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RESEARCH ARTICLE

Towards efficient N cycling in intensive maize: role of cover crops and application methods of digestate liquid fraction

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Abstract
Digestate, a by-product of biogas production, is widely recognized as a promising renewable nitrogen (N) source with high potential to replace synthetic fertilizers. Yet, inefficient digestate use can lead to pollutant N losses as ammonia (NH_3) volatilization, nitrous oxide (N_2O) emissions and nitrate (NO_3^-) leaching. Cover crops (CCs) may reduce some of these losses and recycle the N back into the soil after incorporation, but the effect on the N balance depends on the CC species. In a one-year field study, we tested two application methods (i.e., surface broadcasting, BDC; and shallow injection, INJ) of the liquid fraction of separated co-digested cattle slurry (digestate liquid fraction [DLF]), combined with different winter cover crop (CC) options (i.e., rye, white mustard or bare fallow), as starter fertilizer for maize. Later, side-dressing with urea was required to fulfil maize N-requirements. We tested treatment effects on yield, N-uptake, N-use efficiency parameters, and N-losses in the form of N_2O emissions and NO_3^- leaching. CC development and biomass production were strongly affected by their contrasting frost tolerance, with spring-regrowth for rye, while mustard was winter killed. After the CCs, injection of DLF increased N_2O emissions significantly compared with BDC (emission factor of 2.69% vs. 1.66%). Nitrous oxide emissions accounted for a small part (11%–13%) of the overall yield-scaled N losses (0.46–0.97 kg N Mg grain^{-1}). The adoption of CCs reduced fall NO_3^- leaching, being 51% and 64% lower for mustard and rye than under bare soil. In addition, rye reduced NO_3^- leaching during spring and summer after termination by promoting N immobilization, thus leading to −57% lower annual leaching losses compared with mustard. DLF application method modified N-loss pathways, but not the cumulative yield-scaled N losses. Overall, these insights contribute to inform an evidence-based design of cropping systems in which nutrients are recycled more efficiently.

KEYWORDS
cover crop residues, digestate broadcasting, digestate injection, N_2O emissions, NO_3^- leaching

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1 | INTRODUCTION

The new European Green Deal with the "Farm to Fork strategy" and the "Zero Pollution action plan" sets very ambitious environmental objectives for European agriculture (Ricci et al., 2022). These include a 20% reduction in the use of fertilizers and 50% abatement of nutrient losses by 2030, with a simultaneous restoration of the agroecosystem functions of soils and water bodies (European Commission, 2019). Among all nutrients, nitrogen (N) is simultaneously the main driver of agricultural productivity and the most often overused nutrient in agroecosystems, thus possibly leading to pollutant N losses in the form of nitrate (NO_3^-) leaching, nitrous oxide (N_2O) emissions and ammonia (NH_3) volatilization (Nieder & Benbi, 2022). These losses have negative cascading consequences for climate change, soil quality, and biodiversity (van Groenigen et al., 2015). Therefore, there is an urgent need for identifying solutions that could maximize N-use efficiency (NUE) and reduce N losses to meet the ambitious objectives of the EU's agricultural policy.

Cover crops (CCs) are promising tools to increase the environmental sustainability of an agro-ecosystem. Growing between the previous harvest and the subsequent sowing of primary cash crops, CCs are plant species with the main purpose of protecting the soil from erosion and reducing NO_3^- leaching during off-seasons (Thorup-Kristensen et al., 2003). Among common CC species, rye (*Secale cereale* L.) and white mustard (*Sinapis alba* L.) have been reported as effective N-scavengers (Blanco-Canqui, 2018), but their general effectiveness in lowering NO_3^- leaching losses may differ due to their different frost sensitivities, which also may affect the timing of CC termination and biomass decomposition (Brennan et al., 2013). In addition, CC effects on N_2O emissions may be highly variable due to complex interactions between climatic conditions (e.g., rain-evaporation regime), management practices (e.g., termination method and timing), and CC species, among others (Abdalla et al., 2019; Blanco-Canqui, 2018). For instance, CC residues with high C/N ratio can cause temporary N immobilization, thus limiting N availability for N_2O-producing microorganisms, but also for the following cash crop (Abalos, Rittl, et al., 2022; Boselli et al., 2020). Conversely, residues with low C/N ratio can decompose faster, thus boosting N availability not only for crop yields but also for N_2O emissions (Abalos, Recous, et al., 2022). Understanding this potential trade-off between yield and N losses is important to optimize the ecosystem services provided by CCs, but to date, field experiments have seldom studied these effects covering both the CC and cash crop phases.

Beyond the need to increase N use-efficiency at the agroecosystem level, environmental objectives within the Green Deal require increasing shares of renewable energy and energy efficiency (European Commission, 2019). Bioenergy generation via anaerobic digestion of livestock manure has gained remarkable momentum as a tool to obtain alternative energy from natural sources while improving waste management (Lamolinara et al., 2022). At the same time, the liquid fraction of anaerobic digestates may be easier to manage from a farmer's perspective and can partially or totally replace synthetic fertilizers due to its high inorganic-N content (Sigurnjak et al., 2017). However, digestate use can increase the risk of NO_3^- leaching and N-gaseous losses as NH_3 and N_2O (Möller, 2015). Documenting N losses from digestate in realistic field conditions is required to have a better picture of the overall environmental consequences of anaerobic digestion, and to inform evidence-based policymaking regarding the use of bioenergy by-products.

Adequate digestate application methods should be adopted to reduce N-losses after field application. Shallow injection can abate NH_3 volatilization compared with surface broadcast, providing correct closure of the furrow behind injection tines (Chadwick et al., 2011). However, the effectiveness of direct injection in reducing N_2O emissions compared with surface broadcast is contentious because the lower oxygen availability in deeper soil layers can stimulate N_2O production from denitrification (Chadwick et al., 2011). Similarly, NO_3^- leaching losses may be affected by specific soil conditions that could negatively interact with injection, such as shallow tile drains or shallow water tables (Dell et al., 2012). Application timing is another crucial step for valorising N from animal manures: splitting N fertilization during the growing season of summer crops with organic fertilizers (e.g., digestates), and synthetic ones, like urea, may be an effective way to attain good crop yields while reducing N losses (Martínez et al., 2017).

The objective of this study is to evaluate how to reduce N losses from intensive agro-ecosystems with different CC alternatives and application techniques of digestate liquid fraction (DLF). We hypothesized that (i) CCs help to consistently abate N-leaching losses during winter, compared with bare soil (BS); (ii) N losses differ between CCs with contrasting C/N ratios and frost tolerance; (iii) DLF injection can increase N_2O emissions and NO_3^- leaching compared with surface application, thereby reducing NUE; and (iv) frost-sensitive CCs do not reduce crop yields in digestate-based cropping systems because their residues do not induce digestate-N immobilization.

2 | MATERIALS AND METHODS

2.1 | Site and soil characteristics

The field experiment was performed as one-year trial between October 2018 and October 2019 at the CERZOO

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research station (45°00′17″ N, 9°42′21″ E; 71 m asl), Piacenza, in the Po valley region, Northern-Italy. Local climate is temperate, with mean annual temperature and cumulative precipitation of 14°C and 795 mm, respectively (1999–2019, averaged period). The soil is classified as a fine, mixed, mesic Udertic Haplustalf (Soil Survey Staff, 2014), with a silty clay texture (sand 127 g kg^{-1}, silt 445 g kg^{-1}, and clay 428 g kg^{-1}) in the upper layer (0–30 cm). At the beginning of the experiment, the soil's main physical and chemical properties were as follows: pH_{H2O} 7.00; organic matter content (Walkley-Black) 30.04 g kg^{-1}; total N (Kjeldahl) 1.74 g kg^{-1}; C/N ratio 10; bulk density (soil-core method) 1.30 g cm^{-3} at 0–10 cm depth; available P (Olsen) 32 mg kg^{-1}; exchangeable K (Ba chloride, pH 8.1) 294 mg kg^{-1}; cation exchange capacity (Ba chloride, pH 8.1) 30 cmol^{+} kg^{-1}. An automated meteorological station (Pessl Instruments, GmbH) was located at the experimental field to record climatic data.

2.2 | Experimental design, treatment, and crop management

The experiment was designed as a split-plot with three replicates (blocks). Main factor plots (360 m^2: 30 m length by 12 m width) consisted in the CC preceding maize, with three levels: rye (R, *Secale cereale* L., cv. *Primizia*), white mustard (M, *Sinapis alba* L., cv. *Asta*), and BS. The secondary factor (split factor) was the DLF application method, with three levels: surface broadcast (BDC), shallow injection (INJ) and a non-fertilized control. The subplot size was 120 m^2 (30 m length by 4 m width).

Cover crops were drilled right after a shallow tillage operation (15-cm disc harrowing to incorporate residues of the previous maize crop) on October 6, 2018, at a seeding rate of 150 kg ha^{-1} for R and 25 kg ha^{-1} for M, with spacing rows of 0.17 m. CCs were terminated by glyphosate [N-(phosphonomethyl) glycine] application (2.4 L a.i. ha^{-1}) on 19 March 2019; at that time, M was almost decomposed, being previously frost-killed, while R was still growing. This is a common pattern in the Po Valley and in several agricultural regions under temperate and cold climates. Glyphosate was sprayed also on BS plots and field edges to suppress weeds and to guarantee a similar soil compaction level to all plots.

No soil tillage was adopted, thus allowing for CC decomposition onto the soil surface. The amount of residual CC biomass was quantified from 10 m^2 for each plot just before weeding. Subsamples were oven-dried at 65°C for dry matter determination and then analysed for C and N concentration with an elemental analyser (vario MAX CNS, Elementar Analysensysteme GmbH). No fertilizer was applied during the winter CC season.

Digestate liquid fraction application occurred on April 9, 2019, after CC termination for both BDC and INJ, 10 days before maize sowing. DLF was obtained from a nearby farm as the liquid fraction (after mechanical separation) of co-fermented cattle slurry (about 60% in volume) and silage maize (about 40% in volume) for anaerobic methane production. Prior to distribution, DLF was analysed for main physical and chemical characteristics (Table S1). The N concentration was used to calculate an application rate of 50 Mg fresh weight ha^{-1}, to supply 170 kg N ha^{-1} (approximately 80 kg of total ammoniacal N ha^{-1}). For DLF injection, a slurry tanker (12 m^3 capacity) was equipped in the rear with 14 cutting wheels and drop pipes spaced 0.3 m apart, which cut the soil into narrow slots up to approximately 0.1 m depth and allowed DLF to settle in. For BDC, the cutting wheels were retained above the soil and a wooden board was mounted below them to homogenize the application of DLF over the entire soil surface, thus simulating a broadcast application.

Maize (cv. LG 31630) was directly seeded on untilled soil on April 19, 2019, at a rate of 85,000 seeds ha^{-1} with 0.75 m spacing between rows in each plot. With the exclusion of unfertilized control plots, to meet typical N-requirements for maize-grain production, 140 kg N ha^{-1} were additionally supplied as urea (U, 46% N) by side-dressing (0–10 cm from the row) at the growth stage V5-V6 (Leaf collar method, Begcy & Dresselhaus, 2017) on June 12, 2019. After U application, 20 mm sprinkler irrigation was applied following good practice to minimize NH_3 volatilization (Holcomb et al., 2011). Four other sprinkler irrigations for a total amount of 180 mm occurred between June 12 and August 11, 2019 (Figure S1). The water doses to be applied were estimated from the crop evapotranspiration (ETc) of the previous week (net water requirements). Daily ETc was calculated as ETc = ETo × Kc (mm day^{-1}), given ETo as the evapotranspiration of reference crop, derived with the FAO Penman-Monteith formula (Allen et al., 1998) on daily weather data from the field meteorological station. The crop coefficient (Kc) for maize under our specific climatic conditions was obtained from Facchi et al. (2013). Irrigation requirements were then calculated on a weekly basis by subtracting precipitation from ETc. Considering both irrigation and rain, the maize crop received about 540–550 mm of water from seeding to harvest.

For data reporting and analysis, three different periods were considered: "Winter period" (from CCs sowing to DLF application, lasting 153 days), "DLF period" (from DLF to U application, 64 days), and "Urea period" (from U fertilization to the end of the experiment, 134 days).

2.3 | Maize yield and N-efficiency parameters

Maize was harvested for grain on October 4, 2019, 169 days after seeding. Maize plants were harvested by hand from a

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15 m^2 area for each one of the 27 subplots. Fresh weight of ears and stover from the sampling area was directly measured in the field with a hand dynamometer. The moisture content of stoves, husks and cobs was determined from 10 plant subsamples by oven drying at 105°C until constant weight. From the same plants, the grain was collected, weighed, and dried at 105°C to calculate the Harvest Index (grain yield over total biomass, %). Grain yield was corrected to 14% moisture content. All parts were subsequently analysed for total N content (Kjeldhal method).

The following N-efficiency parameters were calculated (Weih et al., 2018): (i) NUE ($kg\,kg^{-1}$) was defined as the ratio of grain yield to total N supply, both from fertilizers (DLF and U) and soil (N_{soil}). N_{soil} accounted for inorganic N at planting ($N_{ini} = NO_3^-\text{-N} + NH_4^+\text{-N}$) and N derived from organic matter mineralization (N_{min}). N_{min} was estimated on control plots (Martínez et al., 2017) with the following equation:

$$N_{min} = N_{res} + N_{plant} - N_{ini}, \quad (1)$$

where N_{plant} is the sum of N uptake in maize grain and biomass, and N_{res} is residual soil mineral N ($NO_3^-\text{-N}+NH_4^+\text{-N}$) after maize harvest; (ii) nitrogen uptake efficiency (NUpE; $kg\,kg^{-1}$), as the ratio of N_{plant} to total N supply; and (iii) nitrogen utilization efficiency (NUtE; $kg\,kg^{-1}$), as the ratio of grain yield to N_{plant}.

2.4 | Nitrous oxide emissions

During the whole monitoring campaign, N_2O emissions were measured through the static chamber method (Venterea et al., 2020). A total of 27 sampling points (one per plot) were defined. A steel collar (0.45 m diameter and 0.20 m height) was deployed 5 cm into the soil. During the "Winter period," measurements were done above two CC rows, while measurements during the "DLF" and "Urea" periods were taken between maize rows. The rings were removed only for mechanical operations and then returned to the same place.

A 0.06 m^3 PVC chamber was mounted on the ring at the measurement time and the steel collar between these two units was water filled to ensure a tight seal. Then the measurement chamber was connected to a photoacoustic gas analyser (Innova 1412 Photoacoustic Multigas Monitor, LumaSense Technologies A/S) by mean of two Teflon tubes (3 mm inner diameter), one for the inlet and one for the outlet: this system avoids an artificial over-concentration of gases in the measurement chamber which, otherwise, would result biased. A 12 V fan was mounted inside the chamber to ensure adequate air mixing and avoid gas stratification in the measured head space. For each chamber, four samplings were taken, namely at chamber closure (time 0) and then after 7, 14, and 21 min. Since the gas monitor was set to give concentration values (mg $N_2O\text{-N}\,m^{-3}$ air) according to standardized values of 20°C and 1 atm, additional correction was required to tune results to field-measured values of air temperature and pressure.

Seven gas measurements were done from CCs seeding until DLF application: 31, 54, 72, 101, 135, 159, and 184 days after CC planting. Nine samplings were done after DLF application: on the same day of DLF distribution, to capture sudden N_2O peaks that may occur within few hours (Krol et al., 2015), and then after 3, 5, 7, 20, 30, 35, 50, and 62 days. Similarly, nine samplings were done after U application: 1 day after fertilization and then after 8, 12, 26, 44, 62, 79, 83, and 103 days. Considering the background measurement, carried out the day before fertilization events, 10 measurements were realized for the "DLF period" (April 8–June 11, 2019) and 10 for the "Urea period" (June 11–October 23, 2019).

Surface soil temperature was measured with a hand data logger (Elitech®, RC-5+ PDF Temperature Data Logger, Therm La Mode) deployed inside the chambers at measuring time. Atmospheric pressure was recorded from a nearby (5 km) weather station. Measurements generally took place between 9:00 and 12:00, to sample when conditions represented mean daily temperatures. Daily N_2O-N fluxes ($g\,ha^{-1}\,day^{-1}$) were derived from linear regressions of concentration data ($R^2 > 0.90$) at the fixed sampling intervals after the chamber closure. If the linearity assumption was not met at the last sampling point, the fourth measurement time (21 min) was removed to obtain linear regression for all events (Weidhuner et al., 2022). Cumulative emissions (kg $N_2O\text{-N}\,ha^{-1}$) were obtained by trapezoidal integration of consecutive monitoring events (Maris et al., 2015). The direct N_2O emission factor (EF) was calculated according to IPCC guidelines (Liang & Noble, 2019) by subtracting the unfertilized control N_2O-N emissions from those of the fertilized plots and then dividing by the amount of applied N as fertilizer.

Yield-scaled N losses were obtained from the sum of all the various N losses during the monitoring period (cumulative N_2O-N, cumulative NH_3-N volatilization and cumulative NO_3^--N leaching from April to October), divided by the yield of the maize crop.

2.5 | Nitrate leaching

Nitrate-N in the soil solution was monitored during the experiment through ceramic cups, one for every elemental plot, placed at 0.45 m depth with an angle degree of 45° to avoid preferential flow pathways. The monitoring

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period took place between November 2018 and September 2019: sampling dates were decided according to expected irrigations and rainy events. A vacuum of −60 kPa was applied in advance, then soil water was extracted 24 h after the precipitation event using a 100-mL syringe and a long thin tube (2 mm inner diameter) which was inserted into the suction cup. The soil solution was then carried to the laboratory into 100 mL PE flasks and kept frost until analysis.

The NO_3^--N concentration ($mg\,L^{-1}$) in the leachates was analysed with dual wavelength UV spectroscopy (275, 220 nm), after samples acidification with 1 M HCl. Ammonium (NH_4^+) concentrations were measured through an improved version of the colorimetric Berthelot reaction (Rhine et al., 1998). All analyses were performed in 96-well microplates with a Biotek Synergy 2—spectroscopy apparatus. However, given that the NH_4^+-N concentration was very low (<0.5 $mg\,L^{-1}$) throughout the monitoring period, it was considered negligible.

Cumulative NO_3^--N leaching losses ($kg\,N\ ha^{-1}$) were calculated using the trapezoidal rule (Perego et al., 2012) as follows:

$$\text{N leached} = \frac{0.5 \cdot (c1 + c2)}{100} \cdot v, \quad (2)$$

where c_1 and c_2 are concentration values (mg NO_3^--N L^{-1}) from two consecutive samplings, and v is the water drainage amount (mm) between these occasions. This value was obtained through the application of the soil water atmosphere plant (SWAP) model, which demonstrated a good performance in similar environments of the Po Valley, with fine textured and clayey soils (Maris, Abalos, et al., 2021; Perego et al., 2012). A description of SWAP model simulations and calibration with parameters used is presented in Supporting Information (Table S4).

2.6 | Soil parameters

Soil samples were taken along the experimental season to monitor water-filled pore space (WFPS) and mineral N content (NO_3^--N + NH_4^+-N). Specifically, WFPS (%) was derived from the formula:

$$\text{WFPS} = \frac{\text{GWC} \cdot \text{BD}}{1 - (\text{BD}/2.65)} \cdot 100, \quad (3)$$

where GWC is gravimetric water content ($g\,g^{-1}$), determined over the 0–10 cm soil layer by drying undisturbed cores at 105°C until constant weight; BD is soil bulk density ($g\,cm^{-3}$) and 2.65 ($g\,cm^{-3}$) is the assumed particle density, according to Danielson and Sutherland (1986).

Soil samples were taken by means of a hand auger in the layers 0–10 and 10–30 cm depth for each plot. Samples were then kept frozen until analysis. At the time of analysis, soil mineral N was extracted by shaking 5 g of homogeneously mixed soil with 20 mL of K_2SO_4 (0.05 M) for 2 h and then filtered on Whatman paper n. 42. The extract was analysed for NO_3^--N and NH_4^+-N with the same methods as for the leachate samples from the suction cups.

Both WFPS, NO_3^-, and NH_4^+-N values are reported in Supporting Information. While WFPS was expressed on a daily basis, mean NO_3^- and NH_4^+-N values were averaged over the different monitoring periods: "Winter," "DLF," and "Urea."

2.7 | Ammonia volatilization

Ammonia volatilization results during the experiment have already been reported by Maris, Capra, et al. (2021). Here, we use the NH_3 data presented in that article in combination with the N_2O and NO_3^- leaching data reported here to compute cumulative N-losses and yield-scaled cumulative N losses. NH_3 volatilization was monitored using the semi-open static chamber method. Briefly, chambers were made of a PVC pipe (30 cm long, diameter of 20 cm) open at two ends. At monitoring time, after each fertilization event, that is, after DLF application before sowing (April 9–23, 2019, for a total of 335 h), and at tillering (from June 12 to September 19, 2019, for a total of 150 h) after U application, chambers were inserted 5 cm below the soil surface and equipped with two polyfoam discs of 20 cm diameter each. One was placed 10 cm above the soil surface, and the other at the open end of the chamber. The first one captured NH_3 volatilization from the soil, while the second was used to prevent contamination from ambient air. To trap ammonia volatilization, the two foams were previously soaked in 80 mL of an oxalic acid-acetone solution (3% w/v). Ammonia volatilization was calculated by subtracting the threshold level of the unfertilized control from the fertilized plots. Cumulative NH_3 volatilization (kg NH_3-N ha^{-1}) was then obtained by simple addition of sampling events. Further details on sampling methodologies and quantification techniques can be found in Maris, Capra, et al. (2021).

Since static chambers have been pointed as unreliable to derive conclusions about absolute NH_3-N losses, calibration is required (Alexander et al., 2021). Starting from NH_3 data derived by chambers, we calculated NH_3 emissions by multiplying values from chamber measurements to specific correction indices accounting for underestimation: those were determined according to "NH_3 source solution methodology" (Alexander et al., 2021; Yang

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et al., 2019), and subsequently fine-tuned against data coming from wind-tunnels (three were present at DLF application time).

2.8 | Statistical analysis

Statistical analyses were carried out with R 4.0.5 (R Core Team, 2020).

The significance of the effect of CC type, DLF application method, and their interaction on NO_3^- leaching, soil characteristics, N_2O emissions, maize production parameters, and N uptake were evaluated with analysis of variance (ANOVA) with a *p*-value of 0.05 as threshold. To do this, the experimental design was implemented in a linear mixed-effect model with "nlme" package (Pinheiro et al., 2021), with "blocks" as random factor. Data were checked for normality (normal QQ-plot and Shapiro–Wilk's test) and for homogeneity of variances (residuals vs. fitted values graph and Levene's test) prior to analysis. When required, data were log-transformed to meet normality and homoscedasticity assumptions. Significant differences between the means of various treatments were further separated through a Tukey's HSD test ($p<0.05$) with the "multcomp" package (Hothorn et al., 2008).

Principal component analysis (PCA) was computed using the "FactoMineR" package (Lê et al., 2008), while "factoextra" (Kassambara & Mundt, 2020) was used for graphical biplot representation (Figure 3a–c), reporting main relationships between soil variables (WFPS, NO_3^--N and NH_4^+-N at 0–10 cm [up] and 10–30 cm [deep]), N-loss pathways (cumulative N_2O-N emissions, NH_3-N volatilization and NO_3^--N leaching) and treatments, represented for the main (BS, M, R) and secondary factors (C, BDC, INJ).

3 | RESULTS

3.1 | Environmental conditions, water-filled pore space, and soil-available N pools

During the experiment, the average daily air temperature was 13.9°C, ranging from −1.8 to 29.8°C, and cumulative precipitation was 835 mm, 40 mm more than the 20-year mean annual precipitation (Figure S1).

Water-filled pore space ranged from 46% to 87% in the 0–10 cm soil layer and was generally higher than 60% (Figure S2a–c), with values lower than 60% only on three sampling dates—March 14, June 20, and August 30, 2019.

The average NO_3^--N concentration in the 0–10 cm soil layer (Table S2a) was not affected by the CCs during the "Winter period." During the "DLF" and "Urea" periods (and on an annual basis), there was a significant CC × DLF interaction, but with different patterns between the two periods. On an annual basis, for both BS and M, the highest concentration was measured with BDC, whereas the highest concentration in R occurred with INJ. Over the 10–30 cm layer (Table S2b), a significant CC × DLF interaction was found on an annual basis and concentrations were generally lower than in the uppermost layer.

The concentration of NH_4^+-N in the 0–10 cm layer (Table S3a) was significantly affected by a CC × DIG interaction during the "DLF period," and by DLF on an annual basis. A similar pattern was found at 10–30 cm depth during the "DLF period," although the differences between treatments were smaller (Table S3b).

3.2 | Cover crop residual biomass and C and N content

Since M was frost-killed long before R chemical termination in March, its residual biomass (Table 1) was much lower than that of R. Nitrogen concentration was also higher for R (17.1 g N kg^{-1}) than for M (11.8 g N kg^{-1}). The C/N ratio was lower for R than for M. The N input of R was 105.2 kg ha^{-1}, significantly higher than that of M (16.6 kg ha^{-1}). Carbon input with R accounted for almost 3.0 Mg C ha^{-1}, while that with M for 0.6 Mg ha^{-1} only.

3.3 | Maize grain yield, vegetative biomass, and N-use efficiency parameters

Cover crops tended (p = 0.089) to negatively affect maize grain yield and total N uptake, in the following order:

TABLE 1 Cover crop (CC) residual biomass, N and C concentration, C/N ratio, and N and C supplied through their residue after spring termination. Values are expressed on a dry matter basis. Values are means ± standard deviations.

Cover crop, CC	Residual biomass (Mg ha^{-1})	N concentration (g kg^{-1})	C concentration (g kg^{-1})	C/N ratio	N input (kg ha^{-1})	C input (kg ha^{-1})
Rye	6.2 ± 0.3 a	17.1 ± 1.5 a	481.3 ± 17.0	28.2 ± 1.9 b	105.2 ± 8.5 a	2961.3 ± 201.4 a
Mustard	1.4 ± 0.1 b	11.8 ± 0.1 b	460.3 ± 2.6	39.0 ± 0.2 a	16.6 ± 0.7 b	647.2 ± 30.5 b
p (*t*)	<0.0001	0.0018	0.0514	0.0003	<0.0001	<0.0001

Note: Within columns, means followed by different letters are statistically different according to Student's *t*-test (α = 0.05).

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BS > M > R. On the other hand, maize grain yield, vegetative biomass, and total N uptake were all positively affected by DLF and U application, regardless of the method and the CC species (Table 2). The N efficiency parameters showed the opposite pattern (Table 2), with higher values under unfertilized control than under BDC and INJ. No CC × DLF interactions were found.

3.4 | Nitrous oxide emissions and emission factors

The N_2O-N fluxes are reported in Figure 1a–c, and cumulative emissions and EFs are given in Table 3. The N_2O-fluxes during the "Winter period" were low for all treatments (between −30 and 30 g N_2O-N ha^{-1} day^{-1}). Cumulative N_2O-N emissions were negative under both CCs, and positive under BS, with a significant difference only between R and BS (Table 3).

The first N_2O peak was measured during the "DLF period" for both BDC and INJ on the same day of DLF application, and was higher in INJ than BDC for all CC treatments (i.e., by 18%, 36%, and 45% for M, BS, and R, respectively); 2–3 days later, a second lower peak under INJ arose. Cumulative N_2O-N emissions decreased in the order control < BDC < INJ, without CC × DLF interactions.

A second "double-peak" N_2O pattern was recorded during the "Urea period" for both M and R, but not under BS which showed a linear increase in N_2O flux from June 13 to 24 (Figure 1a–c). Cumulative N_2O-N emissions for this period did not evidence any significant difference between BDC and INJ, with 6.3 and 7.3 kg N_2O-N ha^{-1} emitted, respectively, and no interaction between the main and secondary factors (Table 3).

Annual cumulative emissions ranged from 3.5 to 15.6 kg N_2O-N ha^{-1} and were significantly affected by the CC × DLF interaction: under both M and R, the ranking was control < BDC < INJ, but without significant differences between INJ and BDC under BS (Table 3).

The N_2O-N EF based on annual N inputs ranged between 1.1% and 3.6% of applied N (Table 3) and was significantly affected by CC and DLF, with both CCs being higher than BS but not different from each other. Injection almost doubled the EF from BDC (2.7% vs. 1.7%, respectively) and no CC × DLF interaction was observed.

3.5 | Nitrate leaching

Monthly-averaged NO_3^--N leaching (kg NO_3^--N ha^{-1}) and water input (rainfall plus irrigation supply) showed a similar pattern for all treatments (Figure 2a–c): the first NO_3^--N leaching peak was measured in November after large precipitation events (105.8 mm), then the period between December and March had lower NO_3^--N leaching due to a 49% lower precipitation compared with the average value during the 1991–2019 period. Afterward, NO_3^--N leaching increased again during April, May, and June, with a second peak. In September, NO_3^--N leaching increased under all treatments with DLF + urea application at a variable slope, ranging from the highest in BS to the lowest in R.

Cumulative NO_3^--N leaching was significantly affected by CC and by DLF in all periods, while the interaction CC × DLF was never significant (Table 4). In detail, BS had the highest values during the "Winter period," being 103% and 180% higher than M and R, respectively. Cumulative losses during the "DLF" and "Urea" periods were also the largest for BS, while R had always the lowest losses. Both BDC and INJ had higher cumulative NO_3^--N leaching compared with control in both periods. On an annual basis, cumulative NO_3^--N leaching losses were higher under BS than under R, while losses from M did not differ from either treatment. Both BDC and INJ increased cumulative NO_3^--N leaching compared with the control by 49% and 61%, respectively.

3.6 | Yield-scaled N-losses

Cumulative and yield-scaled N-losses were not affected by CC × DLF interactions and were the same for BDC and INJ (Table 5).

Yield-scaled NH_3-N losses were affected by CCs and DLF application method. In detail, (i) BS had lower NH_3-N losses than R, while M was in between and (ii) both BDC and INJ increased NH_3-N losses compared with control with BDC seeming to boost values compared with INJ.

Yield-scaled N_2O-N losses, which accounted for around 11%, 12%, and 13% of total yield-scaled N losses for R, BS, and M respectively, showed no difference between CCs; DLF application increased losses for both BDC and INJ compared with the unfertilized control.

Yield-scaled NO_3^--N leaching was affected by CCs, with both R and M reducing the BS value, with no CC × DLF interactions.

3.7 | Nitrogen loss pathways and relationships with environmental variables

Figure 3a–c shows a PCA for the different periods. For the "Winter period" (Figure 3a), the two principal components explained 72.3% of the variance: here a negative association was found between soil N-loss pathways

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TABLE 2 Maize grain yield, stalk biomass production, total N uptake, and N uptake efficiency (NUpE), N utilization efficiency (NUtE) and N-use efficiency (NUE) parameters (see a more detailed description of these parameters in the main text). Values are expressed on a dry matter basis. Values are means ± standard deviations.

Source of variation	Cover crop, CC	DLF application method	Grain yield ($Mg\,ha^{-1}$)	Stalk biomass ($Mg\,ha^{-1}$)	Total N uptake ($kg\,ha^{-1}$)	NUpE ($kg\,kg^{-1}$)	NUtE ($kg\,kg^{-1}$)	NUE ($kg\,kg^{-1}$)
Main factor, CC	BS	—	14.1 ± 2.8	12.3 ± 2.1	283.1 ± 77.1	0.7 ± 0.2	50.4 ± 4.4	37.1 ± 11.5
	M	—	13.3 ± 3.4	11.6 ± 2.5	263.2 ± 85.5	0.7 ± 0.1	52.2 ± 6.4	39.2 ± 12.2
	R	—	12.4 ± 3.7	11.3 ± 2.5	250.0 ± 96.3	0.7 ± 0.2	51.7 ± 6.8	39.4 ± 12.7
	p (*F*)		0.0889	0.3031	0.0973	0.7774	0.4595	0.3238
Secondary factor, DLF	—	C	9.2 ± 1.7 b	9.4 ± 1.7 b	158.1 ± 33.2 b	0.9 ± 0.0 a	58.6 ± 3.8 a	54.3 ± 3.4 a
	—	BDC	14.8 ± 1.2 a	12.6 ± 1.4 a	309.7 ± 32.0 a	0.6 ± 0.1 b	47.5 ± 1.6 b	29.7 ± 2.6 b
	—	INJ	15.8 ± 1.4 a	13.2 ± 1.9 a	328.5 ± 37.4 a	0.7 ± 0.1 b	48.3 ± 2.5 b	31.7 ± 2.4 b
	p (*F*)		<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
CC × DLF	BS	C	10.7 ± 0.8	11.3 ± 1.1	193.6 ± 25.8	0.9 ± 0.0	55.6 ± 3.1	52.2 ± 2.5
		BDC	15.3 ± 0.3	11.7 ± 1.1	308.0 ± 22.8	0.6 ± 0.0	48.2 ± 1.3	28.6 ± 1.2
		INJ	16.4 ± 2.0	13.9 ± 3.1	347.6 ± 58.3	0.6 ± 0.1	47.4 ± 2.6	30.6 ± 2.8
	M	C	9.1 ± 1.7	8.7 ± 1.3	151.3 ± 22.2	0.9 ± 0.0	59.9 ± 2.7	55.2 ± 3.2
		BDC	14.9 ± 1.0	13.3 ± 1.1	319.5 ± 18.4	0.6 ± 0.0	46.6 ± 1.6	30.2 ± 2.5
		INJ	15.9 ± 0.7	12.8 ± 1.5	318.7 ± 14.4	0.6 ± 0.0	50.0 ± 3.2	32.2 ± 0.4
	R	C	7.8 ± 1.1	8.3 ± 1.0	129.6 ± 8.9	0.9 ± 0.0	60.2 ± 4.6	55.6 ± 4.5
		BDC	14.3 ± 2.0	12.8 ± 1.7	301.5 ± 54.7	0.6 ± 0.1	47.6 ± 2.0	30.4 ± 4.2
		INJ	15.1 ± 1.4	12.7 ± 1.1	319.0 ± 33.9	0.7 ± 0.1	47.5 ± 1.2	32.2 ± 3.5
	p (*F*)		0.7023	0.0517	0.4919	0.7498	0.1833	0.984

Note: Within columns, means followed by the same letter are not significantly different according to Tukey's HSD test ($\alpha = 0.05$).

Abbreviation: BDC, DLF surface broadcast; BS, bare soil; C, control; DLF, digestate liquid fraction; INJ, shallow injection; M, mustard; R, rye.

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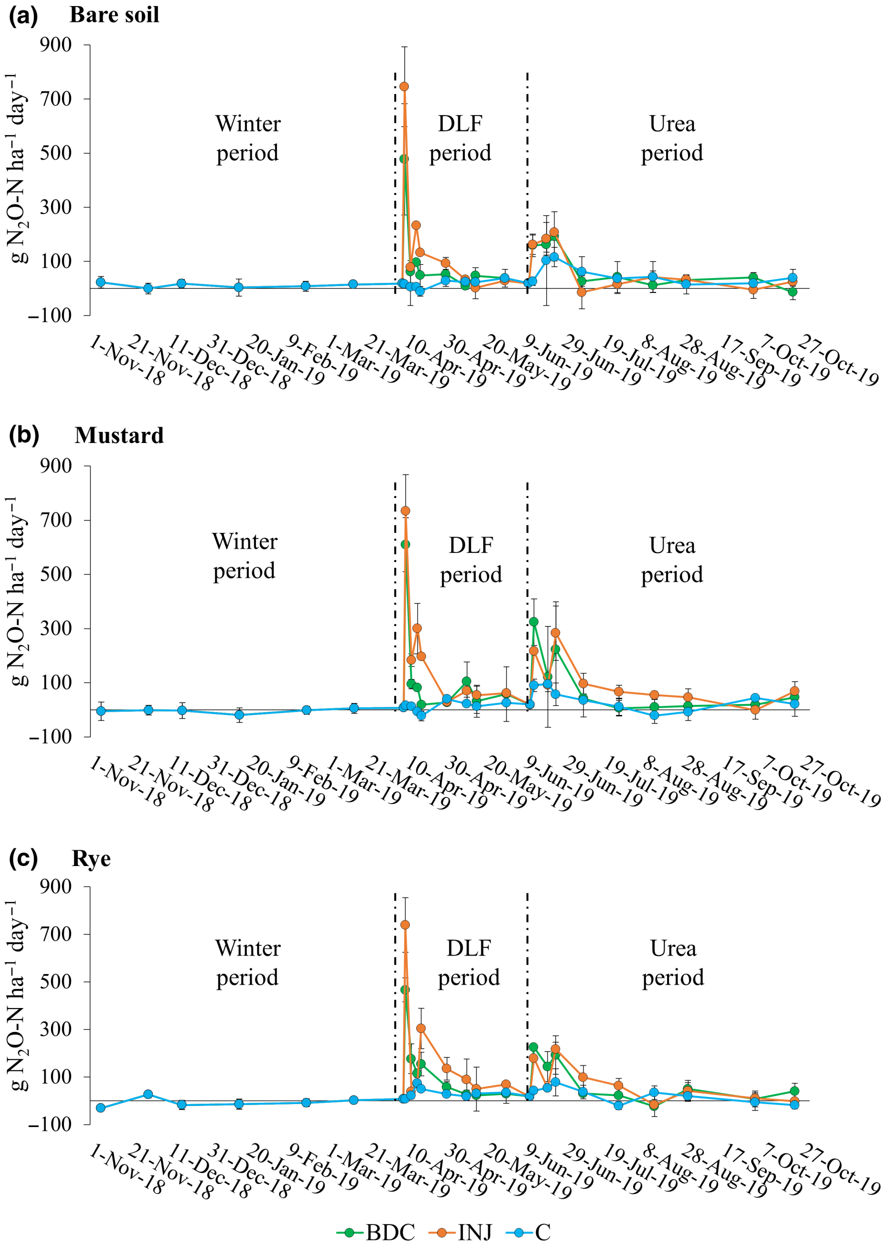

FIGURE 1 Dynamics of daily N_2O-N fluxes (g ha^{-1} day^{-1}) for bare soil (a), mustard (b), and rye (c) along the winter monitoring period, and after digestate liquid fraction (DLF) and urea fertilization. Treatment abbreviations refer to control (C), DLF surface broadcast (BDC) and shallow injection (INJ).

(N_2O-N emissions and NO_3^--N leaching) and CC biomass, especially for R.

The two first principal components for the "DLF monitoring period" (Figure 3b) accounted for 58.8% of the variance. The strongest link was found between N_2O emissions and NH_3 volatilization. These N losses were weakly related to NO_3^--N leaching, which was more closely related to soil NO_3^--N and NH_4^+-N content in the subsoil layer.

The relationships during the "Urea monitoring period" (Figure 3c) showed a similar pattern to that observed during the "DLF period" for N-gaseous losses, but with WFPS negatively related to N_2O emissions and NH_3 volatilization.

4 | DISCUSSION

4.1 | Cover crop biomass production and residual C and N content

Our results illustrate the importance of whether a CC is winter-killed or not for the residue decomposition in spring (at the sampling time). Previous studies reported that white mustard usually reaches its peak biomass before the winter season starts (Brennan & Boyd, 2012), which was also observed in our study. Conversely, rye continues to grow in early spring, thus explaining the higher dry biomass production—as well as higher residue-derived C and N inputs—of this CC. The dry biomass production of rye

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TABLE 3 Cumulative N_2O-N emissions for the different monitoring periods and on an annual basis, and N_2O-N emission factors (EFs) for annual emissions. Values are means ± standard deviations.

Source of variation	Cover crop, CC	DLF application method	Winter period cumulative N_2O-N emissions (kg ha^{-1})	DLF period cumulative N_2O-N emissions (kg ha^{-1})	Urea period cumulative N_2O-N emissions (kg ha^{-1})	Annual cumulative N_2O-N emissions (kg ha^{-1})	N_2O-N EF for annual emissions (%)
Main factor, CC	BS	—	1.6 ± 0.3 a	3.4 ± 1.7	5.6 ± 1.9	10.6 ± 3.1	1.2 ± 0.5 b
	M	—	−0.6 ± 0.9 ab	4.0 ± 2.6	6.4 ± 3.1	9.7 ± 5.2	2.8 ± 1.1 a
	R	—	−0.7 ± 1.0 b	4.8 ± 2.8	5.0 ± 2.4	9.1 ± 4.9	2.5 ± 0.7 a
	p (*F*)		0.0391	0.1316	0.3404	0.1498	0.0136
Secondary factor, DLF	—	C	—	1.5 ± 0.6 c	3.4 ± 1.6 b	5.0 ± 2.4 c	—
	—	BDC	—	4.1 ± 1.1 b	6.3 ± 1.1 a	10.5 ± 1.4 b	1.7 ± 0.6 b
	—	INJ	—	6.5 ± 1.6 a	7.3 ± 2.7 a	13.9 ± 2.2 a	2.7 ± 1.1 a
	p (*F*)			<0.0001	0.0012	<0.0001	0.0060
CC × DLF	BS	C	—	1.4 ± 0.2	4.9 ± 1.5	7.9 ± 1.6 b	—
		BDC	—	3.5 ± 0.8	6.5 ± 1.0	11.7 ± 1.2 a	1.1 ± 0.2
		INJ	—	5.1 ± 0.5	5.4 ± 3.1	12.2 ± 2.8 a	1.3 ± 0.7
	M	C	—	1.1 ± 0.3	3.1 ± 0.9	3.6 ± 1.3 c	—
		BDC	—	4.2 ± 1.8	6.4 ± 1.5	10.0 ± 1.7 b	1.9 ± 0.8
		INJ	—	6.6 ± 0.9	9.6 ± 1.9	15.6 ± 1.6 a	3.6 ± 0.1
	R	C	—	2.0 ± 0.7	2.2 ± 1.3	3.5 ± 0.3 c	—
		BDC	—	4.5 ± 0.5	6.0 ± 1.0	9.8 ± 0.8 b	1.9 ± 0.1
		INJ	—	7.8 ± 2.0	6.8 ± 1.6	13.9 ± 1.0 a	3.2 ± 0.3
	p (*F*)			0.4974	0.0851	0.0027	0.1000

Note: Within columns, means followed by the same letter are not significantly different according to Tukey's HSD test ($\alpha = 0.05$).

Abbreviation: BDC, DLF surface broadcast; BS, bare soil; C, control; DLF, digestate liquid fraction; INJ, shallow injection; M, mustard; R, rye.

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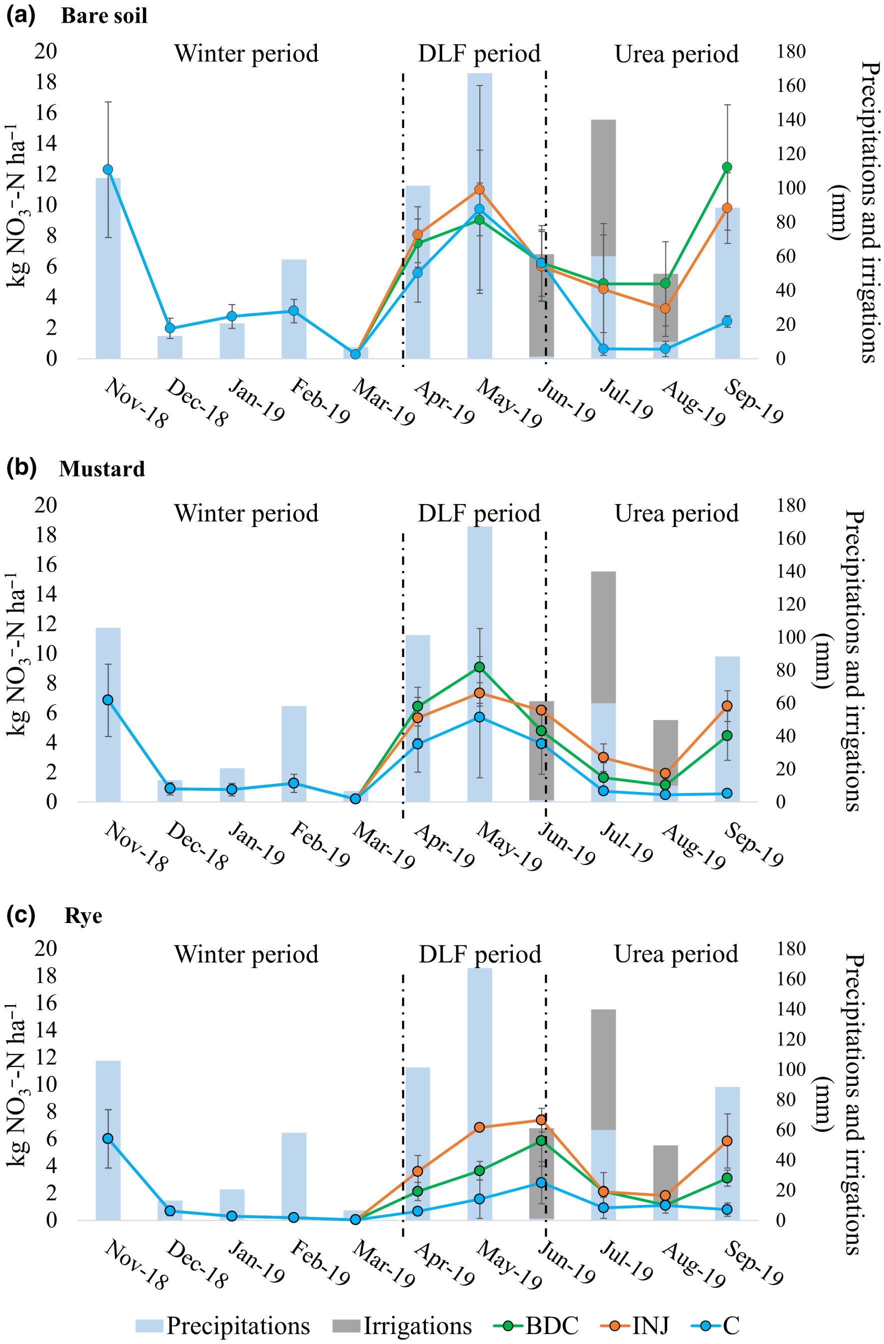

FIGURE 2 Dynamics of monthly cumulative NO_3^--N leaching losses (kg ha^{-1}) for bare soil (a), mustard (b), and rye (c) along the winter monitoring period, and after digestate liquid fraction (DLF) and urea fertilizations. Treatment abbreviations refer to control (C), DLF surface broadcast (BDC) and shallow injection (INJ). Vertical bars refer to monthly cumulative water supply, split into precipitations (light blue) and irrigations (gray).

(6.2 Mg ha^{-1}) was almost two times higher than that reported in the same experimental station in an earlier study (Boselli et al., 2020). This was because these authors considered a long-term no-tillage system, where soil compaction can lower germination efficiency, and because the winter conditions were drier in that earlier study. Similar rye biomass in spring was otherwise reported by Brennan and Boyd (2012), with an average production of 7 Mg ha^{-1}.

4.2 | Maize yield and nitrogen-use efficiency parameters

The presence of CCs tended to decrease maize grain yields and total N uptakes (Table 2). This is fairly consistent with previous studies showing yield declines of 4%–23% after non-legume CCs, due to a partial soil mineral N exhaustion (Abdalla et al., 2019; Fiorini et al., 2022).

Digestate liquid fraction application method had no effect on maize yields. The most likely reason could have been that cumulative N-losses were overall similar for surface broadcast and shallow injection (Table 5). Considering that the average maize yield in the region of our study was around 9.7 Mg ha^{-1} during 2019 (data retrieved from the Italian National Institute of Statistics), our results indicate that both DLF application methods were effective in attaining satisfactory yields for all soil CC scenarios. In terms of NUE indicators, our values were similar to those of Berenguer et al. (2009) for similar levels for organic and mineral inputs. However, they were approximately two

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TABLE 4 Cumulative NO_3^--N leaching ($kg\,ha^{-1}$) for the different monitoring periods and on an annual basis. Values are means ± standard deviations.

Source of variation	Cover crop, CC	DLF application method	Winter period cumulative NO_3^--N leaching ($kg\,ha^{-1}$)	DLF period cumulative NO_3^--N leaching ($kg\,ha^{-1}$)	Urea period cumulative NO_3^--N leaching ($kg\,ha^{-1}$)	Annual cumulative NO_3^--N leaching ($kg\,ha^{-1}$)
Main factor, CC	BS	—	20.4 ± 6.1 a	17.0 ± 5.5 a	20.6 ± 10.2 a	57.9 ± 16.9 a
	M	—	10.1 ± 1.8 ab	12.7 ± 4.3 ab	11.8 ± 5.4 b	34.5 ± 9.3 ab
	R	—	7.3 ± 2.2 b	6.2 ± 3.7 b	11.7 ± 5.6 b	25.1 ± 9.2 b
	p (*F*)		0.0294	0.0336	0.0158	0.0187
Secondary factor, DLF	—	C	—	9.1 ± 6.6 b	7.1 ± 3.1 b	28.7 ± 15.7 b
	—	BDC	—	12.6 ± 6.1 a	17.6 ± 3.1 a	42.8 ± 19.5 a
	—	INJ	—	14.2 ± 5.8 a	19.4 ± 5.4 a	46.2 ± 16.6 a
	p (*F*)			0.0039	0.0001	<0.0001
CC × DLF	BS	C	—	15.3 ± 3.4	9.9 ± 3.2	45.5 ± 12.5
		BDC	—	16.5 ± 5.3	28.4 ± 7.3	65.3 ± 14.2
		INJ	—	19.1 ± 8.5	23.5 ± 8.5	63.0 ± 20.7
	M	C	—	9.6 ± 5.7	5.7 ± 2.1	25.4 ± 9.3
		BDC	—	15.5 ± 3.8	12.0 ± 2.7	37.6 ± 7.8
		INJ	—	13.0 ± 1.5	17.5 ± 0.5	40.6 ± 2.5
	R	C	—	2.3 ± 1.4	5.6 ± 2.6	15.1 ± 5.4
		BDC	—	5.8 ± 0.7	12.3 ± 3.5	25.3 ± 2.4
		INJ	—	10.5 ± 1.2	17.2 ± 2.5	34.9 ± 3.1
	p (*F*)			0.1736	0.1006	0.3004

Note: Within columns, means followed by the same letter are not significantly different according to Tukey's HSD test ($\alpha = 0.05$).

Abbreviation: BDC, DLF surface broadcast; BS, bare soil; C, control; DLF, digestate liquid fraction; INJ, shallow injection; M, mustard; R, rye.

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TABLE 5 Cumulative and yield-scaled N-losses, differentiated per N loss form: NH_3 volatilization, N_2O emissions and NO_3^- leaching. Values are means ± standard deviations.

Source of variation	Cover crop, CC	DLF application method	Cumulative N-losses ($kg\,N\,ha^{-1}$)	Yield-scaled cumulative N losses ($kg\,N\,Mg\,grain^{-1}$)	Yield-scaled NH_3-N losses ($kg\,N\,Mg\,grain^{-1}$)	Yield-scaled N_2O-N losses ($kg\,N\,Mg\,grain^{-1}$)	Yield-scaled NO_3-N leaching losses ($kg\,N\,Mg\,grain^{-1}$)
Main factor, CC	BS	—	93.9 ± 35.9	6.4 ± 1.5	1.6 ± 1.3 b	0.7 ± 0.2	4.1 ± 0.8 a
	M	—	73.7 ± 33.3	5.3 ± 1.5	2.0 ± 1.4 ab	0.7 ± 0.3	2.6 ± 0.4 b
	R	—	87.0 ± 53.9	6.5 ± 3.1	3.8 ± 2.9 a	0.7 ± 0.2	2.0 ± 0.4 b
	p (*F*)		0.2591	0.2481	0.0351	0.6796	0.0167
Secondary factor, DLF	—	C	37.6 ± 15.8 b	3.9 ± 1.0 b	0.5 ± 0.3 b	0.5 ± 0.2 b	2.9 ± 1.2
	—	BDC	112.2 ± 25.2 a	7.6 ± 1.5 a	4.0 ± 1.8 a	0.7 ± 0.1 a	2.9 ± 1.2
	—	INJ	104.9 ± 29.0 a	6.7 ± 2.0 a	2.9 ± 2.0 a	0.9 ± 0.2 a	2.9 ± 0.8
	p (*F*)		<0.0001	0.0005	<0.0001	0.0004	0.8659
CC × DLF	BS	C	55.1 ± 11.6	5.1 ± 0.7	0.2 ± 0.0	0.7 ± 0.2	4.2 ± 0.9
		BDC	119.3 ± 23.3	7.8 ± 1.4	2.7 ± 1.1	0.8 ± 0.1	4.3 ± 0.9
		INJ	107.2 ± 31.4	6.5 ± 1.2	1.9 ± 0.9	0.8 ± 0.2	3.8 ± 0.9
	M	C	32.8 ± 8.5	3.6 ± 0.4	0.4 ± 0.1	0.5 ± 0.2	2.7 ± 0.6
		BDC	96.6 ± 22.9	6.5 ± 1.5	3.3 ± 0.9	0.7 ± 0.1	2.5 ± 0.6
		INJ	91.9 ± 6.7	5.8 ± 0.3	2.2 ± 0.4	1.0 ± 0.1	2.5 ± 0.2
	R	C	24.7 ± 7.1	3.1 ± 0.5	0.8 ± 0.2	0.5 ± 0.1	1.9 ± 0.4
		BDC	120.6 ± 30.4	8.4 ± 1.5	6.0 ± 1.4	0.6 ± 0.0	1.8 ± 0.2
		INJ	115.8 ± 43.4	7.8 ± 3.4	4.5 ± 3.0	1.0 ± 0.1	2.3 ± 0.3
	p (*F*)		0.4537	0.3245	0.8082	0.1828	0.1576

Note: Within columns, means followed by the same letter are not significantly different according to Tukey's HSD test ($\alpha = 0.05$).

Abbreviation: BDC, DLF surface broadcast; BS, bare soil; C, control; DLF, digestate liquid fraction; INJ, shallow injection; M, mustard; R, rye.

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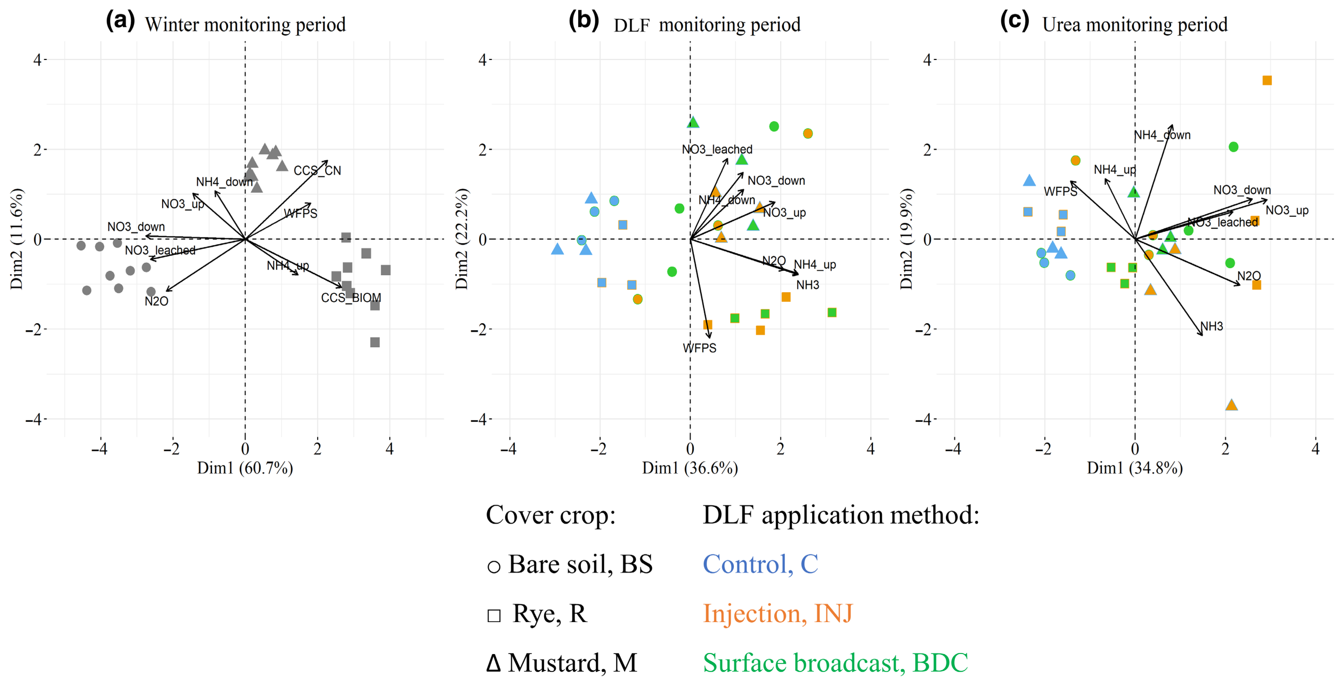

FIGURE 3 Principal component analysis for winter (a), digestate liquid fraction (DLF) (b), and Urea (c) monitoring periods. In biplots, both variables and individuals are reported. Among variables, soil properties (water-filled pore space, NO_3^--N, and NH_4^+-N at 0–10 cm [up] and 10–30 cm [down]), cover crop (CC) parameters (biomass [BIOM] and C/N ratio), and total N losses (cumulative N_2O-N emissions, cumulative NH_3-N volatilization, and NO_3^--N leaching) are reported; individuals are main (circles for bare soil, squares for rye and triangles for mustard) and secondary treatments (light blue for control, C, orange for DLF shallow injection, INJ, and green for DLF surface broadcast, BDC) of the split-plot experimental design.

times higher than those found by Martínez et al. (2017) with pig slurry in Spain. These very low values were attributed by the authors to an inefficient irrigation system and high N losses through leaching.

4.3 | Nitrous oxide emissions as affected by fertilization and cover crop residue

The negative N_2O emissions found in winter with the rye CC may be explained by high denitrification under N-limiting conditions and high WFPS (Liu et al., 2022). In our case, WFPS was never below 75% (except on March 14), an optimum for denitrification (Butterbach-Bahl et al., 2013), and soil NO_3^- during winter was numerically lower for rye than for the other CC treatments (Table S2a,b), promoting the use of N_2O as electron-acceptor by denitrifying organisms under NO_3^- limiting conditions.

Shallow DLF injection increased N_2O emissions compared with surface broadcasting during the "DLF period" (Table 3), which is consistent with previous studies using other animal liquid effluents (Chadwick et al., 2011; Herr et al., 2019) and seems to show similarities with digestates (Fiedler et al., 2017). The most shared explanation refers to the creation of "hot spots" for N_2O emissions within the injection line due to the action of cutting disks, thus creating anaerobic microsites with high C and N concentrations (Petersen et al., 1996), that are ideal conditions for powerful denitrification phenomena (Velthof et al., 2003).

The annual N_2O EFs with CCs (1.9%–3.6% of N applied with fertilizers) were always higher than the 1.6% (uncertainty range = 1.3% to 1.9%) default of IPCC's Tier I for organic and mineral fertilizers application in wet climates (our annual precipitation: potential evapotranspiration ratio is higher than 1; Liang & Noble, 2019). The reason may be a combination of agricultural and environmental conditions stimulating N_2O emissions: high soil organic C availability from digestate application and CC residue decomposition, coupled with high WFPS content following irrigation and precipitation. Moreover, the fine texture of the soil and associated low oxygen diffusivity and low redox potential may have promoted higher denitrification rates (Rochette et al., 2018). Another contributing factor may be that the 140 kg N ha^{-1} added with urea fertilization may have been greater than the maize N-needs, resulting in an exponential, rather than linear, increase in N_2O EFs (van Groenigen et al., 2010).

4.4 | Nitrate leaching as affected by fertilization and residual cover crop biomass

The highest NO_3^- leaching losses during the "Winter period" (November 2018–March 2019) occurred for

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all treatments after the largest precipitation event in November 2018 (cumulative 105.80 mm) (Figure 2). At that peak, 12.3 kg NO_3^--N ha^{-1} were leached in the BS, but only 6.9 and 6.0 kg NO_3^--N ha^{-1} were lost under rye and mustard respectively. This means that the beneficial effect of CCs for NO_3^- leaching was already substantial even after only 1 month of growth. This is because the conditions of the Po Valley area are suitable for fast CC establishment and growth, provided early seeding in late august or September; mild temperatures of October 2018 (average of 15.3°C in our experiment) and copious rains (138 mm) helped, as pointed also by Tadiello et al. (2022). Indeed, it is likely that both CCs may have reached NO_3^--leaching monitoring depth at the beginning of November, considering rooting growth dynamics of 1–2 mm day^{-1} per °C (Thorup-Kristensen et al., 2003).

After DLF application, NO_3^- leaching increased sharply for all treatments. Yet, NO_3^- leaching values in April and May 2019—regardless of DLF application method—were generally highest under BS and lowest under rye, with mustard in between. These results indicate that the higher NO_3^- leaching was due to mineralization of soil organic matter and plant residues, which intensified due to higher soil temperatures in spring. Under these conditions, the differences between CCs were probably due to the effect on soil N immobilization, which was likely higher for rye than for mustard due to the differences between CCs in terms of residue amount, quality, and frost tolerance.

Our results showed that N-immobilization caused by the CC residues, particularly for rye, lasted even during the "Urea period." Considering that by then most of the aboveground CC residues were largely decomposed, this implies that an important factor underlying N-immobilization in our experiment was rye roots decomposition. This seems to be especially important under conditions similar to that of our experiment (no-till or without soil cultivation), where mineralization is slowed down and N-immobilization is prolonged (Martinez-Feria et al., 2016).

On an annual basis, rye reduced NO_3^- leaching by 57% compared with traditional BS, which is in agreement with the range of 18%–95% proposed by Blanco-Canqui (2018) for CCs, and close to the 56% value reported for non-legume CCs by the global metanalysis of Thapa et al. (2018).

4.5 | Yield-scaled N-losses and relation between variables

Nitrate leaching represented the main N loss pathway under BS and mustard, accounting for 63% and 49% of total yield-scaled N-losses, respectively. This was not the case when rye was used as CC, where NH_3 volatilization represented 58% of total yield-scaled N-losses. Although rye residues left over the soil surface acted as a mechanical barrier against DLF infiltration into the soil and increased NH_3 volatilization (Maris, Capra, et al., 2021), the strong reductions in NO_3^--leaching induced by this CC compensated for this effect, leading to generally lower yield-scaled N-losses compared with the other treatments.

The yield-scaled N_2O emissions measured in our study (0.5–0.9 kg N Mg $grain^{-1}$) were close to those of Sistani et al. (2011), who reported a range between 0.2 and 1.4 kg N Mg $grain^{-1}$, depending on fertilizer type. Our results are also in line with those of Preza-Fontes et al. (2022) using in-season split-N application and adoption of a rye CC (0.9–1.0 kg N Mg $grain^{-1}$).

The strong link between NO_3^--leaching and N_2O emissions was mainly driven by the BS plots (Figure 3a), highlighting the benefits derived from ground cover during the fallow period (Blanco-Canqui, 2018; Koudahe et al., 2022). The relationship between gaseous N-losses (N_2O and NH_3) and soil NH_4^+ concentration in the 0–10 cm layer during the DLF period was particularly clear when rye was the CC. In these plots, the high NH_4^+-N content of DLF was more strongly retained in the uppermost soil layer thanks to a barrier effect of rye residue, thus enhancing NH_4^+ conversion to NH_3, and possibly promoting a combination of nitrification and denitrification processes (also nitrifier-denitrification driven by ammonia-oxidizing bacteria), which could have led to the development of N_2O (Maris, Abalos, et al., 2021; Ussiri & Lal, 2013).

During the urea period, NH_3 and N_2O losses were lower and WFPS was generally high, confirming the efficacy of sprinkler irrigation in abating NH_3 volatilization and perhaps a prevalence of N_2O reduction to N_2 trough denitrification (Figure 3c).

To better understand the role that N_2 production may have played, dedicated experiments are required. Since N_2 can be a substantial N loss accounting for up to 85% of total denitrification (Bouwman et al., 2013), it may be of crucial importance for NUE.

5 | CONCLUSIONS

Our study confirms the positive role of winter CCs for limiting nitrate leaching, yet it also raises issues about the controversial effect of their residue once terminated, since (yield-scaled) cumulative N losses were not different to those of BS at the end of the experiment. To promote the potential of CCs to balance productivity and

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environmental aspects, adjusting CC management (e.g., planting time, termination time, and method), fine-tuned at the species level, may be necessary.

An advantage of adopting mustard as CC prior to maize lies in the frost sensitivity of this species, which makes it possible to save on glyphosate spraying if the freezing effect is complete. Therefore, a mixture of mustard and rye, probably with a greater proportion of mustard, could combine the greater N-scavenging potential of rye to reduce nitrate leaching, promoted by its sudden vegetative restart in spring, with the lower competitiveness and earlier degradation of mustard residues due to winter killing. This option may be able to optimize the N loss reductions and N supply benefits of these contrasting species.

Based on our results, using digestate as a starter fertilizer for maize can be a viable option to use this bioenergy by-product. The management of liquid fractions of digestates could then be further improved by acidification and/or application of nitrification inhibitors (Chiodini et al., 2019). These fertilizers technologies deactivate the enzyme responsible for the first step of nitrification, thus decreasing the availability of nitrate susceptible to be leached or stepwise reduced through denitrification and have been shown to be successfully when applied with slurries and digestates (Guardia et al., 2023).

AUTHOR CONTRIBUTIONS

Andrea Fiorini, Vincenzo Tabaglio, and Diego Abalos conceived the ideas and designed the methodology; Federico Capra, Stefania Codruta Maris, Federico Ardenti, and Michela Lommi collected the data; Federico Capra, Stefania Codruta Maris, and Andrea Fiorini analysed the data; Federico Capra, Diego Abalos, and Andrea Fiorini led the writing. All authors contributed critically to the drafts and gave final approval for publication.

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CONFLICT OF INTEREST STATEMENT

The authors declare that they have no 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 openly available from the Dryad Digital Repository: https://datadryad.org/stash/share/VuqxwOr_x5rDksW1oM2O9EuaKqWHxH62KbQBW41H1fo.

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SUPPORTING INFORMATION

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