Title: Root traits and tree functional groups determine variability in root exudation and N uptake rates in mature temperate trees

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Abstract.

- Root exudation and nutrient uptake have profound impacts on soil biogeochemistry, yet quantification of these fluxes in mature trees is rare.
- We measured exudation and nitrogen (N) uptake rates of 11 tree species - gymnosperms and angiosperms associating with arbuscular mycorrhizal (AM) or ectomycorrhizal (EcM) fungi – at the Morton Arboretum, USA, to explore relationships between root traits, exudation and nitrogen uptake.
- Exudation rates were affected by phylogenetic group and mycorrhizal type, as EcM-gymnosperms exuded more carbon than the other three groups. Exudation correlated positively with specific root area and negatively with root tissue density. Nitrogen uptake did not differ between phylogenetic groups or mycorrhizal types. Root traits predicting N uptake varied with N form: root tissue density and phylogenetic group predicted ammonium uptake, whereas exudation and phylogenetic group predicted nitrate uptake. In root economic space, exudation and uptake aligned weakly with the fast side of the conservation gradient, though the two processes were not correlated.
- Our results indicate that exudation and N uptake are dynamic processes not predicted easily by species identity, plant functional groups or root traits. As such, incorporating these processes into trait frameworks and large-scale models may require ancillary information about the soil environment and whole-tree physiology.

Introduction

Global environmental changes are altering plant community composition, with poorly understood impacts on belowground processes and biogeochemical cycling (Fei et al., 2017; Jo et al., 2019). A common approach for assessing ecosystem sensitivity to change is to measure the morphological and biochemical root traits of the dominant species in a stand (Funk et al., 2017). While this approach yields valuable information about plant-soil interactions belowground, it neglects physiological processes that can link carbon (C) and nutrient cycling (Anderegg, 2022; Carmona et al., 2021; Freschet et al., 2021; Funk et al., 2017). Root exudation of C and root uptake of nitrogen (N) are often presumed to modulate soil biogeochemistry (Bardgett, 2014; McCormack et al., 2017; Weemstra et al., 2022; Wen et al., 2022), yet both processes are rarely measured in field studies. Root exudates represent 5–20% of photosynthetically-fixed C (Abramoff & Finzi, 2016; Canarini et al., 2019; Chari et al., 2024; Phillips et al., 2011), and enhanced exudation often (but not always) increases nutrient availability (Chari & Taylor, 2022; Keiluweit et al., 2015; Meier et al., 2017; Villarino et al., 2021). Moreover, exudates can enhance N mineralization (Brzostek et al., 2014; Finzi et al., 2015; Meier et al., 2017; Yin et al., 2014), thereby facilitating root N uptake and linking the two processes. Given that both processes can be measured simultaneously on the same root system (Phillips et al; 2008; Liese et al., 2018), field-based measurements of exudation and N uptake, in addition to their relationships to root traits, should lead to an improved understanding of how plant communities affect (and reflect) vital soil processes (Freschet et al., 2021; McCormack et al., 2017).

A major impediment to identifying species-specific exudation profiles that can be aligned with plant trait frameworks is the high degree of variation within and among species (Kumordzi et al., 2019; Niu et al., 2020; Weemstra et al., 2021). To circumvent this, tree species are often grouped into plant functional types (mycorrhizal association, leaf habit, phylogeny, etc.) in exudation studies. For example, ectomycorrhizal associated trees (hereafter “EcM trees”) have been reported to exude more C than arbuscular mycorrhizal trees (hereafter “AM trees”) in temperate forests (Brzostek et al., 2013; Phillips & Fahey, 2006; Yin et al., 2014), and deciduous trees have been shown to have higher exudation rates than evergreen trees in Asian temperate forests (Sun, et al., 2017a; Wang et al., 2021). Yet, how exudation rates are linked to specific root traits is

unknown, as there has been limited consensus over which root traits, if any, determine exudation (Wen et al., 2022; Williams et al., 2021; Williams & de Vries, 2020). Several studies reported positive relationships between exudation rates and specific root length (SRL; Meier et al., 2020; Tückmantel et al., 2017; Wang et al., 2021), whereas others reported strong relationships between exudation and root tissue density (RTD) and root N (Sun et al., 2017a; 2021a). Such variation indicates that exudation may depend, in part, on site-specific characteristics and plant physiological status (Neumann and Römheld, 2007). As such, investigations of functionally distinct tree species at the same site are needed to investigate whether robust relationships can be developed between root traits and exudation.

The traits that predict fine-root N uptake rates in mature trees are poorly-understood as well (Freschet et al., 2021). Hong et al. (2018) reported that mass-specific N uptake rates were positively correlated with specific root area (SRA) and SRL - two traits that reflect the nutrient-acquisition capacity of tree species (Hong et al., 2018). However, the relationship between traits and N uptake rates may also depend on the availability of different N forms (i.e., NH_4^+, NO_3^-, and amino acids) (Andersen et al., 2017) or tree species preferences (hereafter "N form preference") (Templer & Dawson, 2004). EcM trees typically prefer NH_4^+ over NO_3^- (Buscot et al., 2000; Gessler et al., 1998; Templer & Dawson, 2004; Zerihun & Bassirirad, 2001), but whether this is true for AM trees species is less clear. Soils under AM trees are commonly rich in NO_3^- (Midgley & Phillips, 2016; Mushinski et al., 2021; Phillips et al., 2013) such that AM trees may have preferences for NO_3^- vs. NH_4^+ (Andersen et al., 2017). Establishing linkages between N uptake (total and of different N forms) and more easily measurable root traits should increase our understanding of tree species effects on N cycling in the rhizosphere, leading to improved representations of these dynamics in process-based and large scale models (Braghiere et al., 2021).

Investigations of both exudation and N uptake from mature trees are rare, which has limited our understanding of how these processes affect belowground dynamics. In this study, we evaluated the effects of tree species, functional groups, and root traits on root C exudation and N uptake rates of mature trees. Given that exudation and N uptake are challenging to measure relative to root morphological traits, we investigated whether exudation and uptake align with traits that

define the root economic space (RES) that has been described previously (Bergmann et al., 2020; McCormack & Iversen, 2019; Weemstra et al., 2016; Weigelt et al., 2021; Wen et al., 2022; Yaffar et al., 2022). We chose tree species within each functional group that spanned a range of root trait space, allowing us to disentangle the effects of functional groups (e.g., AM and EcM angiosperms and gymnosperms) from root traits across the RES. Our objectives were to (1) characterize the extent to which root exudation and N uptake rates vary among tree species and across functional groups, (2) determine which root traits in the RES, if any, are closely related to root exudation and N uptake rates, and (3) build a framework for predicting root exudation and N uptake rates using readily-measurable root traits and tree functional groups. We hypothesized that (i) root exudation and N uptake rates would differ among tree species and functional groups due to differences in root traits, (ii) that both exudation and N uptake rates will align with the conservation gradient of the RES because this gradient is expected to be more closely related to root physiological activity, (iii) C exudation and N uptake would be correlated (within and among species) since the two processes have the potential to link the C and nutrient cycles in soil.

Materials and methods

Site description

This study was conducted in monoculture plots at the Morton Arboretum, Lisle, Illinois (41.81N, 88.05W). The Morton Arboretum is situated on a mixture of prairie and oak-dominated savanna and woodlands. The plots were established between 1922 and 1948 to test and study “all the timber trees of the world which might come under consideration for reforestation purposes in this part of the country” (Morton Arboretum Staff, 1929). All individuals in the monodominant plots are at least 70 years-old, mostly ranging 85 to 95 years-old (Midgley & Sims, 2020). Soils in the plots are poorly drained Alfisols that form from a thin layer of loess (0.31 m) underlain by glacial till and Mollisols that formed from alluvium (Midgley & Sims, 2020). The soil series in the plots are primarily Ozaukee silt loams and Sawmill silty clay loam (Midgley & Sims, 2020). The climate in the area has a continental climate with temperatures ranging from -6°C in January to 22□ in July and 800-1,000 mm mean annual precipitation.

Tree species in this study were selected to capture the heterogeneity in root traits among species from distinct functional groups: a phylogenetic group (angiosperm vs. gymnosperm), tree-mycorrhizal association (AM vs. EcM), and leaf habit (deciduous vs. evergreen). Within each group, species were chosen based on mean SRL and root tissue N concentration (root[N]) – the traits that were found to correlate positively with root exudation rates in previous studies (Meier et al., 2020; Sun et al., 2021; Wang et al., 2021). As such, we selected eleven species that spanned a wide range of SRL and root [N] for each group to ensure all selected species represented a diverse trait space overall (Table 1), by which phylogenetic covariations among root functional traits could be minimized while maximizing the species trait dissimilarities. Out of the 8 combinations, the only combination that was absent was evergreen-AM/EcM-angiosperms (Table 1).

Exudate collection and N uptake measurement

Fine-root exudates were collected during the growing season of 2022 (i.e., from May to July 2022) using an *in-situ* culture-based cuvette system (Phillips et al., 2008). To mitigate the impact of variable weather, 5-7 samples from distinct individuals of each tree species were collected within 2 meters of the nearest focal mature tree on the same day. The terminal roots were excavated carefully from the mineral topsoil below the organic layer. The excavated root segments were examined to ensure that the fine-root system consisted of the first three branching orders with an intact absorptive function. Organic matter and soil particles adhering to the root system were removed with DDI water with extreme caution while keeping the roots moist with wet paper towels. In cases where the distal fine roots were damaged or broken off, samples were discarded, and a new sample was prepared. The intact root systems were placed in cuvettes filled with sterile, C-free glass beads (>2mm diameter). The root systems with glass beads were flushed three times with C-free nutrient solution (0.5 mM NH_4NO_3, 0.1 mM KH_2PO_4, 0.2 mM K_2SO_4, 0.15 mM $MgSO_4$, 0.3 mM $CaCl_2$) to ensure the root segments and glass beads were well-mixed. To ensure the same amount of solution in the cuvettes, we added 15mL of nutrient trap solution in the field using a bottle-top dispenser. The cuvette was covered in aluminum foil to allow the root system to equilibrate with the cuvette environment. The same procedure was applied to the control (i.e., no root) cuvette with the same glass beads and nutrient solution. The cuvettes were placed at the exact excavated area and covered with soils and organic matter and

incubated for approximately 24 hrs. After the one-day incubation period, the sampled roots with the cuvette were clipped with care and brought to the laboratory for analysis. Each cuvette was flushed with 15mL of the working nutrient solution three times for the complete removal of accumulated exudates in the cuvette. All solutions were filtered immediately through a sterile 0.22 µm syringe filter (Millex-GV 0.22µm PVDF 33mm Gamma Sterilized 50/Pk, Millipore Co., Billerica, MA), and remained refrigerated at 4°C until analyses (< 24 h). All samples were analyzed for non-particulate organic C on a TOC analyzer (Shimadzu Scientific Instruments, Columbia, MD), while inorganic NH_4^+ and NO_3^- were analyzed on a microplate (Sterile 96-well microplate: Fisher #08-772-7). The total mass-specific exudate rate was calculated with the total C captured from the trap solution minus the total C flushed from the root-free control cuvettes divided by root biomass and day (mg C * g_{root}^{-1} * day^{-1}). Similarly, the net inorganic N uptake rate was calculated by the total change of inorganic NH_4^+ and NO_3^- concentration with respect to the control sample divided by root biomass and day (mg N * g_{root}^{-1} * day^{-1}).

Root morphological and chemical traits

Roots originally placed in the cuvette were carefully collected from the cuvette, washed, and stored at 4°C until processing. Fine-root morphology was analyzed for all the fine roots with a transparent flat-bed scanner and the WinRHIZO program (Regent Instruments, Quebec, QC, Canada). Scans were collected at a resolution of 600 dpi. All root samples were dried at 65°C for at least 48 h, and the dried root biomass was used for root trait calculations. Specific root length (SRL, in m g^{-1} : the length of the fine roots divided by the corresponding root dry weight), Specific root area (SRA, in cm^2 g^{-1} : the area of the fine roots divided by the corresponding root dry weight), root tissue density (RTD, in g cm^{-3} : root dry weight divided by root volume), root branching intensity (BI, in the number of tips per total fine-root length), root diameter (diameter, in cm), and root N concentration (root[N], in % : root N content per dry mass) were calculated from the results from WinRHIZO.

Ecosystem-scale fluxes

Mass-specific rates of root exudation and N uptake (mgC g_{root}^{-1} day^{-1}; mgN g_{root}^{-1} day^{-1}) were scaled to estimate yearly root exudate C and N uptake fluxes per soils surface area (F_{ex} : kgC m^{-2} y^{-1}; F_N : kgN m^{-2} y^{-1}), following the approach by Chari et al. (2024). The fluxes were calculated

by multiplying mass-specific root exudation rate (SER; μgC g_{root}^{-1} day^{-1}), fine root biomass (FRB; kg m^2), and GS (the days of growing season) (Chari et al., 2024). To measure mean root biomass per soil surface area for each species, six soil cores were collected randomly at each species-plot using 5cm×15cm (diameter×height) soil core in June 2022. The collected soils were homogenized using 2mm sieve and immediately processed to three fractions with different depth profiles (0 - 5 cm, 5 - 10 cm, and 10 - 15 cm). The fine roots of focal tree species were sorted by hand and dried at 65°C for at least 48 h. Fine-root biomass in each soil-depth profile was measured by summing fine-root biomass per m^2. The average growing season length was assumed to be 185 days (Apr. 17th - Oct. 20th) for Lisle, Illinois reported by Illinois State Climatologist (https://stateclimatologist.web.illinois.edu/data/frost-dates/). The relationships between yearly C and N fluxes with exudation and N uptake rates were examined to assess the role of root biomass production in C and N fluxes. Given that the active growing season for roots varies with species and is likely longer than for aboveground organs (McCormack et al., 2014), the assumed growing season may underestimate the annual total exudates.

Mass-specific exudation rates in other temperate forests

To compare exudation rates and their variabilities (coefficient of variation (CV)) of tree species from our study with those from previous studies, we collected data from 18 studies measuring root exudation rates of temperate trees *in situ*, using both the metadata set by Chari et al. (2024) and WebPlotDigitizer (https://automeris.io/) to extract the data from the studies that were not included in Chari et al. (2024). We chose the peer-reviewed studies that (1) were conducted *in situ*, (2) measured mass-specific exudation rates of mature trees, and (3) used the cuvette-based methods by Phillips et al. (2008).

Statistical analyses

We calculated the interspecific differences in min/max, mean and median for all root traits and processes. Blomberg's *K* and their p-values were reported for phylogenetic signals on each root trait using the R package; ape, V.PhyloMaker, phytools, and phylosignal. Interspecies variation of exudation and net N uptake rates was tested by pairwise comparisons after an analysis of variance (ANOVA) using Tukey's Honest Significant Difference (HSD) test. Root traits and mass-specific exudation rates were natural-log-transformed to ensure normality and improve

homogeneity of variance. The variation of mass-specific exudation and net N uptake rates among tree functional groups were tested by predicting exudation or N uptake rates of tree species nested in mycorrhizal types and phylogenetic groups while setting species (species-specific-plot) as a random factor by a linear mixed-effects model using restricted maximum likelihood (‘lme4::lmer’ function via REML). We tested the significance of each nested group in the model after accounting for all other groups by Type III ANOVA with Satterthwaite's Method using ‘lme4::anova’. Pairwise trait relationships between exudation/N uptake rates and root traits were also performed using Pearson’s correlations at the individual tree level. Variation partitioning analyses were performed to quantify the contributions of root traits - inter- vs. intraspecific variation of exudation - using the ‘varpart’ function in the VEGAN package. To assess the multi-dimensional trait space and its consistency with a previously determined root economic space (RES) of fine-root traits, we used principal components analysis (PCA) and a multi-step approach (Weigelt et al., 2023). We first generated the RES with four traits that most commonly represent the RES (RTD, root[N], SRL, Diameter), and then generated the same RES with additional traits (SRA, BI, net N uptake rates, and exudation rates) using princomp () with standardized PCA. After checking whether group dispersion is homogeneous, using betadisper::vegan and anova, the differences of the trait syndrome among plant groups (i.e. mycorrhizal types, phylogenetic groups and leaf habits) were tested by Permutational Multivariate Analysis of Variance (PERMANOVA) using bray distance of the adonis function in the VEGAN package. The best predictors for exudation and net N uptake rates were selected via both stepwise multiple regressions using the ‘step’ function in the lmerTest package and mixed-effect model by a nested model comparison approach using the ‘lmer’ function via REML in the lme4 package. In mixed-effect models, the procedure considered species (monodominant-species-plot) as a random effect, considering that each plot comprised the same tree species with a similar age group (85-95 years). The days of data collection campaigns were chosen and blocked randomly by the plot. The assumptions of the selected models were tested by checking residual distribution, unbiasedness, and homoscedasticity. For the best predictors for exudation rates, only the results from mixed-effect model were documented given that species-plots separated exudation rates significantly. By contrast, given the insignificance of species in N uptake, we selected the best predictors by the stepwise multiple regression. All statistical analyses were performed using R v.3.5.3 (R Core Team, 2017).

Results

Carbon exudation and net N uptake rates of tree species

Most root traits varied more among species than within species, though exudation and N uptake rates did not follow this pattern. Rather, exudation and N uptake exhibited greater intraspecific variation than interspecific variation (N uptake, 92% *vs.* 8%; exudation, 77% *vs.* 23%, respectively; Fig. 1). Tree species varied in their exudation rates with species alone, accounting for 23% of the total variation of exudation rates ($F_{(10, 49)} = 2.6$; $p = 0.01$). *Larix gmelinii* exhibited significantly higher rates of root exudation than *Chamaecyparis pisifera* ($p=0.03$) and *Carya ovata* ($p<0.01$) (Fig. 2a: Table 2). The mean exudation rate of *Larix gmelinii* (3.82 mg C g_{root}^{-1} * day^{-1}) was more than two-fold greater than the second-greatest mean exudation rate of *Picea abies* (1.52 mg C g_{root}^{-1} * day^{-1}) (Fig. 2a: Table 2). In contrast, N uptake rates did not differ significantly among tree species ($F_{(10, 49)} = 1.5$; $p = 0.17$). Most species showed positive N uptake, but *Juniperus chinensis* exhibited a net negative uptake rate of -133.28 mg N g_{root}^{-1} * day^{-1}. However, species did vary in their ratio of net NH_4^+ and NO_3^- uptake rates (NH_4^+:NO_3^-) ($F_{(10, 49)} = 2.6$; $p = 0.02$) (Fig. S4; Table 4). On average, with the exception of *Picea abies*, most species exhibited higher NH_4^+ uptake rates than NO_3^-, as eight of all species (except *Picea abies*, *Platanus occidentalis* and *Tsuga canadensis*) showed two-folds higher NH_4^+ uptake compared to NO_3^- (Fig. S4; Table 3). Net NH_4^+ uptake rates showed marginally significant species differences ($F_{(10, 49)} = 1.8$; $p = 0.08$), whereas species did not vary in net NO_3^- uptake ($F_{(10, 49)} = 1.4$; $p > 0.1$) (Fig. S4).

Exudation rates varied with phylogenetic group ($F_{(1, 60)} = 6.2$; $p = 0.02$; Table 4). On average, EcM gymnosperms exuded 235% more C than EcM angiosperms and 152% more C than AM gymnosperms (Fig. 3a; Table 2) in tandem with a significant mycorrhizal × phylogenetic group interaction; ($F_{(1, 60)} = 8.8$, $p = 0.004$; Table 4). However, there were no significant differences between EcM and AM angiosperms (Table 4). By contrast, N uptake rates did not vary with any combination of functional groups (Table 4). However, largely driven by EcM gymnosperms, the NH_4^+:NO_3^- did differ among mycorrhizal type ($F_{(1, 60)} = 6.6$, $p = 0.02$; Table 4; Fig S4b). Unlike

exudation rates, there were no significant interactions among tree functional groups for N uptake (Table 4).

Trait correlations and root economic spectrum (RES)

Trait correlations

Most root morphological (i.e., SRA, SRL, diameter, RTD, and BI) and chemical traits (i.e., Root[N]) were correlated with one another at the individual tree level (Fig. S1). Root diameter negatively correlated with RTD, SRL, and BI significantly (Fig. S1). Root [N] correlated negatively with RTD and BI ($p<0.001$) and positively with root diameter and SRA ($p<0.05$ and $p<0.001$, respectively; Fig. S1). Notably, root exudation correlated positively with SRA and negatively with RTD ($p<0.05$; Fig. S1), while net N uptake did not correlate with any other root traits including root exudation. With the exception of branching intensity (Blomberg's $K = 0.7$, P = 0.01; Table S2), we did not detect a strong phylogenetic signal in interspecific root trait variation (Blomberg's K values < 0.7; $p > 0.05$ for 7 traits).

Multidimensional root economic space (RES)

Root functional traits generated a multidimensional RES in which the directions and relationships among vectors were consistent with previous studies (Wen et al., 2021; Bergmann et al., 2020) (Fig. 4a). Two axes of the RES generated with four core traits (RTD-root[N] vs. SRL-Diameter) explained 88% of the total variation (Fig. S6a) while the two principal axes of the RES generated with six core traits (RTD, root[N], SRL, Diameter, SRA, and BI) and exudation/N uptake rates explained 64% of the total variation (Fig. 4a; Table S8). The axis generated by RTD-root[N] was closely mapped onto the PC1 reflecting the conservation gradient, while the axis generated by SRL-Diameter was loaded closely onto the PC2 representing the collaboration gradient (Bergmann et al., 2020; Fig. 4a). The RES indicated that both N uptake and exudation rates were loaded onto the faster side of the conservation gradient (PC1) and exhibited a strong correlation with SRA (Fig. 4b). However, both N uptake and exudation rates showed a weak association the RES generated by the PC1 and collaboration gradient (PC2). Exudation was better explained by PC3 which accounted for 14% of the total variation, while N uptake rates was best explained by PC4 which captured and additional 12% of the total variation

(Fig 4c; Table S8). Interestingly, AM angiosperms exhibited a wider range of variability in their trait coordination than EcM angiosperms and gymnosperms (Fig. S6b). PERMANOVA analyses using Bray-Curtis distances verified the dissimilarities in trait syndromes among combinations of tree functional groups, corroborating that combining mycorrhizal type and phylogenetic group could significantly account for trait coordination differences ($F_{(3, 59)} = 7.0$, $p = 0.003$) (Fig. S6b).

Mixed-effect models for predicting exudation and N uptake rates

The best mixed-effect model for predicting exudation contained SRA and the interaction term between the phylogenetic group and mycorrhizal types ($p<0.01$; Table S4). Moreover, a mixed-effect model that contained SRL and the interaction term also significantly predicted exudation rates ($p<0.01$; Table S4), while SRL and SRA were significantly correlated ($r=0.81$, $p<0.001$; Fig. S1). In contrast, in the multiple regression models for net N uptake rates, RTD was selected to be the best predictor for both NH_4^+ (phylogenetic group: $p<0.001$, RTD: $p=0.02$) and the total inorganic N uptake (NH_4^+ and NO_3^- combined; phylogenetic group: $p=0.01$, RTD: $p=0.04$; Table S5). In predicting NO_3^- uptake, exudation rates ($p=0.04$) and the phylogenetic group were the best predictors, while the phylogenetic group alone was marginally significant ($p=0.06$; Table S5).

Ecosystem-scale fluxes

To compare interspecific exudation (C) and N uptake fluxes, we incorporated root biomass per m^2 into exudation and N uptake rates. Generally, tree species enhanced their C and N fluxes by increasing root biomass and the rates of exudation and N uptake. Both exudation and N uptake rates significantly accounted for more than half of the variation in the plot-level fluxes (C fluxes: $R^2 = 0.54$; $p=0.01$; N fluxes: : $R^2 = 0.4$; $p=0.04$) (Fig. S7a; S7b). The relationship between N uptake rate and root density was significant ($R^2 = 0.40$, $p = 0.03$), while the relationship of exudation rates with root density was not significant ($R^2 = 0.11$, $p > 0.05$) (data not shown). Notably, exudation rates could significantly explain the estimated fluxes only among EcM trees, but not AM trees (EcM: $R^2=0.84$, $p=0.03$; AM: $R^2=0.05$, $p =0.68$; data not shown). Driven by considerably higher exudation rates, *Larix gmelinii* showed the greatest mean exudation flux, followed by *Picea abies* (Fig. S7a). In contrast, the high root biomass of *Acer saccharum* resulted in a substantially high annual exudation flux and the highest N uptake (Fig. S7a; S7b).

Discussion

Our study aimed to identify key drivers of mass-specific root exudation and N uptake rates in mature field-grown trees. We hypothesized that exudation and N uptake would 1) differ among tree species and functional groups, 2) align with the conservation axis of the RES, and 3) correlate to one another (within and among species). We found partial support for our first hypothesis, as exudation rates varied more within species than among species, but only EcM gymnosperms had statistically greater exudation rates. Likewise, N uptake rates showed little variation among species or functional groups. In partial support of our second hypothesis, both exudation and N uptake rates aligned with the 'fast' side of the conservative gradient, yet they were weakly associated with other root traits. Finally, we found only limited support for our third hypothesis, as we found no correlation between exudation and N uptake, suggesting that distinct mechanisms may govern the two processes. Collectively, our study indicates that root physiological processes are complex and cannot be fully explained by species identity or root traits alone. Instead, a combined consideration of these factors offers a more accurate prediction of fine-root physiological functioning.

Exudation rates for mature, field-grown trees are rare, as all methods used to measure exudation introduce some artifacts; our study is no exception. While the root cuvette method of Phillips et al. (2008) has been used for forest studies (Chari et al., 2024), important methodological questions remain about how to minimize damage to root systems before placement into cuvettes, the chemistry and sterility of the trapping solution, the duration of the equilibrium period and the duration of the collection period (Oburger & Jones, 2018). Moreover, the high degree of spatio-temporal variation in exudation (Jacoby et al., 2017; Jiang et al., 2021; Yang et al., 2020) affects how many samples are needed to draw inferences about species-specific patterns and trait-exudation correlations. In this study, high intraspecific variability in exudation and N uptake (Fig. 1) may have precluded us from detecting species or functional group differences, and our ability to detect trait-exudation relationships was also limited by the fact that many root traits showed relatively low intraspecific variation, whereas exudation rates were highly variable within a species. Nevertheless, given that we made efforts to minimize this variability (e.g., by selecting

tree species grown in a common soil, by sampling roots at the same time of year, by applying the same methodology to all individual trees), our estimates are robust and provide novel information regarding physiological processes infield-grown and mature trees.

Functional groups and traits partially account for variation in exudation

EcM gymnosperms generally had the highest exudation rates (two-fold higher than other groups), though the reasons remain elusive. High exudation rates have been reported for other EcM gymnosperms in temperate forests (Abramoff & Finzi, 2016; Brunn et al., 2022; Jiang et al., 2021; Yang et al., 2020; Zhang et al., 2016) and under controlled conditions (Sell et al., 2022; H. Yin et al., 2013). One of the highest exuders in our study, *Larix gmelinii*, has been reported to have elevated exudation rates in a boreal forest as well (Yin et al., 2023). One factor that may contribute to elevated exudation rates in EcM gymnosperms is their greater reliance on mycorrhizal fungi (relative to EcM angiosperms), as EcM gymnosperms tend to possess large diameter short roots that are well-colonized by mycorrhizal fungi (Cheng et al., 2016; Comas & Eissenstat, 2009; Ma et al., 2018). EcM fungi are also a sink for plant-derived C (Prescott et al., 2020), yet they too can exude organic C (e.g., oxalic acid) resulting in an additional source of C in the cuvettes (Ahonen□Jonnarth et al., 2000; Sun et al., 1999; Van Schöll et al., 2008).

A second factor that could explain greater exudation in EcM gymnosperms is mycorrhizal-root leakiness. Exudation is believed to follow source-sink gradients and if so, it is possible that a portion of the C being provisioned to EcM tips is leaked during transport (Farrar et al., 2003). Whether mycorrhizal-roots of EcM gymnosperms differ from EcM angiosperms in leakiness is unclear. In a global synthesis, Hawkins et al. (2023) reported that C allocation to EcM mycelium biomass was ~2.5 times greater in broad-leaved trees (mostly angiosperms) than needle-leaf trees (mostly gymnosperms). If C allocated to mycelial growth represents a trade-off with C released as exudation, this could explain the greater exudation in EcM gymnosperms. However, if the magnitude of leakiness is determined by the strength of the C sink (like the “hole in the pipe model” for nitrous oxide gas in Firestone and Davidson (1991)), greater exudation in EcM gymnosperms would result from greater allocation to mycelium. Indeed, some of the most striking examples of prodigious EcM mycelium come from studies of EcM gymnosperms like

Pinus (Anderson & Cairney, 2007). Given that the Hawkins et al. (2023) synthesis did not control for biome, it is possible that reports of lesser C allocation to mycelium in EcM gymnosperms was confounded by climate (e.g. if boreal forest needle-leaf trees were compared to temperate/tropical forest broad-leafed trees). Moreover, when we compared exudation between EcM gymnosperms and angiosperms using data from the synthesis of Chari et al. (Fig S7a), no differences were apparent. This indicates that functional differences in exudation depend on many factors – plant, fungal identity, site, climate, etc. - and that our reports of greater exudation in EcM gymnosperms may not be generalizable.

While some root traits were correlated with exudation, the relationships were generally weak. Exudation rates were correlated negatively with RTD and positively with SRA (Lv et al., 2023; Sun et al., 2021a). Such relationships may be attributed to the interplay of root area, tissue permeability, and diffusion strength - all of which can influence exudation rates (Farrar et al., 2003; Jones et al., 2009). Given that a higher RTD often corresponds to reduced root leakiness (Farrar et al., 2003), roots possessing less dense tissues and greater surface areas are more likely to exude carbon. Consistent with our findings, recent studies in cool-temperate and subtropical forests have also reported significant relationships between exudation rates and RTD, root [N], and SRA, but not SRL (Akatsuki & Makita, 2020; L. Sun et al., 2021a). Other studies have reported correlations between SRL and exudation rates (Meier et al., 2020; Phillips et al., 2008; Tückmantel et al., 2017; Wang et al., 2021). Both SRL and SRA reflect acquisitive root behavior (Eissenstat, 1991; Eissenstat & Yanai, 1997; Lv et al., 2023; Weemstra et al., 2016), and our results indicate a weak but positive relationship with this axis, possibly suggesting that root exudation could be an alternative nutrient acquisition strategy (Wen et al., 2022).

Surprisingly, exudation and uptake did not align strongly with either of the two main axes of the RES: the ‘conservation’ gradient generated by RTD and root [N] (e.g., ‘fast’ vs. ‘slow’) or the ‘collaboration’ gradient generated by SRL and diameter (e.g., ‘collaboration’ vs. ‘do-it-yourself’). Rather, additional axes/principal components were needed to explain exudation and N uptake patterns. The RES simplifies trait space by collapsing trait variation (Bergmann et al., 2020). However, these two gradients are not the only aspects of plant trait variation. Weigelt et al. (2021) recently proposed that a third axis – reflecting plant height and rooting depth – to capture the

diversity of form and function in plants. Whether exudation and nutrient uptake align on this axis is unknown, but this could be a fruitful line of inquiry in future studies. Moreover, it is possible that exudation and uptake are sufficiently dynamic processes controlled by myriad processes (e.g., photosynthesis, plant C allocation, microbial consumption, plant nutritional status, etc.), and thus relationships between exudation and relatively static root traits may be nearly impossible to detect.

N uptake rate has little association with tree traits but depends on N form

N uptake rates showed little correlation with root traits and did not differ significantly among species (or functional groups) owing to high intraspecific variation. This finding was consistent with a global analysis of root traits that found no correlation between N uptake rates and root traits (Ma et al., 2018). With respect to N form preferences, our findings are also consistent with other studies that have reported higher rates of NH_4^+ uptake compared to NO_3^- uptake (Gessler et al., 1998; Marschner et al., 1991; Templer & Dawson, 2004; Zerihun & Bassirirad, 2001). This preference is believed to result from the greater absorption potential of NH_4^+ at low concentrations (Britto & Kronzucker, 2013; Eshel & Beeckman, 2013), the lower energetic cost of NH_4^+ assimilation by plants (Britto & Kronzucker, 2013), and the greater abundance of NH_4^+ in mature forest soils (Kronzucker et al., 1997; Rice & Pancholy, 1972).

Although physiological processes for N form preference have been well-documented, the interaction between species identity (e.g., species, functional groups) and soil conditions remains unresolved (Britto & Kronzucker, 2013). We found that EcM gymnosperms differed from other groups in N form preference (Fig. S4a; S4b); AM gymnosperms (*J.chinensis*, *T.disticum* and *C.pisifera*) showed a higher NH_4^+ preference than EcM evergreen gymnosperms (*T.canadensis* and *P.abies*), while *Picea abies* took up more NO_3^- (Fig. S4a; S4c). Contrary to our findings, in an European forest *Picea abies* did not exhibit higher NO_3^- uptake but showed 3-4 times greater NH_4^+ uptake (Marschner et al., 1991). In a ^{15}N labeling study with seedlings from four tree species in the Catskill Mountain soil, NY, both *Acer saccharum* and *Tsuga canadensis* strongly preferred NH_4^+, while *Fagus grandifolia* (an EcM angiosperm) took up more NO_3^- than NH_4^+ (Templer & Dawson, 2004). While we also observed a high NH_4^+ preference of *Acer saccharum*

(Fig. S4c), the observed low NH_4^+ preference of *Tsuga canadensis* was inconsistent with the results from Templer and Dawson (2004). Of course, like the issue with exudation, it is difficult to separate the species effects on N uptake from the mycorrhizal effects, and differences among studies (and among tree species) could relate to variability in fungal taxa. Additionally, some tree and fungal species have preferences for amino acids, which we did not measure yet represent a non-trivial amount of N uptake in temperate forests (Finzi & Berthrong, 2005). Given that ^{15}N tracer studies are the preferred method for quantifying N uptake and N form preferences in forest soils (Templer et al., 2012), adding ^{15}N-enriched ammonium, nitrate and amino acids to incubating roots (i.e., inside the cuvettes) may help resolve uncertainty in N uptake and N form preferences between trees and mycorrhizal fungi.

Exudation and N uptake are independent and dynamic

We found no support for our third hypothesis - that exudation and N uptake were correlated. Rather, our results indicate that exudation and N uptake are dynamic (Fig. S8), independent processes (Fig. S3). Exudates can influence N uptake indirectly by stimulating microbial growth, turnover and enzymatic activity (Brzostek et al., 2013; Phillips et al., 2011; Phillips & Fahey, 2006; Yin et al., 2014), thereby affecting diffusion in the rhizosphere (Farrar et al., 2003). While some studies have linked exudation and N uptake (Liese et al., 2018; Meier et al., 2017; Sun et al., 2021b; Yin et al., 2013), the two processes are rarely measured at the same time. In our study, the time lag between exudate release and microbial N cycling may have limited our ability to detect a relationship between the two processes during the short (24h) incubation. Additionally, our cuvette environment was unlike an actual rhizosphere in that it was devoid of active populations of microbes, soil organic matter and microbial grazers, which likely weakened the potential for exudates effects on N uptake (Jackson et al., 2008).

Scaling exudation and N uptake

Our scaled exudation and uptake measurements suggest that trees may alter the soil environment by adjusting root morphologies, building root biomass, and up-regulating physiological processes like exudation. *Acer saccharum* achieved relatively high plot-level C efflux compared

to most other plots solely through the maintenance of higher standing root mass while it augmented N influx by increasing both root mass and uptake rates. In contrast, EcM gymnosperms such as *Picea abies* and *Larix gmelinii* increased plot-level C efflux primarily by enhancing rates of exudation, without a corresponding increase in N uptake. These results collectively indicate that both tree traits and soil edaphic factors can differ significantly across species and environments, and such differences may influence the rate and magnitude of physiological functions across scales.

Conclusion

We demonstrate that root exudation and N uptake are complex processes that cannot be fully explained by species identity, functional groups or individual root traits. Although we revealed some relationships between root traits and exudation (and traits and N uptake), high intraspecific variation in the physiological processes (but not the traits) likely weakened the relationships. Further, we did not find evidence that exudation rates (or N uptake) align with the RES. Given the emerging interest in including root physiological traits into the RES (which is based mostly on morphological and chemical traits), our results indicate that it may be challenging to include such dynamic processes into the RES. As more mechanistic studies are developed (e.g. tracking exudate responses to tree girdling or nutrient solution culture alterations) and methods for capturing exudates are improved, our ability to understand the role and function of exudates in the environment should come into greater focus.

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Competing interests

The authors declare that they have no conflict of interest.

Author contributions

M.G.M., K.V.B., M.L.M., Y.E.O. and R.P.P. designed the study; M.G.M., K.V.B., M.L.M., Y.E.O., M.M., R.K.B., and S.H. performed the research; M.G.M., M.M., and Y.E.O. analyzed the data; Y.E.O., M.G.M. and R.P.P. wrote the paper with input from all authors.

References

Abramoff, R. Z., & Finzi, A. C. (2016). Seasonality and partitioning of root allocation to rhizosphere soils in a midlatitude forest. *Ecosphere*, *7*(11), e01547. https://doi.org/10.1002/ecs2.1547

Ahonen Jonnarth, U., Van Hees, P. A. W., Lundström, U. S., & Finlay, R. D. (2000). Organic acids produced by mycorrhizal *Pinus sylvestris* exposed to elevated aluminium and heavy metal concentrations. *New Phytologist*, *146*(3), 557–567. https://doi.org/10.1046/j.1469-8137.2000.00653.x

Akatsuki, M., & Makita, N. (2020). Influence of fine root traits on in situ exudation rates in four conifers from different mycorrhizal associations. *Tree Physiology*, *40*(8), 1071–1079. https://doi.org/10.1093/treephys/tpaa051

Anderegg, L. D. L. (2022). Why can't we predict traits from the environment? *New Phytologist*, nph.18586. https://doi.org/10.1111/nph.18586

Andersen, K. M., Mayor, J. R., & Turner, B. L. (2017). Plasticity in nitrogen uptake among plant species with contrasting nutrient acquisition strategies in a tropical forest. *Ecology*, *98*(5), 1388–1398. https://doi.org/10.1002/ecy.1793

Anderson, I. C., & Cairney, J. W. G. (2007). Ectomycorrhizal fungi: Exploring the mycelial frontier. *FEMS Microbiology Reviews*, *31*(4), 388–406. https://doi.org/10.1111/j.1574-6976.2007.00073.x

Bardgett, R. D. (2014). *Going underground: Root traits as drivers of ecosystem processes*. *29*(12), 8.

Bergmann, J., Weigelt, A., & Laughlin, D. C. (2020). The fungal collaboration gradient dominates the root economics space in plants. *SCIENCE ADVANCES*, 10.

Braghiere, R. K., Fisher, J. B., Fisher, R. A., Shi, M., Steidinger, B. S., Sulman, B. N., Soudzilovskaia, N. A., Yang, X., Liang, J., Peay, K. G., Crowther, T. W., & Phillips, R. P. (2021). Mycorrhizal Distributions Impact Global Patterns of Carbon and Nutrient Cycling. *Geophysical Research Letters*, *48*(19). https://doi.org/10.1029/2021GL094514

Britto, D. T., & Kronzucker, H. J. (2013). Ecological significance and complexity of N-source preference in plants. *Annals of Botany*, *112*(6), 957–963. https://doi.org/10.1093/aob/mct157

Brunn, M., Hafner, B. D., Zwetsloot, M. J., Weikl, F., Pritsch, K., Hikino, K., Ruehr, N. K., Sayer, E. J., & Bauerle, T. L. (2022). Carbon allocation to root exudates is maintained in mature temperate tree species under drought. *New Phytologist*, *235*(3), 965–977. https://doi.org/10.1111/nph.18157

Brzostek, E. R., Fisher, J. B., & Phillips, R. P. (2014). Modeling the carbon cost of plant nitrogen acquisition: Mycorrhizal trade-offs and multipath resistance uptake improve predictions of retranslocation: Carbon cost of mycorrhizae. *Journal of Geophysical Research: Biogeosciences*, *119*(8), 1684–1697. https://doi.org/10.1002/2014JG002660

Brzostek, E. R., Greco, A., Drake, J. E., & Finzi, A. C. (2013). Root carbon inputs to the rhizosphere stimulate extracellular enzyme activity and increase nitrogen availability in temperate forest soils. *Biogeochemistry*, *115*(1–3), 65–76. https://doi.org/10.1007/s10533-012-9818-9

Buscot, Munch, Charcosset, & Gardes. (2000). Recent advances in exploring physiology and biodiversity of ectomycorrhizas highlight the functioning of these symbioses in ecosystems. *FEMS Microbiology Reviews*, *24(5)*, 601–614.

Canarini, A., Kaiser, C., Merchant, A., Richter, A., & Wanek, W. (2019). Root Exudation of Primary Metabolites: Mechanisms and Their Roles in Plant Responses to Environmental Stimuli. *Frontiers in Plant Science*, *10*, 157. https://doi.org/10.3389/fpls.2019.00157

Carmona, C. P., Bueno, C. G., Toussaint, A., Träger, S., Díaz, S., Moora, M., Munson, A. D., Pärtel, M., Zobel, M., & Tamme, R. (2021). Fine-root traits in the global spectrum of plant form and function. *Nature*, *597*(7878), 683–687. https://doi.org/10.1038/s41586-021-03871-y

Chari, N. R., & Taylor, B. N. (2022). Soil organic matter formation and loss are mediated by root exudates in a temperate forest. *Nature Geoscience*. https://doi.org/10.1038/s41561-022-01079-x

Chari, N. R., Tumber-Dávila, S. J., Phillips, R. P., Bauerle, T. L., Brunn, M., Hafner, B. D., Klein, T., Obersteiner, S., Reay, M. K., Ullah, S., & Taylor, B. N. (2024). Estimating the global root exudate carbon flux. *Biogeochemistry*. https://doi.org/10.1007/s10533-024-01161-z

Cheng, L., Chen, W., Adams, T. S., Wei, X., Li, L., McCormack, M. L., DeForest, J. L., Koide, R. T., & Eissenstat, D. M. (2016). Mycorrhizal fungi and roots are complementary in foraging within nutrient patches. *Ecology*, *97*(10), 2815–2823. https://doi.org/10.1002/ecy.1514

Comas, L. H., & Eissenstat, D. M. (2009). Patterns in root trait variation among 25 co□existing North American forest species. *New Phytologist*, *182*(4), 919–928. https://doi.org/10.1111/j.1469-8137.2009.02799.x

Eissenstat, D. M. (1991). On the relationship between specific root length and the rate of root proliferation: A field study using citrus rootstocks. *New Phytologist*, *118*(1), 63–68. https://doi.org/10.1111/j.1469-8137.1991.tb00565.x

Eissenstat, D. M., & Yanai, R. D. (1997). The Ecology of Root Lifespan. In *Advances in Ecological Research* (Vol. 27, pp. 1–60). Elsevier. https://doi.org/10.1016/S0065-2504(08)60005-7

Eshel, A., & Beeckman, T. (Eds.). (2013). Inorganic Nitrogen Acquisition and Signaling: Physiological and Molecular Aspects. In *Plant Roots* (0 ed., pp. 386–403). CRC Press. https://doi.org/10.1201/b14550-33

Farrar, J., Hawes, M., Jones, D., & Lindow, S. (2003). HOW ROOTS CONTROL THE FLUX OF CARBON TO THE RHIZOSPHERE. *Ecology*, *84*(4), 827–837. https://doi.org/10.1890/0012-9658(2003)084[0827:HRCTFO]2.0.CO;2

Fei, S., Desprez, J. M., Potter, K. M., Jo, I., Knott, J. A., & Oswalt, C. M. (2017). Divergence of species responses to climate change. *Science Advances*, *3*(5), e1603055. https://doi.org/10.1126/sciadv.1603055

Finzi, A. C., Abramoff, R. Z., Spiller, K. S., Brzostek, E. R., Darby, B. A., Kramer, M. A., & Phillips, R. P. (2015). Rhizosphere processes are quantitatively important components of terrestrial carbon and nutrient cycles. *Global Change Biology*, *21*(5), 2082–2094. https://doi.org/10.1111/gcb.12816

Finzi, A. C., & Berthrong, S. T. (2005). THE UPTAKE OF AMINO ACIDS BY MICROBES AND TREES IN THREE COLD-TEMPERATE FORESTS. *Ecology*, *86*(12), 3345–3353. https://doi.org/10.1890/04-1460

Firestone, M. K., & Davidson, E. A. (1989). Microbiological basis of NO and N 2 O production and consumption in soil. Exchange of trace gases between terrestrial ecosystems and the atmosphere, 47, 7-21.

Freschet, G. T., Roumet, C., Comas, L. H., Weemstra, M., Bengough, A. G., Rewald, B., Bardgett, R. D., De Deyn, G. B., Johnson, D., Klimešová, J., Lukac, M., McCormack, M. L., Meier, I. C., Pagès, L., Poorter, H., Prieto, I., Wurzburger, N., Zadworny, M., Bagniewska□Zadworna, A., … Stokes, A. (2021). Root traits as drivers of plant and ecosystem functioning: Current understanding, pitfalls and future research needs. *New Phytologist*, *232*(3), 1123–1158. https://doi.org/10.1111/nph.17072

Funk, J. L., Larson, J. E., Ames, G. M., Butterfield, B. J., Cavender□Bares, J., Firn, J., Laughlin, D. C., Sutton□Grier, A. E., Williams, L., & Wright, J. (2017). Revisiting the H oly G rail: Using plant functional traits to understand ecological processes. *Biological Reviews*, *92*(2), 1156–1173. https://doi.org/10.1111/brv.12275

Gessler, A., Schneider, S., Von Sengbusch, D., Weber, P., Hanemann, U., Huber, C., Rothe, A., Kreutzer, K., & Rennenberg, H. (1998). Field and laboratory experiments on net uptake of nitrate and ammonium by the roots of spruce (*Picea abies*) and beech (*Fagus sylvatica*) trees. *New Phytologist*, *138*(2), 275–285. https://doi.org/10.1046/j.1469-8137.1998.00107.x

Gougherty, S. W., Bauer, J. E., & Pohlman, J. W. (2018). Exudation rates and δ13C signatures of tree root soluble organic carbon in a riparian forest. *Biogeochemistry*, *137*(1–2), 235–252. https://doi.org/10.1007/s10533-017-0415-9

Han, M., Sun, L., Gan, D., Fu, L., & Zhu, B. (2020). Root functional traits are key determinants of the rhizosphere effect on soil organic matter decomposition across 14 temperate hardwood species. *Soil Biology and Biochemistry*, *151*, 108019. https://doi.org/10.1016/j.soilbio.2020.108019

Hawkins, H.-J., Cargill, R. I. M., Van Nuland, M. E., Hagen, S. C., Field, K. J., Sheldrake, M., Soudzilovskaia, N. A., & Kiers, E. T. (2023). Mycorrhizal mycelium as a global carbon pool. *Current Biology*, *33*(11), R560–R573. https://doi.org/10.1016/j.cub.2023.02.027

Hong, J., Ma, X., Yan, Y., Zhang, X., & Wang, X. (2018). Which root traits determine nitrogen uptake by alpine plant species on the Tibetan Plateau? *Plant and Soil*, *424*(1–2), 63–72. https://doi.org/10.1007/s11104-017-3434-3

Jackson, L. E., Burger, M., & Cavagnaro, T. R. (2008). Roots, Nitrogen Transformations, and Ecosystem Services. *Annual Review of Plant Biology*, *59*(1), 341–363. https://doi.org/10.1146/annurev.arplant.59.032607.092932

Jacoby, R., Peukert, M., Succurro, A., Koprivova, A., & Kopriva, S. (2017). The Role of Soil Microorganisms in Plant Mineral Nutrition—Current Knowledge and Future Directions. *Frontiers in Plant Science*, *8*, 1617. https://doi.org/10.3389/fpls.2017.01617

Jiang, Z., Wang, Q., Xiao, J., Zhang, Z., & Yin, H. (2021). Differential responses of N benefit mediated by root exudate inputs to N addition between two subalpine forests. *Rhizosphere*, *19*, 100404. https://doi.org/10.1016/j.rhisph.2021.100404

Jo, I., Fei, S., Oswalt, C. M., Domke, G. M., & Phillips, R. P. (2019). Shifts in dominant tree mycorrhizal associations in response to anthropogenic impacts. *Science Advances*, *5*(4), eaav6358. https://doi.org/10.1126/sciadv.aav6358

Jones, D. L., Nguyen, C., & Finlay, R. D. (2009). Carbon flow in the rhizosphere: Carbon trading at the soil–root interface. *Plant and Soil*, *321*(1–2), 5–33. https://doi.org/10.1007/s11104-009-9925-0

Keiluweit, M., Bougoure, J. J., Nico, P. S., Pett-Ridge, J., Weber, P. K., & Kleber, M. (2015). Mineral protection of soil carbon counteracted by root exudates. *Nature Climate Change*, *5*(6), 588–595. https://doi.org/10.1038/nclimate2580

Kronzucker, H. J., Siddiqi, M. Y., & Glass, A. D. M. (1997). Conifer root discrimination against soil nitrate and the ecology of forest succession. *Nature*, *385*(6611), 59–61. https://doi.org/10.1038/385059a0

Kumordzi, B. B., Aubin, I., Cardou, F., Shipley, B., Violle, C., Johnstone, J., Anand, M., Arsenault, A., Bell, F. W., Bergeron, Y., Boulangeat, I., Brousseau, M., De Grandpré, L., Delagrange, S., Fenton, N. J., Gravel, D., Macdonald, S. E., Hamel, B., Higelin, M., … Munson, A. D. (2019). Geographic scale and disturbance influence intraspecific trait variability in leaves and roots of North American understorey plants. *Functional Ecology*, *33*(9), 1771–1784. https://doi.org/10.1111/1365-2435.13402

Li, Z., Liu, Z., Gao, G., Yang, X., & Gu, J. (2021). Shift from Acquisitive to Conservative Root Resource Acquisition Strategy Associated with Increasing Tree Age: A Case Study of Fraxinus mandshurica. *Forests*, *12*(12), 1797. https://doi.org/10.3390/f12121797

Liese, R., Lübbe, T., Albers, N. W., & Meier, I. C. (2018). The mycorrhizal type governs root exudation and nitrogen uptake of temperate tree species. *Tree Physiology*, *38*(1), 83–95. https://doi.org/10.1093/treephys/tpx131

Lv, C., Wang, C., Li, Y., & Zhou, Z. (2023). Coordination among root exudation C, mycorrhizal colonization, and functional traits and their responses to drought in five temperate tree species. *Forest Ecology and Management*, *546*, 121316. https://doi.org/10.1016/j.foreco.2023.121316

Ma, Z., Guo, D., Xu, X., Lu, M., Bardgett, R. D., Eissenstat, D. M., McCormack, M. L., & Hedin, L. O. (2018). Evolutionary history resolves global organization of root functional traits. *Nature*, *555*(7694), 94–97. https://doi.org/10.1038/nature25783

Marschner, H., H□ussling, M., & George, E. (1991). Ammonium and nitrate uptake rates and rhizosphere pH in non-mycorrhizal roots of Norway spruce [Picea abies (L.) Karst.]. *Trees*, *5*(1). https://doi.org/10.1007/BF00225330

McCormack, M. L., Adams, T. S., Smithwick, E. A. H., & Eissenstat, D. M. (2014). Variability in root production, phenology, and turnover rate among 12 temperate tree species. *Ecology*, *95*(8), 2224–2235. https://doi.org/10.1890/13-1942.1

McCormack, M. L., Guo, D., Iversen, C. M., Chen, W., Eissenstat, D. M., Fernandez, C. W., Li, L., Ma, C., Ma, Z., Poorter, H., Reich, P. B., Zadworny, M., & Zanne, A. (2017). Building a better foundation: Improving root□trait measurements to understand and model plant and ecosystem processes. *New Phytologist*, *215*(1), 27–37. https://doi.org/10.1111/nph.14459

McCormack, M. L., & Iversen, C. M. (2019). Physical and Functional Constraints on Viable Belowground Acquisition Strategies. *Frontiers in Plant Science*, *10*, 1215. https://doi.org/10.3389/fpls.2019.01215

Meier, I. C., Finzi, A. C., & Phillips, R. P. (2017). Root exudates increase N availability by stimulating microbial turnover of fast-cycling N pools. *Soil Biology and Biochemistry*, *106*, 119–128. https://doi.org/10.1016/j.soilbio.2016.12.004

Meier, I. C., Tückmantel, T., Heitkötter, J., Müller, K., Preusser, S., Wrobel, T. J., Kandeler, E., Marschner, B., & Leuschner, C. (2020). Root exudation of mature beech forests across a nutrient availability gradient: The role of root morphology and fungal activity. *New Phytologist*, *226*(2), 583–594. https://doi.org/10.1111/nph.16389

Midgley, M. G., & Phillips, R. P. (2016). Resource stoichiometry and the biogeochemical consequences of nitrogen deposition in a mixed deciduous forest. *Ecology*, *97*(12), 3369–3378. https://doi.org/10.1002/ecy.1595

Midgley, M. G., & Sims, R. S. (2020). Mycorrhizal Association Better Predicts Tree Effects on Soil Than Leaf Habit. *Frontiers in Forests and Global Change*, *3*, 74. https://doi.org/10.3389/ffgc.2020.00074

Mushinski, R. M., Payne, Z. C., Raff, J. D., Craig, M. E., Pusede, S. E., Rusch, D. B., White, J. R., & Phillips, R. P. (2021). Nitrogen cycling microbiomes are structured by plant mycorrhizal associations with consequences for nitrogen oxide fluxes in forests. *Global Change Biology*, *27*(5), 1068–1082. https://doi.org/10.1111/gcb.15439

Nakayama, M., & Tateno, R. (2018). Solar radiation strongly influences the quantity of forest tree root exudates. *Trees*, *32*(3), 871–879. https://doi.org/10.1007/s00468-018-1685-0

Neumann, G., & Römheld, V. (2000). The release of root exudates as affected by the plant's physiological status. The Rhizosphere: Biochemistry and organic substances at the soil-plant interface, 41-93.

Niu, K., Zhang, S., & Lechowicz, M. J. (2020). Harsh environmental regimes increase the functional significance of intraspecific variation in plant communities. *Functional Ecology*, *34*(8), 1666–1677. https://doi.org/10.1111/1365-2435.13582

Oburger, E., & Jones, D. L. (2018). Sampling root exudates – Mission impossible? *Rhizosphere*, *6*, 116–133. https://doi.org/10.1016/j.rhisph.2018.06.004

Phillips, R. P., Brzostek, E., & Midgley, M. G. (2013). The mycorrhizal□associated nutrient economy: A new framework for predicting carbon–nutrient couplings in temperate forests. *New Phytologist*, *199*(1), 41–51. https://doi.org/10.1111/nph.12221

Phillips, R. P., Erlitz, Y., Bier, R., & Bernhardt, E. S. (2008). New approach for capturing soluble root exudates in forest soils. *Functional Ecology*, *22*(6), 990–999. https://doi.org/10.1111/j.1365-2435.2008.01495.x

Phillips, R. P., & Fahey, T. J. (2006). TREE SPECIES AND MYCORRHIZAL ASSOCIATIONS INFLUENCE THE MAGNITUDE OF RHIZOSPHERE EFFECTS. *Ecology*, *87*(5), 1302–1313. https://doi.org/10.1890/0012-9658(2006)87[1302:TSAMAI]2.0.CO;2

Phillips, R. P., Finzi, A. C., & Bernhardt, E. S. (2011). Enhanced root exudation induces microbial feedbacks to N cycling in a pine forest under long-term CO2 fumigation: Rhizosphere feedbacks in CO2-enriched forests. *Ecology Letters*, *14*(2), 187–194. https://doi.org/10.1111/j.1461-0248.2010.01570.x

Prescott, C. E., Grayston, S. J., Helmisaari, H.-S., Kaštovská, E., Körner, C., Lambers, H., Meier, I. C., Millard, P., & Ostonen, I. (2020). Surplus Carbon Drives Allocation and Plant–Soil Interactions. *Trends in Ecology & Evolution*, *35*(12), 1110–1118.

Rice, E. L., & Pancholy, S. K. (1972). Inhibition of Nitrification by Climax Ecosystems. *American Journal of Botany*, *Vol. 59*(No. 10), 1033–1040.

Sell, M., Ostonen, I., Rohula-Okunev, G., Rusalepp, L., Rezapour, A., & Kupper, P. (2022). Responses of fine root exudation, respiration and morphology in three early successional tree species to increased air humidity and different soil nitrogen sources. *Tree Physiology*, *42*(3), 557–569. https://doi.org/10.1093/treephys/tpab118

Sokol, N. W., Kuebbing, Sara. E., Karlsen-Ayala, E., & Bradford, M. A. (2019). Evidence for the primacy of living root inputs, not root or shoot litter, in forming soil organic carbon. *New Phytologist*, *221*(1), 233–246. https://doi.org/10.1111/nph.15361

Sun, L., Ataka, M., Han, M., Han, Y., Gan, D., Xu, T., Guo, Y., & Zhu, B. (2021a). Root exudation as a major competitive fine□root functional trait of 18 coexisting species in a subtropical forest. *New Phytologist*, *229*(1), 259–271. https://doi.org/10.1111/nph.16865

Sun, L., Ataka, M., Kominami, Y., & Yoshimura, K. (2017a). Relationship between fine-root exudation and respiration of two Quercus species in a Japanese temperate forest. *Tree Physiology*, *37*(8), 1011–1020. https://doi.org/10.1093/treephys/tpx026

Sun, L., Ataka, M., Kominami, Y., Yoshimura, K., & Kitayama, K. (2021b). A constant microbial C/N ratio mediates the microbial nitrogen mineralization induced by root exudation among four co-existing canopy species. *Rhizosphere*, *17*, 100317. https://doi.org/10.1016/j.rhisph.2021.100317

Sun, L., Kominami, Y., Yoshimura, K., & Kitayama, K. (2017b). Root-exudate flux variations among four co-existing canopy species in a temperate forest, Japan. *Ecological Research*, *32*(3), 331–339. https://doi.org/10.1007/s11284-017-1440-9

Sun, Y.-P., Unestam, T., Lucas, S. D., Johanson, K. J., Kenne, L., & Finlay, R. (1999). Exudation-reabsorption in a mycorrhizal fungus, the dynamic interface for interaction with soil and soil microorganisms. *Mycorrhiza*, *9*(3), 137–144. https://doi.org/10.1007/s005720050298

Tabachnick, B. G., & Fidell, L. S. (1996). Using Multivariate Statistics (3rd ed.). *New York: Harper Collins.*

Templer, P. H., & Dawson, T. E. (2004). Nitrogen uptake by four tree species of the Catskill Mountains, New York: Implications for forest N dynamics. *Plant and Soil*, *262*(1/2), 251–261. https://doi.org/10.1023/B:PLSO.0000037047.16616.98

Templer, P. H., Mack, M. C., Iii, F. S. C., Christenson, L. M., Compton, J. E., Crook, H. D., Currie, W. S., Curtis, C. J., Dail, D. B., D'Antonio, C. M., Emmett, B. A., Epstein, H. E., Goodale, C. L., Gundersen, P., Hobbie, S. E., Holland, K., Hooper, D. U., Hungate, B. A., Lamontagne, S., … Zak, D. R. (2012). Sinks for nitrogen inputs in terrestrial ecosystems: A meta□analysis of ^{15}N tracer field studies. *Ecology*, *93*(8), 1816–1829. https://doi.org/10.1890/11-1146.1

Tückmantel, T., Leuschner, C., Preusser, S., Kandeler, E., Angst, G., Mueller, C. W., & Meier, I. C. (2017). Root exudation patterns in a beech forest: Dependence on soil depth, root morphology, and environment. *Soil Biology and Biochemistry*, *107*, 188–197. https://doi.org/10.1016/j.soilbio.2017.01.006

Van Schöll, L., Kuyper, T. W., Smits, M. M., Landeweert, R., Hoffland, E., & Breemen, N. V. (2008). Rock-eating mycorrhizas: Their role in plant nutrition and biogeochemical cycles. *Plant and Soil*, *303*(1–2), 35–47. https://doi.org/10.1007/s11104-007-9513-0

Villarino, S. H., Pinto, P., Jackson, R. B., & Piñeiro, G. (2021). Plant rhizodeposition: A key factor for soil organic matter formation in stable fractions. *Science Advances*, *7*(16), eabd3176. https://doi.org/10.1126/sciadv.abd3176

Wang, Q., Xiao, J., Ding, J., Zou, T., Zhang, Z., Liu, Q., & Yin, H. (2021). Differences in root exudate inputs and rhizosphere effects on soil N transformation between deciduous and evergreen trees. *Plant and Soil*, *458*(1–2), 277–289. https://doi.org/10.1007/s11104-019-04156-0

Weemstra, M., Freschet, G. T., Stokes, A., & Roumet, C. (2021). Patterns in intraspecific variation in root traits are species□specific along an elevation gradient. *Functional Ecology*, *35*(2), 342–356. https://doi.org/10.1111/1365-2435.13723

Weemstra, M., Kuyper, T. W., Sterck, F. J., & Umaña, M. N. (2022). Incorporating belowground traits: Avenues towards a whole□tree perspective on performance. *Oikos*. https://doi.org/10.1111/oik.08827

Weemstra, M., Mommer, L., Visser, E. J. W., Ruijven, J., Kuyper, T. W., Mohren, G. M. J., & Sterck, F. J. (2016). Towards a multidimensional root trait framework: A tree root review. *New Phytologist*, *211*(4), 1159–1169. https://doi.org/10.1111/nph.14003

Weigelt, A., Mommer, L., Andraczek, K., Iversen, C. M., Bergmann, J., Bruelheide, H., Fan, Y., Freschet, G. T., Guerrero□Ramírez, N. R., Kattge, J., Kuyper, T. W., Laughlin, D. C., Meier, I. C., Plas, F., Poorter, H., Roumet, C., Ruijven, J., Sabatini, F. M., Semchenko, M., … McCormack, M. L. (2021). An integrated framework of plant form and function: The belowground perspective. *New Phytologist*, *232*(1), 42–59. https://doi.org/10.1111/nph.17590

Weigelt, A., Mommer, L., Andraczek, K., Iversen, C. M., Bergmann, J., Bruelheide, H., Freschet, G. T., Guerrero-Ramírez, N. R., Kattge, J., Kuyper, T. W., Laughlin, D. C., Meier, I. C., Van Der Plas, F., Poorter, H., Roumet, C., Van Ruijven, J., Sabatini, F. M., Semchenko, M., Sweeney, C. J., … McCormack, M. L. (2023). The importance of trait selection in ecology. *Nature*, *618*(7967), E29–E30. https://doi.org/10.1038/s41586-023-06148-8

Wen, Z., White, P. J., Shen, J., & Lambers, H. (2022). Linking root exudation to belowground economic traits for resource acquisition. *New Phytologist*, *233*(4), 1620–1635. https://doi.org/10.1111/nph.17854

Williams, A., Langridge, H., Straathof, A. L., Fox, G., Muhammadali, H., Hollywood, K. A., Xu, Y., Goodacre, R., & de Vries, F. T. (2021). Comparing root exudate collection techniques: An improved hybrid method. *Soil Biology and Biochemistry*, *161*, 108391. https://doi.org/10.1016/j.soilbio.2021.108391

Williams, A., & de Vries, F. T. (2020). Plant root exudation under drought: implications for ecosystem functioning. *New Phytologist*, 225(5), 1899-1905. https://doi.org/10.1111/nph.16223

Yaffar, D., Cabugao, K. G., & Meier, I. C. (2022). Representing root physiological traits in the root economic space framework. *New Phytologist*, *234*(3), 773–775. https://doi.org/10.1111/nph.18070

Yang, L., Wang, X., Mao, Z., Jiang, Z., Gao, Y., Chen, X., & Aubrey, D. P. (2020). Root Exudation Rates Decrease with Increasing Latitude in Some Tree Species. *Forests*, *11*(10), 1045. https://doi.org/10.3390/f11101045

Yin, H., Li, Y., Xiao, J., Xu, Z., Cheng, X., & Liu, Q. (2013). Enhanced root exudation stimulates soil nitrogen transformations in a subalpine coniferous forest under experimental warming. *Global Change Biology*, *19*(7), 2158–2167. https://doi.org/10.1111/gcb.12161

Yin, H., Wheeler, E., & Phillips, R. P. (2014). Root-induced changes in nutrient cycling in forests depend on exudation rates. *Soil Biology and Biochemistry*, *78*, 213–221. https://doi.org/10.1016/j.soilbio.2014.07.022

Yin, S., Wang, C., Lv, C., & Zhou, Z. (2023). Short-term responses of root traits and carbon exudation to drought in a Larix gmelinii plantation. *Plant and Soil*, *484*(1–2), 393–405. https://doi.org/10.1007/s11104-022-05800-y

Zerihun, A., & Bassirirad, H. (2001). Interspecies variation in nitrogen uptake kinetic responses of temperate forest species to elevated CO_2: Potential causes and consequences: N UPTAKE KINETIC RESPONSE OF TREES TO CO_2. *Global Change Biology*, *7*(2), 211–222. https://doi.org/10.1046/j.1365-2486.2001.00384.x

Zhang, Z., Qiao, M., Li, D., Yin, H., & Liu, Q. (2016). Do warming-induced changes in quantity and stoichiometry of root exudation promote soil N transformations via stimulation of

soil nitrifiers, denitrifiers and ammonifiers? *European Journal of Soil Biology*, *74*, 60–68. https://doi.org/10.1016/j.ejsobi.2016.03.007

Figures and tables

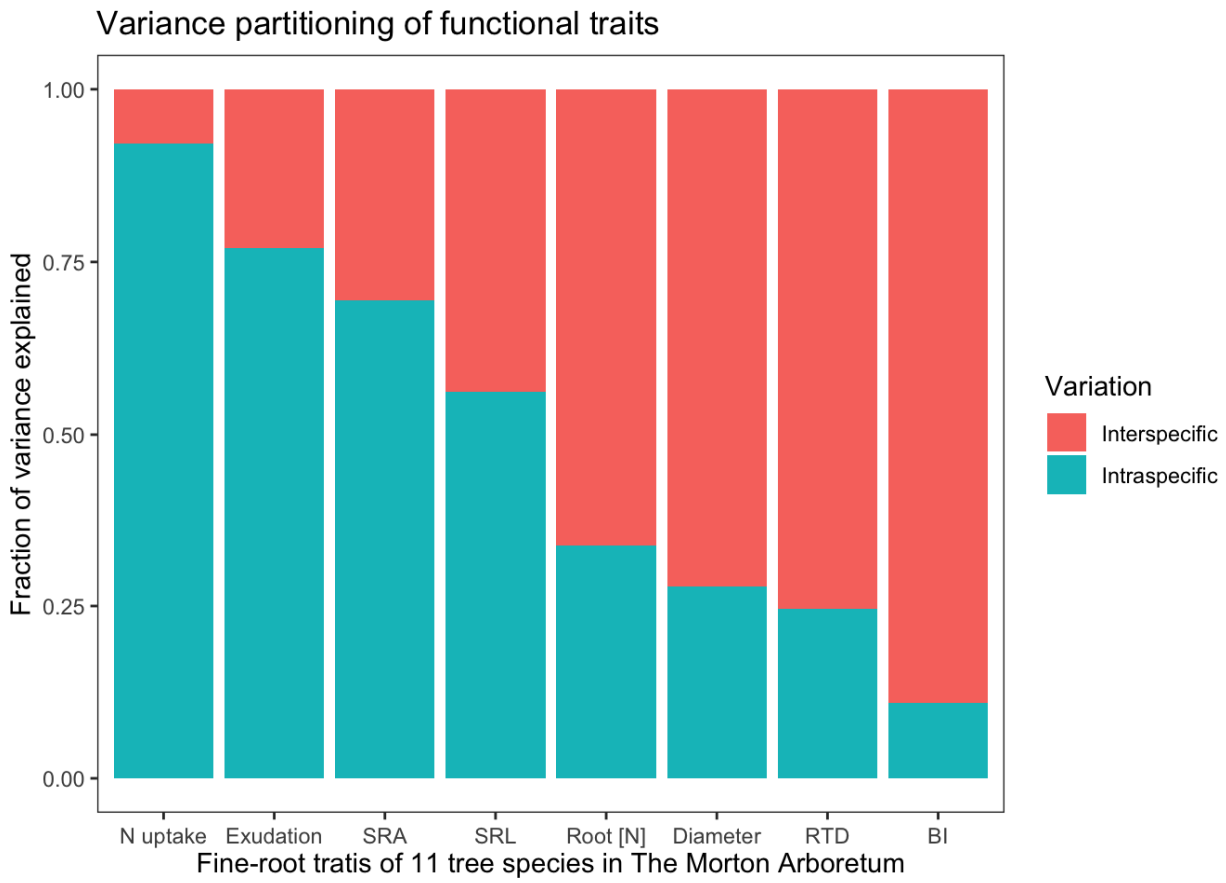

Fig 1) Variance partitioning of fine-root functional traits of 11 tree species. All individuals were mature trees (> 85 years old) occurring in monospecific plots at The Morton Arboretum, Lisle, IL, USA. Fraction of variance explained for each root functional trait was calculated from the results of a mixed effect model where monospecific plots were considered as a random effect. N uptake, net inorganic N uptake rate (mg N d^{-1} g_{root}^{-1}); Exudation, mass-specific exudation rate (mg C d^{-1} g_{root}^{-1}); SRA, specific root area (cm^2 g^{-1}); SRL, specific root length (m g^{-1}); Root [N], root nitrogen concentration (%); Diameter; fine-root diameter (mm); RTD, root tissue density (mg cm^{-3}); BI, branching intensity (tips per cm).

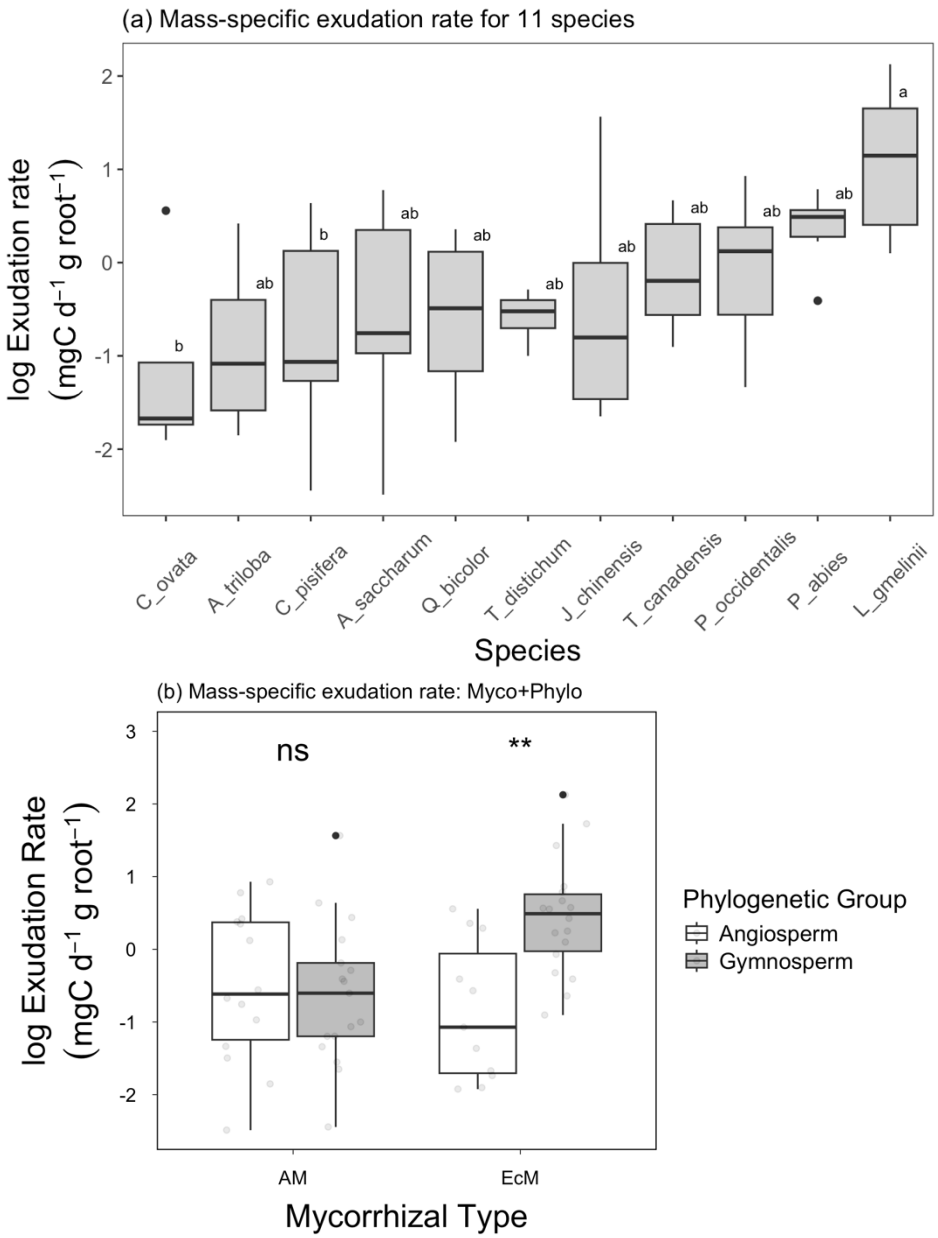

Fig 2) Boxplot of root exudation variation among: (a) 11 tree species and (b) combinations of mycorrhizal type (AM vs. EcM) and phylogenetic group (Angiosperm vs. gymnosperm) at The Morton Arboretum, Lisle, IL, USA. The measured rates were natural log transformed. In (a), the species were aligned in the order of the values of dependent variable. The abbreviations of species denote the first letter of genus and species name, all of which are consistent with Table 1. Different lowercase letters denote significant differences, as determined by a post hoc TukeyHSD test. In (b), predicted mass-specific exudation rates of tree species nested in mycorrhizal types and phylogenetic groups while setting species (species-specific-plot) as a random factor and using restricted maximum likelihood. There was a significant difference between EcM angiosperms and gymnosperms ($F_{(1, 60)} = 6.2$, p-value = 0.02) and a significant interaction (i.e., mycorrhizal type x phylogenetic group: $F_{(1, 60)} = 8.8$, p-value = 0.004), based on the mixed-effect model results analyzed by Type III ANOVA with Satterthwaite's Method. ** indicates significant different difference EcM gymnosperms from the rest of groups. Closed circles denote outliers. The central box in each boxplot represents the median and the interquartile range. The whiskers extend to the minimum and maximum value. Significance level: ns > 0.05, **P < 0.05.

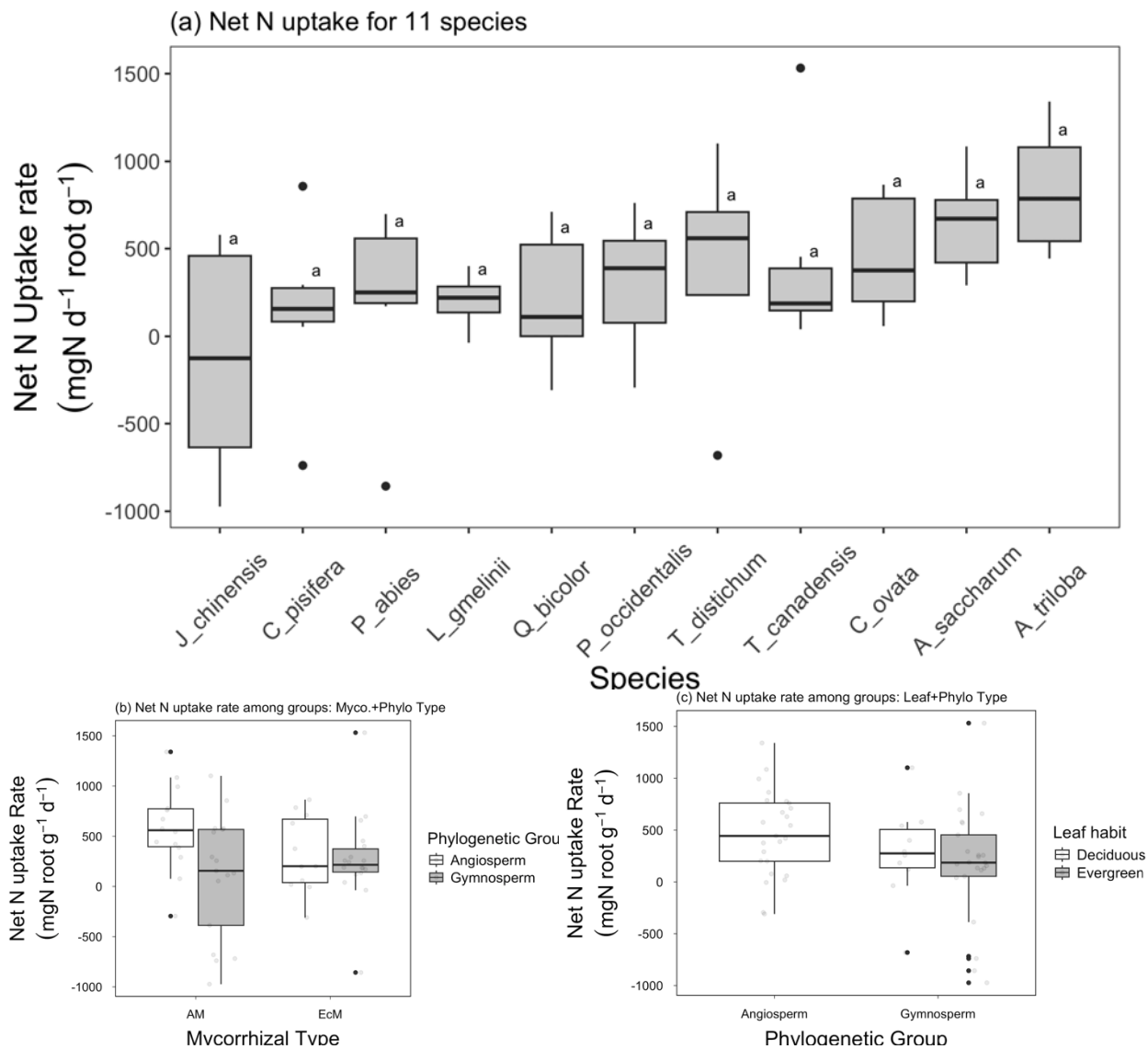

Fig 3) Boxplot of variation of net N uptake rates among: (a) 11 tree species and (b & c) combinations of mycorrhizal type (AM vs. EcM), leaf habit (deciduous vs. evergreen), and phylogenetic group (angiosperm vs. gymnosperm) at The Morton Arboretum, Lisle, IL, USA. In (a), the species were aligned in the order of the values of dependent variable. The abbreviations of species denote the first letter of genus and species name, all of which are consistent with Table 1. Different lowercase letters denote significant differences, as determined by a post hoc TukeyHSD test. The residuals of net N uptake rates were normally distributed (Anderson-Darling test for normality; p-value = 0.41). In (b) and (c), predicted mass-specific N uptake rates of tree species nested in mycorrhizal types, leaf habits, and phylogenetic groups. Closed circles denote outliers. The central box in each boxplot represents the median and the interquartile range. The whiskers extend to the minimum and maximum value. There were no significant differences among tree functional groups (see Table 4).

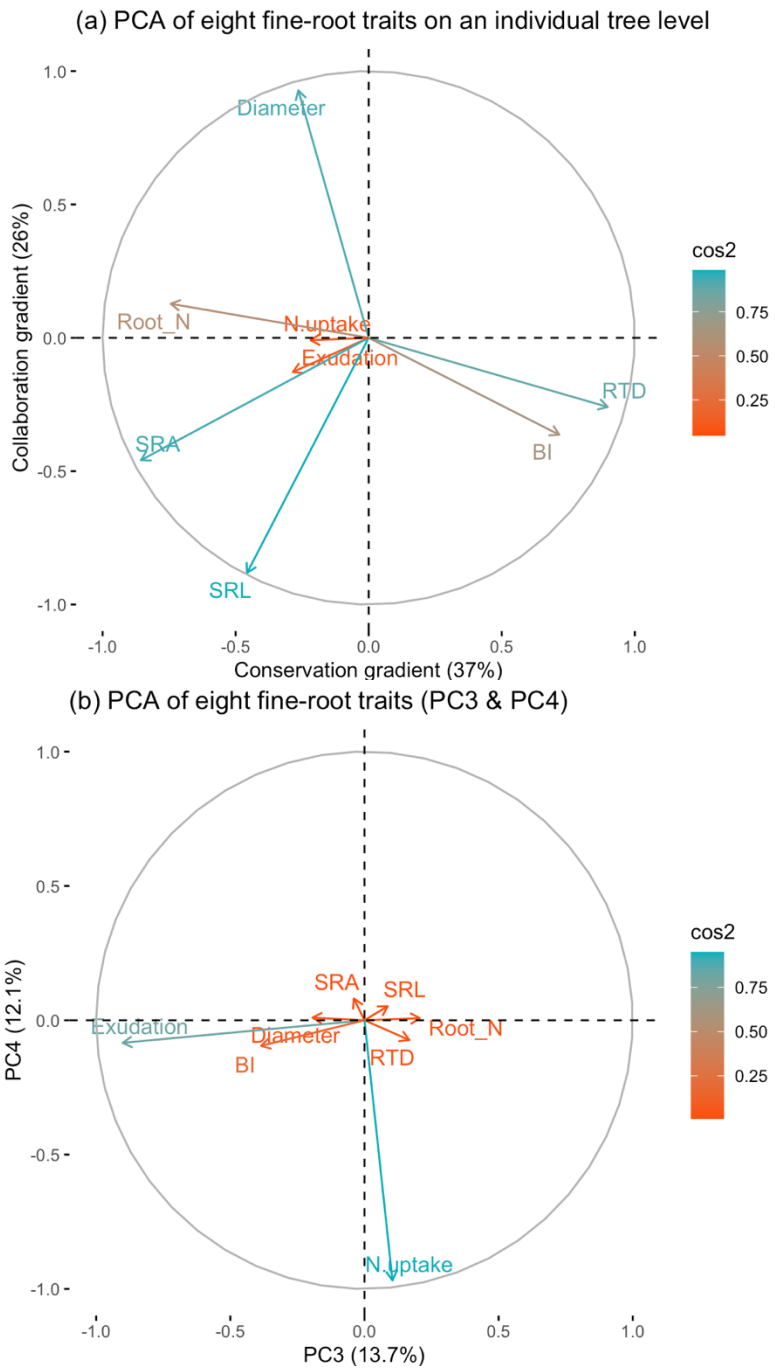

Fig 4) Root economic space (RES) projected by standardized principal components analysis (PCA) of six core fine-root functional traits, exudation and N uptake rates on an individual tree level . PC1 & PC2 represent conservation gradient and collaboration gradient respectively (Bergmann et al., 2020). 'cos2' values indicate that the quality of the representation of the variable on that principal component as a cos2 value close to 1 indicates that the variable is very well represented by the PC(s) in question. (a) RES generated by six core root functional traits that comprise conservation and collaboration gradient with two physiological functions (i.e., exudation and N uptake rates). (b) Two additional axes (Dim 3 & 4) from the RES (Fig 4a) generated by six traits with two physiological functions (see more information in Table S8).

Table 1) Characteristics tree species in the monodominant forestry plots at The Morton Arboretum, Lisle, IL, USA. All trees are at least 70 years old.

	Abbreviation	Soil series	Soil order	Mycorrhizal type	Phylogenetic group	Leaf habit	Clade	Family	n
Acer saccharum	A_saccharum	Ozaukee	Alfisol	AM	Angiosperm	Deciduous	Rosids	Sapindaceae	5
Asimina triloba	A_triloba	Ozaukee	Alfisol	AM	Angiosperm	Deciduous	Magnoliids	Annonaceae	4
Platanus occidentalis	P_occidentalis	Ashkum	Mollisol	AM	Angiosperm	Deciduous	Eudicots	Platanaceae	5
Chamaecyparis pisifera	C_pisifera	Ozaukee	Alfisol	AM	Gymnosperm	Evergreen	Gymnosperms	Cupressaceae	7
Juniperus chinensis	J_chinensis	Ozaukee	Alfisol	AM	Gymnosperm	Evergreen	Gymnosperms	Cupressaceae	6
Taxodium distichum	T_distichum	Sawmill	Mollisol	AM	Gymnosperm	Deciduous	Gymnosperms	Cupressaceae	4
Carya ovata	C_ovata	Ozaukee	Alfisol	EcM	Angiosperm	Deciduous	Rosids	Juglandaceae	5
Quercus bicolor	Q_bicolor	Sawmill	Mollisol	EcM	Angiosperm	Deciduous	Rosids	Fagaceae	6
Larix gmelinii	L_gmelinii	Ozaukee	Alfisol	EcM	Gymnosperm	Deciduous	Gymnosperms	Pinaceae	6
Picea abies	P_abies	Ozaukee	Alfisol	EcM	Gymnosperm	Evergreen	Gymnosperms	Pinaceae	6
Tsuga canadensis	T_canadensis	Ozaukee	Alfisol	EcM	Gymnosperm	Evergreen	Gymnosperms	Pinaceae	6

Note: AM, arbuscular mycorrhizal type; EcM, ectomycorrhizal type

Table 2) Exudation rates, net N uptake rates, and estimated daily exudate (C) and inorganic N uptake fluxes of 11 tree species in The Morton Arboretum (mean ± SD, n =60). All samples were collected in May to July 2022. n denotes the number of tree individuals sampled. Six soil cores were collected randomly at each plot using 5cm diameter soil corer. Mean root-density (g cm-3) was estimated by calculating the mean fine-root biomass in a given volume of soil. To estimate plot-level fluxes of exudate C (total exudation; mgC cm-3 day-1) and net nitrogen uptake (total N uptake; mgN cm-3 day-1), we multiply mean mass-specific exudation (mg C * g root-1 * day-1) and mean net N uptake rates (mg N * g root-1 * day-1) to mean root-density for each species.

		Exudation rate ($mg\ C\ g_{root}^{-1} * day^{-1}$)		Net N uptake ($mg\ N\ g_{root}^{-1} * day^{-1}$)		NH_4^+ uptake ($mg\ N\ g_{root}^{-1} * day^{-1}$)		Net NO_3^- uptake ($mg\ N\ g_{root}^{-1} * day^{-1}$)		Mean root-density ($g\ cm^{-3}$)		Total exudation ($mg\ C * cm^{-3} * day^{-1}$)		Total N uptake ($mg\ N * day^{-1}$)	
Species (plot)	n	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Acer saccharum	5	0.91	0.87	648.9	311.4	429.9	188.1	219.0	192.0	8.6	7.9	7.8	14.6	5,563.3	7,767.3
Asimina triloba	4	0.60	0.63	838.4	408.9	573.8	368.4	264.5	163.2	1.6	1.5	1.0	1.9	1,326.1	1,905.8
Platanus occidentalis	5	1.19	0.88	295.3	413.2	181.5	227.8	113.8	208.4	1.2	0.6	1.4	1.8	342.5	668.6
Chamaecyparis pisifer	7	0.75	0.71	141.7	470.8	107.4	203.5	34.2	269.6	3.5	2.2	2.7	4.1	499.6	1,969.8
Juniperus chinensis	6	1.22	1.78	-133.3	660.4	64.9	331.2	-198.2	346.0	1.0	0.6	1.2	2.5	-128.0	553.4
Taxodium distichum	4	0.58	0.16	384.8	755.3	227.4	312.9	117.7	333.8	1.4	0.8	0.8	0.7	548.1	1,370.4
Carya ovata	5	0.52	0.69	456.7	355.6	322.5	243.9	134.2	134.9	3.0	1.6	1.6	2.9	1,381.8	1,824.6
Quercus bicolor	6	0.73	0.54	207.7	394.7	147.2	150.2	60.5	265.9	2.6	1.8	1.9	2.7	537.0	1,386.3
Larix gmelinii	6	3.82	2.83	203.0	151.4	151.4	87.7	51.6	79.5	2.0	1.3	7.5	10.5	397.3	557.0
Picea abies	6	1.52	0.52	195.1	562.1	34.8	289.0	160.3	291.4	3.7	3.3	5.6	7.0	714.8	2,708.0
Tsuga canadensis	6	1.05	0.66	422.6	560.6	227.4	312.9	195.2	254.0	2.2	2.0	2.3	3.6	924.8	2,091.4

Note. SD: standard deviation; Exudation and N uptake rates are mass-specific.

Table 3) The species-specific means ± SDs of six core root traits. n denotes the number of tree individuals sampled. Net N uptake: net inroganic N uptake rate; Exudation: mass-specific exudation rate

		Specific root length (SRL) ($cm\ g^{-1}$)		Root diameter (Diameter) (mm)		Root tissue density (RTD) ($g\ cm^{2}$)		N concentration (root [N]) (%)		Specific root area (SRA) ($cm^{2}\ g^{-1}$)		Branching intensity (BI) (Tips cm^{-1})	
Species (plot)	n	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Acer saccharum	5	1,636.4	529.4	0.34	0.03	0.7	0.1	1.2	0.1	169.9	42.7	3.2	0.5
Asimina triloba	4	1,110.2	104.3	0.64	0.02	0.3	0.1	2.9	0.2	222.1	28.1	0.7	0.1
Platanus occidentalis	5	1,045.5	540.4	0.52	0.10	0.5	0.1	1.7	0.2	157.5	52.9	3.0	0.5
Chamaecyparis pisifera	7	1,528.9	460.5	0.42	0.06	0.5	0.1	1.9	0.2	195.1	39.5	2.2	0.6
Juniperus chinensis	6	863.6	273.3	0.55	0.09	0.5	0.1	1.4	0.1	142.7	25.2	2.4	0.9
Taxodium distichum	4	1,696.2	374.9	0.51	0.08	0.3	0.0	1.9	0.3	264.1	26.8	2.0	0.4
Carya ovata	5	1,160.7	209.5	0.37	0.05	0.9	0.2	1.3	0.3	133.4	25.2	5.3	1.2
Quercus bicolor	6	1,439.0	397.3	0.33	0.02	0.9	0.1	1.3	0.1	146.7	32.4	5.6	0.5
Larix gmelinii	6	975.8	384.6	0.58	0.11	0.4	0.1	1.5	0.1	168.0	40.1	3.7	0.6
Picea abies	6	1,639.3	220.2	0.42	0.04	0.5	0.1	2.0	0.1	214.4	19.1	4.4	0.7
Tsuga canadensis	6	689.4	276.7	0.59	0.07	0.6	0.2	1.2	0.4	125.2	46.8	4.2	0.6

SD: standard deviation

Table 4) Type III Analysis of Variance Table for predicting root exudation rates, net N uptake rates (ammonium and nitrate), and the ratio of NH4+:No3- of 11 tree species in The Morton Arboretum (n =60) using a linear mixed model (dependent variables~myco x phylo x leaf + (1|species)). The species were nested in mycorrhizal types, phylogenetic groups and leaf habit. P-values (P<0.05) are highlighted in bold. The significance of fixed effects in the model was tested by Type III ANOVA with Satterthwaite's method. Exudation rates were log-transformed to ensure normality and improve homogeneity of variance.

	Exudation rate		Net N uptake		NH4+ : NO3-		NH_4^+ uptake		$NO3^-$ uptake	
Fixed Effect	F value	Pr(>F)	F value	Pr(>F)	F value	Pr(>F)	F value	Pr(>F)	F value	Pr(>F)
Mycorrhizal type (myco)	0.69	0.41	0.02	0.88	6.66	**0.02**	0.59	0.45	1.30	0.26
Phylogenetic group (phylo)	7.99	**0.01**	0.79	0.38	0.24	0.63	0.95	0.33	0.46	0.50
Leaf habit (leaf)	2.36	0.13	0.58	0.45	0.00	0.96	1.04	0.31	0.13	0.71
myco:phylo	9.27	**0.00**	0.05	0.83	0.00	0.99	0.04	0.84	0.04	0.85
myco:leaf	1.90	0.17	1.87	0.18	2.70	0.12	0.66	0.42	3.24	0.08

Note. Numerator Degrees of Freedom = 1; Denominator Degrees of Freedom =60; Net N uptake rates (mgN*d^{-1}*g $root^{-1}$) consist of uptake rates of NH_4^+ and NO_3^- ;
Exudation rates: log-transformed mass-specific exudation rate (mgC*d^{-1}*g $root^{-1}$). The random effect (i.e., (1|species)) in these models were not significant.