Notat

SEGES Innovation
Planter & Miljø

Foreløbig manuskript til publicering i et videnskabeligt tidsskrift	Ansvarlig	nanb
	Oprettet	29-11-2023
Projekt: 8506 – Klimaeffektive gødningsstrategier	Side	1 af 1

STØTTET AF
Promilleafgiftsfonden for landbrug

Foreløbig manuskript til publicering i et videnskabeligt tidsskrift

Data der er indsamlet i 2022-2023 skal, sammen med data der indsamles i 2024, indgå i en videnskabelig artikel, og i den forbindelse arbejdes der løbende på et manuskript. Manuskriptet bygges op ved først at sætte scenen: hvad ved vi allerede, og hvor er vores videnshuller? Dernæst er der et materiale og metode afsnit, hvor det forklares, hvad der er gjort og under hvilke omstændigheder, samt hvilket udstyr der er blevet brugt. Så kommer der et resultatafsnit, hvor de udvalgte resultater bliver opsat i tabeller og figurer med en beskrivelse af, hvad der ses. Til slut bliver resultaterne diskuteret og sat i relation til andre resultater, ligeledes fra videnskabelige artikler, og på baggrund af den nye, samlede viden laves der konklusioner. Undervejs i manuskriptet er der sat forslag ind til, hvilke referencer der skal medtages i den endelige artikel.

/Nanna Baggesen

Introduction

1.1 Setting the scene for the need to mitigate N_2O emissions

- N_2O production from the agricultural sector (impact from mineral fertilizers).
- Why is the production of N_2O a global problem.
- A brief description of how N_2O is produced in agricultural soils (e.g., nitrification, denitrification, nitrifier dentification).

(Mosier *et al.*, 1998, Ravishankara *et al.*, 2009, Syakila & Kroeze, 2011, Butterbach-Bahl *et al.*, 2013, Myhre *et al.*, 2014, Wrage-Mönnig *et al.*, 2018, Tian *et al.*, 2020; Grados et al., 2022).

1.2 Nitrification inhibitors as a mitigation strategy

- A focus on their potential in the agricultural sector.
- How these inhibitors reduce N_2O emissions (i.e., delaying nitrification).

(Subbarao *et al.*, 2006, Akiyama *et al.*, 2010, Ruser & Schulz, 2015, Bryne et al., 2020; Friedl *et al.*, 2020, Shen *et al.*, 2020, Grados et al., 2022; Soares et al., 2023).

1.3 Previous information on 3,4-dimethlypyrazole phosphate (DMPP)

- Focus on both meta-analyses.
    - "Based on global meta-analyses, across a range of NIs, N_2O emissions were on average reduced by 44% (Qiao *et al.*, 2015), 35-38% (Akiyama *et al.*, 2010, Ruser & Schulz, 2015), and 38% (Thapa *et al.*, 2016) as compared to either mineral or organic fertilization without inhibitors."
- Specific studies either conducted in Denmark or within a similar climate.
    - Peixoto and Petersen et al., 2023

(Weiske *et al.*, 2001, Zerulla *et al.,* 2001; Akiyama *et al.*, 2010, Qiao *et al.*, 2015, Ruser & Schulz, 2015, Feng *et al.*, 2016, Thapa *et al.*, 2016; Azeem *et al.,* 2022; Tufail *et al.,* 2023).

1.4 Objectives and hypotheses

- Need to bridge existing knowledge with rationales for conducting these studies.

Materials and Methods

2.1. Experimental sites

Proposed table for the three different locations and years

Table 1
Locations, weather conditions and soil properties of the four experimental field sites.

Experimental sites		Foulum	Vejen	Askov	Taastrup
Geographical coordinates		56°29 N, 9°34E	55°26 N, 9°08E	55°28 N, 9°06E	55°40 N, 12°18E
Annual rainfall[#]	mm	803	933	933	646
Annual temperature[#]	°C	8.6	9.0	9.0	9.3
Rainfall during spring[§]					
2011–2019	mm	177	165	165	139
2020	mm	102	122	122	116
2021	mm	196	285	268	95
Temperature during spring					
2011–2019	°C	10.9	11.3	11.3	11.7
2020 (min/max)	°C	+ 5.5/+ 18.9	+ 0.9/+ 27.3	+ 0.9/+ 27.3	-0.7/+ 24.1
2021 (min/max)	°C	-2.6/+ 28	-4.7/+ 29.4	-4.7/+ 29.4	-2.2/+ 29.3
Soil type		Sandy loam	Sand	Sandy loam	Sandy loam
Texture					
Clay	$g\ (100\ g)^{-1}$	7.0 (0.03)	3.0 (0.00)	10.5 (0.28)	15.9 (0.47)
Silt	$g\ (100\ g)^{-1}$	27.7 (0.88)	3.6 (0.49)	22.4 (0.37)	18.4 (0.30)
Fine sand	$g\ (100\ g)^{-1}$	32.2 (0.65)	9.1 (0.27)	27.2 (0.23)	32.0 (0.72)
Coarse sand	$g\ (100\ g)^{-1}$	30.2 (0.76)	82.4 (0.58)	35.7 (0.18)	29.4 (0.55
Organic matter	$g\ (100\ g)^{-1}$	3.8 (0.09)	2.3 (0.17)	5.1 (0.24)	4.4 (0.06)
Total C	$g\ (100\ g)^{-1}$	1.9 (0.05)	1.2 (0.09)	2.5 (0.12)	1.36 (0.03)
Bulk density	$Mg\ m^{-3}$	1.25 (0.03)	1.53 (0.01)	1.37 (0.02)	1.7 (0.03)
pH (1 M KCl)		5.30 (0.14)	5.68 (0.27)	5.46 (0.11)	4.3 (0.25)

[#] Average for the period 2011–2019.
[§] Day of fertilisation of individual campaigns in Table S2; final sampling *c.* 1 July.

The experimental sites represented different typical sand soil types in Denmark ranging from sandy loam to clay soil and were spread across Denmark. Holeby and Ringsted represented a drier site, whereas Aarhus represented a wetter site.

The experiment was carried out in two different experiments, each with three sub trials resulting in six experiments per year during a three year-period (2022-2024). The experiments were carried out in established agricultural fields in winter wheat and spring barley, respectively, which were treated following common agricultural practice.

At each field site a weather station was established to record and log local air temperature in XX cm height and precipitation every ten minutes. Furthermore, a soil sensor (Tomst TMS-4, TOMST s.r.o., Pragh, Czech Republique) was placed in a representative field plot at each field site to record and log soil temperature and -moisture in 6 cm depth every minute to represent the total experimental area. During each N_2O measurement round, a handheld soil moisture meter (HH2 Moisture Meter) connected to a soil sensor (SM150T, Delta T-Devices Ltd, Cambridge, UK) was used to record N_2O sub plot specific soil moisture. In each sub plot, outside the metal frames, soil moisture was measured in the upper 5 cm of the soil three times to get a representative soil moisture content.

2.2. Experimental setup

The experimental setup was identical for the two experiments and was a part of the national field trials in Danish agriculture and followed the accepted guidelines herein (link to guidelines). Each experiment consisted of field plots which were split in two: harvest subplots (>15 m2) and N_2O measurement subplots (>4 m^2), and for each treatment there was four replicates. The N_2O

measurement subplots were also used for taking soil samples. The field plots were placed in lines (1-4 lines) in a representative area of the field and were spared from mechanical disturbances.

2.3. Fertilizer and NI Treatments

Identical for each experiment was the "zero N" plots, which did not receive added N, to get a measurement of the background N_2O. The rest of the treatments followed standard N addition (200 kg N ha^{-1} in winter wheat and 120 kg N ha^{-1} in spring barley), but differed in type of N, number of fertilizer allocations, and the use of the nitrification inhibitor (trade mark name) with the active compound DMPP (3,4-dimethyl-1H-pyrazole phosphate) (Table– overview of treatments). Fertilizer in spring barley was surface-applied when sowing in start-April and NI was coated directly on the fertilizer. All plots in winter wheat were surface fertilized in mid-Marts, mid-April, and mid-May except for the liquid foliar fertilizer treatment, which were not fertilized in mid-May but in the beginning of May. The treatment with four fertilizer applications was fertilized during all four occasions. As was the case for spring barley, the NI was coated directly on the fertilizer at each application.

2.4. N_2O flux measurements

The sampling strategy was to do field measurements once every week during the growing season with more frequent measurement for 2 weeks directly after field actions (2 measurements per week). The higher frequency was to reveal short emission bursts after tillage and fertilization. N2O measurements were carried out using the static chamber method. The chamber units were made of white, non-transparent PVC in the dimensions 50 cm x 50 cm x 20?? Cm (L x W x H) and the possibility of using up to two XX cm extension pieces to use when the crops grew. Inside each chamber there was a fan to ensure circulation and mixture of the air (two fans when using two extension pieces?). The chambers had a rubber septum for N_2O sampling. In the N_2O subplots metal frames were installed 10 cm into the ground prior to the first measurement and kept there throughout the season (potentially removed if something was happening in the field?). These frames served as a base for the chambers and together with straps to fix the chambers to the frames they created a closed system inside the chambers. The sampling procedure for all N_2O measurements was the same for both experiments. Four 10 ml gas samples were taken through the septum from each chamber using a plastic syringe with a needle (producer) during a time span of 45 < x < 120 min and stored in 6 ml vials (producer) until analysis. Gas samples were analyzed for N_2O and CO_2 with a dual-inlet Agilent 7890 gas chromatograph with an autosampler (Confirm with TI for the analysis of N_2O).

2.5. Soil sampling

Soil samples were collected weekly outside the frames within the N_2O subplots (need to be precise with the distance from the frames as this could cause issues). A soil auger (XX mm) was used to collect 4-6 soil samples at a depth of 25 cm in each N_2O subplot and pooled together to have one composite soil sample per plot.

Mineral N determination, pH, other aspects.

2.6. Aboveground biomass sampling

In all the plots, the above-ground biomass was assessed by cutting vegetation in squares of 0.25 m2 (0.5 * 0.5 m) in each joint (at 4 repetitions, cut 2 fields in each repetition) within the selected plot. The crop was cut with scissors approximately 1 cm above the ground. All cut plant parts of the crop were included in the sample sent to the laboratory (from Ashley).

Did we determine the N content?

2.7. Statistical analysis

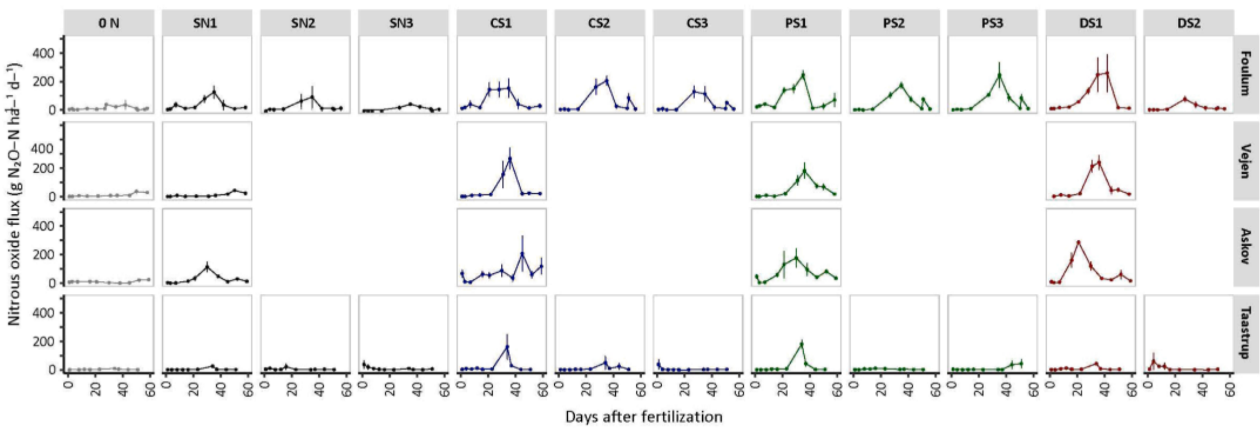
TI for the analysis.

3. Results

3.1 Climate and environmental variables

3.2 N_2O emissions in winter wheat

Proposed figure for each year that shows the comparison of temporal N_2O fluxes between treatments and across locations.



3.1.1-3.1.3 A separate section for each year (2022-2024)

- Holeby: Treatment effect or the effect of the NI on the N_2O flux and cumulative N_2O fluxes.
- Ringsted: Treatment effect or the effect of the NI on the N_2O flux and cumulative N_2O fluxes.

- Aarhus: Treatment effect or the effect of the NI on the N_2O flux and cumulative N_2O fluxes.
- Compare the N_2O flux and cumulative N_2O fluxes between locations, which could be based on specific treatments. Specifically, is there an effect of location on the effect of NI with 50% NH_4^+.
- Compare the N_2O flux and cumulative N_2O fluxes between years (is there a year effect?).

Proposed Table to show the cumulative N2O emissions and N2O emission factors.

Table 1
Total N added (kg N ha^{-1}), cumulative N_2O emissions (kg N_2O-N ha^{-1}), and N_2O emission factors (EF; %) for both 2020 with spring barley in ploughed soil and in 2021 with winter wheat in ploughed and direct seeded soil. Statistically significant differences ($p < 0.05$) between an N source (NS, UAN, or PS) and associated NIs: DMPP, NP, and Piadin are indicated by different letters. Values represent means ± the standard error of the mean ($n = 3$).

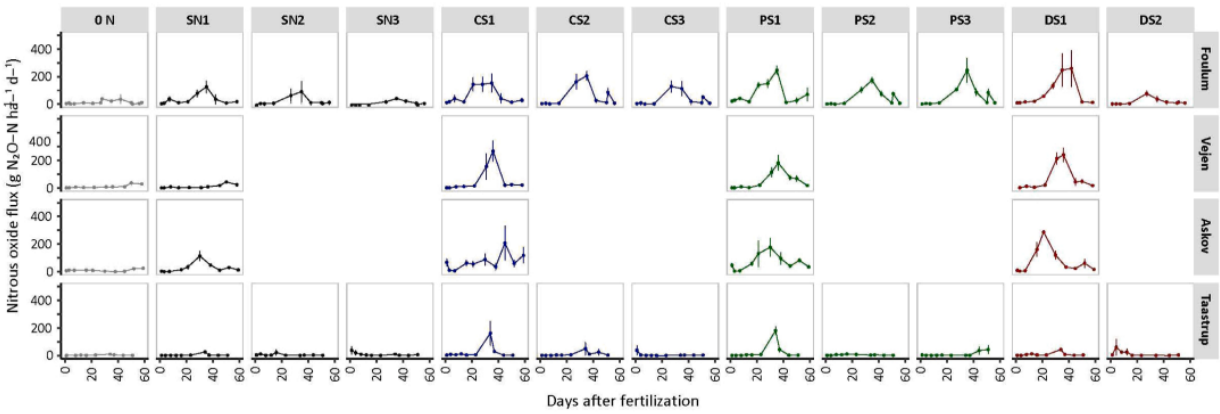
2020 – Spring barley	Total N added	N_2O-N	N_2O EF (%)
	kg ha^{-1}	kg ha^{-1}	(%)
0 N	0	0.10 ± 0.03**b**	NA
PS	165	2.61 ± 0.78**a**	1.52 ± 0.49**a**
NS	124	0.26 ± 0.12**b**	0.12 ± 0.09**b**
UAN	124	0.37 ± 0.07**b**	0.21 ± 0.03**b**
PS	165	2.61 ± 0.78**a**	1.52 ± 0.49**a**
PS + DMPP	165	1.09 ± 0.34**ab**	0.60 ± 0.22**ab**
PS + NP	165	0.26 ± 0.11**b**	0.10 ± 0.07**b**
PS + Piadin	165	0.25 ± 0.05**b**	0.09 ± 0.04**b**
NS	124	0.26 ± 0.12**a**	0.12 ± 0.09**a**
NS + DMPP	124	0.15 ± 0.04**a**	0.04 ± 0.01**a**
UAN	124	0.37 ± 0.07**a**	0.21 ± 0.03**a**
UAN + NP	124	0.14 ± 0.02**b**	0.03 ± 0.02**b**
UAN + Piadin	124	0.05 ± 0.02**b**	−0.04 ± 0.02**b**

Discuss the statistical differences between treatments, years, and locations

3.3 Mineral N dynamics

3.3.1-3.3.3 A separate section for each year (2022-2024)

3.4 N_2O emissions in Spring barley



3.4.1-3.4.3 A separate section for each year (2022-2024)

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- Ringsted: Treatment effect or the effect of the NI on the N_2O flux and cumulative N_2O fluxes.
- Aarhus: Treatment effect or the effect of the NI on the N_2O flux and cumulative N_2O fluxes.
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PS + NP	165	0.26 ± 0.11**b**	0.10 ± 0.07**b**
PS + Piadin	165	0.25 ± 0.05**b**	0.09 ± 0.04**b**
NS	124	0.26 ± 0.12**a**	0.12 ± 0.09**a**
NS + DMPP	124	0.15 ± 0.04**a**	0.04 ± 0.01**a**
UAN	124	0.37 ± 0.07**a**	0.21 ± 0.03**a**
UAN + NP	124	0.14 ± 0.02**b**	0.03 ± 0.02**b**
UAN + Piadin	124	0.05 ± 0.02**b**	−0.04 ± 0.02**b**

3.5 Mineral N dynamics

3.5.1-3.5.3 A separate section for each year (2022-2024)

3.6 N_2O emission factors

- Compare N2O emission factors among the different treatments, locations, years, and if possible, crops (winter wheat vs. Spring barley).