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Lysimeter deep N fertilizer placement reduced leaching and improved N use efficiency

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Abstract Deep fertilization has been tested widely for nitrogen (N) use efficiency but there is little evidence of its impact on N leaching and the interplay between climate factors and crop N use. In this study, we tested the effect of three fertilizer N placements on leaching, crop growth, and greenhouse gas (GHG) emissions in a lysimeter experiment over three consecutive years with spring-sown cereals (S1, S2, and S3). Leaching was additionally monitored in an 11-month fallow period (F1) preceding S1 and a 15-month fallow period (F2) following S3. In addition to a control with no N fertilizer (Control), 100 kg N ha⁻¹ year⁻¹ of ammonium nitrate was placed at 0.2 m (Deep), 0.07 m (Shallow), or halved between 0.07 m and 0.2 m (Mixed). Deep reduced leachate amount in each cropping period, with significant reductions (p < 0.05) in the drought year (S2) and

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Department of Ecology, Swedish University of Agricultural Sciences, Box 7044, 750 07 Uppsala, Sweden cumulatively for S1-S3. Overall, Deep reduced leaching by 22, 25 and 34% compared to Shallow, Mixed and Control, respectively. Deep and Mixed reduced N leaching across S1-S3 compared with Shallow, but Deep further reduced N loads by 15% compared to Mixed and was significantly lowest (p < 0.05) among the fertilized treatments in S1 and S2. In S3, Deep increased grain yields by 28 and 22% compared to Shallow and Mixed, respectively, while nearly doubling the agronomic efficiency of N (AE_N) and the recovery efficiency of N (RE_N). Deep N placement is a promising mitigation practice that should be further investigated.

Keywords Deep N fertilization · Drought · Fertilizer placement · Lysimeter · Nitrate leaching

Introduction

As essential nitrogen (N) is to crop production, its use in agriculture is difficult to manage due to its mobility and rapid transformation in the soil leading to air and water pollution, partially derailing its intended path to the plant. In humid climatic conditions, dissolved nitrate (NO_3^-) and nitrite (NO_2^-) are transported through the soil into ground- and surface water and further into streams, lakes and coastal areas, contributing to eutrophication. Nitrous oxide (N_2O) emissions from microbial nitrification and denitrification of fertilizer N are major sources of greenhouse gases (GHG) emitted within the agricultural sector (Nabuurs et al. 2022). Population growth, which necessitates greater crop production, and thus fertilizer, is on an upwards trajectory (FAO 2019; UN 2022), presenting a particular challenge at a time when we are seeking to reduce GHG emissions to keep the global temperature increase below the 1.5 °C target (IPCC 2018). In order to meet both United Nations sustainable development goals for increasing the food supply (goal 2), while reducing the negative environmental impacts of fertilization (e.g., goals 13 and 15) (UN 2015), we need to rapidly test and employ new methods to increase fertilizer N use efficiency (NUE).

One such method is deep fertilizer placement, which multiple studies have shown can positively influence crop production and minimize fertilizerinduced environmental damage. In the literature, what qualifies as a deep placement depth can differ widely, depending on the existing local practice for fertilization for the particular cropping system. Although placement depth, as well as climate and growing systems vary, deep N placement relative to surface fertilization has been reported to increase yields and NUE, decrease ammonification, and in some cases decrease N₂O emissions (Chen et al. 2021; Pandit et al. 2022; Rychel et al. 2020; Sosulski et al. 2020; Wu et al. 2021; Zhang et al. 2022;). However, NO_3^- remaining in the soil after crop uptake is susceptible to leaching and literature on the fate of NO_3^- following deep fertilization is scant. Ke et al. (2018) reported high NO_3^- losses in a flooded rice system, whereas Wu et al. (2022) found that deep placement at 0.25 or 0.15 m decreased NO₃⁻ content in the 0-1 m depth compared to a shallow (0.05 m) placement in a field experiment with maize. Wang et al. (2022) reported that deep urea placement promoted the proliferation of deep roots in winter wheat, which increased crop N uptake and water utilization, but NO₃⁻ leaching varied depending on seasonal rainfall amount. There is little to no information, however, regarding the longer-term effects of N fertilizer placement on NO₃⁻ leaching.

In a Swedish field experiment, deep N fertilization was shown to increase yield and N uptake while simultaneously decreasing N_2O emissions (Rychel et al. 2020). Although soil mineral N levels were measured multiple times during the growing season in that field experiment, the fate of the remaining N in the soil not removed by crops after the growing season (or below our sampling depth) was unknown. Therefore, we performed an additional experiment using undisturbed soil monoliths (lysimeters) taken from the same field as the previous experiment, in which we could quantify the nutrient load in the leachate following treatments with several N fertilization strategies.

In the eastern region of central Sweden, where cereals are the dominant crops, the local agronomic practice is to place fertilizer at 0.07 m and seeds at 0.05 m simultaneously using, for example, a Combi drill. Thus, we used a baseline 0.07 m depth for a shallow fertilizer N placement and 0.2 m for deep placement, with the motivation that at 0.2 m, soil moisture and temperature are relatively more constant compared to the shallow placement depth. Thus, fertilizer N placed at 0.2 m would be less susceptible to mobilization following rainfall events and pulses in nitrification and denitrification with temperature and moisture fluctuations. Moreover, the common depth for harrowing (performed in spring) and tilling (performed in the autumn) in the region is around 0.05-0.07 m and 0.2-0.25 m, respectively, and thus we opted for N fertilizer placement on the border of these two zones in the soil profile.

In this experiment, we sought to test the effect of N fertilizer depths and depth combinations on (i) mineral N leaching and (ii) crop N uptake and yield, as well as (iii) soil emissions of carbon dioxide (CO₂), methane (CH₄) and N₂O. The latter two objectives could both corroborate field observations and provide further insight into the interplay between climate conditions, crop growth, and N losses. We hypothesized that deep-placed N fertilizer would have a beneficial affect for crop growth, and thus N uptake, resulting in less leaching of mineral N compared with a shallow N placement.

Materials and methods

Lysimeter collection and installation

Sixteen undisturbed soil columns with a diameter of 0.295 m were excavated to a depth of 1.18 m from an agricultural field in Säby (59°83'N, 17°71'E) in SE Uppsala, Sweden in May 2016 according to the method described by Persson and Bergström (1991). Briefly, the columns were extracted with a

tractor-mounted hydraulic soil auger, capped at both ends, and transported 8 km on a flatbed truck to the lysimeter station at the Uppsala campus of the Swedish University of Agricultural Sciences—SLU.

The field site where the lysimeters were excavated has been cultivated with agricultural crops, mainly cereals, for over a century. The mean annual air temperature is 5.5 °C and average precipitation is around 528 mm per year (Fig S1). In this area, as of much of Sweden, crops are primarily rain fed. The soil is characterized as a Eutric Cambisol, with a silt loam texture in the topsoil and 6.1 pH_{H2O} (Table 1). The subsoil (around 0.8 m and below) is influenced by the presence of gyttja, a gel-like material originating from partially decomposed organic matter accumulated under waterbody sediment. It has an elevated organic N and carbon (C) content, high porosity, and acidic pH.

Prior to installing the lysimeters, we removed approximately 0.08 m of soil from the bottom and filled in 0.05 m of each soil column with washed pea gravel (2–5 mm diameter). Stainless steel mesh was placed between the gravel layer and the 0.3 m-thick perforated PVC lid that capped the bottom of the columns. To simulate tillage, the topsoil (0–0.25 m) was removed from all lysimeters, pooled and manually homogenized, then replaced. There was a gap of approximately 0.05 m between the top of the PVC pipe and the upper soil surface, and 0.08 m from the underside was taken up by the gravel and the cap, so

Table 1 Soil physical properties along the soil profile sampled at lysimeter extraction from the field. Soil bulk density (BD) (kg dm⁻³), porosity (%), organic carbon (SOC) (g kg⁻¹), total nitrogen (g kg⁻¹), carbon to nitrogen ratio, calcium carbonate

that the effective soil volume was 0.718 m³ (1.05 m length and 0.068 m^2 surface area). We attached a broad, nearly-flat funnel and a steel-framed supportive seat to the underside of the columns before lowering them into the concrete-walled ports. The outlet of the attached funnel fit snugly into piping that connected to outlets in the lysimeter basement, where individual lysimeter leachates could be continuously collected in 5 L glass Erlenmeyer flasks. The lysimeters were installed in June 2016 and from that point on exposed to weather and allowed to drain freely by gravity. The experiment was initiated the following year (June 2017) to provide time for the lysimeters to both settle and equilibrate as well as to collect sufficient information on individual lysimeter draining behavior and background leachate N loads. During this period, the lysimeters were not planted and were periodically weeded. Thus in the remainder of this paper we refer to this time as a fallow period.

Experimental setup

Sixteen individual lysimeters were randomly assigned to three N fertilizer depth treatments and the control, consisting of four lysimeters each. In addition to a control treatment without N fertilization (Control), was a shallow N placement (Shallow) at 0.07 m, a mixed N placement (Mixed) where half the amount of N fertilizer was placed at 0.07 m and the other half at 0.2 m, and a deep N placement (Deep) at 0.2 m.

 $(CaCO_3)$ (g kg⁻¹), pH (H₂O), and texture represented by percentage clay, silt, and sand. Bulk density and porosity were not collected at 1.0–1.1 m depth due to groundwater infiltration. Adapted from Getahun et al. (2021)

Depth (m)	BD (kg dm ⁻³)	Porosity (%)	SOC (g kg ⁻¹)	Total N (g kg ⁻¹)	C:N	$CaCO_3 (g kg^{-1})$	pH (H ₂ O)	Clay (%)	Silt (%)	Sand (%)
0-0.1	1.3	50.9	28.2	2.4	11.9	0.10	6.1	21.9	54.5	23.6
0.1–0.2	1.4	48.3	26.4	2.2	11.8	0.20	6.1	20.5	56.9	22.6
0.2–0.3	1.4	46.8	14.2	1.2	11.6	0.09	6.3	21.3	56.2	22.5
0.3–0.4	1.6	41.5	7.9	0.7	11.6	0.07	6.5	18.9	54.1	27.0
0.4–0.5	1.5	43.0	3.8	0.4	9.6	0.07	6.7	23.5	59.7	16.8
0.5-0.6	1.4	46.4	3.4	0.4	8.6	0.10	6.8	25.3	62.6	12.1
0.6-0.7	1.4	48.7	3.7	0.5	7.9	0.09	6.9	31.1	61.1	7.8
0.7–0.8	1.4	48.7	3.2	0.4	7.6	0.07	6.7	27.4	56.7	15.9
0.8–0.9	1.3	49.4	6.3	0.9	7.3	0.11	6.1	39.6	57.7	2.7
0.9–1.0	1.2	54.0	6.4	0.9	7.3	0.12	5.2	34.2	63.1	2.7
1.0-1.1			9.8	1.3	7.4	0.16	4.8	40.6	57.4	2.0

The lysimeters were planted in 2017 with spring barley (Hodeum vulgare L. var. 'Makof'), in the second year with spring wheat (Triticum aestivum L. var. 'Quarna'), and in the third year with oats (Avena sativa L. var. 'Symfoni'). All lysimeters were fertilized at 0.07 m depth with potassium phosphate at a rate of 20 kg P and 40 kg K ha⁻¹ year⁻¹. We applied 100 kg N and 15 kg S ha⁻¹ year⁻¹ in the form of ammonium sulfate and ammonium nitrate dissolved in 10 mL water to each lysimeter receiving N fertilizer. To apply the N-S fertilizer solution, we removed 0.05 m of soil from the top of each lysimeter, to simulate harrowing and seed placement depth in the field, and injected the fertilizer in 1 mL increments with a syringe at ten different sites distributed evenly over the soil surface at either 0.07 m or 0.2 m, or both. The control treatment was injected with the equivalent amount of water in place of N solution. Following fertilization, a thin layer of soil was replaced, then two rows of seeds were placed on the surface before backfilling the remainder of the ~ 0.05 m soil. To promote seed germination, we then irrigated each lysimeter with 1 L H₂O (14.6 mm rain equivalents) over a span of two days in 250 mL increments. In S1 (2017), spring barley was sown on June 12th and harvested September 29th. Spring wheat was sown on May 10th and harvested August 16th in S2 (2018). In 2019 (S3), oats were sown on May 13th and harvested September 2nd. In this paper, we refer to the initial period from August 2016 to May 2017, beginning from the installation of the lysimeters until the first seeding and fertilization, as Fallow 1 (F1); the first experimental growing season as S1 (June 2017–April 2018); the second growing season as S2 (May 2018–April 2019); the third growing season as S3 (May 2019–April 2020); and the final fallow period as F2 (May 2020-August 2021), ending with the final leachate collection.

Measurements

Leachate

concentration was determined colorimetrically using the salicylate method and $NO_3^- + NO_2^-$ concentration via colorimetric vanadium chloride-reduction (ISO, 2013). Lysimeters did not drain at the same rate, so sampling would occur when there was sufficient leachate for collection at individual lysimeters. Leaching occurred primarily in fall and winter due to climatic conditions and plant uptake of available water during summer.

Chlorophyll content, plant height and harvest

Relative plant leaf chlorophyll content was measured twice during the first growing season (S1), three times during the second season (S2), and five times in the third (S3). We used a handheld SPAD-502 m (Minolta Camera Co., Osaka, Japan) to take three averaged readings per plant leaf while four randomly chosen leaves were measured per plant. On the same day as the SPAD measurements, we measured the plant height from two plants growing in each lysimeter.

At harvest, we removed all biomass down to the base of the plant with scissors. Harvested biomass was dried and then threshed to separate grain from straw. Subsamples of ground grain and straw were analyzed for N content using an organic elemental combustion instrument (LECO CNS Analyzer, Leco Corporation, St. Joseph, MI, USA).

Greenhouse gas measurements

Measurements of N₂O, CH₄, and CO₂ were taken during the growing seasons, beginning the day after sowing and fertilizing and ending around the time of harvest. During CH₄ and N₂O gas collection, a cylindrical PVC chamber (0.022 m³ volume, with riser 0.036 m^3) equipped with a small axial circulation fan and ventilation tube was fitted directly onto the lysimeter pipe. Chamber gas concentrations were collected five times per closure at 0, 10, 20, 30, and 40 min after chamber closure via the flow-through method where a loop is made with tygon tubing between a 20 ml glass vial, the chamber, and air pump. Collection vials were transported to the lab and stored for 2-14 days at room temperature before analysis for N₂O and CH₄ on a gas chromatograph (Clarus 500, Perkin Elmer, USA) equipped with an FID and ECD using an automatic headspace injector (Turbo Matrix

110, Perkin Elmer, USA). We sampled 19 times during S1, between June 13th and August 29th, with the highest frequency immediately following fertilization. In S2 we sampled 28 times, twice weekly, between May 11th and September 3rd.

Fluxes of CO₂ from each lysimeter were measured separately with a portable infrared gas analyzer (EGM-4, PP Systems, USA) on a majority of the same sampling occasions using an opaque chamber, and, once seed emergence occurred, a transparent chamber. Carbon dioxide was measured for approximately 125 s resulting in 27 respiration measurements for flux determination. The opaque chamber (SRC-2 Soil Respiration Chamber, PP Systems, USA) measured directly on the soil surface. The transparent chamber was 200 mm in diameter and 200 mm tall with an extension up to 600 mm to accommodate growing crops, and was similarly equipped with a ventilation tube and a battery-operated axial fan. Carbon dioxide was measured 17 times in S1 and 15 times during S2. Greenhouse gas measurements were not performed during S3.

Calculations and statistical analyses

Statistical analyses were performed with the R-software version 2022.07.02 "Spotted Wakerobin" (R Core Team 2022). We used R package *ggplot2* (Wickham 2016) and the plot_grid function from the *cowplot* package (Wilke 2020a, b) to produce data figures. To determine treatment effects on crop yield, N content, SPAD and plant height, as well as cumulative leachate amount and N load, we used the Anova function (*car* package, Fox and Weisberg 2019) to determine analysis of variance and the glht function (*multcomp* package, Hothorn et al. 2008) for posthoc analysis by using Tukey's all pair comparisons. Treatment differences were considered significant for P < 0.05.

To determine treatment effects over time in leachate amount (mm), N load (kg ha⁻¹), and volume-weighted concentration (mg L⁻¹), we used a repeated measures anova, using the lme function (*nlme* package, Pinheiro et al. 2021) to make a linear mixed model with time as a repeated factor. We used the corAR1 correlation structure to model the error term. Using the emmeans function (*emmeans* package, Lenth 2022), we tested treatment differences at each sampling time as well as within-treatment differences at different time points. The cropping seasons (S1-S3) were analyzed separately from the non-cropping periods (F1 and F2).

Lysimeter N load was calculated by multiplying the N concentration by leachate quantity at each sampling time. Mean volume-weighted concentration (mg L^{-1}) was determined by dividing the N load (mg) by leachate amount.

Greenhouse gas fluxes were calculated using the R package *gasfluxes* (Fuss 2020) using the fit "robust linear." Fluxes with P values greater than 0.05 were not considered.

The emission factor (EF) for indirect N_2O emissions from leached N (N_2O —L) in the fertilized treatments was calculated according to

$$EF_{N,O-L} = \left(N_{fert} \left[kgNha^{-1}\right] - N_{unfert} \left[kgNha^{-1}\right]\right) * EF5(0.011)$$

where the value for EF5 is the default value for leaching/runoff (IPCC 2019), N_{fert} = cumulative N load in leachate for fertilized treatment, and N_{unfert} = cumulative N load in leachate for the unfertilized treatment.

The N balance consisted of measured N inputs (N fertilizer and seed) and outputs (crop biomass N and leachate N) in the experimental system and their sums (kg N ha⁻¹ yr⁻¹) calculated for individual lysimeters. In addition, we calculated the nitrogen use efficiency (NUE) as an indicator for resource efficiency (Quemada et al. 2020):

$$NUE[\%] = \left(\frac{\sum \left(crop \, N \, outputs[kgNha^{-1}]\right)}{\sum \left(N \, fertilizer \, inputs[kgNha^{-1}]\right)}\right) * \, 100$$

To incorporate the control treatment that did not receive N input, we additionally calculated the agronomic efficiency of N (AE_N) according to Lahda et al. (2005) as well as the recovery efficiency of N (RE_N) (Lahda et al. 2005; Dobermann 2005):

$$AE_{N}[kgkg^{-1}] = \frac{\left(grain \, yield_{fert} - grain \, yield_{unfert}\right)}{N_{applied}}$$

$$RE_{N}[\%] = \frac{\left(plant \, N \, uptake_{fert} - plant \, N \, uptake_{unfert}\right)}{\left(N_{applied}\right)} \, * \, 100$$

Results

Leachate quantity and climatic conditions

Across treatments, collected leachate amounts averaged 27, 10 and 14% of precipitation plus irrigation in S1, S2 and S3, respectively (Table 2). Over the three periods (S1 - S3) % total leachate quantity relative to total water inputs was lowest in Deep and highest in Control following the pattern of Control > Mixed > Shallow > Deep, corresponding to 20, 18, 17 and 13%.

The Control had significantly higher (p=0.04) mean cumulative leachate amount (\pm SE) for the three growing seasons (S1-S3), 377 \pm 37 mm H₂O, compared to the lowest in deep, 249 \pm 12 (Fig. 1). Mixed and Shallow were intermediates with mean cumulative leachate of 332 \pm 36 and 319 \pm 25, respectively. The Deep placement had the lowest quantity of leachate for all periods except for the initial F1 period. Within fertilized treatments, Deep leached 25% less water compared to Shallow and 29% less than Mixed.

During the initial fallow period (August 2016 to May 2017) there were no statistical differences in water flow, although there was some variation between individual lysimeters and overall among the different treatments (Fig. 2, Table 2). Mean leachate amount was somewhat higher in Shallow $(133 \pm 12 \text{ mm})$ and lowest in the Control $(83 \pm 17 \text{ mm})$. The Mixed and Deep treatments had

intermediate water flow in F1 with 110 ± 15 and 109 ± 7 mm. Total precipitation and irrigation during the F1 period was approximately 322 mm, the lowest quantity of all periods (Fig S1, Table 2).

In the winter and spring following the first cropping season (S1), the pattern of water flow changed from the preceding F1 period. The S1 period (June 2017-April 2018) was generally wetter than the preceding period, particularly in the autumn and winter months (Fig S1). Though there were no significant differences in mean cumulative leachate, the treatments in S1 were by amount Control>Shallow>Mixed>Deep corresponding to 179 ± 10 , 168 ± 6 , 160 ± 17 , and 148 ± 5 mm, respectively (Figs. 1 and 2, Table 2).

In the second cropping year (S2), nearly every month had both lower precipitation and higher average temperatures compared to the long-term normal (Fig S1), particularly during the cropping period from May to July 2018. Although it appears that July received sufficient precipitation, the majority occurred late in the month on a single day, when 79 mm out of the monthly total of 82 mm rain fell. Lysimeter leachate quantity was greatly affected by the drought and only two of the four treatments, Control and Mixed placement, were releasing water by February 2019, and in large quantities. Sufficient quantities of water for sampling did not flow from all lysimeters until around April 2019. The mean cumulative amount for S2 (\pm SE) was significantly higher

Table 2 Total precipitation and irrigation (mm), cumulative lysimeter leachate (mm) and mineral nitrogen (N) load in leachate (kg ha^{-1}) per period. Lowercase letters represent treatment differences (p < 0.05)

Period	Precip + Irrigation (mm)		Control	Shallow	Mixed	Deep
F1	322	Leachate (mm)	83±17	133 ± 12	110±15	109±7
		N load (kg ha ⁻¹)	25 ± 11	69 ± 11	49 ± 4	53 ± 12
S1	599	Leachate	179 ± 10	168 ± 6	160 ± 17	148 ± 5
		N load	41 ± 11^{b}	124 ± 12^{a}	108 ± 13^{a}	96 ± 21^{ab}
S2	637	Leachate	101 ± 27^{a}	36 ± 6^{ab}	94 ± 14^{a}	27 ± 4^{b}
		N load	15 ± 5^{ab}	11 ± 2^{ab}	24 ± 4^{a}	9 ± 2^{b}
S 3	642	Leachate	97 ± 14	114 ± 33	79 ± 14	74 ± 4
		N load	14 ± 3	46 ± 22	15 ± 3	20 ± 3
F2	746	Leachate	165 ± 10	211 ± 48	195 ± 38	150 ± 5
		N load	10 ± 1	62 ± 52	15 ± 9	12 ± 1

F1 = Initial fallow, S1 = 1st year with crops, S2 = 2nd year with crops, S3 = 3rd year with crops, F2 = latter fallow. Control = no N fertilizer, Shallow = shallow N fertilizer placement (0.07 m), Mixed = mixed placement of N fertilizer (half at 0.07 m, half at 0.2 m), Deep = deep placement of N fertilizer (0.2 m)





in the Control $(101 \pm 27 \text{ mm})$ and Mixed placement $(94 \pm 14 \text{ mm})$ compared with Shallow $(36 \pm 6 \text{ mm})$ and Deep $(27 \pm 4 \text{ mm})$ (Fig. 2, Table 2). Within the fertilized treatments, Mixed was also significantly higher than Shallow and Deep (p=0.001). In terms of within-treatment comparisons, the Control treatment water flow in this period was significantly higher than at any other sampling time during S1-S3 with the exception of one sampling occasion immediately following, in December 2019 (Fig. 2). Similarly, leachate quantity in the Mixed placement was significantly higher than all other sampling times in S1-S3.

Rainfall was relatively closer to long-term normal during S3 in comparison with the previous year, although some compensation with irrigation was necessary during May—July 2019 (Fig S1). Mean cumulative leachate (\pm SE) in S3 followed the order of Shallow > Control > Mixed > Deep corresponding to 114 \pm 33, 97 \pm 14, 79 \pm 14, and 74 \pm 4 mm although there were no significant differences between treatments (Table 2).

Though not significant, the deep placement continued to leach less water than all other treatments into the F2 period despite the absence of crops, although most treatment effects tapered off after December 2020 (Fig. 2). Mean cumulative water flow followed the order of Shallow > Mixed > Control > Deep and corresponded to 210 ± 47 , 195 ± 37 , 165 ± 10 , and 150 ± 5 mm (\pm SE).



Fig. 2 Mean leachate quantity (mm H₂O), leachate N concentration (mg L⁻¹ H₂O), and weighted N load (mg N ml⁻¹). Uppercase letters indicate treatment differences within the same sampling time (p < 0.05). Each bar represents a single sampling, except Apr–May 2018 and Feb–Apr 2019 which are comprised of the sum of multiple samplings in order to incor-

porate leaching from all lysimeters. F1=Initial fallow, S1=1st year with crops, S2=2nd year with crops, S3=3rd year with crops, F2=latter fallow. Control=no N fertilizer, Shallow=shallow N fertilizer placement (0.07 m), Mixed=mixed placement of N fertilizer (half at 0.07 m, half at 0.2 m), Deep=deep placement of N fertilizer (0.2 m)

N Load and N volume-weighted concentrations

Cumulative N load for all cropping seasons (S1-S3) was lowest in the Control, 70 ± 18 kg N ha⁻¹, and highest in Shallow, 181 ± 21 kg N ha⁻¹ with significant differences (p=0.009) between the two treatments. The Mixed and Deep placements were intermediates with mean cumulative N loads of 147 ± 14 and 124 ± 24 kg ha⁻¹ respectively, resulting in a reduction of leachate N losses of 21 and 37% compared to Shallow. Among the fertilized treatments, however, there were no significant differences.

N losses in leachate during F1 were relatively low, but not significantly, in the Control $(25 \pm 11 \text{ kg} \text{ N ha}^{-1})$ compared to the other treatments $(69 \pm 11, 49 \pm 4, 53 \pm 12 \text{ kg N ha}^{-1}$ for Shallow, Mixed, and Deep respectively), even though no fertilizer had been applied to any of the treatments. Nitrogen load steadily increased in all lysimeters after the initial disturbance at the beginning of the setup (Fig. 2) and by the end of F1, treatment differences were nearly significant (p=0.055). This flush of mineralized N peaked in S1 and began to decline in S2. In S1, total mean leachate N increased to 41 ± 11 , 124 ± 12 , 108 ± 21 , and 96 ± 13 kg N ha⁻¹ yr⁻¹ for Control, Shallow, Mixed, and Deep respectively, and there were significant treatment differences where Shallow, Mixed > Deep > Control, with Shallow and Mixed significantly higher than the Control (Fig. 1, Table 2). The emission factors for indirect N₂O emissions for this period due to leached N were 1.40, 1.30, and 1.18 for Shallow, Mixed, and Deep, respectively.

Subsequent to S1, average N loads decreased below F1 levels. During the growing season of S2 there was a drought during the critical part of the growing period from May to July in 2018, with low precipitation and high temperatures compared to the long-term normal (Fig S1). Consequently, there was just one brief period of water flow the following spring (Fig. 2) from late February to April 2019, in which the Mixed placement had the highest leachate N load, significantly higher than Deep (p=0.04), following the pattern Mixed > Control, Shallow > Deep. S2 mean leachate N loads (\pm SE) were 15 \pm 5, 11 \pm 2, 24 \pm 4, and 9 \pm 2 kg N ha⁻¹ yr⁻¹ for Control, Shallow, Mixed, and Deep respectively (Table 2). Indirect N₂O emission factors for leached N were 0.23, 0.86, and 0.13 for Shallow, Mixed, and Deep, respectively.

In the third growing season (S3), precipitation was higher, particularly in October-December, relative to the previous drought year, resulting in a different pattern in leachate N loads, where Shallow and Deep had insignificantly elevated N loads relative to Control and Mixed. Total mean N loads $(\pm SE)$ were 14 ± 3 , 46 ± 22 , 15 ± 3 , and 20 ± 3 kg N ha⁻¹ yr⁻¹ for Control, Shallow, Mixed, and Deep respectively (Table 2). An individual lysimeter in the Shallow treatment, which also had a low yield in S3, leached N at levels 5 times greater than all other lysimeters, and continued to leach high levels of N well into F2 the following year (Fig. 1 and 2). The EF for indirect N_2O emissions due to leached N was 1.11, 0.72, and 0.66 for Shallow, Mixed, and Deep, respectively. In the last period when no fertilization or cropping had occurred, the N load in the majority of lysimeters declined. Cumulative treatment N load means for F2 (\pm SE) for the Control, Shallow, Mixed, and Deep were 10 ± 1 , 62 ± 52 , 15 ± 9 , and 12 ± 1 kg N ha⁻¹ respectively.

Greenhouse gas fluxes

About 9% of N₂O fluxes had a p-value < 0.05, which means that the individual measurements showed a significant increase or decrease. Of those, about half surpassed the variability of the detection limit of the GC (max ppm – min ppm < GC detection limit). Likewise, 25% of CH₄ fluxes had a p-value < 0.05, and of those only one was below the GC detection limit.

Photosynthetic CO_2 uptake in the Control was greatest in the earlier stages of plant growth (Fig. 3) in S1 with the Shallow treatment following a similar trend or with somewhat less uptake than the Control, but in the drought year S2, this trend was less clear. The Mixed and Deep treatments tended to have greater CO_2 uptake later in the growing period relative to Control and Shallow. In 2017 (S1), the Mixed had the greatest CO_2 uptake but the following year the control was highest, while in both years Shallow was the lowest. Total uptake (sum of negative fluxes) in S1 was 1.77, 1.57, 1.84, and 1.77 g CO_2 -C m⁻² h⁻¹ for Control, Shallow, Mixed, and Deep respectively. In S2 the pattern of total uptake changed, and was 2.63, 2.03, 2.08, and 2.34 g CO_2 -C m⁻² h⁻¹ for Control, Shallow, Mixed, and Deep respectively.

Biomass yield and N balance

Nitrogen use efficiency (NUE) was high in all fertilized treatments in all seasons due to the high crop N output relative to the fertilization rate (Table 3, Fig. 4). The N surplus, calculated as the difference between N inputs to and outputs from the system, was highest in all treatments in S1 and lowest in S2, the latter of which occurred during a drought and resulted in the lowest N outputs from almost all components for all treatments.

In S1, N losses via leachate were very high (Fig. 2, Table 2), even in the control, and leachate accounted for the second-highest output from the system after harvested grain N (Table 3). Although yields were similar among treatments in S1 (Table 4), due to the difference in grain and straw N uptake, the Mixed and Deep treatments had higher outputs in grain and straw, the latter significantly higher, compared with the control (p=0.02). Additionally, mixed placement had significantly higher grain N content compared to the control (p=0.045) (Table 4).

Both AE_N , a measure of grain N uptake efficiency that accounts for N uptake in the non-fertilized control, and RE_N , where the additional N in straw is incorporated into the calculation, followed a similar trend over time, with the greatest treatment differences occurring in the final S3 season. This trend is similar to the trend in grain yield between the Mixed and Deep treatments (Table 4). During S1 and S2, both Mixed and Deep treatments had similar yields and crop N uptake, and thus similar system outputs in the form of crop N, and were greater than Shallow placement in both years. However, in S3, when there was neither drought nor an excess of mineralized N as in the previous years, Mixed placement had intermediate yields and the lowest crop N uptake among the fertilized treatments, although leachate losses were as low as the control (Table 3).



Fig. 3 Daytime CO₂ fluxes (mg m⁻² h⁻¹ CO₂-C) during the cropping periods of S1 and S2. Uppercase letters indicate represent treatment differences (p < 0.05).S1=1st year with crops, S2=2nd year with crops. Control=no N fertilizer, Shal-

In the cumulative N balance (sum of S1-S3) (Fig. 4), the Deep placement had the greatest N surplus but the highest amount and proportion (83%) of N output from harvested crops relative to total N outputs among the fertilized treatments. An opposite trend was observed in the Shallow placement, where outputs from crop N were lowest but those from leaching were highest, while Mixed placement was an intermediate to Deep and Shallow. The Control had the second highest N surplus, primarily due to no fertilizer input, and each N output component was the lowest among all treatments. However, the proportion of N losses in the Control were similar to the Deep placement, but approximately 1.7 times lower in each component.

low = shallow N fertilizer placement (0.07 m), Mixed = mixed placement of N fertilizer (half at 0.07 m, half at 0.2 m), Deep = deep placement of N fertilizer (0.2 m)

Discussion

Reduced leaching through deep N placement

In general, Deep placement had the lowest quantity of leachate for all periods except for the initial fallow period (Fig. 1, Table 2), suggesting a greater water use efficiency, which in turn promoted crop N uptake, higher yields, and lower N losses via leachate (Fig. 4). Total leachate quantity in S1 – S3 was significantly lower in the Deep treatment than in the other treatments (p=0.03), compared to highest in Control. Compared to Shallow, Deep placement had 25% less leachate and 29% less than Mixed. Though not significant, Deep continued to have less leachate

	S1				S2				S3			
	Control	Shallow	Mixed	Deep	Control	Shallow	Mixed	Deep	Control	Shallow	Mixed	Deep
N inputs and out	uts (kg N ha	$^{-1}$ yr ⁻¹)										
Seeds	8	8	8	8	8	8	8	8	8	8	8	8
Fertilization		100	100	100		100	100	100		100	100	100
Harvested grain	-118 ± 17	-141 ± 14	-157 ± 9	-155 ± 13	$-80\pm4^{\rm b}$	-127 ± 9^{a}	-158 ± 12^{a}	-157 ± 5^{a}	$-95\pm6^{\mathrm{b}}$	-151 ± 25^{ab}	$-145\pm16^{\rm ab}$	-190 ± 5^{a}
Harvested straw	-27 ± 2^{b}	-38 ± 6^{ab}	-43 ± 2^{a}	-43 ± 1^{a}	-13 ± 1^{b}	-30 ± 3^{a}	-26 ± 4^{a}	-31 ± 1^{a}	$-18\pm2^{\rm b}$	-32 ± 1^{a}	-29 ± 2^{a}	-30 ± 2^{a}
Leachate load	-41 ± 11^{b}	-124 ± 12^{a}	-108 ± 13^{a}	-96 ± 21^{ab}	-15 ± 5^{ab}	-11 ± 2^{ab}	-24 ± 4^{a}	-9 ± 2^{b}	-14 ± 3	-46 ± 22	-15 ± 3	-20 ± 3
N surplus	-179	-195	- 200	- 185	- 100	- 60	- 101	- 89	-119	- 121	- 80	-132
NUE %	n.a	179	200	198	n.a	157	184	188	n.a	183	174	220
$AE_N (kg kg^{-1})$	n.a	0.23	0.39	0.36	n.a	0.47	0.78	0.77	n.a	0.56	0.5	0.95
RE _N (%)	n.a	34	55	53	n.a	64	92	95	n.a	70	60	107

as efficiency (NTIE) (%) - arronomic efficiency of N (AE.) (ka ka^{-1}) and the recovery efficiency of N (RE.)	1^{-1} yr ⁻¹) \pm standard error. Lowercase letters represent significant treatment differences within the given period		
Table 3 Nitrovan (N) halance commonants ner cronning season Nuse efficiency (NHF)	(%). N balance values are presented as treatment mean value (kg N ha ⁻¹) \pm standard	(p < 0.05). Note that actual season lengths varied	



Fig. 4 Cumulative (S1-S3) N balance containing total inputs (N from seeds and fertilizer) and total outputs (N leaching, grain and straw yield N) with balance sum in kg N ha⁻¹ 3 yr⁻¹. Note that actual season lengths varied. S1 = 1st year with crops (June 2017 – April 2018), S2=2nd year with crops (May 2018–April 2019), S3=3rd year with crops (May 2019 – April 2020). Control = no N fertilizer, Shallow = shallow N fertilizer placement (0.07 m), Mixed = mixed placement of N fertilizer (half at 0.07 m, half at 0.2 m), Deep = deep placement of N fertilizer (0.2 m)

than all other treatments during the F2 period despite the absence of crops. Within the fertilized treatments, however, there were no significant differences in cumulative leachate except during the drought period S2. Similar to our results, Chen et al. (2022) found that Deep placement of N fertilizer at 0.15 m relative to placement at 0.05, 0.25 and 0.35 m had the highest precipitation use efficiency, crop N uptake, radiation use efficiency, and also reduced soil nitrate-nitrogen residue levels in the deep layers under two years of maize followed by winter wheat. They also found that root surface area and root length density were highest at 0.15 m placement.

In our experiment, observed treatment differences in leachate amount, and thus crop uptake of soil water, were likely a consequence of differences in either root architecture (e.g., deep rooting), root biomass, or a combination of the two. Although genetics play a fundamental role in plant rooting patterns, many studies have shown that roots exhibit plasticity in response to the soil environment, particularly when nutrients are distributed heterogeneously or in patches (Hodge 2004). Although the roots were not sampled, we can infer belowground biomass from aboveground plant biomass and leaching quantity. Aboveground crop biomass at harvest was negatively correlated with leachate quantity (p < 0.0001, $R^2 = 0.81$) when excluding the drought year S2 (Fig. 5).

In S1, when available soil N was likely quite high, there was only a minor effect of the N fertilizer placement on aboveground biomass and leachate quantity across treatments, and thus the correlation of biomass to leachate amount was not significant for the year individually (p=0.18, $R^2=0.39$). Whereas in S3, the fertilizer treatment significantly affected crop biomass, and the correlation between aboveground biomass and leachate quantity was more clear (p < 0.0001, $R^2=0.65$). In S2, summer drought

Table 4 Grain yield (tons ha⁻¹), straw and grain N content (% N). Lowercase letters represent treatment differences within the given period (p < 0.05)

Period	Parameter	Control	Shallow	Mixed	Deep
S1	Grain yield	6.4 ± 0.3	6.2 ± 0.4	6.7 ± 0.5	6.6 ± 0.5
	Grain % N	1.83 ± 0.20^{b}	2.26 ± 0.14^{ab}	2.38 ± 0.08^{a}	2.29 ± 0.07^{ab}
	Straw % N	0.53 ± 0.02	0.69 ± 0.08	0.72 ± 0.02	0.71 ± 0.03
S2	Grain yield*	5.8 ± 0.2^{b}	6.1 ± 0.7^{ab}	7.7 ± 0.6^{a}	7.5 ± 0.2^{ab}
	Grain % N	1.4 ± 0.03^{b}	2.1 ± 0.09^{a}	2.0 ± 0.02^{a}	2.1 ± 0.03^{a}
	Straw % N	0.30 ± 0.02^{b}	0.43 ± 0.02^{a}	0.45 ± 0.02^{a}	0.50 ± 0.02^{a}
S3	Grain yield	6.7 ± 0.3^{b}	7.9 ± 1.5^{ab}	8.3 ± 1.1^{ab}	10.1 ± 0.2^{a}
	Grain % N	1.41 ± 0.02^{b}	1.97 ± 0.09^{a}	1.77 ± 0.07^{a}	1.89 ± 0.02^{a}
	Straw % N	0.22 ± 0.02^{b}	0.34 ± 0.03^{a}	0.29 ± 0.02^{ab}	0.27 ± 0.01^{ab}

*excluding outlier lysimeter 13 (Shallow treatment)

S1 = 1st year with crops, S2 = 2nd year with crops, S3 = 3rd year with crops. Control = no N fertilizer, shallow = shallow N fertilizer placement (0.07 m), mixed = mixed placement of N fertilizer (half at 0.07 m, half at 0.2 m), deep = deep placement of N fertilizer (0.2 m)



Fig. 5 Total aboveground crop biomass (Dry matter in Mg ha^{-1}) and total leachate quantity of H_2O (mm) per cropping season (S1–S3) for individual lysimeters. Linear trend line and R² value for combined years S1 and S3. S1=1st year with crops, S2=2nd year with crops, S3=3rd year with crops. Control=no N fertilizer, Shallow=shallow N fertilizer placement (0.07 m), Mixed=mixed placement of N fertilizer (half at 0.07 m, half at 0.2 m), Deep=deep placement of N fertilizer (0.2 m)

conditions likely affected the allometric relationship between roots and shoots as there were significant treatment differences in both aboveground biomass at harvest and total leachate, but they were not correlated (p=0.7, $R^2=0.21$) (Fig. 5). Mathew et al. (2018) also found a weaker relationship between root to shoot ratio and shoot biomass or grain yield in wheat under drought-stressed conditions compared to non-stressed conditions. Similar to that, Meurer et al. (2019) found that shoot:root ratios, as well as N concentrations in living roots changed depending on irrigation and N fertilization in a field experiment with mixed grass ley in central Sweden.

Deep rather than lateral exploration by roots, earlier in the season, has been shown to be beneficial for N capture and subsoil water access, although water in the subsoil is potentially more beneficial in the latter part of crop growth (Lynch 2013). We expected that in the earliest stages of crop growth, Shallow placement crop roots would proliferate around 0.07 m where the P and 100% of the N fertilizers were placed, but Deep placement by contrast, would have earlier deeper root exploration and have a relatively higher root biomass at and below 0.2 m. In the Mixed placement, we would expect that initial root proliferation would have occurred around the 0.07 m placement initially, but once N resources were exhausted, roots would explore the soil profile toward the remaining 0.2 m-placed N, but this deeper exploration would be delayed compared to the Deep treatment, and possibly mechanically impaired if subsoil moisture was low (Colombi et al. 2018). While this delay in the mixed placement was not generally detrimental in terms of crop N uptake, during drought conditions in S2 there were high N losses via leaching, likely due to low root biomass relative to the other fertilized treatments. Additionally, in S3, with non-drought conditions and in the absence of excess soil mineral N, the mixed placement had lower yield and lower crop N uptake compared to Deep. The shallow N placement was beneficial in drought conditions (albeit with supplemental irrigation) in terms of soil water usage, since both leachate flow and N load were low, but in non-drought conditions leachate flow and N loads were high. It is possible, however, that the higher N load in Shallow and Deep placements in S3 was a result of a carryover effect of previously immobilized soil N from S2 when both treatments had very little leachate flow.

Drought obscured treatment effect on GHG emissions

Unlike our findings from the field experiment (Rychel et al. 2020), N₂O and even CH₄ fluxes to a lesser extent, were too few to allow for treatment comparisons. During the same cropping season as S1, the field N₂O fluxes averaged $(\pm SD)$ 69.9 ± 49.1 and $56.9 \pm 52.9 \ \mu g \ N_2O-N \ m^{-2} \ h^{-1}$ for a shallow and mixed placement respectively, and 44.9 ± 39.2 and $43.8 \pm 37.9 \ \mu g \ N2O-N \ m^{-2} \ h^{-1}$ respectively for the deep and control treatments, respectively. The lack of surface N₂O fluxes in the present study is possibly due to lysimeter detachment from groundwater, which alters the soil water relative to field conditions due to the lack of capillary rise from the groundwater (Abdou and Flury 2004; Bergström 1990). Since the lysimeters are draining freely by gravity year-round, we could assume that, on average, the lysimeter soil was drier compared to natural conditions in the field, particularly in the subsoil (> 0.3 m) during periods of high evapotranspiration. Supplemental irrigation was applied in small quantities in the summertime in order to avoid creating preferential flow, usually either 3.7 or 7.3 mm per lysimeter per day, split into two watering times, and meanwhile rainfall events in May–July were normally even less. Thus, the minimum infiltration depth of supplemental irrigation (mm H₂O / soil porosity) would have been around 14.4 mm. Due to the combined effect of drier soil conditions in the subsoil along with a shallow infiltration of supplemental irrigation in the overlaying zone, we expect that any upward diffusion of N₂O produced in the (likely aerobic) zone below the infiltration depth would have been limited by the upper wetted zone where it could have undergone complete denitrification. Due to the soil moisture status during summer months, N₂O emissions in our experiment could thus be compared to studies where crops are irrigated. For example, Yang et al. (2019) and Wang et al. (2016) found that overhead sprinkler irrigation or surface drip irrigation, respectively, relative to flood irrigation, significantly reduced N₂O emissions, which primarily wetted the soil surface and did not fill soil macropores in lower soil depths.

Carbon dioxide fluxes, on the other hand, were within an expected range, although the two growing seasons differed (Fig. 3). In S1, rates and length of crop maturation as indicated by CO₂ uptake patterns were more clear between treatments and followed the pattern of Control > Shallow > Mixed, Deep. However, in S2, possibly due to climatic conditions, the rate of maturation was largely similar among treatments. Grain N content at harvest in S1 reflected the pattern of CO₂ uptake, where a longer growth period and later maturation corresponded to higher grain N accumulation, similar to the findings of Hay and Kirby (1991) and Andersson (2005). Cheng et al. (2020) and Wu et al. (2022) also found that deep N placement delayed senescence of maize so that more aboveground biomass was sustained later in the cropping season, leading to deeper and more extensive rooting, which in turn promoted both crop N uptake and higher grain yield. Plant height differences, when measured around the same time as CO_2 fluxes (Table S1), corresponded to differences in CO₂ uptake, but not relative leaf chlorophyll content readings, which instead indicated crop N uptake differences.

Conclusions

In this study, deep N fertilization was beneficial for crop N uptake and yield, but also, in contrasting climatic conditions and soil N availability, this method promoted greater crop-water use efficiency, which led to reduced mineral N losses via leaching. The effect of reduced leaching continued even into the fallow period following the three years of cropping. This study highlighted the importance of monitoring leaching behavior over a longer time period, rather than within an individual cropping season, which may be difficult to interpret.

While our results showed agronomic and environmental benefits, we recognize that the required equipment and management for implementing deep N fertilization available in Sweden may be cost-prohibitive and possibly inaccessible for some farmers. Additionally, further studies on a variety of soil textures, as well as drought studies without irrigation management, would elucidate the effectiveness of deep fertilization more generally.

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Author contributions Katrin Rychel ran the experiment, collected and analyzed the data, and wrote the manuscript. Katharina Meurer contributed to supervising the experiment, assisted in data analysis and manuscript revision.Gizachew Getahun contributed to collecting and analyzing the data.Lars Bergström assisted in data analysis and supervision and planning of the experiment.Holger Kirchmann led the project and contributed to planning the experiment.Thomas Kätterer was the primary supervisor of the experiment and assisted with manuscript revision.All authors reviewed the manuscript.

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Declarations

Competing interests The authors declare no competing interests.

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