Effects of Dairy Slurry Injection on Carbon and Nitrogen Cycling

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ABSTRACT:
Surface broadcast of dairy slurry is a common practice; however, concerns over nuisance odors and nutrient losses have prompted research into alternatives. Manure injection is one practice that addresses these concerns but is not widely adopted. Therefore, two studies were conducted to quantify NH_3-N loss by volatilization, impacts on soil N cycling, and microbial response between surface broadcast and subsurface injection of dairy slurry. A constant air flow volatilization chamber system measured NH_3-N losses and soil inorganic N, mineralizable carbon, and active microbial biomass. A 40-day static air incubation was performed to study nitrogen transformations over a longer period after application. Statistical significance was evaluated at the $\alpha = 0.05$ level. In the volatilization study, subsurface injection reduced NH_3-N losses by 98% and 87% in a clay loam and sandy loam, respectively, resulting in greater soil inorganic nitrogen compared with surface application. There were no significant differences in active microbial biomass between treatments. Surface application prompted greater microbial respiration in the sandy loam, but there were no significant differences between treatments in the clay loam. In the static incubation study, differences in soil NO_3^--N became significant on day 28, and by day 40, injection showed increases in soil NO_3^--N of 13% and 26% in the sandy loam and clay loam, respectively, relative to surface application. While the effect of subsurface injection on soil microbial response was unclear, it remains a tool that can greatly reduce NH_3-N losses by volatilization and increase soil plant available nitrogen.

Key Words: Ammonia volatilization, dairy slurry, manure injection, microbial biomass, shallow disk injection, substrate-induced respiration

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The application of animal manure in agricultural systems removes manure from animal production sites and restores valuable nutrients and organic materials that aid crop growth and soil quality. Nutrients provided by the manure reduce soil nutrient requirements that are otherwise fulfilled by expensive chemical fertilizers. While the application of animal manure provides these benefits at a low cost, there are several concerns over the land application of animal manures. Liquid manures, such as swine and dairy, are most often applied to agricultural fields by surface broadcast. However, the practice of leaving manures on the soil surface has prompted reevaluation of current application technologies (Maguire et al., 2011). Manure left sitting on the soil surface causes nutrient losses of nitrogen by NH_3-N volatilization and increases N and P losses by surface runoff. Losses of N due to volatilization can be large; 45% to 80% of ammonical-N applied can be lost as NH_3-N (Thompson and Meisinger, 2002; Maguire et al., 2011). Losses of N decrease the N use efficiency of the agricultural system and increase the need for N inputs via chemical fertilizer. Additional concerns of manure application are the associated odor problems, and farmers are commonly reported by neighbors for their application of animal manures due to nuisance odors (Hardwick, 1985).

In response to these concerns, incorporation by tillage can be used to minimize atmospheric contact with manure, but this is not feasible in no-till systems. In no-till environments and where available, manure injection is suggested as an alternative application technique. Injection of dairy manure has been shown to decrease odor emissions by 33% relative to surface broadcast (Chen et al., 2014), whereas the injection of poultry litter can decrease NH_3-N losses by 98% relative to surface application (Kulesza et al., 2014). Manure injection can also decrease nutrient losses in surface runoff, with injection of poultry litter reducing total Kjeldahl nitrogen and total Kjeldahl phosphorous in runoff by more than 50% compared with surface application (Kulesza et al., 2014). Despite the benefits associated with subsurface injection, widespread adoption of this technique has been slow because of concerns related to application speed and equipment cost (Maguire et al., 2013). Indeed, Chen et al. (2014) studied the costs of different application techniques and agreed that injection has a higher "startup" cost; however, the total cost of injection may be lower because of retention of nutrients and reduced need for chemical fertilizers. Similarly, Rotz et al. (2011) modeled different application techniques and claims that the increased costs of shallow disk injection are leveled with economic return, maintaining profit.

Although greater microbial biomass was reported in soils treated with manure than on unfertilized (Tessier et al., 1998) and chemically fertilized plots (Fraser et al., 1988; Edmeades, 2003), less is known about the impact of manure application technique on soil microorganisms. Generally, no-till practices are thought to increase soil enzyme activity over time when compared with tillage. Still, the shortage of literature on manure application techniques and microbial response has been noted (Acosta-Martinez and Waldrip, 2014). One recent study did consider the application technique of swine manure on the soil arthropod community and found significant interactions, $\alpha = 0.05$, between application technique and collembolan populations (Schuster, 2015). Land application of manures has been shown to increase soil carbon, with higher rates of application resulting in larger amounts of carbon storage (Ding et al., 2012).

Therefore, this study was performed to expand the knowledge of nitrogen cycling dynamics and microbial responses between application methods of liquid dairy manure. Specifically, the objectives of this study were to quantify NH_3-N losses, track changes in soil nitrogen, and observe responses of microbial biomass and

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mineralizable carbon between surface broadcast and subsurface injection of dairy manure.

MATERIALS AND METHODS

Soil and Dairy Manure Collection and Analysis

The following studies were performed using two soil types. One was a Braddock clay loam (Clayey, mixed, mesic Typic Hapludult), and the other was a Dragston sandy loam (Coarse-loamy, mixed, thermic, Aeric Ochraquults). From here onward, the Braddock and Dragston soils will be referred to as the clay loam and the sandy loam, respectively. Values of field capacity, defined as the soil water content after soil saturation and 24-h free drainage, were 41% and 29% for the clay loam and sandy loam, respectively. Dairy manure used in the studies was gathered from a stirred slurry tank on a working dairy farm in Virginia. Manure analysis performed by the agricultural service laboratory of Clemson University indicated a water content of 94.13%, total Kjeldahl nitrogen of 2.21 kg Mg^{-1} and organic nitrogen 1.23 kg Mg^{-1} (Bremner and Breitenbeck, 1983; Peters et al., 2003). The manure was refrigerated prior to use to minimize changes in properties.

Volatilization Chamber Study

Ammonia volatilization was measured in a laboratory setting using enclosed Kimble Chase glass jars, hereafter "chambers," 8.8-cm inside diameter × 15-cm depth, as described by Woodward et al. (2011) and Kulesza et al. (2014). Briefly, humidified air is pumped through chambers in temperature-controlled boxes into acid traps. Chambers are designed with threaded tops and are fitted with lids housing gaskets in order to create an airtight seal. Polytetrafluoroethylene thread seal was wrapped around the chambers threads to aid sealing. Temperature of the boxes was set and maintained at 26°C; air flow was calibrated to 1 L min^{-1} using a digital flow meter. Air traveled from the pump, through distribution lines, and through external and internal humistats filled with deionized water (DI) water to humidify air entering the chambers. Air passing through the chambers is directed through bubbling stones into 4-oz bottles containing 100 mL 0.04 M phosphoric acid. The phosphoric acid removed NH_3-N from the passing air and trapped it in solution as NH_4^+-N. A detailed description of box design and schematics can be found in Woodward et al. (2011). There were three treatments: surface-applied dairy manure, injected dairy manure, and a no-manure control, 2 soils (clay loam, sandy loam) × 3 treatments (surface, injected, and control) × 3 replicates = 18 samples. Field capacity, determined gravimetrically by free draining saturated soil samples for 24 h, was obtained in order to predict water needs to establish 70% field capacity at the beginning of the experiment. Five hundred grams of soil was added to each chamber; injection slits were simulated by inserting two rectangular metal plates vertically into the soil to a depth of 2.5 cm from the bottom of the jar. The plates were then shifted and arranged in a "V" shape, and wooden shims placed between plates (removed before manure application) to hold form. Soils were brought to 70% field capacity excluding the water content to be added by dairy slurry in treated soils. Jars were left covered overnight for water infiltration. In treated soils, manure was applied based on surface area, at a common Virginia application rate of 56,123 L ha^{-1}, approximately 34 mL jar^{-1} (VADCR, 2005). In surface treatments, manure was poured on top of undisturbed soil. In injected treatments, after pouring manure in the simulated slit, the metal plates were gently removed; afterward, the plates were used to carefully close injection slits with soil that had mounted outside the "V" created by the metal plates. This method was used to mirror field conditions created by a shallow disk manure injector (Maguire et al., 2011). Jars were placed in temperature-controlled boxes in a randomized complete block design with three blocks, and the lids were secured. Acid trap bottles were changed at 1, 3, 6, 9, 12, 18, 24, 36, 48, 60, 72, 96, 120, 144, 168, 192, 216, 240, 264, 288, 312, and 336 h from the beginning of the study to establish a time series of NH_3-N emissions. Acid collected was weighed and refrigerated until analysis. The samples were analyzed for NH_3-N using a Lachat Instruments flow injected colorimeter (QuickChem 8500 FIA Automated Ion Analyzer; Lachat Instruments, Hatch Company, Loveland, CO) using QuickChem phenol method 10-107-06-1-G (Prokopy, 1993). Upon study completion, soils within each jar were homogenized, a portion of each soil was retained moist and refrigerated at 1.1°C until microbial analyses were performed. All other soils were laid out thinly on drying racks to air dry. After drying, soil samples were ground to pass a 2-mm screen, and 4-g subsamples were placed in 50-mL centrifuge tubes and shaken with 40 mL of 2 M KCl for 30 min to extract soil inorganic N. The extract was vacuum filtered through Millipore S-PAK 0.45-μm membrane filters and analyzed for NH_4^+-N and NO_3^--N on a Lachat Instruments QuickChem 8500 autoanalyzer using QuickChem Salicylate Method 12-107-06-2-A and QuickChem Method 12-107-04-1-B, respectively (Hofer, 2001; Knepel, 2001).

A carbon mineralization study was performed with the moist soils from the volatilization study in order to estimate the amount of mineralizable carbon present in each sample. The analysis was done following the procedures of Strickland et al. (2010) and was conducted after the volatilization study with soil from each chamber that was homogenized prior to subsampling. Samples were sieved to pass a 4-mm screen, and 6-g dry-weight subsamples were placed in 50-mL centrifuge tubes and adjusted to 65% field capacity. Samples were placed in an incubator for a total of 30 days with samples collected at 1, 5, 10, 20, and 30 days. Twenty-four hours prior to each sampling, the headspace was flushed with CO_2 free air for 3 min to clear the tubes of CO_2. At each sampling time, 5 mL of air was withdrawn and analyzed on an LI-COR CO_2 H_2O gas analyzer (LI-7000 CO_2/H_2O analyzer; LI-COR, Lincoln, NE). Samples were taken after 1, 5, 10, 20, and 30 days; flushing of headspace was repeated 24 h before each sampling time. Measurements taken across time generated a curve, and the area underneath was used to estimate total CO_2 emitted over the 30-day period.

Active microbial biomass was estimated on the soil samples that had been stored wet following the volatilization study. Microbial activity was approximated via CO_2 respiration measurement using the method of Fierer and Schimel (2002). Four-gram dry-weight soil was placed into 50-mL centrifuge tubes and placed in a 20°C incubator overnight to condition. The next day, 8 mL of autolyzed yeast solution (concentration of 12 g yeast, 1 L H_2O) was added to each tube, and samples were shaken for 1 h. Tubes were capped, flushed, and incubated at 20°C for 5 h after which samples were analyzed for CO_2 following the procedure described previously.

Soil Incubation

Since the volatilization study allowed analysis of soil nitrogen only at the end of a 14-day period, the same three dairy manure treatments (surface, injected, and control) on the same two soils were studied in a 40-day incubation. Five sampling times (0, 7, 14, 28, and 40 days) and three replicates were prepared to total 90 incubation cups that were placed in a climate-controlled laboratory at 22.2°C. Three hundred grams of dry soil was placed in 10.5 × 10.5 × 8.5-cm (depth) planter cups lined with coffee filters to prevent soil loss from drainage holes. Control soils were then brought to 70% field capacity using DI water. Manure-treated samples had the water from manure addition taken into account before wetting and received the difference in DI water before manure application. Manure injection was performed using two rectangular metal sheets to form a "V" opening in the soil, in which manure was applied, by surface area, at the common VA application rate (56,123 L ha^{-1}), approximately 62-mL slurry. After pouring manure in the slit, soil from each side of the slit was pushed loosely over the "V," simulating field injection of manure. In surface-applied soils, manure was poured on top of undisturbed soil. After manure application was completed, laboratory cellophane was used to cover each planter cup. Three small holes were made in each cellophane lid to allow gaseous exchange but reduce water loss. Samples were weighed to ensure adequate water content, and water was adjusted to 70% field capacity every 2 to 3 days using DI H_2O. Soils were then incubated at 22.2°C for 40 days and analyzed on days 7, 14, 28, and 40 for NO_3^--N and NH_4^+-N. At each sampling time, samples were spread out thinly on paper in the laboratory to dry, excluding the coffee filter. All samples were set out to dry in this manner on their respective day. Once dried, soil samples were ground to pass a 2-mm sieve, and 4-g subsamples were placed in 50-mL centrifuge tubes. Forty milliliters of 2 M KCl was added to each tube, after which the tubes were shaken for 30 min. The resulting extract was vacuum filtered through a Millipore S-PAK 0.45-μm membrane filter and processed on a Lachat Instruments QuickChem 8500 autoanalyzer for NH_4^+-N and NO_3^--N using the aforementioned colorimetric methods.

Statistical Analysis

Data were analyzed using JMP Pro 12 software (SAS Institute Inc., 2016). One-way analysis of variance was performed by soil and treatment means separated using the Tukey-Kramer honestly significant difference test. In the volatilization study, analysis was performed on the cumulative NH_3-N loss at the end of the 14-day period. One-way analysis of variance was performed on microbial measurements by soil type; where applicable, treatment means were separated using Tukey-Kramer honestly significant difference. In the 40-day incubation study, sample dates were analyzed separately, and means compared within a sampling time. All analyses were considered significant at the $\alpha = 0.05$ level, and error bars in figures represent the S.D. of the mean. The REG procedure in SAS 9.4 was used to evaluate the trends in soil NO_3^--N and NH_3-N dynamics over time and to compare the slopes and intercepts of those regressions (SAS Institute Inc., 2013).

RESULTS AND DISCUSSION

Chamber Study

Ammonia volatilization

Throughout the 14-day volatilization study, manure injection decreased NH_3-N losses relative to surface application (Fig. 1). After 14 days, NH_3-N losses were reduced by 98% and 87% relative to surface application in the clay loam and sandy loam soils, respectively (Table 1). Ammonia losses from both soils and all treatments fit well to a logarithmic model; injection and surface treatments ranged from $R^2 = 0.85$ to $R^2 = 0.98$, $\alpha = 0.05$, $P < 0.001$ (Fig. 1). Control treatments fit a logarithmic model because of rewetting prior to NH_3-N measurement. Ammonia losses from the injection treatment were indistinguishable and marginally elevated from the no-manure control in the clay loam and sandy loam, respectively. These results are similar to those seen in Kulesza et al. (2014), where poultry litter was injected and surface applied. In their study, NH_3-N losses from litter injection were statistically the same as the no-treatment control. In addition, Powell et al. (2011) reported lower NH_3-N volatilization in injection (40%–95%) and aeration-incorporation (26%–64%) treatments compared with surface application in 3 of 4 years of field studies. Powell et al. (2011) also reported that most of the cumulative NH_3-N volatilization occurred in the first 48 h after manure application. This is similar to our study, where 70% of total NH_3-N loss had occurred from surface applications of manure 36 and 48 h after application of manure for the sandy loam and clay loam, respectively (Fig. 1). In manure-injected treatments, 70% of total NH_3-N loss had occurred 96 and 144 h after application for the sandy loam and clay loam, respectively (Fig. 1).

Soil nitrogen

The total organic and inorganic N applied via surface application or injection was the same. Both methods of manure application increased soil NO_3^--N to concentrations well above the control (Table 1). Nevertheless, injection increased soil NO_3^--N by 23% in the clay loam and 110% in the sandy loam over surface application after subtracting the control, presumably because of NH_4^+-N conservation (Table 1; Fig. 1). This is beneficial for crop production as nonlegumes such as corn (*Zea mays*) rely on NO_3^--N additions for economic yield. The sandy loam showed greater differences in soil NO_3^--N between surface application and injected treatments, because of more NH_3-N losses from surface application in the sandy loam (Fig. 1). Total inorganic N (calculated as NH_3-N + NO_3^--N + NH_4^+-N) was the same for injection and surface application in both soils, indicating there was no impact of management on organic N mineralization over 14 days (Table 1.). These results are similar to those obtained by Kulesza et al. (2014), where poultry litter injection

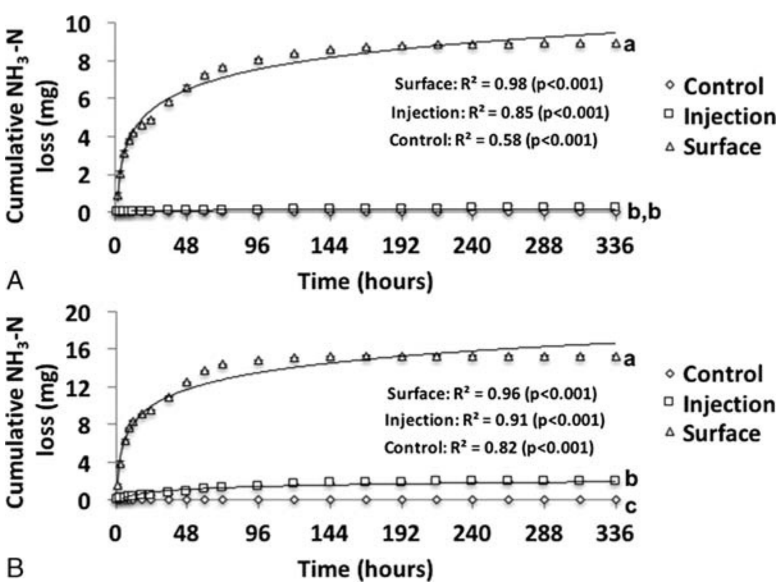

FIGURE 1. Cumulative NH_3-N loss during a 14-day forced-airflow volatilization study with soils clay loam (A) and sandy loam (B). Different letters indicate significant differences at the end of the 2-week period, $\alpha = 0.05$. Error bars indicate the S.D. *P* values indicate an F test for overall significance.

TABLE 1. Soil Nitrogen After 14-day Volatility Study Including NH_3-N Loss, Soil NO_3^--N, Soil NH_4^+-N, and Total Inorganic-N Capture

Soil Type	Treatment	NH_3-N Loss	Soil NO_3^--N	Soil NH_4^+-N	Total Inorganic N
		mg Jar^{-1}			
Clay loam	Control	0.0 b	18.9 b	1.4 b	20.3 b
	Injection	0.2 b	36.4 a	7.8 a	44.3 a
	Surface	8.9 a	33.1 a	5.5 a	47.4 a
Sandy loam	Control	0.0 c	11.4 c	0.6 a	12.1 b
	Injection	1.9 b	40.2 a	0.8 a	42.9 a
	Surface	15.3 a	25.2 b	0.8 a	41.3 a

Within each soil type, different letters in each column indicate significant differences, $\alpha = 0.05$.

increased soil NO_3^--N by 47% in a loam and 58% in a sandy loam. In that study, a large portion of nitrogen captured was in the NH_4^+-N form, possibly because of the shorter study length of 7 days. If nitrification were allowed to continue by increasing study length, it is expected that this NH_4^+-N would be converted to NO_3^--N. In the present study, the soil total inorganic N was increased by 31% and 108% in the clay loam and sandy loam relative to the control, respectively. This is again similar to Kulesza et al. (2014), who found inorganic N to increase by 71% and 105% for a loam and sandy loam, respectively.

Soil carbon

In the clay loam soil, there were no significant differences in mineralizable C detected between treatments, although variability was high (Fig. 2). In the sandy loam, the surface application of manure increased mineralizable C by 68% relative to the control, whereas injection resulted in an increase of 20% relative to the control (Fig. 2). In addition, injection reduced mineralizable carbon by 70% relative to surface application after subtracting the control (Fig. 2). The increases in microbial respiration are presumably due to a larger mineralizable carbon pool originating from manure application; alternatively, addition of manure could have provided the nitrogen necessary to metabolize carbon already present in the soil, or both. Indeed, Liang et al. (1996) reported that adding manure-extracted organic carbon to soils resulted in higher mineralizable carbon than soils receiving no additions of carbon; however, this was probably from carbon already present in the soil. Similarly, Tessier et al. (1998) reported higher soluble carbon and total carbon in plots fertilized with incorporated manure than an untreated control. Conversely, Wander et al. (2007) reported that particulate organic matter carbon did not accumulate in a system amended with dairy manure and attributed it to excess labile N stocks provided by manure. In the present study, manure application in the sandy soil resulted in higher mineralizable C than the no-treatment control, presumably because of the organic carbon and nitrogen added from manure. In addition, surface application led to greater mineralizable C than injection application, possibly indicating that mineralization of organic carbon had already occurred or is not occurring as quickly in the injection treatment. However, this higher mineralizable C did not translate into greater mineralization of N, as total inorganic N was the same for surface and injected manure (Table 1). Anecdotal evidence exists that injection of manure may cause decomposition of added carbon to slow, as it can be identified visually in the injection slit several months after application (Fig. 3). No treatment effect was present in the clay loam soil; other than high variability, this could be attributed to the soil nearing its effective carbon saturation point as described by Stewart et al. (2007). Briefly, the closer a soil is to its effective carbon saturation point, the smaller the observed increases in soil carbon will be.

Active microbial biomass

No significant differences were detected in the active microbial biomass estimates among any of the treatments in either soil (Fig. 4). This could be explained, in part, by the nature of the volatilization study. During the NH_3-N volatilization study (14 days), the soils were kept in an environment conducive to microbial growth; this opportunity for growth before the active biomass estimate occurred could explain the lack of differences between treatments. Future endeavors looking at microbial parameters should consider monitoring microbial activity directly after manure application. Nevertheless, microbial biomass has been shown elsewhere to increase after being treated with manure. Tessier et al. (1998) noted that great variability was seen in manure-treated plots, probably because of inconsistencies in both the incorporation of manure and the properties of

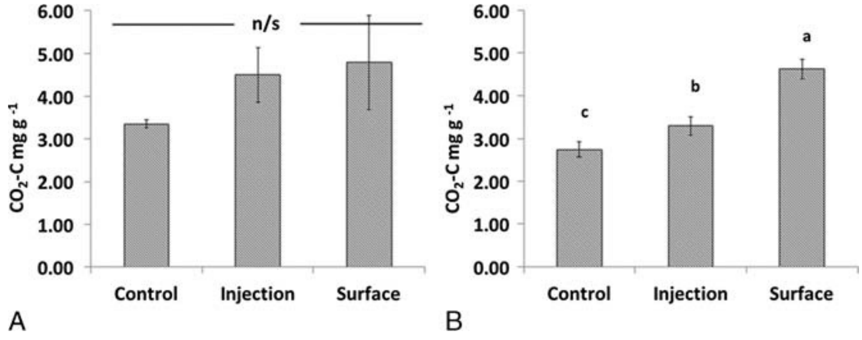

FIGURE 2. Cumulative mineralized carbon during 30-day incubation performed on soils run through volatilization chambers, clay loam (A) and sandy loam (B). Data shown represent the integration of five points in time on days 1, 5, 10, 20, and 30 of the incubation. Significant differences are indicated by different letters. n/s Indicates no significant differences, $\alpha = 0.05$. Error bars indicate the S.D.

FIGURE 3. Photo of a manure injection slit 4 months after application. Manure-added carbon is seen in the center of the photo (Maguire et al., 2013).

the manure itself. Likewise, Kuzyakov and Blagodatskaya (2015) agree that animal manures are a source of microbial "hotspots," and these "hotspots" are likely to have greater total and active microbial biomass. In a study by Rochette et al. (2000), long-term (18-year) application of swine manure did not have a significant effect, $\alpha = 0.05$, on year-end residual microbial biomass carbon; however, microbial biomass carbon peaked directly after manure application and varied by application rate. In theory, microbial biomass should follow carbon mineralization rates; biomass increases during the mineralization of carbon and decreases when little mineralization is occurring.

Forty-Day Incubation Discussion

Over the course of the 40-day incubation, manure injection resulted in higher soil NO_3^--N concentrations relative to both surface application and the control in both soil textures (Figs. 5A, C). In both soils, changes in soil NO_3^--N concentration due to manure application became significant on day 7 compared with the control; however, separation between application methods was not significant until day 28 (Figs. 5A, C). On day 28, injection had increased soil NO_3^--N by 30% and 34% relative to surface application in the sandy loam and clay loam, respectively. Conjunctively, mineralization and nitrification followed a quadratic relationship with strong R^2 relationships, all $P < 0.05$ (Fig. 5). The intercept of the regression for NO_3^--N was similar for all three treatments, but the slope was different between the control treatment and either injection or surface application. Both injection and surface application resulted in continually increasing NO_3^--N through day 40 (Figs. 5A, C). By day 40, the increase in soil NO_3^--N by injection over surface application had changed to 13% and 26% for the sandy loam and clay loam, respectively. Similarly, a study done in Saskatchewan found that the injection of liquid swine manure resulted in consistently higher preseeding soil N over broadcast application and subsequent incorporation (Mooleki et al., 2002). In addition, Schmitt et al. (1995) showed that manure injection increased soil NO_3^--N in the top 30 cm of soil by 17% relative to surface broadcast. As discussed previously for the volatilization study, increases in soil NO_3^--N were due to nitrogen retained by reducing NH_3-N volatilization losses. These increases in NO_3^--N are present in both studies performed; however, they are probably greater in the volatilization study because of the forced-airflow design. Soil NO_3^--N is one parameter of plant available nitrogen, and increasing NO_3^--N at the proper time could result in increased crop yield. Application of manure resulted in higher soil NH_4^+-N concentrations, especially early in the 40-day incubation. Generally, there were no significant differences in soil NH_4^+-N between manure application methods throughout the study, the exception being a relatively small difference on day 40 of the clay loam soil where the injection treatment was more similar to the control than the surface treatment (Figs. 5B, D). Comparisons of the slope of the regressions from the NH_4^+-N over time were similar for both injected and surface-applied manure, but both different from the control. Both the injected and surface-applied manure exhibited a decline in soil NH_4^+-N over time, reaching near zero on day 40 for the clay loam soil (Fig. 5B) and on day 28 for the sandy loam soil (Fig. 5D). In the sandy soil, complete nitrification had occurred by day 28, with no significance in NH_4^+-N seen afterward (Fig. 5D). In the clay loam, nitrification had not been completed entirely by day 40. This was probably due to clay soils having a higher cation exchange capacity and ability to hold NH_4^+-N. This phenomenon is discussed in a study by Fortuna et al. (2012), suggesting that increasing NH_4^+-N fixation due to mineralogy is negatively correlated to NH_3-N oxidizing bacteria. The temporal dynamics of soil N in this study indicate that manure application method did not largely impact nitrification or mineralization rates (Fig. 5). Thus, there is no need to alter the time frame of manure application when using manure injection because of concerns over nitrogen availability relative to crop growth.

CONCLUSIONS

The results of this study indicate that manure injection leads to reductions of N losses by NH_3-N volatilization relative to surface application, and these are coupled with increases in plant available nitrogen as NO_3^--N. This increase in soil NO_3^--N could be beneficial for crop production and could help cover the extra costs associated with manure injection. Application technique did not affect microbial biomass estimates. Mineralizable C increased in the sandy soil

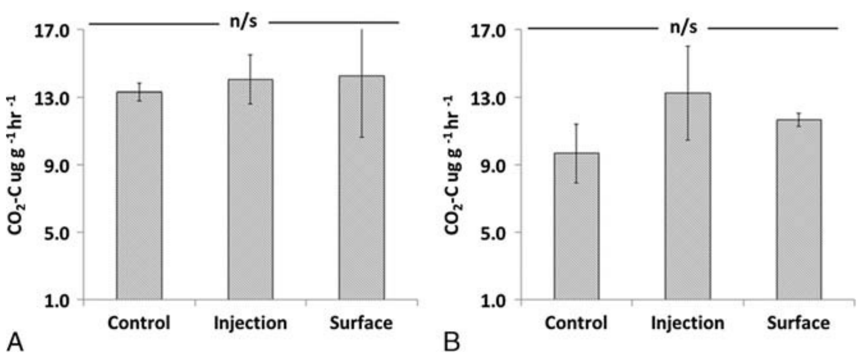

FIGURE 4. Active microbial biomass estimate from an induced respiration incubation using an autolyzed yeast substrate on a clay loam (A) and a sandy loam (B). n/s Indicates no significant differences, $\alpha = 0.05$. Error bars indicate the S.D.

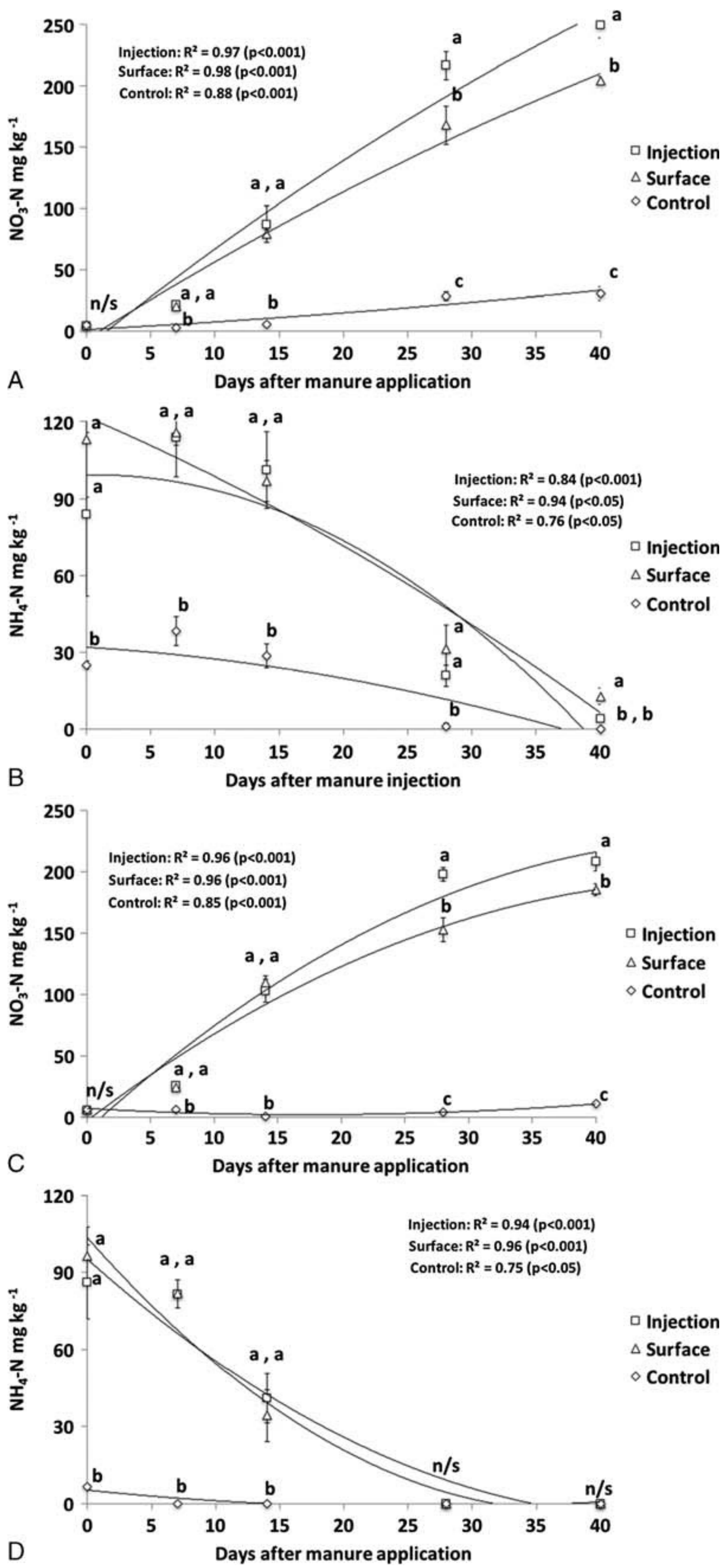

FIGURE 5. Soil nitrogen dynamics as NO_3^--N and NH_4^+-N between manure application methods over the course of a 40-day static air incubation with soils clay loam (A and B) and sandy loam (C and D). Significant differences are noted by differing letters at each time point. n/s Indicates that no significant differences exist, $\alpha = 0.05$. Error bars indicate the S.D. *P* values indicate an F test for overall significance.

where manure was applied, with surface application having the greatest mineralizable C. If more conclusive effects of application technique on carbon cycling are found in the future, the preference of one manure application technique may increase. Nevertheless, injection remains a viable choice for manure application and appears superior to surface broadcast in terms of N retention.

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