



Ammonium nutrition enhances rhizosphere mobilization and uptake of silicon in white lupin grown in low phosphorus soil

Ljiljana Kostic Kravljanc · Jelena Pavlovic · Predrag Bosnic · Igor Kostic · Maja Trailovic · Tijana Dubljanin · Miroslav Nikolic 

Received: 27 August 2024 / Accepted: 23 September 2024 / Published online: 1 October 2024
© The Author(s) 2024

Abstract

Background and Aims While nitrogen (N) supply can enhance plant silicon (Si) accumulation, the mechanisms by which different forms of N affect Si mobilization in the rhizosphere are not well understood. This study aims to elucidate how pH changes induced by ammonium (NH_4^+) and nitrate (NO_3^-)

affect Si availability in the rhizosphere, especially under low phosphorus (P) conditions.

Methods White lupin (*Lupinus albus*) plants were grown in non-fertilized low-P soil, supplied with a low dose of N, either as NH_4^+ or NO_3^- , with or without supply of monosilicic acid. We measured Si levels in various rhizosphere soil pools, along with different plant and rhizosphere soil parameters.

Results The addition of NH_4^+ significantly lowered rhizosphere pH and decreased both Si adsorbed to pedogenic Fe/Mn oxides and amorphous phytogenic Si, resulting in higher concentrations of plant available Si in the white lupin rhizosphere. This led to greater Si uptake and improved plant growth compared to both the $-\text{N}$ and $+\text{NO}_3^-$ treatments. The supply of Si further enhanced these effects, with NH_4^+ showing a consistently different pattern of influence compared to NO_3^- . Additionally, $-\text{N}$ white lupin plants accumulated more P than those treated with N, while Si supply significantly improved P acquisition across all treatments.

Conclusions Our study demonstrates that rhizosphere acidification induced by NH_4^+ nutrition can significantly enhance Si mobilization from the rhizosphere soil in the absence of Si supply and reduce Si adsorption when Si is applied. These findings may have practical implications for improving both Si mobilization in the rhizosphere and the effectiveness of Si fertilizers.

Responsible Editor: Jian Feng Ma.

Supplementary Information The online version contains supplementary material available at <https://doi.org/10.1007/s11104-024-06982-3>.

L. K. Kravljanc (✉) · J. Pavlovic · P. Bosnic · I. Kostic · M. Trailovic · T. Dubljanin · M. Nikolic (✉)
Institute for Multidisciplinary Research, University of Belgrade, 11030 Belgrade, Serbia
e-mail: ljkostic@imsi.bg.ac.rs

M. Nikolic
e-mail: mnikolic@imsi.bg.ac.rs

J. Pavlovic
e-mail: jelena.pavlovic@imsi.bg.ac.rs

P. Bosnic
e-mail: predrag.bosnic@imsi.bg.ac.rs

I. Kostic
e-mail: igork@imsi.bg.ac.rs

M. Trailovic
e-mail: m.trailovic@imsi.bg.ac.rs

T. Dubljanin
e-mail: tdubljanin@imsi.bg.ac.rs

Keywords Nitrogen forms · Rhizosphere · Acidification · Silicon pools · Root-soil interactions

Introduction

Silicon (Si) is the second most abundant element in the Earth's crust, constituting about 29% of the soil mass. Despite this, Si is not universally recognized as an essential mineral element for plants. However, its beneficial effects, particularly in enhancing plant resilience to various stresses, are well-documented in the literature (for review see e.g., Coskun et al. 2019; Debona et al. 2017; Pavlovic et al. 2021). Silicon in soil is present in various fractions, including mobile Si in the form of monosilicic acid which is the only directly available Si fraction for plants' uptake, adsorbed Si (monosilicic acid adsorbed on surface of aluminium (Al) and iron (Fe)/manganese (Mn) oxides and hydroxides), occluded Si in pedogenic Al and Fe/Mn oxides and hydroxides, amorphous Si (biogenic and non-biogenic forms), crystalline Si (in the primary or secondary silicates and quartz) (Cornelis et al. 2010; Sauer et al. 2006). Plant availability of Si in soil is governed by an equilibrium between Si in soil solution, Si adsorbed on soil particles and Si in soil solid phase (Cornelis et al. 2010; Georgiadis et al. 2013; Haynes 2014; Sauer et al. 2006). Numerous factors such as pH, temperature, microbial activity, the presence of cations, Al and Fe/Mn oxides and hydroxides or organic compounds influence Si transformation in soil and thus its availability for plants. For instance, soil acidification leads to the enhanced decay of clay minerals (e. g. kaolinite) as an important soil Si pool (Nguyen et al. 2017).

On the other hand, plant roots can actively modify the chemical properties of the surrounding soil (rhizosphere) through several processes, including proton (H^+) extrusion and exudation of low molecular weight compounds, such as organic acids (Neumann and Ludewig 2023). While processes affecting Si solubility have been extensively studied in the context of pedogenesis or global biogeochemical Si cycling, research on Si mobility, transformation, and availability specifically within the rhizosphere remains limited. De Tombreur et al. (2020) concluded that Western Australian native plants employing P-mobilizing strategies such as carboxylate release, had significantly higher leaf Si concentrations when grown on older, low-P soils, despite these soils having very low Si availability, compared to plants on younger, P-rich soils. Furthermore, in vitro experiments by de Tombreur et al. (2021), which used artificial

carboxylates (oxalate, citrate, and malate) to extract Si from old quartz-rich soils, pure clay minerals (chlorite and kaolinite), and pure quartz (including mixtures with phytoliths), suggested that root-released carboxylates under low P conditions can enhance the mobilization of Si from various soil pools. However, deciphering which are the main rhizosphere processes involved in soil Si mobilization remains challenging (Yu et al. 2024).

Nitrogen (N) affects rhizosphere pH by balancing the cation:anion ratio, as it can be taken up in the forms of either a cation (ammonium, NH_4^+) or an anion (nitrate, NO_3^-). When plants take up more NO_3^- than NH_4^+ , there is a net H^+ removal due to the symport transport of NO_3^- across the root plasma membrane, which increases the rhizosphere pH. In contrast, when plants take up NH_4^+ , they release an equivalent amount of H^+ into the rhizosphere to balance the excess positive charges, thereby decreasing the rhizosphere pH. Consequently, the form of N can affect plant P acquisition due to changes in rhizosphere pH, although the interactions between N form and P supply in relation to rhizosphere pH are complicated and not fully understood. Generally, NH_4^+ supply promotes the solubility of phosphates through rhizosphere acidification (Hinsinger 2001). By contrast, P deficiency may either decrease, increase, or have no effect on rhizosphere pH in NO_3^- -fed plants (e.g., Dinkelaker et al. 1989; Neumann and Römheld 1999; Li et al. 2007; Tang et al. 2009). For legumes, the situation is more complex because N_2 fixation generally leads to rhizosphere acidification due to the preferential uptake of cations over anions (Hinsinger et al. 2003; Li et al. 2008a). White lupin (*Lupinus albus*) is a legume species well adapted to infertile soils; it is particularly effective in increasing P availability in the rhizosphere through the release of citrate and H^+ , primarily from its proteoid roots (Dinkelaker et al. 1989). Furthermore, the NH_4^+ form of N additionally enhances root H^+ release in white lupin under P deficiency (Liu et al. 2016).

Long-term weathering of soil minerals is progressively depleting plant-available Si across large areas of the world (Savant et al. 1996). In addition to natural weathering, intensive agricultural practices that involve growing high-yielding Si-accumulating crops (e.g., cereals and sugarcane) significantly contribute to Si depletion in the soil (Struyf et al. 2010; Guntzer et al. 2012; Vandevenne et al. 2015; Tubana

et al. 2016). Estimates indicate that harvesting rice (*Oryza sativa*) and sugarcane (*Saccharum spp.* hybrid) can remove nearly 1000 kg of Si per hectare from the soil (Savant et al. 1997). The most common Si fertilizers are industrial by-products from the steel industry, which may have potential hazardous effects (Haynes 2014). In recent decades, maintaining Si availability in agricultural soils has become increasingly challenging even with the adoption of sustainable practices like intercropping, cover cropping, integrated crop-livestock systems, effective nutrient management, and the incorporation of Si-rich crop residues (e.g., rice straw and husks) into the soil. However, the impact of applying NH_4^+ and NO_3^- mineral fertilizers on Si availability is less studied (Keeping et al. 2015), and there is virtually no data available at the rhizosphere level.

Therefore, the objective of this study was to investigate how N nutrition affects the availability of Si in the rhizosphere and Si uptake by plants. We hypothesized that the availability of Si in the rhizosphere soil could be further influenced by pH changes induced by NH_4^+ and NO_3^- forms of N, in addition to the root responses to low P conditions, such as carboxylate exudation. To test this hypothesis, we conducted a soil rhizobox experiment using white lupin as a model crop in pH-neutral, low-P soil. Additionally, to elucidate the role of N nutrition in mobilizing Si from various soil pools in the white lupin rhizosphere, both without and with Si fertilization, we performed sequential extractions of soil Si using different extractants.

Material and methods

Plant material and growth conditions

White lupin (*Lupinus albus* L., cv. Vesna) seeds were surface sterilized with H_2O_2 , soaked in saturated CaSO_4 for 24 h and then germinated between two sheets of filter paper moistened with saturated CaSO_4 . After germination, three uniform seedlings were planted in each rhizobox (10×26×1.4 cm; custom made black Plexiglas plates with removable front cover) filled with 0.5 kg of dry soil. The experimental soil was rather pH-neutral (pH in H_2O 6.7), clay loam textured (sand 29%, silt 22%, clay 49%), and low in organic matter content (C_{org} 0.58%), total N

(0.07%), total (257 mg kg^{-1}) and available (Olsen; 6.5 mg kg^{-1}) P; the detailed soil characteristics are shown in Table S1.

The experiment design included three N treatments: -N (control), + NH_4 (30 mg N kg^{-1} dry soil, applied as NH_4SO_4) and + NO_3 (30 mg N kg^{-1} dry soil, applied as CaNO_3), each without (-Si) or with (+Si) addition of 150 mg Si kg^{-1} dry soil, with four replicates (rhizoboxes with three plants) of each treatment. Silicon was applied as monosilicic acid, freshly prepared by passing Na_2SiO_3 solution through a plastic column filled with cation-exchange resin (Amberlite IR-120, H^+ form; Fluka, Buchs SG, Switzerland). The soil was thoroughly mixed with the amendments prior to filling the rhizoboxes and then watered to 70% of the field water capacity (FWC).

The plants were grown for three weeks under controlled environmental conditions in a growth chamber with a light/dark regime of 16/8 h, photon flux density of 300 $\mu\text{mol m}^{-2} \text{s}^{-1}$ at plant height provided by led panels (Apollo 8, Cidly Co., Ltd., Shenzhen, China), temperature of 26 ± 2 °C and relative air humidity of about 70%. During the entire experimental period the plants were irrigated according to weight loss to achieve 70% FWC, and the rhizobox positions were changed randomly.

Plant harvest and rhizosphere soil sampling

The plants were harvested three weeks after planting. At harvest, the plants were carefully removed from the rhizoboxes, the intact roots were shaken gently to remove excess soil; the soil remaining adhered to the root surface, referred to as rhizosphere soil (Kostic et al. 2017), was carefully collected. Soil samples were air dried and thoroughly ground prior to analyses. Shoots were separated, dried at 68 °C for 48 h, and their dry weight was recorded.

Determination of N, P, Si and Mn in leaves

Dry plant material (0.2 g) was digested with 3 ml of concentrated HNO_3 and 2 ml of H_2O_2 in a microwave oven (Ethos Easy, Milestone Srl, Sorisole, Italy) for 1 h. For P analysis, the samples were transferred in volumetric plastic flasks and adjusted to 25 ml with deionized H_2O . For Si determination, the samples were transferred into 25 ml plastic flasks, diluted with 15 ml deionized H_2O , 1 ml concentrated HF

was added, and the samples were then left overnight; after addition of 2.5 ml 2% (w/v) H_3BO_3 , the flask volume was adjusted to 25 ml with deionized H_2O . Concentrations of P, Si and Mn were determined by inductively coupled plasma mass spectrometry (ICP-MS; Agilent 8900 ICP-QQQ, Agilent Technologies, Inc., Santa Clara, CA, USA), after a final dilution of the samples of 1:20 (v/v) with deionized H_2O . The concentrations of N were determined by direct combustion of the dried leaf samples (5–6 mg) in a Vario Microcube CHNS analyser (Elementar Analysensysteme GmbH, Hanau, Germany).

Determination of pH, NO_3^- , NH_4^+ and available P in soil

The soil pH was measured in H_2O (soil:solution ratio of 1:2.5). The dry soil samples were extracted by 2 M KCl for 30 min shaking (soil:solution ratio of 1:10) and the concentrations of NH_4^+ and NO_3^- were determined by HI-4101 Ammonia Combination Ion Selective Electrode (ISE) and HI-4113 Nitrate Combination ISE (Hanna Instruments Ltd., Leighton Buzzard, UK), respectively. Available P was extracted with 0.5 M NaHCO_3 (soil:solution ratio of 1:20) by 30 min shaking and the P concentration was determined by colorimetric molybdenum blue method at 580 nm.

Determination of soil Si fractions

Soil Si fractions were determined by four subsequent extraction steps; 1 g of dry soil sample was placed in a 50 mL centrifuge tube and extracted sequentially using different extractant for each step. The following Si fractions were separated: 1) mobile Si [extracted with 0.01 M CaCl_2 , according to Haysom and Chapman (1975)], representing plant available Si, mainly in the form of monosilicic acid; 2) adsorbed Si [extracted with 0.01 M acetic acid, according to Snyder (2001)], representing monosilicic acid adsorbed to the surfaces of soil minerals (Georgiadis et al. 2013); 3) occluded Si [extracted with 0.2 M NH_4 -oxalate buffer, according to Schwertmann (1964)], representing Si in pedogenic Al and Fe/Mn oxides/hydroxides (Georgiadis et al. 2013); 4) amorphous Si, including phytogenic (biogenic) and mineralogenic (pedogenic) Si sub-fractions [extracted with 1% NaCO_3 , according to DeMaster (1981)].

Briefly, the mobile Si fraction was extracted with 10 ml 0.01 M CaCl_2 (soil:solution ratio of 1:10) by

shaking for 16 h. After centrifugation at $2000\times g$ for 5 min, the supernatant was used for Si analysis (step 1) and the residue was washed with deionized H_2O and subsequently dried at 65 °C. The whole residue from the step 1 was extracted with 10 ml 0.01 M acetic acid as described above and the supernatant was used for determination of adsorbed Si fraction (step 2). The soil residue from the step 2 was washed and dried as described above, extracted with 50 ml 0.2 M NH_4 -oxalate buffer solution (pH 3) for 1 h in darkness, centrifuged, and the supernatant was used for determination of occluded Si fraction (step 3). The residue from the step 3, previously washed and dried, was used for determination of two amorphous Si fractions, i.e. phytogenic (biogenic) Si fraction and pedogenic (mineralogenic) Si fraction. The time-course extraction method by DeMaster (1981) is based on the difference between the rapid dissolution of biogenic Si (within the first 2 h of the extraction) and slower release of Si from clay minerals at a constant rate over the whole extraction time. Dry soil residue from the step 3 (30 mg) was incubated for 5 h with 40 ml 1% NaCO_3 at 85 °C in shaker bath and 1 ml aliquot of the extraction solution was sampled and analysed at 1, 2, 3, 4 and 5 h. Prior to Si measurement the aliquot was neutralized with 9 mL of 0.02 M HCl. The concentrations of Si extracted over time (mg g^{-1}) were plotted against extraction time and the linear intercept at time 0 was extrapolated, representing the biogenic Si fraction. The concentration difference from time 0 to 5 h represented pedogenic Si.

Statistical analyses

Data were subjected to two-way ANOVA and differences between treatment means were compared by a posteriori Tukey's test at significance level of 0.05, using the statistical software Statistica 6 (StatSoft, Inc., Tulsa, OK, USA).

The multivariate structure of the collected data was analysed by Principal Component Analysis (PCA) by PC-ORD7 software (MjM Software Design, Gleneden Beach, USA), using correlation cross-product matrix, Euclidean distance and no rotation. Correlation cross-product matrix was centred and standardized (relativized by standard deviate), what brought all the analysed variables to an equal footing. The number of significant ordination axes to be interpreted was determined after Monte Carlo randomization test (999 runs).

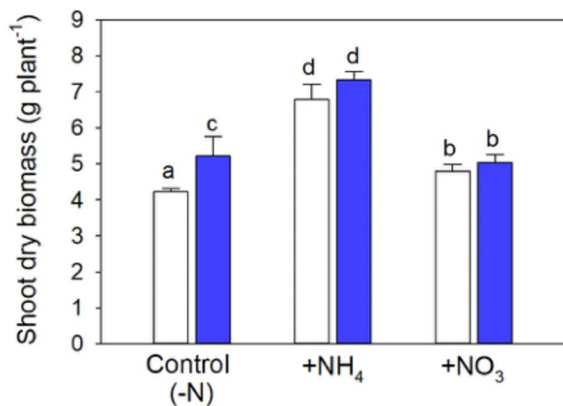


Fig. 1 Effect of N and Si nutrition on shoot growth of white lupin (*Lupinus albus*). Plants were grown in the rhizoboxes for four weeks in the control (–N), +NH₄, and +NO₃ treatments, each without (–Si) or with (+Si) supply of monosilicic acid. Open bars, –Si; filled bars, +Si. Data are means ± SD ($n=4$). Statistically significant differences among the treatment means are denoted by different letter (Tukey’s test following two-way ANOVA; $P<0.05$). The visual effects of N and Si nutrition on white lupin plants are illustrated in Fig. S1

Results

Plant parameters

The addition of N (both forms) significantly enhanced shoot growth in white lupin grown in soil without Si fertilization (Fig. 1, S1). However, the addition of NH₄⁺ form led to a 42% increase in shoot biomass accumulation compared to the plants supplied with NO₃[–]. Additionally, amendment of Si significantly increased shoot dry biomass in the control plants (–N) by 24% but did not affect plant growth in N-supplied plants. The leaf N concentrations were significantly higher in N-fed plants, with the increase being more pronounced in those receiving NH₄⁺ (Fig. 2a). However, the application of Si had no impact on leaf N concentrations.

Interestingly, N supply significantly reduced leaf P concentrations by 50% in lupin plants grown without Si. In contrast, Si supply increased leaf P concentrations by 33% in the control (–N) plants but had no effect on N-fed plants (Fig. 2b). Consequently, the leaf N:P ratio was approximately 1.5 times higher in N-fed plants compared to control plants without N, while Si application had no effect on this ratio (Fig. 3a). Leaf Si concentrations increased significantly with N addition, showing about a 1.5-fold

increase in NH₄⁺-fed plants and a 1.2-fold increase in NO₃[–]-fed plants (Fig. 2c). Even in the Si supplied plants, the addition of NH₄⁺ further increased leaf Si concentrations by 50% compared to both the control (–N) and NO₃[–] treatments. Leaf Mn concentrations also increased significantly with N addition, being 2.2 times higher in NH₄⁺-fed plants and 1.7 times higher in NO₃[–]-fed plants (Fig. 2d). On the other hand, the supply of Si significantly reduced leaf Mn concentrations in all N treatments.

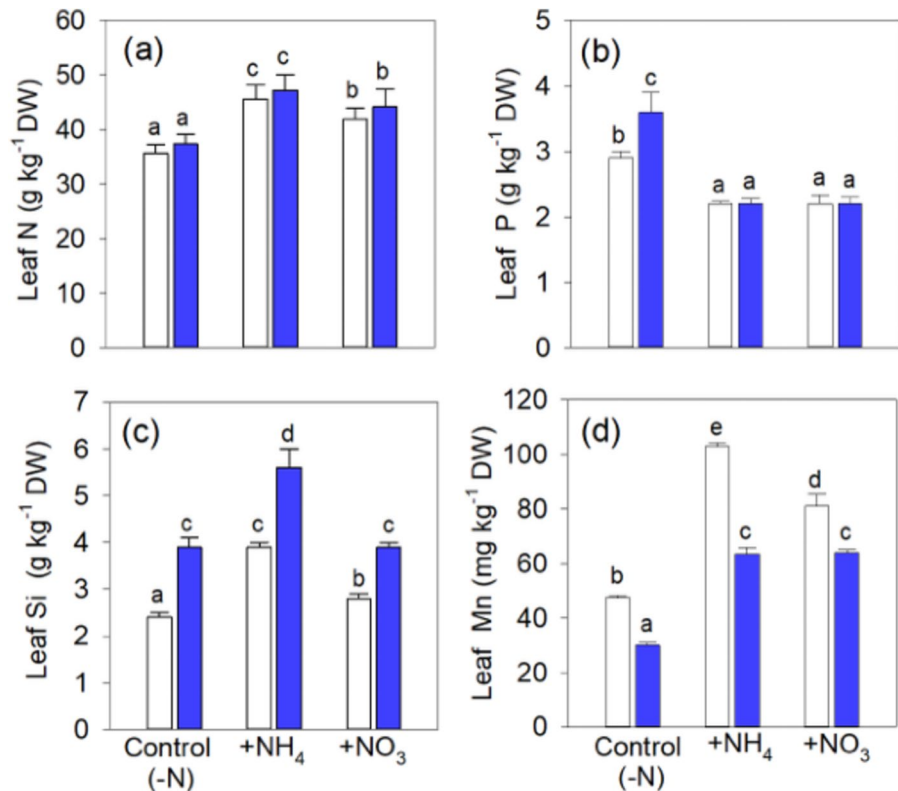
Rhizosphere soil parameters

The rhizosphere pH of the control (–N) treatment without Si decreased by 0.6 pH units compared to the initial soil pH (Fig. 4a; see Table S1). The addition of NH₄⁺ lowered the rhizosphere pH by 0.5 pH units relative to the control (–N) treatment without Si (Fig. 4a) and by 1.1 pH units compared to the initial soil pH (see Table S1). In contrast, NO₃[–] caused a significant increase in rhizosphere pH, rising by 0.4 pH units compared to the control (–N) treatment without Si (Fig. 4a), though it remained 0.2 pH units below the initial soil pH (see Table S1). The addition of Si significantly increased the rhizosphere pH by approximately 0.4 pH units only in the control (–N) treatment, with no significant changes observed in the NH₄⁺ and NO₃[–] treatments.

The addition of NH₄⁺ dramatically increased the rhizosphere NH₄⁺ concentration by more than 15 times compared to both the control (–N) and NO₃[–] treatments, while the application of Si did not significantly affect NH₄⁺ concentrations in any of the treatments (Fig. 4b). The addition of NO₃[–] raised the rhizosphere NO₃[–] concentration by 2.3-fold compared to the control (Fig. 4c). Interestingly, the NO₃[–] concentration in the rhizosphere also increased by 1.7-fold in the NH₄⁺ treatment compared to the control (Fig. 4b). The supply of Si had no impact on the rhizosphere concentrations of NH₄⁺ and NO₃[–] across all treatments.

Regardless of Si application, different Si fractions in the rhizosphere soil were affected differently by N nutrition (Fig. 5). The concentration of plant-available Si (mobile Si fraction) increased by about 35% with NH₄⁺ treatment (Fig. 5a). In contrary, the addition of NO₃[–] only slightly decreased this fraction compared to the control (–N). The Si adsorbed onto inorganic soil particle surfaces containing Fe/Mn oxides/hydroxides (adsorbed Si fraction) significantly decreased with N addition by 43% (Fig. 5b). However, Si associated with

Fig. 2 Effect of N forms on the concentrations of N (a), P (b), Si (c) and Mn (d) in leaves of white lupin. Plants were grown in the rhizoboxes for four weeks in the control (–N), +NH₄, and +NO₃ treatments, each without (–Si) or with (+Si) supply of monosilicic acid. Open bars, –Si; filled bars, +Si. Data are means ± SD (*n* = 4). Statistically significant differences among the treatment means are denoted by different letter (Tukey's test following two-way ANOVA; *P* < 0.05)



pedogenic Al and Fe/Mn oxides (occluded Si fraction) in the rhizosphere soil did not change significantly with N treatments (Fig. 5c). The concentration of amorphous Si, originating from plant phytoliths (phytogenic Si fraction), significantly decreased with NH₄⁺ treatment by 35% (Fig. 5d). Amorphous Si originated from soil minerals (pedogenic Si fraction) remained unchanged across all treatments (Fig. 5e). Application of Si as monosilicic acid also influenced Si fractions differently, increasing both mobile and adsorbed Si fractions across all N treatments while showing no significant effect on occluded and amorphous Si (phytogenic and pedogenic) fractions (Fig. 5e). However, the patterns observed were consistent with those in the –Si treatments (Fig. 5).

Relations between plant and rhizosphere-soil parameters

The addition of N showed varying effects on the depletion of plant available Si in the rhizosphere (Table 1). The bulk-to-rhizosphere gradient of plant available (CaCl₂-extracted) Si significantly increased by more

than twofold with NH₄⁺ treatment, regardless of Si supply. In contrast, rhizosphere depletion of plant-available Si decreased significantly with NO₃[–] supply, showing reductions of 3.2 times in the –Si treatment and 2.5 times in the +Si treatment. Addition of NH₄⁺ also led to about a twofold increase in Si uptake by white lupin plants compared to the control (–N) in both –Si and +Si treatments. However, Si uptake did not significantly differ between the NO₃[–]-fed plants and the control plants, regardless of Si supply.

In contrast to plant available Si, the depletion of plant available P in the rhizosphere was independent of N nutrition and was approximately 7 times lower with Si supply (Table 2). In the –Si treatment, NH₄⁺-fed white lupin plants exhibited a 20% higher P uptake compared to the control, while no significant differences were observed between the control (–N) and NO₃[–]-fed plants. However, the application of Si significantly increased P uptake in the +Si plants, regardless of N addition, following a similar pattern to that observed in the –Si treatments.

The multivariate correlations in the experimental data are visualized in Fig. 6. Nitrogen nutrition per

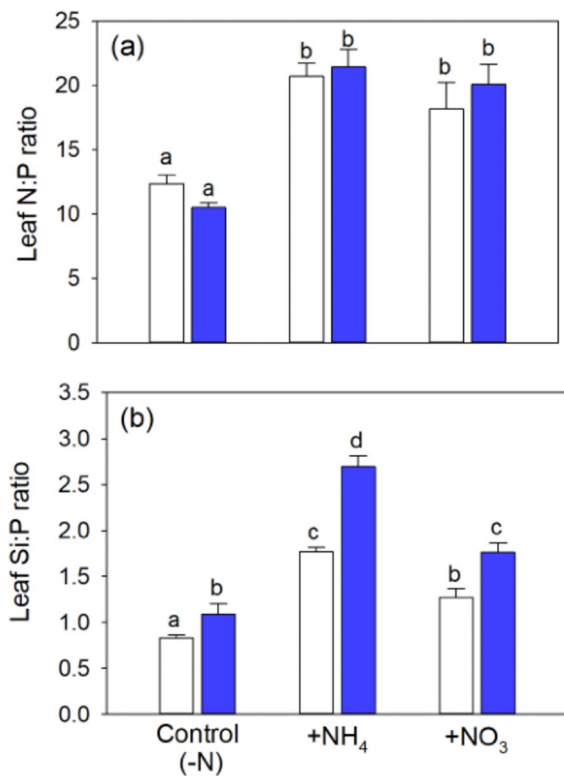


Fig. 3 Effect of N forms on the N:P (a) and N:Si (b) ratios in leaves of white lupin. Plants were grown in the rhizoboxes for four weeks in the control (-N), +NH₄, and +NO₃ treatments, each without (-Si) or with (+Si) supply of monosilicic acid. Open bars, -Si; filled bars, +Si. Data are means $\pm$ SD ($n=4$). Statistically significant differences among the treatment means are denoted by different letter (Tukey's test following two-way ANOVA; $P<0.05$)

se (without Si supply) was responsible for about 65% of the total variation in -Si trials (Fig. 6a). Clearly, the addition of NH₄⁺ increased leaf Si concentrations and the rhizosphere-soil mobile Si fraction, enhanced plant growth (as shoot dry biomass) and increased the leaf concentrations of N and Mn, while it decreased soil pH, rhizosphere-soil adsorbed and phytogenic fractions of Si, and leaf P concentration. Both N and Si nutrition accounted for about 69% of the variability among the measured plant and soil parameters (Fig. 6b). The effects of N forms are summarized along the PC axis 1, while the second PC axis separates -Si from +Si treatments.

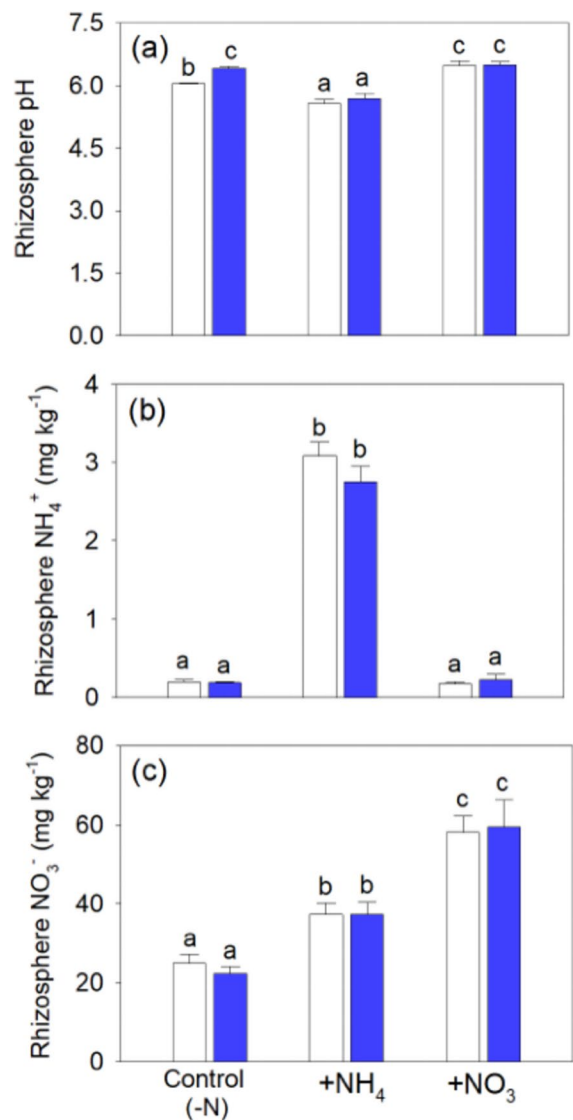
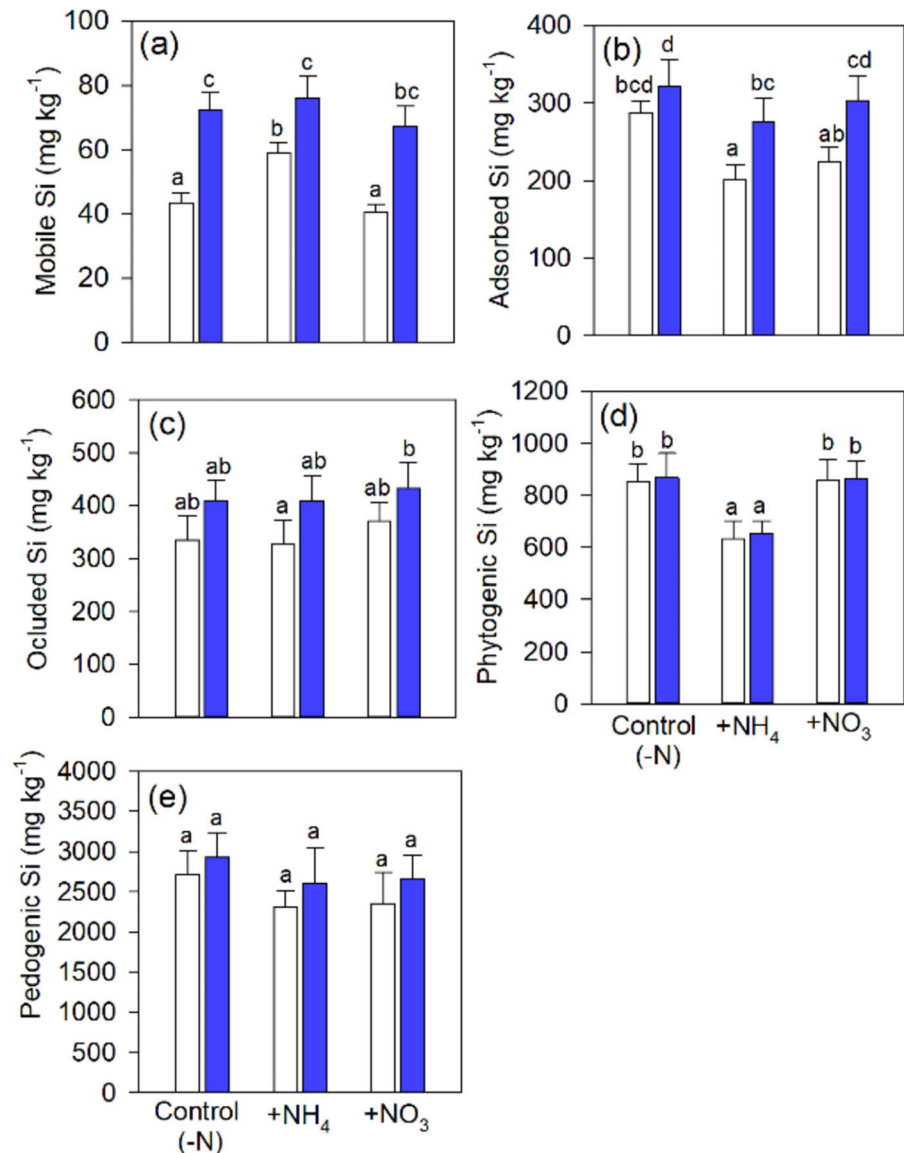


Fig. 4 Effect of N forms on the pH (a) and concentrations of NH₄⁺ (b) and NO₃⁻ (c) in the white lupin rhizosphere soil. Plants were grown in the rhizoboxes for four weeks in the control (-N), +NH₄, and +NO₃ treatments, each without (-Si) or with (+Si) supply of monosilicic acid. Open bars, -Si; filled bars, +Si. Data are means $\pm$ SD ($n=4$). Statistically significant differences among the treatment means are denoted by different letter (Tukey's test following two-way ANOVA; $P<0.05$)

Discussion

Very recently, Yu et al. (2024) reported that long-term addition of N in low-P subtropical forest soil affected Si pools in topsoil, especially plant available Si extracted with CaCl₂. Present study investigated

Fig. 5 Effect of N forms on the Si fractions in the white lupin rhizosphere soil. Mobile (CaCl_2 -extracted) Si (a); adsorbed (acetic acid-extracted) Si (b); occluded (NH_4 -oxalate-extracted) Si (c); amorphous phytogenic Si (d); amorphous pedogenic Si (e). Plants were grown in the rhizoboxes for four weeks in the control (–N), + NH_4 , and + NO_3 treatments, each without (–Si) or with (+Si) supply of monosilicic acid. Open bars, –Si; filled bars, +Si. Data are means $\pm$ SD ($n=4$). Statistically significant differences among the treatment means are denoted by different letter (Tukey's test following two-way ANOVA; $P<0.05$)



how different forms of mineral N fertilizer (NH_4^+ vs. NO_3^-) affect the mobilization of Si from various soil pools at the rhizosphere level, under continuous low-P conditions. We found that Si uptake by white lupin, a model crop known for its very efficient P acquisition under limited P conditions, was enhanced by the addition of NH_4^+ form of N (Table 1; Fig. 2). To differentiate between the effects of low-P and the N-form on rhizosphere pH and thus Si availability, we used pH-neutral (pH 6.7), low-P (total and plant available P of 257 and 6.5 mg kg^{–1}, respectively) soil, without P fertilization. To minimize potential negative impacts of high N supply on Si uptake

(e.g., downregulation of Lsi1 and Lsi2 transporters at high N conditions, Wu et al. 2017) we applied a low dose of N fertilizers, equivalent to approximately 40 kg N ha^{–1}. This amount of applied N also does not alter the high N_2 fixation rate of lupine roots (Pál-mason et al. 1992). Additionally, de Tombeur et al. (2022) reported that N supply decreased leaf Si concentrations in durum wheat genotypes in proportion to shoot biomass, which was attributed to a dilution effect. Previous research on N and Si interactions has primarily used NO_3^- or NH_4NO_3 fertilizers (Minden et al. 2021; de Tombeur et al. 2022; Yu et al. 2024). Consequently, these studies have not addressed the

Table 1 Effect of N forms on the bulk-to-rhizosphere gradient of plant available Si and Si uptake by white lupin plants. Plants were grown in the rhizoboxes for four weeks in the control (–N), +NH₄, and +NO₃ treatments, each without (–Si) or with(+Si) supply of monosilicic acid. Data are means ± SD (*n* = 4). Statistically significant differences among the treatment means are denoted by different letter (Tukey's test following two-way ANOVA; *P* < 0.05)

Treatments	Bulk-to-rhizosphere gradient of plant available Si (mg kg ^{–1})		Si uptake (mg plant ^{–1})	
	–Si	+Si	–Si	+Si
Control (–N)	3.6 ± 0.4b	3.0 ± 0.1b	1.01 ± 0.06a	2.03 ± 0.30b
+NH ₄	8.1 ± 0.7c	7.2 ± 0.4c	2.33 ± 0.18c	4.09 ± 0.46d
+NO ₃	1.1 ± 0.2a	1.2 ± 0.1a	1.32 ± 0.15ab	1.95 ± 0.13b

impact of NH₄⁺-induced rhizosphere acidification on Si availability.

The rhizosphere pH may differ from the bulk soil pH by up to two units, influenced by various plant and soil factors, particularly the form of N supply, which determines the cation:anion uptake ratio (Neumann and Ludewig 2023). The experimental soil used in this study was low in total N, with NO₃[–] being the predominant form of inorganic N available to plants (see Table S1). Despite high soil NO₃[–]/NH₄⁺ ratio in the control treatment without N supply, the rhizosphere pH was about 0.6 units lower than the initial soil pH, likely due to N₂ fixation leading to enhanced uptake of excess cations and net H⁺ release by white lupin roots (O'Sullivan et al. 2022). The addition of NH₄⁺ further decreased the rhizosphere pH by 0.5 units compared to the control (–N) and by 1.1 units relative to the initial soil pH. In contrast, the addition of NO₃[–] increased the rhizosphere pH by 0.4 units, although it remained 0.2 units lower than the initial soil pH (Table S1; Fig. 4a). These findings are in accordance with those of Sas et al. (2002), who demonstrated that H⁺ extrusion by P-deprived white lupin roots was most pronounced with NH₄⁺ supply and less pronounced with NO₃[–] supply.

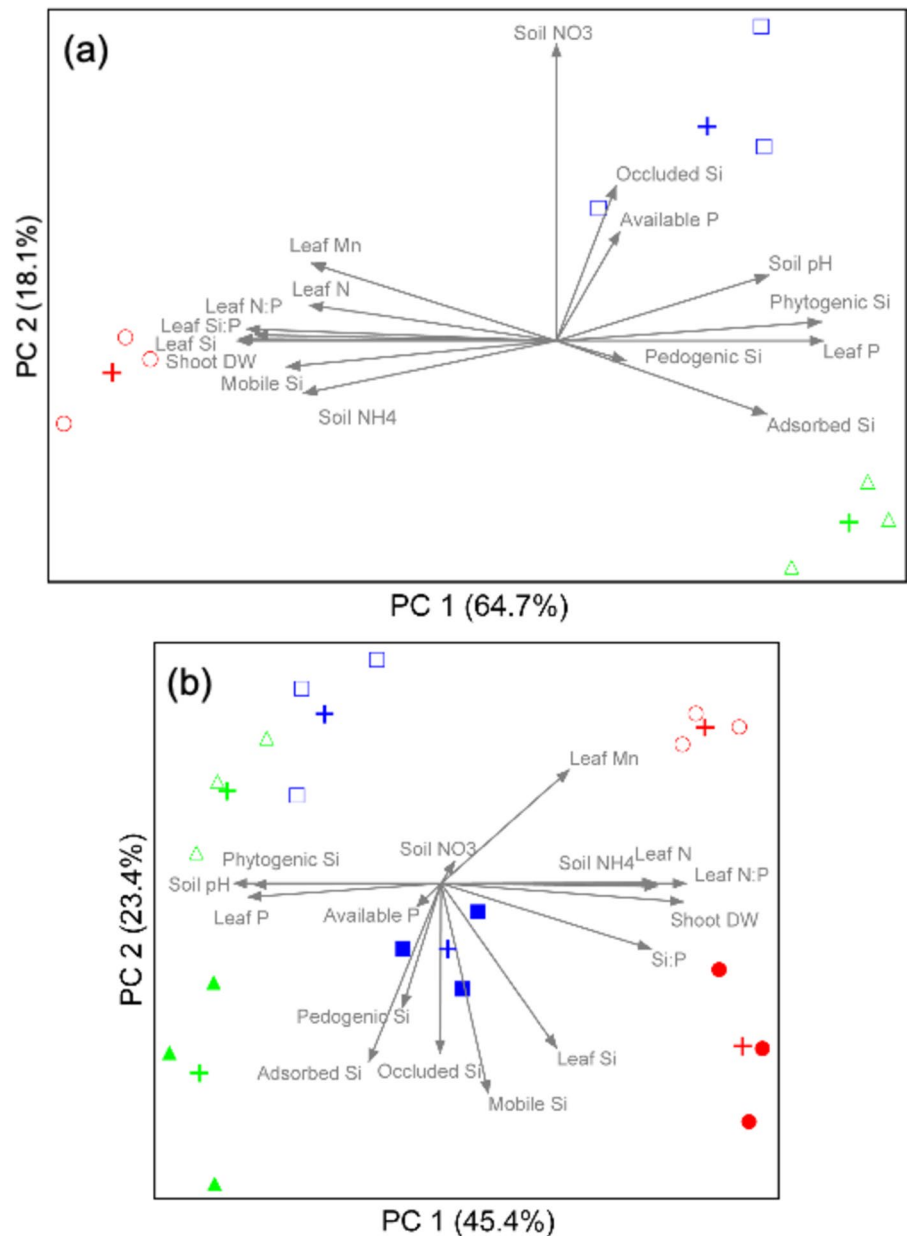
In our experimental plant-soil system, white lupine was continuously exposed to P deficiency, regardless of N nutrition, while maintaining critical leaf P concentrations (2–3 mg P g^{–1} DW) necessary for high levels of citrate exudation (Li et al. 2008a, b). This allowed the plants to effectively mobilize P from the rhizosphere soil (Table 2; Fig. 2b). Although the addition of N did not significantly affect the depletion of P in the rhizosphere soil (Table 2), NH₄⁺ supply notably increased both shoot dry biomass and P uptake, thereby increasing leaf N:P ratio above 20 (Table 2; Fig. 1, S1, 2b; see also Yu et al. 2024). Interestingly, the addition of Si significantly increased leaf P concentration only in the control plants without N supply (Fig. 2b), which can be attributed to Si-enhanced citrate exudation (Kostic et al. 2017). However, Si supply significantly enhanced P uptake across all treatments (see Table 2), potentially due to the upregulation of phosphate transporter genes (*PHT1*) by Si (Kostic et al. 2017). Consequently, the gradient of available P between the bulk and the rhizosphere soil decreased, suggesting that Si supply enhanced the overall processes of P acquisition.

Mineral Si in soils exists in three main phases: primary silicates (e.g., quartz, feldspars), secondary

Table 2 Effect of N forms on the bulk-to-rhizosphere gradient of plant available P and P uptake by white lupin plants. Plants were grown in the rhizoboxes for four weeks in the control (–N), +NH₄, and +NO₃ treatments, each without (–Si) or with(+Si) supply of monosilicic acid. Data are means ± SD (*n* = 4). Statistically significant differences among the treatment means are denoted by different letter (Tukey's test following two-way ANOVA; *P* < 0.05)

Treatments	Bulk-to-rhizosphere gradient of plant available P (mg kg ^{–1})		P uptake (mg plant ^{–1})	
	–Si	+Si	–Si	+Si
Control (–N)	1.4 ± 0.2a	0.23 ± 0.03a	1.22 ± 0.07a	1.89 ± 0.09d
+NH ₄	1.5 ± 0.1a	0.20 ± 0.05a	1.49 ± 0.07b	1.62 ± 0.09c
+NO ₃	1.5 ± 0.2a	0.21 ± 0.02a	1.32 ± 0.16a	1.63 ± 0.05c

Fig. 6 Principal component analysis (PCA) of the measured response of plant and rhizosphere soil parameters as affected by N forms (–Si) (a), and by N forms and Si supply (+/–Si) (b). The share of the total variance represented by each axis is parenthesized; treatment centroids are denoted by crosses. The angles and lengths of the radiating lines indicate the direction and strength of relationships of the measured parameters with ordination scores. One statistically significant PC axis represents 64.5% of the total variance (a); two statistically significant PC axes represent 68.8% of the total variance (b). Control (–N), triangle (green); +NH₄ treatment, circle (red); +NO₃ treatment, square (blue). Open symbols, –Si; filled symbols, +Si



silicates (e.g., clay minerals), and amorphous silica (Sommer et al. 2006). A large portion of the amorphous silica in many soils is composed of bio-opal, primarily in the form of phytoliths (Georgiadis et al. 2017). There is an equilibrium between Si in the soil solution (mainly as monosilicic acid), Si adsorbed onto soil particles (mostly Fe/Mn oxides and hydroxides), and Si in the soil solid phase, which includes both crystalline forms (e.g., quartz, feldspars, mica, and secondary clay minerals) and amorphous forms

(including soil phytoliths). These various Si pools can transform into one another depending on soil conditions such as weathering, pH, and temperature. At low pH, soil clay minerals can dissolve, releasing Si into the soil solution; this released Si may then be adsorbed onto mineral surfaces or precipitated as amorphous Si (Sauer et al. 2006).

Carboxylates exuded by roots under low-P conditions can further influence these processes (de Tombeur et al. 2021). The release of citric acid

(with three carboxyl groups) or malic acid (with two carboxyl groups) can liberate Si adsorbed on pedogenic Fe/Mn oxides and hydroxides, making it available to plants as monosilicic acid (de Tomberur et al. 2021; Yu et al. 2024). Additionally, N supply can stimulate the exudation of organic acids in response to increasing P limitation, which can further enhance the availability of soil Si (Yu et al. 2024).

In this study, we observed a significant increase in leaf Si concentrations in N-fed white lupin plants without Si supply (Fig. 2c), with the effect being more pronounced in plants fed with NH_4^+ . This increase was attributed to the greater depletion of plant available Si in the rhizosphere and subsequent Si uptake by NH_4^+ -fed plants (see Table 1). Therefore, the observed high level of Si depletion in the rhizosphere of white lupin fertilized with NH_4^+ can be explained by the combined effect of rhizosphere acidification (Fig. 4a) and N-promoted exudation of citrate (Yu et al. 2024). Consequently, the leaf Si:P ratio increased with N supply, regardless of Si supply, with the highest values observed in the NH_4^+ treatments ($-/+$ Si) due to rhizosphere acidification (Figs. 3b, 4a).

The effects of Si dynamics in soil, especially in response to N mineral fertilizers, have not been extensively investigated (e.g., Keeping et al. 2015; Yu et al. 2024), and there is lack of data on Si dynamics specifically at the rhizosphere level. Our study reveals that the addition of NH_4^+ form of mineral N fertilizer significantly affected specific Si fractions in the rhizosphere soil, resulting in an approximately 35% increase in mobile (plant available) Si and reductions of 43% and 35% in adsorbed Si and amorphous phytogenic Si, respectively (Fig. 5). Even in soil that was fertilized with Si, there was a decrease in adsorbed Si (17%) and phytogenic Si (33%), along with a slight increase in mobile Si (8%). This trend was further supported by the enhanced depletion of mobile Si from bulk to rhizosphere soil, which led to an increased Si uptake by white lupine plants (Table 1). The increased mobilization of Si through NH_4^+ supply was followed by a dramatic rise in leaf Mn concentration (Fig. 2d). Meunier et al. (2018) found a positive correlation between soil Si extracted with acetic acid and Mn, which suggests assension of

Si adsorbed on the surfaces of Fe/Mn and Al oxides/ (oxi)hydroxides (Hiemstra et al. 2007). Additionally, increased leaf Mn levels have been used as a proxy for rhizosphere carboxylate concentrations (Zhou et al. 2022). This implies that leaf Mn concentration may also be a useful indicator of rhizosphere acidification processes and the mobilization of Si. Our findings are consistent with those of Keeping et al. (2015), who reported increased Si uptake in sugarcane due to NH_4^+ fertilization. Although NO_3^- fertilization also caused a significant decrease in adsorbed Si (28% of the control), it did not result in increased depletion of Si in the rhizosphere or its uptake by white lupin plants (Fig. 5b; Table 1).

Despite the enhanced Si depletion, NH_4^+ fertilization increased plant available Si, likely due to enhanced dissolution of both adsorbed and phytogenic Si fractions (Table 1; Fig. 5b, d). The concentration of Si in soil solution is significantly influenced by the dissolution of Si from the solid phase, particularly from amorphous phytogenic silica, which is several times more soluble than quartz or other crystalline forms of Si (Frayssé et al. 2009, 2010; Tubana et al. 2016). However, the dissolution of phytoliths in soil is highly variable and these processes are not well understood (Haynes 2014). Klotzbücher et al. (2018) reported that Si availability, extracted with acetate, increased linearly with soil pH, attributing this to the increased solubility of phytoliths with rising soil pH. Our study did not confirm this observation; instead, we found that increasing the rhizosphere pH from NH_4^+ to NO_3^- led to decreased plant-available Si and dissolution of phytogenic Si, resulting in lower Si uptake (Fig. 4a, 5a, d; Table 1). In fact, the relationship between pH and the Si adsorption/desorption processes varies within the dynamic environment of the rhizosphere. Haynes and Zhou (2020) suggested that Si adsorption increases with soil pH, resulting in stronger retention of Si, while Si desorption from soil particles is inversely related to soil pH. In support of this, Meunier et al. (2018) demonstrated a positive correlation between soil pH and plant available Si, indicating that soil pH can serve as a proxy for plant-available Si up to a pH of 7.5. Therefore, reducing rhizosphere pH through NH_4^+ treatment could enhance the desorption of Si and decrease its adsorption.

The overall effect of adding NH_4^+ to non-Si-fertilized soil is clear (Fig. 6a): there is a decrease in rhizosphere-soil pH and in the adsorbed and phyto-genic fractions of soil Si, and an increase in the rhizo-sphere-soil mobile Si fraction, indicating enhanced Si dissolution (supported by the increased leaf accu-mulation of Mn), which together led to higher leaf Si accumulation. A similar pattern of NH_4^+ effects is observed when Si is added (Fig. 6b), indicating improved Si use efficiency. Therefore, modification of the soil-rhizosphere pH by N nutrition is an important component of the rhizosphere processes involved in soil mobilization of Si.

In conclusion, our study reveals that additional rhizosphere acidification induced by NH_4^+ nutri-tion can significantly enhance soil Si mobilization in a legume-low-P soil system. We clearly show that NH_4^+ fertilization considerably lowers rhizosphere pH, thereby increasing the dissolution of Si adsorbed on Fe/Mn oxides and Si from amorphous phyto-genic pools. Moreover, NH_4^+ enhances the efficacy of Si fertilizers, likely by reducing the adsorption of mono-silicic acid. These findings could have practical impli-cations for improving both rhizosphere Si mobiliza-tion and the effectiveness of Si fertilizers through the low-dose application of NH_4^+ fertilizers, particularly in cereal-legume intercropping systems.

Acknowledgements This work was supported by the Fund of Science of the Republic of Serbia *Si4Crop* Grant (7739571) to LKK. We thank Dr Nina Nikolic for help in statistical analyses and data interpretation.

Author contributions Ljiljana Kostic Kravljanc conceived the study, performed most of the laboratory work, analyzed data and wrote the first draft of the manuscript; Jelena Pavlovic was responsible for plant and soil chemical analyses; Predrag Bosnic was responsible for rhizobox construction, plant cultivation and plant sample collection; Igor Kostic performed some of the laboratory work and contributed in plant and soil chemical analyses; Maja Trailovic was responsible for plant sample preparation for chemical analyses; Tijana Dubljanin was responsible for rhizosphere soil collection and soil sample preparation for chemical analyses; Miroslav Nikolic conceived the study, wrote the final draft of the manuscript and took care of several revisions. All authors read and approved the final manuscript.

Declarations

Competing interests The authors have no conflict of interest to declare.

Open Access This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit <http://creativecommons.org/licenses/by/4.0/>.

References

- Cornelis J-T, Ranger J, Iserentant A, Delvaux B (2010) Tree species impact the terrestrial cycle of silicon through various uptakes. *Biogeochemistry* 97:231–245. <https://doi.org/10.1007/s10533-009-9369-x>
- Coskun D, Deshmukh R, Sonah H, Menzies JG, Reynolds O, Ma JF, Kronzucker HJ, Bélanger RR (2019) The controversies of silicon's role in plant biology. *New Phytol* 221:67–85. <https://doi.org/10.1111/nph.15343>
- de Tombreur F, Turner BL, Laliberté E, Lambers H, Mahy G, Faucon MP, Zemunik G, Cornelis JT (2020) Plants sustain the terrestrial silicon cycle during ecosystem retrogression. *Science* 369:1245–1248. <https://doi.org/10.1126/science.abc0393>
- de Tombreur F, Cornelis JT, Lambers H (2021) Silicon mobilisation by root–released carboxylates. *Trends Plant Sci* 26:1116–1125. <https://doi.org/10.1016/j.tplants.2021.07.003>
- de Tombreur F, Lemoine T, Violle C, Fréville H, Thorne SJ, Hartley SE, Lambers H, Fort F (2022) Nitrogen availability and plant–plant interactions drive leaf silicon concentration in wheat genotypes. *Funct Ecol* 36:2833–2844. <https://doi.org/10.1111/1365-2435.14170>
- Debona D, Rodrigues FA, Datnoff LE (2017) Silicon's role in abiotic and biotic plant stresses. *Annu Rev Phytopathol* 55:85–107. <https://doi.org/10.1146/annurev-phyto-080516-035312>
- DeMaster DJ (1981) The supply and accumulation of silica in the marine environment. *Geochim Cosmochim Acta* 45:1715–1732. [https://doi.org/10.1016/0016-7037\(81\)90006-5](https://doi.org/10.1016/0016-7037(81)90006-5)
- Dinkelaker B, Römheld V, Marschner H (1989) Citric acid excretion and precipitation of calcium citrate in the rhizosphere of white lupin (*Lupinus albus* L.). *Plant Cell Environ* 12:285–292. <https://doi.org/10.1111/j.1365-3040.1989.tb01942.x>
- Frayse F, Pokrovsky OS, Schott J, Meunier JD (2009) Surface chemistry and reactivity of plant phytoliths in aqueous solutions. *Chem Geol* 258:197–206. <https://doi.org/10.1016/j.chemgeo.2008.10.003>
- Frayse F, Pokrovsky OS, Meunier J-D (2010) Experimental study of terrestrial plant litter interaction with aqueous

- solutions. *Geochim Cosmochim Acta* 74:70–84. <https://doi.org/10.1016/j.gca.2009.09.002>
- Georgiadis A, Sauer D, Herrmann L, Breuer J, Zarei M, Stahr K (2013) Development of a method for sequential Si extraction from soils. *Geoderma* 209–210:251–256. <https://doi.org/10.1016/j.geoderma.2013.06.023>
- Georgiadis A, Rinklebe J, Straubinger M, Rennert T (2017) Silicon fractionation in Mollic Fluvisols along the Central Elbe River, Germany. *CATENA* 153:100–105. <https://doi.org/10.1016/j.catena.2017.01.027>
- Guntzer F, Keller C, Poulton PR, McGrath SP, Meunier J-D (2012) Long-term removal of wheat straw decreases soil amorphous silica at Broadbalk, Rothamsted. *Plant Soil* 352:173–184. <https://doi.org/10.1007/s11104-011-0987-4>
- Haynes RJ (2014) A contemporary overview of silicon availability in agricultural soils. *J Plant Nutr Soil Sci* 177:831–844. <https://doi.org/10.1002/jpln.201400202>
- Haynes RJ, Zhou Y-F (2020) Silicate sorption and desorption by a Si-deficient soil – Effects of pH and period of contact. *Geoderma* 365:114204. <https://doi.org/10.1016/j.geoderma.2020.114204>
- Haysom MBC, Chapman LS (1975) Some aspects of the calcium silicate trials at Mackay. *Proc Queens Soc Sugar Cane Technol* 42:117–122
- Hiemstra T, Barnett MO, van Riemsdijk WH (2007) Interaction of silicic acid with goethite. *J Colloid Interface Sci* 310:8–17. <https://doi.org/10.1016/j.jcis.2007.01.065>
- Hinsinger P (2001) Bioavailability of soil inorganic P in the rhizosphere as affected by root-induced chemical changes: a review. *Plant Soil* 237:25–41. <https://doi.org/10.1023/A:1013351617532>
- Hinsinger P, Plassard C, Tang C, Jaillard B (2003) Origins of root-mediated pH changes in the rhizosphere and their responses to environmental constraints: a review. *Plant Soil* 248:43–59. <https://doi.org/10.1023/A:1022371130939>
- Keeping MG, Rutherford RS, Sewpersad C, Miles N (2015) Provision of nitrogen as ammonium rather than nitrate increases silicon uptake in sugarcane. *AoB PLANTS* 7:plu080. <https://doi.org/10.1093/aobpla/plu080>
- Klotzbücher T, Klotzbücher A, Kaiser K, Merbach I, Mikutta R (2018) Impact of agricultural practices on plant-available silicon. *Geoderma* 331:15–17. <https://doi.org/10.1016/j.geoderma.2018.06.011>
- Kostic L, Nikolic N, Bosnic D, Samardzic J, Nikolic M (2017) Silicon increases phosphorus (P) uptake by wheat under low P acid soil conditions. *Plant Soil* 419:447–455. <https://doi.org/10.1007/s11104-017-3364-0>
- Li L, Li SM, Sun JH, Zhou LL, Bao XG, Zhang HG, Zhang FS (2007) Diversity enhances agricultural productivity via rhizosphere phosphorus facilitation on phosphorus-deficient soils. *Proc Natl Acad Sci USA* 104:11192–11196. <https://doi.org/10.1073/pnas.0704591104>
- Li H, Shen J, Zhang F, Clairotte M, Drevon J, Le Cadre E, Hinsinger P (2008a) Dynamics of phosphorus fractions in the rhizosphere of common bean (*Phaseolus vulgaris* L.) and durum wheat (*Triticum turgidum durum* L.) grown in monocropping and inter-cropping systems. *Plant Soil* 312:139–150. <https://doi.org/10.1007/s11104-007-9512-1>
- Li H, Shen J, Zhang F, Tang C, Lambers H (2008b) Is there a critical level of shoot phosphorus concentration for cluster-root formation in *Lupinus albus*? *Funct Plant Biol* 35:328–336. <https://doi.org/10.1071/FP07222>
- Liu H, Tang C, Li C (2016) The effects of nitrogen form on root morphological and physiological adaptations of maize, white lupin and faba bean under phosphorus deficiency. *AoB PLANTS* 8:plw058. <https://doi.org/10.1093/aobpla/plw058>
- Meunier JD, Sandhya K, Prakash NB, Borschneck D, Dus-souillez P (2018) pH as a proxy for estimating plant-available Si? A case study in rice fields in Karnataka (South India). *Plant Soil* 432:143–155. <https://doi.org/10.1007/s11104-018-3758-7>
- Minden V, Schaller J, Olde VH (2021) Plants increase silicon content as a response to nitrogen or phosphorus limitation: a case study with *Holcus lanatus*. *Plant Soil* 462:95–108. <https://doi.org/10.1007/s11104-020-04667-1>
- Neumann G, Ludewig U (2023) Rhizosphere chemistry influencing plant nutrition. In: Rengel Z, Cakmak I, White PJ (eds) *Marschner's Mineral Nutrition of Plants*, 4th edn. Academic Press, London, pp 545–586. <https://doi.org/10.1016/B978-0-12-819773-8.00013-7>
- Neumann G, Römhild V (1999) Root excretion of carboxylic acids and protons in phosphorus-deficient plants. *Plant Soil* 211:121–130. <https://doi.org/10.1023/A:1004380832118>
- Nguyen MN, Picardal F, Dultz S, Dam TTN, Nguyen AV, Nguyen KM (2017) Silicic acid as a dispersibility enhanced in a Fe-oxide-rich kaolinitic soil clay. *Geoderma* 286:8–14. <https://doi.org/10.1016/j.geoderma.2016.10.029>
- O'Sullivan BJ, Jin J, Tang C (2022) White lupin (*Lupinus albus* L.) exposed to elevated atmospheric CO₂ requires additional phosphorus for N₂ fixation. *Plant Soil* 476:477–490. <https://doi.org/10.1007/s11104-022-05311-w>
- Pálmason F, Danso SKA, Hardarson G (1992) Nitrogen accumulation in sole and mixed stands of sweet-blue lupin (*Lupinus angustifolius* L.), ryegrass and oats. *Plant Soil* 142:135–142. <https://doi.org/10.1007/BF00010183>
- Pavlovic J, Kostic L, Bosnic P, Kirkby EA, Nikolic M (2021) Interactions of silicon with essential and beneficial elements in plants. *Front Plant Sci* 12:697592. <https://doi.org/10.3389/fpls.2021.697592>
- Sas L, Rengel Z, Tang C (2002) The effect of nitrogen nutrition on cluster root formation and proton extrusion by *Lupinus albus*. *Ann Bot* 89:435–442. <https://doi.org/10.1093/aob/mcf066>
- Sauer D, Saccone L, Conley DJ, Herrmann L, Sommer M (2006) Review of methodologies for extracting plant-available and amorphous Si from soils and aquatic sediments. *Biogeochemistry* 80:89–108. <https://doi.org/10.1007/s10533-005-5879-3>
- Savant NK, Snyder GH, Datnoff LE (1996) Silicon Management and Sustainable Rice Production. *Adv Agron* 58:151–199. [https://doi.org/10.1016/S0065-2113\(08\)60255-2](https://doi.org/10.1016/S0065-2113(08)60255-2)
- Savant NK, Datnoff LE, Snyder GH (1997) Depletion of plant-available silicon in soils: a possible cause of declining rice yields. *Commun Soil Sci Plant Anal* 28:1245–1252. <https://doi.org/10.1080/00103629709369870>
- Schwertmann U (1964) Differenzierung der Eisenoxide des Bodens durch Extraktion mit Ammoniumoxalat-Lösung.

- Z Pflanzenernaehr Boden 105:194–202. <https://doi.org/10.1002/jpln.3591050303>
- Snyder GH (2001) Methods for silicon analyses in plants, soils and fertilizers. In: Datnoff LE, Snyder GH, Korndörfer GH (eds) *Silicon in Agriculture*. Elsevier Science B.V, Amsterdam, pp 185–196
- Sommer M, Kaczorek D, Kuzyakov Y, Breuer J (2006) Silicon pools and fluxes in soils and landscapes – a review. *J Plant Nutr Soil Sci* 169:310–329. <https://doi.org/10.1002/jpln.200521981>
- Struyf E, Smis A, Van Damme S, Garnier J, Govers G, Van Wesemael B, Conley DJ, Batelaan O, Frot E, Clymans W, Vandevenne F, Lancelot C, Goos P, Meire P (2010) Historical land use change has lowered terrestrial silica mobilization. *Nat Commun* 1:129. <https://doi.org/10.1038/ncomms1128>
- Tang C, Han XZ, Qiao YF, Zheng SJ (2009) Phosphorus deficiency does not enhance proton release by roots of soybean [*Glycine max* (L.) Murr.]. *Environ Exp Bot* 67:228–234
- Tubana BS, Babu T, Datnoff LE (2016) A review of silicon in soils and plants and its role in US agriculture. *Soil Sci* 181:1. <https://doi.org/10.1097/SS.0000000000000179>
- Vandevenne FI, Barão L, Ronchi BP, Govers G, Meire P, Kelly EF, Struyf E (2015) Silicon pools in human impacted soils of temperate zones. *Global Biogeochem Cycles* 29:1439–1450. <https://doi.org/10.1002/2014GB005049>
- Wu X, Yu Y, Baerson SR, Song Y, Liang G, Ding C, Niu J, Pan Z, Zeng R (2017) Interactions between nitrogen and silicon in rice and their effects on resistance toward the brown planthopper *Nilaparvata lugens*. *Front Plant Sci* 8:28. <https://doi.org/10.3389/fpls.2017.00028>
- Yu H, Qin Z, de Tombeur F, Lambers H, Lu X, Lai Y, Kuang Y (2024) Atmospheric nitrogen deposition: what are the impacts on silicon dynamics in a subtropical forest? *Plant Soil*. <https://doi.org/10.1007/s11104-024-06872-8>
- Zhou XM, Ranathunge K, Cambridge ML, Dixon KW, Hayes PE, Nikolic M, Shen Q, Zhong H, Lambers H (2022) A cool spot in a biodiversity hotspot: why do tall *Eucalyptus* forests in Southwest Australia exhibit low diversity? *Plant Soil* 476:669–688. <https://doi.org/10.1007/s11104-022-05559-2>

Publisher's Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.