



Nitrous oxide emissions after struvite application in relation to soil P status

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Abstract

Purpose Although struvite ($\text{MgNH}_4\text{PO}_4 \cdot 6\text{H}_2\text{O}$) is mostly considered to be a novel phosphorus (P) fertiliser, it does contain a significant amount of nitrogen (N). Yet, relatively little is known about the soil N dynamics in struvite-amended soils. Here, we focus on how struvite application impacts emissions of the greenhouse gas nitrous oxide (N_2O), in relation to soil P status.

Methods We conducted a 54-day greenhouse pot experiment on two similar soils with different P status (“low-P soil”; “high-P soil”) seeded with *Lolium perenne* L. We applied seven fertiliser treatments (Control; Struvite granules; Struvite powder; Urea; Triple superphosphate (TSP); TSP + Struvite granule; TSP + Urea). Except for the unfertilised Control and

the TSP treatments, N application rate was $150 \text{ kg N} \cdot \text{ha}^{-1}$. Nitrous oxide (N_2O) fluxes, aboveground yield, plant N and P uptake and readily plant-available soil N and P contents were measured.

Results In the low-P soil, none of the fertiliser treatments induced a significant increase in N_2O emission compared to the control. In the high-P soil, struvite application resulted in lower emissions than urea application, statistically not different from the control treatment. Struvite powder significantly increased both plant N and P uptake compared to granular struvite and the resulting yield was similar to conventional fertilisation (TSP and Urea). Any struvite application also resulted in lower readily plant-available soil nitrate contents than urea.

Conclusion Our results suggest that struvite fertilisation can reduce the risk of gaseous N losses without compromising agronomic performance. Pulverizing struvite granules further promotes its dissolution, which could be useful for crops with early nutrient needs.

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Introduction

Phosphorus (P) is an essential element for plant growth. Since many soils are P-deficient and unable to supply P to plants at a sufficient rate, P fertilisation

is needed to overcome P deficiency in plants (Hinsinger 2001). Agricultural food production heavily relies on rock phosphate as a source to produce mineral P fertiliser (Nesme et al. 2018). However, global rock phosphate reserves are limited (Childers et al. 2011; Cordell 2011) and might be exhausted within a century (Cordell and White 2013). This causes an increasing need for an alternative, more sustainable management of P in agriculture. To achieve this, circularity needs to be restored through the recycling of nutrients from waste products, especially for P (Muhmood et al. 2019; Tonini et al. 2019). For a more sustainable P management, the recovery of P from wastewater rich in nutrients such as ammonium (NH_4) and orthophosphate (P-PO_4) through the precipitation of struvite ($\text{MgNH}_4\text{PO}_4 \cdot 6\text{H}_2\text{O}$) is a promising option (Liu et al. 2013; Uysal 2015; Muhmood et al. 2019). Struvite has a high potential for replacing conventional synthetic mineral P fertilisers (Rahman et al. 2014; Huygens and Saveyn 2018).

Since the solubility of struvite in water is low when compared to conventional mineral P fertilisers such as monoammonium phosphate and triple superphosphate (TSP), it is considered a slow-release P fertiliser (Degryse et al. 2017; Rech et al. 2019). The agronomic efficiency of struvite depends therefore on the rate at which struvite dissolves in soil as well as the subsequent diffusion of P-PO_4 to the surface of the plant roots where uptake occurs. Kratz et al. (2019) suggested that soils with a different P status could result in a different agronomic efficiency for a given struvite, with a lower P status soil leading to a higher efficiency due to an increase in struvite dissolution. Indeed, the solubility of struvite depends, among others, on the equilibrium concentrations of PO_4 , ammonium (NH_4) and magnesium (Mg) ions in soil pore water, with lower concentrations of these ions driving the dissolution of struvite, and the pH, with a more acidic pH favouring dissolution (Bhuiyan et al. 2007). Furthermore, struvite granular size has been reported to have an impact on its dissolution rate in soil. In a pot experiment conducted by Talboys et al. (2016), struvite granules did not provide P to plants during early growth as fast as conventional diammonium phosphate fertiliser. Degryse et al. (2017) reported that when struvite was ground to powder and mixed with soil, the struvite dissolved significantly faster than granular struvite, due to the increase in soil:struvite contact area. It is also

reported that struvite powder had similar agronomic performances in terms of wheat yield, P content, and P uptake as monoammonium phosphate (Degryse et al. 2017). Indeed, the dissolution rate of struvite is highly dependent on its specific surface area (SSA) (Ariyanto et al. 2017). Compared to struvite granules, struvite powder has higher SSA, leading to faster dissolution in soil pore water. In most literature-reported studies, powdered struvite rather than granular struvite was used to compare the agronomic efficiency of struvite with conventional mineral P fertilisers, which may lead to a bias in the estimation of agronomic performance (Hertzberger et al. 2020). As struvite is mostly sold on the market in the granular form (farmer's equipment being adapted to spread fertiliser as granules), it is important not to overlook the agronomic efficiency of granular struvite.

Although struvite is used primarily as a P fertiliser, it also contains significant amounts of nitrogen (N). Yet, soil N dynamics following struvite application, including nitrous oxide (N_2O) emissions, have received little attention. Urea, accounting for 57% of global N fertiliser demand in 2013/2014 (Heffer and Prud'homme 2016), leads to on average 0.56% of the applied N being lost as N_2O (Luo et al. 2007). Although this is a small part of the N budget, the strong global warming potential of N_2O compared to CO_2 makes it a significant contributor to global warming (IPCC 2022). Some studies indicated that the highest anthropogenic emissions of N_2O are derived from arable land (Reay et al. 2012; Tian et al. 2020). It is therefore valuable to study how struvite application impacts the soil N turnover including N_2O emissions. Liu et al. (2011) estimated the cumulative N_2O emission from soil columns amended with struvite by multiplying the measured total N leaching loss with a N_2O emission factor from the IPCC. The estimated N_2O emissions were lower for the struvite-treated soil than for the urea-treated soil. However, to our knowledge, no study has directly measured the N_2O fluxes from soil after struvite application. It is unknown how powdered and granular struvite affect soil N_2O emissions.

Interactions between P fertilisation and soil N_2O emission have previously been studied but results were inconsistent. In a greenhouse pot experiment, P application significantly improved plant N uptake by alleviating plant P shortage. This resulted in a lower N surplus in the soil, thereby mitigating soil

N₂O emissions after N fertiliser application (Baral et al. 2014). However, Wang et al. (2015) found that P addition increased the soil microbial biomass during wet seasons, potentially leading to a higher microbial activity. The addition of N and P led to a 60% of increase in soil N₂O emission (compared to the control without fertilisation), while the addition of only N increased soil N₂O emission by 39% as compared to the control (Wang et al. 2015). Some studies also showed an increase in soil N₂O emissions after P application, especially on P-limiting soils (He and Dijkstra 2015; Mehnaz and Dijkstra 2016; Mehnaz et al. 2018). Given the fact that struvite results in slow release of both N and P, it is not yet clear how struvite addition would impact the soil N cycle.

In order to address the research gaps identified above, we conducted a greenhouse pot experiment with grass to measure N₂O fluxes from two acidic sandy soils (low and high-P status) amended with powdered or granular struvite. Control treatments with conventional P and N fertilisers were used as a reference. We hypothesised that:

- **H1:** Granular struvite dissolves at a higher rate in a soil with a lower P status. This leads to a higher plant N uptake and thereby to lower cumulative soil N₂O emissions in low-P soil than high-P soil;
- **H2:** Under the same N application rate, struvite application in the granular form leads to lower cumulative N₂O emissions compared to urea application, while having the same agronomic performance;
- **H3:** The soil amended with powdered struvite has higher cumulative N₂O emissions and agronomic performance compared to that of the soil fertilised with granular struvite;
- **H4:** Addition of P in the form of TSP reduces cumulative soil N₂O emissions derived from both urea and granular struvite, as the addition of extra P in both cases will improve the nitrogen use efficiency of struvite and urea.

To test these hypotheses, we applied the following treatments: a control without fertiliser, struvite granules (H1, H2, H3), urea (H2), struvite powder (H3), struvite and TSP combined (H4), urea and TSP combined (H4) and TSP only (H4).

Materials and methods

Soil description

Two acidic sandy soils were used in the greenhouse pot experiment, i.e., a soil with a low soil P status (low-P soil) and a soil with a high soil P status (high-P soil). The low-P soil was collected from the subsoil (30–60 cm) of an arable land in Achterberg, the Netherlands (51°59′32.28″N, 5°35′1.37″E), which had not received P fertilisation for 30 years. The high-P soil was collected from the topsoil of an arable field in Wageningen, the Netherlands (51°59′15.70″N, 5°39′32.46″E) and was regularly fertilised using both organic and mineral fertilisers following local practices prior to sampling. Both soils were air-dried and homogenized for the pot experiment. The properties of the two soils are shown in Table 1. They have a similar texture. However, the low-P-soil had a slightly lower pH and both soils had a different soil organic matter (SOM) content as well as different N and P characteristics. The high-P soil also had a higher SOM and higher mineral N content than the low-P soil. The distinction between the low-P soil and high-P soil was made based on the amount of P that can be extracted from soil by using acid ammonium acetate lactate (P-AL) (Egnér et al. 1960). In the Netherlands, P-AL is routinely used as a soil extraction method in

Table 1 Initial physico-chemical properties of the two acidic sandy soils

Property	Unit	Low-P	High-P
Clay	%	< 1	< 1
Silt	%	15	12
Sand	%	84	87
SOM	%	1.9	3.6
pH ¹	-	5.05	5.66
P-PO ₄ ¹	mg·kg ⁻¹	bd ²	1.1
N-NH ₄ ¹	mg·kg ⁻¹	2.1	4.7
N-NO ₃ ¹	mg·kg ⁻¹	1.7	21.1
Total dissolved N ¹	mg·kg ⁻¹	6	41
Mg ¹	mg·kg ⁻¹	28.8	35.6
P-AL	mg·kg ⁻¹	23	300

¹, Measured in 0.01 M CaCl₂ extract (Houba et al. 2000). ², bd=below detection limit of 0.4 mg·kg⁻¹. A description of the 0.01 M CaCl₂ soil extraction method can be found in Section 2.6 and the analytical details of the other methods can be found in the Supplementary Information

agricultural practice to determine the soil P status for the P fertiliser recommendation system of grassland. Based on P-AL, the soil P status of the low-P soil is considered as very low whereas the soil P status of the high-P soil is considered as high (Reijneveld et al. 2010).

Experimental design

An eight-week greenhouse pot experiment was set up at Wageningen University Campus (Bornsesteeg 48, Wageningen, the Netherlands, 51°59'16.3''N, 5°39'48.5''E), as a fully randomized block design with seven fertilisation treatments and five replicates which were distributed over the five blocks, amounting to a total of 70 PVC pots of 19.5 cm diameter and 23 cm depth. Details of the fertiliser treatments are shown in Table 2. Nitrogen fertilisation rates followed the Dutch fertiliser advice for intensively managed grassland (Commissie Bemesting Grasland en Voedergewassen 2017). To calculate the application rate of struvite, we used the theoretical composition (12.6% of P, 5.7% of N and 9.9% of Mg), which was quite close to the actual composition (Table S1). For all treatments except the control and TSP treatments, we provided 75 kg N·ha⁻¹ at each growing cycle (two growing cycles in total). TSP was applied to ensure a surplus of soil available P at a rate of 50 kg P·ha⁻¹ per growing cycle. All treatments including the control received K fertilisation at 50 kg·ha⁻¹ per growing cycle. In short, we had two treatments using struvite only, either as granule (Sg) or as powder (Sp). We compared these treatments with urea (U) and an unfertilised control (C). To assess the effect of P

addition on soil N₂O emission, we added TSP to both a struvite granule treatment (TSg) and an urea treatment (TU). Finally, we had a P-fertilised control without N but with TSP added (T). Urea, TSP and potassium sulphate were applied in conventional granular form. The struvite used in this study was recycled from a potato peels sludge (NuReSys, Deerlijk, Belgium) and its average granular diameter was 2 mm.

Pot preparation and maintenance

During the pot filling (24th July 2019), potassium nitrate, ammonium nitrate and micronutrient solutions were mixed thoroughly with 6.2 kg of air-dry soil per pot and the application rate of N and K were the same at 50 kg·ha⁻¹ for all treatments including C, to ensure a proper grass cover before the start of the experiment. The soil was packed in the pot in two layers to ensure a homogeneous bulk density of 1.29 g·cm⁻³. Estimations of field bulk density following the method of Rawls (1983) were 1.39 and 1.24 g·cm⁻³ for the low and high-P soils, respectively. A watering tube of 5 cm diameter was inserted in the middle of each pot to avoid artificially modifying the topsoil structure and thus impacting gaseous emissions or increasing the dissolution of fertilisers through more frequent watering in the greenhouse than under field conditions. Ryegrass (*Lolium perenne* cv Barforma) seeds were sown on the soil surface three times until the grass cover was sufficient and homogeneous for all pots (2 × 8 g·m⁻² on the 25th July and two weeks after, and 16 g·m⁻² a month before the start of the experiment).

Table 2 The composition and rate of fertilisers applied in the different treatments during the entire pot experiment

Treatment	Fertiliser use [g·pot ⁻¹] Struvite (12.6% P; 5.7% N)	Urea (46% N)	Triple superphosphate (TSP) (45% P)	Potassium sulphate (25% K)	Fertilisation rate [kg·ha ⁻¹]		
					N	P	K
C	-	-	-	1.12	-	-	100
Sg	7.37	-	-	1.12	150	332 ¹	100
Sp	7.37	-	-	1.12	150	332 ¹	100
U	-	0.91	-	1.12	150	-	100
T	-	-	0.62	1.12	-	100	100
TSg	7.37	-	0.62	1.12	150	332 + 100 ²	100
TU	-	0.91	0.62	1.12	150	100	100

¹, 332 kg·ha⁻¹ P from struvite; ², 332 kg·ha⁻¹ P from struvite and 100 kg·ha⁻¹ P from TSP. The treatment abbreviations refer to the control (C), struvite granule (Sg), struvite powder (Sp), urea (U), TSP (T), TSP + struvite granule (TSg) and TSP + urea (TU)

The experiment started on 28th October 2019 (day 1). On the first day of the experiment, the above-ground biomass was cut at 7 cm above the soil surface. Fifty percent of the fertiliser application rates listed in Table 2 were homogeneously applied on the soil surface (broadcasting) to start the first growing cycle. On 25th November 2019 (day 29), the first growing cycle ended and grass was harvested for plant analysis and soil samples were collected for intermediate soil analysis. Subsequently, the rest of the fertilisers was applied to start the second growing cycle. On 20th December 2019 (day 54), grass was harvested, and soil samples were collected again for analysis and the experiment was terminated. On the two sampling days, the grass was cut at a height of 7 cm above the soil surface. Soil samples were taken within each pot from two randomly selected places (i.e., not purposefully avoiding struvite remnants). The soil sampling depth was 10 cm. After the first growing cycle, the drilling holes were filled with quartz sand after soil sampling. Soil N₂O fluxes were measured 27 times during the 54 days. All pots were watered through the watering tube and the moisture content of the soil was maintained gravimetrically at 117 ml water kg⁻¹ dry soil (i.e., 60% of water holding capacity) on a daily basis. Two 15 mm-rainfall events were simulated by spraying demi-water on the surface of the whole pot on 20th November 2019 (day 24) and 13th December 2019 (day 47), temporarily bringing the soils beyond 100% of water holding capacity.

N₂O flux measurements and calculations

N₂O fluxes were measured overall 27 times during the 54-day pot experiment. The fluxes were determined with the flux-chamber method described by Velthof and Oenema (1995). During the measurement, the top of each pot was sealed by a PVC chamber and the watering tube was closed with a rubber stopper. The headspace volume of the chamber was approximately 4.2 L. The N₂O concentration in the headspace was measured after closing the chamber for 30–40 min, with the exact closing time being recorded, using a photoacoustic gas monitor (Innova, Type 1302) for N₂O measurement. A soda-lime filter was connected before the inlet to eliminate the interference of carbon dioxide. We calculated the increase of N₂O concentration by subtracting the ambient N₂O concentration from the N₂O concentration inside the chamber.

The emission rate of N₂O from soil was calculated based on the assumption that the increase of the concentration in the chamber during the closing period was linear (Velthof and Oenema 1995). To calculate the cumulative emissions, we assumed that the emission rates changed linearly between two measurement dates.

Plant analysis

The plant samples were dried at 70°C for 48 h in order to determine dry yields. The dried plant samples were finely ground for N and P content analysis. A mixture of sulfuric acid and hydrogen peroxide was used to digest the plant material (Novozamsky et al. 1983). The N and P concentrations in the digests were measured by a segmented flow analyser (SFA; Skalar, SAN⁺⁺). Although some N and P must have been present in the grass roots and stubbles, we hereafter refer to plant uptake as the amount of N and P in the harvested grass shoots. The nitrogen use efficiency (NUE) of the grass was calculated as:

$$\text{NUE (\%)} = \left[\frac{U - U_0}{F} \right] \times 100$$

in which NUE is the N use efficiency expressed as a percentage, U is the amount of N taken up by the grass in the fertiliser treatment for both harvests, U₀ is the amount of N taken up in the control treatment for both harvests, and F is the total amount of N applied (Table 2), with U, U₀ and F all expressed in kg N·ha⁻¹.

In this study, we defined the agronomic performance of a fertiliser as the effect of a given fertiliser on the yield and the nutrient uptake of the grass.

Soil analysis

All soil samples collected on day 29 and day 54 of the pot experiment were air-dried (40°C) and sieved through a 2 mm-screen. To determine the readily plant-available amounts of N and P, soils were extracted with 0.01 M CaCl₂ using a soil-to-solution ratio of 1:10 (g:ml) and a shaking time of 2 h (Houba et al. 2000). After centrifugation at 3000 rpm for 10 min and subsequent 0.45-µm filtration (Aqua 30, Whatman) of the soil extracts, the total dissolved Mg concentration was measured by an inductively

coupled plasma-optical emission spectrometry (ICP-OES, Thermo scientific, ICAP 6000) whereas the concentrations of N-NH₄, N-NO₃ and P-PO₄ were measured by SFA (Skalar, SAN++). Strictly speaking, where we report the N-NO₃ content, the sum of N-NO₃ and N-NO₂ is meant.

Furthermore, an additional experiment was performed in which we quantified the dissolution of struvite in 0.01 M CaCl₂ solution, separately and in combination with soil, up to an extraction time of 96 h. A description and short discussion of the results of the experiment can be found in the Supplementary Information.

Statistical analysis

Statistical analysis of data was performed within SPSS 25.0 software (SPSS Inc., Chicago, IL, USA). Before conducting any statistical test, normality of residuals and homogeneity of variances were examined. Normality was firstly checked with the Shapiro–Wilk test. If the assumption of normality was violated, we further checked the normality visually with a Normal Quantile–Quantile plot. The homogeneity of variances was examined with the Levene test. If the variances were homogeneous, we conducted one way ANOVA with Tukey HSD post hoc test. In the case of heterogeneity of variances, Games-Howell test was the alternative. When the p-value was smaller than 0.05, we rejected the null hypothesis of equal means. As for dealing with the violation of normality, data transformation (Log transformation and Box-Cox transformation) would be applied. If data transformation methods failed to normalize data, the bootstrap module in SPSS would be used. In this case, statistical significance was obtained by examining the 95% confidence interval of the difference between two means. If the confidence interval was intersected with zero, two means were not statistically different. Relations between soil parameters and plant parameters with cumulative N₂O emission were probed by Pearson Bivariate Correlation. Pearson *r* was used to interpret the magnitude of correlation. In order to control the confounding variables, Partial Correlation was also performed as a supplement to the bivariate correlation. The interpretation of the magnitude of correlation was in line with the guideline proposed by Gignac and Szodorai (2016).

Results

Grass performance

Figure 1a shows the aboveground yield of grass. For the low-P soil, the largest yield was observed for the Sp and TU treatments. The sole application of TSP did not increase yield compared to that of the C treatment whereas the application of urea without any P addition (U treatment) significantly increased yield. The yield of the Sp treatment was significantly larger than that of the Sg treatment, and the yield of the Sg treatment was significantly higher than that of the U treatment. The yields of the TSg and TU treatments were not statistically different. In the high-P soil, similar to that of the low-P soil, the two lowest yields were observed for the C and TSP treatments that did not receive any N fertilisation. The application of struvite powder and urea resulted in a significantly larger yield compared to the C treatment. The yields of the Sg, TSg, and TU treatments were not statistically different from each other and that of the C treatment. The two soils resulted in very different yields for the same treatments. When comparing the yields of the C treatments, there is an average yield difference of 3301 kg·ha⁻¹ between the low-P soil and the high-P soil.

Aboveground N uptake of grass is shown in Fig. 1b. In the low-P soil, the TU treatment had the largest N uptake, followed by that of Sp. Similar to the yield, the two treatments with the lowest amounts of N taken up were the C and T treatments. For the Sg, U and TU treatments, the N uptake was not statistically different from each other. However, in the high-P soil, N uptake of the U treatment was significantly higher than that of the Sg and TSg treatments. The lowest N uptake was also observed for the C and TSP treatments which did not include any N fertilisation. For each treatment, N uptake was higher for the high-P soil than for the low-P soil. For example, N uptake in the C treatment of the high-P soil was higher than 135 kg·ha⁻¹, but it was only about 10 kg·ha⁻¹ in the low-P soil.

For both soils, aboveground P uptake of both Sg and Sp treatment was significantly higher than that of the C treatment (Fig. 1c). On both soils, the highest P uptake was realised in the Sp and TSg treatments, followed by the Sg treatment. On the high-P soil, the T treatment had lower P uptake than the Sg treatment.

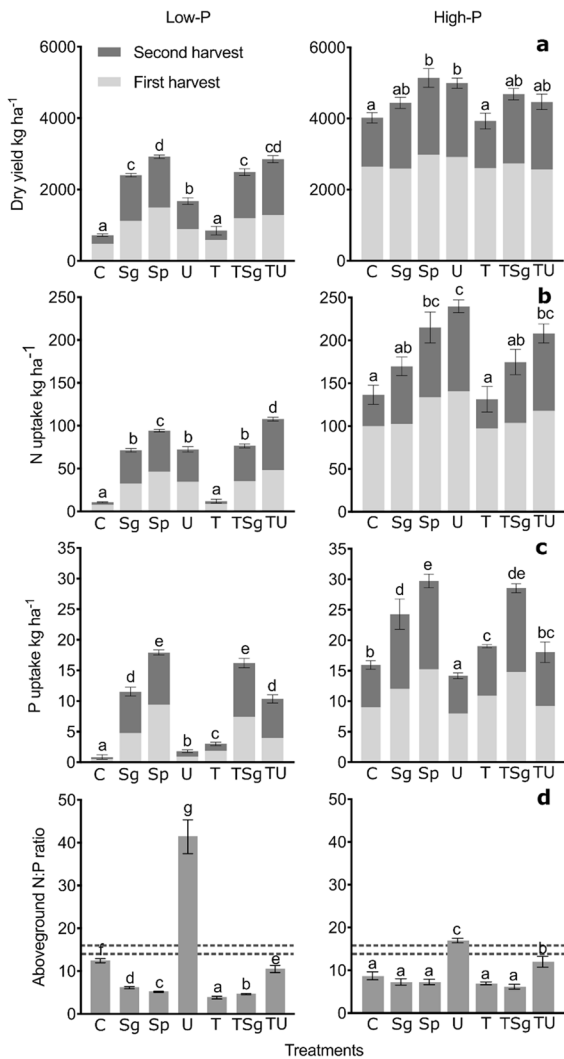


Fig. 1 Grass characteristics per treatment and per soil ($n=5$). **a** Aboveground dry yield. **b** Aboveground N uptake. **c** Aboveground P uptake. **d** Aboveground N:P ratio, dashed line represent commonly acknowledged N and P limitation thresholds (14 and 16 respectively). The bars in a, b and c are split by harvest; error bars denote the standard errors of the means of the sum of both harvests. d is averaged for both harvests and error bars are standard errors of the means. Letters indicate significant differences in overall dry yields. Letters for a and b were obtained by the Tukey HSD test ($p < 0.05$). Significant differences for c and d were obtained by the bootstrapped Games-Howell test (the 95%-confidence interval of the difference of two means does not intersect with zero). The seven treatments are control (C), struvite granule (Sg), struvite powder (Sp), urea (U), TSP (T), TSP+struvite granule (TSg) and TSP+urea (TU)

The TU treatment was not different from that in the C treatment, while the U treatment had the lowest P uptake. On the low-P soil, the TU treatment had the same P uptake as the Sg treatment. Interestingly, the C, U and T treatments all had a much lower P uptake than the treatments in which a fertiliser source with both N and P was provided. Again, the P uptake differed strongly for all treatments of the two soils. For example, the P uptake of the control in the high-P soil was higher than $15 \text{ kg}\cdot\text{ha}^{-1}$, but it was lower than $5 \text{ kg}\cdot\text{ha}^{-1}$ in the low-P soil.

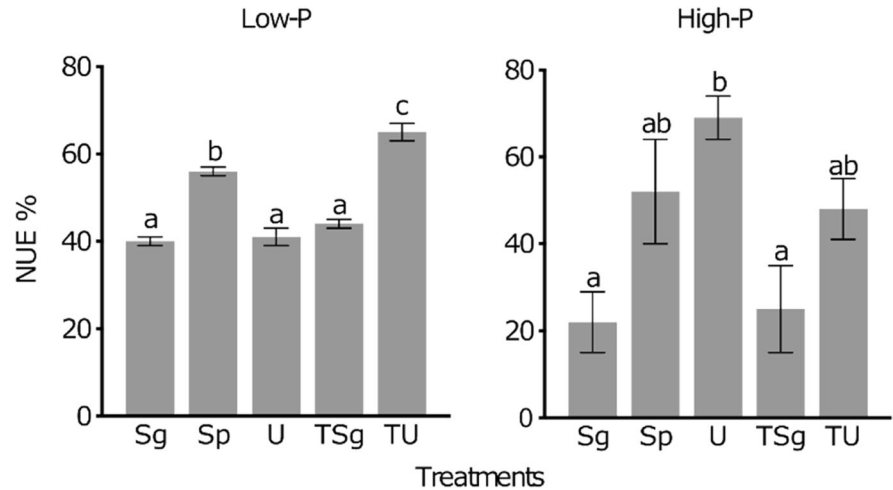
The U treatment led to the largest aboveground N:P ratio which was even above 30 in the low-P soil (Fig. 1d). The second largest N:P ratio was found in the C treatment without any fertilisation. In addition, the Sp treatment resulted in a significantly lower aboveground N:P ratio than that of the Sg treatment. The lowest aboveground N:P ratio was observed with the T treatment. All treatments with P addition had a lower aboveground N:P ratio than that of the C treatment. In the high-P soil, the largest N:P ratio was still obtained for the U treatment. However, the second largest value was from the TU treatment. Struvite application did not significantly change the aboveground N:P ratio compared to the control treatment. In both soils, the application of only urea strongly increased the aboveground N:P ratio.

A similar pattern in NUE (Fig. 2) for the different treatments receiving N fertiliser was found as that of aboveground N uptake (Fig. 1b). For the low-P soil, the TU treatment had the highest NUE followed by that of Sp. The treatment with only urea application (U) led to the lowest NUE, which was not statistically different from that of the other two treatments of struvite granule application (Sg and TSg). For the high-P soil, the U treatment had a significantly higher NUE compared to that of Sg and TSg. The P use efficiency (PUE) was calculated in the same manner as the NUE; results are shown in the Supplementary Information (Table S2).

Readily plant-available soil nutrients

Table 3 shows the results of the extraction of the soil samples taken at the two sampling dates with 0.01 M CaCl_2 . For the treatments with struvite addition (Sg, Sp and TSg) of both soils, the CaCl_2 -extractable amount of N-NH_4 was significantly higher on days 29 and 54 compared to the C treatment. The same

Fig. 2 NUE (%) for the treatments receiving N fertilisation for the two soils. Error bars are standard errors of the means. Lowercase letters indicate significant differences. The significant differences were obtained by the Tukey HSD test ($p < 0.05$). Seven treatments are control (C); struvite granule (Sg); struvite powder (Sp); urea (U); TSP (T); TSP + struvite granule (TSg) and TSP + urea (TU)



is true for the extracted amounts of P-PO₄. Interestingly, the extracted amounts of N-NH₄ and P-PO₄ for the treatments receiving powdered struvite (Sp) and granular struvite (Sg and TSg) of the low-P soil did not differ significantly on day 29. Likewise, no significant differences were found for the Sg and Sp treatments of this soil on day 54. However, the extracted amounts of N-NH₄ and P-PO₄ for the Sp treatment of the high-P soil were significantly higher than for the

Sg treatment on day 29. These differences remained intact for the extracted amounts of P-PO₄ on day 54. With respect to N-NO₃ for the low-P soil, the largest amount was extracted for the U, TU and TSg treatments on day 29. On day 54, the U treatment still had the largest extractable amount of N-NO₃, but we did not observe significant differences in the extracted amounts of N-NO₃ across the other treatments. For the high-P soil, the Sp, U and TU treatments had the

Table 3 Results of 0.01 M CaCl₂ extraction of soil samples in the seven treatments of the low-P soil and the high-P soil (soil samples collected on day 29 and day 54). The results of pH and Mg are presented in Supplementary Information (Table S3)

		Day 29			Day 54		
		N-NH ₄ mg·kg ⁻¹	N-NO ₃ mg·kg ⁻¹	P-PO ₄ mg·kg ⁻¹	N-NH ₄ mg·kg ⁻¹	N-NO ₃ mg·kg ⁻¹	P-PO ₄ mg·kg ⁻¹
Low-P	C	0.5(0.1)b	0.1(0.0)a	0.00(0.00)a	0.3(0.0)a	0.2(0.0)a	0.03(0.03)ab
	Sg	13.6(1.7)d	0.3(0.1)b	1.02(0.38)c	14.0(5.1)de	0.1(0.0)a	7.59(3.37)cd
	Sp	6.9(3.0)cd	0.2(0.0)b	0.74(0.26)c	3.4(0.5)d	0.2(0.0)a	1.12(0.40)c
	U	7.3(2.8)cd	2.3(0.9)c	0.00(0.00)a	7.6(2.7)d	3.1(1.4)b	0.00(0.00)a
	T	0.2(0.0)a	0.1(0.0)a	0.02(0.02)b	0.5(0.1)b	0.1(0.0)a	0.12(0.04)b
	TSg	10.3(3.2)cd	0.5(0.2)bc	0.99(0.26)c	16.3(2.2)e	0.1(0.0)a	9.37(2.35)d
	TU	5.3(1.3)c	1.9(0.9)c	0.00(0.00)a	1.8(0.2)c	0.1(0.1)a	0.02(0.02)a
High-P	C	2.6(0.5)a	9.3(3.4)a	1.00(0.04)a	1.4(0.1)a	0.9(0.3)a	0.82(0.04)a
	Sg	13.4(0.8)c	9.8(4.8)a	3.57(0.50)c	11.3(2.8)bc	2.1(0.8)ab	5.20(1.53)d
	Sp	23.9(3.8)d	19.4(7.0)ab	12.52(2.49)d	20.1(6.4)c	2.3(0.7)b	14.93(3.85)e
	U	3.4(0.9)ab	45.5(18.4)b	1.10(0.09)ab	3.0(0.3)b	29.2(13.2)c	0.98(0.07)b
	T	3.3(0.4)a	5.5(2.0)a	1.00(0.07)ab	1.8(0.2)a	0.7(0.2)a	1.57(0.22)c
	TSg	21.4(4.3)d	7.5(3.2)a	11.33(6.21)cd	22.5(6.3)c	1.5(0.9)ab	43.29(21.30)e
	TU	9.8(4.0)bc	35.2(13.3)b	1.30(0.13)b	8.4(2.9)b	19.6(9.9)c	1.76(0.39)c

Data show average values with standard errors in brackets ($n=5$). Letters indicate significant differences. Significant differences were obtained by the bootstrapped Games-Howell test (The 95% confidence interval of the difference between two means does not intersect with zero).

highest amounts of extracted N-NO₃ on day 29. The amount of extractable N-NO₃ for all three treatments with struvite addition did not differ significantly from the amount of N-NO₃ extracted from the C treatment. For day 54, still no differences were found for these treatments. Similar to the low-P soil, the U and TU treatments again resulted in the largest amounts of extracted N-NO₃ on day 54.

Cumulative soil N₂O emissions

The two soils showed distinctive cumulative soil N₂O emission patterns (Fig. 3). In general, the N₂O fluxes measured from the low-P soil were substantially lower than those from the high-P soil. Rain events on day 24 and 47 led to an increase in N₂O fluxes for both soils, especially for the U and TU treatment on the high-P soil. For the low-P soil, none of the fertiliser treatments resulted in a significant increase in cumulative N₂O emission (one-way ANOVA, $F=0.453$, $p=0.837$). For the high-P soil, however, both treatments receiving urea (U and TU) resulted in a significant increase in the cumulative N₂O emission, with the N₂O emission peaking at 239 g N ha⁻¹ for the U treatment. In both soils, neither Sg nor Sp led to a significant increase in the N₂O emission compared to the C treatment, meaning that the grinding of struvite did not have a significant effect on the N₂O emissions. We also calculated the N₂O emission factors, with application of soluble N fertiliser (urea) resulting in significantly higher N₂O emission factors than the other treatments in the high-P soil (Table S4). The daily N₂O fluxes from each measurement are presented in Table S5. For the high-P soil, we found a

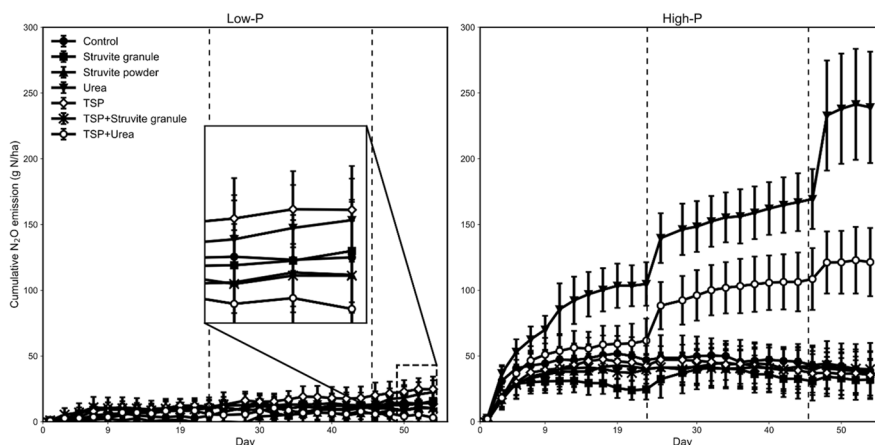
significant positive correlation ($r=0.59$, $p<0.001$; $n=35$) between the cumulative N₂O emission and CaCl₂-extractable N-NO₃ content. However, no such correlation was found in the low-P soil ($p=0.277$). Furthermore, there was a significant positive correlation between the cumulative N₂O emission and the N:P ratio of the aboveground biomass for the high-P soil ($r=0.68$, $p<0.001$, $n=35$).

Discussion

Agronomic performances of struvite in relation to soil P status

We hypothesized that granular struvite would dissolve at a higher rate on a soil with a lower P status. This would then lead to a higher plant N uptake and thereby lower cumulative soil N₂O emissions from the low-P soil than that from the high-P soil. However, we did not measure the actual dissolution rate of struvite as this would be technically challenging because any P released to solution upon struvite dissolution is to a large extent immediately captured through adsorption by the soil solid phase. By using labelled ³³P, a diffusion test showed that most P released by a single struvite granule could not travel more than 1 mm distance in the soil, while the P released by TSP travelled more than 25 mm, indicating that any P dissolved from struvite that has a low solubility would be captured by the soil (Rech et al. 2019). The fast adsorption of P by the soil solid phase after struvite dissolution is further supported by the results of the test that we performed to quantify the

Fig. 3 Cumulative N₂O emissions from two soils. Error bars represent standard errors of means ($n=5$). Dotted lines indicate two simulated rain events (day 24 and day 47). Fertilisers were applied on day 1 and day 29. For the low P soil, the results of the last three measurements are amplified in the inset figure



dissolution of struvite during soil extraction with 0.01 M CaCl_2 (Section 8.2, Supplementary Information). The concentration of P-PO_4 in 0.01 M CaCl_2 kept increasing over time (up to 96 h) when struvite was extracted in the absence of soil. However, when the struvite-amended soil was extracted, hardly any P-PO_4 could be detected in the solution (Fig. S1A).

Nevertheless, we did extract soil samples taken at two sampling times from all treatments of the two soils with 0.01 M CaCl_2 (Table 3) to mimic soil pore water conditions (McDowell and Sharp-ley 2001). The results were surprising because of the large amounts of N-NH_4 extracted compared to those of N-NO_3 . The ratios between N-NH_4 and N-NO_3 observed in the CaCl_2 soil extracts of the Sg-treated soils on day 29 were on average 45.3 and 1.3 for the low and high-P soils respectively. Treatments without struvite showed considerably lower ratios, ranging from 2 to 5 and >0.1 to 0.8 for the low and high-P soils, respectively (Table 3), which is comparable to the ratios of the soils before the start of the experiment (Table 1) as well as ratios in cultivated soil with similar texture (Łukowiak et al. 2017). The difference in the ratio between N-NH_4 and NO_3 for the Sg-treated soils and for the soils that did not receive struvite is even more pronounced at day 54 than at day 29. The Sg-treated soils had average ratios of 140 and 5.3 for the low and high-P soils, respectively, while the treatments without struvite stayed within similar range as on day 29, with ratios from 1.5 to 18 and 0.1 to 2.5 for the low and high-P soils, respectively (Table 3). Under normal conditions, it is expected that soil NH_4 from fertilizer application would be shortly nitrified. Moreover, these apparently high mineral N-NH_4 contents in the soil were contradictory to plant N limitations in the treatments with struvite (Fig. 1d). We visually observed that not all of the applied struvite dissolved during the greenhouse pot experiment. Unfortunately, these undissolved struvite particles could not be separated from soil during sample preparation due to their small diameters. Thus, it is suspected that some of the remaining struvite must have at least partially dissolved during the 2-h CaCl_2 extraction procedure of our soil samples. Struvite dissolution does occur during 2-h CaCl_2 extraction (Section 8.2, Supplementary Information) as well as during other commonly used soil P tests (Gu et al. 2021). Thus, for the treatments

containing struvite in our pot experiment, the CaCl_2 extraction most likely did not reflect the actual readily plant-available soil N-NH_4 . Since grass growth in the treatments containing struvite was N-limited (Fig. 1d), it is likely that the actual level of readily plant-available N-NH_4 was low and that any dissolved NH_4 from struvite during the growing period would have been quickly taken up by the grass. The residual struvite in soil poses a substantial analytical challenge in evaluating the true nutrient availability of the soil. Especially when struvite is applied under field conditions, separating undissolved struvite residues from soil is difficult. By comparing various soil P extraction methods, Gu et al. (2021) suggested that sink-based P test might be the most reliable method to assess the plant-available P.

Although the CaCl_2 extraction method failed to realistically reveal the readily plant-available nutrients derived from struvite in the soils, the calculated NUE could be an alternative proxy. Indeed, the grass would have taken up any plant-available N as both soils were N-limited, and a comparison between the difference in N uptake of the C and Sg treatments in each soil can give some quantitative idea to which extent struvite dissolved during the pot experiment. Struvite granules (Sg treatment) led to a NUE twice as high on the low-P soil than on the high-P soil (Fig. 2). Assuming that any N from dissolved struvite would have been taken up, this suggests that struvite granules did indeed dissolve faster in the low-P soil than in the high-P soil. However, this did not lead to a higher N uptake in the Sg treatment for the low-P soil than for the high-P soil as the amount of N supplied by the latter without any fertilisation overcompensated this difference in struvite dissolution. The N uptake of grass in the C treatment of the high-P soil was more than 12 times higher than in the low-P soil (Fig. 1b). This can be explained by the higher amounts of readily plant-available N as well as the higher SOM in the high-P soil before the start of this experiment (Table 1). We can only partially accept H1 as struvite granules seem to dissolve faster on the low-P soil but this apparently did not translate into a higher N uptake for the very same reason: a soil with low levels of plant-available N and P would lead to a faster dissolution and thus a better performance of struvite granules but would also intrinsically provide less N for uptake by the plants. In this respect, struvite granules can be considered as an “on demand”

fertiliser i.e., mineral fertiliser with a low solubility, providing more N when the soil is lacking N, and less N when the soil is already quite fertile.

Agronomic performances and N₂O emission: struvite vs urea

Our second hypothesis stated that under the same N application rate, struvite application in the granular form would lead to lower cumulative N₂O emissions compared to urea application, while having the same agronomic performances.

In the high-P soil, Sg led to a statistically similar yield as U, while in the low-P soil struvite led to a higher yield than U (Fig. 1a). This can be explained by the strong P limitation of the U treatment in the low-P soil (Fig. 1d). When TSP was combined with urea in the TU treatment for the low-P soil, this limitation was alleviated (Fig. 1d) and a similar yield as with struvite was reached (Fig. 1a). Additionally, in the low-P soil, Sg and U led to the same N uptake but this was not the case in the high-P soil where U led to a significantly higher N uptake than Sg (Fig. 1b). However, since the high N uptake of the U treatment did not lead to an increased yield (Fig. 1a) and showed P limitation (Fig. 1d), this hints at the possibility that the plant had more N than what it could actually use. In other words, part of the N that was taken up by the grass in the U treatment is due to luxury uptake. This is further supported by the effects of applying TSP combined with urea in the TU treatment of the high-P soil: the TU treatment had a N uptake comparable to the Sg treatment (Fig. 1b). With respect to P, the Sg treatment had a clear advantage over U in both soils. When TSP was combined with urea in the TU treatment, similar levels of P uptake were found in the low-P soil as compared to the Sg treatment but not in the high-P soil where Sg had a more positive effect on P uptake than TU. Although surprising, this result could be explained by the nature of the P fertilisers. From literature it is known that most of the TSP dissolves in soil within 24 h after application while struvite dissolves slowly over 60 days or even more depending on soil properties like pH (Lawton and Vomocil 1954; Degryse et al. 2017). Dissolution of struvite in the high-P soil may have proceeded at a lower rate than in the low-P soil, because the initial pH and concentrations of N-NH₄, P-PO₄, and Mg in the CaCl₂ soil extracts of the high-P soil were higher

than for the low-P soil (Table 1). For P uptake by plants, it may be more advantageous to have a low but regular flow of P arriving from soil in the pore water than one large flush of P: in the first case, plant roots have a higher chance to intercept this P before it binds to the soil while in the second case, plants cannot take up that much P within a given short period of time meaning that the excess P will be bound by the soil and as such be less available for uptake. Overall, Sg showed a rather similar agronomic performance as U. The slow release of P from struvite dissolution over time may be considered as an advantage to provide P to the soil as compared to TSP.

N₂O emissions originate from nitrification and denitrification and thus depend on the availability of N-NH₄ and N-NO₃ in soil. Because of residual struvite dissolving during CaCl₂ extraction (Section 8.2, Supplementary information), we cannot use the measured readily plant-available N-NH₄ contents as a reliable source of information for how much N-NH₄ was actually present in these soils. The CaCl₂-extractable N-NO₃ contents, however, are not impacted by struvite dissolution during CaCl₂ extraction (Section 8.2, Supplementary information). In both soils, Sg showed lower N-NO₃ concentrations than the U treatment (Table 3). This is consistent with the findings of another pot experiment where the total amount of N leached from soil amended with struvite was about three times less than that of urea in a column experiment (Liu et al. 2011). This seems to suggest that the actual soil N-NH₄ concentration for Sg was also lower than for U. This is in line with the N₂O emissions we observed for the high-P soil: we found a significantly lower cumulative N₂O emission from the Sg treatments compared to that of U (Fig. 3). Thus, in the high-P soil, the slow release of nutrients originating from struvite did not increase N₂O emissions. This is in line with studies reporting that slow-release N fertilisers such as slow-release urea can significantly decrease N₂O emission compared to that of traditional urea, as used in our study (Awale and Chatterjee 2017; Trinh et al. 2017). In the low-P soil, the average cumulative emission of Sg was lower than that of U (15.7 and 22.5 g N-N₂O·ha⁻¹ respectively) but this was not significantly different. The emissions were low because the soil had very little N to start with (Table 1) and a large part of the fertiliser added was taken up by the plants, especially when addition of N was combined with addition of P (Fig. 3). This is

further supported by the significant positive correlation we found in the high-P soil between N-NO₃ and cumulative N₂O emissions, in line with many previous studies that identify denitrification as the main source of N₂O emissions following N application (Mehnaz and Dijkstra 2016; Ji et al. 2020; Zhou et al. 2020). Although we did not find significant differences in the cumulative N₂O emissions for the low-P soil, we did find a significantly lower N-NO₃ concentration for Sg than for U. This does suggest that N₂O emissions may in general be lower for Sg than for U. With Sg having a rather similar agronomic performance as U, and N₂O emissions being lower than for the former, we can accept our second hypothesis.

Agronomic and climatic impact of powdering struvite

Our third hypothesis stated that the soil amended with powdered struvite would have higher cumulative N₂O emissions and agronomic performance compared to that of the soil fertilised by granular struvite. In the low-P soil, the Sp treatment led to higher yield, N uptake and P uptake compared to that of the Sg treatment (Fig. 1a, b, c). In comparison to TU, Sp had a similar yield, a slightly lower N uptake and a higher P uptake. A similar pattern was observed in the high-P soil, although results of yield and N uptake were not statistically different between Sg and Sp. The higher NUE for Sp in both the low and the high-P soil (significant difference for the low-P soil only) (Fig. 2) indeed seems to suggest a faster dissolution of the struvite powder. Grinding struvite increases its SSA i.e., surface area per unit of mass. Similar to rock phosphate, struvite with a higher specific surface area will dissolve faster than the one with a lower SSA, all other soil properties being equal (Degryse et al. 2017). This is because the dissolution of struvite occurs at the particle surface, which comes into contact with soil pore water as the solvent. A higher SSA means that there is a higher mineral surface area exposed to the soil pore water.

As the N-NO₃ concentrations in the Sg-treated and Sp-treated soils were similar in both soils (Table 3), we can argue that N dissolved from the struvite, in granules or in powder, was taken up in similar amounts by the grass, which resulted in identical N₂O emissions for Sp and Sg from both soils. Therefore, contrary to our hypothesis, the dissolution of powdered struvite does appear to have gone faster than

that of granular struvite, leading to a better agronomic performance of the fertiliser, but without impacting N₂O emissions. Therefore, our third hypothesis was rejected. Our results seem to suggest that reducing the granular size could increase the agronomic performance of struvite fertiliser without sacrificing its ability to mitigate N₂O emission. Some studies have suggested that in order to deal with the low solubility of struvite, it could be applied in combination with soluble fertilisers such as monoammonium or diammonium phosphate, especially in the early growth stage when plants may require a faster nutrient supply (Ackerman et al. 2013; Talboys et al. 2016). Combining struvite with conventional fertiliser may increase risks of nutrient leaching and N₂O emissions. Our results demonstrate that a good alternative solution to a lack of nutrients in the early growth stage of plants can be to provide (at least some of) the struvite in powdered form. This could be done by pelletising powdered struvite so that it could be applied with conventional fertiliser application equipment. In this form it would still be more soluble than granular struvite. Alternatively, regular equipment could be adapted to apply powder instead of granules. Research on how powdered struvite can be applied in practice needs further attention.

Effects of P addition on N₂O emissions

Our fourth hypothesis stated that the addition of P in the form of TSP would reduce cumulative soil N₂O emissions derived from both urea and granular struvite, as the addition of extra P in both cases would improve the NUE of struvite and urea. In the low-P soil, TSP addition did not have any measurable influence on N₂O emission in either of the treatments because the emissions were all very low, even for the U treatment (Fig. 3). However, TSP addition did increase the N uptake of grass fertilised with urea (Fig. 1b). As mentioned in Section 4.2, plants of the U treatment in the low-P soil probably accumulated luxury N and did not leave sufficient N available in the soil to result in significant N₂O emissions. The combination of TSP and urea meant that plants could use this N to invest in growth (Fig. 1a). In the high-P soil, TU had much lower N₂O emissions than U (Fig. 3). However, we did not observe such an effect for struvite granules (TSg vs Sg). This can be explained by the slow release of N from struvite granules as compared to urea

(see Section 4.2). Interestingly, although adding TSP to urea reduced N_2O emissions, we did not find that P addition led to a higher aboveground N uptake in the high-P soil (Fig. 1b). We also did not find that the N was significantly immobilized by the soil microbes in the treatments with TSP (Table S6). Thus, it is possible that the rest of N might be present in the plant roots, which were not sampled in this study.

Koerselman and Meuleman (1996) proposed to use the N:P ratios of vegetation to represent nutrient limitation on a community level. Although they used a N:P ratio of 14 and 16 as the thresholds for N (<14) and P limitation (>16), a study has shown that indicating nutrient limitation by a canonical N:P ratio was questionable (Yan et al. 2017). Using more conservative thresholds of 10 and 20, instead of the commonly used thresholds of 14 and 16, was leading to better estimation of N and P limitation. The N:P ratio should thus be used with caution. Yet, in general a high N:P ratio of vegetation indicates P limitation for the plant community, while a low ratio implies N limitation. In both our soils, any form of struvite fertilisation led to a low N:P ratio of the grass (<10), which shows that grass growth under struvite fertilisation was possibly N-limited (Fig. 1d). U and TU resulted in P-limited or less N-limited conditions compared to struvite fertilisation, although only U in the low-P soil had a N:P ratio higher than 20. Since N was still a limiting resource to grass after the struvite fertiliser application, grass might utilise various strategies to acquire as much N as they can take up from soil (Gutschick 1981; Tolley and Mohammadi 2020). In the high-P soil where urea application led to an increase of cumulative N_2O emission, a significant positive correlation was found between aboveground N:P ratio and the cumulative N_2O emission (partial correlation; soil surplus NO_3 as the control variable). This demonstrates that the more severe the P shortage was for plant growth (or the less N-limiting the system was), the more N was lost through N_2O . The reason for struvite mitigating N_2O emission is therefore that it potentially shifts the system towards N limitation.

Conclusion

This study reports the first measurement of soil N_2O fluxes after struvite fertilisation. Under the same N

application rate, struvite has lower cumulative N_2O emissions compared to those of urea. The direct application of granular struvite did not lead to yield penalties on either soil. Increasing the contact area between struvite and soil through grinding resulted in higher N and P uptake in comparison to regular struvite granules. Our results show that the powdered struvite application could increase the agronomic performance of this fertiliser without increasing N_2O emissions. Although struvite fertilisation may pose a concern of N deficiency if applied in regular doses, on both soils the aboveground dry yield from the struvite powder treatment was the highest of all treatments. This greenhouse study showed that the use of struvite led to lower N_2O emission as compared to a conventional N fertiliser like urea and these results need further confirmation in the form of a field experiment.

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Data Availability This study's raw data will be stored in the Zenodo repository. The link is available from the corresponding author upon request.

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