Greenhouse gas balances and yield-scaled emissions for storage and field application of organic fertilizers derived from cattle manure

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1. Introduction

Ruminants excrete 75–95 % of the ingested N as dung and urine, and recycling the manure as an organic fertilizer is a widely used practice to supply essential nutrients for crop production (Castillo et al., 2000; Eckard et al., 2007). This is particularly needed in organic farming where synthetic N is not available to meet the N demand of plants (Chmelíková et al., 2021). Anaerobic co-digestion of livestock manure and other biomasses is a well-known strategy to produce biogas as a renewable energy source, and an additional benefit of this treatment is to increase the proportion of nutrients in plant available form compared to the untreated substrates (Holly et al., 2017). In organic farming, however, co-digestates are restricted to fiber-rich residues such as deep litter or plant biomass, and this may complicate the management of nutrients and environmental losses by significantly changing the physical and chemical properties of digestates. For example, Häfner et al. (2022) reported N fertilizer values of digestates based on food waste, manure and crop residues ranging from 18 % to 83 % across two years. Also, fiber-rich residues may increase the potential for CH₄ and N₂O emissions during storage and after field application (Wang et al., 2014; Baral et al., 2017).

Additional processing is possible to obtain new products that meet...
specific needs (Mata-Alvarez et al., 2000; Svensson et al., 2004; Odlare et al., 2011). For example, solid-liquid separation produces a nitrogen- and potassium-rich liquid fraction and a phosphorus-rich and dry matter-rich solid fraction (Møller et al., 2000; Baral et al., 2018). The solid fraction of a digestate can be used directly as a biofertilizer in agriculture, or it can be composted and used as a soil amendment (Dinuccio et al., 2008). Depending on separation technology, an NH₄-enriched liquid fertilizer with a reduced dry matter content is produced (Hjorth et al., 2011), and with physical characteristics that may promote infiltration and benefit plant uptake (Morris and Lathwell, 2004). Further treatments of digestates and separation products are possible, such as membrane treatment, biological treatment, stripping and evaporation, but there will be a trade-off between fertilizer value, environmental effects, and the cost of processing.

Manure processing to increase crop production should not have negative environmental impacts. Gaseous losses during storage and after field application contribute significantly to greenhouse gas (GHG) emissions, with far-reaching environmental effects (Zervas and Tsipiapoulou, 2012). It has been reported that methane (CH₄) was the main GHG source during storage, and N₂O was the main contributor after field application (Amon et al., 2006; Baral et al., 2018). Removing degradable organic matter by manure treatment such as anaerobic digestion or separation is expected to reduce the potential for methanogenesis and CH₄ emissions during storage (Amon et al., 2006; Holly et al., 2017), and in some cases reduce N₂O emissions after field application (Petersen, 1999; Möller and Stinner, 2009; Thomsen et al., 2010).

In order to ensure high availability of manure N for crop production, it is important to minimize N losses during both storage and field application. It was reported that the anaerobic digestion of dairy manure increased the NH₃ emissions during storage due to higher total ammoniacal nitrogen and pH (Sun et al., 2014; Neerackal et al., 2015). Temperature also affects NH₃ emissions especially when NH₃ level and pH are high (Clemens et al., 2006). Studies reporting N₂O emissions from digestate and untreated manure during storage were not consistent; Sommer et al. (2000) concluded that there were more N₂O emissions from digestate during storage, while other studies found insignificant differences (Amon et al., 2006; Holly et al., 2017).

A review of field studies found that field-applied organic fertilizers have a higher N₂O emission factor (1.21 ± 0.14 %) than the IPCC default emission factor of 1 % (IPCC et al., 2006; Charles et al., 2017). By comparison, N₂O emissions from inorganic fertilizers are much more variable and may be much less than 1 % depending on site conditions such as annual rainfall (Bell et al., 2015). Many studies have found that annual emissions of N₂O from well-drained arable soils occur mainly during the period in spring and early summer when N applied for crop production is available in the soil (Chirinda et al., 2010; Bell et al., 2015; Herr et al., 2020). A review by Möller (2015) concluded that the predominant response to anaerobic digestion of manure is a reduction of N₂O emissions after field application, possibly as a result of the lower availability of degradable carbon to maintain anaerobic conditions supporting denitrification (Baral et al., 2016). There is little information about the possible effect of separating digestate on soil N₂O emissions; (Meng et al., 2022) found in laboratory incubation experiments, surprisingly, higher N₂O emissions from the liquid fraction and concluded that denitrification was enhanced by the greater contact between the liquid fraction and soil.

Nitrogen use efficiency and crop yield were in some studies higher with digestate compared to untreated manure if application method, timing, and rate were optimal (Abuhaker et al., 2012; Webb et al., 2013). However, other studies found that crop yields were not significantly different between undigested and digested cattle slurry (Pötsch, 2005; Möller et al., 2008). The amount and composition of organic matter in organic fertilizers may also affect crop N availability through priming effects or inorganic N immobilization (Bernal and Kirchmann, 1992; Kirchmann and Lundvall, 1993).

Greenhouse gas emissions from organic fertilizers should be held up against crop N use efficiency to evaluate yield-scaled emissions. This is best done with a system-oriented approach where the emissions from both storage and field application are determined together with N use efficiency, to suggest best strategies for low-emission crop production. In this study we investigated (i) CH₄, N₂O and NH₃ emissions during storage of untreated cattle manure, a digestate based on cattle manure, and the liquid fraction of mechanically separated digestate; (ii) N₂O emissions from these organic fertilizers, as well as an NS fertilizer and mineral fertilizer treatment after field application; and (iii) the N uptake and yield of spring barley at harvest. Our hypotheses were: (i) GHG emissions from digested organic fertilizers would be less compared to untreated cattle manure during storage; (ii) N₂O emissions from untreated and digested fertilizer materials after field application would be comparable; and (iii) digested fertilizer materials would have higher crop yields compared to undigested fertilizer.

2. Materials and methods

2.1. Organic fertilizers

Four different fertilizer materials derived from cattle manure were part of this research including (1) untreated cattle manure (CA) as a reference; (2) a digestate produced mainly from cattle manure (DD); (3) the liquid fraction of separated DD (LF); and (4) a nitrogen-sulfur (NS) concentrate produced as described below. Liquid cattle manure was obtained from the experimental farm of the research center belonging to Aarhus University; the breed of cattle was Holstein. Digestate was obtained from an experimental biogas facility where the substrate for anaerobic digestion was a mixture of 92.5 % cattle manure and 7.5 % grass-clover silage by weight. The daily load was 270 kg for a 10 m³ digester with an active volume of 9 m³. The operating temperature was 51 °C, and the hydraulic retention time (HRT) was 33 days. The liquid fraction of digestate was obtained after mechanical separation of digestate using a GEA Westphalia decanting centrifuge (UCD 305–00–02, Germany). Before separation, the digestate had been stored at 20 °C for 30 days to collect residual biogas during the cooling phase. An NS fertilizer was produced by passing the biogas over a bio-trickling desulfurization filter with a biofilm of bacteria under microaerophilic conditions, where H₂S was oxidized to elemental sulfur or H₂SO₄, and during this process, the pH was reduced from 7.9 to 2.0. The biofilter was supplied with a liquid manure fraction from a 50 µm microfiltration (Sepcom, Italy). The acid liquid was subsequently enriched with a concentrated ammonia solution (25 %) until a final pH of about 6.5. NH₃ would originate from the process of drying the separated solids. Selected physical-chemical properties of the different manure-derived products are shown in Table 1.

2.2. Storage experiment

A pilot-scale experiment was conducted in covered and partly below-ground tanks (6.5 m³) with continuous ventilation to quantify the GHG and NH₃ emissions during storage. Detailed information about the facility is given in Petersen et al. (2009) and pictures in Fig. S1. Storage took place between 21 August and 7 November 2020 (11 weeks). The minimum, maximum, and average air temperatures during the period of the storage experiment were 0.6, 25.4, and 11.9 °C, respectively, and the cumulative rainfall was 157 mm. The daily precipitation and air temperature during the storage period (as well as the field application period introduced below) are shown in Fig. S2. Approximately 2 m³ portions of untreated cattle manure (CA), digestate (DD), and the liquid fraction of the digestate (LF) were stored with two replicates per treatment. The liquid volume in each tank was measured at the beginning of the experiment. The liquid volume in each tank was measured at the beginning of the experiment.
ventilation air with a peristaltic pump operating continuously at 15 ml min$^{-1}$. For the determination of NH$_3$ loss, acid traps with 20 mM phosphoric acid were inserted near the sampling position, and the acid was replaced every 7 days. For determination of CH$_4$ and N$_2$O in the ventilation air, the subsamples of ventilation air from each tank were led to a rack with three-way solenoid valves downstream from the pump; the valves were programmed to collect ventilation air in 3-liter aluminum foil gas sampling bags (SKC Ltd.) for 1 min in every 60 min. The gas sampling bags were also replaced weekly. Following the 11-week storage period with monitoring of emissions, a portion of each organic fertilizer was transferred to a 1 m$^3$ closed plastic pellet tank for storage in a shelter until the following spring.

### 2.3. Field experiment

The field site was located at Foulumgaard (56'49'N, 09'58'E) in Central Jutland, Denmark. The soil is classified as loamy sand with 9% clay, 13% silt, and 78% sand. Soil bulk density after cultivation is 1.35 g cm$^{-3}$ (0–25 cm), and the soil contains around 1.8 g kg$^{-1}$ total N, 23 g kg$^{-1}$ soil organic matter, 33 mg kg$^{-1}$ Olsen-P, 120 mg kg$^{-1}$ extractable K. It had a pH of 6.5 (0.01 M CaCl$_2$) (Li et al., 2015). The mean annual precipitation and air temperature (1996–2016) of the site were 774 mm and 8.2$^\circ$C, respectively (Auret et al., 2020). The minimum, maximum, and average air temperatures during the experimental period from the day of fertilization (13 April 2021) to three days after harvest (13 August 2021) were 2.6, 28.0, and 13.3$^\circ$C, respectively, and the cumulative rainfall was 302 mm.

The field experiment included four treatments with organic fertilizers: (1) CA; (2) DD; (3) LF; (4) NS. The application rates were close to 100 kg total N ha$^{-1}$; specific application rates are given in Table 1. Besides, three levels of mineral fertilizer were included at application rates of (5) 100 kg ha$^{-1}$ total N (M100) and (6) an unfertilized control (M0). The mineral fertilizer was AXAN NS 27–4 with 27% nitrogen (equal proportions of NH$_4$ and NO$_3$) and 4% sulfur. The experimental area was subdivided into three blocks, each with eight 15 m × 3 m plots separated by 1.5 m wide strips. The organic fertilizers were applied by direct injection (10 cm depth, 24 cm between injectors) after plowing on 13 April 2021 with except for NS fertilizer, where the application was split over two days to allow for infiltration of a large liquid volume. Hence, half of the NS fertilizer was injected before plowing, and the other half the following day after plowing. All mineral fertilizers were surface-applied on 13 April 2021. The crop was spring barley (Hordeum vulgare L) which was sown on 14 April 2021 and harvested on 13 August 2021. The field experiment was managed according to Danish organic farming rules, except for the treatments with mineral N. No pesticides were used and the barley was protected against weeds by blind-harrowing at an early stage, and by establishing the barley at 24 cm row distance combined with inter-row hoeing in May (De Notaris et al., 2019).

The first soil and gas samples were collected on 15 April 2021, that is, 2 days (1 day for a part of the NS fertilizer) after fertilizer application, as well as 6 and 9 days after fertilization. Subsequently, samples were collected once per week during week 3–11 after fertilization, and in week 13 and 19 (three days after harvest). Gas sampling was always initiated around 10:00 (mid-morning) to represent the daily average soil temperature (Reeves and Wang, 2015). Flux measurements were done using two-part static chambers (75 cm × 75 × 20 cm) supported by stainless steel frames inserted to c. 10 cm depth immediately after seeding; the chambers were equipped with a septum for gas sampling and a battery-driven fan to mix headspace gas, but no vent. Ten ml headspace gas was sampled at chamber deployment and additionally three times within a c. 2-hour period; gas samples were transferred to 6 ml pre-evacuated extainers (Labco, Lampeter, UK) for analysis by gas chromatography.

Soil sampling took place in parallel with gas sampling. Six sub-samples (0–20 cm depth) were taken using a 2 mm diameter auger and pooled. Soil samples were stored at −20$^\circ$C before analysis of gravimetric moisture and mineral N content. The barley crop was harvested on 13 August 2021 with a combine harvester and divided into grain and straw. The above-ground dry matter was determined after drying at 60$^\circ$C for 40 h. The total N of the harvested plant material is expressed as a percentage of DM. The apparent fertilizer N use efficiency (NUE) was calculated as:

$$\text{NUE} = (N_{\text{fert}, i} - N_{\text{control}})/N_{\text{applied}, i}$$

where $N_{\text{fert}, i}$ is the crop N uptake (kg N ha$^{-1}$ in grain + straw) with fertilizer $i$; $N_{\text{control}}$ is the crop N uptake in the unfertilized soil (control); and $N_{\text{applied}, i}$ is the total N application rate (kg TN ha$^{-1}$) with fertilizer $i$.

### 2.4. Analytical methods

Concentrations of N$_2$O and CH$_4$ were determined by Model 7890 gas chromatography system with dual-inlet injection (Agilent; Nærum, Denmark) configured as previously described by Petersen et al. (2012). Detection limits were 0.042 and 61 µL L$^{-1}$ for N$_2$O and CO$_2$, respectively.

The moisture content of soil and organic fertilizers were measured by drying at 105$^\circ$C for 24 h, and the volatile solids (VS) of organic fertilizers after an additional 6 h at 500$^\circ$C after drying at 105$^\circ$C for 24 h. The pH and electrical conductivity were measured by a MeterLab CDM210

### Table 1

<table>
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<tr>
<th>Slurry type</th>
<th>DM</th>
<th>VS</th>
<th>TN</th>
<th>TC</th>
<th>NH$_4$-N</th>
<th>pH</th>
<th>NH$_4$-N/TN</th>
<th>Appl. Rate</th>
</tr>
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<td>39.2</td>
<td>2.3</td>
<td>–</td>
<td>1.1</td>
<td>7</td>
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<td>–</td>
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<tr>
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<td>–</td>
<td>2.1</td>
<td>7.8</td>
<td>0.68</td>
<td>–</td>
</tr>
<tr>
<td>LF</td>
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<td>19</td>
<td>2.8</td>
<td>–</td>
<td>1.9</td>
<td>7.9</td>
<td>0.68</td>
<td>–</td>
</tr>
<tr>
<td>NS</td>
<td>Storage, beginning (August 21, 2020)</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
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<td>–</td>
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<td>0.61</td>
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</tr>
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<td>–</td>
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<td>–</td>
</tr>
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<td>NS</td>
<td>Storage, end (November 7, 2020)</td>
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<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Field application (April 13, 2021)</td>
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<td>–</td>
<td>2.5</td>
<td>78</td>
<td>1.2</td>
<td>7.4</td>
<td>0.48</td>
</tr>
<tr>
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<td>0.8</td>
<td>7.3</td>
<td>0.67</td>
<td>114.9</td>
</tr>
</tbody>
</table>

*Not included in storage experiment.*
(Hach; Loveland, Colorado, USA) in a 1:1 (w/v) soil:water slurry. For analysis of NH$_4^+$ and NO$_3^-$ in soil samples, about 10 g fresh wt. soil was extracted in 40 ml 1 M KCl; the mixture was rotated end-over-end for 30 min and centrifuged at 1500 rpm for 5 min. The supernatant was then filtered through a microfilter filter (Filter 691, VWR Europe) and measured colorimetrically by continuous flow analysis using an Auto-analyser III (Bran+Luebbe, Germany). The Henderson-Hasselbalch equation was used to calculate concentrations of NH$_3$ in the manure at the beginning of storage from pH and the total ammoniacal N concentration (Hao et al., 2005); the calculation is in Supplementary information. The total N of organic fertilizers and plant materials was determined by the Kjeldahl method (Novozamsky et al., 1983).

2.5. Data analyses

In the storage experiment, weekly average gas fluxes were calculated as the product of ventilation rate, logged as 15-minute averages for the individual storage units, and gas concentrations in the time-integrated gas samples composed of hourly 15 ml subsamples. A separate gas sampling line, collecting ambient air at 1 m height in a position among the pilot-scale storage tanks, represented the background concentrations of CH$_4$, N$_2$O and NH$_3$. The temperature used was a 7-day moving average of air temperature. Only treatment CA was observed to form a thin crust inside the tank during storage.

In the field experiment, fluxes of N$_2$O were calculated by the flux estimation software HMR using new functions pfvar, pfalpa, SatPct and SatTimeMin which constrain the datasets analyzed with a nonlinear model. The parameters selected for pfvar and pfalpa represent pre-filtering to avoid using a nonlinear model with low fluxes for which measurement uncertainty is high. Here, pfvar was set at 0.0001 μg N$_2$O l$^{-1}$ corresponding to a coefficient of variation of 3 %, while the default significance level, p < 0.05, was used for pfalpa. SatPct and SatTimeMin together limit the curvilinearity of nonlinear fluxes, and here the parameter values were 90 and 2, respectively, which means that an exponential curve increasing to a maximum would not be allowed to reach 90 % of full saturation until 2 h after chamber deployment. HMR is available as an add-on package in R (R Core Team, 2014). The cumulative N$_2$O emissions were calculated by linear interpolation between daily fluxes.

The emissions of CH$_4$, N$_2$O and NH$_3$ during storage, as well as CO$_2$ equivalents (100-year time horizon) from CH$_4$ and N$_2$O, and indirect N$_2$O emissions from NH$_3$, were analyzed by mixed-effects models using the “lme” function from the R package “nlme”. Global warming potentials of 28 and 265 were used for CH$_4$ and N$_2$O, respectively (Woodward et al., 2014). Manure type, week and their interactions were fixed effects in the mixed-effects models, and tank ID was used as a random effect. To satisfy the requirement for normality of residuals and variance homogeneity, for N$_2$O emissions, the residuals of the linear model with week as fixed effect were used as the dependent variable. For CH$_4$, NH$_3$ and CO$_2$ equivalents, the cubic root of the raw data was used as the dependent variable. Autocorrelation was taken into account in all models. Sidak was used in post-hoc comparisons. To compare treatment effects on gas emissions after field application, ANOVA was used followed by the Tukey-HSD test using R 4.0.3 software. Levene’s test was used to test for homogeneity of variance, and the Shapiro-Wilk test was used to test for normality. The level of significance was set to 0.05.

3. Results

3.1. Properties of organic fertilizers

Table 1 and Table S1 present the composition of organic fertilizers before and after storage. At the beginning of the storage period, dry matter (DM) and volatile solids (VS) contents in treatment DD were lower compared to treatment CA, while total N and NH$_4$-N concentrations were higher. DM and VS were further reduced in the LF treatment, whereas N concentrations were unchanged, indicating a selective removal of fibers that had low nitrogen content. During storage, the NH$_4$+TN and NH$_4$ content declined in DD and LF. Between the end of the storage experiment in November 2020 and the field application of organic fertilizers in April 2021, the trends of dry matter loss continued, and there was an increase in pH in all three treatments. Treatment NS had the lowest dry matter content, as well as NH$_4$ and total N content, while it had the highest ratio of NH$_4$-N/total N.

3.2. Temporal dynamics of emissions during storage

Transformations of C and N during storage were accompanied by gaseous emissions of CH$_4$, N$_2$O and NH$_3$. Treatment CA showed the highest CH$_4$ emissions which gradually increased and peaked after about 50 days of storage (Fig. 1a). CH$_4$ emissions from treatment DD were lower and dropped after the first month, while the emissions from treatment LF remained close to zero throughout the measurement period. The CH$_4$-C/(CH$_4$-C + CO$_2$-C) ratios showed a declining trend, varying between 11 % and 45 % (mean: 34 %) in treatment CA, between 0% and 23% (mean: 13%) in treatment DD, and between 0 % and 31 % (mean: 6 %) in treatment LF (Fig. 1b, CO$_2$-emission are in Fig. S3).

Treatment CA emitted the least NH$_3$ during most weekly periods while DD emitted the most (Fig. 1c); the largest difference in NH$_3$ emission between CA and other treatments occurred in the first week. Based on NH$_4$-N concentrations and pH of individual treatments, the concentrations of free NH$_3$ at the beginning of storage in treatments CA, DD and LF were 6, 63 and 80 mg NH$_3$ l$^{-1}$, respectively (Supplementary calculation). N$_2$O emissions during storage in all three treatments showed similar trends, with a decline during day 0–28 and a transient increase around day 30–50 (Fig. 1d).

3.3. Temporal dynamics of N$_2$O emissions after field application

From all treatments in the field experiment, N$_2$O emissions occurred mainly during the first 50 days (Fig. 2a). N$_2$O fertilizer stood out with extremely high emissions, and treatments CA, LF, DD, M0 and M100 were not significantly different (p > 0.05) (Fig. 3a). Even without including NS fertilizer, they were also not significantly different from each other (p > 0.05, results not shown). NS had the highest N$_2$O emission factor (EF) of 2.4 %, whereas treatments CA, LF and DD all had EF values lower than 1 %, and M100 had a negative EF since this treatment showed less N$_2$O emission than M0 (Fig. 3b). The largest emissions of N$_2$O from treatments CA, LF and DD occurred during day 28–42. Yield-scaled GHG emissions applied per 100 kg TN ha$^{-1}$ were lower in treatment LF than in CA, DD and LF during the storage and field monitoring periods (Table 2).

3.4. Soil mineral N dynamics and water content

Treatment NS and M100 had high NH$_4$+ levels during day 1–42 (Fig. 2b). Treatments CA, DD, and LF had comparable NH$_4$+ during the whole period and the NH$_4$-value fluctuated during day 1–25 and then diminished slowly. The soil NO$_3$ concentrations of treatment M100 during day 1–42 closely followed the NH$_4$+ trend, while there was a slower decline during day 28–60 (Fig. 2c). For all organic fertilizers, soil NO$_3$ concentrations remained below 20 μg N g$^{-1}$ during the whole monitoring period. There were no clear differences in soil gravimetric water content, which were averaged 20–22 % (corresponding to 50–55 % WFPS in this soil), with all manure-derived fertilizers during day 0–42 (Fig. 2d).

3.5. Cumulative GHG emissions during storage and after field application

Cumulative CH$_4$ emissions during storage from treatment CA were, respectively, 4 and 33 times larger than those from treatments DD and LF, whereas N$_2$O emissions were similar and NH$_3$ emissions were
4–5 times lower than from the digested fertilizers DD and LF (Table 2). The cumulative emissions of N₂O after field application were also similar in treatments CA, DD and LF (p > 0.05), whereas the cumulative N₂O emissions from the NS fertilizer were significantly higher. About 93 %, 94 % and 93 % of total N₂O emissions during storage and after field application came from the field for treatments CA, DD and LF, respectively (data not shown). For treatment CA, CH₄ emissions during storage constituted the main source of the overall GHG emissions during storage.

Fig. 1. Emissions of CH₄ (a), CH₄-C/(CH₄+C + CO₂-C) ratios (b), emissions of NH₃ (c) and emissions of N₂O (d) from untreated manure (CA), digestate of cattle manure co-digested with 7.5% grass-clover (DD), and the liquid fraction of digestate (LF) during storage. The error bars represent the standard deviation of the mean.

Fig. 2. N₂O emissions (a), concentration of NH₄ (b) and NO₃ (c), and soil water content (d) after fertilization with different N sources. CA: untreated cattle manure; DD: anaerobic digestate of cattle manure co-digested with 7.5% grass-clover; LF: liquid fraction of DD; NS: liquid fraction from biogas desulfurization filter enriched with sulfur and ammonium; M100: mineral fertilizer with an application rate of 100 kg TN ha⁻¹; M0: unfertilized control.
and after field application, and the N$_2$O emissions after field application constituted the second source (Fig. 4). In contrast, N$_2$O emissions after field application were more important for treatments DD and LF. Adding up all measured emissions, the anaerobically digested fertilizers in treatments CA had the highest, and treatment LF the lowest overall GHG emissions.

### 3.6. Spring barley yields

Treatments were not significantly different (p > 0.05) except that the yield of M0 was lower than those of all other treatments (Fig. 5). The nitrogen use efficiency (NUE) was 48 % for M100 and not different from NUE of organic fertilizers (p > 0.05). (Fig. S4). Only considering N$_2$O in the GHG calculation during the field period, treatment NS had the highest GHG emission per hkg of grain yield (p < 0.05), while CA, DD and LF were not significantly different (p > 0.05) (Table 2). NS also had higher yield-scaled N$_2$O emissions (0.58 g N$_2$O-N kg$^{-1}$ grain yield) compared to the other treatments (p < 0.05), which were not significantly different (p > 0.05) (Table 3). Considering both storage and field

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**Fig. 3.** Cumulative N$_2$O emissions (a) and N$_2$O emission factors (EF) (b) of the different fertilizers after field application. The letters show significant differences at p < 0.05. CA: untreated cattle manure; DD: anaerobic digestate of cattle manure co-digested with 7.5 % grass-clover; LF: liquid fraction of DD; NS: liquid fraction from biogas desulfurization filter enriched with sulfur and ammonium; M100: mineral fertilizer with an application rate of 100 kg TN ha$^{-1}$; M0: unfertilized control.

**Fig. 4.** Total GHG emissions from organic fertilizer treatments during storage and after field application. The application rate for these three treatments was approximately 100 kg TN ha$^{-1}$. CA: untreated cattle manure; DD: anaerobic digestate of cattle manure co-digested with 7.5 % grass-clover; LF: liquid fraction of DD.

**Fig. 5.** The grain dry matter yields of spring barley after the application of different fertilizers. The letters show significant differences at p < 0.05. CA: untreated cattle manure; DD: anaerobic digestate of cattle manure co-digested with 7.5% grass-clover; LF: liquid fraction of DD; NS: liquid fraction from biogas desulfurization filter enriched with sulfur and ammonium; M100: mineral fertilizer with an application rate of 100 kg TN ha$^{-1}$; M0: unfertilized control.

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**Table 2**

Cumulative emissions of CH$_4$, NH$_3$ and N$_2$O, and overall greenhouse gas (GHG) emissions during storage and after field application. Only N$_2$O was considered in the GHG calculation during the field period. Different letters within columns indicate a significant difference at p < 0.05. The grain yield indicates the dry matter (DM) of spring barley grain yield. CA: untreated cattle manure; DD: anaerobic digestate of cattle manure co-digested with 7.5% grass-clover; LF: liquid fraction of DD; NS: liquid fraction from biogas desulfurization filter enriched with sulfur and ammonium. The yield-scaled GHG emission indicated the total GHG kg$^{-1}$ grain DM yield during storage and field application, except that there was no measurement of emissions from NS during storage.

<table>
<thead>
<tr>
<th>Slurry type</th>
<th>Storage</th>
<th>Field application</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>CH$_4$</td>
<td>N$_2$O</td>
</tr>
<tr>
<td>CA</td>
<td>1249a</td>
<td>1.1a</td>
</tr>
<tr>
<td>DD</td>
<td>309ab</td>
<td>1.4a</td>
</tr>
<tr>
<td>LF</td>
<td>37.4b</td>
<td>1.2a</td>
</tr>
<tr>
<td>NS</td>
<td>–</td>
<td>–</td>
</tr>
</tbody>
</table>

$^1$GWP(N$_2$O) = 265; GWP(CH$_4$) = 28; NH$_3$ contributed as an indirect source of N$_2$O assuming an emission factor of 1 %.

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Table 3 Cumulative emissions of $N_2O$ per kg grain dry matter yield after field application to spring barley. Different letters within columns indicate a significant difference at $p < 0.05$. CA: untreated cattle manure; DD: anaerobic digestate of cattle manure co-digested with 7.5% grass-clover; LF: liquid fraction of DD; NS: liquid fraction from biogas desulfurization filter enriched with sulfur and ammonium; M0 and M100 indicate the mineral fertilizer with an application rate of 0 and 100 kg TN ha$^{-1}$, respectively.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>CA</th>
<th>DD</th>
<th>LF</th>
<th>NS</th>
<th>M0</th>
<th>M100</th>
</tr>
</thead>
<tbody>
<tr>
<td>$g N_2O-N \cdot kg^{-1} \cdot grain yield$</td>
<td>0.13b</td>
<td>0.21b</td>
<td>0.15b</td>
<td>0.58a</td>
<td>0.13b</td>
<td>0.05b</td>
</tr>
</tbody>
</table>

periods, the yield-rounded GHG emissions were 44.4, 17.1, 8.5 and 24.3 kg CO$_2$ eq hkg$^{-1}$ grain yield, respectively (Table 2).

4. Discussion

4.1. GHG and NH$_3$ emissions during storage

CH$_4$ emissions from untreated cattle manure (treatment CA) were on average 0.90 g CH$_4$m$^{-3}$ h$^{-1}$ during the 11-week autumn storage period, which is higher than the average emission of 0.58 g CH$_4$m$^{-3}$ h$^{-1}$ given by Kupper et al. (2020) in a literature review of results from pilot-scale and full-scale studies. Early autumn is the period where CH$_4$ emissions from liquid manure storage are typically highest (Maldanner et al., 2018), and this may explain the relatively high level observed. In accordance with this, the CH$_4$/C/(CH$_4$ + CO$_2$) ratio was initially around 0.4 in treatment CA, indicating a highly active methanogenic community.

Treatments DD and LF had much lower CH$_4$ emissions during storage corresponding to 25 % and 3 % of those from treatment CA. Methanogenesis depends on abundant labile C and neutral pH (El-Mashad et al., 2004). The reduction of VS content from digestate (treatment DD) compared to CA was 61 % (based on initial VS), which is comparable to the reduction of 66% reported by Maldanner et al. (2018) for full-scale storage tanks, although the CA and DD treatments in this study were derived from the anaerobic co-digestion of cattle slurry and 7.5 % grass-clover silage, which is not a widely used co-digestion substrate. The LF fertilizer was more directly comparable with the digestate of treatment DD as it was produced by decanter centrifugation of the digestate. Surprisingly, the liquid fraction of digestate (treatment LF) showed an 8-fold reduction of CH$_4$ emissions during storage compared to treatment DD although the VS content was only 20 % lower; presumably, the fraction of VS removed was important as a source of substrates for fermentation and methanogenesis. The average CH$_4$/C/(CH$_4$ + CO$_2$) ratios of 34 % and 13 % in untreated manure and digestate are in reasonable agreement with the values of 25 % and 10 % assumed in a recent analysis of biogas scenarios (Möller et al., 2022). Anaerobic digestion thus appears to be an effective method to reduce CH$_4$ emissions during manure storage, although it must be stressed that an effective digestion process and collection of biogas during the cooling phase have to be ensured (Sommer et al., 2000). The climate impact of separating the digestate depends on the management of the solids removed (Hansen et al., 2006).

Inhibition by ammonia potentially contributes to suppressing methanogenesis in manure during storage. Yenigün and Demirel (2013) reviewed ammonia inhibition in anaerobic digestion of organic wastes, and the potential roles of both total ammoniacal N (TAN) and free NH$_3$ were discussed. Varel et al. (1977) concluded that the production of CH$_4$ was inhibited when the liquid contained above 1700 mg total ammonium nitrogen (TAN) per liter and pH 7.8. Koster and Lettinga (1984) reported that acetotrophs were more negatively affected above 1700 mg TAN per liter, and it is noteworthy that Methanosaera, reported by Habteewold et al. (2018) to dominate CH$_4$ production in dairy cattle manure, is characterized by acetotrophic methanogenesis. The role of free NH$_3$ in manure is not clear; Yenigün and Demirel (2013) cited inhibitory effects with different organic wastes that ranged from 40 to several hundred mg l$^{-1}$ free NH$_3$. It is, therefore, possible that the concentrations of total ammoniacal N in treatments DD and LF at the time of storage, which were 2100 and 1900 mg TAN l$^{-1}$, respectively, or/and the concentrations of free NH$_3$, which were 63 and 80 mg l$^{-1}$, respectively, were involved in suppressing methanogenesis.

The emissions of N$_2$O during storage were comparable in treatments CA, DD and LF, and lower than the average rate of 0.002 g N$_2$O m$^{-3}$ h$^{-1}$ reported by Kupper et al. (2020). N$_2$O emissions during liquid manure storage depend on the N availability, oxygen content, pH, and redox status (Chadwick et al., 2011), and N$_2$O production occurs at the manure-air interface since nitrification requires oxygen. Reported effects of biogas treatment on N$_2$O emissions from stored digestate and untreated manure are not consistent; Sommer et al. (2000) observed higher N$_2$O emissions from digestate during storage compared to untreated manure, while other studies found no significant differences (Amon et al., 2006; Holly et al., 2017). N$_2$O emissions are mainly produced in well-developed surface crusts (Kupper et al., 2020), but in the present study, only treatment CA formed a thin crust, which indicates that the small amounts of N$_2$O observed came from the liquid phase.

NH$_3$ is not a greenhouse gas, but NH$_3$ emitted from manure is an indirect source of N$_2$O formed elsewhere following re-deposition. It is assumed that 1% of NH$_3$-N will be re-emitted as N$_2$O-N (Houghton et al., 1996). The pH in CA, DD and LF was 7, 7.8 and 7.9, respectively. Based on NH$_3$-N concentrations and pH of individual treatments, the concentrations of free NH$_3$ at the beginning of storage in treatments CA, DD and LF were 6, 63 and 80 mg NH$_3$ l$^{-1}$, respectively (see Supplementary information). Much higher free NH$_3$ in treatments DD and LF led to higher NH$_3$ emissions from treatments DD and LF than from treatment CA during storage. The emission of NH$_3$ is affected by many factors for example the homogeneity of manures, aeration, and NH$_3$ immobilization. The reason why DD emitted slightly more NH$_3$ than LF is unknown. Also, the thin crust forming in treatment CA may have delayed emissions of NH$_3$ (Baldé et al., 2018). These trends are consistent with the decreases observed in concentrations of TN, NH$_4$ and NH$_3$/TN of treatments DD and LF, but not CA, during storage. There is also a possibility that more organic N was mineralized during the storage of the cattle slurry compared to the digested fertilizers.

4.2. N$_2$O emissions after field application

With one exception (treatment NS), N$_2$O emissions during the growing season were moderate from both organic and mineral fertilizers and not significantly different from the unfertilized control. Half of the NS fertilizer was injected after plowing and therefore not disturbed by tillage, and this could be a reason why NS emitted much higher N$_2$O than other treatments. There was no evidence for stimulation of N$_2$O emissions in treatment M100 receiving 100 kg N ha$^{-1}$ mineral fertilizer, indicating that bulk soil conditions in this experiment did not support N$_2$O emissions even with the higher concentrations of NO$_3$ or NH$_4$. Li et al. (2015) investigated the drivers of N$_2$O emissions from the same soil by laboratory incubation experiments and found a positive interaction between CO$_2$ emissions (an indicator of O$_2$ demand in the soil) and concentration of NO$_3$, which suggested the availability of degradable organic matter in organic fertilizers was important. There was limited accumulation of soil NO$_3$ in treatments with organic fertilizers in this study, possibly because of effective plant uptake, and hence NO$_3$ availability may have been a limiting factor. Except for treatment NS, high N$_2$O emissions from other treatments occurred with rainfall in May 2021, at this period, oxygen depletion in the soil probably activated denitrifiers (Sezton et al., 1988). A similar response to rainfall was not observed in June 2021. However, the NH$_4$ and NO$_3$ content in the soil were low in all treatments by this time, indicating that soil NO$_3$ availability limited N$_2$O emissions. There were no clear differences in soil gravimetric water content, which averaged 20–22 % in soil samples from all manure-derived fertilizers during the first six weeks.
corresponding to 50–55 % WFPS in this soil (Li et al., 2015). A recent laboratory study concluded that the interactions between liquid organic fertilizers and the soil are critical for N₂O emissions (Meng et al., 2022), and greater redistribution of N and labile C with infiltrating water may trigger higher N₂O emissions.

Pools of NH₄⁺ and NO₃⁻ in treatments with mineral fertilizer remained at the same high level until the time when N uptake by spring barley was expected, indicating that dissolution of inorganic N and subsequent nitrification were slow. The content of soil NH₄⁻-N with organic fertilizers remained much lower than in soil with mineral fertilizer, the reasons could be microbial immobilization, plant uptake, or environmental losses. The crop yield of the control treatment M₀ was significantly lower than that of fertilized treatments (p < 0.05), but there were no significant differences in crop yields among fertilized treatments, thus environmental losses were generally independent of fertilizer type or composition. Nitrification has been considered the main source of N₂O emissions from the soil below 60% WFPS (Davidson, 1999), but Meng et al. (2022) used ¹⁵N to study sources of N₂O with the same organic fertilizers and soil as this study at 55% WFPS, and the results showed that denitrification was the main source of N₂O; thus coupled nitrification-denitrification may have contributed to N₂O emissions (Petersen et al., 1991). Since crop N uptake was similar to mineral and organic fertilizers, any N immobilization was probably re-mineralized during the spring period.

Except for treatment NS, the N₂O emission factors (EF) for the growing season were below 1 %, which is the default annual emission factor proposed by IPCC et al. (2006). This is consistent with previous studies on this soil type (Chirinda et al., 2010; Mutegei et al., 2010; Brozyňa et al., 2013). In a meta-analysis of N₂O emissions from organic fertilizers, Charles et al. (2017) reported the average EF was 1.12 ± 0.18 %, and the EF was 2.8 times higher in fine-textured compared to coarse-textured soils. Therefore, the sandy soil type in this study may explain the lower EF. In the present study, treatments CA, DD, and LF emitted similar amounts of N₂O, indicating that characteristics of the soils, i.e., environmental controls, were more important than the composition of the organic fertilizers as driver for N₂O emissions.

4.3. Yield-scaled emissions

The DM yield and NUE of spring barley in all fertilized treatments were similar. The fact that the NUE of treatment NS was similar to other fertilizer treatments indicates that the higher N₂O emissions were not an indicator of much higher total gaseous losses, but also that the NS fertilizer with this application did not have particular benefits over the other organic fertilizers despite a higher NH₄⁺-N/TN ratio.

4.4. GHG balances

Greenhouse gas balances were calculated and expressed as CO₂ equivalents (CO₂ eq); CO₂ itself was not considered due to the recent offset by the C fixation of crops used for cattle feed (IPCC et al., 2006). Anaerobic digestion with or without separation was effective to mitigate CH₄ emissions as well as total GHG emissions in this study, which was in accordance with Amon et al. (2006), who found that GHG emissions during storage and field periods were reduced by 60% by the digestion of cattle manure and the main contributor to this reduction was CH₄. Holly et al. (2017) found that anaerobic digestion and separation of dairy cattle slurry reduced total GHG emissions during storage and after field application by 25 % and 31 %, respectively, with most of the reduction coming from CH₄ emissions during storage. However, in their studies, N₂O emissions during the storage of digestate after separation increased and therefore did not result in further GHG reduction.

The monitoring of emissions during storage was terminated in mid-November, 2020. Methane emissions were close to zero by this time, but there were still low emissions of NH₃ (about 0–2 g N m⁻² d⁻¹) and N₂O (about 20 mg N m⁻² d⁻¹), and the total GHG emissions could therefore have been underestimated. However, Baral et al. (2018) in a study conducted using the same pilot-scale storage facility showed that CH₄ and other emissions from cattle slurry remained low from winter to April. Husted (1994) also, in an annual study of full-scale storage tanks in Western Denmark, found that CH₄ emissions from cattle slurry were negligible during winter and early spring. Both of these studies thus took place under the same climatic conditions as this study. N₂O emissions from stored manure highly depend on the temperature, and losses during winter were expected to be low (Sommer, 1997). N₂O emissions increased during early November, indicating some crust formation in the absence of rainfall (Sommer et al., 2000), but both NH₃ and N₂O emissions were expected to be low during winter as observed previously (Clemens et al., 2006; Petersen et al., 2013). The air temperature between mid-November and April was always below 10 °C and with occasional episodes of freezing temperatures, except that Clemens et al. (2006) did not specify temperature conditions.

From a system perspective, effective measures are required to curb the NH₃ and N₂O emissions from the solid fraction after separation (Hansen et al., 2006). Amon et al. (1999) reported that the solid fraction of the anaerobic digestate emitted more N₂O than compost. For treatment CA, CH₄ during storage was the main source of GHG emission, whereas, for treatments DD and LF, the main source of GHG was N₂O emissions after field application. This highlights that GHG mitigation efforts should consider all stages of manure management, and that manure treatment may change the emissions substantially.

In this study, CH₄ and NH₃ emissions after field application were not considered. CH₄ production in manure after field application is considered to be minor because the soil oxygen status does not support methanogenesis (Dammgen et al., 2012), and CH₄ released immediately after manure application is probably a result of degassing (Sommer et al., 2013). In one study on well-drained arable soil in Germany, CH₄ emissions were minor after manure application in spring, and the observed emission was believed to result from dissolved CH₄ (Wulf et al., 2002). NH₃ losses highly depend on the application method, it was reported that 20–40% of NH₃-N was lost after broadcast application (Thompson et al., 1990; Sanz et al., 2010). Following the legislation in Denmark, the organic fertilizers were surface applied and immediately incorporated, or injected; immediate incorporation is the most effective way to reduce NH₃ emissions and may achieve a 90% abatement of NH₃ emissions compared to a surface application (Sommer and Hutchings, 2001; Webb et al., 2013).

The yield-scaled N₂O emissions ranging from 0.13 to 0.21 g N₂O-N kg⁻¹ grain yield for the organic fertilizers did not show effects of manure treatment. The driving factors of N₂O after fertilizer application include the amount of easily degradable C, available N as well as the water content in soils. The effect of digestion on yield-scaled N₂O emissions in this experiment is in contrast to a previous study with the application of 100 kg NH₄-N ha⁻¹ on the same soil type (Baral et al., 2017); in that study the yield-scaled N₂O emissions were 0.26 and 0.04 g N₂O-N kg⁻¹ spring barley grain yield for cattle manure and digestate, respectively. In another experiment where spring barley was fertilized with inorganic fertilizers at a rate of 120 kg TN ha⁻¹, the yield-scaled N₂O emissions were about 0.4 and 0.3 g N₂O-N kg⁻¹ grain yield for ammonium nitrate or urea, respectively (Hinton et al., 2015). Considering both storage and field emissions, the yield-scaled GHG emissions were 44.4, 17.1, 8.5 and 24.3 kg CO₂ eq ha⁻¹ grain yield for treatments CA, DD, LF and NS, respectively, which was not surprising since the DM yield of spring barley in all fertilized treatments were similar.

5. Conclusions

Anaerobic digestion and separation were effective methods to mitigate total GHG emissions from storage and field application in this study, and the main effect was the reduction of CH₄ emissions during storage resulting from the lower degradability and content of residual
vulgaris solids and a pH above neutral in digestate. For untreated cattle slurry, the CH4 emitted during storage was the largest component of the total GHG emissions, while for treatments with digestate and a liquid fraction of the digestate, the N2O emissions after field application were the main source, and this difference should be considered in GHG mitigation strategies. The treatment with liquid manure from biogas desulphurization containing a proportion of ammonium and sulfur, but at low concentrations, had much higher N2O emissions than other treatments during the growing season, and the mechanism behind this should be investigated. The DM yields of processed organic fertilizers were similar, and yield-scaled GHG emissions were lower, compared to untreated cattle manure when considering emissions during storage and after field application. Therefore, we recommend anaerobic co-digestion of manure and grass with or without post-processing as a viable GHG mitigation strategy. Since anaerobic digestion also produces biogas, and post-processing requires energy, further environmental assessments are needed to evaluate the full potential of these organic fertilizers.

Declaration of Competing Interest

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

Data will be made available on request.

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Appendix A. Supporting information

Supplementary data associated with this article can be found in the online version at doi:10.1016/j.agee.2022.108327.

References


