included in the R package ‘pgirmess’ (Giraudoux 2015).

For *Fagus*, the genus with probably the most distinct masting behaviour (Nussbaumer et al. 2016; 2020), we additionally applied the non-parametric Wilcoxon test from the R package ‘stats’ (R Core Team 2022) to test the difference in input from pollen to TF fluxes between years with massive pollen dispersal (SPIn > 80 % of the annual mean) and all years, and between years with massive pollen dispersal and years with low pollen production (SPIn ≤ 80 % of the annual mean).

For each tree species, we applied the one-sided t-test included in the R package ‘stats’ (R Core Team 2022) to test whether the contribution of pollen to TF fluxes was different from zero. For *Fagus*, the same was also done for years with massive pollen dispersal and years with low pollen production separately.

Results

Relationship between SPIn and TF element fluxes

For *Fagus*, the LME models revealed a significant positive relationship between TF fluxes of K^+, NH_4^+-N, DON, DOC and SPIn (Fig. 2, Table 2). However, a significant negative relationship was found between TF fluxes of NO_3^--N and SPIn for *Fagus*. For *Quercus*, the TF element fluxes showed no relationship with SPIn. For *Pinus*, a significant positive relationship was found between TF fluxes of K^+ and SPIn, but not for the other elements. For *Picea*, TF fluxes of K^+ and NH_4^+-N showed a significant positive relationship with SPIn, but not the other elements.

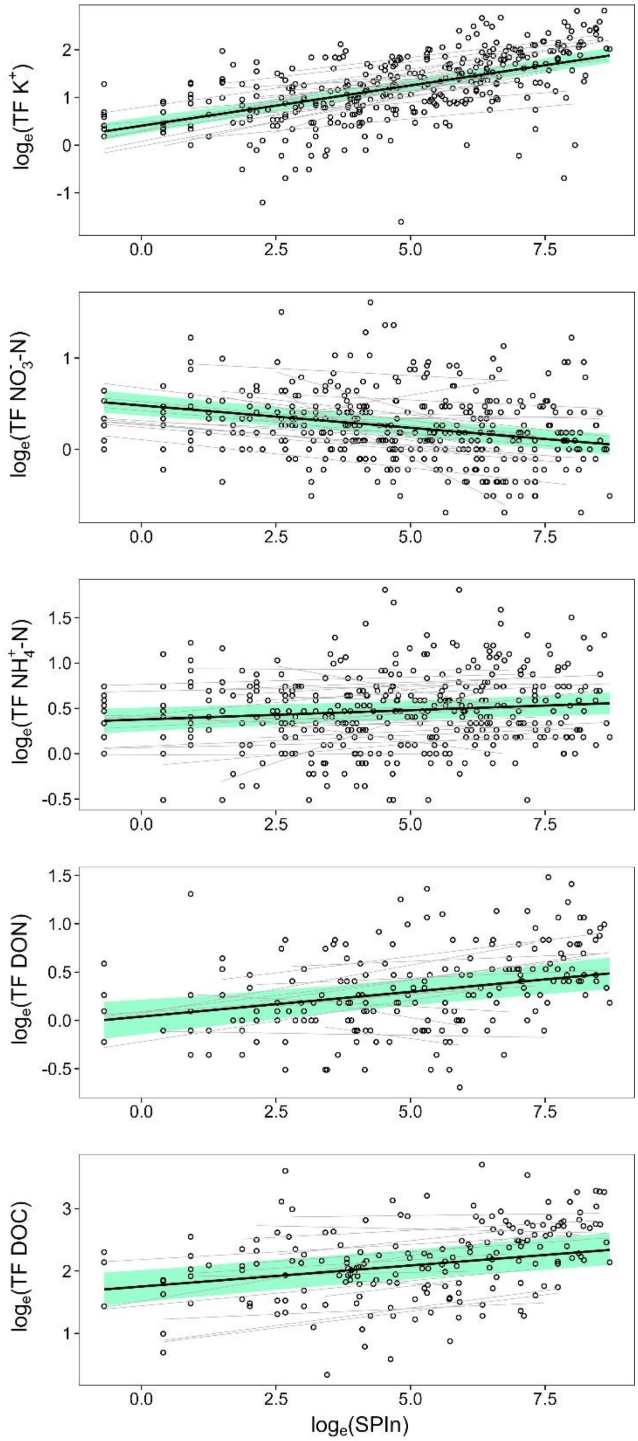


Fig. 2 Output of the mixed-effects models (LME) for *Fagus* stands. Left: log$_e$-transformed throughfall deposition fluxes (kg ha^{-1}) of K$^+$, NO$_3^-$-N, NH$_4^+$-N, DON, and DOC during the MPS (as in Table 1) in function of log$_e$-transformed SPIn (pollen grains * day * m^{-3}). Black lines represent the response curves and green fields are the 95 %-confidence intervals. Circles represent individual plot-years. Grey lines were added to show the relationship for individual plots using simple linear regression (also when not significant). Based on ICP Forests data.

Table 2. Modelling results (LME) for TF element fluxes (kg ha^{-1}) in function of SPIn (pollen grains * day * m^{-3}) during the MPS, with the number of included plot-years (n) and estimated slope of the relationship. The value of the t-test is shown between brackets when significant (t > ±1.96, ns: not significant). Based on ICP Forests data.

Tree genus		K^+	NO_3^--N	NH_4^+-N	DON	DOC
Fagus	Slope	0.169 (12.9)	-0.049 (-6.7)	0.021 (2.6)	0.051 (4.5)	0.069 (4.9)
	n	371	382	382	196	186
Quercus	Slope	ns	ns	ns	ns	ns
	n	208	210	210	133	97
Pinus	Slope	0.123 (2.5)	ns	ns	ns	ns
	n	192	192	192	175	146
Picea	Slope	0.074 (5.1)	ns	0.048 (5.1)	ns	ns
	n	280	307	307	142	137

Impact of pollen on TF

Mean annual contribution of pollen to the TF fluxes of K^+ was significantly higher in *Quercus* (3.67 kg K^+ ha^{-1} yr^{-1}) and *Fagus* stands (2.20 kg K^+ ha^{-1} yr^{-1}) than in *Pinus* (0.79 kg K^+ ha^{-1} yr^{-1}) and *Picea* stands (0.62 kg K^+ ha^{-1} yr^{-1}) (Fig. 3a, Table S3–6). The input from pollen covered a significantly lower mean part of annual TF K^+ fluxes in *Picea* stands (4.8 %) than in *Quercus* stands (15.0 %) and *Fagus* stands (10.5 %), but there was no significant difference to the mean for *Pinus* (10.9 %) (Fig. 3f). Pollen added significantly more DOC to TF in *Quercus* stands (9.54 kg C ha^{-1} yr^{-1}) than in *Fagus* (3.21 kg C ha^{-1} yr^{-1}) and *Picea* stands (2.91 kg C ha^{-1} yr^{-1}), but the difference with the mean for *Pinus* (3.29 kg C ha^{-1} yr^{-1}) was not significant (Fig. 3e). The input from pollen covered a significantly higher mean part of annual TF DOC fluxes in *Quercus* stands (12.0 %) than in *Picea* stands (4.3 %), but the difference with the mean for *Fagus* (7.0 %) and *Pinus* (6.3 %) was not significant (Fig. 3j). The contribution of pollen to TF DON was significantly higher in *Quercus* stands (0.53 kg N ha^{-1} yr^{-1}) than in *Picea* (0.14 kg N ha^{-1} yr^{-1}) and *Pinus* stands (0.13 kg N ha^{-1} yr^{-1}), but the difference with the mean for *Fagus* (0.33 kg N ha^{-1} yr^{-1}) was not significant (Fig. 3d). The input from pollen covered a significantly higher mean part of annual TF DON fluxes in *Quercus* stands (13.1 %) than in *Picea* stands (3.9 %), but the difference with the mean for *Fagus* (8.0 %) and *Pinus* (4.9 %) was not significant (Fig. 3i). The mean annual input of NH_4^+-N to TF by pollen was considerable in *Quercus* stands (0.68 kg N ha^{-1} yr^{-1} or 10.4 % of annual TF fluxes), limited in *Fagus* (0.12 kg N ha^{-1} yr^{-1} or 2.1 % of annual TF fluxes) and *Pinus* stands (0.10 kg N ha^{-1} yr^{-1} or 1.9 % of annual TF fluxes), and negligible in *Picea* stands (0.01 kg N ha^{-1} yr^{-1} or 0.3 % of annual TF fluxes, Fig. 3 c and h). Pollen removed more NO_3^--N from TF in *Quercus* stands (0.15 kg N ha^{-1} yr^{-1}) than in *Picea* (0.02 kg N ha^{-1} yr^{-1}) and *Pinus* stands (where even an addition of 0.01 kg N ha^{-1} yr^{-1} was found), but the difference to the removal in *Fagus* stands (0.12 kg N ha^{-1} yr^{-1}) was not significant (Fig. 3b). The percentage of annual TF NO_3^--N fluxes removed by pollen was, however, not statistically different among tree species (Fig. 3g). Overall, the estimated contribution of pollen to TF fluxes was highest in *Quercus* > *Fagus* > *Pinus* > *Picea*, covering on average 4.1–10.6 % of the annual TF fluxes of K^+ > DOC > DON > NH_4^+-N, while pollen removed on average 1.8 % of annual TF NO_3^--N fluxes in European forests (significant differences among elements are listed in Table S7). The impact of pollen on TF element fluxes was significantly different from zero for all tree genera, except for NH_4^+-N and NO_3^--N in *Pinus* and *Picea* stands (Fig. 3).

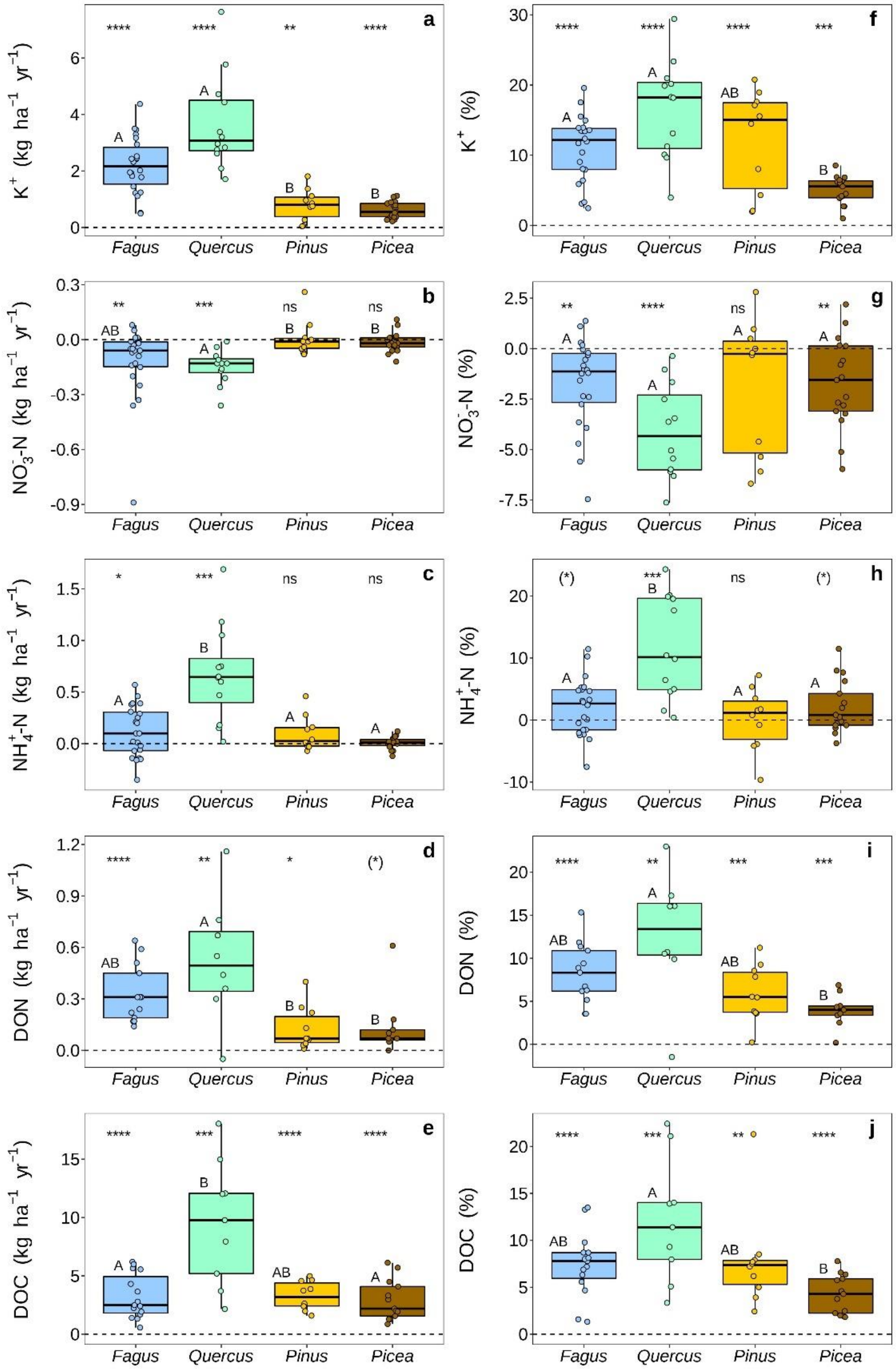


Fig. 3 Input from pollen to TF fluxes in *Fagus*, *Quercus*, *Pinus* and *Picea* stands as derived from TF monitoring data (a positive value means that pollen added elements, a negative value that pollen removed elements). Dots represent individual plots. a–e: mean annual contribution (kg ha^{-1} yr^{-1}). f–j: expressed as percentage of mean annual TF flux. Capital letters (A, B) denote significantly different groups ($p < 0.05$). Symbols indicate whether the mean of all values is different from zero (****: p <

0.0001; ***: $p < 0.001$; **: $p < 0.01$; *: $p < 0.05$; (*): $p < 0.1$; ns: not significant). Based on ICP Forests data

For *Fagus*, the contribution of pollen to TF K^+ fluxes was about three times higher ($p < 0.0001$) in years with massive pollen dispersal (3.70 kg ha^{-1} yr^{-1} or 15.3 % of annual TF fluxes) than in other years (1.15 kg ha^{-1} yr^{-1} or 6.1 % of annual TF fluxes) (Fig. 4a and f). In *Fagus* stands, pollen also added more than twice the amount of DOC to TF ($p < 0.01$) in years with massive pollen dispersal (4.78 kg ha^{-1} yr^{-1} or 9.8 % of annual TF fluxes) than in other years (1.89 kg ha^{-1} yr^{-1} or 4.4 % of annual TF fluxes) (Fig. 4e and j). In absolute terms, the contribution of pollen to TF DON fluxes in *Fagus* stands was almost two times higher in years with massive pollen dispersal (0.45 kg ha^{-1} yr^{-1}, 10.4 % of annual TF fluxes) than in other years (0.25 kg ha^{-1} yr^{-1} or 6.3 % of annual TF fluxes), but the difference was not significant (Fig. 4d and i). The difference in the contribution of pollen to TF NH_4^+-N fluxes in *Fagus* stands between years with massive pollen dispersal (0.22 kg ha^{-1} yr^{-1} or 3.8 % of annual TF fluxes) and other years (0.05 kg ha^{-1} yr^{-1} or 1.0 % of annual TF fluxes) was also not significant (Fig. 4c and h). *Fagus* pollen removed over three times more NO_3^--N from TF ($p < 0.0001$) in years with massive pollen dispersal (0.23 kg ha^{-1} yr^{-1} or 4.2 % of annual TF fluxes) than in other years (0.06 kg ha^{-1} yr^{-1} or 1.0 % of annual TF fluxes) (Fig. 4b and g). In years with massive pollen dispersal, the impact of pollen on TF fluxes was significantly different from zero for all elements. In years with low pollen dispersal, the impact of pollen on TF fluxes was significantly different from zero for K^+, DOC and DON, but not for NH_4^+-N and NO_3^--N (results not shown).

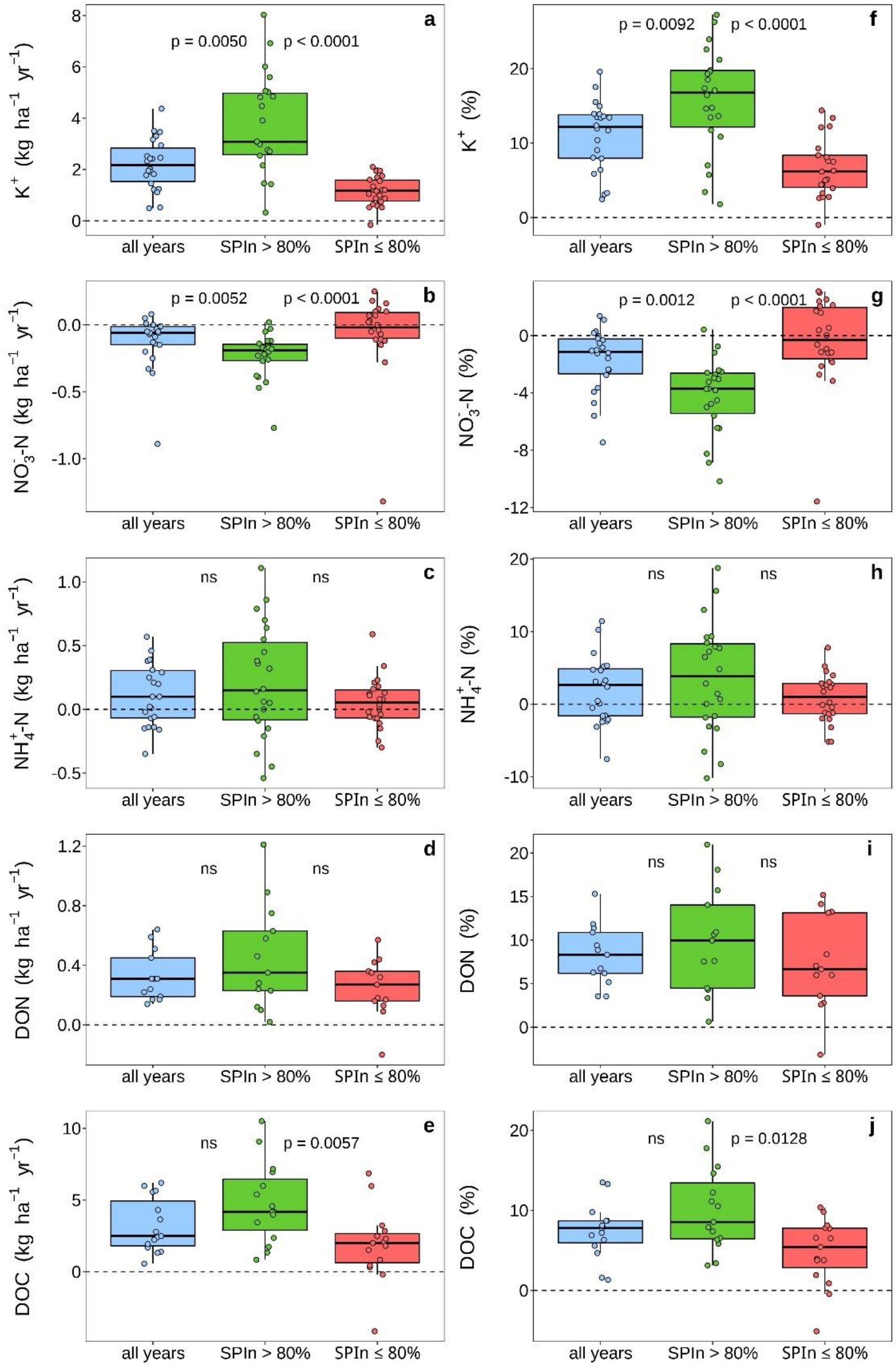


Fig. 4 Input from pollen to TF fluxes in *Fagus* stands as derived from monitoring data (n = 22) for all years compared with years for which SPIn > 80 % of the annual mean, and years for which SPIn ≤ 80 % of the annual mean. Dots represent individual plots. a–e: mean annual contribution (kg ha^{-1} yr^{-1}). f–j: expressed as percentage of mean annual TF flux. The significance level of the difference between

groups is indicated by the p-value of the Wilcoxon test (ns = not significant). Based on ICP Forests data

Discussion

Effects of pollen on TF element fluxes

This study, to our knowledge the first of its kind performed at large spatial scale, yielded a quantitative estimate of the impact of airborne pollen on TF fluxes of K^+, NO_3^--N, NH_4^+-N, DOC, and DON based on long-term measurements in managed, mostly pure, even-aged forest stands across Europe. Isolating the effects of pollen during the MPS enabled to minimize other factors influencing TF fluxes year-round or throughout the growing season, including atmospheric deposition, leachates from foliage and associated nitrifying microorganisms (negligible in broadleaved stands in the period of leaf emergence), stems and branches, phytophagous insects and epiphytic mosses and lichens (Pitman et al. 2010; Van Stan et al. 2012; Wagenhoff et al. 2014; Guerrieri et al. 2021). Nevertheless, outbreaks of certain phytophagous insects like *Melolontha melolontha* and *Tortrix viridana* may impact TF fluxes in spring particularly in *Quercus* stands and might have partly obscured the effects of pollen in this group of forests. Another factor that might influence atmospheric fluxes specifically during the MPS more generally is leaching from flowers (i.e. flower parts other than pollen), but as far as we know this has never been quantified. The latter would require a laboratory dissolution experiment, and prior removal of any residual pollen from the flower parts, which is probably difficult to achieve. In this study, we assumed that leaching from flower parts is negligible, considering that flowers from *Fagus*, *Quercus*, *Pinus* and *Picea* decompose relatively slowly. Because the total productions of inflorescences, flowers, anthers and pollen grains in trees are all logically correlated (Molina et al. 1996), any leaching from flower parts other than pollen will not obscure but rather amplify the biochemical signal from pollen, and does not significantly affect the conclusions of our study. The significantly higher fluxes that we found in *Fagus* stands during years with massive pollen dispersal, provided further evidence that the observed effects were induced by pollen. Pollen, however, hosts various microorganisms, including bacteria and fungi, that might affect pollen germination and tube elongation or pollen defence and further study is needed to elucidate their possible role in pollen-associated biochemistry (Madmony et al. 2005; Obersteiner et al. 2016; Cullen et al. 2021).

Many unknowns exist about the complex processes governing forest canopy N cycling (Guerrieri et al. 2021). However, our results support the findings from previous TF studies at local/regional level and *in vitro* experiments suggesting that pollen is an important source of K^+ and organic C and N in TF, confirming our main hypothesis (Table 3). Moreover, we found that pollen also may affect TF inorganic N fluxes by adding NH_4^+-N and removing NO_3^--N. The latter provides evidence for an effect that, to date, was demonstrated only at the intracellular level, i.e. the enzymatic reduction of NO_3^- through the 2-step nitrate reductase pathway (Bright et al. 2009). Furthermore, it might explain why NO_3^- concentrations at a North American watershed were found to be lower in precipitation and TF collected with bulk samplers compared to wet-only samplers, the latter excluding dry deposition (Richter and Lindberg 1988). Hence, pollen dispersal appears to be an important factor also in canopy inorganic N cycling. If NO_3^- in TF is enzymatically reduced by pollen, then short-lived NO_2^- peaks are expected in TF, at the moment when intermediately formed NO_2^- is not yet further reduced into NO, or converted back into NO_3^- by nitrifiers, which appears to be common in the phyllosphere (Guerrieri et al. 2015, 2021). Unfortunately, it was impossible to include NO_2^- in the present study because few countries analyse this compound in TF samples and the available data were too limited. However, in a mixed *Quercus robur – Fagus sylvatica* stand in Gontrode (Flanders, Belgium), which was not included in this study where we focused on single-species even-aged stands, remarkable peaks in the TF NO_2^- fluxes of variable magnitude were indeed observed during the spring in most years (Fig. S2, unpublished results INBO). This indicates the possible effects of pollen and/or associated microorganisms on TF inorganic N fluxes. Specific laboratory experiments and a more detailed analysis of inorganic N forms in TF might help to shed light on the abilities of pollen to reduce extracellular NO_3^-.

Table 3. Overview of the effects of rehydrating pollen on elemental concentrations in solution reported by various studies.

Study	Tree species	Effect
<u>Analysis of TF data</u>		
Carlisle et al. 1966	*Quercus petraea*	K^+, P (increase)
Ferm 1997	*Picea abies*	K^+, organic N (increase)
Le Mellec et al. 2010	*Fagus sylvatica*, *Picea abies*	organic C and N (increase)
Verstraeten et al. 2016	*Fagus sylvatica*, *Quercus robur*	organic N (increase)
<u>*In vitro* experiments with pollen</u>		
Doskey and Ugoagwu 1989	*Pinus resinosa*, *Pinus strobus*	K^+, P, organic C (increase)
Bright et al. 2009	*Magnolia delavayi*, *Protea nitida*,	intracellular: NO_3^- (decrease),
	Banksia serata, *Ulmus scabra*	NO_2^-, NO (increase)
Rösel et al. 2012	*Pinus sylvestris*	P, organic C, N (increase)

We found an overall higher impact of pollen on TF composition in deciduous broadleaved stands (*Quercus*, *Fagus*) compared to evergreen coniferous stands (*Pinus*, *Picea*). It was rather expected that *Quercus* and *Pinus* would alter the fluxes the most because these genera produce many flowers and high amounts of pollen each year. We think this discrepancy could be explained by interference from pollen originating from broadleaved trees growing in the understorey or direct surroundings of coniferous stands. A large part of the *Pinus* and *Picea* plots is located in the boreal zone of Fennoscandia (40 % of the *Pinus* stands and 76 % of the *Picea* stands, Fig. S1), where *Betula* is a naturally widespread and abundant genus (Beck et al. 2016). The main pollen season of *Betula* in Fennoscandia ranges from late April to early June and is partly overlapping with that of *Pinus* and *Picea* (Biedermann et al. 2019). Given the method (formula (2)) applied to calculate the contribution of pollen to TF fluxes, admixture of *Betula* pollen may have resulted in an underestimation of the impact in coniferous stands. To verify this, it would be necessary to study local pollen spectra at the stand level, as explained below.

Although according to previous studies TF P fluxes are increased by pollen (Table 3), it was not possible to include analysis of P in this study because the available data were too limited.

Indicative value of SPIn for the impact of pollen on TF element fluxes

For *Fagus* stands, we found a clear relationship between SPIn and TF element fluxes of K^+, NO_3^--N, NH_4^+-N, DOC, and DON, confirming our main hypothesis, but this was not the case for *Picea*, *Pinus* and *Quercus* stands. Differences in the intrinsic dispersal abilities of pollen at the tree genus level may partly explain this result. The effective transport distance of pollen from wind-pollinated trees is inversely related to its size and weight, but other factors also seem to play a role, like pollen morphology and local topography (Erdtman 1969; Damialis et al. 2017). For *Fagus* pollen (Ø 30–50 µm), the effective transport distance is generally limited to a few hundred meters (Wagner et al. 2010), but this might be overcome by the common occurrence of *Fagus* in large parts of Europe. For genera with larger and heavier pollen grains such as *Abies* and *Picea* (Ø 80–150 µm), airborne pollen concentrations in residential areas are generally less representative for those in forests because few pollen grains are able to reach the samplers, although the pollen grains of both taxa are equipped with air sacs (‘sacci’) that facilitate air transport. In contrast, pollen from *Pinus* (Ø 40–85 µm, also equipped with two sacci) and also *Betula* (Ø 20–25 µm) and *Quercus* (Ø 26–40 µm) can float in the atmosphere, allowing it to travel much further (Wrońska-Pilarek et al. 2016; Damialis et al. 2017; Bogawski et al. 2019). This means that *Pinus, Betula* or *Quercus* pollen collected at a particular location can originate from both local and remote sources, depending on the meteorological conditions, particularly wind, rainfall, and solar radiation (Szczepanek et al. 2017; Bruffaerts et al. 2018; Verstraeten et al. 2019). Measurements of airborne pollen concentrations are not included in the standard monitoring programme for Level II plots, so we had to rely on data from aerobiological monitoring stations installed predominantly in urban environments to assess the risk for associated respiratory allergies (Buters et al. 2018; Biedermann et al. 2019). This difference in location means that the representativeness of SPIn for the airborne pollen concentrations in the forest may vary, depending on the distance, difference in altitude, and other factors. In general, SPIn as well as pollen accumulation rates in sediments are positively related to the abundance and proximity of local pollen sources, especially forests, but also trees growing along streets and in parks (Verstraeten et al. 2019; Abraham et al. 2021). To get more representative airborne pollen concentration data for forests, passive pollen traps (gravitational fluxes) would have to be installed under the canopy. Alternatively, the TF filtration residue using filters with an adequate pore size (5 µm) could be analysed. Ground-level samplers within the forest plots might have allowed to relate more directly local pollen emissions to throughfall biochemistry, but it would also have been more influenced by the pollination behaviour of some individual trees and might be less representative of the forest masting that this study aims to relate at larger scale. Elaboration of pollen dispersion models based on the spatial distribution and abundance of tree species combined with meteorological data may provide further insights into the emission sources of pollen, but these models are currently available only for a limited number of tree genera, e.g. *Betula* and *Pinus* (Sofiev et al. 2013; Szczepanek et al. 2017; Verstraeten et al. 2019).

Besides variation in the representativeness of airborne pollen concentrations, differences in flowering strategies among the tree genera likely affect the results. Trees belonging to the genera *Fagus* and *Picea* exhibit a distinct masting behaviour and produce much more flowers during years with successful development of fruits and seeds and vice versa. On the contrary, *Quercus* and *Pinus* produce large amounts of flowers every year resulting in airborne pollen concentrations that are higher and more constant over the years (Geburek et al. 2012; Lebourgeois et al. 2018; Bogdziewicz et al. 2019). This contrast is reflected in the overall higher SPIn and lower coefficient of variation (CV) of SPIn for *Quercus* and *Pinus* stands compared to *Fagus* and *Picea* stands (Fig. 5a and b). Limited ability to catch the annual variations in constantly very high pollen loads with the available pollen predictors is probably the second main reason why we could not find a clear relationship between TF element fluxes and SPIn for *Quercus* and *Pinus*.

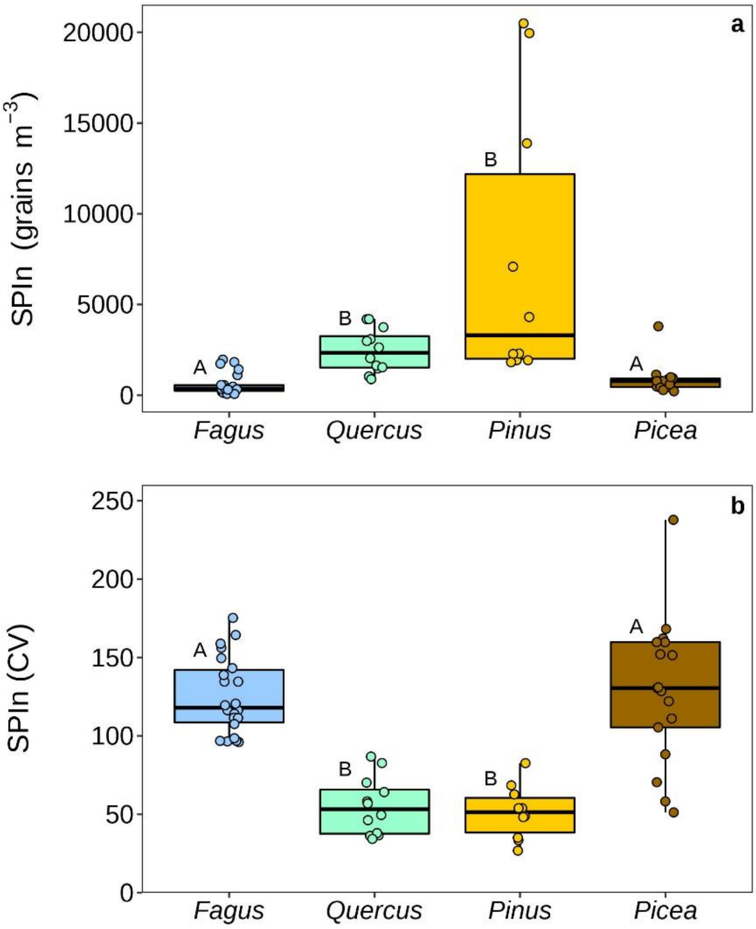


Fig. 5 Boxplots of mean annual SPIn (grains m^{-3}) per plot (a) and coefficient of variation (CV) of SPIn per plot (b) for the four tree genera. Capital letters (A–C) denote significantly different groups ($p < 0.05$)

Conclusions

Tree pollen was found to make a substantial contribution to the annual TF fluxes of K^+ > DOC > DON > NH_4^+-N in managed, mostly pure, even-aged forest stands across Europe, an effect that appeared to be stronger in deciduous broadleaved stands than in evergreen coniferous stands (*Quercus* > *Fagus* > *Pinus* > *Picea*). On the other hand, we obtained evidence that pollen removes a small part of annual TF NO_3^--N fluxes. The estimation of throughfall fluxes is thus significantly altered by pollen. Airborne pollen concentrations (SPIn) measured in urban areas were able to predict TF fluxes for tree genera with distinct masting behaviour like *Fagus* and *Picea*, but not for genera with a more constant pollen production like *Quercus* and *Pinus*. Pollen dispersal seems to play a more prominent and complex role in forest nutrient cycling than previously thought, also with regard to inorganic N.

Acknowledgements

This study was funded by the Flemish government. It is based on data collected by partners of the official UNECE ICP Forests Network (http://icp-forests.net/contributors), partly co-financed by the European Commission (data downloaded on 8 May 2020). The authors would like to express their gratitude to the members of the ICP Forests Expert Panel on Deposition for giving their consent to use the throughfall data for this study. The Italian Aerobiological Monitoring Network POLLnet, belonging to the National System for Environmental Protection - SNPA (http://www.pollnet.it/default_it.asp#), is kindly acknowledged for the provision of airborne pollen concentration data from their stations listed in Table S2. We also want to thank the Air Pollution and Plant Analysis unit at the Austrian Research Centre for Forests (BFW), Astma-Allergi Danmark, Gábor Bobvos, Andrea-Pil Holm, Gergely Mányoki, Marijke Hendrickx, Iben Margrete Thomsen, Michel Thibaudon, Trond Einar Brobakk, Charlotte Sindt, Malgorzata Latałowa, Marcelina Zimny, Joanna Święta-Musznicka, Michael Tatzber and Hans-Peter Dietrich for technical advice on the aerobiological monitoring data and Pieter Verschelde for advice on the statistical analysis.

Conflicts of interest/Competing interests

Not applicable.

Availability of data and material

The TF data can be obtained by submitting an official data request through the ICP Forests website http://icp-forests.net/. Data on airborne pollen concentrations can be requested from the national/regional monitoring networks representatives (https://www.zaum-online.de/pollen/pollen-monitoring-map-of-the-world/).

Code availability

Not applicable.

Authors' contributions

A.V., E.G. and N.B. designed the study; A.V. compiled the data, analysed the data and drafted the manuscript; and all other authors provided data and/or commented critically to the drafts and gave final approval for publication.

References

Abraham V, Hicks S, Svobodová-Svitavská H, et al. (2021) Patterns in recent and Holocene pollen accumulation rates across Europe – the Pollen Monitoring Programme Database as a tool for vegetation reconstruction, Biogeosciences 18:4511–4534. https://doi.org/10.5194/bg-18-4511-2021

Bates D, Maechler M, Bolker B, Walker S (2015) Fitting linear mixed-effects models using lme4. J Stat Soft 67, 1–48. https://doi.org/10.18637/jss.v067.i01

Beck P, Caudullo G, de Rigo D, Tinner W (2016) *Betula pendula*, *Betula pubescens* and other birches in Europe: distribution, habitat, usage and threats. In: San-Miguel-Ayanz J, de Rigo D, Caudullo G, Houston Durrant T, Mauri A (Eds.), European atlas of forest tree species. Publ. Off. EU, Luxembourg, pp. e010226+

Biedermann Y, Winther L, Till SJ, Panzner P, Knulst A, Valovirta E (2019) Birch pollen allergy in Europe. Allergy 74:1237–1248. https://doi.org/10.1111/all.13758

Bogawski P, Borycka K, Grewling L, Kasprzyk I (2019) Detecting distant sources of airborne pollen for Poland: Integrating back-trajectory and dispersion modelling with a satellite-based phenology. Sci Total Environ 689: 109–125. https://doi.org/10.1016/j.scitotenv.2019.06.348

Bogdziewicz M, Szymkowiak J,Fernández-Martínez M, Peñuelas J, Espelta JM (2019) The effects of local climate on the correlation between weather and seed production differ in two species with contrasting masting habit. Agric For Meteorol 268:109–115. https://doi.org/10.1016/j.agrformet.2019.01.016

Bright J, Hiscock SJ, James PE, Hancock JT (2009) Pollen generates nitric oxide and nitrite: a possible link to pollen-induced allergic responses. Plant Physiol Bioch 47:49–55. https://doi.org/10.1016/j.plaphy.2008.09.005

Bruffaerts N, De Smedt T, Delcloo A, Simons K, Hoebeke L, Verstraeten C, Van Nieuwenhuyse A, Packeu A, Hendrickx M (2018) Comparative long-term trend analysis of daily weather conditions with daily pollen concentrations in Brussels, Belgium. Int J Biometeorol 62:483–491. https://doi.org/10.1007/s00484-017-1457-3

Buters JTM, Antunes C, Galveias A, Bergmann KC, Thibaudon M, Galán C, Schmidt‐Weber C, Oteros J (2018) Pollen and spore monitoring in the world. Clin Transl Allergy 8:9 https://doi.org/10.1186/s13601-018-0197-8

Carlisle A, Brown AHF, White EJ (1966) The organic matter and nutrient elements in the precipitation beneath a Sessile oak (*Quercus petraea*) canopy. J Ecol 54:87–98. https://doi.org/10.2307/2257660

Clarke N, Žlindra D, Ulrich E, Mosello R, Derome J, Derome K, König N, Geppert, F, Lövblad G, Draaijers GPJ, Hansen K, Thimonier A, Waldner P, Verstraeten A (2022) Part XIV: Sampling and analysis of deposition. Version 2022-1. In: UNECE ICP Forests Programme Coordinating Centre (ed.): Manual on methods and criteria for harmonized sampling, assessment, monitoring and analysis of the effects of air pollution on forests. Thünen Institute of Forest Ecosystems, Eberswalde, Germany, 34 p. + Annex.

Cullen NP, Fetters AM, Ashman T-L (2021) Integrating microbes into pollination. Curr Opin Insect Sci 44:48–54. https://doi.org/10.1016/j.cois.2020.11.002

Damialis A, Kaimakamis E, Konoglou M et al. (2017) Estimating the abundance of airborne pollen and fungal spores at variable elevations using an aircraft: how high can they fly? Sci Rep 7:44535. https://doi.org/10.1038/srep44535

Doskey PV, Ugoagwu BJ (1989) Atmospheric deposition of macronutrients by pollen at a semi-remote site in northern Wisconsin. Atmos Environ 23:2761–2766. https://doi.org/10.1016/0004-6981(89)90556-8

Draaijers GPJ, Erisman JW, Spranger T, Wyers GP (1996) The application of throughfall measurements for atmospheric deposition monitoring. Atmos Environ 30:3349–3361. https://doi.org/10.1016/1352-2310(96)00030-1

Erdtman G (1969) Handbook of palynology, 486pp. Munksgaard, Copenhagen.

Faegri K, van der Pijl L (1979) The Principles of Pollination Ecology. 3rd edn. - Pegamon Press, Oxford.

Ferm M (1997) Improvement and validation of the throughfall technique for nitrogen deposition measurements to forest ecosystems. In: Slanina S, eds. Biosphere-atmosphere exchange of pollutants and trace substances. Transport and chemical transformation of pollutants in the troposphere, vol 4. Springer, Berlin, Heidelberg.

Ferretti M, König N, Granke O, Nicolas M (2021) Part III: Quality Assurance within the ICP Forests monitoring programme. Version 2021-1. In: UNECE ICP Forests Programme Co-ordinating Centre (ed.): Manual on methods and criteria for harmonized sampling, assessment, monitoring and analysis of the effects of air pollution on forests. Thünen Institute of Forest Ecosystems, Eberswalde, Germany, 14 p. + Annex.

Galán C, Ariatti A, Bonini M, Clot B, Crouzy B, Dahl A, Fernandez-González D, Frenguelli G, Gehrig R, Isard S, Levetin E, Li DW, Mandrioli P, Rogers CA, Thibaudon M, Sauliene I, Skjoth C, Smith M, Sofiev M (2017) Recommended terminology for aerobiological studies. Aerobiologia 33:293–295. https://doi.org/10.1007/s10453-017-9496-0

Geburek T, Hiess K, Litschauer R, Milasowszky N (2012) Temporal pollen pattern in temperate trees: expedience or fate? Oikos 121:1603–1612. https://doi.org/10.1111/j.1600-0706.2011.20140.x

Gehrig R (2019) Representativeness of pollen traps: a review of the national pollen network of Switzerland. Aerobiologia 35:577–581. https://doi.org/10.1007/s10453-019-09593-z

Giraudoux P (2018) pgirmess: spatial analysis and data mining for field ecologists. R package version 1.6.9. https://CRAN.R-project.org/package=pgirmess.

Grewling L, Jackowiak B, Nowak M, Uruska A, Smith M (2012) Variations and trends of birch pollen seasons during 15 years (1996–2010) in relation to weather conditions in Poznań (western Poland). Grana 51:280–292. https://doi.org/10.1080/00173134.2012.700727

Guerrieri R, Templer P, Magnani, F (2021) Canopy exchange and modification of nitrogen fluxes in forest ecosystems. Curr Forestry Rep 7:115–137. https://doi.org/10.1007/s40725-021-00141-y

Guerrieri R, Vanguelova EI, Michalski G, Heaton THE, Mencuccini M (2015) Isotopic evidence for the occurrence of biological nitrification and nitrogen deposition processing in forest canopies. Glob Change Biol 21:4613–4626. https://doi.org/10.1111/gcb.13018

Haman J, Avery M (2020) ciTools: confidence or prediction intervals, quantiles, and probabilities for statistical models. R package version 0.6.1, https://CRAN.R-project.org/package=ciTools

Kelly D, Sork VL (2002) Mast seeding in perennial plants: why, how, where? Annu Rev Ecol Evol Syst 33:427–447. https://doi.org/10.1146/annurev.ecolsys.33.020602.095433

Kluska K, Piotrowicz K, Kasprzyk I (2020) The impact of rainfall on the diurnal patterns of atmospheric pollen concentrations, Agric For Meteorol 291, 108042. https://doi.org/10.1016/j.agrformet.2020.108042

Lebourgeois F, Delpierre N, Dufrêne E, Cecchini S, Macé S,·Croisé L, Nicolas M (2018) Assessing the roles of temperature, carbon inputs and airborne pollen as drivers of fructification in European temperate deciduous forests. Eur J For Res 137:349–365. https://doi.org/10.1007/s10342-018-1108-1

Le Mellec A, Meesenburg H, Michalzik B (2010) The importance of canopy-derived dissolved and particulate organic matter (DOM and POM) – comparing throughfall solution from broadleaved and coniferous forests. Ann For Sci 67:411. https://doi.org/10.1051/forest/2009130

Lindberg SE, Lovett GM, Richter DD, Johnson DW (1986) Atmospheric deposition and canopy interactions of major ions in a forest. Science 231:141–145. https://doi.org/10.1126/science.231.4734.141

Madmony A, Chernin L, Pleban S, Peleg E, Riov J (2005) Enterobacter cloacae, an Obligatory Endophyte of Pollen Grains of Mediterranean Pines. Folia Microbiol 50:209–216. https://doi.org/10.1007/BF02931568

McDonald JH (2014) Handbook of Biological Statistics. Sparky House, Maryland.

Molina RT, Rodríguez AM, Palaciso IS, López G (1996) Pollen production in anemophilous trees. Grana 35:38–46. https://doi.org/10.1080/00173139609430499

Nussbaumer A, Meusburger K, Schmitt M, Waldner P, Gehrig R, Haeni M, Rigling A, Brunner I, Thimonier A (2020) Extreme summer heat and drought lead to early fruit abortion in European beech. Scientific Reports 10:5334. https://doi.org/10.1038/s41598-020-62073-0

Nussbaumer A, Waldner P, Etzold S, Gessler A, Benham S, Thomsen I M, Jørgensen B B, Timmermann V, Verstraeten A, Sioen G, Rautio P, Ukonmaanaho L, Skudnik M, Apuhtin V, Braun S, Wauer A (2016) Patterns of mast fruiting of common beech, sessile and common oak, Norway spruce and Scots pine in Central and Northern Europe. Forest Ecol Manage 363:237–251. http://dx.doi.org/10.1016/j.foreco.2015.12.033

Obersteiner A, Gilles S, Frank U, Beck I, Häring F, Ernst D, et al. (2016) Pollen-associated microbiome correlates with pollution parameters and the allergenicity of pollen. PLoS ONE 11:e0149545. http://dx.doi.org/10.1371/journal.pone.0149545

Pitman RM, Vanguelova EI, Benham SE (2010) The effects of phytophagous insects on water and soil nutrient concentrations and fluxes through forest stands of the level II monitoring network in the UK. Sci Total Environ 409:169–181. https://doi.org/10.1016/j.scitotenv.2010.09.029

R Core Team (2022) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. https://www.R-project.org/.

Ranta H, Hokkanen T, Linkosalo T, Laukkanen L, Bondestam K, Oksanen A (2008) Male flowering of birch: Spatial synchronization, year-to-year variation and relation of catkin numbers and airborne pollen counts. Forest Ecol Manage 255:643–650. https://doi.org/10.1016/j.foreco.2007.09.040

Rejón JD, Delalande F, Schaeffer-Reiss C, Alché JDD, Rodríguez-García MI, Van Dorsselaer A, Castro AJ (2016) The pollen coat proteome: at the cutting edge of plant reproduction. Proteomes 4:5. https://doi.org/10.3390/proteomes4010005

Richter DD, Lindberg SE (1988) Wet deposition estimates from long-term bulk and event wet-only samples of incident precipitation and throughfall. J Environ Qual 17:619–622. https://doi.org/10.2134/jeq1988.00472425001700040017x

Rösel P, Rychła A, Wurzbacher C, Grossart H-P (2012) Effects of pollen leaching and microbial degradation on organic carbon and nutrient availability in lake water. Aquat Sci 74:87–99. https://doi.org/10.1007/s00027-011-0198-3

RStudio Team (2022) RStudio: Integrated Development Environment for R. RStudio, PBC, Boston, MA URL http://www.rstudio.com/.

Sofiev M, Siljamo P, Ranta H, Linkosalo T, Jaeger S, Rasmussen A, Rantio-Lehtimaki A, Severova E, Kukkonen J (2013) A numerical model of birch pollen emission and dispersion in the atmosphere. Description of the emission module. Int J Biometeorol 57:45–58. https://doi.org/10.1007/s00484-012-0532-z

Szczepanek K, Myszkowska D, Worobiec E, Piotrowicz K, Ziemianin M, Bielec-Bąkowska Z (2017) The long-range transport of Pinaceae pollen: an example in Kraków (southern Poland). Aerobiologia 33:109–125. https://doi.org/10.1007/s10453-016-9454-2

Ulrich B (1983) Interaction of forest canopies with atmospheric constituents: SO2, Alkali and earth alkali cations and chloride. In: Pankrath J (ed) B Ulrich. Effects of accumulation of air pollutants in forest ecosystems. D Reidel Publishing Company, Dordrecht, pp 33–45.

Van Stan JT, Levia DF, Inamdar SP, Lepori-Bui M, Mitchell MJ (2012) The effects of phenoseason and storm characteristics on throughfall solute washoff and leaching dynamics from a temperate deciduous forest canopy. Sci Total Environ 430:48–58. https://doi.org/10.1016/j.scitotenv.2012.04.060

Verstraeten A, Verschelde P, De Vos B, Neirynck J, Cools N, Roskams P, Hens M, Louette G, Sleutel S, De Neve S (2016) Increasing trends of dissolved organic nitrogen (DON) in temperate forests under recovery from acidification in Flanders, Belgium. Sci Total Environ 553:107–119. https://doi.org/10.1016/j.scitotenv.2016.02.060

Verstraeten WW, Dujardin S, Hoebeke L, Bruffaerts N, Kouznetsov R, Dendoncker N, Hamdi R, Linard C, Hendrickx M, Sofiev M, Delcloo AW (2019) Spatio-temporal monitoring and modelling of birch pollen levels in Belgium. Aerobiologia 35:703–717. https://doi.org/10.1007/s10453-019-09607-w

Wagenhoff E, Blum R, Delb H (2014) Spring phenology of cockchafers, Melolontha spp. (Coleoptera: Scarabaeidae), in forests of south-western Germany: results of a 3-year survey on adult emergence, swarming flights, and oogenesis from 2009 to 2011. J For Sci 60:154–165. https://doi.org/10.17221/5/2014-JFS

Wagner S, Collet C, Madsen P, Nakashizuka T, Nyland RD, Sagheb-Talebi K (2010) Beech regeneration research: From ecological to silvicultural aspects. Forest Ecol Manage 259:2172–2182. https://doi.org/10.1016/j.foreco.2010.02.029

Wrońska-Pilarek D, Danielewicz W, Bocianowski J, Maliński T, Janyszek M (2016) Comparative pollen morphological analysis and its systematic implications on three European oak (Quercus L., Fagaceae) species and their spontaneous hybrids. PLOS ONE 11:e0161762. https://doi.org/10.1371/journal.pone.0161762

Zuur AF, Ieno EN, Walker N, Saveliev AA, Smith GM (2009) Dealing with Heterogeneity. In: Mixed effects models and extensions in ecology with R. Springer, New York NY, USA, p71–100.

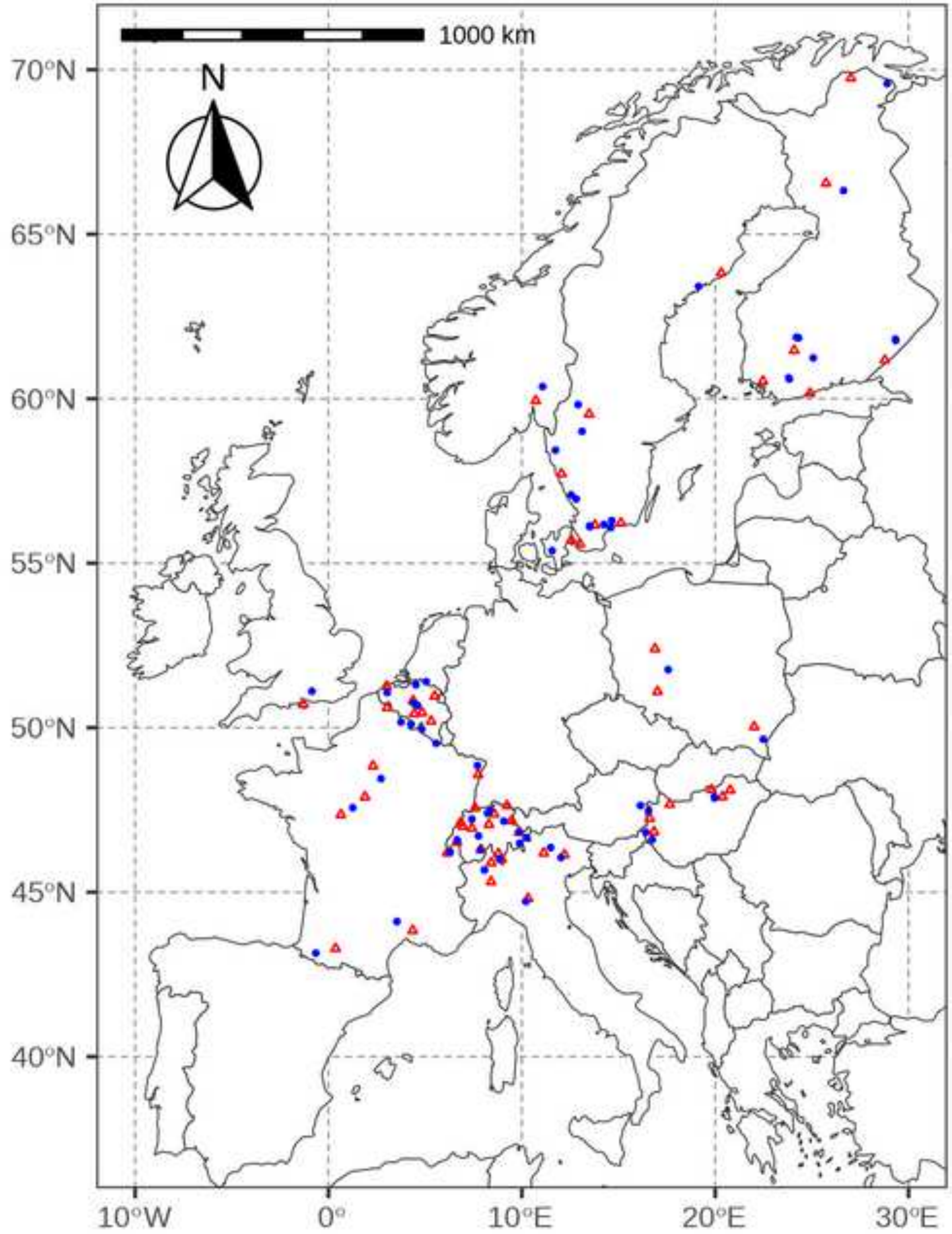

1000 km
N
70°N
65°N
60°N
55°N
50°N
45°N
40°N
10°W
0°
10°E
20°E
30°E

Figure 2

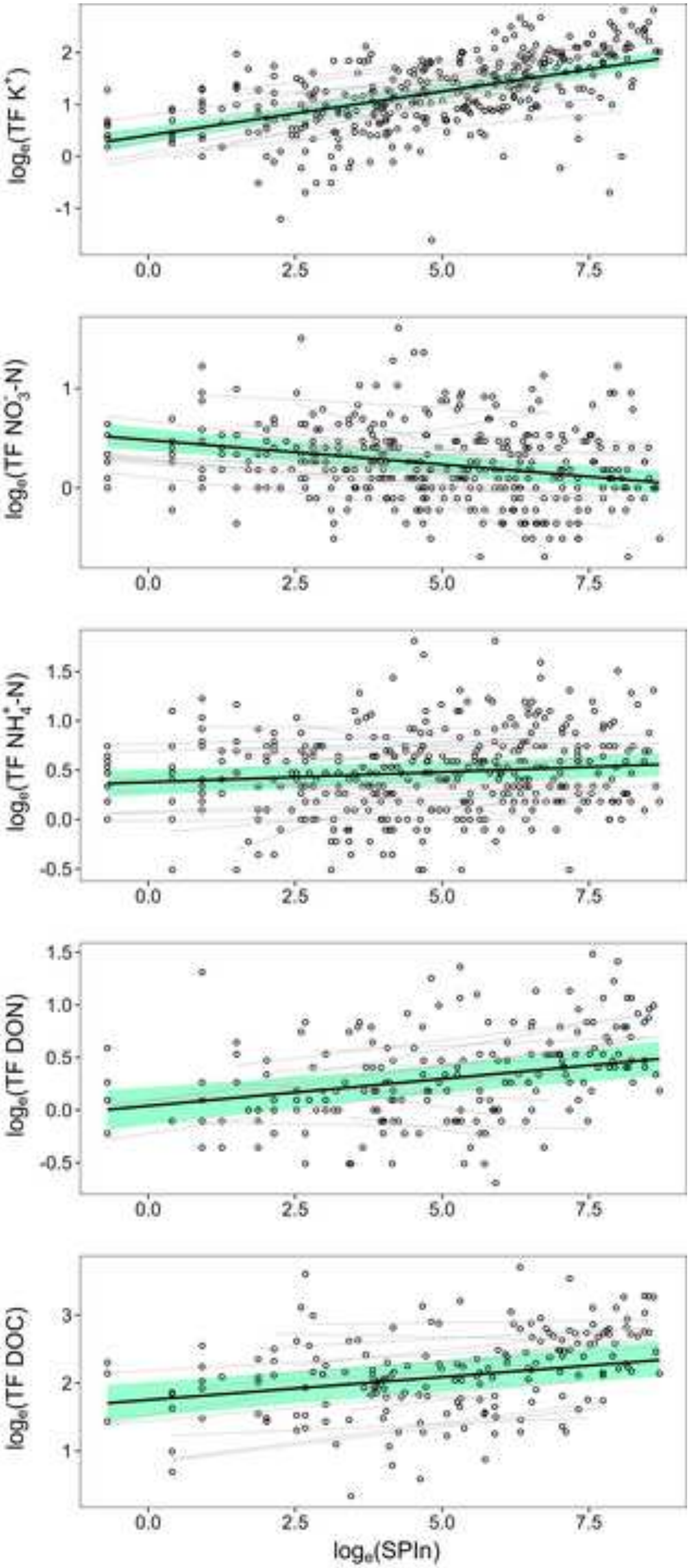

$\log_e(\text{TF K}^+)$
2
1
0
-1
0.0
2.5
5.0
7.5
$\log_e(\text{TF NO}_3^-\text{-N})$
1
0
0.0
2.5
5.0
7.5
$\log_e(\text{TF NH}_4^+\text{-N})$
1.5
1.0
0.5
0.0
-0.5
0.0
2.5
5.0
7.5
$\log_e(\text{TF DON})$
1.5
1.0
0.5
0.0
-0.5
0.0
2.5
5.0
7.5
$\log_e(\text{TF DOC})$
3
2
1
0.0
2.5
5.0
7.5
$\log_e(\text{SPIn})$

Figure 3

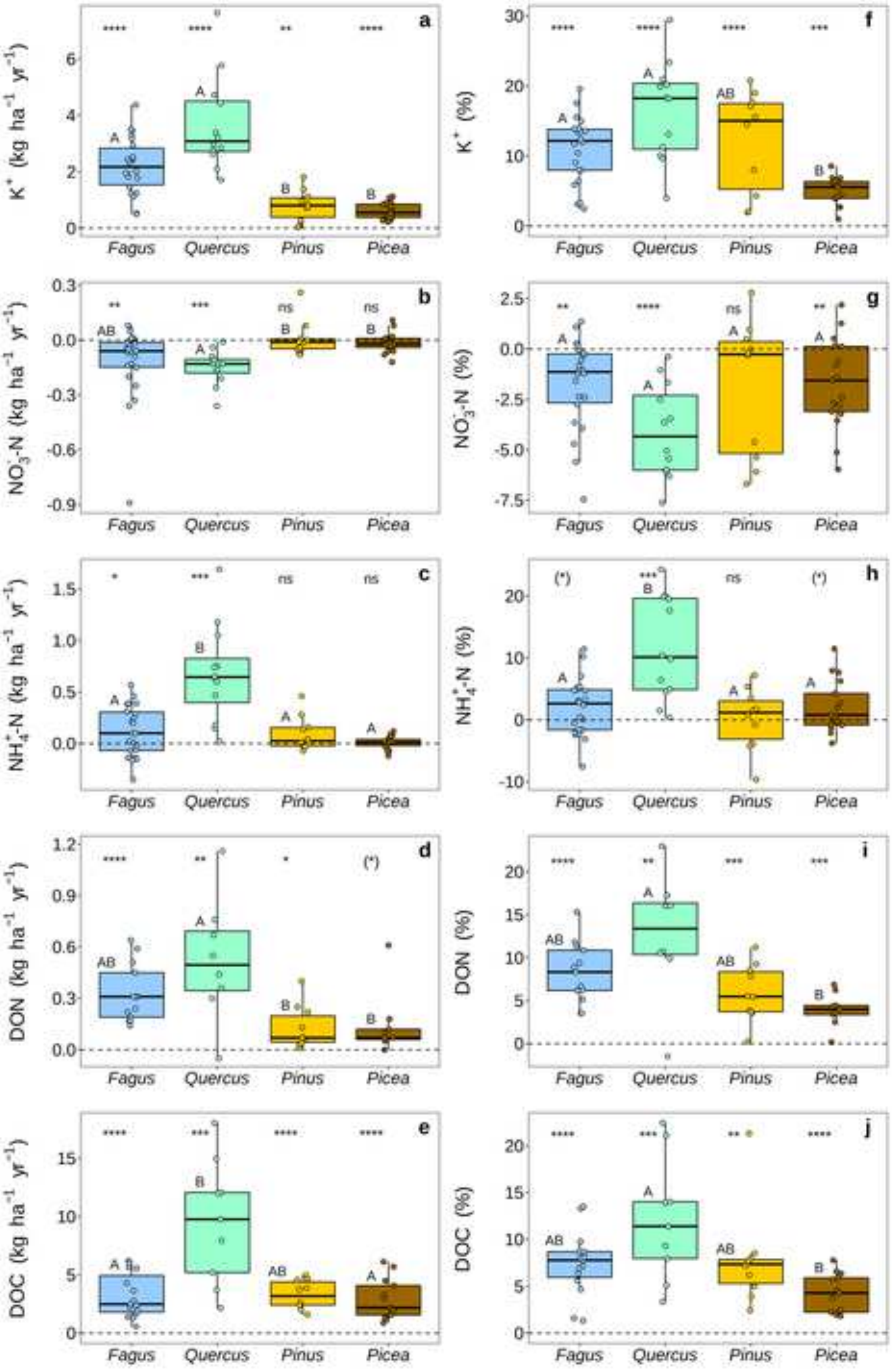
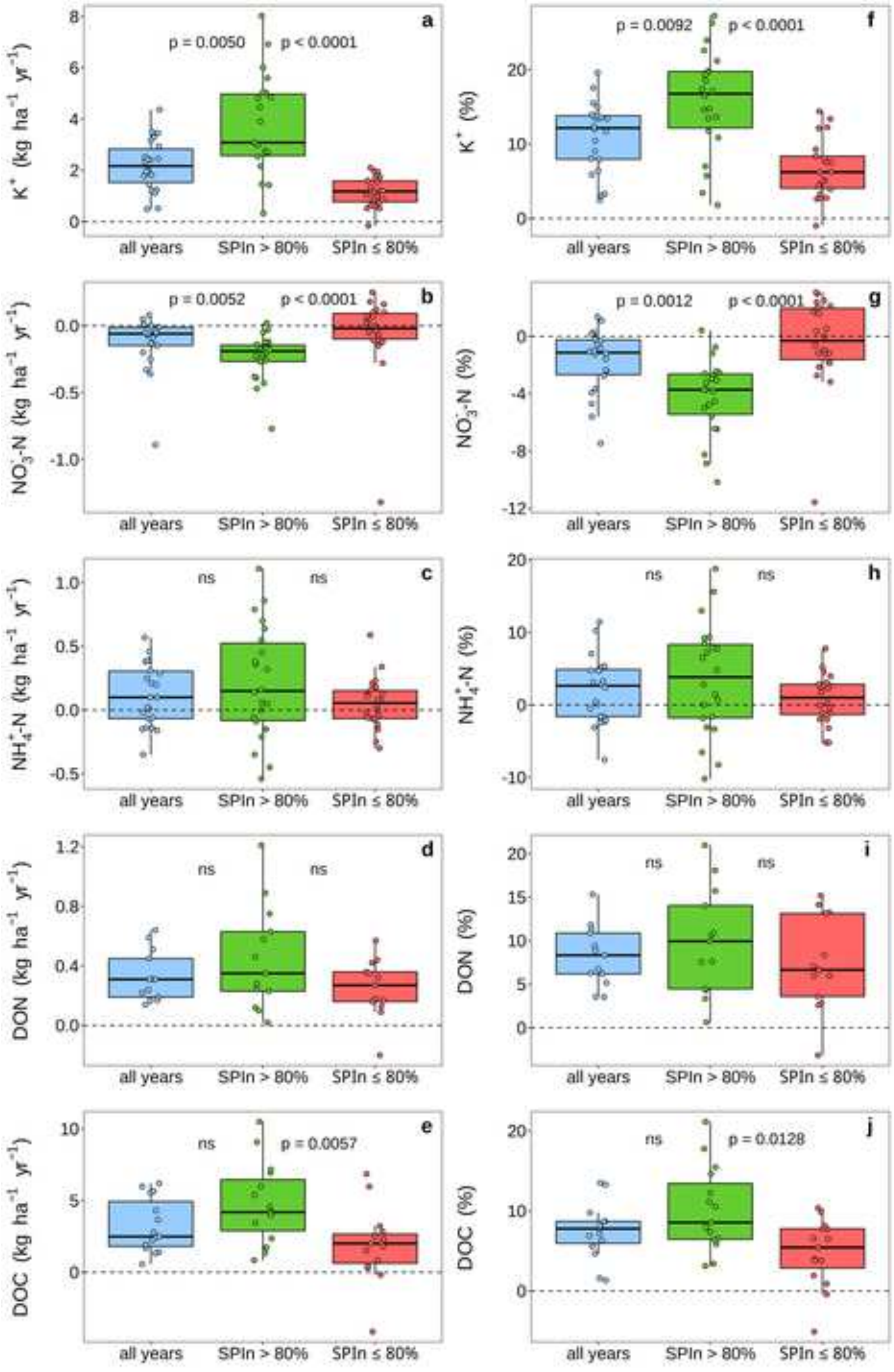

a
K^+ (kg ha^{-1} yr^{-1})
0
2
4
6
**** **** ** ****
A A B B
f
K^+ (%)
0
10
20
30
**** **** **** ***
A A AB B
b
NO_3^--N (kg ha^{-1} yr^{-1})
0.3
0.0
-0.3
-0.6
-0.9
** *** ns ns
AB A B B
g
NO_3^--N (%)
2.5
0.0
-2.5
-5.0
-7.5
** **** ns **
A A A A
c
NH_4^+-N (kg ha^{-1} yr^{-1})
1.5
1.0
0.5
0.0
* *** ns ns
A B A A
h
NH_4^+-N (%)
20
10
0
-10
(*) *** ns (*)
A B A A
d
DON (kg ha^{-1} yr^{-1})
1.2
0.9
0.6
0.3
0.0
**** ** * (*)
AB A B B
i
DON (%)
20
15
10
5
0
**** ** *** ***
AB A AB B
e
DOC (kg ha^{-1} yr^{-1})
15
10
5
0
**** *** **** ****
A B AB A
j
DOC (%)
20
15
10
5
0
**** *** ** ****
AB A AB B
Fagus
Quercus
Pinus
Picea

Figure 4


a
p = 0.0050
p < 0.0001
K^+ (kg ha^{-1} yr^{-1})
0
2
4
6
8
f
p = 0.0092
p < 0.0001
K^+ (%)
0
10
20
b
p = 0.0052
p < 0.0001
NO_3^--N (kg ha^{-1} yr^{-1})
0.0
-0.5
-1.0
g
p = 0.0012
p < 0.0001
NO_3^--N (%)
0
-4
-8
-12
c
ns
ns
NH_4^+-N (kg ha^{-1} yr^{-1})
1.0
0.5
0.0
-0.5
h
ns
ns
NH_4^+-N (%)
20
10
0
-10
d
ns
ns
DON (kg ha^{-1} yr^{-1})
1.2
0.8
0.4
0.0
i
ns
ns
DON (%)
20
15
10
5
0
e
ns
p = 0.0057
DOC (kg ha^{-1} yr^{-1})
10
5
0
j
ns
p = 0.0128
DOC (%)
20
10
0
all years
SPIn > 80%
SPIn ≤ 80%

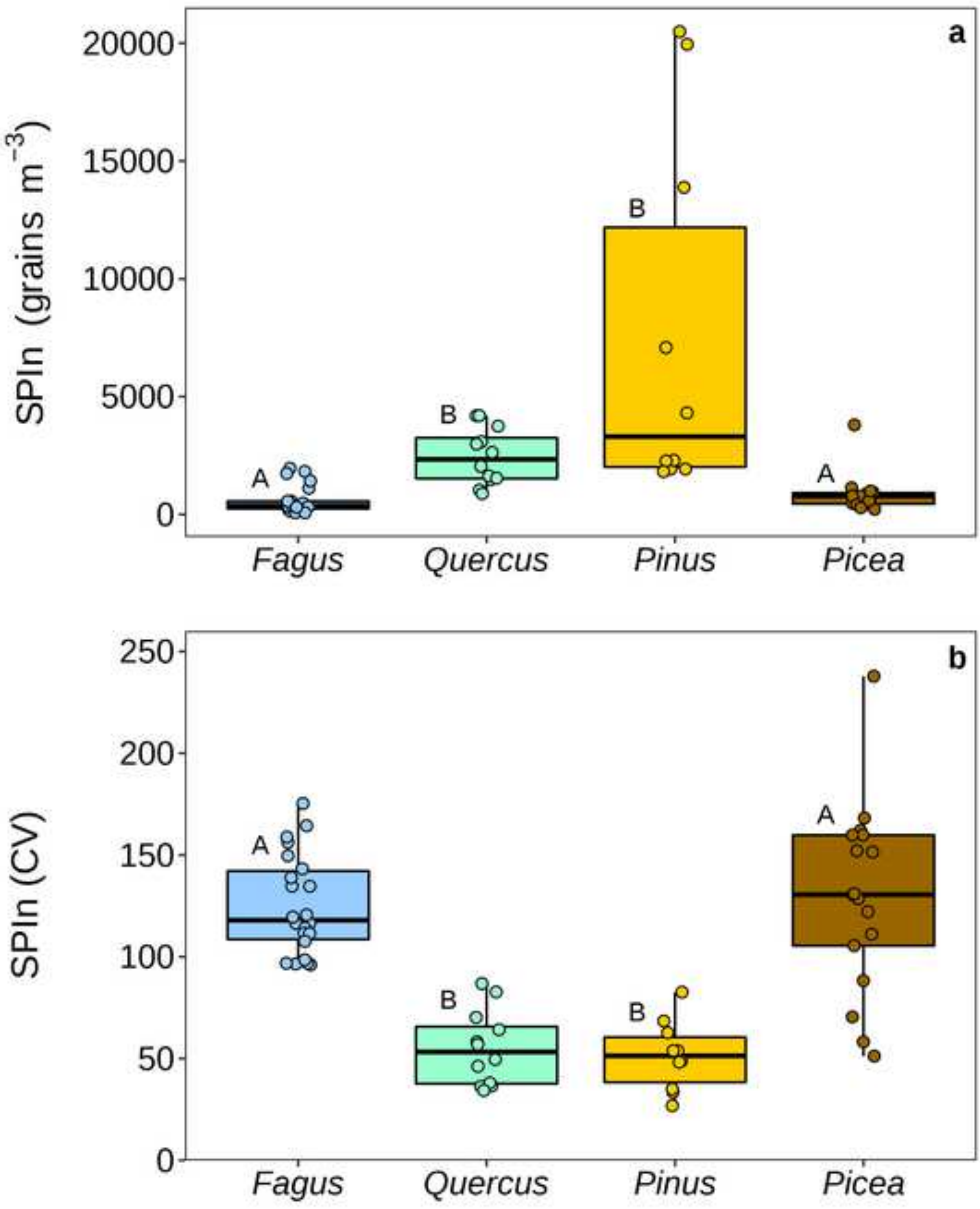
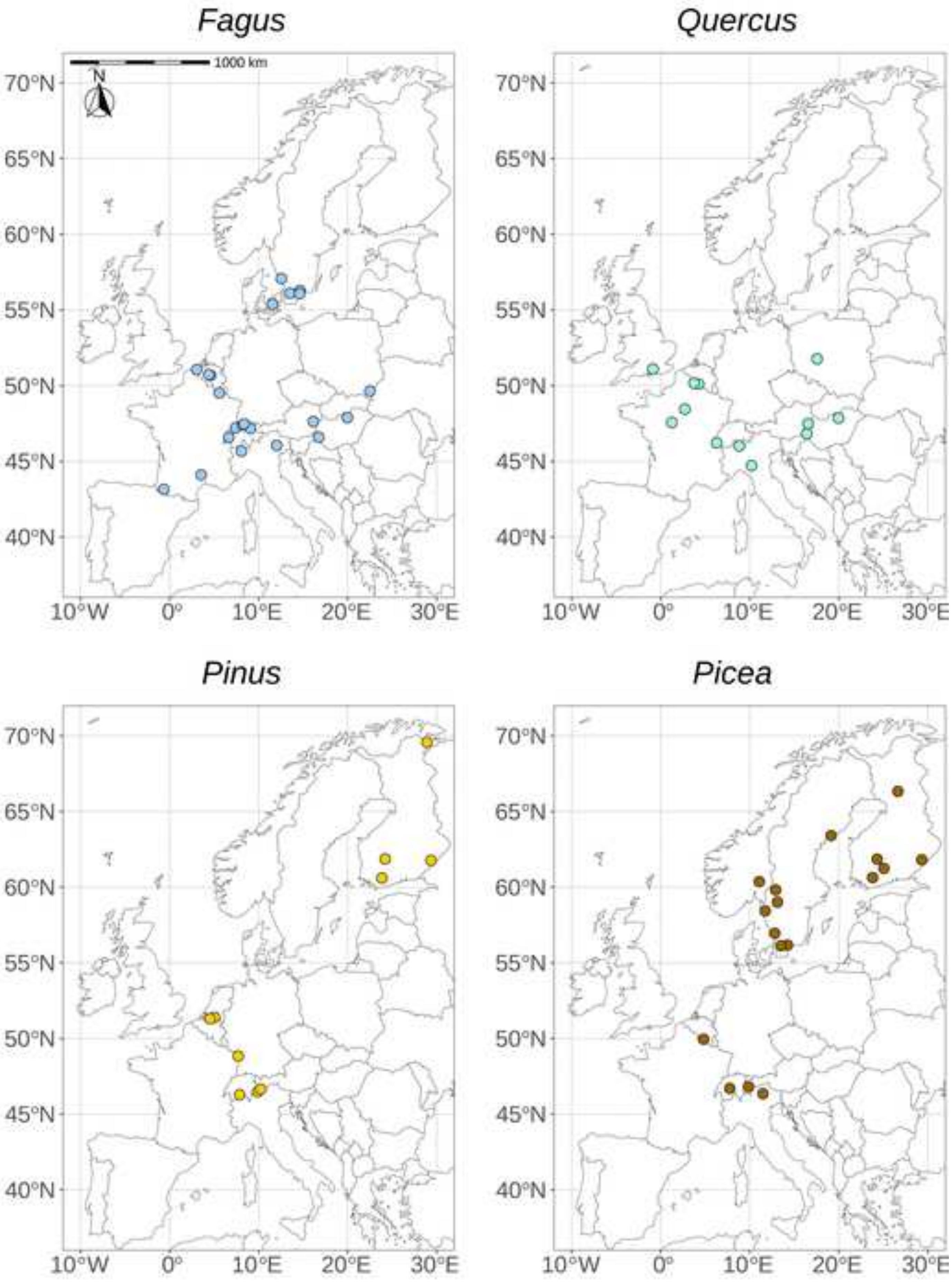
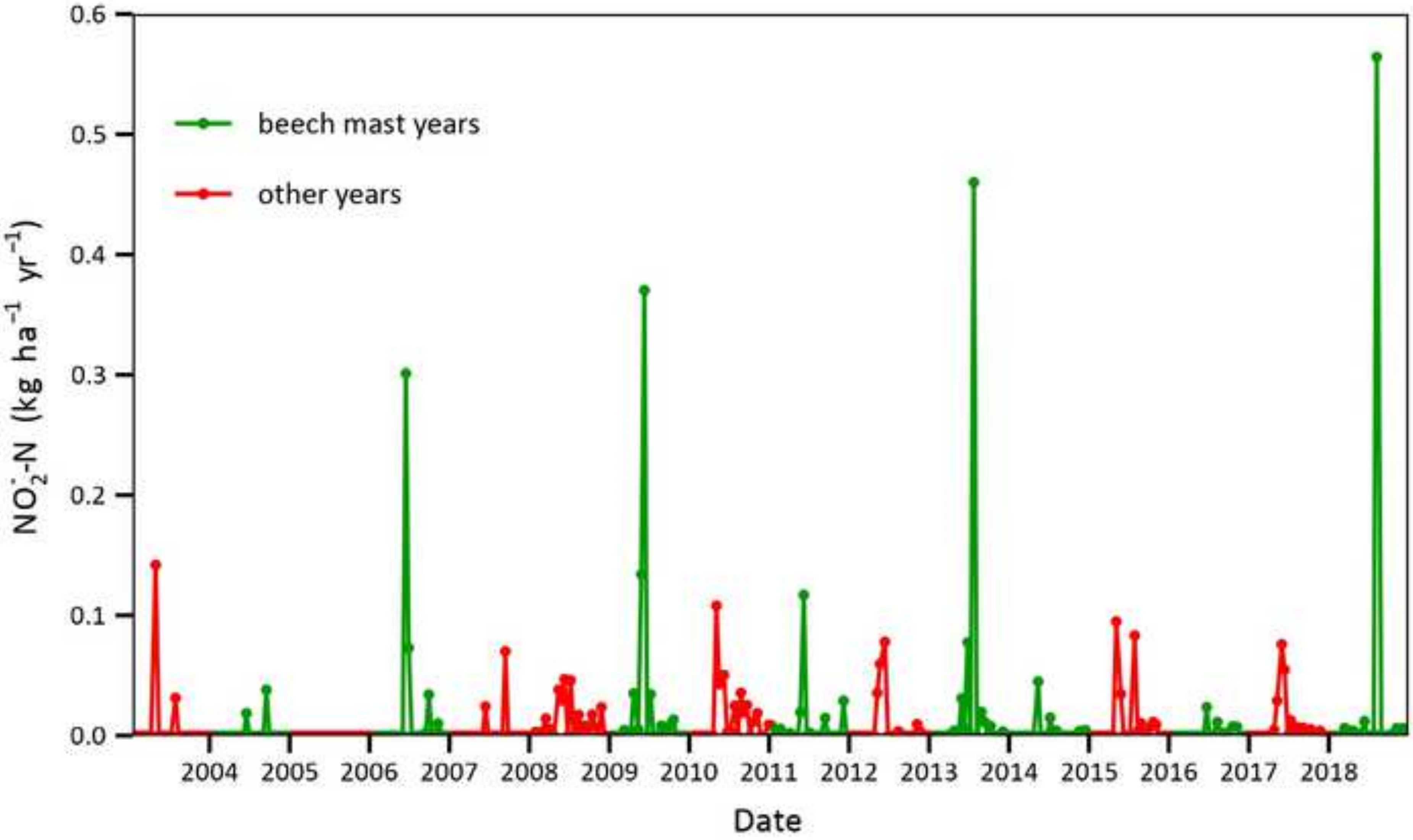

a
SPIn (grains m^{-3})
20000
15000
10000
5000
0
A
B
B
A
Fagus
Quercus
Pinus
Picea
b
SPIn (CV)
250
200
150
100
50
0
A
B
B
A
Fagus
Quercus
Pinus
Picea

Figure S1


Fagus
Quercus
Pinus
Picea
1000 km
N
70°N
65°N
60°N
55°N
50°N
45°N
40°N
10°W
0°
10°E
20°E
30°E


0.6
0.5
0.4
0.3
0.2
0.1
0.0
NO_2^--N (kg ha^{-1} yr^{-1})
beech mast years
other years
2004
2005
2006
2007
2008
2009
2010
2011
2012
2013
2014
2015
2016
2017
2018
Date