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Effect of slurry application techniques on nitrous oxide emission from temperate grassland under varying soil and climatic conditions

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Abstract

The effect of slurry application techniques and slurry N stabilizing strategies on nitrous oxide emission from grasslands is poorly understood and, therefore, can result in large uncertainties in national/regional inventories. Field experiments were, thus, conducted to estimate the effect of different fertilization techniques on nitrous oxide (N_2O) emissions. Fertilizer was applied (135–270 kg N ha^{-1} $year^{-1}$) as calcium ammonium nitrate (CAN), untreated or treated cattle slurry. The slurry was either treated with sulfuric acid (target pH = 6.0), applied using trailing shoes or treated with 3,4-dimethyl pyrazole phosphate and applied via slot injection. N_2O fluxes were sampled using the closed chamber technique. Cumulative N_2O emissions ranged 0.1–2.9 kg N ha^{-1} $year^{-1}$ across the treatment, sites and years. The N application techniques showed inconsistent effects on soil mineral N content, cumulative N_2O emission and N yield. The fertilizer replacement value of slurry was low due to low N use efficiencies at the sites. However, a close positive relationship ($r = 0.5$; $p = .013$) between slurry value and biomass yield was observed, highlighting the benefit of high slurry value on crop productivity. N_2O-N emission factors were low for all treatments, including CAN, but were 2–6 times higher in 2019 than in 2020 due to lower precipitation in 2020. Variations in N_2O emission were largely explained by soil and climatic factors. Even with the low N_2O emissions, this study highlights the benefit (significant mitigation of N_2O emissions) of replacing the increasingly expensive chemical fertilizer N with input from slurry under favourable conditions for denitrification.

KEYWORDS
greenhouse gas emissions, livestock, manure, nitrogen cycling, nitrogen use efficiency

1 | INTRODUCTION

The current generation is faced with feeding an increasing global population, projected to exceed 9.7 billion by 2050 (United Nations, 2019). This obligation requires sustainable use of fertilizer to increase the agricultural food base with less contamination of the environment (Billen et al., 2015). Currently, the plant takes up only between 30.2% and 53.2% of fertilizer N applied to agricultural soils (Anas et al., 2020). The remaining is vulnerable to losses to the atmosphere as nitrous oxide (N_2O), ammonia (NH_3), dinitrogen (N_2), nitrogen oxides (NO_x) and leached or run off into ground or surface water as nitrate (NO_3^-), ammonium (NH_4^+) and dissolved organic N (Anas et al., 2020). Globally, about 62% of atmospheric N_2O emissions originate from soils, equivalent to about 13 Mt N_2O–N annually (Fowler et al., 2013), with agricultural soils responsible for more than 50% of the European Union's N_2O budget (Kolasa-Więcek, 2018). Nitrous oxide is a potent greenhouse gas, 310 times stronger than CO_2 on a 100-year scale (IPCC, 2022), and detrimental to the ozone layer, the most critical threat to stratospheric damage (Ravishankara et al., 2009). Nitrous oxide can arise as a by-product of processes regulated by soil conditions, such as nitrification and/or denitrification pathways (Saggar et al., 2013). Increases in N_2O are usually associated with excessive N inputs, particularly in agricultural soils, suggesting inefficient nutrient management rather than ecosystem inefficiency. Consequently, increased N_2O emission from elevated N fertilizer use increases climate change, exacerbates stratospheric ozone layer depletion and increases the social cost of N_2O emission (Kanter et al., 2021).

Unarguably, efficient N management in agriculture is required to increase production to preserve biodiversity and environmental resources. Therefore, the call to increase N use efficiency and reduce fertilizer losses cannot be overemphasized. Organic fertilizers have the potency to meet the nutritional needs of crop plants, and their usage is encouraged, particularly as a significant increase in manure N production over the last decades has occurred (Chojnacka et al., 2020). However, organic fertilizers have to undergo mineralization, leading to nutrient losses to the environment, mainly in times with low N demand of the plants, with effects on the overall Mineral Fertilizer Replacement Value (Jensen, 2013). Nitrous oxide is one of the most challenging greenhouse gasses to reduce (Seitzinger & Phillips, 2017). However, some strategies to minimize N_2O emission include timing N input to avoid moments of high emissions (Maris et al., 2021), timing fertilizer application and plant growth (Lassaletta et al., 2014) and reducing soil compaction (Hernandez-Ramirez et al., 2021). The fertilizer placement technique may also influence nutrient losses. For example, slurry injection on grasslands has been shown to deliver slurry close to the root zone of plants for optimum uptake and, hence, reduce NH_3 losses compared with broadcasting or trailing hose application techniques (Rodhe, 2004). However, slurry injection can cause "pollution swapping", increasing N_2O emission compared with surface application methods as more conducive conditions are created in the soil for denitrification (i.e., readily available organic carbon in combination with soil zones with low O_2 concentrations; Herr et al., 2019).

The use of N stabilizing compounds to extend the residence time of applied N in the soil is a promising option to increase N use efficiency and reduce N losses to the environment (Harty et al., 2016). Accordingly, nitrification inhibitors (NIs) and urease inhibitors have been suggested to counteract N_2O emissions from grasslands (Subbarao et al., 2007; Krol et al., 2020). Nitrification inhibitors inhibit or delay the conversion of NH_4^+ to NO_3^- (Ruser & Schulz, 2015), desynchronize C and NO_3^- availability and thus can reduce N_2O emissions from denitrification after slurry injection (Herr et al., 2020). Although meta-analysis revealed similar global N_2O reduction potentials between approximately 35% and 56% (i.e. Akyama et al., 2010; Fan et al., 2022; Soares et al., 2023), their effectiveness varied widely depending on the environmental conditions. Soil texture, soil pH, organic carbon content, soil moisture, soil and air temperatures and seasonal precipitation significantly affected N_2O reduction by NIs (Fan et al., 2022; Soares et al., 2023). Recently, Soares et al. (2023) summarized results from NI studies in grazing systems. From the 61 studies, only three covered measurements after slurry application with 3,4-dimethylepyrazole phosphate (DMPP), the most commonly used NI in Germany. All three studies were conducted in Spain under different climatic conditions compared to those at German grassland sites. Further, the N_2O measurements of these three studies lasted between 3 weeks and approximately 3 months. Fan et al. (2022) pointed out that long-term measurements (exceeding the cropping season), taking possible residual effects of inhibitors into account, are particularly important. Such effects, as a reduced CO_2 release during late autumn or even in winter, or unexpected long effective duration of the inhibitor, have been reported for the use of DMPP at German study sites in horticulture (Pfab et al., 2012) and in winter wheat production (Weiske et al., 2001). Accordingly, data on the N_2O emission from slurry application with NI under the different German climatic and diverse environmental conditions measured over a more extended period are urgently required.

Slurry acidification is an efficient technique to reduce NH_3 volatilization (Keskinen et al., 2022); however, its effect on N_2O emission is not consensual as enhancing effects (Gómez-Muñoz et al., 2016), no effects (Malique et al., 2021), and mitigating effects (Owusu-Twum et al., 2017; Park et al., 2018) have been observed. Furthermore, the results of slurry acidification on soil microbial structure and function, Mineral Fertilizer Replacement Value (MFRV) and herbage yields, particularly in permanent grasslands, are unclear due to limited research (Fangueiro et al., 2015). However, higher organic matter mineralization and solubility have been observed with acidified slurry, resulting in increased nutrient supply to microbes and crops, increasing crop growth and N uptake and reducing N_2O emissions (Gómez-Muñoz et al., 2016; Nyameasem et al., 2021).

Nitrous oxide emission contributes significantly to the greenhouse gas inventory of many European countries (European Environment Agency, 2021). Various policies at the global and regional levels have placed an obligation on member countries to set greenhouse gas mitigation targets and report progress in annual inventories. A recent study using published data across Germany (Mathivanan et al., 2021) broadly showed that 0.38% to 0.92% of applied N is lost directly as

TABLE 1 Meteorological, soil chemical and physical characteristics of the experimental sites in 2019 and 2020 in the study regions Schleswig-Holstein (SH), Baden-Württemberg (BW), and Niedersachsen (LS). All soil analysis: 0–0.3 m depth.

Region	Site (abbrev.)	Year	Coordinates	Soil type[a]	MAP/Exp. year (mm yr^{-1})	MAT/Exp. year (°C)	Soil texture Clay (%)	Silt (%)	Sand (%)	Soil BD g cm^{-3}	pH ($CaCl_2$)	CEC ($mmol_c$ kg^{-1})	C_{org} (g kg^{-1})	N_t (g kg^{-1})	C/N g g^{-1}
SH	Holtsee (HS)	2019	N 54°23′16″ E 09°50′46″	Luvisol	847/714	8.8/11.1	11	33	57	1.39	5.9	74.3	25.3	2.1	12
		2020	N 54°23′16″ E 09°50′46″	Luvisol	847/646	8.8/9.8	8	25	67	1.47	5.8	50.9	16.4	1.4	12
	Bredenbek (BR)	2019	N 54°03′16″ E 09°51′30″	Luvisol	847/709	8.8/11.1	11	30	59	1.45	5.4	53.4	18.4	1.7	11
		2020	N 54°03′16″ E 09°51′30″	Luvisol	847/646	8.8/9.8	12	32	56	1.32	5.6	68.8	24.6	2.5	10
LS	Osnabrück (OS)	2019	N 52°15′47″ E 08°13′53″	Plaggic Anthrosol	883/602	9.5/11.4	12	20	68	1.16	5.0	99.0	18.0	1.7	11
		2020	N 52°14′10″ E 08°15′05″	Gleysol	883/712	9.5/9.6	7	42	51	1.24	5.5	89.0	16.6	1.7	10
BW	Hohenheim (HH)	2019	N 48°42′55″ E 9°13′11″	Calcaric Regosol	736/532	9.7/11.5	21	67	12	1.30	6.5	176.0	29.9	3.0	10
		2020	N 48°43′02″ E 9°13′06″	Calcaric Regosol	736/464	9.7/10.8	15	69	16	1.31	6.4	154.0	25.2	2.5	10

Note: MAP/Exp year: Long-term mean annual precipitation and annual precipitation in the single experimental years (year 2019 = February 2019–January 2020; year 2020 = February 2020–January 2021); MAT/Exp. year: Long-term mean annual air temperature (2 m) and annual mean air temperature in the single experimental years (2019/2020).
Abbreviations: CEC, cation exchange capacity; Corg, organic carbon; Nt, total nitrogen; Soil BD, soil bulk density.
[a]IUSS Working group WRB (2015).

N_2O. The study also showed that N_2O emission factor was lower for the north of Germany than for the south, probably due to the frequent occurrence of frost/thaw cycles in south Germany (Jungkunst et al., 2006). However, adequate N_2O measurement data, covering many environmental and management practices and providing comparable and consistent data for the different slurry applications and treatment techniques on grasslands, is needed to improve accountability. Nitrogen uptake, a primary pathway of N recovery from grassland ecosystems, is dependent on the productivity of the swards. Although the last decade has seen some studies reporting relatively low N_2O emission factors for organic inputs from European grasslands and grass-clover swards (e.g., Fuchs et al., 2020; Nyameasem et al., 2021; Reinsch et al., 2020), the effect of perennial ryegrass, one of the most dominant and productive forage grass species in temperate regions of the world, including Western Europe (Wilkins & Humphreys, 2003), on N_2O emission is not well understood. As many countries, including Germany, are beginning to adapt emission factors for more accurate reporting, this study contributes additional empirical data to previous studies elsewhere (Bourdin et al., 2014; Cahalan et al., 2015; Melaku et al., 2020; Minet et al., 2016) for modelling and decision making. Accordingly, our study reports N_2O emissions and MFRV from different slurry application techniques on permanent grasslands under different commercial field conditions, differing in soil and weather conditions. We hypothesized that

i. slurry acidification does not increase N_2O emission but increases nutrient supply for biomass growth and increases N recovery,
ii. adding nitrification inhibitor to injected slurry reduces N_2O emission and increases the MFRV of slurry applied to grasslands,
iii. slurry injection relative to applying slurry with Trailing Shoe on grassland increases direct N_2O emission but increases MFRV through the abatement of NH_3 volatilization and minimal losses due to surface run-off, thus increasing the availability for plant N-fertilizer uptake.

2 | MATERIALS AND METHODS

2.1 | Study site characteristics

The experiments were conducted on meadows at four grassland sites over 2 years in Germany between spring 2019 and spring 2021 in three regions: Baden-Wuerttemberg (Hohenheim; HH) in the south, Lower Saxony (Osnabrück; OB) in the west and Schleswig-Holstein (Holtsee; HS) and Bredenbek; BR) in the north (Table 1). The trial sites were changed to nearby fields with similar soil properties in the second year. The regions were chosen since they followed a gradient covering a broad range of climatic conditions for regions with high grassland portions in Germany. The long-term annual precipitation and mean air temperature were 847 mm and 8.8°C in Schleswig-Holstein, 883 mm and 9.5°C in Lower Saxony and 736 mm and 9.7°C in Baden-Wuerttemberg, respectively (Table 1). Soil types at the study sites were Luvisols in Schleswig-Holstein, Calcaric Regosols in Baden-Wuerttemberg, Plaggic Anthrosol in 2019 and Gleysol in 2020 in Lower Saxony, with soil texture ranging between clayey silt and loamy sand (Table 1). Soil classes were derived from the World Reference Base for Soil Resources (WRB, 2015). The meadows were dominated by perennial ryegrass (*Lolium perenne* L.) at Schleswig-Holstein and Lower Saxony, and Italian ryegrass (*Lolium multiflorum* L.) in Baden-Wuerttemberg, with a high species richness (Table S1 in Data S1), probably due to the low cutting frequency (two to three cuts per year) practised in this region.

2.2 | Experimental design and fertilizer application

The field experiments were designed as a fully randomized complete block design with four replicates at every study site. The plot size was 81 m^2 (9 × 9 m) with a 6 m (at Schleswig-Holstein) or 12 m (at Baden-Wuerttemberg & Lower Saxony) distance between the plots. At every site, the following six treatments were established:

1. A control, i.e. without N fertilizer (N0),
2. Fertilization with calcium ammonium nitrate (CAN), applied by broadcast,
3. Fertilization with untreated cattle slurry using trailing shoes (TS),
4. Fertilization with acidified cattle slurry (target pH = 6, adjusted with sulfuric acid) using trailing shoes (TS + A),
5. Fertilization with untreated cattle slurry using open slot injection (SI), and
6. Fertilization with cattle slurry treated with DMPP and applied by open slot injection (SI + NI).

Depending on the cutting frequency and the corresponding N demand, N fertilizer was applied in splits of 2 to 3, supplying a total of 135–270 kg N ha^{-1} $year^{-1}$ with the highest, intermediate and lowest rates in Lower Saxony, Schleswig-Holstein and Baden-Wuerttemberg, respectively (Table S2 in Data S1). In accordance with the German Fertilizer Ordinance (Dü, 2017), the maximum cattle slurry application rate was limited to 170 kg N ha^{-1} $year^{-1}$. As reported by Peters et al. (2021), N uptake of a German grassland site was highest between the beginning of the vegetation period and the first cut, and it decreases with further cuttings over time. Thus, in accordance with Peters et al. (2021) and in agreement with the recommendations of the regional agricultural extension services, we decided to split the 170 kg N ha^{-1} $year^{-1}$ of applicable slurry N into two doses with 100 and 70 kg N ha^{-1} for the first and second dose, respectively. Differences between the targeted 170 kg total N ha^{-1} and the applied amounts of total N with slurry during the first two N applications were the result of a deviation from total slurry N determined in the slurry storages on the farms providing the experimental slurry approximately one month before spreading and the total N determined in the tanks of the experimentally used tanks during slurry application. The first dose of cattle slurry was applied in early spring (at the beginning of the vegetation period), and the second dose was applied within 2 weeks after the first silage cut. The cattle slurry used in both

years was sourced from the same local farm close to the study sites to minimize the effect of different feeding strategies on slurry characteristics (Table S3 in Data S1).

In cases where the N demand exceeded the 170 kg N ha^{-1} $year^{-1}$ limit for total N application via organic fertilizers, mineral N fertilizer was applied. According to common agricultural practice, an additional 30 kg N ha^{-1}, after the second cut, was applied as CAN, the most used mineral N-fertilizer in Germany. No mineral N was applied in HH, the site with only two cuttings. In Lower Saxony, technical problems emerged with the slurry spreader before the first N application in 2020. Therefore, departing from the common N-fertilization strategy followed at all study sites, 100 kg CAN-N ha^{-1} was applied as the first dose, followed by slurry as the second and third doses in 2020 (Table S2 in Data S1). To keep the comparability with the slurry amounts at the remaining study sites, slurry doses in Lower Saxony in 2020 were split into 100 kg N ha^{-1} after the first and 70 kg N ha^{-1} after the second cutting. Additional sulfur (24–30 kg ha^{-1}) was applied as kieserite to all plots, including the acidified plots to ensure that low sulfur fertility was not a limiting factor for any treatment. In both years, DMPP was used as a nitrification inhibitor (2019: 6 L ha^{-1} Entec-FL® and 2020: 2 L ha^{-1} Vizura®).

A customized slurry spreader (Samson Agro A/S, Viborg, Denmark), designed for experimental purposes, was used for applying the slurry. The spreader, with a working width of 3 m, was equipped with 1 m^3 tank, 12 trailing shoes and 12 double discs for slot injection (injection depth of approximately 5 cm). A piece of technical equipment was installed on the tank to enable a fast change of application technique hydraulically. Before application, slurry sub-samples were taken directly from the tank and were frozen at −18°C for analysis by AgroLab LUFA GmbH (see Table S3 in Data S1). The slurry strips of trailing shoes had a mean width of 0.09 m (corresponding to 36% of the total area covered by slurry), while the mean width of slurry from injection slots was 0.05 m (corresponding to 20% of the total area covered by slurry).

2.3 | N_2O flux measurements

Nitrous oxide flux measurements were conducted using circular closed chambers, according to Hutchinson and Mosier (1981). The chambers were constructed from polyvinyl chloride with the same dimensions (for more details, see Flessa et al., 1995). The inner diameter was 0.4 m, but heights ranged from 0.17 to 0.43 cm across sites due to different insertion depths of the base rings and the use of extensions; thus, the chamber volumes ranged from 21 to 54 L. After slurry application, base rings were placed in each experimental plot to achieve a proportional distribution of the soil covered with slurry relative to the total soil area (i.e., 36% of the soil covered with slurry in the trailing shoe treatments and 20% in the injection treatments). Gas sampling after a fertilizer application followed a typical frequency of five times per week for 2 weeks, followed by twice per week for another 2 weeks and then once a week until the next fertilizer application (Harty et al., 2016). However, additional flux measurements complemented weekly sampling after events which frequently induced increased N_2O flux rates (e.g., high precipitation or soil freezing–thawing cycles). As described by Flessa et al. (2002), this sampling strategy leads to a deviation of the cumulative N_2O emission of about 10% compared to continuous measurements.

Gas sampling was conducted between 09:00 and 11:00 am. This sampling time was chosen because it was shown that mid-morning sampling effectively represents the daily N_2O emission average (Machado et al., 2019). During each sampling, four gas samples were periodically (0, 20, 40, and 60 min after chamber closure) taken with a syringe from the chambers' atmosphere and transferred into evacuated gas vials. Simultaneously to gas sampling, air temperature in the chambers was recorded. Gas samples were analysed for N_2O concentrations in the laboratories of the participating research groups by various gas chromatographs (GCs) equipped with Ni^{63} electron capture detectors and automated samplers. In an earlier study, laboratory inter-comparability was verified by conducting blind inter-comparison measurements between the labs (Ruser et al., 2017). All GCs used in our study were also tested by Ruser et al. (2017) and achieved a coefficient of variance below 2% on 10 repeated measurements of an ambient N_2O standard gas.

Molar gas concentrations were transformed into mass concentrations according to the ideal gas law, taking chamber temperature and standard pressure into account. Flux rates were calculated with the robust linear regression model (Huber, 1981), but linear regression was applied if only three of the four gas samples were available for flux determination. Cumulative annual N_2O emissions were calculated between February 1 and January 31 separately for each year and site by stepwise interpolation, assuming constant flux rates beginning with the date of each gas sampling until the subsequent gas sampling. The yield-related N_2O emission was estimated by dividing the annual N_2O-N emission by the dry mass yield of the grass (annual sum of all clippings). The N_2O emission factor (N_2O-N EF) was estimated using equation Eq. (1) as described by Velthof and Mosquera (2011).

$$N_2O-N\,EF = (N_2O - N_{fertilized} - N_2O - N_{unfertilized}) \times \text{total}\,N_{Fert}{}^{-1} \times 100 \quad (1)$$

with N_2O-N EF being N_2O-N emission factor (%); N_2O–$N_{fertilized}$ is the mean cumulative direct N_2O–N kg N_2O–N ha^{-1} $year^{-1}$ emission of the particular experimental year from the fertilized treatments; N_2O–$N_{unfertilized}$ being annual N_2O-N emitted from the unfertilized control (N0) (kg N_2O–N ha^{-1} $year^{-1}$), and total N_{fert} is the total fertilizer-N applied via CAN or cattle slurry (kg N ha^{-1} $year^{-1}$).

2.4 | Climatic and soil variables

Climate stations were installed in the middle of each plot at each study site at the beginning of the experiment to measure daily precipitation (mm) and air temperature (°C) at 2 m height, as well as soil temperature (°C) at 5, 10 and 15 cm depths. Weather data from the closest weather station of the German Weather Service was used for gap filling.

Before the start of the trials, soil samples were taken from the topsoils (0–30 cm depth) at each experimental site to determine the

physical and chemical properties of the soils. The soil samples were mortared, pre-dried at an appropriate temperature and stored in a desiccator until further analysis. Soil texture was determined according to DIN EN ISO/IEC (17025:2018) and soil pH using 0.01 M $CaCl_2$ as an extractant. Cation exchange capacity (CEC) determination also followed the standardized method of the VDLUFA (2017), while soil organic C and total N content were analysed by dry combustion (Yeomans & Bremner, 1991).

Soil samples were regularly taken from the topsoil (0 to 0.3 m depth) every fortnight at the same time as the gas sampling and on selected supplementary gas sampling dates to determine mineral N and soil moisture. Three samples were taken per plot separately for the slurry-covered and uncovered soil during the growing season. These soil samples were then pooled over the four replicated plots resulting in one composite sample of slurry-covered soil and one sample of uncovered soil per treatment and sampling date. In addition, we took three samples from each treatment outside the growing season and pooled them into one composite sample per each treatment. The soil samples were homogenized and stored at −20°C. Frozen samples were thawed overnight at 4°C prior to analysis. N samples were extracted with 2 M KCl solution using an extraction ratio of 1:4. Nitrate and ammonium concentrations in the filtrates were determined using flow injection analysis. The corresponding area-weighted data represented the mineral N concentrations of the separately sampled areas (slurry-covered and uncovered soil).

Soil moisture was determined for the soil layer 0–0.3 m gravimetrically by drying a soil aliquot at 105°C overnight. Also, to determine soil bulk density, four soil cores were collected per plot using stainless steel cylinders (100 cm^3) in the early autumn of each experimental year. The soil cores were crushed in the lab and dried at 105°C until a constant weight was attained. Water-filled porosity indicates soil aeration in soils with different bulk densities (Granli & Bøckman, 1994). Therefore, water-filled pore space (WFPS) [%] was calculated as shown in Eq. (2), where VWC is the volumetric water content of the soil, calculated as the product of the gravimetric water content and soil bulk density (BD); and PD is particle density of 2.65 g cm^{-3}.

$$\mathrm{WFPS} = 100 \times \frac{\mathrm{VWC}}{\left(1 - \frac{\mathrm{BD}}{\mathrm{PD}}\right)} \tag{2}$$

2.5 | Biomass sampling and analysis

To determine the aboveground biomass of the meadows, plant samples were cut manually (in Schleswig-Holstein and Lower Saxony, two subsamples per plot with a size of 0.25 m^2 each) or using a forage harvester in Baden-Wuerttemberg. To ensure comparability of results across all sites, the yields were checked at the Baden-Wuerttemberg site to ensure that scaling up of subsamples from the forage harvester produced similar results to scaled-up yields at Schleswig-Holstein and Lower Saxony. The number of harvests during the experimental period varied across the study sites. In Schleswig-Holstein, five silage cuts were carried out in both years and at both sites; in Lower Saxony, four cuts were performed, whereas in Baden-Wuerttemberg, three cuts were executed in 2019 and only two in 2020, with the latter being a result of low precipitation. Harvest dates at each site were based on local growing conditions and were between the heading and flowering of the dominant grass species. The plant samples (in Schleswig-Holstein and Lower Saxony) or an aliquot of the cut (in Baden-Wuerttemberg) were dried at 58°C until constant weight. Dry samples were milled <1 mm, and the N content was analysed using near-infrared-reflectance spectroscopy with a NIR-Systems 5000 monochromator (Foss, Silver Spring, USA).

2.6 | Calculations and statistical analyses

2.6.1 | Apparent N use efficiency (aNUE) and N fertilizer replacement value (MFRV)

N yields obtained from total N input were considered to calculate apparent N use efficiency (aNUE) (Sistani et al., 2010) as follows:

$$a\mathrm{NUE} = \frac{\text{N uptake from fertilized treatment} - \text{N uptake from unfertilized treatment}}{\text{Total N applied}} \tag{3}$$

where the total plant N uptake is the total amount of N in the harvested biomass of all clippings (kg N ha^{-1}); total N applied is total N input, including all N dressings (kg N ha^{-1}).

The N fertilizer replacement value (MFRV) of applied cattle slurry was estimated using the following equation:

$$\mathrm{MFRV} = \frac{a\mathrm{NUE}\text{ of slurry treatment}}{a\mathrm{NUE}\text{ of CAN treatment}} \tag{4}$$

2.6.2 | Statistical analysis

All statistical analyses and visualizations were done using R software (R Core Team, 2021; Version 4.2.1). The data were analysed using a mixed model using the package "nlme". The statistical model included treatment, year of experimentation and experimental site as fixed factors and the experimental block as a random factor. Statistical significance of the tested treatments was declared when $p < .05$. We tested the effects of the N application techniques on the response ratio (RR) of

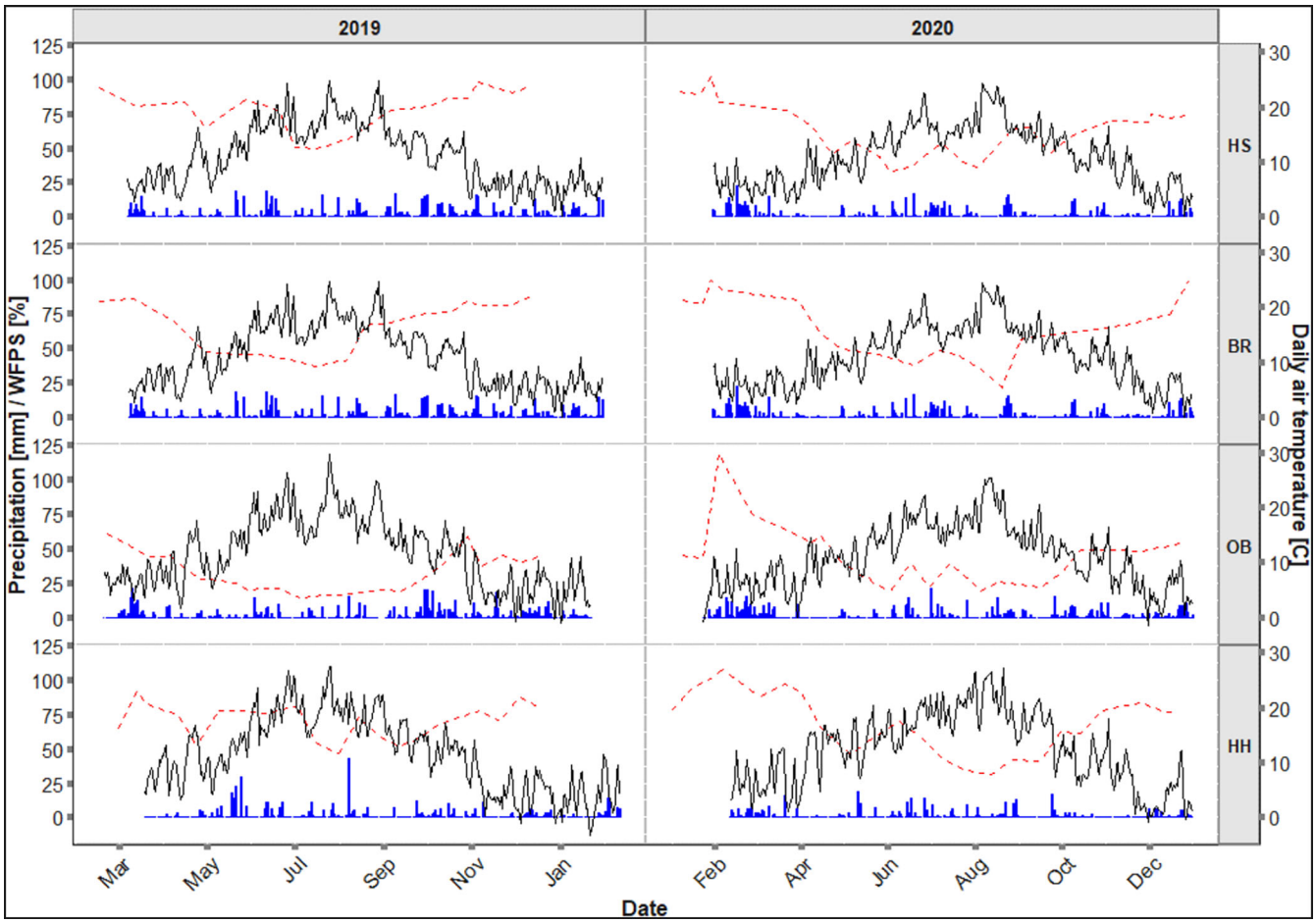

FIGURE 1 Average daily air temperature (black lines), total daily precipitation (blue bars) and water-filled pore space (broken red lines) trend during the experimental years at Holtsee (HS), Bredenbek (BR), Osnabrück (OB) and Hohenheim (HH). Green and red arrows indicate slurry and calcium ammonium nitrate fertilizer applications, respectively.

cumulative N_2O emissions by linearizing N_2O RRs through transforming via natural logarithms ('Euler's number as a base, $e = 2.718$) to obtain Ln N_2O RR (Eq. 5). This RR approach enabled us to account for management and environmental divergence across the study sites, by normalizing the ratios to 1 (Hernandez-Ramirez et al., 2021). The Ln zero baselines were compared with zero baselines (control) using t-tests (Ho: $\mu RR = 0$) and 95% confidence intervals to determine whether the N treatments differed significantly from the control,

$$\text{Ln}\,N_2O\,RR = \text{Ln}\left(\frac{N_2O\,\text{treatment}}{N_2O\,\text{control}}\right) \quad (5)$$

Associations between the measured N_2O emission, soil and average weather factors were assessed with data from all treatments, years and sites using Pearson's correlation and regression tests. Consequently, we assessed the importance of these factors (soil and weather variables, N application rates and *L. perenne* proportion in the swards) on N_2O emissions and fitted a statistical model to the data to predict N_2O emissions. A stepwise forward selection procedure using the "olsrr" package (version 0.5.3) of R was implemented to improve the model. The best model was determined after removing collineated variables, ensuring variance inflation factors did not exceed two. Accordingly, the best model is parameterized by N application rate, clay content in soils, soil pH, total rainfall and mean air temperature (Tables S7 and S8 in Data S1). The model efficiency was evaluated using the Nash–Sutcliffe model efficiency coefficient test and the root mean square error.

3 | RESULTS

3.1 | Climatic conditions

Rainfall was generally highest, intermediate and lowest in Schleswig-Holstein, Lower Saxony and Baden-Wuerttemberg, respectively and more frequent in 2019 than in 2020, with the latter being drier across the four sites (Figure 1). Thus, precipitation from April to December was lower by 33%, 36% and 19% at Baden-Wuerttemberg, Schleswig-Holstein and Lower Saxony in 2020 relative to

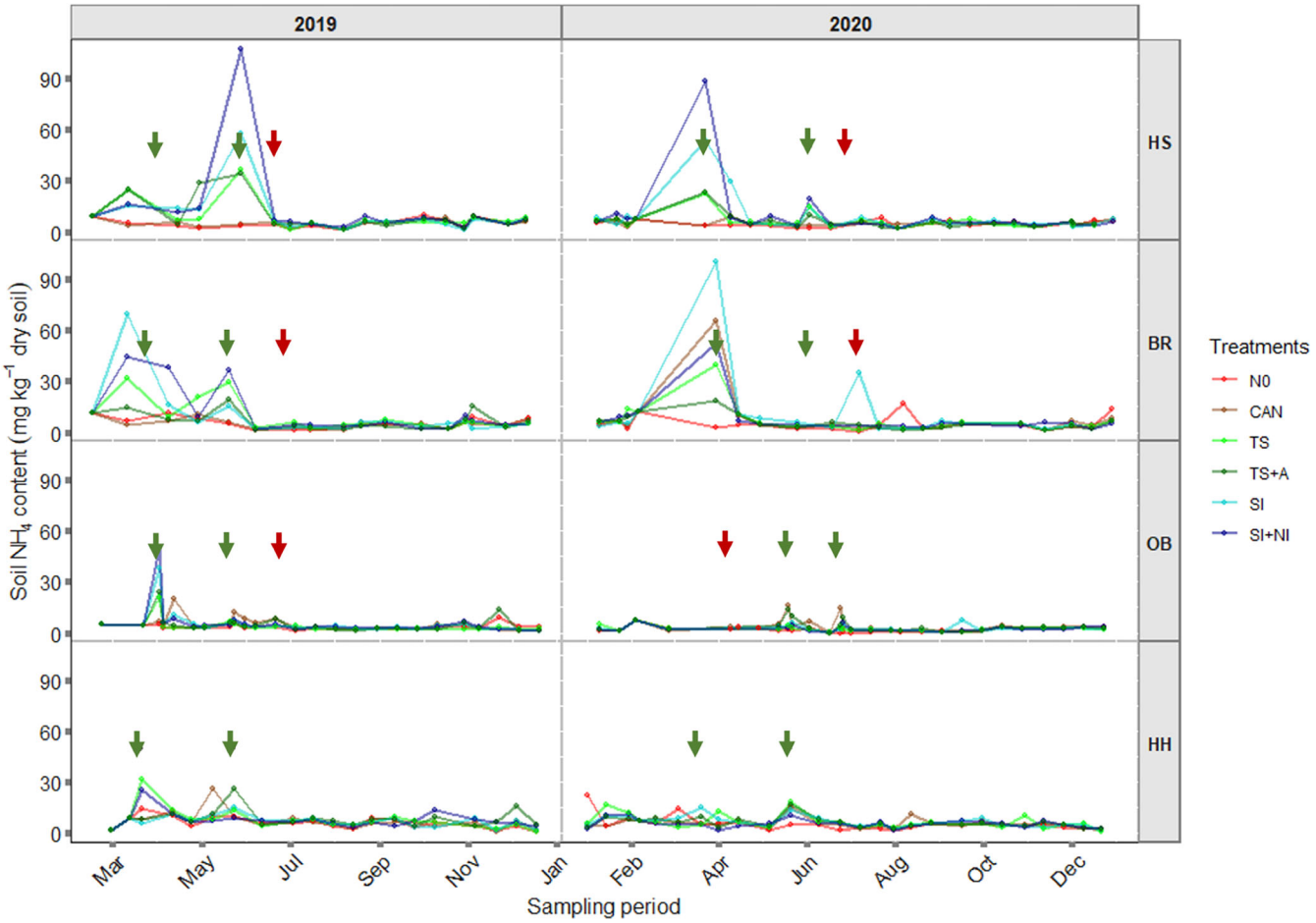

FIGURE 2 Soil ammonium concentrations in soils across the experimental sites and years as affected by treatments at Holtsee (HS), Bredenbek (BR), Osnabrück (OB) and Hohenheim (HH). Arrows point to N fertilization dates; green and red arrows indicate slurry and calcium ammonium nitrate fertilizer applications, respectively.

2019. However, there were alternate wet and dry periods after March, with more severe drier conditions in June and July each year. There was generally no or little rainfall on the days of fertilizer application, but WFPS was high at the beginning of the experiment in each year (above 50%). The WFPS declined after that and reached the lowest levels during the summer months (June to August), with levels below 25% at Lower Saxony in both experimental years (Figure 1). As a result, WFPS was generally higher at the first fertilizer application (above 50%, except at Lower Saxony) and lower at the subsequent fertilizer applications. Comparatively, WFPS were generally low in Lower Saxony, even during the humid year of 2019 (Figure 1).

The air temperatures were relatively low (Figure 1) between November and March (−5 and 10°C) and high between June and August (15 to 28°C). The average air temperature monitored during the measurements (April–December) was slightly higher (3% to 9%) than the long-term averages of the same months and daily average temperatures at the first fertilizer application were mainly lower (<12°C) than for the subsequent applications (>15°C).

3.2 | Soil N dynamics and daily N_2O emissions

Ammonium and nitrate levels in soils were relatively low at HH and OB, even after fertilizer application (Figures 2 and 3), than at HS or BR. The highest peaks observed at BR and HS (>80 mg N kg^{-1}) were mainly from treatments with injected slurry. NO_3^- levels were generally low (<30 mg N kg^{-1}), but a higher peak (>50 mg N kg^{-1}) was observed in the CAN treatment at BR in 2019. Although low, NO_3^- concentrations were slightly higher in the CAN treatment than in the other treatments at HH and OB (Figure 3). The highest NO_3^- peak was observed in the CAN treatment in BR after fertilizer application in 2020. Treatment effects on soil N concentration depended on site and year differences but with no apparent pattern. With a few exceptions, NO_3^-, NH_4^+ and the corresponding N concentrations (sum of NH_4^+-N and NO_3^--N) were comparable for N0 and the fertilized treatments ($p > .05$) (Table S3 in Data S1). N concentrations in soils were comparable between TS and TS + A as well as between SI and SI + NI at all sites (Table S3 in Data S1).

As expected, N_2O peaks were preceded by NH_4^+ and NO_3^- peaks, occurring earlier for NH_4^+ than NO_3^- peaks (Figures 2 and 3).

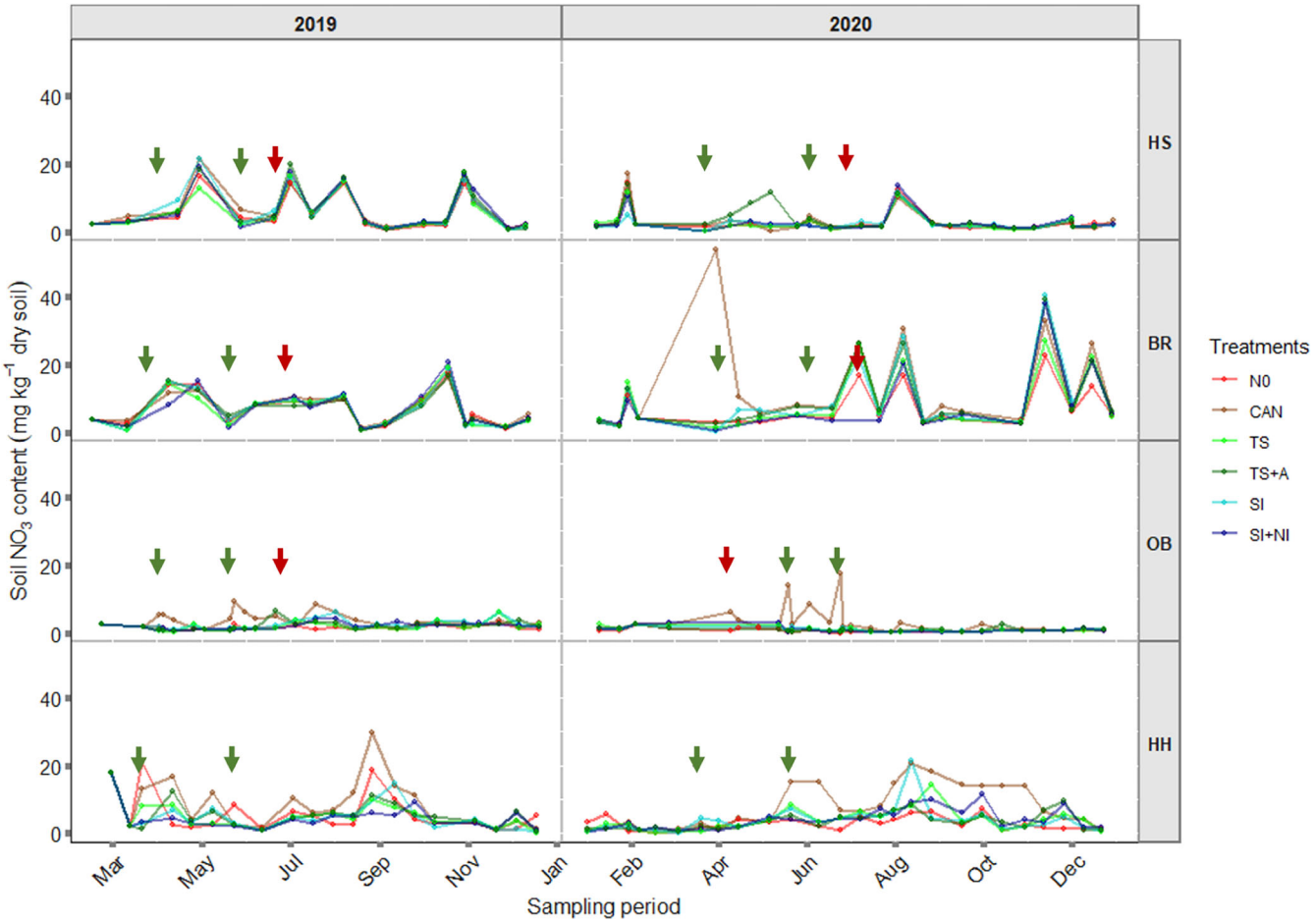

FIGURE 3 Nitrate concentrations in soils across the experimental sites and yeas as affected by treatments at Holtsee (HS), Bredenbek (BR), Osnabrück (OB) and Hohenheim (HH). Arrows point to N fertilization dates; green and red arrows indicate slurry and calcium ammonium nitrate fertilizer applications, respectively.

Although there were NO_3^- peaks from the 2020 N applications, the corresponding N_2O peaks were generally lower in 2020 than in 2019 (Figure 4), with the highest peak in 2020 across all sites being lower than 2.2 mg N m^{-2} d^{-1}. The peaks were consistently low at OB in both experimental years. Average (±s.e) N_2O emissions were 0.20 ± 0.01, 0.29 ± 0.04, 0.33 ± 0.03 and 0.36 ± 0.04 mg N m^{-2} d^{-1} for OB, HS, BR and HH, respectively. In most cases, nitrous oxide peaks occurred a few days after fertilizer N application, but in 2019 a peak was also observed at HH, more than 4 weeks after fertilization. Whereas the highest daily N_2O peaks at HH (0.83 mg N m^{-2}) and OB (0.36 mg N m^{-2}) were observed for the CAN treatment, the highest peaks at BR (13.4 mg N m^{-2}) and HS (11.1 mg N m^{-2}) were observed for the SI + NI treatment.

3.3 | Cumulative N_2O and emission factors

Cumulative N_2O emissions across sites and years ranged from 0.17 to 1.44, 0.17 to 2.83 and 0.12 to 2.89 kg N ha^{-1} $year^{-1}$ for N0, CAN and slurry treatments, respectively. The mean cumulative N_2O emissions (from all sites) were about three times higher in 2019 (1.42 ± 0.14) than in 2020 (0.49 ± 0.04 kg N ha^{-1}). The results from all sites and years showed that fertilizer application significantly ($p < .05$) elevated cumulative N_2O emission by 33 to 117%, depending on treatment type, site and experimental year (Table 2). In 2020, where N_2O fluxes were generally low, the N0 treatment showed comparable ($p > .05$) cumulative N_2O emissions to the slurry and mineral fertilizer treatments in many instances, except at HS (Table 2). Except in one instance, CAN-related emissions were primarily similar (p > .05) to slurry emissions. Cumulative N_2O emissions from acidified slurry were consistently comparable to the non-acidified slurry at each site and year ($p > .05$), except at HH in 2019, where TS + A treatment had a 49% lower ($p < .05$) cumulative N_2O emission compared with TS (Table 2).

Within the fertilized treatments, yield-scaled emission was highest for CAN but comparable to SI + NI treatment at BR, HS and HH; comparable to SI at HS and HH; comparable to TS + A at HS and comparable to TS at HH ($p < .05$). In 2019, the yield-scaled emissions were 53% lower for TS + A relative to TS at HH, while SI + NI treatment was 20% relative to SI at OB. Yield-scaled N_2O emissions were generally comparable for SI + NI and SI treatments across sites, except at OB in 2019, where a 21% lower ($p < .05$) yield-scaled N_2O emissions were observed for SI + NI relative to SI. Overall, using data

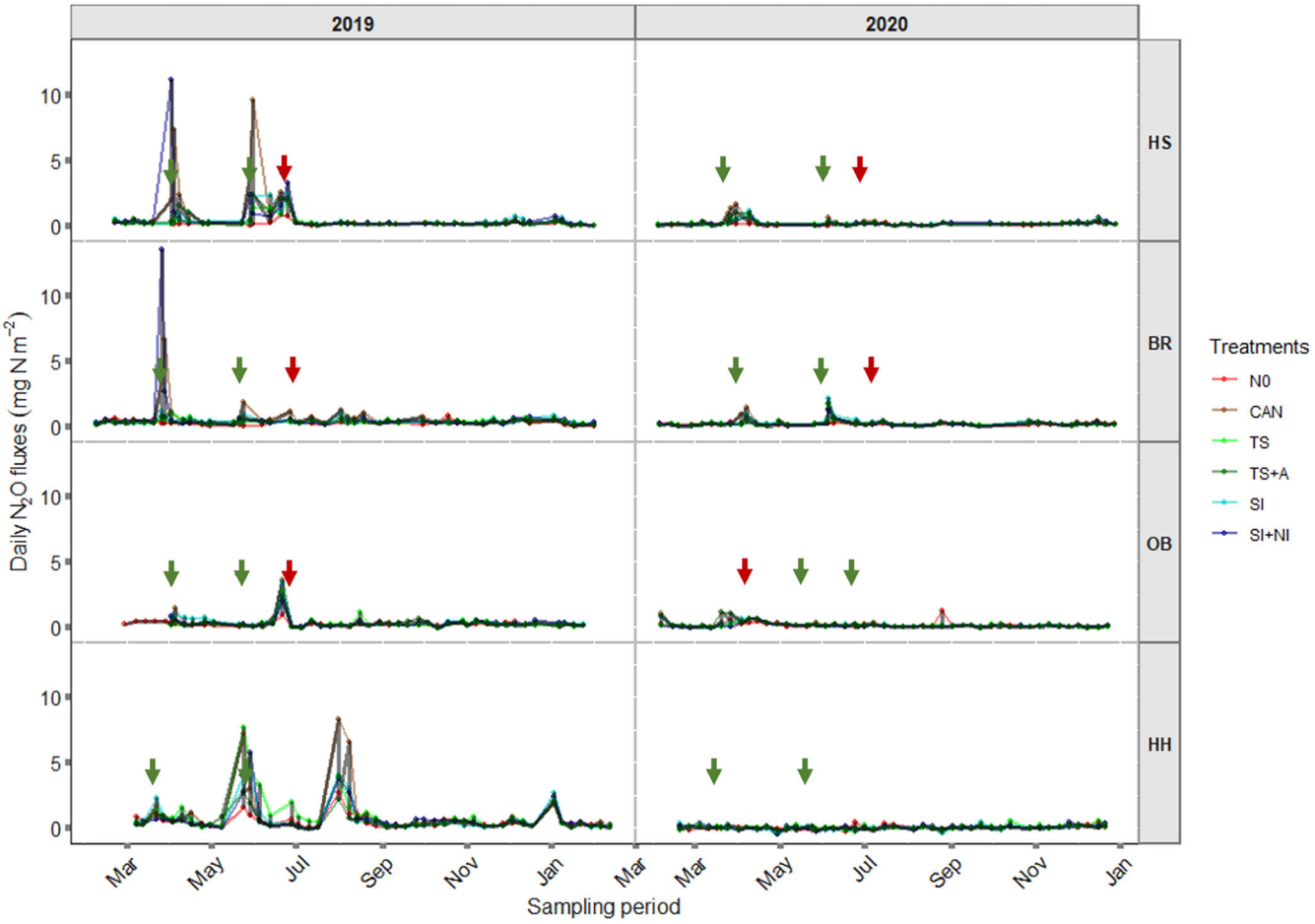

FIGURE 4 Daily N_2O-N fluxes at each experimental site and year as affected by treatment effects at Holtsee (HS), Bredenbek (BR), Osnabrück (OB) and Hohenheim (HH). Arrows show fertilization events. Arrows point to N fertilization dates; green and red arrows indicate slurry and calcium ammonium nitrate fertilizer applications, respectively.

across sites showed no significant differences between SI + NI and SI, TS + A and TS or SI and TS treatments (p > .05) in terms of N_2O response ratio (Ln N_2O–RR; Figure S1 in Data S1) in both experimental years. Although the Ln N_2O-RR for all treatment means were positive and significantly different from zero (*t*-test, $p < .05$) under higher emissions in 2019, the slurry application/treatment techniques showed a lower tendency to emit N_2O compared with the CAN treatment (Figure S1 in Data S1). In 2020, however, the Ln N_2O-RR of the fertilizer treatments was not different from the control.

Nitrous oxide EF (N_2O-N EF) ranged from −0.04% to 0.97% (Table 2) and was twice as high for CAN compared with slurry treatments (0.35% vs. 0.16%) and about six times higher in 2019 than in 2020 (0.35% vs. 0.06%). Significant site differences were only evident in 2019 for CAN treatment where EF was five times higher at Hohenheim than at Osnabrück. Like the cumulative N_2O emission trends, there was no significant differences ($p > .05$) between the slurry treatments within each site; however, using pooled data across sites and years showed that NI-treated slurry had slightly lower N_2O EF (0.17% vs. 0.19%) than the untreated ($p < .05$).

3.4 | Dry matter yield and MFRV

Dry matter yield, N content and N yield ranged from 3.1 to 10.8 Mg ha^{-1}, 1.6% to 3.3% and 59.6 to 315.0 kg N ha^{-1}, respectively (Table 3). Generally, N application increased DM yield, N yield and N concentration in harvested biomass, but treatment effects were variable, depending on the site or experimental year. For instance, N application generally increased ($p < .05$) biomass N concentration, except in HH (Table 3). Dry matter and N yields were highest, intermediate and lowest for CAN, slurry and N0 treatment, respectively; however, dry matter yields were comparable between TS and TS + A, SI + NI and SI or TS and SI ($p > .05$). The year effect on dry matter yield was evident but not consistent across sites and treatments (Table 3). On the other hand, the yield parameters were significantly ($p < .05$) affected by site and treatment, with significant interactions (Table S5 in Data S1).

The observed NUEs ranged from 46% to 85% for CAN and 10–54% for the slurry treatments while MFRV ranged from 14% to 103%, depending on the site, year of the experiment and treatments. Treatment averages only differed between TS and SI + NI (22% higher for SI + NI, $p < .05$). Whereas MFRV for SI + NI and SI treatments were comparable

TABLE 2 Cumulative N_2O emission, yield-scaled N_2O emission and N_2O emission factors as affected by site, year and treatment differences.

Location	Year	N0	CAN	TS	TS + A	SI	SI + NI
Cumulative N_2O emission (kg N ha^{-1} yr^{-1})							
HS	2019	$0.50 \pm 0.03^{\#Ca}$	$2.44 \pm 0.32^{\#Ac}$	$0.90 \pm 0.07^{\#b}$	$1.28 \pm 0.13^{\#b}$	1.36 ± 0.38^{abc}	1.25 ± 0.25^{ABbc}
	2020	$0.26 \pm 0.03^{\$a}$	$0.56 \pm 0.12^{\$ABab}$	$0.47 \pm 0.06^{\$ab}$	$0.49 \pm 0.12^{\$ABab}$	0.42 ± 0.04^{ABb}	0.49 ± 0.15^{ABab}
BR	2019	0.90 ± 0.10^{ABab}	$2.02 \pm 0.09^{\#Ab}$	$1.24 \pm 0.16^{\#a}$	1.22 ± 0.15^{a}	$1.39 \pm 0.14^{\#a}$	$1.58 \pm 0.10^{\#Aab}$
	2020	0.42 ± 0.11	$0.71 \pm 0.12^{\$A}$	$0.58 \pm 0.08^{\$}$	0.67 ± 0.13^{A}	$0.75 \pm 0.08^{\$A}$	$0.54 \pm 0.05^{\$A}$
OB	2019	0.69 ± 0.03^{BCa}	0.99 ± 0.08^{Bb}	$0.77 \pm 0.10^{\#ab}$	0.75 ± 0.15^{ab}	$1.09 \pm 0.13^{\#b}$	$0.86 \pm 0.01^{\#Bb}$
	2020	0.43 ± 0.11	0.48 ± 0.18^{AB}	$0.36 \pm 0.04^{\$}$	0.57 ± 0.20^{AB}	$0.50 \pm 0.11^{\$AB}$	$0.31 \pm 0.07^{\$AB}$
HH	2019	$1.44 \pm 0.19^{\#Aa}$	$2.83 \pm 0.35^{\#Ab}$	$2.89 \pm 0.63^{\#ab}$	$1.47 \pm 0.34^{\#ab}$	$2.11 \pm 0.58^{\#ab}$	$2.13 \pm 0.42^{\#ABab}$
	2020	$0.17 \pm 0.03^{\$}$	$0.17 \pm 0.07^{\$B}$	$0.26 \pm 0.07^{\$}$	$0.14 \pm 0.06^{\$B}$	$0.12 \pm 0.09^{\$B}$	$0.18 \pm 0.06^{\$B}$
Yield scaled N_2O emission (g N kg N^{-1} produced)							
HS	2019	6.00 ± 0.18^{ABa}	$11.29 \pm 1.44^{\#Aab}$	6.03 ± 0.66^{ABab}	7.58 ± 0.26^{Bbc}	8.25 ± 2.10^{abc}	6.66 ± 1.13^{abc}
	2020	4.53 ± 0.57^{A}	$2.76 \pm 0.47^{\$}$	4.51 ± 0.79^{A}	4.03 ± 0.68	4.01 ± 0.84^{A}	4.68 ± 1.73
BR	2019	5.44 ± 0.60^{ABab}	$7.34 \pm 0.33^{\#Ab}$	4.98 ± 0.69^{Aab}	$5.20 \pm 0.52^{\#Aab}$	4.90 ± 0.37^{a}	$5.72 \pm 0.33^{\#ab}$
	2020	3.11 ± 0.85^{AB}	$2.32 \pm 0.43^{\$}$	2.89 ± 0.48^{AB}	$2.80 \pm 0.31^{\$}$	3.46 ± 0.29^{AB}	$2.440.24^{\$}$
OB	2019	3.92 ± 0.27^{Bab}	3.13 ± 0.23^{Ba}	3.59 ± 0.39^{Aab}	4.36 ± 1.50^{ABabc}	5.04 ± 0.17^{c}	3.99 ± 0.38^{b}
	2020	5.38 ± 1.44^{AB}	2.61 ± 1.07	2.60 ± 0.43^{AB}	3.91 ± 1.48	3.43 ± 1.02^{AB}	2.33 ± 0.71
HH	2019	$8.07 \pm 1.11^{\#Aab}$	$15.70 \pm 2.52^{\#Ac}$	$15.99 \pm 2.58^{\#Bbc}$	$7.61 \pm 1.33^{\#ABa}$	13.83 ± 4.14^{abc}	$12.11 \pm 2.83^{\#abc}$
	2020	$1.04 \pm 0.28^{\$B}$	$0.91 \pm 0.41^{\$}$	$1.30 \pm 0.36^{\$B}$	$0.64 \pm 0.24^{\$}$	0.83 ± 0.60^{B}	$0.86 \pm 0.29^{\$}$
N_2O-N emission factor (% of N applied)							
HS	2019		$0.97 \pm 0.17^{\#Ab}$	0.18 ± 0.03^{a}	0.36 ± 0.07^{a}	0.40 ± 0.19^{ab}	0.35 ± 0.12^{ab}
	2020		$0.15 \pm 0.05^{\$}$	0.10 ± 0.04	0.11 ± 0.06	0.08 ± 0.02	0.11 ± 0.07
BR	2019		0.56 ± 0.09^{ABb}	0.16 ± 0.04^{a}	0.15 ± 0.04^{a}	0.23 ± 0.03^{a}	0.31 ± 0.06^{ab}
	2020		0.14 ± 0.07	0.07 ± 0.07	0.12 ± 0.10	0.15 ± 0.06	0.05 ± 0.07
OB	2019		0.15 ± 0.03^{B}	0.04 ± 0.05	0.04 ± 0.08	0.21 ± 0.09	0.09 ± 0.02
	2020		0.02 ± 0.08	-0.02 ± 0.04	0.05 ± 0.06	0.03 ± 0.04	-0.04 ± 0.03
HH	2019		$0.82 \pm 0.14^{\#Ab}$	0.97 ± 0.52^{ab}	0.02 ± 0.12^{a}	0.45 ± 0.28^{ab}	0.47 ± 0.30^{ab}
	2020		$0.00 \pm 0.04^{\$}$	0.06 ± 0.05	-0.02 ± 0.04	-0.03 ± 0.07	0.00 ± 0.03

Note: Means (±standard error) with different letters/symbols are different at $p < .05$; $, # being year effect within the same treatment and site, A, B, C being site effect within a treatment and year, and a, b, c being treatment differences within the same year and site.

($p < .05$) at all sites, a lower MFRV ($p < .05$) was observed for TS + A compared with TS but only at BR in 2019 (Figure 5). Also, the year effect was only evident in one instance, where the average MFRV for SI + NI was higher ($p < .05$) in 2019 compared to 2020 at BR.

3.5 | Relationships between variables

Positive correlations were observed between N_2O variables on the one hand and environmental variables on the other hand (Table 4), with correlations between N_2O-N EF and the environmental factors depending on the N application treatment (Tables S5 and S6 in Data S1). Further analysis shows that the variations across sites in cumulative N_2O emission were strongly dependent on the soil's clay content, soil pH, total N applied, mean daily temperature and annual precipitation (Table S7 in Data S1); and these variables could predict the cumulative N_2O-N emission (logN_2O-N) with appreciably high efficiency (adjusted $R^2 = 90\%$, Table S8 in Data S1). Accordingly, a comparison between the modelled and observed values showed a high correlation (Figure 6). Again, higher MFRV correlated with higher annual precipitation and soil NO_3^--N content. Whereas the relationships between MFRV and soil factors or perennial ryegrass proportion in the swards were weak (Table 4), MFRV correlated positively with N uptake in aboveground biomass ($r = 0.5$, $p < .05$) and biomass N concentration ($r = 0.8$, $p < .001$).

4 | DISCUSSION

4.1 | Site and year effects on N_2O emission and emission factors

This study applied N fertilizer from CAN or cattle slurry, treated with either acid or NI, at an average rate of 201 kg N ha^{-1} to

TABLE 3 Total harvested biomass, N content of harvested biomass and N yield as affected by N source, slurry application methods, year of experimentation and experimental site.

Site	Year	N0	CAN	TS	TS + A	SI	SI + NI
Dry matter yield (*t* DM ha^{-1} yr^{-1})							
HS	2019	4.20 ± 0.25^{Ba}	7.48 ± 0.32^{Ab}	6.46 ± 0.40^{Ab}	6.51 ± 0.47^{b}	6.46 ± 0.36^{b}	7.04 ± 0.40^{b}
	2020	3.10 ± 0.41^{Ba}	7.77 ± 0.67^{BCc}	5.03 ± 0.60^{Cb}	5.54 ± 0.73^{Cb}	5.28 ± 0.46^{Bb}	5.00 ± 0.51^{Bb}
BR	2019	6.56 ± 0.09^{Aa}	8.42 ± 0.31$^{\#ABb}$	8.25 ± 0.22^{ABb}	7.92 ± 0.31^{b}	8.84 ± 0.43^{b}	8.44 ± 0.34^{b}
	2020	6.25 ± 0.38^{Aa}	10.83 ± 0.06$^{\$Ac}$	8.29 ± 0.39^{ABb}	9.28 ± 0.53^{ABbc}	8.81 ± 0.35^{Ab}	8.81 ± 0.29^{Ab}
OB	2019	8.53 ± 0.33$^{\#Aa}$	10.75 ± 0.37$^{\#Bb}$	9.75 ± 0.72$^{\#Bab}$	9.14 ± 1.14^{ab}	8.99 ± 0.88^{ab}	9.39 ± 0.79^{ab}
	2020	4.99 ± 0.48$^{\$ABa}$	7.60 ± 0.38$^{\$Cb}$	6.47 ± 0.29$^{\$BCa}$	6.90 ± 0.42^{BCa}	7.06 ± 0.65^{ABab}	6.61 ± 0.55^{Ba}
HH	2019	8.26 ± 0.63^{A}	8.74 ± 0.91^{AB}	8.00 ± 0.50^{AB}	9.05 ± 0.70	8.16 ± 0.30	8.94 ± 0.46
	2020	8.79 ± 0.52^{C}	9.78 ± 0.41^{AB}	9.42 ± 0.31^{A}	10.06 ± 0.29^{A}	9.55 ± 1.15^{AB}	10.31 ± 0.70^{A}
N content of harvested biomass (%, DM basis)							
HS	2019	1.99 ± 0.06^{Ba}	2.89 ± 0.05$^{\#Bd}$	2.34 ± 0.07^{Bb}	2.60 ± 0.11$^{\#ABbcd}$	2.53 ± 0.09$^{\#Bbc}$	2.66 ± 0.08^{Bcd}
	2020	1.93 ± 0.02^{Aa}	2.63 ± 0.01$^{\$Ac}$	2.18 ± 0.07^{ABb}	2.17 ± 0.04$^{\$Bb}$	2.12 ± 0.04$^{\$Bb}$	2.23 ± 0.11^{ABab}
BR	2019	2.53 ± 0.06$^{\#Aa}$	3.28 ± 0.04$^{\#Ad}$	3.05 ± 0.08$^{\#Abc}$	2.96 ± 0.09$^{\#Ab}$	3.21 ± 0.04$^{\#Acd}$	3.28 ± 0.10$^{\#Acd}$
	2020	2.19 ± 0.08$^{\$Aa}$	2.83 ± 0.07$^{\$Ac}$	2.49 ± 0.06$^{\$Ab}$	2.48 ± 0.07$^{\$Ab}$	2.46 ± 0.05$^{\$Ab}$	2.52 ± 0.05$^{\$Ab}$
OB	2019	2.07 ± 0.10$^{\#Ba}$	2.94 ± 0.09$^{\#ABb}$	2.19 ± 0.05^{Ba}	2.13 ± 0.05^{BCa}	2.40 ± 0.14^{Ba}	2.37 ± 0.06^{BCa}
	2020	1.60 ± 0.05$^{\$Ba}$	2.50 ± 0.02$^{\$Ac}$	2.21 ± 0.04^{Bb}	2.19 ± 0.03^{Bb}	2.17 ± 0.03^{Bb}	2.13 ± 0.04^{Bb}
HH	2019	2.18 ± 0.10^{AB}	2.13 ± 0.05^{C}	2.21 ± 0.11^{B}	2.08 ± 0.05^{C}	1.97 ± 0.12^{B}	2.04 ± 0.14^{C}
	2020	2.05 ± 0.18^{ABab}	1.92 ± 0.05^{Bab}	2.18 ± 0.15^{ABb}	2.16 ± 0.16^{ABab}	1.80 ± 0.03^{Ca}	2.01 ± 0.15^{ABab}
N yield (kg N ha^{-1} yr^{-1})							
HS	2019	83.58 ± 5.54^{Ba}	215.45 ± 5.54^{Cc}	150.04 ± 5.47$^{\#Bb}$	169.52 ± 16.52^{bc}	163.78 ± 12.91^{Bb}	187.77 ± 14.78$^{\#Bbc}$
	2020	59.59 ± 7.38^{Ca}	204.85 ± 18.04^{Bc}	109.23 ± 12.17$^{\$Bb}$	119.62 ± 14.13^{Cb}	112.19 ± 11.60^{Bb}	110.28 ± 8.27$^{\$Bb}$
BR	2019	165.67 ± 4.68^{Aa}	276.39 ± 12.19^{ABc}	251.53 ± 10.62^{Abc}	234.18 ± 12.72^{b}	283.45 ± 11.36$^{\#Abc}$	276.23 ± 10.94$^{\#Ac}$
	2020	135.99 ± 5.56^{ABa}	306.56 ± 9.61^{Ac}	206.87 ± 12.61^{Ab}	231.39 ± 19.45^{ABb}	216.11 ± 7.09$^{\$Ab}$	221.51 ± 5.85$^{\$Ab}$
OB	2019	175.81 ± 8.65$^{\#Aa}$	314.98 ± 1.69$^{\#Bb}$	213.24 ± 13.94$^{\#ACa}$	195.10 ± 24.28^{a}	218.32 ± 32.30^{ABa}	223.72 ± 24.46^{ABa}
	2020	80.10 ± 8.36$^{\$BCa}$	189.39 ± 8.69$^{\$Bc}$	142.79 ± 7.85$^{\$Bb}$	151.42 ± 10.75^{BCb}	153.15 ± 12.86^{Bb}	140.74 ± 12.67^{BCb}
HH	2019	181.01 ± 20.00^{A}	186.58 ± 20.87^{AC}	176.50 ± 10.83^{BC}	187.29 ± 12.08	160.67 ± 11.55^{B}	180.75 ± 7.78^{B}
	2020	179.27 ± 15.79^{A}	188.11 ± 11.39^{B}	205.73 ± 16.16^{A}	217.44 ± 16.69^{A}	171.44 ± 20.02^{AB}	205.77 ± 15.59^{AC}

Note: Means (±standard error) with different letters/symbols are different at $p < .05$; $, # being year effect within the same treatment and site, A, B, C being site effect within a treatment and year, and a, b, c being treatment differences within the same year and site.

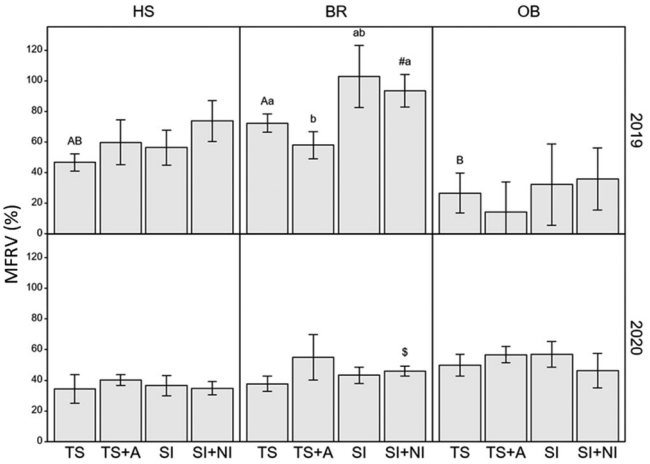

FIGURE 5 Mineral fertilizer replacement value (MFRV) of applied cattle slurry total nitrogen at Holtsee (HS), Bredenbek (BR), and Osnabrück (OB) affected by application/treatment technique and site differences. Data from HH is not included in this analyses because too many extreme values. Means (±s.e. as error bars) with different letters are different at $p < .05$, year j, k, treatment a, b, c and site A, B, C; MFRV was calculated as the ratio of slurry N use efficiency to mineral fertilizer (Calcium ammonium nitrate) N use efficiency.

grasslands. Cumulative N_2O-N emissions depended on the year, the study site and the technique of N application. The cumulative N_2O-N emissions of the investigated sites (for both years and all treatments) are in the lower range of published values for temperate grassland (0.04 and 21.2 kg N_2O-N ha^{-1} $year^{-1}$) and comparable to other German grasslands (Jungkunst et al., 2006; Dechow & Freibauer, 2011; Rees et al., 2013; Mathivanan et al., 2021). The range of cumulative N_2O emissions observed for the N0 treatment in this study (0.12–1.44 kg ha^{-1}) is within the ranges reported for grasslands in the UK (0.25–0.78 kg ha^{-1}; Thorman et al., 2020) and in Canada (0.17–0.30 kg ha^{-1}; Hunt et al., 2019). The overall average annual N_2O EF for CAN (0.35% ± 0.07%) and cattle slurry (0.16% ± 0.03%) was lower than the Tier 1 IPCC default of 1.6% (1.3%–1.9%) for synthetic N fertilizers and within the lower range of 0.1%–1.1% for the slurry in a wet temperate climate (IPCC, 2019). The low N_2O-N EFs observed in this study are not uncommon, as previous studies on temperate grasslands have reported low N_2O EFs (<0.5%) for organic N inputs (Reinsch et al., 2020; Voglmeier et al., 2019). For example, Irish grasslands receiving 160–640 kg N ha^{-1} $year^{-1}$ cattle slurry emitted only −0.20% to 0.43% of the applied N (O'Neill et al., 2020).

According to Holtan-Hartwig et al. (2002), some soils generally have the intrinsic potential for low N_2O emission rather, irrespective of environmental conditions, probably due to the soil microbial community structure (Cavigelli & Robertson, 2001). This trend might also be due to the high productivity of such grasslands. At high production levels, the N uptake from grassland is very high, leaving only small amounts of N for denitrification by soil microbes. Furthermore, the low or sometimes negative N_2O EFs could be attributed to higher N_2O emissions from the control treatment (O'Neill et al., 2020), as a higher mineralization rate of soil organic matter can lead to increased inorganic N-concentration in soil solution, potentially leading to higher gaseous losses N_2O emissions (Liu et al., 2018). Nevertheless, it is worth mentioning that static chamber methods lack temporal resolution and might fail to capture large but brief N_2O peaks, leading to the underestimation of cumulative N_2O emission and N_2O-N EFs (O'Neill et al., 2020).

4.2 | Driving factors of N_2O emission

Our data showed a positive correlation between yearly N_2O emissions, EFs and yield-scaled emissions on the one hand and annual precipitation and with WFPS on the other hand (Table 4). A strong influence of precipitation and WFPS on N_2O emission have been reported in arable fields (Bell et al., 2015) and grasslands (Cardenas et al., 2016; Li et al., 2017). The relative importance of nitrification and denitrification largely depends on WFPS, with low WFPS (low soil water content and coarse soil texture) favouring aeration and, therefore, nitrification, and high WFPS (high soil water content and fine soil texture) promoting denitrification (Davidson, 1991). Thus, the low WFPS and low N_2O emissions observed at Osnabruck in 2019 might be due to the poor water-holding capacity of sandy soils. Under wetter and warmer soil conditions, anaerobic conditions are created due to enhanced microbial and root oxygen consumption and reduced gas diffusion (Schlüter et al., 2018; van der Weerden et al., 2014). In the presence of adequate N substrate, this situation might lead to increased denitrification and a consequent increase in N_2O fluxes. Both precipitation and clay content of soil (which influence the water-holding capacity) influence WFPS directly. Although not always the case, fine-textured soils generally facilitate the development of anaerobic conditions and long-lasting local anoxia than coarser-textured soils as clay particles hold water tightly in aggregates, favouring N_2O production and emission through denitrification (Gu et al., 2013).

However, N_2O emissions could be lower under high soil moisture conditions if complete denitrification is promoted, especially when labile-C is abundant (e.g. from added manures). On the other hand, low rainfall conditions decrease plant N uptake, reduce N_2O emission and increase soil N pool. As observed in this study, there were relatively higher NO_3^- peaks in 2020 (Figure S4 in Data S1), suggesting limited denitrification and hence the low N_2O emission in 2020 (Figure 2). It is worth mentioning that N_2O rewetting peaks occur under low moisture conditions, who's intensity is highly dependent on the intensity of the previous drying episode (Barrat et al., 2021). However, these N_2O hot moments due to rewetting could be more relevant under semi-arid conditions and, therefore, not applicable to our study.

Although precipitation in both years was lower than the long-term averages, the shortfalls were higher in 2020 (38% to 47%) than in 2019 (13% to 24%). Accordingly, the low rainfall observed for both experimental years might partly explain the generally low N_2O-N EFs and cumulative N_2O emissions in 2020 relative to 2019. Nevertheless,

TABLE 4 Pearson correlation coefficients showing relationships between nitrous oxide emission, nitrogen uptake and nitrogen fertilizer replacement value and environmental variables using data across sites, years and treatments.

Variables	N_2O-N	Yield scaled N_2O-N	N_2O-N EF	N uptake	NFRV[b]
Degrees of freedom	**46**	**46**	**38**	**46**	**22**
Total N applied	0.08	−0.07	−0.27	−0.11	0.29
Clay content	0.54*	0.53*	0.43*	0.20	0.08
Soil bulk density	0.14	0.12	0.23	−0.07	0.11
Soil cation exchange capacity	0.25	0.30	0.18	0.01	−0.40
Soil pH	0.19	0.32	0.28	−0.25	0.21
Soil C_{org} content	0.44*	0.47*	0.46*	0.11	0.13
Soil N content	0.37	0.37*	0.33*	0.16	0.08
Soil NH_4-N content	0.18	0.08	0.14	0.26	0.23
Soil NO_3-N content	0.45*	0.38*	0.52*	0.18	0.60*
Annual precipitation	0.64*	0.55*	0.55*	0.22	0.54*
Water filled pore space	0.41*	0.29	0.40	0.35*	0.32
Mean soil temperature[a]	0.08	0.13	0.22	−0.07	−0.02
Perennial ryegrass proportion in swards	−0.02	−0.06	0.07	0.03	0.07

Abbreviations: NFRV, Nitrogen fertilizer replacement value; N_2O-N EF, Nitrous oxide emission factor.
*Coefficient significant at $p < .01$.[a]Mean soil temperature was measured at 15 cm soil depth.
[b]NFRV did not include data from HH because the values were extreme.

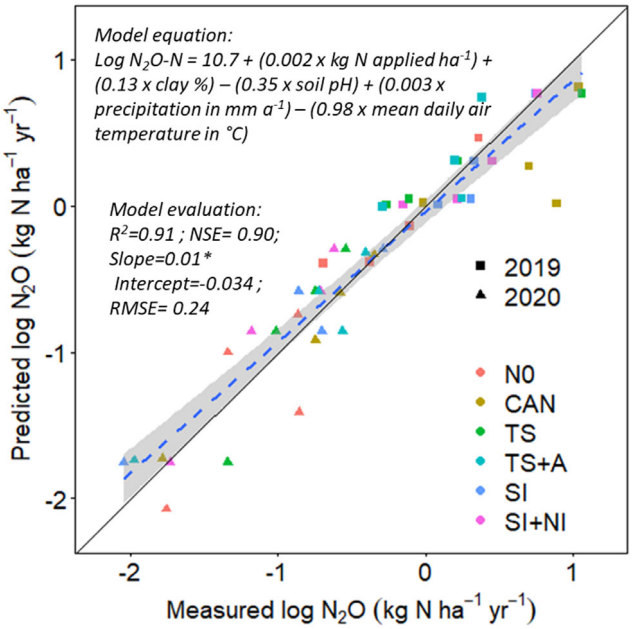

FIGURE 6 Measured versus annual nitrous oxide emissions (kg N_2O-N ha^{-1}) at grasslands sites. Each symbol represents the average cumulative N_2O emissions of each treatment (N0, CAN, TS, TS + A, SI, SI + NI) either in 2019 or 2020. NSE, Nash–Sutcliffe model efficiency coefficient; RMSE, Root mean square error; * $p < .001$.

the unusually low precipitation in 2018, where there was half as much precipitation compared with the last few years' annual averages, led to drought and low yields across the country (Webber et al., 2020). This lowers the plants' capacity to use the total applied fertilizer N. However, the average N_2O EF observed across sites in 2019 (0.62%) is within the range of 0.42% to 0.84% calculated for Germany using Tier 2 methodology (Mathivanan et al., 2021) and similar to the IPCC (2019) default. Several factors might interact to influence N_2O emission from soils (Bourdin et al., 2014; Fan et al., 2018) and the effects of management, climatic and soil factors on N_2O emissions were observed in the current study (Table 4).

The relatively high mean N_2O-N EF observed for Baden-Wuerttemberg (0.27% ± 0.08%) relative to Schleswig-Holstein (0.24% ± 0.03%) and Lower Saxony (0.06%± 0.02%) agree with earlier reports (Mathivanan et al., 2021) that emission factors in southern Germany are generally higher than in the north. This was attributed to the effects of different climatic conditions and soil properties on N_2O formation (Jungkunst et al., 2006; Dechow & Freibauer, 2011). The sites in Baden-Wuerttemberg received less precipitation and fertilizer N, but the soils were characterized by relatively high organic C, N and clay content (Table 1). As corroborated by the positive correlations between N_2O EF, on the one hand, and soil organic C and total N, on the other hand, and between NO_3^- content and soil organic C (Table 4), the soils might have encouraged decomposition of organic matter to provide labile organic C and inorganic N substrates for nitrification and denitrification (Gu et al., 2009). Other studies also observed these relationships, where high organic C content resulted in higher N_2O fluxes (Harrison-Kirk et al., 2013). Also, the relatively high clay content in the soils might have favoured denitrification.

4.3 | Effects of N application/slurry treatment on N_2O emissions

Fertilizer N application generally increased N_2O emission in this study (Figure S1 in Data S1) due to the increased availability of N substrate

required for N_2O production (LaHue et al., 2016). However, the N application effect on N_2O emission varied across sites and years. Similarly, Thies et al. (2019) observed no fertilizer N effect on N_2O emission in spring, although the N_2O emission level was high. This situation might be due to increased organic C and N pools in soils from long-term N fertilization (Drury et al., 1998). The nitrification of applied NH_4^+ and the subsequent denitrification depends on the mineralization dynamics of soil N, moisture and temperature (Cookson et al., 2000). The grassland sites in this study have been fertilized at moderate to high rates during the last years and therefore had relatively high residual N levels, as shown by the comparable mineralized N levels and N_2O emissions in the N0 treatment relative to the slurry treatments in the many instances (Table S4 in Data S1).

We hypothesized that slurry acidification as a measure to mitigate NH_3 emissions does not increase N_2O emissions as more N becomes available for the crop to utilize (Nyameasem et al., 2021). Accordingly, we did not observe any significant effect of slurry acidification on N_2O emission or EF. Similar to our study, Malique et al. (2021) reported an insignificant slurry acidification effect on N_2O emission. However, the effects of manure acidification on N_2O emissions have been assessed differently in the literature. For example, while Fanguero et al. (2010) observed increased N_2O emissions from sandy soils when laboratory incubations were performed using acidified pig slurries, Park et al. (2018) reported reduced (79.9%) N_2O emission. Acid type, target pH and N application rate have been reported to influence N_2O emission from acidified slurry (Petersen et al., 2012). Low pH resulting from slurry acidification might encourage the proliferation of pH-tolerant non-nitrifying species, including fungi, may promote N immobilization (de Vries, 2009) to reduce N_2O emission. However, this was not evident in our study, as the N values in the acidified treatment were more or less similar to those without acid. On the other hand, soil acidity level influences the N_2O:N_2 product ratio of denitrification, with lower soil pH conditions (e.g., pH = 6.1 vs. 8.0; Liu et al., 2010) producing more N_2O and increasing the ratio of N_2O to N_2 (Saggar et al., 2013). It is worth mentioning that both experimental years were relatively dry for a considerable denitrification N_2O production to occur, which might partly explain the insignificant slurry acidification effect on N_2O emission. The current study's slurry pH (target of 6) was probably not potent enough to modify the soil microbial dynamics. In any case, the consistent results from our study suggest that slurry acidification, a valuable technique to mitigate NH_3 loss (Nyameasem et al., 2022), has no adverse impact on direct N_2O emission at the N rates and conditions observed in this study but might reduce indirect N_2O emission from slurry.

Our study showed a relatively similar N_2O response ratio for injected slurry (open slot, injection depth of 5 cm) and surface application with a trailing shoe (Figure S1 in Data S1). However, slight increases in cumulative N_2O emissions, N_2O EF and mineral N values were observed in many instances (Table 2). In contrast, a recent study (Maris et al., 2021) observed an increased (32%) N_2O emission from injected slurry (5/10-cm injection depth) compared with the broadcast application. Also, on grassland and maize land, shallow injection (open slot, injection depth of 5 cm) of slurry increased the mean N_2O-N EF relative to a surface application (Velthof & Mosquera, 2011). The increased N_2O emissions from the injected slurry are attributable to the anaerobic environment created by the slurry concentrated in the injection furrow and the possible occlusion of soil pores due to the smearing effect of the injector tines (Maris et al., 2021). This condition might also lower N uptake by plants with injection than with the trailing shoe application technique resulting in increased N_2O emissions (Fangueiro et al., 2015; Herr et al., 2019; Maris et al., 2021). In their review, Chadwick et al. (2011) observed that fertilizer application methods that retain more N in the soil may not necessarily lead to reduced N_2O emission as environmental and soil factors can be more important in controlling N_2O emission. Deep placement of slurry (15–20 cm depth) is not a common practice on grasslands, probably because of the risk of placing nutrients beyond the reach of plant roots. Accordingly, studies comparing depth effects on slurry emissions from grasslands are scarce, but the diffusion path length could impact N_2O emission with longer diffusion paths leading to a greater chance of N_2O being reduced to N_2 (Velthof & Moquera, 2011).

Our results show that slurry injection (relative to the trailing shoe technique) as an NH_3 emission mitigation strategy might increase N_2O emissions. Although the pooled data (across sites and years) showed that adding NI reduced cumulative N_2O emission, the reductions were insignificant at most sites. Also, the expected higher NH_4^+ and lower NO_3^- in the NI-treated soils relative to the untreated slurry was not observed. The N_2O flux rates were positively correlated with the NO_3-N contents, precipitation and water-filled porosity, which hints at denitrification as the primary source for N_2O release at our study sites. The dry conditions in the periods of slurry and DMPP applications, especially in the second experimental year, might have been a major reason for establishing aerobic conditions, thus suppressing enzyme activity in the denitrification pathway. The resulting unusually low N_2O emissions, even in the treatment with slurry injection, thus minimized the NI's potential to reduce N2O production further. Although on a much higher emission level, Herr et al. (2020) also reported lower N_2O emission reduction from injected NI-treated slurry in their drier experimental year.

The mechanisms and factors determining the efficacy of NIs in reducing N_2O emissions are not well understood (Ruser & Schulz, 2015). In a microcosm experiment, DMPP addition to N fertilizer significantly reduced N_2O emissions from sandy soil but did not affect N_2O emissions from loamy soils (Friedl et al., 2020). Our soils were primarily loamy, with relatively high organic C contents, and such soils might have more pronounced sorption of DMPP (Marsden et al., 2016), reducing the efficacy of DMPP in inhibiting autotrophic nitrification. Lowest efficacies of all NIs included in their meta-analysis for soil groups with a fine texture and high carbon contents were also reported by Fan et al. (2022) and by Soares et al. (2023). Moreover, temperature and soil moisture effects on NI efficacy have also been observed (Kelliher et al., 2008), with extreme soil conditions (cool and wet or hot and dry conditions) favouring DMPP efficacy (Menéndez et al., 2012). In this study, slurry was primarily applied in early to late spring. However, it is unclear whether temperature-moisture relations were influential on DMPP activity. To increase DMPP's efficacy on

N_2O emission in soils with high organic matter content and an intensive soil N turnover, Friedl et al. (2020) suggested repeating applications, increasing the application rate and/or applying before fertilization to limit the effect of N fertilizer priming on N_2O emissions; however, this might not be feasible with slurry injection. Invariably, this study shows that NI addition to injected slurry has the potency to reduce N_2O emission relative to CAN fertilizer.

4.4 | Forage N concentration and yield

In most grassland systems, the amount of plant-available N in the soil is insufficient for optimal grass yield. Thus, N fertilization is required for optimal forage yield and protein content. It is well-known that grass growth and biomass yield are associated with plant-available N (Kai-yun et al., 2015). Pooled data across the sites and years showed that N application generally increased biomass N concentrations, dry matter and N yields. Higher N yields were observed in 2019 compared with 2020, probably due to the weather in 2018, as explained earlier, where there was half as much precipitation as the last few years' annual averages, leading to drought and low yields across Germany. As a result, plants might not have taken up the applied fertilizer N, thus increasing plant-available N in spring 2019.

However, the results also show that fertilizer application did not affect biomass accumulation, particularly at Hohenheim, as shown by the insignificant effect of the applied N on DM and N yields at this site (Table 3). N uptake in the control plots was 29–108 kg/ha/year higher at Hohenheim than at the other sites. Depending on soil, climatic and land management factors, successive fertilizer application increases soil N content. Under high temperature and moisture conditions, elevated microbial activity and soil biogeochemical process rates may increase inorganic N concentration in soil solution with a positive residual effect for subsequent pasture production (Giraud et al., 2021). Furthermore, soils with relatively high total oxidized N and lower C/N ratio are associated with higher N uptake and DM yield response from SOM mineralization (McDonalds et al., 2014). Thus, depending on the slurry C/N ratio or the amount of organic matter in the soil, applied N may not be entirely recovered in the plant, as plants may take up mineralized N after release from existing organic compounds (Chen et al., 2019; Zistl-Schlingmann et al., 2020).

The soils at Hohenheim had a relatively low C/N ratio and high N content and experienced relatively high temperatures (Table 1), which might have favoured organic matter decomposition. We did not determine the residual N supply potential of the soils, but nitrogen mineralization from organic matter is an essential resource for agricultural productivity. For example, without fertilization, nitrogen uptake by grasses ranged from 40 to 212 kg N/ha/year in temperate grasslands (Hopkins et al., 1990; Humphreys et al., 2007). Under high fertilization, Murphy et al. (2013) reported about 49% N uptake by grass from mineralized organic matter. Similar to our study, Zistl-Schlingmann et al. (2020) reported low N recovery of cattle slurry applied to grassland in southern Germany and suggested N immobilization. Also, Fu et al. (2017) reported plant N uptake often exceeding slurry application rates in montane soils of southern Germany, suggesting a further supply of N from SOM decomposition.

Generally, DM and N yields were higher for CAN than for the slurry treatment in many instances, probably due to the lower fertilizer N uptake efficiency of slurry N compared to mineral fertilizer N, as the organically bound N fraction, which makes up about 45% of total N, limits N availability (Nannen et al., 2011). On the other hand, the slurry application techniques showed similar effects on DM yield, N yields and biomass N concentration, suggesting that the strategies were equally efficient at delivering N to the plants. As all sites of our study were fertilized with organic fertilizers for many years, high mineralization rates might mask the possible effects of the slurry treatments on N usage. In the case of NIs, the lack of effect regarding yield or N yield is usually observed at field conditions and could be because optimum or excessive N rates are usually applied, thus masking the effect of the inhibitor. Contrary to reports that slurry injection reduces yield due to sward damage (Nyord et al., 2010), yields in the current study were similar for the TS and SI application techniques, and there was no evidence that plants took up extra NH_4^+ due to slurry acidification or NI addition to produce higher biomass (Table 3). Apparently, a positive slurry acidification effect on plant performance is partly related to sulfur supply due to its role in plant protein formation (Hawkesford et al., 2012). However, the sulfur-based acidification effect might not be plausible in this study as soils already have adequate sulfur levels, partly explaining the non-significant effect of slurry acidification on plant performance observed in our study.

4.5 | Mineral fertilizer replacement values for cattle slurry

NUEs were lowest at Hohenheim (3%–5% for CAN, −14% to 23% for slurry treatment), and the calculated MFRVs were extremely low (data not shown), most probably due to high N mineralization from soil N pools from long-term high amounts of organic fertilizer application as explained previously. Thus, data from HH was eliminated from the final analyses and results were based only on data from BR, HS, and OB (Figure 5). The MFRVs for cattle slurry in this study are generally low and fall within the lower range of 20% to 90% reported in the literature (Delin et al., 2012; Jensen, 2013; Webb et al., 2013). The low MFRV and N_2O emissions might be due to "missing N losses" via other pathways, as losses via volatilization, NO_3 leaching, and N immobilization could be high (Nyameasem et al., 2022; Selbie et al., 2015). Also, MFRV is based on the NUE of slurry treatments and, therefore, on the N rates considered for NUE calculations. As the NUEs in this study were based on the total N content of slurry (not on available N), the MFRVs were likely underestimated. Nevertheless, the MFRVs observed for TS and SI in this study agree with the 36%–54% and 53%–77%, respectively, reported for grasslands in the Netherlands (Shröeder et al., 2007), as well as reports by other authors for European grasslands (Lalor, 2011; Pedersen et al., 2020; Sørensen et al., 2019). On the other hand, there was no consistent effect of acidification or NI addition to slurry on MFRV in our study,

although in one of the six instances, slurry acidification or NI addition surprisingly depressed MFRV.

Although our third hypothesis could not be confirmed, a relatively high MFRV was observed at Schleswig-Holstein compared with Lower Saxony (57% vs. 40%). This difference could partly be attributed to differences in botanical composition at these sites (Table S1 in Data S1), especially as the high perennial ryegrass density at Schleswig-Holstein (94% vs. 50%) might have enhanced DM yield (Creighton et al., 2012) and MFRV. As shown by this study (Table 4), a positive correlation between MFRV and N uptake in aboveground biomass ($r = 0.5$, $p = .01$) and biomass N concentration ($r = 0.8\%$, $p < .01$) was observed. This positive effect suggests that increasing the value of slurry could increase N recovery to promote clean production in forage production. Moreover, MFRV is highly dependent on the mineralization of slurry to release N for plant uptake growing degree days and soil moisture moderates this process (Pedersen et al., 2021). As shown in this study, a positive correlation was evident between MFRV and WFPS ($r = 0.8$, $p < .01$) and between MFRV and soil NO_3^--N ($r = 0.6$, $p < .01$), suggesting higher precipitation facilitated higher organic N mineralization to increase plant-available N supply, which might partly explain the apparent year effect on MFRV, especially at BR, where values were higher in 2020 (46%–57%) than in 2019 (14%–36%). Thus, we assume that the relative difference in slurry MFRV between Schleswig-Holstein and Lower Saxony could be partly attributed to the difference in soil moisture, a function of precipitation and soil type.

5 | CONCLUSION

Nitrous oxide emission is only a small economic loss pathway for N but impacts the environment considerably. To achieve the aims of the Farm to Fork Strategy by the European Union, efficient N management on grassland is required to reduce fertilizer N input while keeping greenhouse gas emissions low and maintaining yields. The current study evaluated the site-specific effects of N fertilizer application techniques on N_2O emissions from permanent grasslands. Fertilizer N application generally increased N_2O emission, but average site N_2O emissions were in the lower range of values reported for temperate grasslands. The average annual EF for CAN and cattle slurry (pooled across sites and years) was lower than the Tier 1 IPCC default for wet temperate climates. The low N_2O emissions from permanent grassland, even at high N application rates, are attributable to the dry weather conditions in these years. Further long-term research would be needed to rule out the weather impact. Again, our study shows that slurry acidification, shown in the literature as an efficient tool for NH_3 mitigation, could have no adverse effect on direct N_2O emissions. Also, this study showed that the slurry application strategies delivered N to the plants at equal efficiency, validated by a similar effect on MFRV. Overall, our results emphasize the benefit of replacing chemical fertilizer N input (i.e. CAN treatment in this case) with N from slurry in terms of lower N_2O emissions and utilization of an on-farm N source rather than relying on increasingly expensive chemical fertilizer N.

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CONFLICT OF INTEREST STATEMENT

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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REFERENCES

Akyama, H., Yan, X., & Yagi, K. (2010). Evaluation of effectiveness of enhanced-efficiency fertilizers as mitigation options for N_2O and NO emissions from agricultural soils: Meta-analysis. *Global Change Biology*, *16*, 1837–1846.

Anas, M., Liao, F., Verma, K. K., Sarwar, M. A., Mahmood, A., Chen, Z. L., Li, Q., Zeng, X. P., Liu, Y., & Li, Y. R. (2020). Fate of nitrogen in agriculture and environment: Agronomic, eco-physiological and molecular approaches to improve nitrogen use efficiency. *Biological Research*, *16*(1), 47. https://doi.org/10.1186/s40659-020-00312-4

Barrat, H. A., Evans, J., Chadwick, D. R., Clark, I. M., Cocq, K.l., & Cardenas, L. M. (2021). The impact of drought and rewetting on N_2O emissions from soil in temperate and Mediterranean climates. *European Journal of Soil Science*, *72*, 2504–2516. https://doi.org/10.1111/ejss.13015

Bell, M. J., Rees, R. M., Cloy, J. M., Topp, C. F. E., Bagnall, A., & Chadwick, D. R. (2015). Nitrous oxide emissions from cattle excreta

applied to a Scottish grassland: Effects of soil and climatic conditions and a nitrification inhibitor. *The Science of the Total Environment*, *508*, 343–353. https://doi.org/10.1016/J.SCITOTENV.2014.12.008

Billen, G., Lassaletta, L., & Garnier, J. (2015). A vast range of opportunities for feeding the world in 2050: Trade-off between diet, N contamination and international trade. *Environmental Research Letters*, *10*, 25001. https://doi.org/10.1088/1748-9326/10/2/025001

Bourdin, F., Sakrabani, R., Kibblewhite, M. G., & Lanigan, G. J. (2014). Effect of slurry dry matter content, application technique and timing on emissions of ammonia and greenhouse gas from cattle slurry applied to grassland soils in Ireland. *Agriculture, Ecosystems & Environment*, *188*, 122–133. https://doi.org/10.1016/J.AGEE.2014.02.025

Cahalan, E., Ernfors, M., Müller, C., Devaney, D., Laughlin, R. J., Watson, C. J., Hennessy, D., Grant, J., Khalil, M. I., McGeough, K. L., & Richards, K. G. (2015). The effect of the nitrification inhibitor dicyandiamide (DCD) on nitrous oxide and methane emissions after cattle slurry application to Irish grassland. *Agriculture, Ecosystems & Environment*, *199*, 339–349. https://doi.org/10.1016/J.AGEE.2014.09.008

Cardenas, L. M., Misselbrook, T. M., Hodgson, C., Donovan, N., Gilhespy, S., Smith, K. A., Dhanoa, M. S., & Chadwick, D. (2016). Effect of the application of cattle urine with or without the nitrification inhibitor DCD, and dung on greenhouse gas emissions from a UK grassland soil. *Agriculture, Ecosystems and Environment*, *235*, 229–241. https://doi.org/10.1016/j.agee.2016.10.025

Cavigelli, M. A., & Robertson, G. P. (2001). Role of denitrifier diversity in rates of nitrous oxide consumption in a terrestrial ecosystem. *Soil Biology and Biochemistry*, *33*, 297–310.

Chen, L., Liu, L., Qin, S., Yang, G., Fang, K., Zhu, B., Kuzyakov, Y., Chen, P., Xu, Y., & Yang, Y. (2019). Regulation of priming effect by soil organic matter stability over a broad geographic scale. *Nature Communications*, *10*, 1–10. https://doi.org/10.1038/s41467-019-13119-z

Chojnacka, K., Moustakas, K., & Witek-Krowiak, A. (2020). Bio-based fertilizers: A practical approach towards circular economy. *Bioresource Technology*, *295*, 122223. https://doi.org/10.1016/J.BIORTECH.2019.122223

Cookson, W. R., Rowarth, J. S., & Cameron, K. C. (2000). The effect of autumn applied ^{15}N-labelled fertilizer on nitrate leaching in a cultivated soil during winter. *Nutrient Cycling in Agroecosystems*, *56*, 99–107. https://doi.org/10.1023/A:1009823114444

Creighton, P., Gilliland, T. J., Delaby, L., Kennedy, E., Boland, T. M., & O'Donovan, M. (2012). Effect of *Lolium perenne* sward density on productivity under simulated and actual cattle grazing. *Grass and Forage Science*, *67*, 526–534.

Davidson, E. A. (1991). Fluxes of nitrous oxide and nitric oxide from terrestrial ecosystems. In J. E. Rogers & W. B. Whitman (Eds.), *Microbial production and consumption of greenhouse gases: Methane, nitrogen oxides, and halomethanes* (pp. 219–235). American Society for Micro-biology.

De Vries, F. T. (2009). Soil fungi and nitrogen cycling. Causes and consequences of changing fungal biomass in grasslands. Ph.D thesis Wageningen Universiteit, Wageningen, p. 126.

Dechow, R., & Freibauer, A. (2011). Assessment of German nitrous oxide emissions using empirical modelling approaches. *Nutrient Cycling in Agroecosystems*, *91*, 235–254.

Delin, S., Stenberg, B., Nyberg, A., & Brohede, L. (2012). Potential methods for estimating nitrogen fertilizer value of organic residues. *Soil Use and Management*, *28*, 283–291.

Drury, C. F., Oloya, T. O., Tan, C. S., van Luyk, C. L., McKenney, D. J., & Gregorich, E. G. (1998). Long-term effects of fertilization and rotation on denitrification and soil carbon. *Soil Science Society of America Journal*, *62*, 1572–1579.

Dü, V. (2017). Verordnung über die Anwendung von Düngemitteln, Bodenhilfsstoffen, Kultursubstraten und Pflanzenhilfsmitteln nach den Grundsätzen der guten fachlichen Praxis beim Düngen. (Düngeverordnung - DüV). Bundesministerium der Justiz und für Verbraucherschutz.

European Environment Agency. (2021). Annual European Union greenhouse gas inventory 1990–2019 and inventory report 2021 (No. EEA/PUBL/2021/066). Accessed on February 20, 2022 from https://www.eea.europa.eu/publications/annual-european-union-greenhouse-gas-inventory-2021

Fan, C., Li, B., & Xiong, Z. (2018). Nitrification inhibitors mitigated reactive gaseous nitrogen intensity in intensive vegetable soils from China. *Science of the Total Environment*, *612*, 480–489. https://doi.org/10.1016/j.scitotenv.2017.08.159

Fan, D., He, W., Smith, W. N., Drury, C. F., Jiang, R., Grant, B. B., Shi, Y., Song, D., Chen, Y., Wang, X., He, P., & Zou, G. (2022). Global evaluation of inhibitor impacts on ammonia and nitrous oxide emissions from agricultural soils: A meta-analysis. *Global Change Biology*, *28*, 5121–5141.

Fangueiro, D., Hjorth, M., & Gioelli, F. (2015). Acidification of animal slurry: A review. *Journal of Environmental Management*, *149*, 46–56. https://doi.org/10.1016/j.jenvman.2014.10.001

Flessa, H., Dörsch, P., & Beese, F. (1995). Seasonal variation of N_2O and CH_4 fluxes in differently managed arable soils in Southern Germany. *Journal of Geophysical Research*, *100*, 23115–23124.

Flessa, H., Ruser, R., Schilling, R., Loftfield, N., Munch, J. C., Kaiser, E. A., & Beese, F. (2002). N_2O and CH_4 fluxes in potato fields: Automated measurement, management effects and temporal variation. *Geoderma*, *105*, 307–325.

Fowler, D., Coyle, M., Skiba, U., Sutton, M. A., Cape, J. N., Reis, S., Sheppard, L. J., Jenkins, A., Grizzetti, B., Galloway, J. N., Vitousek, P., Leach, A., Bouwman, A. F., Butterbach-Bahl, K., Dentener, F., Stevenson, D., Amann, M., & Voss, M. (2013). The global nitrogen cycle in the twenty-first century. *Philosophical Transactions of the Royal Society B: Biological Sciences*, *368*(1621), 20130164. https://doi.org/10.1098/RSTB.2013.0164

Friedl, J., Scheer, C., Rowlings, D. W., Deltedesco, E., Gorfer, M., de Rosa, D., Grace, P. R., Müller, C., & Keiblinger, K. M. (2020). Effect of the nitrification inhibitor 3,4-dimethylpyrazole phosphate (DMPP) on N-turnover, the N_2O reductase-gene nosZ and N_2O: N_2 partitioning from agricultural soils. *Scientific Reports*, *10*, 1–11. https://doi.org/10.1038/s41598-020-59249-z

Fu, J., Gasche, R., Wang, N., Lu, H., Butterbach-Bahl, K., & Kiese, R. (2017). Impacts of climate and management on water balance and nitrogen leaching from montane grassland soils of S-Germany. *Environmental Pollution*, *229*, 119–131.

Fuchs, K., Merbold, L., Buchmann, N., & Bellocchi, G. (2020). Evaluating the potential of legumes to mitigate N_2O emissions from permanent grassland using process-based models. *Global Biogeochemical Cycles*, *34*, e2020GB006561.

Giraud, M., Groh, J., Gerke, H. H., Brüggemann, N., Vereecken, H., & Pütz, T. (2021). Soil nitrogen dynamics in a managed temperate grassland under changed climatic conditions. *Water*, *13*, 931. https://doi.org/10.3390/w13070931

Gómez-Muñoz, B., Case, S. D. C., & Jensen, L. S. (2016). Pig slurry acidification and separation techniques affect soil N and C turnover and N_2O emissions from solid, liquid and biochar fractions. *Journal of Environmental Management*, *168*, 236–244.

Granli, T., & Bøckman, O. C. (1994). Nitrous oxide from agriculture. Norwegian Journal of Agricultural Science, Agricultural University of Norway-Advisory Service, As, Norway. p. 128 (Supplement 12).

Gu, J., Nicoullaud, B., Rochette, P., Grossel, A., Hénault, C., Cellier, P., & Richard, G. (2013). A regional experiment suggests that soil texture is a major control of N2O emissions from tile-drained winter wheat fields during the fertilization period. *Soil Biology and Biochemistry*, *60*, 134–141. https://doi.org/10.1016/J.SOILBIO.2013.01.029

Gu, J., Zheng, X., & Zhang, W. (2009). Background nitrous oxide emissions from crop-lands in China in the year 2000. *Plant and Soil*, *320*, 307–320.

Harrison-Kirk, T., Beare, M. H., Meenken, E. D., & Condron, L. M. (2013). Soil organic matter and texture affect responses to dry/wet cycles:

Effects on carbon dioxide and nitrous oxide emissions. *Soil Biology and Biochemistry*, *57*, 43–55. https://doi.org/10.1016/j.soilbio.2012.10.008

Harty, M. A., Forrestal, P. J., Watson, C. J., McGeough, K. L., Carolan, R., Elliot, C., Lanigan, G. J., Laughlin, R. J., Richards, K. G., & Lanigan, G. J. (2016). Reducing nitrous oxide emissions by changing N fertilizer use from calcium ammonium nitrate (CAN) to urea-based formulations. *Science of the Total Environment*, *563*, 576–586. https://doi.org/10.1016/j.scitotenv.2016.04.120

Hawkesford, M., Horst, W., Kichey, T., Lambers, H., Schjoerring, J., Møller, I. S., & White, P. (2012). Functions of macronutrients. In P. Marschner (Ed.), *Marschner's mineral nutrition of higher plants* (3rd ed., pp. 151–158). Academic Press.

Hernandez-Ramirez, G., Ruser, R., & Kim, D. G. (2021). How does soil compaction alter nitrous oxide fluxes? A meta-analysis. *Soil and Tillage Research*, *211*, 105036. https://doi.org/10.1016/j.still.2021.105036

Herr, C., Mannheim, T., Müller, T., & Ruser, R. (2019). Effect of cattle slurry application techniques on N_2O and NH_3 emissions from a loamy soil. *Journal of Plant Nutrition and Soil Science*, *182*, 964–979. https://doi.org/10.1002/jpln.201800376

Herr, C., Mannheim, T., Müller, T., & Ruser, R. (2020). Effect of nitrification inhibitors on N_2O emissions after cattle slurry application. *Agronomy*, *10*, 1174. https://doi.org/10.3390/agronomy10081174

Holtan-Hartwig, L., Dörsch, P., & Bakken, L. R. (2002). Low temperature control of soil denitrifying communities: Kinetics of N_2O production and reduction. *Soil Biology and Biochemistry*, *34*, 1797–1806.

Huber, P. J. (1981). *Robust statistics*. John Wiley & Sons, Inc. https://doi.org/10.1002/0471725250

Hunt, D., Bittman, S., Chantigny, M., & Lemke, R. (2019). Year-round N_2O emissions from long-term applications of whole and separated liquid dairy slurry on a perennial grass sward and strategies for mitigation. *Frontiers in Sustainable Food Systems*, *3*, 86. https://doi.org/10.3389/FSUFS.2019.00086/BIBTEX

Hutchinson, G., & Mosier, A. (1981). Improved soil cover method for field measurement of nitrous oxide fluxes. *Soil Science Society of America Journal*, *45*, 311–316.

IPCC. (2019). In E. Calvo Buendia, K. Tanabe, A. Kranjc, J. Baasansuren, M. Fukuda, S. Ngarize, A. Osako, Y. Pyrozhenko, P. Shermanau, & S. Federici (Eds.), *2019 refinement to the 2006 IPCC Guidelines for National Greenhouse Gas Inventories*. IPCC Accessed on March 30, 2022 from https://www.ipcc-nggip.iges.or.jp/public/2019rf/vol4.html

IPCC. (2022). Global Warming Potentials (IPCC Second Assessment Report). Accessed on August 19, 2022 at https://unfccc.int/process/transparency-and-reporting/greenhouse-gas-data/greenhouse-gas-data-unfccc/global-warming-potentials

IUSS Working Group. (2015). World Reference Base for soil resources 2014, update 2015. International Soil Classification System for Naming Soils and Creating Legends for Soil Maps. *World Soil Resources Reports*, 106. FAO, Rome.

Jensen, L. S. (2013). Animal manure fertilizer value, crop utilization and soil quality impacts. In S. G. Sommer, M. L. Christensen, T. Schmidt, & L. S. Jensen (Eds.), *Animal manure recycling: Treatment & Management*. John Wiley & Sons Ltd. https://doi.org/10.1002/9781118676677.ch15

Jungkunst, H. F., Freibauer, A., Neufeldt, H., & Bareth, H. (2006). Nitrous oxide emissions from agricultural land use in Germany- a synthesis of available annual field data. *Journal of Plant Nutrition and Soil Science*, *169*(3), 341–351. https://doi.org/10.1002/jpln.200521954

Kanter, D. R., Wagner-Riddle, C., Groffman, P. M., Davidson, E. A., Galloway, J. N., Gourevitch, J. D., van Grinsven, H. J. M., Houlton, B. Z., Keeler, B. L., Ogle, S. M., Pearen, H., Rennert, K. J., Saifuddin, M., Sobota, D. J., & Wagner, G. (2021). Improving the social cost of nitrous oxide. *Nature Climate Change*, *11*, 1008–1010. https://doi.org/10.1038/s41558-021-01226-z

Kelliher, F. M., Clough, T. J., Clark, H., Rys, G., & Sedcole, J. R. (2008). The temperature dependence of dicyandiamide (DCD) degradationin soils: A data synthesis. *Soil Biology and Biochemistry*, *40*, 1878–1882.

Keskinen, R., Termonen, M., Tapio, S., Luostarinen, S., & Räty, M. (2022). Slurry acidification outperformed injection as an ammonia emission-reducing technique in boreal grass cultivation. *Nutrient Cycling in Agroecosystems*, *122*, 139–156. https://doi.org/10.1007/s10705-021-10190-1

Kolasa-Więcek, A. (2018). Neural modeling of greenhouse gas emission from agricultural sector in European Union member countries. *Water, Air, and Soil Pollution*, *229*, 205. https://doi.org/10.1007/S11270-018-3861-7

LaHue, G. T., van Kessel, C., Linquist, B. A., Adviento-Borbe, M. A., & Fonte, S. J. (2016). Residual effects of fertilization history increase nitrous oxide emissions from zero-N controls: Implications for estimating fertilizer-induced emission factors. *Journal of Environmental Quality*, *45*, 1501–1508. https://doi.org/10.2134/JEQ2015.07.0409

Lalor, S. (2011). Nitrogen fertilizer replacement value of cattle slurry in grassland as affected by method and timing of application. *Journal of Environmental Quality*, *40*, 362–373. https://doi.org/10.2134/jeq2010.0038

Lassaletta, L., Billen, G., Grizzetti, B., Anglade, J., & Garnier, J. (2014). 50-year trends in nitrogen use efficiency of world cropping systems: The relationship between yield and nitrogen input to crop-land. *Environmental Research Letters*, *9*, 1–9.

Li, S., Song, L., Gao, X., Jin, Y., Liu, S., Shen, Q., & Zou, J. (2017). Microbial abundances predict methane and nitrous oxide fluxes from a windrow composting system. *Frontiers in Microbiology*, *8*, 409. https://doi.org/10.3389/FMICB.2017.00409

Liu, B., Mørkved, P. T., Frostegård, A., & Bakken, L. R. (2010). Denitrification gene pools, transcriptionand kinetics of NO, N_2O and N_2 production as affected by soil pH. *FEMS Microbiology Ecology*, *72*, 407–417.

Liu, S., Schloter, M., & Brüggemann, N. (2018). Accumulation of NO_2^- during periods of drying stimulates soil N_2O emissions during subsequent rewetting. *European Journal of Soil Science*, *69*, 936–946.

Machado, P. V. F., Wagner-Riddle, C., MacTavish, R., Voroney, P. R., & Bruulsema, T. W. (2019). Diurnal variation and sampling frequency effects on nitrous oxide emissions following nitrogen fertilization and spring-thaw events. *Soil Science Society of America Journal*, *83*, 743–750.

Malique, F., Wangari, E., Andrade-Linares, D. R., Schloter, M., Wolf, B., Dannenmann, M., Schulz, S., & Butterbach-Bahl, K. (2021). Effects of slurry acidification on soil N_2O fluxes and denitrification. *Journal of Plant Nutrition and Soil Science*, *184*, 696–708. https://doi.org/10.1002/JPLN.202100095

Maris, S. C., Abalos, D., Capra, F., Moscatelli, G., Scaglia, F., Cely Reyes, G. E., Ardenti, F., Boselli, R., Ferrarini, A., Mantovi, P., Tabaglio, V., & Fiorini, A. (2021). Strong potential of slurry application timing and method to reduce N losses in a permanent grassland. *Agriculture, Ecosystems and Environment*, *311*, 107329. https://doi.org/10.1016/j.agee.2021.107329

Marsden, K. A., Jones, D. L., & Chadwick, D. R. (2016). The urine patch diffusional area: An important N_2O source? *Soil Biology and Biochemistry*, *92*, 161–170. https://doi.org/10.1016/j.soilbio.2015.10.011

Mathivanan, G. P., Eysholdt, M., Zinnbauer, M., Rösemann, C., & Fuß, R. (2021). New N_2O emission factors for crop residues and fertilizer inputs to agricultural soils in Germany. *Agriculture, Ecosystems and Environment*, *322*, 107640. https://doi.org/10.1016/j.agee.2021.107640

Melaku, N. D., Shrestha, N. K., Wang, J., & Thorman, R. E. (2020). Predicting nitrous oxide emissions after the application of solid manure to grassland in the United Kingdom. *Journal of Environmental Quality*, *49*(1), 1–13. https://doi.org/10.1002/JEQ2.20002

Menéndez, S., Barrena, I., Setien, I., González-Murua, C., & Estavillo, J. M. (2012). Efficiency of nitrification inhibitor DMPP to reduce nitrous oxide emissions under different temperature and moisture conditions.

Soil Biology and Biochemistry, *53*, 82–89. https://doi.org/10.1016/J.SOILBIO.2012.04.026

Minet, E. P., Jahangir, M. M. R., Krol, D. J., Rochford, N., Fenton, O., Rooney, D., Lanigan, G., Forrestal, P. J., Breslin, C., & Richards, K. G. (2016). Amendment of cattle slurry with the nitrification inhibitor dicyandiamide during storage: A new effective and practical N 2 O mitigation measure for landspreading. *Agriculture, Ecosystems and Environment*, *215*, 68–75. https://doi.org/10.1016/j.agee.2015.09.014

Nannen, D. U., Herrmann, A., Loges, R., Dittert, K., & Taube, F. (2011). Recovery of mineral fertilizer N and slurry N in continuous silage maize using the ^{15}N and difference methods. *Nutrient Cycling in Agroecosystems*, *89*, 269–280. https://doi.org/10.1007/s10705-010-9392-2

Nyameasem, J. K., Malisch, C. S., Loges, R., Taube, F., Kluß, C., Vogeler, I., & Reinsch, T. (2021). Nitrous oxide emission from grazing is low across a gradient of plant functional diversity and soil conditions. *Atmosphere*, *12*, 223. https://doi.org/10.3390/atmos12020223

Nyord, T., Kristensen, E. F., Munkholm, L. J., & Jørgensen, M. H. (2010). Design of a slurry injector for use in a growing cereal crop. *Soil and Tillage Research*, *107*, 26–35. https://doi.org/10.1016/J.STILL.2010.01.001

O'Neill, M., Gallego-Lorenzo, L., Lanigan, G. J., Forristal, P. D., & Osborne, B. A. (2020). Assessment of nitrous oxide emission factors for arable and grassland ecosystems. *Journal of Integrative Environmental Sciences*, *17*(3), 165–185. https://doi.org/10.1080/1943815X.2020.1825227

Owusu-Twum, M. Y., Loick, N., Cardenas, L. M., Coutinho, J., Trindade, H., & Fangueiro, D. (2017). Nitrogen dynamics in soils amended with slurry treated by acid or DMPP addition. *Biology and Fertility of Soils*, *53*, 339–347. https://doi.org/10.1007/s00374-017-1178-0

Park, S. H., Lee, B. R., Jung, K. H., & Kim, T. H. (2018). Acidification of pig slurry effects on ammonia and nitrous oxide emissions, nitrate leaching, and perennial ryegrass regrowth as estimated by ^{15}N-urea flux. *Asian-Australasian Journal of Animal Sciences*, *31*, 457–466. https://doi.org/10.5713/AJAS.17.0556

Pedersen, B. N., Christensen, B. T., Bechini, L., Cavalli, D., Eriksen, J., & Sorensen, P. (2020). Nitrogen fertilizer value of animal slurries with different proportions of liquid and solid fractions: A 3-year study under field conditions. *The Journal of Agricultural Science*, *158*, 707–717. https://doi.org/10.1017/S0021859621000083

Pedersen, J., Nyord, T., Feilberg, A., Labouriau, R., Hunt, D., & Bittman, S. (2021). Effect of reduced exposed surface area and enhanced infiltration on ammonia emission from untreated and separated cattle slurry. *Biosystems Engineering*, *211*, 141–151. https://doi.org/10.1016/j.biosystemseng.2021.09.003

Peters, T., Taube, F., Kluß, C., Reinsch, T., Loges, R., & Fenger, F. (2021). How does nitrogen application rate affect plant functional traits and crop growth rate of perennial ryegrass-dominated permanent pastures? *Agronomy*, *11*, 2499.

Petersen, S. O., Andersen, A. J., & Eriksen, J. (2012). Effects of cattle slurry acidification on ammonia and methane evolution during storage. *Journal of Environmental Quality*, *41*, 88–94.

Pfab, H., Palmer, I., Buegger, F., Fiedler, S., Müller, T., & Ruser, R. (2012). Influence of a nitrification inhibitor and of placed N-fertilization on N_2O fluxes from a vegetable cropped loamy soil. *Agriculture, Ecosystems & Environment*, *150*, 91–101.

R Core Team. (2021). The R Foundation for Statistical Computing Platform: x86_64-w64-mingw32/x64 (64-bit). https://www.R-project.org/

Ravishankara, A. R., Daniel, J. S., & Portman, R. W. (2009). Nitrous oxide (N_2O): The dominant ozone-depleting substance emitted in the 21st century. *Science*, *326*, 123–125.

Rees, R. M., Augustin, J., Alberti, G., Ball, B. C., Boeckx, P., Cantarel, A., Castaldi, S., Chirinda, N., Chojnicki, B., Giebels, M., Gordon, H., Grosz, B., Horvath, L., Juszczak, R., Kasimir Klemedtsson, Å., Klemedtsson, L., Medinets, S., Machon, A., Mapanda, F., … Wuta, M. (2013). Nitrous oxide emissions from European agriculture-an analysis of variability and drivers of emissions from field experiments. *Biogeosciences*, *10*, 2671–2682. https://doi.org/10.5194/bg-10-2671-2013

Reinsch, T., Malisch, C., Loges, R., & Taube, F. (2020). Nitrous oxide emissions from grass–clover swards as influenced by sward age and biological nitrogen fixation. *Grass and Forage Science*, *75*, 372–384. https://doi.org/10.1111/gfs.12496

Rodhe, L. (2004). Development and evaluation of shallow injection of slurry into ley. Doctoral dissertation. Uppsala: Sveriges lantbruksuniv., acta universitatis Agriculturae Sueciae Agraria, 482, 1401–6249.

Ruser, R., Fuß, R., Andres, M., Hegewald, H., Kesenheimer, K., Köbke, S., Räbiger, T., Quinones, T. S., Augustin, J., Christen, O., Dittert, K., Kage, H., Lewandowski, I., Prochnow, A., Stichnothe, H., & Flessa, H. (2017). Nitrous oxide from winter oilseed rape cultivation. *Agriculture, Ecosystems & Environment*, *249*, 57–69.

Ruser, R., & Schulz, R. (2015). The effect of nitrification inhibitors on the nitrous oxide (N_2O) release from agricultural soils-a review. *Journal of Plant Nutrition and Soil Science*, *178*, 171–188. https://doi.org/10.1002/JPLN.201400251

Saggar, S., Jha, N., Deslippe, J., Bolan, N. S., Luo, J., Giltrap, D. L., Kim, D. G., Zaman, M., & Tillman, R. W. (2013). Denitrification and N_2O:N_2 production in temperate grasslands: Processes, measurements, modelling and mitigating negative impacts. *Science of the Total Environment*, *465*, 173–195.

Schlüter, S., Henjes, S., Zawallich, J., Bergaust, L., Horn, M., Ippisch, O., Dörsch, P., & Dörsch, P. (2018). Denitrification in soil aggregate analogues-effect of aggregate size and oxygen diffusion. *Frontiers in Environmental Science*, *6*, 17. https://doi.org/10.3389/fenvs.2018.00017

Schröder, J. J., Uenk, D., & Hilhorst, G. J. (2007). Long-term nitrogen fertilizer replacement value of cattle manures applied to cut grassland. *Plant and Soil*, *299*(1–2), 83–99. https://doi.org/10.1007/S11104-007-9365-7/FIGURES/8

Seitzinger, S. P., & Phillips, L. (2017). Nitrogen stewardship in the Anthropocene. *Science*, *357*, 350–351. https://doi.org/10.1126/science.aao0812

Soares, J. R., Souza, B. R., Mazzetto, A. M., Galdos, M. V., Chadwick, D. R., Campbell, E. E., Jaiswal, D., Oliveira, J. C., Monteiro, L. A., Vianna, M. S., Lamparelli, R. A. C., Figueiredo, G. K. D. A., Sheehan, J. J., & Lynd, L. R. (2023). Mitigation of nitrous oxide emissions in grazing systems through nitrification inhibitors: A meta-analysis. *Nutrient Cycling in Agroecosystems*, *125*, 359–377. https://doi.org/10.1007/s10705-022-10256-8

Sørensen, P., Bechini, L., & Jensen, L. S. (2019). Manure management in organic farming. In U. Köpke (Ed.), *Improving organic crop cultivation* (pp. 179–209). Burleigh Dodds Science Publishing.

Subbarao, G., Ito, O., Sahrawat, K., Berry, W., Nakahara, K., Ishikawa, T., Watanabe, T., Suenaga, K., Rondon, M., & Rao, I. (2007). Scope and strategies for regulation of nitrification in agricultural systems–Challenges and opportunities. *Critical Reviews in Plant Sciences*, *25*, 303–335. https://doi.org/10.1080/07352680600794232

Thies, S., Joshi, D. R., Bruggeman, S. A., Clay, S. A., Mishra, U., Morile-Miller, J., & Clay, D. E. (2019). Fertilizer timing affects nitrous oxide, carbon dioxide, and ammonia emissions from soil. *Soil Science Society of America Journal*, *84*, 115–130. https://doi.org/10.1002/SAJ2.20010

Thorman, R. E., Nicholson, F. A., Topp, C. F. E., Bell, M. J., Cardenas, L. M., Chadwick, D. R., Cloy, J. M., Misselbrook, T. H., Rees, R. M., Watson, C. J., & Williams, J. R. (2020). Towards country-specific nitrous oxide emission factors for manures applied to arable and grassland soils in the UK. *Frontiers in Sustainable Food Systems*, *4*, 62. https://doi.org/10.3389/FSUFS.2020.00062/BIBTEX

United Nations. (2019). World population prospects 2019: Highlights. Department of Economic and Social Affairs, population division, ST/-ESA/SER.A/423.

van der Weerden, T. J., Manderson, A., Kelliher, F. M., & de Klein, C. A. M. (2014). Spatial and temporal nitrous oxide emissions from dairy cattle urine deposited onto grazed pastures across New Zealand based on soil water balance modelling. *Agriculture, Ecosystems and Environment*, *189*, 92–100. https://doi.org/10.1016/j.agee.2014.03.018

VDLUFA. (2017). Kongressband 2017 Freising. Standortgerechte Landnutzung -umweltverträglich und wirtschaftlich. VDLUFA-Verlag, Darmstadt.

Velthof, G. L., & Mosquera, J. (2011). The impact of slurry application technique on nitrous oxide emission from agricultural soils. *Agriculture, Ecosystems & Environment*, *140*, 298–308. https://doi.org/10.1016/J.AGEE.2010.12.017

Voglmeier, K., Six, J., Jocher, M., & Ammann, C. (2019). Grazing-related nitrous oxide emissions: From patch scale to field scale. *Biogeosciences*, *16*, 1685–1703.

Webb, J., Sørensen, P., Velthof, G., Amon, B., Pinto, M., Rodhe, L., Salomon, E., Hutchings, N., Burczyk, P., & Reid, J. (2013). An assessment of the variation of manure nitrogen efficiency throughout Europe and an appraisal of means to increase manure-N efficiency. *Advances in Agronomy*, *119*, 371–442.

Webber, H., Lischeid, G., Sommer, M., Finger, R., Nendel, C., Gaiser, T., & Ewert, F. (2020). No perfect storm for crop yield failure in Germany. *Environmental Research Letters*, *15*, 104012.

Weiske, A., Benckiser, G., Herbert, T., & Ottow, J. C. G. (2001). Influence of the nitrification inhibitor 3,4-dimethylpyrazole phosphate (DMPP) in comparison to dicyandiamide (DCD) on nitrous oxide emissions, carbon dioxide fluxes and methane oxidation during 3 years of repeated application in field experiments. *Biology and Fertility of Soils*, *34*, 109–117.

Wilkins, P. W., & Humphreys, M. O. (2003). Progress in breeding perennial forage grasses for temperate agriculture. *Journal of Agricultural Science*, *140*, 129–150.

World Reference Base for Soil Resources. (2015). IUSS working group. World Reference Base for soil resources 2014, international soil classification system for naming soils and creating legends for soil maps. Update 2015. World soil resources reports, No.106, FAO, Rome, Italy.

Yeomans, J. C., & Bremner, J. M. (1991). Carbon and nitrogen analysis of soils by automated combustion techniques. *Communications in Soil Science and Plant Analysis*, *22*, 99–10. https://doi.org/10.1080/00103629109368458

Zistl-Schlingmann, M., Kengdo, S. K., Kiese, R., & Dannenmann, M. (2020). Management intensity controls nitrogen-use-efficiency and flows in grasslands–A ^{15}N tracing experiment. *Agronomy*, *10*, 606. https://doi.org/10.3390/AGRONOMY10040606

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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