MINERALIZATION IN SOILS AMENDED WITH MANURE AS AFFECTED BY ENVIRONMENTAL CONDITIONS

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MINERALIZATION IN SOILS AMENDED WITH MANURE AS AFFECTED BY ENVIRONMENTAL CONDITIONS

By

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MINERALIZATION IN SOILS AMENDED WITH MANURE AS AFFECTED BY ENVIRONMENTAL CONDITIONS

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Dexter Brown Watts was born on July 23, 1978 in Muscle Shoals, Alabama to Oscar and Joyce Watts. He received his high school diploma in 1996 for West Point High School in Cullman County Alabama. Thereafter, he attended Talladega College in pursuit of a Bachelor degree in Chemistry. In 2000, he received a Bachelors of Arts degree from Talladega College. That same year, he was accepted to Alabama Agricultural and Mechanical University where he pursued of the Master of Science Degree in Soil Science, graduating in 2003. Afterwards, he matriculated to Auburn University in hope of obtaining the Doctor of Philosophy Degree in Agronomy and Soils.

DISSERTATION ABSTRACT

MINERALIZATION IN SOILS AMENDED WITH MANURE AS AFFECTED BY ENVIRONMENTAL CONDITIONS

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Concerns for environmental quality have prompted interest in recent years to develop agricultural practices that mitigate nutrient loss to the environment. It is imperative that management practices are developed that maximize the use of plant nutrients while minimizing environmental degradation. Thus, the main aim of this dissertation was to evaluate the impact that manure application to soil from different environmental conditions, soil types, and management practices have on nutrient cycling. This dissertation consists of four parts: (1) Residual effects of long-term tillage and manure application on carbon and nitrogen mineralization, (2) Mineralization of N in soils amended with dairy manure as affected by wetting/drying cycles, (3) A seasonal nitrogen mineralization study as influenced by soil properties and landscape position, (4) Soil microbial community dynamics as influenced by soil properties and landscape position. In the first study, C and N cycling was evaluated in the laboratory using soil from long-term tillage and manure (poultry litter) plots. No-tillage (NT) with litter contained the highest total organic carbon (TOC) in the 0-5 cm depths, which corresponded to significantly higher C and N mineralization rates. Carbon and N mineralization was higher in the 0-5 cm depthsfor NT, while conventional tillage (CT) was higher in 10-20 cm depths. In the second study, influence of manure addition on N mineralization was most evident at higher soil temperatures. The rate of N mineralization was mainly attributed to soil series, Catlin (silt loam)> Goldsboro (loam)> Bama (sandy loam). No significant differences were observed between constant and cycling moisture regimens. In the third study, dairy compost manure was mainly influenced by season with summer months mineralizing the most N. Landscape and soil texture also influenced N mineralization; during the winter months soil with the greatest percentage of sand located in a low lying area lost most of the added manure N, while the loam soil with the greatest soil moisture mineralized the most N during the summer. In the fourth study, dairy compost manure caused a shift in microbial dynamics, which was most evident during summer compared to winter months. Landscape and soil type also changed microbial properties. A loam soil located in a depressed area produced the highest microbial biomass and microbial activity. Canonical discriminate analysis using phospholipid ester-linked fatty acid (PLFA) profiles confirmed these changes in microbial properties by indicating a shift in lipid composition had occurred between season, manure application, and soil landscape. The knowledge acquired from this dissertation study has improved our understanding of nutrient cycling and could aid in the development of better management practices that increases the N use efficiency of litter.

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I. RESIDUAL EFFECTS OF LONG-TERM TILLAGE AND MANURE APPLICATION ON CARBON AND NITROGEN MINERALIZATION

Abstract

Long-term tillage and manure application are thought to alter the ability of the soil to sequester plant nutrients and mineralize carbon and nitrogen. Thus, a laboratory incubation study was conducted under aerobic conditions to evaluate the residual effect of long-term poultry litter (litter) application (>10 years) as affected by tillage. Soil chemical properties were also evaluated in order to assess the influences of management practices. Soil samples were collected at three depths (0-5, 5-10, and 10-20 cm) from continuous soybean and corn plots (with and without litter) under conventional tillage (CT) and no-tillage (NT) systems. The study area was located at the Sand Mountain Substation in the Appalachian Plateau region of Northeast Alabama on a Hartselle fine sandy loam. Long-term tillage management and litter application greatly affected the amount of plant nutrients retained in soil under NT with litter at the 0-5 cm depth. The NT with litter also had higher total organic carbon (TOC) concentration (22.47 and 18.33 g kg^{-1} C) for corn and soybean plots followed by NT without litter (17.32 and 11.10 g kg^{-1} C), respectively. Further, the amount of C mineralized was significantly higher at the 0-5 cm depth for NT and CT compared to the other depths; similar patterns were observed for N mineralization. Plots subjected to long-term litter application had higher

C and N mineralization rates compared to plots without litter. At greater depths more C and N mineralization occurred under CT conditions than under NT conditions; this is attributable to mixing of soil in the plow layer. These results indicate that long-term tillage management plays an important role in the amount of C and N mineralized and sequestered in soil.

Introduction

Soils in the Southeastern USA, including Alabama, where the climate is humid, are severely eroded; this is partially a result of over 200 years of intense row crop agriculture. In this region, row crops have historically been conventionally tilled and fertilized with inorganic fertilizers. These agronomic practices have left the soil relatively infertile, highly eroded, low in organic matter, and easily compacted by rainfall and machine traffic (Carreker et al., 1977).

In recent years, research has shown that soil organic matter (SOM) is the central indicator of soil quality and health (Soil and Water Conservation Society, 1995). Soil organic matter can greatly affect soil properties such as water infiltration rate, erodibility, water holding capacity, and pesticide adsorption (Stevenson, 1994; Campbell et al., 1996; Francioso et al., 2000; Wander and Yang, 2000). The build up of soil organic matter can potentially be achieved by using agronomic practices that are less tillage intensive.

Within the last two decades, agronomic practices using conservation tillage systems have increased. It was estimated that 42.5% of cropland in the Southeastern region was managed with some form of conservation tillage in 2002 (CTIC, 2002). No-tillage (NT) or strip tillage accounted for 37% of cropland in 2002, which is double that reported in 1992. There was an increase in NT from 18.5% in 1998 to 34.7% in 2000 for Alabama's cash crop (cotton) (CTIC, 2000). These practices can lead to a build-up of surface organic matter in addition to reducing soil degradation by erosion (Edwards et al, 1988).

While evaluating the effects of long-term tillage and crop residue management practices on increasing SOM, Hunt et al. (1996) showed that, after 9 years of no-tillage the SOM in the top few centimeters was significantly higher than the soil under CT practices. Campbell et al. (1999) showed that, after 12 years, soil C storage increased in the 0-15 cm depth under a NT practice; most of the differences were observed in the 0-7.5 cm depth with little change in the 7.5-15 cm depth.

Utilization of manure has also been shown to increase SOM. In the USA, the poultry industry produces approximately 8 billion broilers (GASS, 1999) and about 11.4 million tons of broiler litter (1.5kg litter/broiler) each year. At approximately 12% of the nation's output, Alabama is ranked third in the nation in broiler production (Mitchell, 2005). The broiler industry, mainly concentrated in the Sand Mountain region of northern Alabama, produces approximately 1.9 million tons of poultry litter annually (Kingery et al., 1994). This manure has to be disposed of in an environmentally friendly way. The application of poultry litter to cropland can serve as a means of waste disposal and is a relatively inexpensive source of nutrients, particularly N and P (Nyakatawa and Reddy, 2002).

The residual effects of manure application have been shown to last for several ars (Mugwira, 1979; Wallingford et al., 1975, Ginting et al, 2003) Also, residual effects of manure have been shown to maintain crop yield levels for several years after application ceases, since only a portion of the N and other nutrients are plant available in the first year following application (Motavalli et al., 1989; Eghball et al., 2002, 2004). Eghball and Power (1999) observed that 40% of beef cattle feedlot manure N and 20% compost N became plant available in the first year after application, indicating that about 60% of manure N and 80% of compost N became plant available in the succeeding years, assuming little or no loss of N due to NO_3-N leaching or denitrification. Ginting et al,

(2003) showed that manure effects on soil properties can contribute to improved soil quality for several years without an increase in greenhouse gas emissions (CO_2, CH_4, and N_20) resulting from residual manure application 4 years after manure application.

There are a few studies that have investigated the effects of long-term manure application to cropland on increasing SOM in the southeastern USA. In Alabama the application of manure (poultry litter) to cropland has been investigated, although typically in 2 year studies, which has shown a great deal of variability in the amount of SOM generated (Kingery et al., 1994, 1996; Wood et al., 1996; Nyakatawa et al., 2001a). The high variability of these 2-year manure studies suggests that long-term studies need to be evaluated. Since changes in SOM pools occur very slowly, long-term experiments are needed to determine the impact of management practices or other perturbations on SOM dynamics (Beare et al., 1994; Franzluebbers, 2005).

Initiating long-term agronomic practices that build up SOM can play an integral role in regenerating beneficial nutrients of the soil that have been lost due to intensive agronomic practices. Also, understanding the influence that long-term conservation tillage and poultry litter application has on C and N mineralization in soils is necessary from the standpoint of maintaining soil quality for sustainable agriculture. The objectives of this study were to determine the impact of long-term tillage and manure application has on C and N mineralization in soils using laboratory incubation studies.

Materials and methods

Soil samples were collected from a long-term tillage study established in 1980 at the Sand Mountain Substation in the Application Plateau region of northeast Alabama.

The soil was a Hartsells fine sandy loam (fine-loamy, siliceous, subactive, thermic Typic Hapludults). Prior to the initiation of the study, the site had been under intensive row crop production for more than 50 years. The climate of the region is subtropical with no dry season; mean annual rainfall is 1325 mm, and mean annual temperature is 16°C (Shaw, 1982). The soil was sampled at 0-5, 5-10, and 10-20 cm depth increments. Six soil cores (25 mm dia) were collected per plot and composited by depth; surface plant residue was removed prior to sampling. After returning to the laboratory, samples were passed through a 2 mm sieve to remove root materials. Soil mass was recorded and moisture content was determined gravimetrically. Sub-samples were stored at 4°C until use.

Site Description

Samples were collected February 26, 2005, prior to the annual manure application. The two tillage treatments investigated consisted of conventional tillage (CT) (moldboard plow, or rototiller, with wheat cover in the spring, disking in herbicide, and planting) and no-tillage (NT) (planting in the killed crop residue with a double disk-opener planter, with wheat cover in the spring). In 1991, sub-plot manure treatments were estimated within each plot. The cropping systems were continuous corn (*Zea mays* L.) and continuous soybeans [*Glycine max* (L.) Merr.]. Poultry litter was added to soybean crops at 45 kg ha^{-1} (based on P) and to corn crops at 170 kg ha^{-1} (based on N) prior to planting. Plots not receiving poultry litter received the equivalent amounts of commercial fertilizer to match the rates of N and P to corn and soybean litter plots. The fertilizer, be it N and P or poultry litter, was surface broadcast to avoid soil-sampling problems. Lime and K was applied in the fall according to the Auburn University Soil Test recommendations.

Laboratory soil analysis

Air-dried samples were ground and passed through a 2 mm sieve. Total C and N were determined by DUMAS dry combustion method using a CN LECO 2000 analyzer (LECO Corp, St. Joseph, MI). Soil pH and CEC, extractable P, K, Mg, and Ca were analyzed by Auburn University Soil Testing Laboratory as described by Hue and Evans (1986).

Incubation study

Methods described by Torbert et al., (1999) were utilized for quadruple determinations of potential C and N mineralization. Concentrations of NH_4 and NO_2 + NO_3 were determined by extraction (before and after incubation) using 2 M KCl as described by Keeney and Nelson (1982) and measured colorimetrically using automated laboratory equipment (Bran-Luebbe, Norderstedt, Germany). Twenty-five grams of moist soil samples (oven-dried weight basis), passed through a 2 mm sieve, were placed in plastic containers. Deionized water was added to bring the soil moisture level to approximately –20 kPa at a bulk density of 1.3 Mg m^{-3}. The containers were placed in incubation jars and 10 ml of water was added to the bottom of each jar (not sample) for humidity control. A 10 ml CO_2 trap (vial composed of 1N NaOH) was added to the jars (not sample) and sealed. Jars were incubated in the dark at 25˚C and removed after 7, 30, 60, and 90 days. After removal, 1ml of a saturated $BaCl_2$ solution (~1N) was added to each sample to stop the reaction. The NaOH was then titrated with 1N HCL, using phenolthalein as an indicator, to determine the amount of CO_2 released from soil samples. Potential C mineralization was the difference between CO_2-C captured in sample traps from the soil samples and in blanks. Potential N mineralization was the difference between final and initial inorganic N contents for the incubation. Carbon mineralization was divided by total C to calculate C turnover. The ratio of C mineralized to N mineralized resulting from the incubation was also calculated.

Statistics

The experimental design was a randomized complete block design with a split plot restriction on randomization with four replications. The tillage treatments (NT and CT) are the main plots with manure application as the split plot. Statistical analyses of data were performed using the mixed procedure of Statistical Analysis System (Littell et al., 1996). A significance level of $P<0.10$ was established *a priori*.

Results and discussion

In general, soil analyses for total C, total N, CEC, and C:N ratio (Table 1 and 2) indicates that changes resulting from management practices have occurred. Significant differences were mainly observed at the 0-5 cm depth. There were no significant differences observed between management practices for the C:N ratio due to tillage or poultry litter amendment. These trends were shown in soil under both the soybean and corn cropping systems. Changes in soil properties indicate that long-term management practices have played an integral role in improving SOM, thereby affecting the fertility of the soil. The following discussion is a more in-depth look at the specifics of how management practices impacted soil characteristics and nutrient dynamics.

3.1 Soil pH

When averaged over all litter treatments, tillage significantly impacted soil pH with NT maintaining a higher pH compared to CT at the 0-5 cm depth (Table 2).

However, at the 5-10 and 10-20 cm depths the CT management practices had higher pH compared to NT; just the opposite of that observed at the 0-5 cm depth. Soil pH decreased by depth from 6.4 to 5.9 and 6.5 to 5.6 in NT plots and increased from 5.9 to 6.3 and 6.0 to 6.2 in CT plots for the soybean and corn cropping systems, respectively. The decrease in soil pH observed under NT, as depth increased, was probably due to surface application of lime, limiting vertical movement and thereby preventing soil reaction at lower depths. Also, soils in the Southeastern region are susceptible to leaching resulting from high rainfall events, which may have resulted in the removal of the exchangeable bases at the lower depths; alternatively, plant roots could have removed the exchangeable bases, thereby decreasing soil pH. As for the CT, lime was probably mixed to greater depths, thus increasing pH. These findings are similar to that of Ismail et al. (1994) who observed in continuous corn plots that had been limed 6 years prior to sampling. Soil pH under NT was higher in the surface 5 cm compared to the CT, but below the surface 5 cm the pH was lower under NT compared to the CT. Similar findings were reported by Guzman et al. (2006). Litter amendments, average over the tillage treatments, did not affect pH at any depth (Table 3). However, Kingery et al. (1994) reported that long-term poultry litter application to tall fescue (*Festuca arundanacea* Schreb) increased soil pH by 0.5 units to depths of 60 cm. Our results did not reflect any change in pH from litter application, probably due to the high buffering capacity of the soil. The tillage X litter application by depth interaction indicated a difference between treatments for soil pH (Table 2), which was mainly due to the effect of tillage as previously stated above.

Soil CEC

The tillage effect on CEC, averaged over all litter treatments, was higher under NT at the 0-5 cm depth for the soybean (77%) and corn (83%) cropping systems (Table 2), respectively. No significant differences were observed at the 5-10 or 10-20 cm depths. This is in agreement with Tarkalson et al. (2006) who reported that soil under NT had a 20% higher CEC at the 0-5 cm depth compared to CT after 27 years of tillage. It has also been reported that an increase in CEC, at the soil surface under NT systems compared to CT, can result from an increase in SOC (Jaiyeoba, 2003; Ciotta et al., 2003). Significant differences were also observed for litter amendments, averaged over all tillage treatments (Table 2). At the 0-5 cm depth, litter increased the CEC in the soybean (22%) and corn (45%) cropping systems, compared to treatments without litter. There was a significant CEC increase of 21% at the 5-10 cm depth for litter application in the corn cropping system only, which is similar to the findings of Gao and Chang (1996). They reported that manure application increased soil CEC after 18 years of manure application. A significant litter X tillage interaction by depth (Table 2) indicated differences at the surface 0-5 cm depth. Soil under NT with litter had the greatest CEC increase for both cropping systems. At the 5-10 cm depth, significant differences were observed for the corn cropping system but not for the soybean cropping system. Also, no significant differences in CEC were observed at the lower 10-20 cm depth.

Soil macronutrients

Extractable phosphorus P levels were higher at the surface 0-5 cm depth compared to the 5-10 and 10-20 cm depths. Comparison of the tillage treatment, when averaged over all litter treatments, shows that extractable P levels were significantly higher (58%) in the NT compared to that of the CT plots at the 0-5 cm depth for the soybean cropping system (Table 3). The same trend was observed for the corn cropping system with NT containing 38% more than the CT at the 0-5 cm depth (Table 3). No significant differences were observed below the 0-5 cm depth, which was probably due to large within site variability in these measured parameters. Although not significant at the lower depths, trends were the same with NT having the highest extractable P compared to CT. Edwards et al. (1992) reported that NT extractable P levels, after 10 years of management, were greater than CT at the 0-5, 5-10, and 10-20 cm depths. This was also similar to the finding of Hargrove et al. (1982) who reported a 120% increase in P under NT compared of CT at the 0-7.5 cm depth. These reports are also in agreement with the results of Guzman et al. (2006).

Comparison of treatments amended with and without litter (averaged over all tillage treatments) indicated a trend similar to the tillage effect (Table 3). Most of the extractable P was found at the upper 0-5 cm depth. There was a 78% and 175% increase in the amount of extractable P in areas receiving litter compared to those without litter for the soybean and corn cropping systems, respectively. No significant differences were observed in the soybean system below the 5 cm depth. For the corn cropping system, significant differences were observed between the litter and no litter treatments. The 5-10 cm depth had a 151% higher extractable P in the litter plots compared to those without added litter. The same trend was observed at the 10-20 cm depth with the litter plots containing 230% more extractable P than the no litter plots. Kingery et al. (1994) found six times higher extractable P to a depth of 60 cm due to long-term poultry litter application to tall fescue. Our results are also in agreement with that of Chang et al.

(1991) and Eghball (2002) who found increases in extractable P with the use of manure in surface soils. Comparison of the litter X tillage interaction by depth (Table 3) indicated that the NT with litter treatment was significantly higher at all three depths for the corn cropping system. The soybean cropping system had significantly more P at the 0-5 cm depth only. It is important to note that litter was applied to soybean plots based on P recommendations whereas corn was based on N recommendation. Although not significant, more P was observed at lower depths for the soybean cropping system. Similar findings were reported by Kingery et al. (1996) who found that soil under strip tillage (a form of conservation tillage) with litter application released more extractable P compared to conventional tillage practices in a laboratory incubation study. This indicated that conservation practices that utilize organic amendments tend to sequester more P than soil under conventional techniques in surface soil.

No significant differences in extractable K were observed between tillage treatments when averaged over all litter treatments at any depth for the soybean cropping system (Table 5). Although not significant, NT had more extractable K for all depths. There was a significant difference observed in the corn cropping system with NT containing 22% more K than CT at the 0-5 cm depth. The lower 5-10 and 10-20 cm depths, although not significant, had more extractable K in the NT compared to that of the CT. Our findings are in contrast to that of Edwards et al. (1992) who found more extractable K under CT than NT at the 0-5 cm depth for the corn (with a wheat cover crop) cropping system. However, Edwards et al. (1992) results for a soybean cropping system was similar to our observations. Our findings are also similar to that of Guzman et al. (2006) who found higher extractable K at all depths for the NT compared to that of

CT. The reason for the difference between soybean and corn cropping system was probably related to the fact that litter was applied on a P basis for soybean and on a N basis for corn.

When averaged over all tillage treatments, significant differences were also observed between soil with and without litter application at all depths, with more K being found in areas with litter application (Table 3). Soil from the soybean cropping system contained 91% (0-5 cm), 78% (5-10 cm) and 69%(10-20 cm) more K in the litter plots compared to those without litter. The same trend was observed in the corn cropping system with the litter plots containing 110% (0-5 cm), 195% (5-10 cm), and 202% (10-20 cm) more K than the no litter plots. Schlegel (1992) reported that soil K was increased, when using composted beef cattle feedlot manure compared to the same amount of synthetic fertilizers, in a study from 1987 to 1990. A comparison of tillage X litter interaction by depth shows the same trend as observed with the other macronutrients. Soil managed under NT with litter application was significantly higher than the other treatments at all three depths in both cropping systems (Table 3).

Tillage effect averaged over all litter treatments significantly affected the amount of extractable Ca retained in the soil. Extractable Ca was higher under NT (112% for soybean and 117% for corn) compared to CT management practices in both cropping systems at the surface 0-5 cm depths (Table 3). Although, no significant differences were observed at depths below 5 cm, CT had a higher amount of Ca compared to NT. This was similar to the pH effect observed for the liming application in the current study. These findings are also similar to the reports of Guzman et al. (2006). When averaged over all tillage treatments the amount of extractable Ca was 30% (soybean) and 58% (corn) higher with litter compared to no litter (Table 6) for both cropping systems; this is in agreement with the findings of others (Lund and Doss; 1980, Chang et al; 1991). No significant differences were observed at the depths below 5 cm. For the tillage X litter interaction by depth, the NT with litter was significantly higher at the soil surface (Table 3). At the greater depth, CT with litter had the highest Ca concentration, primarily due to mixing of Ca with tillage as observed for pH measurements addressed above.

Significant differences for extractable Mg under different tillage management were observed when averaged over all litter treatments. Extractable Mg was significantly higher under NT management compared to that of the CT at the 0-5 cm depth for both the soybean (85%) and corn (94%) cropping systems (Table 3). Although, no significant differences were observed below the 5 cm depth, the amount of extractable Mg was higher under CT at the lower 5-10 and 10-20 cm depths for the corn cropping system. This is similar to that observed for the extractable Ca discussed above and to the results of Matowo et al. (1999). They proposed that this phenomenon, which resulted in a lower soil pH value under CT compared to NT, might have resulted in greater mineral weathering and translocation of Mg. The same trend was shown for the litter and no litter effects averaged over all tillage treatments (Table 3). Litter treatments contained 25% (soybean) and 39% (corn) more Mg than plots without litter, which was in agreement with the reports of others (Lund and Doss; 1980, Chang et al., 1991). The tillage X litter interaction by depth indicated that NT with litter was significantly higher in Mg than all the other treatments (Table 3). At the lower depths, the same trend was shown as observed with Ca. Conventional tillage with litter had higher Mg at the lower depths compared to the other treatments.

As suggested by Guzman et al. (2006), changes in extractable Ca and Mg can be greatly influenced by the application of lime. In this study extractable Ca and Mg and pH patterns were similar. Therefore, it is likely that higher extractable concentration of Ca and Mg in the CT compared to NT at lower depths can be attributed to lime incorporation. Likewise, higher concentrations of Ca, Mg, and higher pH was attributed to lime being surface applied thereby only affecting the surface few centimeters of the soil.

Total soil carbon

At the 0-5 cm depth, significant differences were observed for total C. The effect of tillage when averaged over all the litter treatments for the NT treatment was 70% and 99% higher (Table 2) than the CT treatment in the soybean and corn cropping systems, respectively. No significant differences were observed at the 5-10 and 10-20 cm depths. The higher soil C under NT reflects increased C inputs and reduced tillage intensity. Total C was lower under CT, probably due to increased oxidation and microbial activity resulting from soil mixing (Stevenson, 1986). It has also been reported by Wood et al. (1991) that differences observed in surface organic matter between tillage systems can be attributed to a slightly higher return of crop residues under NT compared to CT. The increase in total C was restricted to the surface, indicating that changes in the soil environment was due to tillage. Our observed differences are supported by results of other research on long-term effects of conservation tillage systems (Franzluebber et al., 1994; Torbert et al., 1997; Feng, 2002).

Soil amended with litter compared to soil without litter, when averaged over all tillage treatments, was 33% higher in total C under both cropping systems at the 0-5 cm depth (Table 2). At the 5-10 cm depth, there was a significant difference between soil with and without litter application only in the corn cropping system. Soil amended with litter sequestered 22% more carbon compared to soil without litter application. This is in accordance with Gao and Chang (1996) who reported that, after 18 years of manure application, there was an increase in total C near the soil surface.

A significant interaction of tillage X litter by depth indicated differences at the surface 0-5 cm depth (Table 2). The NT system with litter treatment produced the highest amount of total C. No significant differences were observed at the lower depths, except for the 5-10 cm depth for the corn cropping system under NT with litter retaining the most C. In essence, soil under NT with litter can be a powerful management practice capable of building up surface SOM (sequestering more C). This was similar to findings of Nyakatawa et al. (2001b) who reported that the residual effect of poultry litter, in conjunction with conservation tillage, increases plant biomass leading to more residue inputs that increases SOM in the long term.

Also, it was observed in this study that an increase in surface SOM (total C x 1.72) could be correlated with an increase in CEC. This is in accordance with the findings of other researchers (Blevins et al., 1983; Ciotta et al., 2003; Tarkalson et al., 2006).

Total soil nitrogen

The amount of total N in the soil followed the same pattern as CEC and total C at the 0-5 cm depth for the tillage effect when averaged over all litter treatments; total N concentrations were 62% and 87% higher under NT compared to CT for the soybean and corn cropping systems, respectively (Table 2). No significant differences were observed at the lower 5-10 and 10-20 cm depths; this is in agreement with the findings of others (Torbert et al., 1997, 1999).

As for soil amended with and without litter when averaged over all tillage treatments (Table 2), differences were only observed at the 0-5 cm depth where plots with poultry litter had a significantly higher total N. Total N was 11% and 34% higher in the plots containing litter than plots without litter for the soybean and corn cropping systems, respectively. No significant differences were observed at the 5-10 and 10-20 cm depths. This is similar to the findings of Dick (1983) who found increases in the amount of total N in surface soils under NT compared to intensive tillage practices (moldboard plow).

Comparing the interaction of tillage X litter by depth (Table 2), plots under NT and litter application were significantly higher than all the other treatments at the 0-5 cm depth. This was probably due to the fact that manure is less resistant to decay and was not in a readily usable form as compared to inorganic fertilizers. Also, greater crop residues are returned to the soil under conservation tillage thereby supplying the soil with greater residual organic matter. Therefore, the N that is in organic form (litter and crop residue) slowly decomposes, causing a buildup of total soil N.

Soil C:N ratio

Research has shown that soil C:N ratios generally increase with decreased tillage and increased addition of crop residues (Black, 1973). The C:N ratios in this study (comparing the effect of tillage averaged over all litter treatments) tended to be higher in the NT tillage systems at all depths for both cropping systems, although no significant effects were observed at the 0-5 and 5-10 cm depths (Table 2). There was a significant difference at the 10-20 cm depth for the soybean cropping system. The CT had the lowest C:N ratio, probably due to the mixing of crop residue within the soil profile. The same trend was shown for the manure treatments averaged over all tillage treatments. At the 5-10 cm depth, the no-litter treatment had a lower C:N ratio compared to the litter treatment (Table 2). The tillage X litter interaction by depth (Table 2) indicated there was no significant difference in the corn cropping system. However, there were differences observed at the 10-20 cm depth of the soybean cropping system, with the CT with litter being significantly lower than the other practices.

Soil incubation

Carbon mineralization

Laboratory C and N mineralization procedures can be useful in investigating the impact that long-term tillage management practices and organic amendments may have on soil functions. A comparison of tillage effects when averaged over all litter treatments showed that tillage had a significant affect on soil mineralization patterns. The quantity of CO_2 produced from soil respiration (C mineralized) under NT was 14% and 28% higher compared to CT for both soybean and corn cropping systems at the 0-5 cm depth, respectively (Table 4). This is similar to the findings of Wood and Edwards (1992) where, after 10 years, NT mineralized more C compared to CT for soil collected from the same region as this experiment. Salinas-Garcia et al. (1997) found that after 16 years, NT mineralized 34% more C than CT. Similar observations have been reported by others (Franzluebbers et al., 1995; Torbert et al., 1999; Wright and Hons, 2004). At the 5-10 cm depth, the opposite trend was observed with the CT mineralizing 60% and 33% more than the NT system for the soybean and corn cropping systems, respectively. Although not significant, the amount of C mineralized was higher under CT than NT, probably due to the shallow mixing of crop residue at the 5-10 and 10-20 cm depth resulting from the roto-tiller. The amount of C mineralized due to the litter effect when averaged over all tillage treatments, was similar to that observed with the differences in tillage. Carbon mineralization was 9% and 15% greater under litter compared to no litter at the 0-5 cm depth for soybean and corn cropping systems, respectively (Table 4). Below 0-5 cm depth, the amount of C mineralized did not differ between litter treatments. Differences observed in the amount of C mineralized are an indication of amounts of labile organic C accumulated from different tillage and litter verses no litter plots. The tillage X litter interaction effect by depth (Table 4) shows that soil under NT with litter application had the highest C mineralization than all the other treatments. This is consistent with the findings of Kingery et al. (1996), who found that management practices under NT with litter application in the Sand Mountain area of Alabama had the highest C mineralization. At the lower depths, opposite trends were shown with the CT with litter (tillage X litter interaction) and the CT without litter (tillage X without litter interaction) having the highest mineralization. This is due to the mixing of residue at lower depths resulting from CT.

Nitrogen mineralization

The rate of N mineralized for the tillage effect when averaged over all litter treatments was not significant at the 0-5 cm or 10-20 cm depth (Table 4) for the corn cropping systems. There was a significant difference for the soybean cropping system with the NT mineralizing 13% more than the CT system. The amount of N mineralized was 64% and 43% greater under CT compared to NT at the 5-10 cm depth for the soybean and corn cropping systems, respectively. This is consistent with previous research, which indicated a higher N mineralization for CT at lower depths (Torbert et al., 1999; Wright and Hons, 2004). There was a significant difference between the plots with and without litter (averaged over the tillage treatments; Table 4); litter plots produced 13% and 29% more mineralized N than plots with no litter for the soybean and corn cropping systems, respectively. Below the 5 cm depth, only the 5-10 cm depth for the corn cropping system was significantly different, with litter plots mineralizing 36% more than the no litter plots. This is probably due to the fact that long-term management with litter increased SOM in the soil (as seen with the C data) that supports large microbial population and activity, resulting in a higher N mineralization of the available substrate.

The same trend was observed with the tillage X litter interaction effect by depth (Table 4); NT litter treatment mineralized the most N compared to the other treatments for the soybean cropping system at the 0-5 cm depth. No significant differences were observed at lower depths. These results are similar to the findings reported by Kingery et al. (1996). The opposite was shown for the corn cropping system where the CT with litter in the 0-5 cm depth mineralized the most N. This was probably due to the effect of corn residue being left on the soil surface and lack of tillage. Risasi et al. (1999) reported that corn root residues immobilize N for as much as 24 weeks. It has also been reported by Fortuna et al. (2003) that decreased N availability can result in reduced corn yields of crops grown under continuous corn with compost, thereby causing immobilization.

Soil carbon turnover

At each depth, the lowest C turnover observed with the tillage effect (averaged over all litter treatments) was the NT management system (Table 8). At the surface 0-5 cm depth, the highest C turnover was observed under CT with a turnover of 41% and 20% more than under NT for the soybean and corn cropping system, respectively; a similar trend was observed at the lower depths. No significant differences were observed at any depth for the litter effect averaged over all tillage treatments in the soybean cropping system. Although residue quality data was not collected in this study, the lower C turnover was probably due to the fact that soybean residues generally have higher N concentration compared to corn. Thus, soybean residues decompose at a more rapid rate (Parr and Papendick, 1978; Wood and Edwards, 1992) and are less likely to accumulate as SOM (Wood and Edwards, 1992). There was a significant difference between litter and no litter treatments when averaged over the tillage treatments; litter treatments produced 43% higher C turnover than the no litter treatments for the 0-5 cm depth for the corn cropping system (Table 4). This is probably a result of corn residue decomposing at a slower rate, therefore contributing to organic matter buildup. A comparison of the tillage X litter interaction shows that C turnover (amount of carbon lost) under NT with litter management practice sequestered the most C at all three depths. Therefore, management practices that minimized tillage and litter can greatly affect the capacity of soil to sequester C.

Soil C:N mineralization

Soil C:N mineralization ratio can be used as an index of the amount of labile C substrate available in the soil. An increase in the C:N mineralization ratio indicates a decrease in labile C substrate available, resulting in immobilization (Burke et al., 1989; Nadelhoffer et al., 1991; Torbert et al., 1999). No significant difference occurred between tillage or litter treatment main effects at any soil depths for the corn cropping system. This is somewhat different from what was reported by others (Torbert et al., 1997;1999). They found differences between conservation tillage and more intensive tillage methods. The highest C:N mineralization ratio in the present study was observed in the upper soil depths, which is similar to findings of Wood and Edwards (1992). For the soybean cropping system there was a significant difference with the NT having a higher C:N mineralization ratio at the 0-5 and 5-10 cm depths; this is in agreement with other reports (Nadelhoffer et al., 1991; Torbert et al., 1999). A comparison of the tillage X litter interaction effect shows that no significant differences occurred at the upper two depths, 0-5 and 5-10 cm. Significant differences were observed at the lower (10-20 cm) depth for the soybean and corn cropping systems. This was mainly attributed to tillage intensity of the NT with litter (NT X litter interaction) and NT without litter (NT X without litter interaction) having a higher C: N mineralization ratio.

Changes in soil properties since initiation of study

The long-term agronomic practices such as conservation tillage and litter application examined in this study resulted in soil changes since the start of the experiment in 1980. Conservation tillage systems such as NT were shown to greatly impact the fertility, mainly near the soil surface (0-5 cm depth). As observed in previous studies (Wood et al., 1991; Torbert et al, 1999), this is probably a result of the crop residues added back to the soil from initiation of the study in 1980 until 2005. Conventional tillage practices tend to promote decomposition by incorporation of crop residue into soil, leading to the physical breakdown of residues and disruption of SOM within soil aggregates (Paustian et al., 2000; Six et al., 2000). Research has shown that NT promotes a less oxidative environment in the soil with slower decomposition rates than CT (Doran, 1980, Torbert et al., 1999) and is less susceptible to loss by surface soil erosion (Saffigna et al., 1989).

Difference in tillage practice over this 25 yr period also altered soil chemical properties. After 25 yr of tillage, NT had a lower surface pH and higher K, Ca, CEC, Mg, P, total C, and total N. The amount of C and N mineralized was highly correlated to the fertility of the soil (total C & N, CEC, and macro nutrients). The amount of potential C and N mineralized was sensitive to SOM build-up in the soil, showing that CT has a higher potential for loss of C and N and other soil nutrients compared to NT.

The effect of long-term litter application was also shown to increase K, Ca, Mg, P, total C, total N, and CEC. A contributing factor is that litter may act as mulch, which reduces soil erosion while at the same time conserving soil moisture and providing additional nutrients that are not found in normal commercial fertilizers (Nyakatawa et al., 2001b). Due to the increase in organic matter and other additional nutrients from litter application, our results from the C and N mineralization show these areas are better adapted to conserving C and N in the soil. These results are supported by other work, showing that, after two years of poultry litter application to cropland (Nyakatawa et al, 2001b), SOM significantly increased near the soil surface (0-15 cm depth). Also, Kingery et al. (1994) found similar results to ours where cumulative mineralized C was 33% higher under conservation tillage where litter was applied compared to conventional methods. The amount of cumulative N mineralized in their study was greatest in soils maintained under conservation tillage amended with litter. Conservation tillage treatment was 47% greater than soils where litter was applied to CT; similar results were shown with C mineralization (Kingery et al., 1996). Manure or litter has a greater effect than just N fertilization and increasing crop residue; it can substantially increase soil organic C by directly adding C to the soil. The use of conservation tillage such as NT in conjunction with manure application can decrease C and N loss from soil. As stated by Nyakatawa et al. (2001b), manure is prominent in increasing SOM, because not only does it increase crop residue, it builds up SOM since surface application is not subjected to rapid microbial decomposition processes that occur when residue is incorporated into the soil.

Conclusions

Differences in agronomic practices such as tillage (NT and CT) for over 25 years and organic amendment (addition of litter) for more than 10 years, have resulted in differences in soil chemical properties in the Sand Mountain Region of Northeast Alabama. After 25 years, NT had higher soil fertility compared to CT. The use of organic amendment was similar to that of the conservation tillage practices in that it significantly increased soil fertility. These findings show that long-term conservation tillage practices can greatly affect the capacity of soil to sequester C and retain nutrients that are essential for plant growth. Poultry litter in agriculture production not only functions as a means of waste disposal, but also plays a major role in supplying the soil with residual C and N and other macro and micronutrients. Taken together, our results show that farmers in the Southeastern region could help improve the environment and promote soil quality improvement of highly eroded soils by implementing conservation practices with organic amendments.

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Table 1. Selected initial (1980) soil chemical properties with soil depth (0-60 cm) of experimental area.

Soil depth	Soil NO3-N	Soil pH	Soil CEC	Soil Organic matter	Double-acid extractable K	P	Ca	Mg
cm	mg kg^{-1}		$cmol_c$ kg^{-1}	g kg^{-1}	------------------- mg kg^{-1} -----------------			
0-15	7.9	5.8	4.5	10	76	36	428	26
15-20	7.8	5.3	4.5	8.1	52	18	380	31
30-45	5.1	4.5	5.7	5.3	38	10	230	39
45-60	5.9	4.6	5.8	3.3	31	1	195	37

Initial soil samples were collected in the spring of 1980 (Edwards et Al., 1992)

Table 2. Effect of litter and tillage on soil pH, CEC, total C, total N, and C:N ratio at the 0-5, 5-10, and10-20 cm depths for the soybean and corn cropping systems.

Tillage	Amendment	pH	CEC	Total C	Total N	C:N
			cmole kg^{-1}	---------g kg^{-1} --------		
			Soybean			
0-5 cm						
NT	no litter	6.5 a	9.15 b	11.10 b	1.18 b	9.39 a
NT	litter	6.4 a	12.52 a	18.33 a	1.49 a	12.33 a
CT	no litter	5.9 b	6.12 c	9.00 bc	0.80 c	11.27 a
CT	litter	6.2 ab	6.10 c	8.46 c	0.71 c	11.90 a
5-10 cm						
NT	no litter	6.0 a	5.42 a	7.25 a	0.73 a	9.95 a
NT	litter	6.0 a	6.55 a	8.48 a	0.66 a	12.81 a
CT	no litter	6.2 a	5.39 a	7.37 a	0.70 a	10.56 a
CT	litter	6.3 a	5.68 a	7.41 a	0.62 a	11.87 a
10-20 cm						
NT	no litter	5.9 b	4.87 a	5.49 a	0.36 a	15.36 a
NT	litter	5.9 b	5.27 a	6.35 a	0.49 a	12.97 a
CT	no litter	6.3 a	5.07 a	5.24 a	0.53 a	9.88 a
CT	litter	6.4 a	4.78 a	4.88 a	0.49 a	9.89 a
			Corn			
0-5 cm						
NT	no litter	6.5 a	9.53 b	17.32 b	1.28 b	13.48 a
NT	litter	6.5 a	13.79 a	22.47 a	1.82 a	12.33 a
CT	no litter	6.0 b	5.23 d	8.30 d	0.77 c	10.80 a
CT	litter	6.4 a	7.54 c	11.67 c	0.94 d	12.46 a
5-10 cm						
NT	no litter	5.9 b	5.79 ab	8.47 b	0.73 a	11.55 a
NT	litter	6.0 ab	6.42 ab	10.36 a	0.76 a	13.59 a
CT	no litter	6.3 a	5.00 b	7.58 b	0.63 a	11.97 a
CT	litter	6.3 a	6.72 a	9.33 ab	0.71 a	13.22 a
10-20 cm						
NT	no litter	5.6 b	4.30 a	5.89 a	0.45 a	13.00 a
NT	litter	5.7 b	5.14 a	6.94 a	0.58 a	11.91 a
CT	no litter	6.2 a	4.35 a	5.26 a	0.47 a	12.24 a
CT	litter	6.1 a	5.09 a	5.99 a	0.47 a	12.64 a

Means within a column followed by the same letter do not differ significantly (0.10 level)

Table 3. Effect of litter and tillage on soil extractable levels of K, Mg, Ca, P, at the 0-5, 5-10, and 10 -20 cm depths for the soybean and corn cropping systems.

Tillage	Amendment	K	Mg	Ca	P
		------------------- mg kg $^{-1}$ -------------------			
			Soybean		
0-5 cm					
NT	no litter	79 b	184 b	1164 b	64 b
NT	litter	138 a	250 a	1643 a	326 a
CT	no litter	103 b	108 c	630 c	65 b
CT	litter	107 b	115 c	692 c	74 b
5-10 cm					
NT	no litter	42 b	86 a	542 a	29 a
NT	litter	95 a	115 a	681 a	81 a
CT	no litter	59 b	95 a	587 a	36 a
CT	litter	77 b	101a	626 a	52 a
10-20 cm					
NT	no litter	32 b	78 a	408 a	24 a
NT	litter	76 a	87 a	460 a	45 a
CT	no litter	38 b	83 a	477 a	24 a
CT	litter	49 ab	77 a	531 a	31 a
			Corn		
0-5 cm					
NT	no litter	96 c	196 b	1221 b	62 c
NT	litter	187 a	266 a	1859 a	187 a
CT	no litter	70 d	97 d	518 d	50 c
CT	litter	161 b	141 c	896 c	126 b
5-10 cm					
NT	no litter	48 b	89 a	530 a	25 b
NT	litter	137 a	110 a	653 a	81 a
CT	no litter	38 b	95 a	529 a	21 b
CT	litter	120 a	119 a	739 a	72 a
10-20 cm					
NT	no litter	36 b	69 a	336 a	22 ab
NT	litter	103 a	78 a	398 a	50 a
CT	no litter	26 b	79 a	415 a	12 d
CT	litter	81 a	81 a	452 a	34 bc

Means within column followed by the same letter do not differ significantly (0.10 level).

Table 4. Effect of litter and tillage on soil C mineralization, C turnover, N mineralization, and and C:N mineralization at the 0-5, 5-10, and 10-20 cm depths during the 90 day laboratory

Tillage		C Mineralization	C Turnover	N Mineralization	C:N Mineralization
		-------------------- mg kg $^{-1}$		-----------------------	----- g g^{-1}-------
			Soybean		
0-5 cm					
NT	no litter	935 b	8.4 b	62.2 b	15.1 a
NT	litter	1062 a	5.8 c	76.2 a	15.7 a
CT	no litter	857 b	9.5 b	62.1 b	13.9 a
CT	litter	890 b	10.5 a	58.7 b	14.2 a
5-10 cm					
NT	no litter	317 b	4.3 b	15.5 b	23.8 a
NT	litter	392 b	4.6 b	24.5 ab	17.6 a
CT	no litter	566 a	7.7 a	32.2 a	17.6 a
CT	litter	569 a	7.6 a	33.9 a	18.0 a
10-20 cm					
NT	no litter	244 b	4.4 b	9.3 a	40.7 a
NT	litter	351 a	5.6 b	10.1 a	36.3 a
CT	no litter	362 a	7.0 a	8.8 a	30.6 b
CT	litter	346 a	7.1 a	11.4 a	30.2 b
			Corn		
0-5 cm					
NT	no litter	1053 b	9.5 a	63.5 b	16.7 a
NT	litter	1199 a	5.3 c	69.1 b	17.6 a
CT	no litter	809 d	9.7 a	47.0 c	17.3 a
CT	litter	949 c	8.1 b	80.0 a	11.9 a
5-10 cm					
NT	no litter	443 b	5.2 b	16.7 b	21.98 a
NT	litter	470 b	4.5 b	27.2 b	17.4 a
CT	no litter	559 b	7.4 a	25.6 b	19.31 a
CT	litter	659 a	7.1 a	43.4 a	15.6 a
10-20 cm					
NT	no litter	294 b	6.0 b	9.7 a	35.1 a
NT	litter	308 ab	4.5 b	10.6 a	30.8 a
CT	no litter	311 ab	5.9 a	12.1 a	26.4 b
CT	litter	397 a	6.6 a	10.7 a	36.4 a

Means within column followed by the same letter do not differ significantly (0.10 level).

II. MINERALIZATION OF N IN SOILS AMENDED WITH DAIRY MANURE AS AFFECTED BY WETTING/DRYING CYCLES

Abstract

Interest in manure management and its effects on nitrogen (N) mineralization has increased in recent years. The focus of this research was to investigate the N mineralization rates of different soil types in Coastal Plain soils and compare them to a soil from Illinois. Soils with and without dairy composted manure addition were subjected to different wetting/drying cycles (constant moisture at 60% water filled pore space (WFPS) and cycling moisture from 60 to 30% WFPS) under laboratory conditions at three different temperatures (11ºC, 18ºC, and 25ºC). Samples were collected from three different soil types: Catlin (Mollisols), Bama (Ultisols), and Goldsboro (Utilsols). Soil chemical and physical properties were determined to help assess variations in N mineralization rates. Addition of composted manure greatly impacted the amount of N mineralized. The amount of manure-derived organic N mineralized to inorganic forms was mainly attributed to the soil series, with the Catlin (silt loam) producing the most inorganic N followed by the Goldsboro (loam) and then Bama (sandy loam). This was probably due to soil texture and the native climatic conditions of the soil. No significant differences were observed between the constant and cycling moisture regimens, suggesting that the imposed drying cycle may not have been sufficient to desiccate microbial cells causing a flush in N mineralization upon rewetting. Nitrogen

mineralization responded greatly to the influence of temperature with the greatest N mineralization occurring at 25ºC. The information acquired from this study may aid in predicting the impact of manure application to help increase N use efficiency when applied under different conditions (e.g.; climate season) and soil types.

Introduction

In the past two decades, an increased interest in maintaining soil and water quality for a sustainable environment has sparked great concerns in manure management. These concerns have been attributed to increased animal production in confined areas; there has been a decrease in the number of individual farms while the number of animals on large farms has increased in concentrated areas. This has resulted in large amounts of manure being generated that have to be disposed of as a waste. The most feasible method of disposal is land application, but improper land application practices can result in nutrient contamination of groundwater and surface water, which increases human health and environmental risks. Therefore, it is imperative that the effects of manure application to soil under different abiotic conditions are understood in order to effectively utilize nutrients without causing adverse effects to the environment.

Nitrogen (N) mineralization can be useful in quantifying the impact management practices and organic amendments have on soil functions and environmental quality. Understanding N mineralization rates under different abiotic conditions could be important in managing N more efficiently.

Changes in N mineralization are directly related to microbial activity and biomass inputs, which are affected by abiotic factors. Nitrogen mineralization processes are influenced by environmental factors, including temperature and soil moisture (Katterer et al., 1998), soil wetting and drying cycles (Kruse et al., 2004), soil texture (Torbert and Wood, 1992), and soil characteristics (Schjonning et al., 1999; Gordillo and Cabrera, 1997).

Nitrogen mineralization has been shown to accelerate with increasing soil temperature under conditions found in agricultural systems (Cassman and Munns, 1980; Eghball, 2000). It has been reported that decomposition is 3.7 times faster at 25ºC than at 15ºC and 13 times faster at 15ºC than at 5ºC (Vigil and Kissel, 1995). The effect of temperature on the rate of N mineralization can also change with latitude. Campbell et al. (1984) reported that soils in the Northern region of the USA were affected more by incubation temperatures than Southern soils. Campbell et al. (1984) concluded that this was probably due to the decomposition rate, as a function of temperature, which is related to the amount of readily decomposible organic matter in the soil. Soils in Northern climates, where the summer is shorter, have less decomposition occurring in a given year compared to soils in Southern climates that experience long summers.

Mineralization is also greatest when soil moisture is near field capacity and declines with soil drying (Cassman and Munns, 1980). Linn and Doran (1984) showed that in most soils microbial activity is optimum when soil water filled pore space (WFPS) is near 60%, resulting in maximum organic matter decomposition. Anaerobic conditions tend to occur at a WFPS above 60%, thereby slowing the decomposition process. Knoepp and Swank (2002) found that there was a significant interaction between soil moisture content and soil temperature in N mineralization. Soil temperature and moisture probably have the greatest influence on nitrification because of their importance in soil aeration.

Franzluebbers et al. (1994) found that N mineralization can also be related to the amount of soil organic matter (SOM) present in the soil. Agronomic practices that build up SOM by adding crop residues back to the soil greatly impact the fertility of the soil;

thereby, increasing the potential for N mineralization (Wood and Edwards, 1992; Torbert et al., 1999).

Soil texture also affects the rate of N mineralization in soil. Soil texture that is more conducive to the retention of soil organic C and N is associated with increased soil aggregation (Beare et al., 1994). In soils with high amounts of aggregates, the clay-sized particles are bound around organic material; thereby, protecting organic matter from decay (Beare et al., 1994; Jastow 1996). When soil aggregates are destroyed the organic matter is exposed to microbial attack.

Surface soils, in most ecosystems, experience periods of drying followed by relatively rapid rewetting cycles. It has been shown that mineralization rates of N are generally increased for a few days following rewetting of a dry soil (Birch, 1958; Bloem et al., 1992; Cui and Caldwell, 1997; Franzluebbers et al., 2000). Soil undergoes complex physical, chemical, and biological changes under the impact of drying and re-wetting, including changes to soil structure (aggregation), SOM and microflora (Soulides and Allison, 1961; Sorensen, 1974; Utomo and Dexter, 1982).

Wetting and drying cycles have an influence on microbial activity; thereby, affecting decomposition of SOM (Soulides and Allison, 1961; Bloem et al., 1992; Magid et al., 1999). Drying followed by rapid rewetting cycles generally cause an increase in organic substrate available for microbial attack (Soulides and Allison, 1961; Sorensen, 1974). These substrates are partially derived from the death of a portion of the soil organisms upon drying (Lund and Goksoyr, 1980; Bottner 1985) by causing microbes to undergo osmotic shock, which can induce microbial cell lysis (Bottner, 1985; Van Gestel et al., 1992) or lead to the release of intracellular solutes (Halverson et al., 2000). The labile substrates that become available (i.e., C and N compounds in the form of organic material) are rapidly mineralized by the remaining soil microbes, which causes a pulse in mineralization of N and C (Birch, 1959; Keift et al, 1987). Wetting and drying cycles have also been shown to cause soil aggregates to break apart, exposing physically protected organic matter to become available for further degradation (Adu and Oades, 1978, Lundquist et al., 1999). The organic matter that was previously unavailable can be rapidly mineralized by the microbial community (Appel, 1998).

A better understanding of soil under different textural, moisture, and temperature conditions from different climatic regions is needed in order to develop management practices that optimize N derived from manure. This could aid in a better quantification of N use efficiency, because, it is presently estimated that efficiency is about 30 to 50% in most agricultural soils, subjecting the soil to excess leaching or runoff (Delgado, 2002). Therefore, information is needed on the relative rate of N being mineralized in soils amended with manure as affected by different climatic conditions. The objective of this study was to determine the impact of manure application on N mineralization in soils subjected to different temperatures and wetting and drying cycles under laboratory incubation conditions.

Materials and Methods

Soil samples were collected from an ongoing precision agriculture experiment established in 2000 at Auburn University's E.V. Smith Experiment Station located in Macon County, Alabama and compared to soil under long-term pasture management located in Champaign County, Illinois. Bulk samples (0-15 cm) were collected from field plots and transported back to the laboratory for analysis.

Soil Description

Average annual precipitation and temperature in Macon County, AL is 1422 mm and 17°C, respectively. Two soil series evaluated from Macon County were Bama and Goldsboro. The Bama series consists of very deep, well-drained soils formed in loamy sediments. Slopes range from 0 to 5 percent. These soils are fine-loamy, siliceous, subactive, thermic Typic Paleudults. Goldsboro series consists of very deep, moderately well drained soils formed in loamy sediments. Slopes range from 0 to 2 percent. These soils are fine-loamy, siliceous, subactive, thermic Aquic Paleudults. The farming practice was comprised of conventional tillage, which receives inorganic fertilizer in a continuous cotton/corn rotation. The average annual precipitation and temperature in Champaign County, IL are 938 mm and 11˚C, respectively. Catlin soil series consists of moderately well drained, moderately permeable soil on till plains and moraines. These soils formed in loess and in the underlying loam glacial till. Slopes range from 2 to 7 percent. These soils are fine-silty, mixed, mesic Typic Arguidolls. Farming practice for this soil consisted of pasture management.

Laboratory Analysis

Air-dried samples were ground to pass thorough a 2 mm sieve and subjected to chemical and physical analysis. Total C and N were determined by the DUMAS dry combustion method using a CN LECO 2000 analyzer (LECO, St. Joseph, MI). Soil characteristics [pH, electrical conductivity (EC), cation exchange capacity (CEC), soil extractable Ca, Mg, K, P, Fe, Mn, Zn, Cu, B, and Na as described by Hue and Evans

(1986) and particle size] were measured by the Auburn University Soil Testing Laboratory. Composted dairy manure was obtained from compost piles located on the Auburn University dairy farm. Total N was measured using the DUMAS dry combustion method as stated above. Soil properties are present in Table 1.

Incubation Study

An aerobic incubation experiment was performed similar to the procedure described by Honeycutt et al. (2005b). Treatments were replicated in triplicates with 250g of soil (oven-dried weight basis) amended with the appropriate amount of composted dairy manure to give 350 kg organic N kg^{-1} soil. Samples were thoroughly mixed and placed into 2 L Mason jars. Deionized water was added to bring the soil moisture to approximately 60% water filled pore space (WFPS). The controls were treated with deionized water only. Samples were incubated at three different temperatures (11˚C, 18˚C, and 25˚C) and exposed to two wetting/drying regimes. Half were maintained at constant moisture (60% WFPS) and the other half were cycled from 60% to 30% WFPS. Samples were aerated daily (2 hrs), at which time moisture content was adjusted gravimetrically as required. Cycled soils were allowed to reach 30% WFPS to complete a drying cycle. After undergoing a drying cycle, soils were re-wetted by adding the appropriate amount of water to bring the soil back to 60% WFPS. On the day that cycled soils were rewetted to 60% WFPS, a sub-sample was taken from the cycled soil and its corresponding soil at constant moisture. Concentration of NH_4 and NO_2 + NO_3 were determined by extraction using 2 M KCl as described by Keeney and Nelson (1982) and measured colorimetrically using Bran-Luebbe Autoanalyzer (Bran-Luebbe, Norderstedt, Germany).

The experiment was analyzed as a completely randomized factorial design with three soil types amended with and without manure and three incubation temperatures. Statistical analyses were performed using GLM procedure of SAS (SAS Institute, 1985), and means were separated using least significant difference (LSD) at an *a priori* 0.10 probability level.

Results and Discussion

Most of the N assimilated by plants is derived from inorganic N pools (NH_4 and NO_3) generated through the processes of N mineralization and nitrification. In this study, net N mineralization rates were determined from the change in the soil inorganic-N pool size over time where N mineralization at day 0 was subtracted from each cycle.

The addition of dairy composted manure to the incubated soil significantly increased the amount of inorganic N produced at the 11, 18, and 25°C temperature regimens combined across both the constant and cycling moisture regimens (Figure 1). The amount of ammonium produced was not significantly higher in the soil containing manure compared to that of non-amended soils. This result differs from that reported by Honeycutt et al. (2005a) who used dairy manure in their study and is probably due to the fact that dairy compost was used in this study and the NH_4 had already undergone nitrification (NH_4 conversion to NO_3) or had been volatilized.

In the succeeding text the amount of inorganic N is expressed as the sum of NH_4 and NO_3. Also, the amount of inorganic N mineralized was corrected by subtracting soil amended with composted dairy manure from those unamended soils. It is assumed that ammonia volatilization was negligible due to the fact that most of the N would be in a stable form (NO_3) and the compost was incorporated in to the soil (Schilke-Gartley and Sims, 1993; Hengnirun et al., 1999). Denitrification was also assumed to be negligible since the WFPS was not greater than 60% (Linn and Doran, 1984).

N mineralization as affected by soil type

Significant differences were observed resulting from soil types (Table 2). The Catlin soil was significantly higher than the Goldsboro and Bama soils, for day 0 and all of the sampling periods (Table 3). There were no significant differences observed between the Goldsboro and Bama soils for all of the sampling periods. These soils were collected from the Southeastern region of the U.S. from soils under precision agriculture management. These results showed that land-use planning for agronomic and environmental sustainability may not be useful in manure management of soil that is historically nutrient poor.

By evaluating the soil characteristics (Table 1), it can be shown that the Catlin soil greatly varies from the other two soils. Catlin soil has a higher CEC, total C and total N. The total amount of inorganic N mineralized in the Catlin soil (Table 3) was 175 and 215% higher than the Goldsboro and Bama soil, respectively. Even though the Goldsboro soil has higher clay content and is considered to be a better soil than the Catlin, N mineralization was probably influenced by CEC, total N and C, and organic matter, resulting from the climate where the soil originated. Soil under cool wet climates tend to build up organic matter (e.g. Catlin), while soil from hot humid climates (Goldsboro and Bama) tend to promote organic matter decomposition and high rainfall events tend to cause leaching. Since N mineralization is a process that is mediated by microbial activity, the microbial populations of soil from the Northern climatic environments were probably more adapted to relatively low soil temperature. Therefore, causing a higher mineralization in the Catlin soil at 11ºC. As soil temperature increased the Catlin soil experienced a faster decomposition due to the residual organic matter.

Unlike results reported by Honeycutt et al. (2005a), performing a similar experiment, we did not observe N immobilization in the Catlin soil. Honeycutt et al. (2005a) reported that soil with higher clay and silt contents showed greater N immobilization when amended with dairy manure compared to the other soils. Sorensen et al. (1994), while utilizing sheep feces; their study also found N immobilization in soil that had the highest clay content. These soils were all compacted to the same bulk density. In the current study, the soil was compacted to the corresponding bulk density observed in the field where they were collected. Our results were similar to the laboratory incubation study of Linn and Doran (1984), who found slightly less respiration with soil of different compactions across a range of WFPS compared with loose, porous soils. Torbert and Wood (1992) observed from sandy coastal plain soils that respiration (measure by amount of CO_2 evolved from soil) decreased 65% when bulk density was increased from 1.4 to 1.8 Mg m^{-3} at 60% WFPS. Thus, their results showed that bulk density could impact microbial activity. This also helps explain the higher N mineralization rate in the Catlin, which had the lowest bulk density (1.2 Mg m^{-3}).

N mineralization as affected by soil moisture

Soil incubation samples in this experiment were dried at different rates. Drying times varied depending on soil characteristics and temperature. Incubation jars containing a given soil at a particular temperature were sampled when the cycling moisture content reached 30% WFPS. In general, the amount of time required between each cycling period (to reach 30% WFPS) decreased as temperature increased.

There was no significant difference observed between the constant and cycling moisture regimens for all soil types regardless of soil texture (Table 2 & Table 4). These results are similar to that of Griffin et al. (2002) and Honeycutt et al. (2005a). Unlike the results of other researchers (Birch, 1964; Carbera, 1993) who found that air drying the soil caused a flush in N mineralization, our results show that drying the soil to a 30% WFPS was not sufficient to desiccate microbial cells, leading to a flush in N mineralization upon rewetting the soil (Griffin et al., 2002). Instead, the soil microbes probably adjusted the intercellular solutes to the given water stress level.

Comparing this study to other studies, the drying cycles were performed at a much slower rate than previous reports. The drying cycles in most of the other studies occurred within 1-3 days, which can have an effect on the survival of soil microorganisms (Mikha et al. 2005). The drying period in this experiment at times took up to three weeks. It has been reported that soil-drying occurring at a slow rate allows for microbial metabolic adjustment that could reduce mortality (Chao and Alexander, 1984; Hartel and Alexander, 1986; Roberson and Firestone, 1992). Also, the rate of soil drying conducted in this experiment is probably more comparable to that experienced under normal field conditions.

N mineralization as affected by soil temperature

The amount of inorganic N mineralized was significantly affected by temperature (Table 2), with the highest two temperatures mineralizing the most in this experiment (Table 5). Dalias et al. (2002) reported that when soils were incubated at 4, 10, 16, 25, and 30ºC, N mineralization increased with temperature. This suggests that the increase in N pool was related to temperature, indicating an influence of thermal conditions on the degree of microbial exploitation of organic N sources.

The soil and temperature interaction was significant (Table 2). The Catlin soil showed the greatest difference between temperature regimens (Table 6). This is similar to the finding of Dalias et al. (2002) who found that the optimum temperature for nitrification showed a good correlation with the geographic origin of the soil. This was also in accordance with Campbell et al. (1984) who stated that the decomposition rate (as a function of temperature) was related to the amount of readily decomposable organic matter in the soil. Not all organic material decomposes at the same rate. For example, water-soluble carbohydrates, amino acids, and amino-sugars have a short residence time in a soil. With an active microbial population under typical summer field conditions, these components are usually consumed within a week or two. Organic matter that is humified or derived from more resistant materials (lignin associated) has longer residence time under the same field conditions and may last years. In northern climates, where summers are cooler and shorter, less decomposition of organic litter and native soil organic matter will occur in a given year, resulting in a buildup of more easily decomposed material over time. In southern climates, summer is longer and warmer, allowing for more complete decomposition of most of the litter material. Over a long period in Southern soils, only the most resistant organic materials are left behind, whereas in Northern soils, both easily decomposable and resistant materials can accumulate (Campbell et al., 1984).

Conclusions

Nitrogen is one of the basic components of life and has a very important function in soil ecosystems. Understanding the effects of manure management in the soil ecosystem and how it relates to the nitrogen cycle is essential in determining N use efficiency. From this study, it was concluded that temperature and region of soil origin could impact the rate of N mineralization of soils amended with composted dairy manure. Contrary to other studies, soil subjected to cycling moisture regimens did not have a significant affect on the amount of inorganic N produced. The soil water stress level imposed in this study suggests that the range may not have been high enough or the drying cycle rapid enough to cause a change in N mineralization. This was probably due to the fact that the soil was not dried enough to desiccate microbial cells, which normally leads to a flush in N mineralization. There was less inorganic N mineralized from soils of the Southeastern region (Goldsboro and Bama: Alabama soils) compared to that from the Midwestern region (Catlin; Illinois soil). Although the particle size analysis of the Goldsboro soil indicates that it is a finer textured soil, the Catlin was more fertile due to the climate from where the soil originated, therefore, having a higher N mineralization potential. Minimal differences were shown between the soil types from the Southeastern region. This indicates that the use of precision management in land use planning for N mineralization of different soil types of nutrient poor soils (resulting from humid climates and high leaching potential) may not be useful. This information can also be useful in determining the timing and impact of manure application in different climates and seasons depending on the soil type. Further studies are needed that encompass more soils from different climatic regions in order to accurately determine the effects of climatic conditions and wetting and drying cycles on soil N mineralization

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Table 1. Soil Properties

Soil Series	pH	EC	BD	CEC	Total C	Total N	Sand	Silt	Clay
		mmhoscm^{-1}	g cm^{-3}	cmol kg^{-1}	------ g kg-1 ------		--------------- % ---------------		
Bama	6.7	0.1	1.46	5.8	7.53	0.72	66.25	21.25	12.50
Goldsboro	6.5	0.1	1.58	6.3	7.08	0.78	33.75	48.75	17.50
Catlin	7.2	0.1	1.2	26.77	40.66	3.14	18.75	66.25	15.00

Table 2.Analysis of variance for the effects of soil type, moisture regimen, and temperature.

	Day 0	**Cycle 1**	**Cycle 2**	**Cycle 3**	**Cycle 4**
Soil (S)	<.0001	<.0001	<.0001	<.0001	<.0001
Moisture (M)	0.227	0.0775	0.5465	0.2775	0.8594
Temperature (T)	0.055	0.0003	0.0001	<.0001	<.0001
S x M	0.2851	0.2225	0.7612	0.1104	0.5512
S x T	0.2511	0.3737	0.0003	0.0049	0.0005
M x T	0.2006	0.0125	0.479	0.0233	0.0894
S x M x T	0.3418	0.3316	0.0433	0.1258	0.0311

Analysis of variance P > F LSD (0.10)

Table 3. Mean inorganic N concentration as affected by soil type over time in mg kg $^{-1}$

Soil	**Day 0**	**Cycle 1**	**Cycle 2**	**Cycle 3**	**Cycle 4**
Catlin	3.497 b	80.20 a	87.66 a	95.62 a	102.16 a
Goldsboro	1.147 c	28.00 b	28.27 b	37.93 b	39.09 b
Bama	1.244 c	26.93 b	23.17 b	30.55 b	34.78 b

Table 4. Mean inorganic N concentration as affected by soil moisture over time in mg kg $^{-1}$

Moisture	**Day 0**	**Cycle 1**	**Cycle 2**	**Cycle 3**	**Cycle 4**
Cycling	2.00 a	47.09 a	45.62 a	56.13 a	58.89 a
Constant	1.92 a	42.10 a	47.14 a	53.26 a	58.47 a

Table 5. Mean inorganic N concentration as affected by soil temperature over time in mg kg $^{-1}$

Temp	**Day 0**	**Cycle 1**	**Cycle 2**	**Cycle 3**	**Cycle 4**
25°C	2.05 a	48.47 a	52.58 a	61.06 a	67.73 a
18°C	2.00 a	48.91 a	47.59 a	59.31 a	60.35 b
11°C	1.84 ab	37.75 b	38.92 b	43.72 b	47.95 c

Table 6. Interaction between soil type and temperature as a measure of inorganic N over time in mg kg^{-1}

Soil	**Temp**	**Day 0**	**Cycle 1**	**Cycle 2**	**Cycle 3**	**Cycle 4**
Catlin	**11°C**	3.40 a	68.44 a	68.55 a	77.32 a	79.01 a
	18°C	3.60 a	85.49 a	91.64 b	99.28 b	106.87 b
	25°C	3.48 a	86.67 a	102.79 b	110.27 b	120.61 b
Goldsboro	**11°C**	1.02 a	24.32 a	27.38 a	30.05 a	35.82 ab
	18°C	1.04 a	30.14 a	29.95 a	48.60 b	43. 01 a
	25°C	1.39 a	29.54 a	27.46 a	35.11 ab	38.41 ab
Bama	**11°C**	1.09 a	20.50 a	20.83 a	23.79 a	29.01 a
	18°C	1.36 a	31.09 a	21.18 a	30.07a	31.15 ab
	25°C	1.28 a	29.20 a	27.42 a	37.80 a	44.17 b

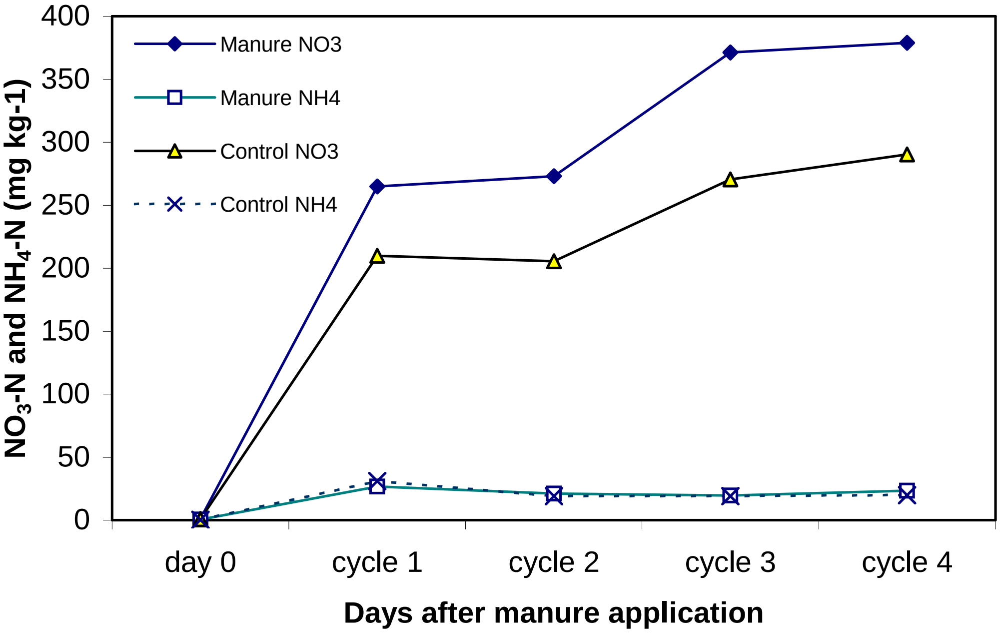

Figure 1. Concentration of nitrate and ammonium following manure application for the Goldsboro soil amended with and without manure combined across three incubation temperatures and the wetting and drying cycles.

III. A SEASONAL NITROGEN MINERALIZATION STUDY AS INFLUENCED BY SOIL PROPERTIES AND LANDSCAPE POSITION

Abstract

Information is needed on mineralization and dynamics of N under normal field conditions in order to better develop management practices that will optimize the amount of N derived from manure. Thus an *in situ* field study was conducted to evaluate N mineralization patterns using three different soil types located in close proximity of each other during the summer and winter months. The three Coastal Plain soils investigated were Bama (Ultisol), Lynchburg (Ultisol) and Goldsboro (Ultisol). Dairy composted manure was incorporated into *in situ* soil cores at a rate of 350 kg N ha^{-1} and compared to an unamended control. Addition of dairy composted manure greatly increased the mineralization of N. This was most evident during summer months, suggesting seasonal timing of application influences mineralization. These seasonal patterns in N mineralization were mostly affected by temperature. During winter (temperature ~ 10° C) N mineralization was minimal, whereas during summer N mineralization was greater due to higher temperatures (25-30°C). Landscape and soil texture also played an important role in mineralization. During winter months the soil type with the greatest percent sand located in a low lying area lost most of the added N from compost compared to other soils. However, during summer the loam soil with the greatest field capacity

mineralized the most N. It was also noted that the soil with the highest bulk density was more effective in retaining N in the soil compartment of the *in situ* core. These results show that when applying manure for crop production field scale variability of soils and landscapes needs to be taken into account.

Introduction

Nutrient availability from manure in crop production has been recognized for many centuries. Prior to the use of inorganic fertilizer, manure was the primary source of nutrients for plants. Recently, there has been a renewed interest in use of manure due to increases in animal production in confined areas generating large amounts of waste. This interest is attributed to concerns in maintaining sustainable agricultural production while at the same time preserving the environment. Therefore, more knowledge is needed on manure mineralization rates as it affects inputs, losses, and transformation of N in the soil during the growing (summer months) and non-growing season (winter months) in order to develop management practices that increase nitrogen use efficiency (NUE).

When applying manure during the growing or non-growing season, farmers usually apply it at uniform rates, assuming the N sources, sinks, and mechanisms for loss are constant across fields (Delgado, 2002), thus failing to account for variability existing in most agricultural fields. This leaves agricultural fields sometimes vulnerable to N loss. It has been estimated worldwide that N use efficiency is about 30 to 50% in most agricultural fields, subjecting the excess to leaching or runoff. A large contribution to NUE could be attributed to soil type, whether it is a coarse textured soil (sandy soil) where N losses are potentially subjected to NO_3 leaching, or a fine textured (clay loam) soil where the N loss may be less due to higher retention capability of the soil. It has been reported that manure increases NO_3 leaching compared to inorganic N fertilizer when applied at equivalent N rates (Roth and Fox, 1990; Jemison and Fox, 1994). This is attributed to greater N mineralization with manure during winter months, generating NO_3 during periods without crop uptake.

Nitrogen mineralization is the driving force behind the transformation of N in manure to a form that is readily accessible by plants. This mineralization process is affected by several factors such as the organic composition of the residue (Whitmore, 1996), soil temperature and water content (Katterer et al., 1998), drying and rewetting events (Kruse et al., 2004), soil texture (Torbert and Wood, 1992), and soil characteristics (Schjonning et al., 1999; Gordillo and Cabrera, 1997b). It has been reported in some laboratory studies that N mineralization from the same organic residues have shown differences in the amounts of N released from different soils (Whitmore and Groot, 1997; Gordillo and Cabrera, 1997b; Thomsen and Olesen, 2000). Observed differences may be attributed to adsorption capacity of the soil to bind organic N (Van Veen et al., 1985), increased aeration in sandier soil (Thomsen et al., 1999), and different C to N ratios (Hassink, 1994; Hassink et. al., 1994).

Numerous studies have evaluated the effects that soil physical and chemical characteristics have on mineralization rates of soils amended with manure. These studies have been important in achieving an understanding of manure mineralization. However, most N mineralization studies have been carried out under laboratory conditions (Castellanos and Pratt, 1981; Chae and Tabatabai, 1986; Bonde and Lindberg, 1988; Cabrera et al., 1993), which often overestimated mineralization patterns that occur under normal field conditions. This is because laboratory incubation does not take into account the interactions between soil variations in organic matter, temperature, moisture, and different soil types often found in the field.

The *in situ* resin core method has been used to observe N mineralization rates in forest ecosystems (Distefano and Gholz, 1986; Binkley et al., 1992). This method was also used to study mineralization rates in dryland agroecosystems (Kolberg et al., 1997) and Eghball (2000) performed a N mineralization study on beef cattle manure and beef cattle compost during the growing season. The use of the *in situ* core method can be costly and labor intensive, but valuable in developing an index of the amount of N mineralized under natural field conditions with the use of undisturbed soil (Eghball, 2000). The information obtained can also be useful in understanding the potential rate of mineralization and dynamics of N from applied manure under different climatic conditions.

Information is needed on the relative rate of N being mineralized (released), retained (tied up), and potentially lost (leached) from soils treated with manure. Nitrogen mineralization rates in previous studies using the *in situ* core method have not taken into account the spatial variability that is often encountered in normal agricultural fields. Most large-scale agricultural fields have varying degrees of soil types resulting from different soil forming factors and landscapes. Nitrogen pools in one soil type may differ from other soil types due to inherit soil-forming properties. Therefore, the objectives of this study was to determine N mineralization during the growing season and non-growing season from composted dairy manure applied to three soils developed from different soil forming properties.

Materials and Methods

Soil samples were collected from an ongoing precision agriculture experiment located at Auburn University's E.V. Smith Experiment Station located in Macon County, Alabama (Terra et al., 2006). Soils were collected from field plots that have not received manure within the last 10 years.

Soil Description

The climate in Macon County consists of long hot summers due to moist tropical air from the Gulf of Mexico. Mean annual precipitation is 1422 mm with most of this (52%), occurring between April and October. The average daily temperature is 17°C. The three soil series evaluated were Bama, Goldsboro and Lynchburg. These three soils were chosen because they are found in close proximity to one another, yet different in texture. The Bama series consists of very deep, well-drained soils that formed in loamy sediments. Slopes range from 0 to 5%. These soils are fine-loamy, siliceous, subactive, thermic Typic Paleudults. Goldsboro series consists of very deep, moderately well drained soils that formed in loamy sediments. Slopes range from 0 to 2 percent. These soils are fine-loamy, siliceous, subactive, thermic Aquic Paleudults. The Lynchburg series consists of very deep, somewhat poorly drained soils that formed in loamy sediments. Slopes range form 0 to 2 percent. These soils are Aquic Paleaquults and Paleaquults. The farming practice is comprised of conventional tillage, which receives inorganic fertilizer in a continuous cotton/corn rotation.

Laboratory Analysis

Air-dried samples were ground and passed through a 2 mm sieve and subjected to chemical and physical analysis. Total C and N were determined by the DUMAS dry combustion method using a CN LECO 2000 analyzer (LECO, St. Joseph, MI). Soil characteristics; pH, soil effective CEC, soil extractable nutrients (Ca, Mg, K, P, Fe, Mn, Zn, Cu, B, and Na) and particle size analysis were measured by Auburn University Soil

Testing Laboratory on 0-15 cm soil depth increment using methodology described by Hue and Evans (1986).

In situ mineralization study

A field *in situ* mineralization study was conducted by placing polyvinyl chloride (PVC) plastic cylinders in the surface 20 cm of the soil profile according to procedures described by Honeycutt et al. (2005b). Nitrogen mineralization and nitrification rates were measured using these *in situ* soil cores. These *in situ* soil core (microplot cylinders) incubation chambers were 6.25 cm dia and 20.32 cm in length. Intact cores were taken by driving the PVC cylinder into the top 20 cm of the soil profile using a hydraulic core sampler. Vegetation was removed from the surface portion and roots were severed in order to prevent N loss to plant uptake. The core samples were collected and brought to the laboratory. The top 4 cm of the soil core in the microplot cylinders were removed and the appropriate amount of dairy manure was added and thoroughly mixed to give 350 kg N ha^{-1} applied to a 15 cm depth (Figure 1.) The soil was gently packed back into the microplot cylinder. The unamended soil underwent the same procedure excepted without manure in order to subject the soil to the same disturbance. Anion and cation exchange resin (JT Baker Inc., Phillipsburg, NJ) was placed in the bottom of each cylinder to capture leachate. The ion exchange resin bags were placed in plastic bags prior to installation to maintain humidity of the exchange resins. Soil cores were transported back to the field and inserted in fallow ground. Dataloggers (HOBO Weather Station, Onset Computer Corporation, Pocasset, MA) were also used with soil cores to continuously monitor soil temperature and soil moisture. The tubes were placed in the ground on the 12 day of January 2004, and 25 day of May 2005.

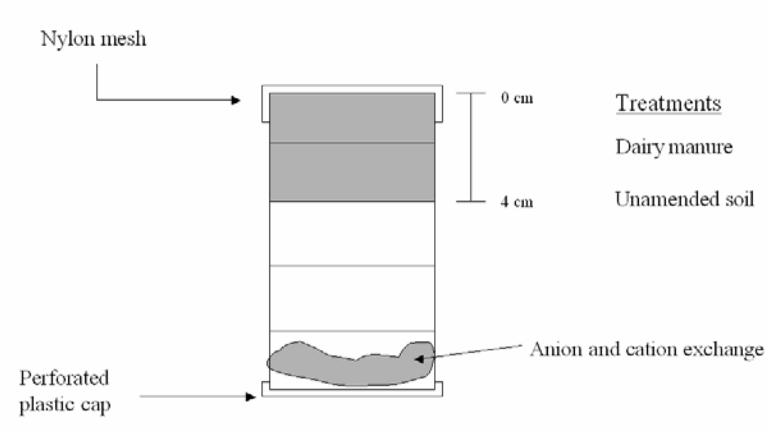

Figure 1. Design of the microplot cylinder used in the winter 2004 and summer 2005 *in situ* mineralization study.

Soil cores were collected and returned to the laboratory for analysis on 0, 3, 7, 14, 21, 49, and 70 days after manure application by randomly selecting and removing six cylinders from each plot. On each sampling day, soil cores were collected and transported to the laboratory in a cooler. Inorganic N content for each incubated sample was analyzed by taking a sub-sample from fresh soil extracted using 2M KCl (5g moist soil in 50 ml of extractant). Extractions were carried out by shaking the soil samples for 1 hr on an orbital shaker at 180 rpm. Resin beads were also extracted by shaking with 250 ml of 2 M KCl for 1 hr. Soil extracts were allowed to settle for 2 hr, and then passed through # 42 Whatman filter paper. The extracts were frozen until analysis (Keeney and Nelson, 1982). Ten-gram soil sub-samples were also taken from the core and dried for 24 h at 105˚C to determine gravimetric moisture content on each sample.

The amount of net N mineralized from substrate addition was compared to the mineralized N of the control treatments. Basically, the control (non-fertilized check) was used to provide an indication of the amount of native soil N that was mineralized. The amount of N mineralized was expressed as mass of N per unit mass of dry soil.

The treatments of the *in situ* study consisted of a total of 3 soils X 2 manure additions (with and without) X 6 replications X 8 sampling dates, which equals a total of 288 experimental units. The experiment was analyzed as a completely randomized factorial design with three soil types amended with and without manure. Statistical analysis were performed using a GLM procedure of SAS (SAS institute, 1985), and means were separated using least significant difference (LSD) at an *a priori* 0.10 probabililty level.

Results and Discussion

Nitrogen observation in the microplot cylinder

Nitrogen mineralization was observed *in situ* through use of microplot cylinders in order to evaluate the effects of soil interactions in its natural environment during summer and winter months. Microplot cylinders used in this study were designed and constructed similar to that of Honeycutt (1999) so that the N leached from the soil in the cylinder would be captured and retained by an ion exchange resin. This setup gives a more precise measurement of the N mineralized by accounting for the N retained in the soil and N that has leached from the soil. The total amount of inorganic N observed in this study, extracted from the soil and resin, suggests that a substantial amount of N was mineralized in the soil and transported to the resin (Table 3). Inorganic N extracted from the resins increased with time in both the summer and winter months. During both seasons more inorganic N was observed in the resin compared to that retained in the soil, suggesting that the N in the soil was very dynamic. Most of the resin inorganic N was mainly in the form of NO_3. This was probably due to NH_4-N ions being strongly adsorbed by soil particles, or rapidly transformed to NO_3 in the process of nitrification, thereby leaving most of the N leachate in the form of NO_3. The use of the microplot cylinders was effective in collecting inorganic N mineralized from the soil. These results suggests that the use of microplot cylinders may be a useful tool in understanding N transformation in soils to better predict management practices in agronomic fields under natural environments.

Manure Mineralization

Dairy composted manure additions significantly increased the plant available N, extracted from the soil and resin, released during both seasons combined across all three soil types (Fig. 2). The added N from the dairy compost was plant available immediately after incorporation into the soil. This is similar to the findings of other researchers (Egball, 2000; Gordillo and Cabrera, 1997).

Dairy compost additions increased the concentration of inorganic N (NH_4 and NO_3) extracted in the soil compartment at all sampling dates compared to the unamended soils (Fig. 3). Concentrations of NH_4 observed in the soil were higher the first few weeks after incorporation of dairy compost and declined throughout the course of the incubation to levels observed in the control soils (Fig. 4). However, concentrations of NO_3 in the soil increased throughout the study. This can be attributed to rapid nitrification (NH_4 to NO_3) occurring in the latter part of the experiment. These finding are similar to that of Honeycutt et al. (2005) who observed the same phenomenon in an incubation experiment.

Significant increases in N mineralization in the soil compartment associated with dairy compost additions also corresponded to an increase in the concentration of inorganic N captured by the ion exchange resin. The increased concentration of inorganic N in the ion exchange resin resulting from manure addition was mainly in the form of NO_3, suggesting that under favorable conditions soil amended with manure could potentially increase the amount of N lost from the soil profile. Control soils (unamended soils) also experienced substantial amount of N loss from the soil compartment. The observed loss in inorganic N can be attributed to mineralization of native soil organic matter (Table 4).

Seasonal Mineralization

In the succeeding text, inorganic N will be expressed as the sum of NH_4 and NO_3. Contribution of residual N from previously added manure or other organic sources are assumed to be minimal since manure had not been previously added to the soil for at least 10 years. Nitrogen loss from volatilization was considered to be minimal due to the fact that compost was utilized in this study and it was incorporated into the soil. Dentrification was also assumed to be negligible due to the sandy nature of the soil, which promotes soil aeration and water infiltration.

A comparison of N mineralization rates between the winter and summer months indicate that mineralization was greatly affected by season. As expected, the total amount of inorganic N mineralized (soil + resin) was significantly higher during the summer months (715 and 520 kg N ha^{-1} for manured and control soil, respectively) compared to that of the winter months (90 and 63 kg N ha^{-1} for manured and control soil, respectively).

The concentration of inorganic N observed during the winter months in the soil compartment of the control soil (soil without manure) was primarily the same at day 0 as it was at the end of the study (Fig. 5), suggesting that N mineralization was minimal and the amount of N retained in the soil was in equilibrium with the amount lost. The concentration of inorganic N observed in the manured soil (soil compartment of cylinder) decreased over time from 22 kg N ha^{-1} (day 0) to 14 kg N ha^{-1} (day 70) during the winter months. This decrease in inorganic N from the soil compartment can be correlated to minimal N mineralization occurring and leaching of the added inorganic N from the manure (due to the dynamic properties of N in the soil).

During the summer (Fig. 5), the amount of inorganic N extracted from the soil compartment increased with time for both the control (13 to 23 kg N ha^{-1}) and manure amended soils (24 to 35 kg N ha^{-1}). The manure soil produced more inorganic N compared to the unamended soils. This increase in inorganic N retained in the soil during the summer can be attributed to mineralization occurring and the accumulation of plant available N. Under normal conditions inorganic N continues to accumulate in the soil until it is taken up by a plant or lost by leaching.

Concentration of inorganic N extracted from the ion exchange resins increased over time for the control and composted manured soils during the summer and winter months (Table 5 and 6). The inorganic N collected from the resin during the summer months was significantly higher than that collected during the winter months, suggesting that as mineralization increases so does the potential for N loss. Inorganic N collected in the resins during winter months was mainly attributed to NO_3 leaching of plant available N contained in the composted manure because mineralization was minimal. Also, water percolation is assumed to be higher during winter months suggesting that the NO_3 that is retained or added to the soil has a greater chance of being lost.

Seasonal mineralization rates corresponded to soil moisture and soil temperature. Although, the water holding content of the soil is higher during winter month's temperature appears to be the most limiting factor. Cool soil temperatures of approximately 10 º C or less were observed for the first 45 days after manure application (Figure 7). This probably accounts for the low mineralization rates resulting in slow decomposition of the added manure. However, during the summer months the soil temperature ranged from 25 º C to 35 º C, which, likely, increased microbial activity, and subsequent N mineralization. In general, maximum N mineralization occurs when soil temperatures are between 25 and 35 º C (Wang et al., 2006; Nicolardot et al., 1994; Stark and Firestone, 1996). Also, Ladd et al. (1996) report that more rapid decomposition of soil organic substance occurs from a combination of warmer soil temperature and better aeration, which is often observed during the summer months due to warmer temperature and low water content of the soil.

Moisture also affected the amount of N retained in the soil compartment. As previously stated during the winter months the amount of inorganic N observed in the soil compartment decreased over time. This decrease in inorganic N concentrations followed rainfall patterns. These findings are similar to that of Watts et al. (2006) who observed N loss during winter months after rainfall events from fall application of manure. During the summer months soil moisture also affected N mineralization (Fig. 8 and Table 5). Fluctuations in soil moisture content were observed corresponding to multiple rainfall events. This was most evident between day 28 and day 49 (highest N mineralization occurred) where there was an increase in moisture content following a dry period, lasting from day 21 to day 28. A combination of the two events resulted in an increase in N mineralization. Higher field moisture contents experienced during this period were probably the overriding factor increasing N mineralization, suggesting that an increase in soil moisture caused an increase in microbial activity. Also during this three week period between sampling periods (day 28 to day 49) the average water filled pore space was approximately 62% compared to 44 % observed between day 21 and day 28. It has been stated that microbial activity is optimum at a soil water filled pore space of 60% (Linn and Doran, 1984).

Soil

In this study, observations of the control soils were utilized to estimate background levels of mineralization originating from sources other than the mineralization of the dairy compost – presumably soil organic matter. No significant differences in the control soils were observed in the soil compartment of the microplot cylinder resulting from different soil types during the winter months at any soil sampling date except for day 0 ($P < 0.05$) (Fig. 5). This was probably due to initial N immobilization, suggesting that all of these soils regardless of their soil type or field origin retained the same amount of inorganic N in the soil under normal conditions.

The addition of manure to soil during winter months was greatly affected by soil type. The Lynchburg soil, a loam soil (located in a depression), had the highest total N mineralization (soil + resin) during the winter months (Table 6). This soil had a lower bulk density compared to the Bama soil (sandy loam), but somewhat similar to that of the Goldsboro (loam), Table 2. The clay content of this soil was similar to that of the Bama (sandy loam). This soil also had a higher CEC, total C and total N in comparison to the other soils (Table 1). The infiltration rate was also greater, probably due to the sandy nature of the soil. During the first 21 days after manure application, the amount of inorganic N gradually increased in the Lynchburg soil (soil compartment, Fig. 5) and contained significantly higher inorganic N at day 14 ($P < 0.02$) and day 21($P<0.04$) after which it was lost to leaching, into the ion exchange resin, by day 28 (Fig. 6). Most of the inorganic N captured in the ion resins was mainly in the form of NO_3. This suggests that sandy soils located in depression areas in agronomic fields are potentially susceptible to leaching during winter months. Also, the sandy soil had more aeration, which promoted mineralization. There was also less water held in the soil, suggesting water percolation through this soil was greater. It has been reported that soil texture influences how rapidly N loss occurs. This influence of soil texture in sandy soil is documented by Delgado et al (1999) and Delgado (2001) who observed more leaching of N in soils with more sandy texture (Follett and Delgado, 2002).

Significant differences were observed between the control soils during the summer months. The Bama (control) soil mineralized and retained significantly more inorganic N in the soil compartment than any of the other soils. This soil also had the lowest amount of N loss from the soil compartment compared to the other soil in the first few days after the intiation of the study. Addition of manure during the summer was also greatly affected by soil type. The Bama soil with manure was similar to the Bama control soil (soil compartment). It contained significantly more inorganic N in the soil compartment ($P<0.04$) throughout the study, except for day 0 and day 49 (Fig. 5). The inorganic N decline on day 49 was due to N loss from the soil compartment corresponding to an increase in inorganic N observed in the resins (Fig. 6). Higher concentration of inorganic N retained in the soil compartment of the Bama soil can be attributed to the higher soil bulk density. The soil was able to retain more N in the early stages after manure application because the soil was more compacted and did not promote leaching. Once the soil reached a threshold where moisture content was optimum, then N was lost from the soil. Also the amount of inorganic N observed in the resin was less in the Bama soil compared to the other soil until the end of the study. This data confirms that more N was retained in the soil compartment, suggesting that the potential for NO_3 leaching is less in high bulk density soils compared to lower bulk density soils.

The Goldsboro soil mineralized the most inorganic N (soil + resin) compared to the other two soils (Table 5). This was likely due to the ability of the soil to maintain the highest soil moisture content compared to the other soils. The percent sand, silt, and clay in the Goldsboro soil was more evenly distributed than the Lynchburg soil. This suggest that the slightly higher clay content of this loam soil was integral in maintaining a higher moisture content compared to the other soils which corresponded to a higher N mineralization capacity.

Conclusions

If managed properly N derived from manure can be an important source of plant nutrient supply, but improperly managed manure N can potentially contaminate surface and groundwater. The information obtained in this study may be useful in maximizing the benefits of manure as a N source, while minimizing adverse effects of water quality contamination associated with variations often experienced in most agricultural fields. The use of microplot cylinders with ion exchange resins in an *in situ* N mineralization study during winter and summer months was effective in providing information on N release from dairy compost under normal field conditions. Addition of manure greatly affected the mineralization of N with concentrations of NH_4 being higher during the winter and summer months shortly after manure application. Over time NH_4 concentrations decreased as NO_3 concentration continued to increase, suggesting the occurrence of rapid nitrification. Seasonal conditions greatly affected N mineralization of dairy compost amended and control soils. Mineralization was minimal during the winter months and the N added from the manure was susceptible to leaching. This was probably due to temperatures being approximately 10 ° C, thereby, reducing microbial transformation of N. On the other hand, during summer months an increase in inorganic N concentration was observed resulting from an increase in mineralization. This increase in mineralization was caused by an increase in microbial activity observed at temperatures above 25 º C. During the summer months N mineralization also was greatly affected by moisture. Higher N concentrations were observed in the soil with the highest field capacity. During winter months the soil with the greatest percent sand had the highest leaching potential compared to the soils with more clay content. As for the summer, the soil with the highest bulk density retained more inorganic N in the soil compartment compared to soil with lower bulk density. Also, the soil with the greatest field capacity overall mineralized the most inorganic N. These results indicate that N mineralization rates should not be based entirely on mineralization capacity of the organic amendment but also on the field scale variability of the soil and landscape experienced in the field.

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Table 1. Characteristics of soil properties used in the *in situ* field study reported on a dry wt basis.

Soil Series	pH	CEC	Total C	Total N	C:N Ratio
		cmol kg^{-1}	-----------g kg^{-1}-----------		
Winter 2004					
Bama	6.31	5.84	4.42	0.48	9.21
Lynchburg	6.10	5.46	5.57	0.51	10.92
Goldsboro	6.24	6.09	3.77	0.41	9.20
Summer 2005					
Bama	6.26	5.70	3.77	0.39	9.67
Lynchburg	6.25	7.79	6.12	0.58	10.56
Goldsboro	6.86	5.12	4.02	0.54	7.41

Table 2. Soil physical characteristics of soils used in this study

	BD	Sand	Silt	Clay
	g cm^{-3}	----------------- % -----------------		
Bama	1.68	66.25	21.25	12.50
Lynchburg	1.64	46.25	41.25	12.50
Goldsboro	1.61	33.75	48.75	17.50

Table 3. The amount of inorganic N mineralized (NO_3+NH_4) and retained in the soil and captured in the resin after manure application.

	Days after initiation							
	Day 0	Day 3	Day 7	Day 14	Day 21	Day 28	Day 49	Day 70
	---------------mg kg^{-1} ---------------							
Winter 2004								
Soil	9.54	9.47	11.01	11.07	11.69	10.56	8.92	8.48
Resin	0.00	15.21	23.28	24.80	31.22	43.31	35.24	55.07
Summer 2005								
Soil	13.47	11.99	9.73	10.29	10.31	14.94	14.64	22.91
Resin	0.00	17.49	42.79	49.40	117.15	51.74	462.05	497.58

Table 4. The amount of inorganic N mineralized in the soil and captured (total N mineralization) in the resin after manure application for winter 2004.

	Days after initiation							
Soil	Day 0	Day 3	Day 7	Day 14	Day 21	Day 28	Day 49	Day 70
	---------- mg kg^{-1} ----------							
no manure								
Bama	11.62 a	26.02 a	28.85 a	30.48 a	40.59 a	44.51 a	49.37 a	58.97 a
Lynchburg	8.43 b	23.82 a	36.13 a	37.55 a	43.95 a	47.32 a	64.90 a	84.46 a
Goldsboro	8.58 b	24.21 a	37.88 a	39.57 a	44.19 a	35.81 a	52.20 a	47.24 a
manure								
Bama	24.17 a	40.92 a	46.79 b	49.75 b	65.28 b	64.14 b	60.99 b	70.61 b
Lynchburg	20.26 a	40.14 a	69.17 a	53.91 a	87.31 a	103.35 a	107.61 a	125.03 a
Goldsboro	21.72 a	35.60 a	46.24 b	47.53 b	57.57 b	66.03 b	58.55 b	74.62 b

Mean separatation represented by the same letter are significantly different (LSD 0.10).

Table 5. The amount of inorganic N mineralized in the soil and captured (total N mineralization) in the resin after manure application for summer 2005

Soil	Days after initiation Day 0	Day 3	Day 7	Day 14	Day 21	Day 28	Day 49	Day 70
	----------	----------	----------	---------- mg kg^{-1}	----------	----------	----------	----------
no manure								
Bama	10.30 b	28.84 a	28.66 b	26.46 b	56.08 b	153.89 a	527.92 a	702.31 a
Lynchburg	18.43 a	35.05 a	29.47 b	41.74 b	159.75 a	149.33 a	460.02 a	531.91ab
Goldsboro	11.69 ab	24.54 a	99.42 a	110.89 a	166.56 a	196.82 a	442.14 a	527.27ab
manure								
Bama	23.36 b	43.20 a	46.61 b	43.72 b	87.25 b	209.89 a	594.46 a	783.58 a
Lynchburg	29.15 a	51.17 a	45.51 b	51.67 b	244.50 a	169.43 b	520.11 a	571.96 a
Goldsboro	20.36 b	47.08 a	151.12 a	196.37 a	240.18 a	263.96 a	625.00 a	792.01 a

Mean separations represented by same letter are significantly different (LSD 0.10).

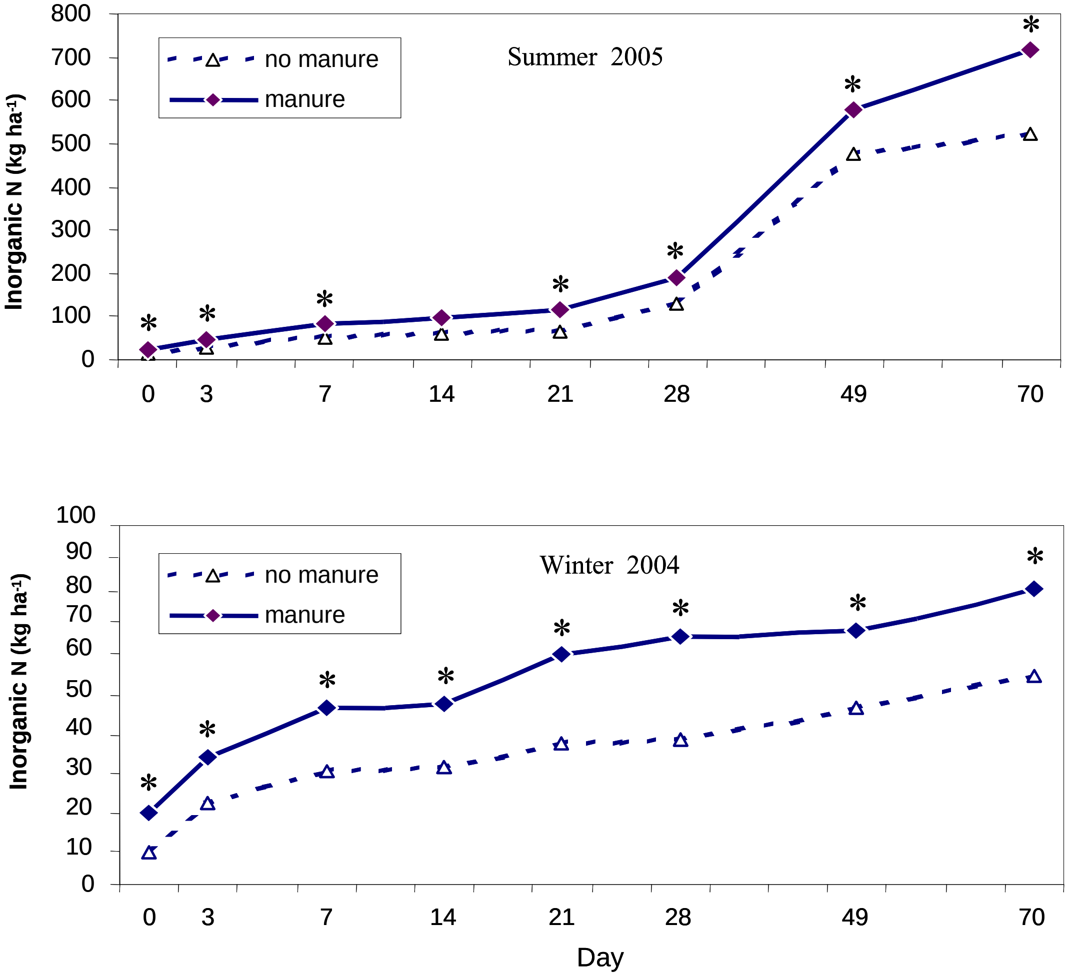

Figure 2. Seasonal N mineralization (Winter 2004 and Summer 2005) of the soil and resin combined in soil amended with and without manure at eight sampling days during the season. Symbol (*) represents significant differences at LSD (0.10), and ns represents not significant.

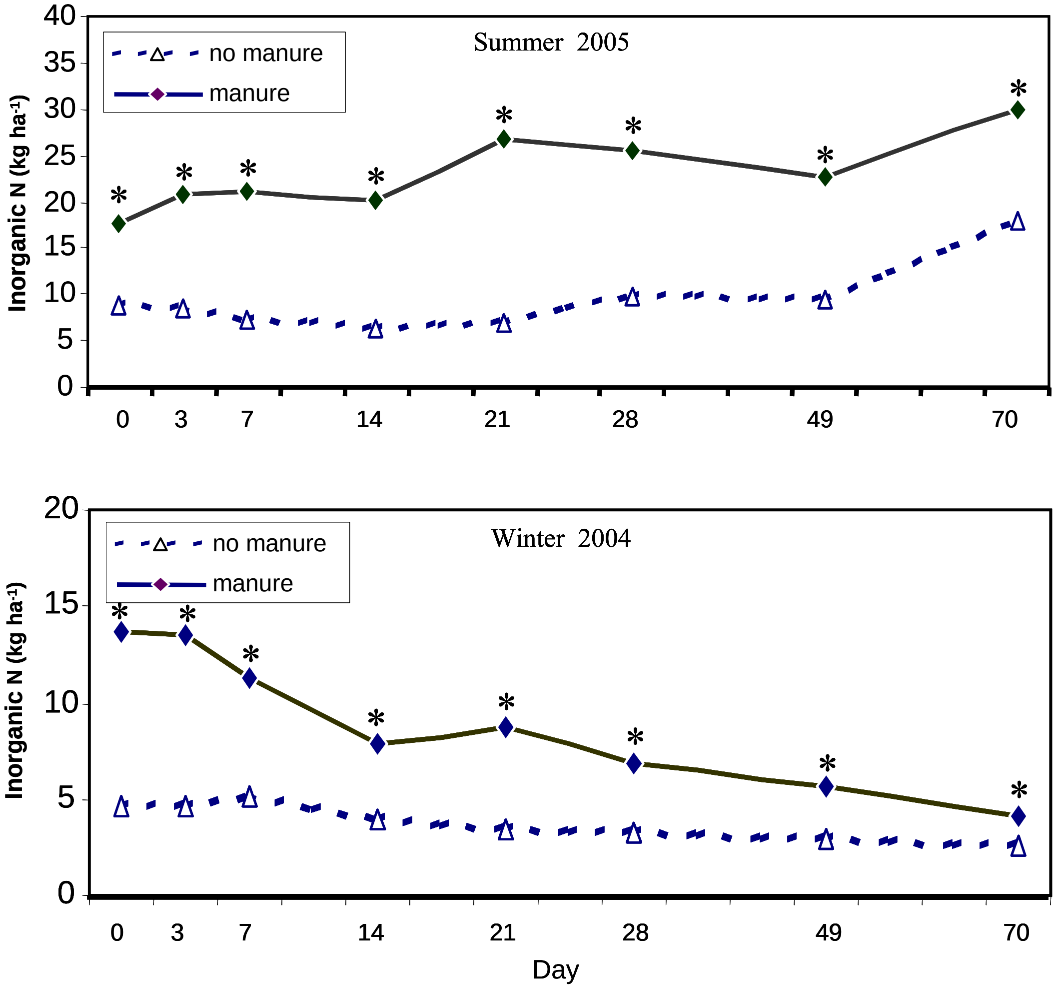

Figure 3. Seasonal concentrations (Winter 2004 and Summer 2005) of inorganic N in the soil compartment of the microplot cylinder for soil amended with and without manure at eight sampling days during the season. Symbol (*) represents significant differences at LSD (0.10), and ns represents not significant.

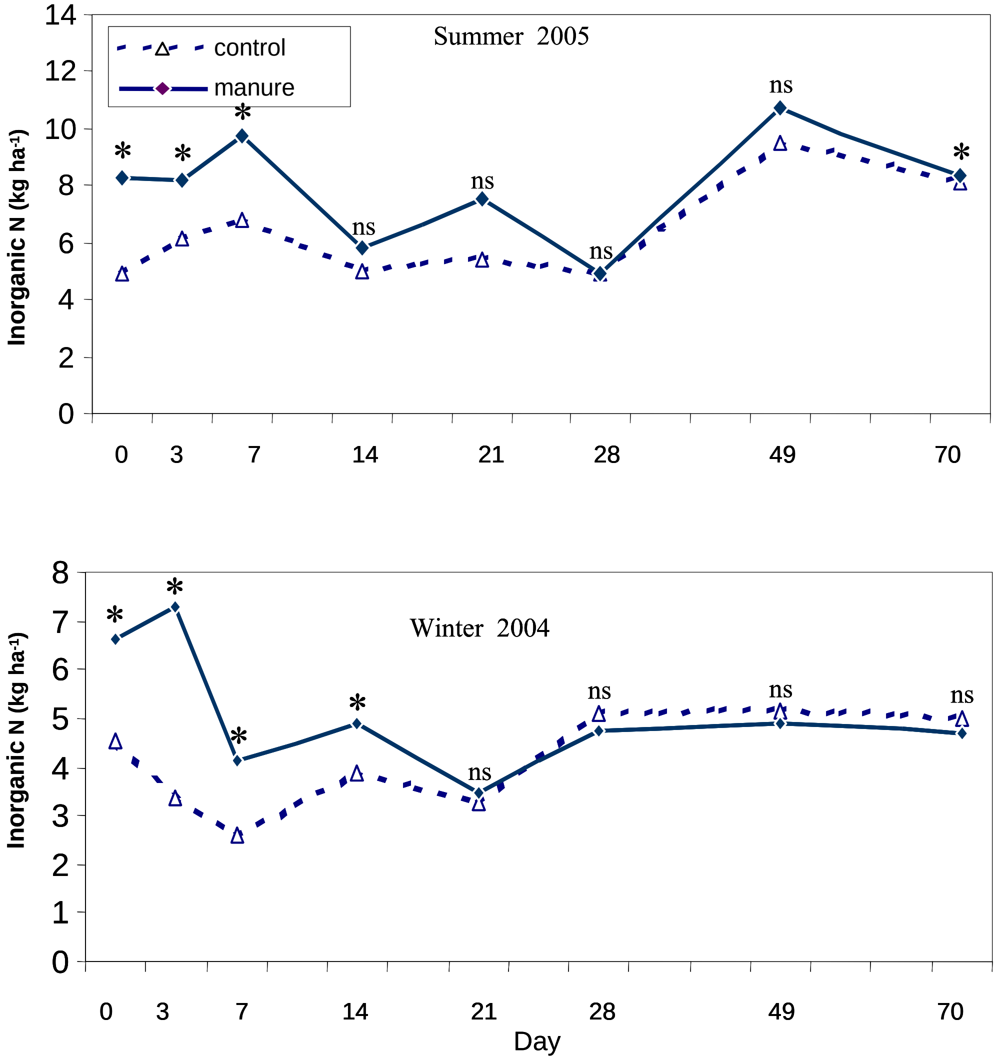

Figure 4 Seasonal concentrations (Winter 2004 and Summer 2005) of ammonium in the soil compartment of the microplot cylinder for soil amended with and without manure at eight sampling days during the season. Symbol (*) represents significant differences at LSD (0.10), and ns represents not significant.

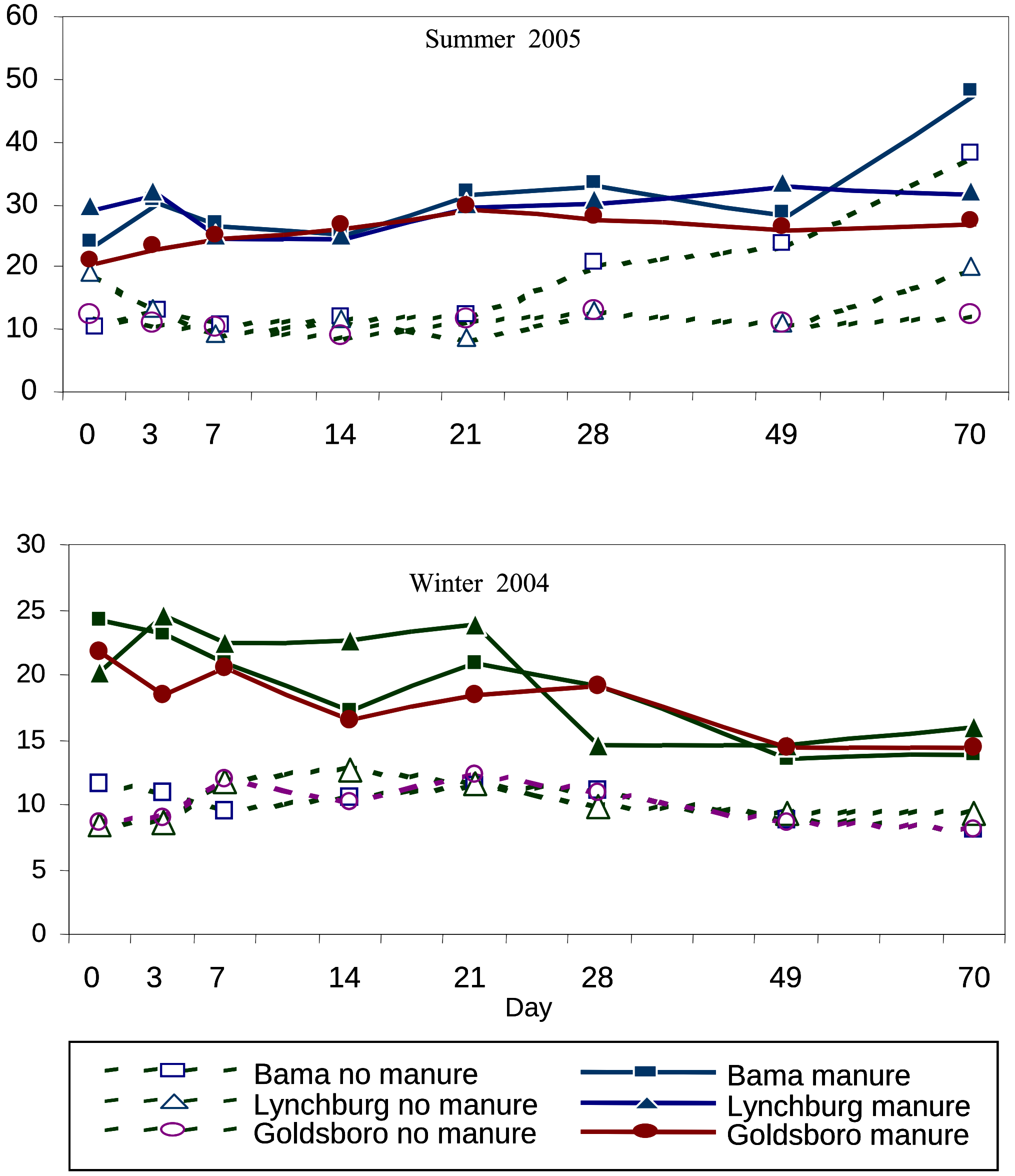

Figure 5. Seasonal concentrations (Winter 2004 and Summer 2005) of inorganic N in the soil compartment of the microplot cylinder for three different soil types amended with and without manure at eight sampling times during the season.

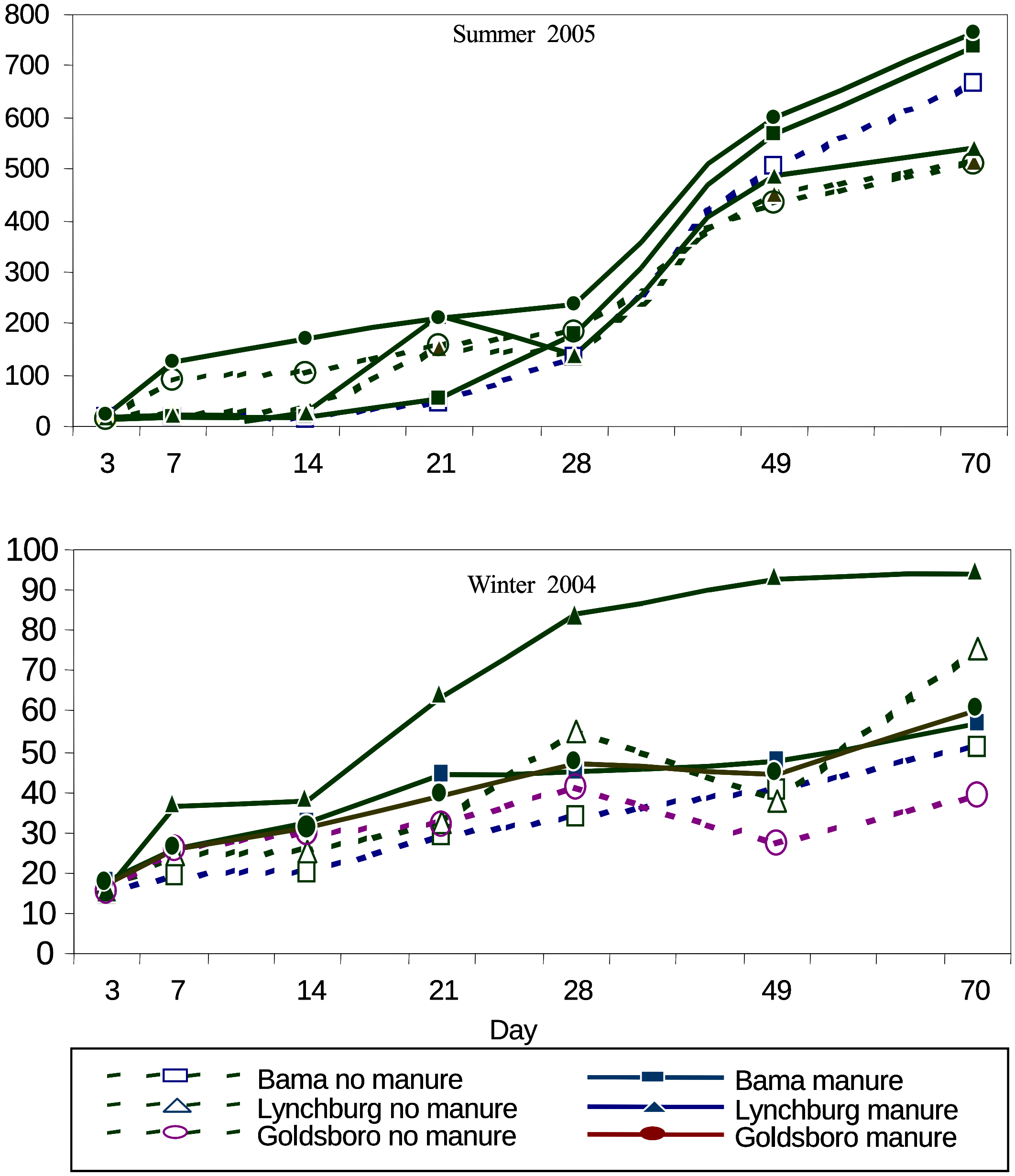

Figure 6. Seasonal concentrations (Winter 2004 and Summer 2005) of inorganic N collected by the ion exchange resins for three different soil types amended with and without manure at eight sampling times during the season.

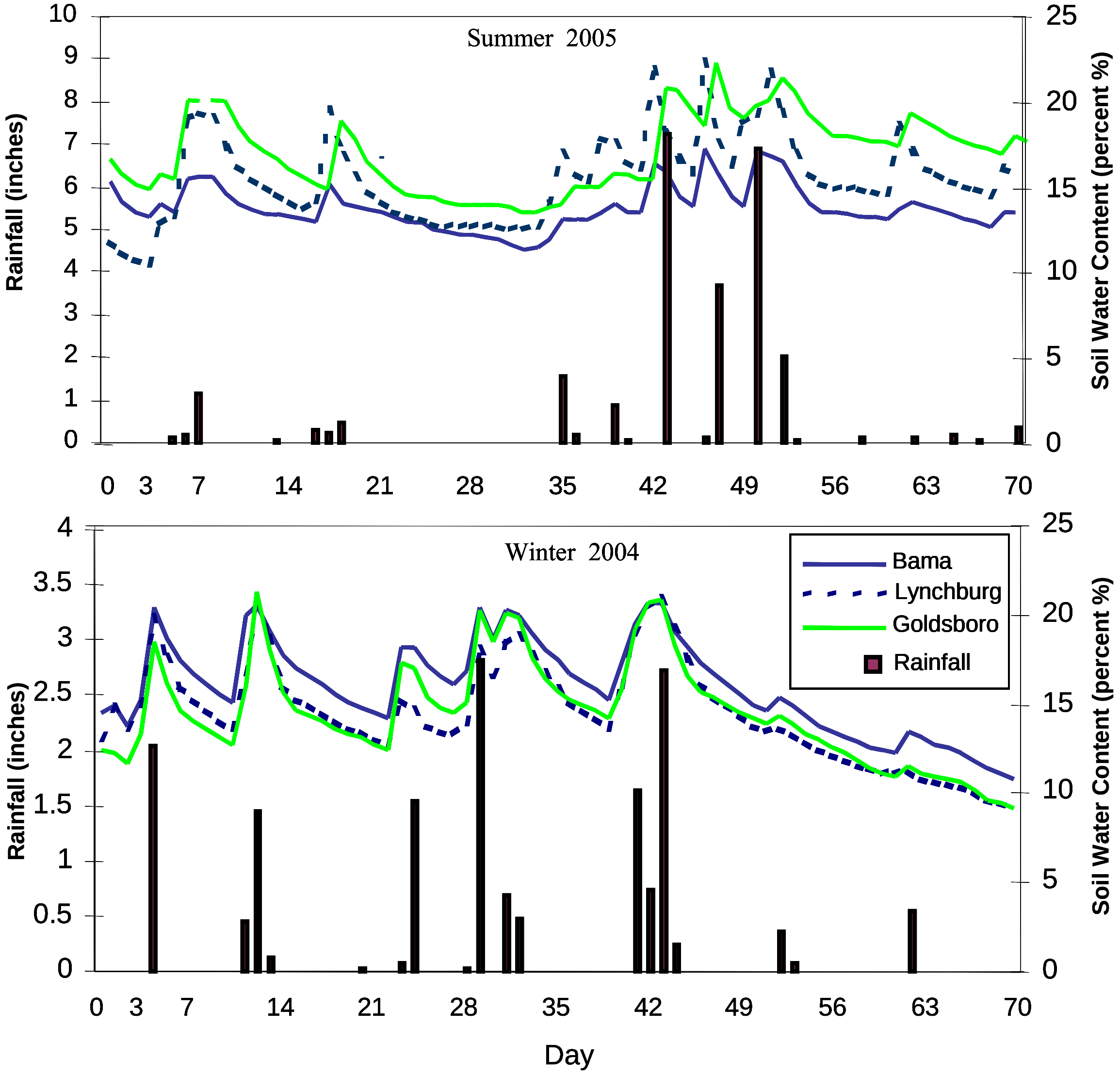

Figure 7. Seasonal mean soil temperature (Winter 2004 and Summer 2005) in the microplot cylinders for three soil types during the season.

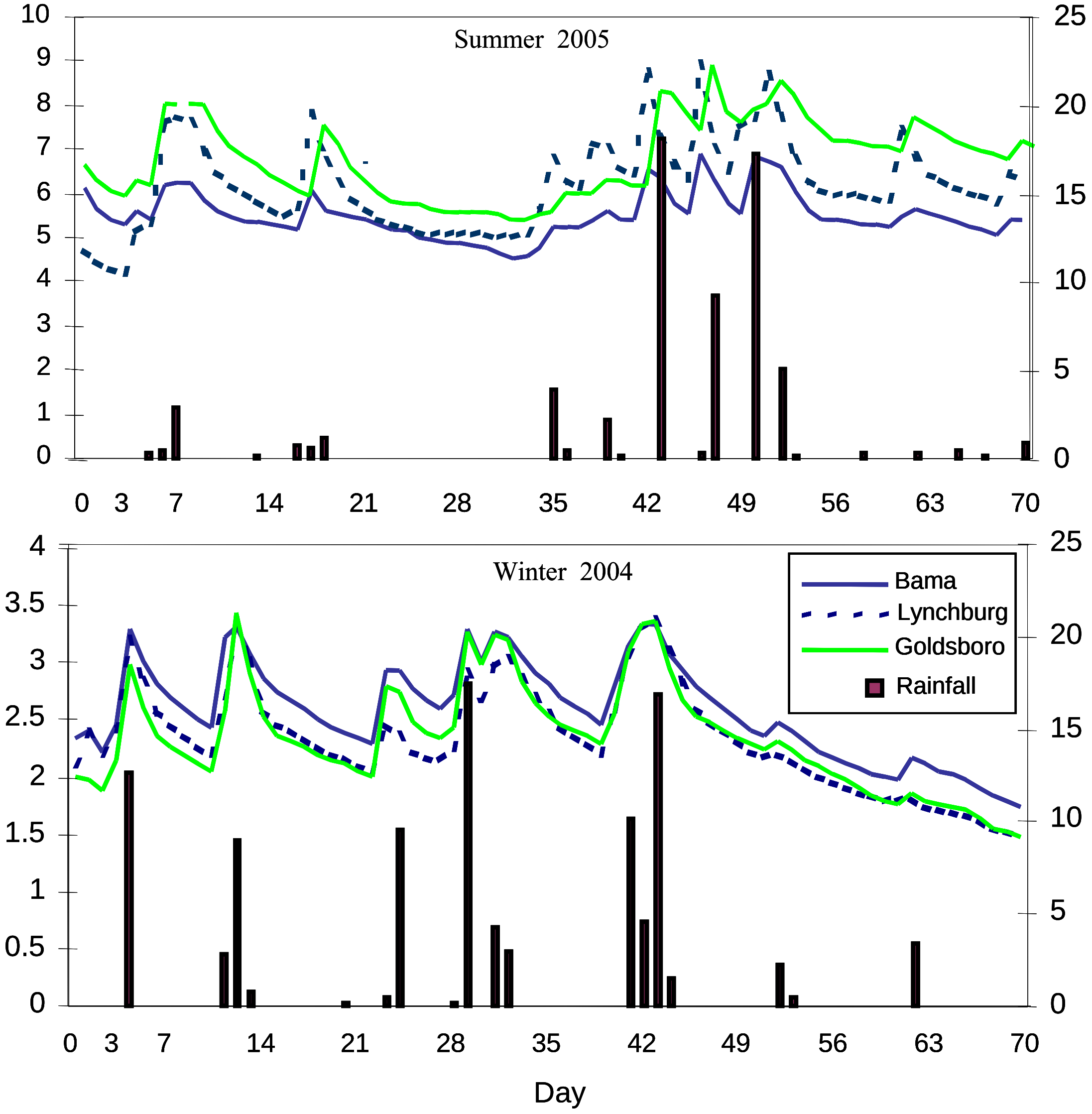

Figure 8. Seasonal mean soil moisture and average rainfall (Winter 2004 and Summer 2005) in the microplot cylinders for three soil types during the season.

V. SOIL MICROBIAL DYNAMICS AS INFLUENCED BY SOIL PROPERTIES AND LANDSCAPE POSITION

Abstract

Factors that affect plant growth, whether it is manure addition, season, or soil-type and landscape variability may provide insight on how to better manage agricultural fields through the evaluation of soil microbial activity, biomass and community structure. Thus an *in situ* study was conducted to evaluate microbiological properties from three different soil types and landscape positions located in close proximity of each other during the summer and winter months. The three Coastal Plain soils investigated were Bama (Sandy loam), Lynchburg (Loam) and Goldsboro (Loam). Dairy composted manure was incorporated into *in situ* soil cores at a rate of 350 kg N ha^{-1} and compared to unamended controls. Microbial properties were determined by microbial biomass N, dehydrogenase enzyme activity, and PLFA analysis. Dairy composted manure addition greatly affected the microbial properties of the soil. An increase in microbial activity and immobilization of N was observed with the addition of manure, suggesting that a shift in microbial dynamics had occurred due to the changes in the available substrate. This was most evident during summer months, which suggests that warmer temperatures stimulated the microbial activities. Landscape and soil type was also shown to affect microbial properties. The Lynchburg soil, a loam soil located in a depressed area, was shown to have the highest microbial biomass and microbial activity. Canonical discriminate

analysis (CDA) of the phospholipid ester-linked fatty acid (PLFA) profileswas utilized to confirm the results of microbial properties. This analysis indicated that a shift in lipid composition occurred between season, manure application, and soil landscape. Therefore, microbial properties could be a useful tool for providing insight into the long-term sustainability of the soil.

Introduction

In recent years there has been a renewed interest into the use of manure in agricultural row crop production resulting from large amounts of manure being generated in confined areas. The use of manure in row crop production can be viewed as having a two-fold affect: as a means of waste disposal, and as a means of building up soil fertility through the addition of organic matter. The addition of organic matter in the form of manure promotes microbial activity. Soil fertility and microbial activity go hand and hand because it is through the microbial population that mineralization (C, N, P, S) of organic material occurs (Frankenburger and Dick, 1983), which is controlled by the soil microbial community structure. Also, the topography of a landscape can influence the fertility and microbial activity of a soil resulting from the water movement and distribution of nutrients carried by water. Thus, information on the affect that manure application has on microbial parameters of soils from different soil types and landscape positions during winter and summer months is needed to make predictions on the long-term sustainability of the soil.

Since the structure and diversity of microbial communities are the driving force behind regulating processes such as decomposition of organic matter and nutrient cycling, in the soil at the ecosystem level, it is imperative to have a better understanding of the factors that regulate its size, activity and structure (Zeller et al., 2001). It has been estimated that microbial activity plays a significant role in the ecosystem process due to the fact that approximately 80- 90% of the processes in soil are reactions mediated by microorganisms (Nannipieri and Badalucco, 2003). Therefore, a better understanding of the diversity and size of microbial populations in a soil ecosystem may provide useful information on environmental and fertility impacts of agronomic practices. Soils containing a high structural diversity are characteristic of a healthy agroecosystem and soil with low structural diversity are characterized by low structural diversity that often hardly responds to environmental changes (Bianchi and Bianchi, 1995; Mader et al., 2002).

It has been reported that in healthy agroecoystems the dynamics of the microbial community is governed by interactions in tillage, soil moisture, temperature, aeration, and substrate availability (Feng et al., 2003). Also, the microbial biomass in the agroecosytems responds quickly to changes in management of the soil and is often utilized as an indicator of the sustainability of the soil.

The stimulation of microbial activity from manure addition has been attributed to greater inputs of organic carbon, thereby affecting the microbial population. In essence, the addition of animal manure may produce high structural diversity in the soil thereby causing alteration in the composition, size and activity of the soil microorganisms and extracellular enzyme activities. Due to the fact that microbiological activities are important in regulating soil properties (Dick, 1992) as a result of the integral role that they play in the biogeochemical cycle, a better understanding of the structure and functions that microbial communities in soils amended with manure is needed. This represents the key to improving soil fertility and sustainability of the soil (Kennedy, 1999; Buckley and Schmidt, 2003), while at the same time identifying areas that may potentially cause nutrient loss to the environment.

When applying manure to the field during the growing or non-growing season farmers usually apply it at a uniform rate, thereby failing to take into account the variability often experienced in most agricultural fields. Most agricultural fields have a varying degree of soil texture and landscape patterns such as different elevations. The varying degree of topography of a landscape is known to affect both the microclimate and the hydrological conditions experienced in agronomic fields (Rowe, 1984) In general, topography has been shown to influence water movement, thereby affecting the redistribution of materials carried within the water. This plays a role in influencing the type of soil microbiological processes occurring within a landscape (Huggett, 1975, Pennock et al., 1994), resulting in spatial patterns of SOM buildup, soil moisture, redox potential, bulk density, N mineralization, N immobilization, denitrification and respiration, observed in agricultural fields (Schimel et al., 1991; Goovaerts and Chiang, 193; Pennock et al, 1992; van Kessel et al., 1993). Little information has been reported on the influence that spatial variability has on microbial activity and community structure in agronomic fields that differ between landscapes and soil types. The response of microbial activity and microbial community structure to spatial variability in soil moisture, nutrient redistribution and soil texture could affect microbial transformations (alter nutrient cycling processes), especially following manure application.

More information is needed in order to better understand the affect manure application has on microbiological properties in agronomic fields subjected to a varying degree of soil types and landscape variability during the summer and winter seasons. This information could be useful in predicting the differences of metabolic activities from microbial communities inhabiting different landscape positions, therefore providing insight into how these microbes could play roles in affecting the sustainability of the soil environment. Thus, to access soil sustainability, microbial parameters such as microbial biomass N content, enzyme activities that are indexes of respiration, and total microbial biomass and microbial community structure from PLFA are believed to be sensitive reliable indicators for evaluating the microbial response to the organic amendment and landscape and soil type variability (Gregorich et al., 1994; Kandeler et al., 1999). Therefore, the objectives of this study were to determine the effects of manure application on three different soils in close proximity of each other from different landscapes and soil textural classes on microbial parameters and community structure during two different seasons.

Materials and Methods

Soil samples were collected from an ongoing precision agriculture experiment located at Auburn University's E.V. Smith Experiment Station in Macon County, Alabama (Terra et al., 2006). Soils were collected from field plots that have not received manure within the last 10 years.

Soil Description

The climate in Macon County consists of long hot summers due to moist tropical air from the Gulf of Mexico. Mean annual precipitation is 1422 mm with most of this (52%) occurring between April and October. The average daily temperature is 17°C. The three soil series to be evaluated from Macon County are Bama, Goldsboro and Lynchburg. These three soils were chosen because they are found in close proximity to one another, yet different in texture. The Bama series consists of very deep, well-drained soils that formed in loamy sediments. Slopes range from 0 to 5 %. These soils are fine-loamy, siliceous, subactive, thermic Typic Paleudults. Goldsboro series consists of very deep, moderately well drained soils that formed in loamy sediments. Slopes range from 0 to 2 percent. These soils are fine-loamy, siliceous, subactive, thermic Aquic Paleudults. The Lynchburg series consists of very deep, somewhat poorly drained soils that formed in loamy sediments. Slopes range from 0 to 2 percent. These soils are fine-loamy, siliceous, semiactive, thermic Aeric Paleaquults. The farming practice was comprised of conventional tillage, which receives inorganic fertilizer in a continuous cotton/corn rotation.

Laboratory Analysis

Air-dried samples were ground and passed thorough a 2 mm sieve and subjected to chemical and physical analysis. Total C and N were determined by the DUMAS dry combustion method using a CN LECO 2000 analyzer (LECO, St. Joseph, MI). Soil characteristics; pH, electrical conductivity (EC) soil effective CEC, soil extractable nutrients (Ca, Mg, K, P, Fe, Mn, Zn, Cu, B, and Na) and particle size analysis were measured by Auburn University Soil Testing Laboratory on the soil 0-15 cm soil depth increment using methodology described by Hue and Evans (1986).

In situ Mineralization Study

A field *in situ* mineralization study was conducted by placing polyvinyl chloride (PVC) plastic cylinders in the surface 20 cm of the soil profile. Net N mineralization and nitrification rates were measured using these *in situ* soil cores. Soil core (microplot cylinders) incubation chambers were 6.25 cm dia and 20.32 cm length. Intact cores were taken by driving the PVC cylinder into the top 20 cm of the soil profile using a hydraulic core sampler. Vegetation was removed from the surface portion and roots were severed in order to prevent N loss to plant uptake. The core samples were collected and brought to the laboratory. The top 4 cm of the soil core in the microplot cylinders were removed and the appropriate amount of dairy manure was added and thoroughly mixed to give 350 kg N ha^{-1} applied to a 15 cm depth. The soil was gently packed back into the microplot cylinder. The unamended soil underwent the same procedure except without manure in order to subject the soil to the same disturbance. Soil cores were transported back to the field and inserted in fallow ground. Dataloggers (HOBO Weather Station, Onset Computer Corporation, Pocasset, MA) were placed in the soil core in order to measure continuous soil temperature and soil moisture. The tubes were placed in the ground on the 12th day of January 2004 (winter study), and 25th day of May 2005 (summer study).

Soil cores were collected and returned to the laboratory for analysis on 0, 7, 14, 21, 49, and 70 days after manure application by randomly selecting and removing six cylinders from each plot. On each sampling day, soil cores were collected, transported to the laboratory in a cooler, at which time microbial biomass N and dehydrogenase activity was analyzed by taking a sub-sample from fresh soil as described below. Soil samples were collected for PLFA analysis on day 70.

Microbial biomass N

Microbial biomass N was determined similar to Runion et al. (2004) using the chloroform fumigation extraction method as described by Horwath and Paul (1994). Thirty grams of fresh soil were placed into 125 ml flasks. Flasks were placed into vacuum desiccators with 50 ml of chloroform, and a vacuum was placed on the desiccator until the chloroform boiled (22 mm Hg). The desiccator was then sealed and incubated (25°C) for 24hr. Following removal of the chloroform, desiccators were flushed with clean air a minimum of 6 times. Soil samples were removed, 30ml of 0.5M K_2SO_4 added to each flask, and flasks and placed on an orbital shaker at 180 rpm for 1hr. The resulting soil suspension was filtered through Whatman No. 42 filter paper in plastic funnels with the solution captured in 50 ml plastic vials. Vials were capped and frozen until N determination using standard Kjeldahl procedures. Nitrogen was determined on a replicate set of non-chloroform incubated soil samples following K_2SO_4 extraction; microbial biomass N was calculated as incubated N minus non-incubated N and expressed as μg N per gram of dry soil weight.

Dehydrogenase activity

Dehydrogenase activity, a measure of microbial respiration and reliable index of microbial activity in soil (Stevenson, 1959), was determined similar to Runion et al., (2002) from a modified procedures described by Tabatabai (1982). Sieved 1g of soil was placed in test tubes (15 X 100 mm), covered with 1 ml of 3% (w/v) 2,3,5-triphenyltetrazolium chloride, and stirred with a glass rod. After a 96 hr incubation (27°C), 10 ml of methanol was added to each test tube, and the suspension was vortexed for 30 sec. Tubes were incubated for an additional 4 hrs to allow suspended soil to settle. The resulting supernatant (5ml) was carefully transferred to clean test tubes using Pasteur pipets. Absorbance was read spectrophotometrically at 485 nm, and formazan concentration was calculated using a standard curve produced from known concentrations of triphenyl formazan per gram soil dry weight.

Phospholipid fatty acid analysis

Field moist soil samples were used for PLFA analysis as described by Feng et al. (2003) using a modified procedure of Findlay and Dobbs (1993) and Bossio and Scow

(1998). Soil samples (8 g of dry weight) were extracted in 19 ml of a single-phase mixture (1:2:0.8, v/v/v) containing chloroform, methanol and citrate buffer (0.15 M, pH 4) in the dark on a rotator for 2 hrs. After centrifugation for 10 min at 2500 rpm the supernatant was decanted to a fresh tube. The soil was washed with 8 ml of single–phase extractant, centrifuged and combined with the extract. Chloroform (7.1 ml) and citrate buffer (5.7 ml) was added to the extract to break phase and allowed to sit overnight. The aqueous phase was removed and the chloroform layer was transferred to a fresh tube and dried slowly under nitrogen in a water bath at 37°C. The phospholipids were separated from neutral and glycolipids using silicic acid column chromatography and subjected to a mild alkaline methanolysis to obtain the fatty acid methyl esters (FAME). The silicic columns were conditioned with 3 ml of chloroform; lipids were transferred to the column with 4 X 150 µl of chloroform. The elution of the neutral-lipids, glycolipid and the phospholipids fraction was performed with 5 ml chloroform, 10 ml of acetone and 5 ml of methanol, respectively. Methanol elutes were collected and dried down under nitrogen in a water bath at 37°C. The phospholipids were subjected to a mild alkaline methanolysis. Samples were dissolved in 1 ml of 1:1 methanol toluene and 0.2 N KOH in methanol, sealed, vortexed, and heated for 15 min at 37°C. The samples were allowed to cool to room temperature and 2 ml of deionized water and 0.3 ml of 1 N acetic acid were added. The fatty acid methyl esters (FAMEs) were extracted twice with 2 ml of hexane by vortexing for 30 s. Hexane fractions were combined and dried under nitrogen in a water bath at 37°C. Prior to GC analysis, samples were dissolved in the appropriate amounts of hexane containing 19:0 methyl ester as an internal standard.

The Hewlett Packard 5890 gas chromatograph (GMI, Inc., Ramsey, MN) equipped with a 25-m HP Ultra 2 capillary column and a flame ionization detector was used to analyze the FAMEs. Column temperature of this device initially started at 170ºC and increased to 270ºC at 5 C $\min^{-1}$. The injector and detector temperatures were maintained at 250 and 300ºC, respectively. Fatty acid peaks were identified using the MIDI peak identification software (MIDI, Inc., Newark, DE) and bacterial fatty acid methyl ester standards (Matreya, Inc., Pleasant Gap, PA).

Nomenclature

Fatty acids are designated according to the conventional X:YωZ system, where 'X' indicates the total number of carbon atoms in the molecule (except for molecules with a midchain branch), 'Y' indicates the number of double bonds, 'Z' indicates the position of the 1st double bond or cyclopropane ring, and ω indicates the position counted from the methyl end of the molecule. The prefix 'i' indicated iso branching, 'a' indicates anteiso branching, '10Me' refers to methyl branching on the 10th carbon from the carboxyl end, and 'cy' stands for cyclopropane ring. Suffixes 'c' and 't' indicate the cis and trans configuration, respectively. The number before an OH refers to the location of a hydroxyl group relative to the carboxyl end of the molecule. When branching occurs in the middle of the carbon chain, the branching C is not added to 'X' (Gunstone and Herslof, 1992).

Statistics

The treatments of the *in situ* study consisted of a total of 3 soils X 2 manure additions (with and without) X 6 replications X 7 sampling dates, which equals a total of 252 experimental units. The experiment was analyzed as a completely randomized factorial design with three soil types amended with and without manure. Statistical analysis was performed using a GLM procedure of SAS (SAS institute, 1985), and means were separated using least significant difference (LSD) at an a *priori* 0.10 probabililty level. To access specific effects of season (winter vs. summer), soil series, and manure application on microbial community structure, Canonical discriminate analysis (CDA) was performed on FAME data. CDA was analyzed using the mole percentage distribution of PLFAs with SAS software version 9.13. CDA was performed on combined PLFA data from day 70 from winter 2004 and summer 2005. All samples were analyzed for PLFA profiles using a set of 33 fatty acids that were present in most of the samples.

Results and Discussion

Some of the basic soil properties of the three soil types utilized in this study are presented in Table 1 and 2. In general, the focus of this study was to access whether season and manure addition had an impact on microbial characteristics and the microbial community as a whole when applied to different soil types and landscape positions. Season, manure application, soil type and landscape position had an affect on the microbial properties. Seasonal effect (winter season compared to summer seasons) was shown to have the greatest affect on microbial properties compared to soil type and manure application. This is similar to that of Bardgett et al. (1999) who reported greater microbial biomass C and N and microbial activity during summer months compared to winter months. The following discussion is a more in-depth look at the specifics of how the previously mentioned management decisions affect microbial properties.

Dehydrogenase

Dehydrogenase is an intercellular enzyme involved in the microbial oxidoreductase metabolism. These soil enzymes depend on the metabolic state of soil microorganisms. Therefore, dehydrogenase can be utilized as a measure of microbial respiration and a reliable index of microbial activity in soil (Stevenson, 1959). A significant increase in dehydrogenase activity was observed on all sampling days except day 49 during the winter and day 7, 28, and 49 during the summer months. Although, not significant on each sampling day, an increase in dehydrogenase activity was observed with the addition of manure to the soil during the winter and summer, suggesting that changes in the size of microbial populations and respiratory activity occurred in response to the added available substrate. Season greatly impacted dehydrogenase activity. Significant differences were observed ($P< 0.001$) for every sampling day except day 14. Dehydrogenase activity measured during the summer was almost double that measured during the winter months. The increase in concentration resulting from the contrasting seasons suggests that different dehydrogenase enzyme systems changed with season (Ross, 1971). Also, higher dehydrogenase enzyme activity, which is a representation of microbial activity, was probably a result of higher soil temperature, which has been shown to stimulate microbial activity. This is similar to the finding of previous research that has reported that temperature and moisture are the two most important abiotic factors affecting microbial activity. Although moisture was slightly higher during the winter (Fig 2&3) soil temperature was the overriding factor affecting microbial activity.

Dehydrogenase activity was also greatly affected by soil type. Significant differences were observed on all sampling days except day 7, 14 and 49 during the winter and day 28 and 49 during the summer season. The Lynchburg soil produced higher dehydrogenase enzyme activity at all sampling dates except day 0, 49, and 70 during the winter and day 7 during the summer months. Although no significant differences were observed between the soil X amendment effects at any sampling days, there was a trend resembling the soil effect. The Lynchburg soil with manure produced the highest microbial activity compared to the other soils. The Lynchburg soil, located in a depression area, contains the highest organic C and N content. The observed difference in microbial activity was probably attributed to the soil being located in a depression area and nutrients probably accumulated in this area resulting from water movement, thus, resulting in increased organic matter. This also corresponds with the higher organic C and N, and CEC values observed from the initial soil characteristics from this soil. It has been reported that generally enzyme activities in the soil are closely related to the organic matter buildup (Kanchikermath and Singh; 2001), which helps explain our results in this study. The decline in microbial activity observed during the winter months for the Lynchburg soil on day 49 and 70 was probably due to moisture stress. This soil was the most sandy in nature compared to that of the other soils. The Goldsboro soil experienced an increase in microbial activity on day 49 and 70. This was probably attributed to the soil having the highest clay content. Thus, the microbes were probably better protected in the micropores of the clay soil compared to that of the more sandy soils. In essence, the effect of soil texture affected the ability of a soil to retain water for microbial activity.

Soil microbial biomass N

Although the microbial biomass in the soil represents approximately 1-5 % of the total organic matter (Jenkinson and Ladd, 1981) it can be utilized as an early indicator of the soil quality as a result of different management practices (Powlson, 1994). Microbial biomass has also been utilized to evaluate the microbial transformations of plant nutrients as well as for their ability to act as a source or sink for N, P, S and C (Paul and Voroney, 1980; Anderson and Domsch, 1980). Similar to dehydrogenase activity, microbial biomass N also increased following the application of dairy compost (Figure 4). The addition of dairy compost significantly increased microbial biomass N on all sampling days except day 7 and 70 during the winter and day 49 during the summer. Although not significant on every sampling day, microbial biomass was higher in manure compared to no manure treatments. It is well known that changes in microbial biomass concentrations observed in the soil correspond to changes in the availability of decomposable substrate. The addition of manure provided the microbes with readily available C and N. This is consistent with the finding of Bohme et al. (2005) who reported that microbial biomass was greater in soil following the application of farmyard manure. The same trend was also shown for soil X season effect. During the summer more microbial biomass N was observed compared to the winter months at all sampling dates. This corresponds to the dehydrogenase activity, suggesting that as microbial activity increased more N was immobilized into microbial cells. Microbial biomass was also greater during the summer compared to the winter months. This is probably due to the fact that microbial activity was higher during the summer months as shown with the dehydrogenase activity. During the winter and summer months the amount of microbial N biomass reached a peak at day 0 and decreased until approximately day 14 and day 21 for both the summer and winter months, respectively. This trend may represent a temporary storage of N in the microbial cells at day 0 and decreased by day 7, representing release of N into the soil environment

(N mineralization). A comparison of soil type shows that significant differences were observed on every sampling date for the winter and summer season ($P < 0.10$). The Lynchburg soil, which contained highest initial soil organic C and N content was more efficient in immobilizing the N into its microbial cells, suggesting that land use and topography of a landscape could cause changes in soil C and N cycling rates and accumulation of organic matter (Chen and Stark, 2000). The microbial biomass was the lowest in the Goldsboro soil. This means that less N was being immobilized into the microbial cells. The reduced microbial biomass N occurring in the Goldsboro soil could be attributed to more nitrification occurring and less immobilization. This also corresponds with the low C:N ratio that was observed in the soil, thus suggesting that although the Goldsboro soil had a higher clay content, microbial biomass N was more closely related to the C, N, and C:N ratio of the soil. Also, the textural differences in these were not great enough to affect the microbial biomass N. There was a soil X season ($P < 0.0001$) effect for every sampling day between the summer and winter months. A soil amendment X season effect was observed only on day 28, suggesting that the interaction between the soil and season was greater than the interaction between manure and season. In essence, the soil effect was the major determinate in increasing the immobilization of N into the microbial biomass.

Microbial biomass composition (PLFA)

Response of total biomass composition of the microbial community to differences in soil type and manure application during winter and summer months were determined using mole percentage of the PLFAs most common to all samples. This method was used to detect differences in microbial biomass pools in the soil. The microbial biomass observed using PLFAs detects the active proportion alone (Tunlid et al., 1985; Tunlid and White, 1992; Zak et al., 1996). The phospholipids released after cell death are quickly used within minutes to hours as substrate by living microorganisms and are metabolized to diglyceride and PO_4^{3-} (White et al., 1979). Therefore, due to rapid turnover, the total concentration of PLFA in soil can be utilized to provide evidence of the active microbial populations (Zak et al., 1996). In essence, PLFA analysis may be used to determine stress responses or periods of activity of microbial communities (Morgan and Winstanley, 1997). Observations in this study showed that among the phospholipid fatty acids identified, mole percentage of PLFAs were present in significantly lower concentrations during the winter months ($P < 0.0001$) compared to that of the summer months (Figure 5). This suggests that increase in mole percentage PLFA concentrations corresponded to higher microbial community composition probably resulting from increased soil temperatures affecting the mobilization of organic matter fractions accumulated during winter. These findings are in agreement with that of Ritz and Robinson (1988), and Ross et al. (1995). Mole percentage of PLFA were significantly higher ($P < 0.002$) in the manure treatments compared to that of the no manure treatments for both the winter and summer season, suggesting that manure application caused the soil microbial populations to increase in size compared to control plots. This increase in total percentage of PLFAs representing a population increase was probably a result of increased nutrients as well as microbial biomass contained in the manure. Significant increase in microbial biomass was observed resulting from differences in landscape position and soil types, implying changes in soil microbiological characteristics have occurred in these areas. Total percentage PLFAs was also significantly ($P < 0.10$) higher in the Lynchburg soil, which is located in a depression area. Large inputs of nutrients have accumulated in the soil over the years, which corresponded to greater changes in microbial community composition. The Lynchburg soil being located in a depression area and accumulation of carbonaceous nutrients with water movement have caused the microbial biomass to increase, thereby affecting soil characteristics and influencing greater changes in microbial community population. The significant increase in the microbial composition also suggests that agronomic management factors can cause distinct effects on microbiological soil characteristics (Widmer et al., 2005). Therefore, this should be taken into account when applying manure to different landscapes with a varying degree of soil types and landscapes in order to prevent environmental degradation.

Soil microbial community structure

In this study, PLFAs analysis identified 48 fatty acids. However, of these, only 33 were present in most samples and used in data analysis. PLFA profiles identified consisted mainly of saturated and unsaturated fatty acids. The saturated fatty acids consisted of three sub-groups: normal straight chain saturated fatty acids (NSFA) – 14:0, 15:0, 16:0, 17:0, 18:0, 20:0; mid-chain branched saturated fatty acids (MBFA) – 10 Me 18:0 and terminally branched saturated fatty acids (TBSFA)- i14:0, i15:0, a15:0, i17:0 and a17:0. The unsaturated fatty acid group is comprised also of three subgroups: (CYCLO) cy17:0 and cy19:0; monounsaturated fatty acids (MONO): i15:1, i16:1, 16:1ω9c, 16:1 ω 5c, 16:1 2OH, 17:1, 18:1 ω9c, 18:1ω5c, 20.1ω9c and (POLY) 18.3ω6c. The proportions of these PLFA identified from each fatty acid group are provided in Figure 5.

The specific biomarkers obtained from the PLFA profiles were used to quantify the relative abundance and changes in microbial groups that have occurred due to treatment. The mean ratio of monounsaturated to saturated fatty acids (Table 3 and Figure 5) increased in soil receiving dairy compost during the winter ($P<0.003$) and summer ($P< 0.02$) seasons, suggesting that monounsaturated fatty acids (commonly associated with Gram-negative bacteria) increased in population resulting from dairy compost addition. This is similar to the finding of Bossio et al., (1998) who reported that differences in farming practices that increase organic inputs also influence monounsaturated PLFA profiles. Although, significant differences were not observed between soil types, the Lynchburg and the Goldsboro soils (Lynchburg- loam soil in depression area; Goldsboro – loam soil in well drained area) contained higher amounts of monounsaturated fatty acids compared to the Bama soil (sandy loam). This suggests that soil type could also affect concentration of monounsaturated PLFA profiles. The response of monounsaturated fatty acids to dairy compost and soil type shows that a shift in community structure has occurred due to the overall shift in lipid composition.

The PLFA markers used in this study to quantify the relative abundances of specific gram-positive to gram-negative bacteria ratio were as follows: i14:0 i15:0, a15:0, i16:0, 10Me16:0, i17:0, and a17:0 for Gram-positive bacteria, and cy17:0, cy19:0, 16:1ω9c, 18:1ω9c, 15:1ω4c, 18:1ω7c, and 17:1ω9c for Gram-negative bacteria (O'Leary and Wilkinson, 1988; Zelles et al., 1994; White et al., 1996; Zelles, 1997, Fierer et. al, 2003). PLFA markers for fungi to bacteria ratio are as follows :18:2 ω6,9c, 18:1 ω9c, 18:3 ω6c, and 20:1 ω9c for fungi, and i15:0, i16:0, 10Me16:0, a15:0, cy17:0, 18:1 ω7c, cy 19:0, 14:0 15:0, 16:1 ω9c, 16:1 ω7c, 16:1 ω5c, a17:0, i17:0 17:0 and 18:0 for bacteria

Frostegard et al., 1993; Zelles, 1997; Fierer et. al, 2003; Feng et al, 2003). The ratios of the relative abundance of the calculated cyclopropyl fatty acids/monoenoic precursors (cy17:0 + cy 19:0/ 16:1 ω7c + 18:1 ω7c) have been previously used by other researchers as indicators of nutritional stress in bacterial communities (Knivett and Cullen, 1965; Kieft et al., 1997; Bossio and Scow, 1998; Fierer et. al, 2003).

The mean ratio of gram-positive to gram-negative bacteria (Table 3) significantly decreased in soil containing dairy compost manure compared to soil without dairy compost manure for the winter ($P<0.10$) and summer ($P<0.008$) months (Figure 7, Table 3). This suggests that addition of compost increased the soluble organic C in the soil (Bhogal and Shepard, 1997; Gregorich et. al, 1998. Liang et al., 1998), thereby providing a more stable and readily available substrate for supporting higher levels of microbial activity for gram-negative bacteria (Peacock, 2001). It has also been reported that Gram-negative bacteria have been mainly associated with monounsaturated fatty acids, which corresponds to increases in organic matter content and high substrate availability (Bohme et al., 2005; Zelles et al., 1992; Bossio et al., 1998). No significant differences were observed between seasons and soil types. Population of actinomycetes, although not significant decreased with the addition of more readily available substrate from the dairy compost compared to the without dairy compost manure for the Bama and Goldsboro soil. On the other hand, the addition of dairy compost manure caused an increase in actinomycetes in the Lynchburg soil. The same trend was shown for the biomarkers of fungi. The population of fungi decreased in the Bama and Goldsboro soil and increased in the Lynchburg soil with the addition of dairy compost manure. This shows that microbial community structure changed due to landscape position and effect of soil types.

Differences observed in the available substrate in the soil are likely responsible for a decrease in fungi/ bacteria and gram-positive/gram-negative bacteria ratios for the Bama and the Goldsboro soil after manure addition. This is common under most conditions because the addition of manure increase the more readily decomposable compounds, which are mainly decomposed by soil bacteria while fungi decompose the more recalcitrant and insoluble materials. On the other hand, the Lynchburg soil, which in located in depressed area, has received a more continual addition of readily-available substrate, thus suggesting that historically as the substrate increased from water movement, thus, bacteria and fungi probably had a long-term potential to develop simultaneously (Griffiths et al., 1999). Gram-positive bacteria are often found in environments where there is less competition due to plenty of available substrate. In this study the Lynchburg soil inheritly has more organic matter and the addition of dairy compost manure increased the competition in the soil, therefore changing the microbial community structure. The other two soils are located in well-drained areas and have not received manure within the last ten years and the addition of available substrate was minimal, suggesting that the microbial response to the added substrate might be more subtle. This data also suggests that higher proportion of gram-positive bacteria in the Bama and Goldsboro soil indicates that the PLFAs in the soil were adapted to extant bacterial biomass rather than successional changes.

The cyclopropyl/monoenoic precursor ratio can be utilized as an indication of the physiological status of the microbial population. Low cyclopropyl/monoenoic ratio is an indication of activity growing cells. This study provided evidence that more stressful conditions were observed during the summer compared to the winter. This increase in stress could be attributed to an increase in the soil temperature during the summer compared to the winter. This is similar to the finding of Petersen et al. (2002) who also observed higher stress during summer months. Petersen et al. (2002) attributed the increased stress to more extreme environmental conditions resulting from a hot and dryer climate observed during the summer.

To obtain a more detailed interpretation about the fatty acids accounting for differences between treatments and microbial groups canonical discriminant analysis was used. CDA was carried out by comparing the summer and winter season to identify differences between the dairy compost additions and soil series. Table 4 presents the results of CDA. The use of CDA statistically confirmed the result of mole percentage by showing significant difference between seasonal variations, manure application and soil type. The first 4 canonical discriminant variates (CDV) accounted for a total of 90% of the total variance. Table 4 and 5 show that the first CDV, which accounts for 48% of the variance, consists mainly 16.1ω5c, and the second for 25% of the variance, consists of 16:1ω7c and 15:0 iso 2OH. The third and fourth CDV explained 16% and consisted mainly of 17:0 iso and 14:0, respectively. A CDA plot of the three soil series with and without dairy compost for the summer and winter season is shown in Figure 8 and 9.

Canonical discriminate analysis indicated that there were significantly different PLFA profiles between treatments and different microbial communities. In general there was clear discrimination observed between the dairy compost manure treatment and the without compost treatments on the canonical variate (CV)1 (Figure 6), with manure plots having higher ordinates values (explaining 48% of the variance). The Lynchburg soil from the summer of 2005 was intermixed with the soils containing dairy compost, this unusual occurrence could not be explained and is probably a result of experimental error. The effect of season was discriminated by with CV2 (Figure 8) (explaining 25% of the variance). The Bama soil during the spring months and Goldsboro soil during the summer months had the highest ordinates. Also it seems that the Bama soil clustered closer together while the other soils intermixed. The PLFA profiles representing microbial community were affected by sampling times or certain season of the year. The warmer soil temperatures experienced during the summer probably lead to an apparent increase in the C substrate available to microbes due to temperature dependent change in growth efficiency or diffusional processes (Ellert and Bettany, 1988; MacDonald et al., 1995). In combination with the results obtained from the total PLFAs suggest that a shift in microbial community composition occurred with changes in season resulting from higher soil temperature. The third CV (Figure 9) clearly discriminated the soil types into distinct groups (explaining 11% of the variance), suggesting the microbial communities were structurally different according to the variation experienced within each soil type. This could have lasting affect on the rate a nutrient is immobilized into organic matter or mineralized and lost from the immediate soil environment. Canonical discriminate analysis identified fatty acids that were important in explaining the variability observed within the PLFA profiles. The PLFAs 16.1ω5c, 18:3ω6c, 18:1ω7c, cy19:0, 20:4ω6, 9,12 were identified by CDA as influential biomarkers for the CV1 and 16:1ω7c / i15:0 2OH, 18:1ω7c, 18:0, 18:3ω6c for CV2, respectively (Table 5). The PLFAs i17:0, a18:0/18:2 ω6, 9c, 16:120H, cy17:0, and 17:0 10 methyl were influential biomarkers for CV3. The PLFA 16.1ω5c is associated with monounsaturated fatty acids, which have been shown to increase with manure addition as previously stated. Also 16.1ω5c, 18:1 ω 7c and cy19:0

are Gram-negative bacteria and which are associated with an increased readily-available substrate. On the other end of the spectrum 18:3ω6c and 20:4ω6, 9,12 are associated with fungi and was shown to decrease with the addition of available substrate. The PLFA identified for the second CV 16:1ω7c, 18:1ω7c accounted for most of the discrimination. Fatty acid 16:1ω7c is associated with monounsaturated fatty acids and 18:1ω7c is associated with gram-negative bacteria both of which increased with the addition of manure as previously stated. The biomarker 18:0 is a non-specific fatty acid, which is found in all organisms. The signature fatty acid biomarker a15:0 is associated with gram-positive bacteria and 18:3ω6c is associated with fungi. The increase in soil temperature probably affected the PLFA concentrations, thereby causing a shift in lipid composition between seasons. The PLFAs identified for the third CV i17:0 is a gram-positive bacteria, 18:0 gram-negative bacteria, 16:1 20H non-specific bacteria, cy17:0 and 17:0 10 methyl were all found in more abundance in the Lynchburg and Goldsboro soil, which are both loam soils. The Lynchburg soil had a higher concentration of the PLFA during the winter months and the Goldsboro had a higher abundance of the soil during the summer months. The data observed from the CDA confirmed that differentiation in lipid composition of microbial community structure had occurred due to season, manure amendment, and soil type. These results help support the previous observation made in the study with microbial biomass N and dehydrogenase, total PLFAs.

Conclusions

Soil microorganisms are the driving force behind maintaining long-term sustainability of an agroecosystem. It is through their processes that soil formation and nutrient cycling occurs. The available substrate in the soil determines the size, activity and biomass in the soil, which is instrumental to controlling the structure of the microbial community. Therefore, evaluation of key microbial parameters such as microbial activity, microbial biomass and community structure may provide insight into the long-term sustainability of the soil ecosystem processes. Microbial parameters evaluated in the study suggest that season, addition of manure, and changes in the topography of a landscape can greatly affect the microbial community structure in the soil. The addition of dairy compost manure resulted in a diverging microbial community structure probably by increasing the soluble C in the soil. Season also increased the microbial parameter resulting in increased metabolic activity during the summer compared to the winter. Soil landscape positions that have resulted in a buildup of organic matter were observed to enhance and alter the microbial community too. The significant changes in microbial parameters were evident by observing increases in microbial biomass N, dehydrogenase (microbial activity), total PLFAs, as well as changes in microbial community structure. Canonical discriminate analysis clearly discriminated PLFA profiles by season, manure addition and soil type and landscape, thus, confirming that changes in microbial community structure diverged, resulting from the agronomic management practices evaluated. Data in this study suggest that soil located in depression areas tend to accumulate organic matter, which is responsible for increasing microbial activity. Also, the addition of manure increases microbial activity, which plays a prominent role in immobilizing and mineralizing plant nutrients. Therefore, consideration should be taken in account when developing management practices in order to maximize the use of plant nutrients without negatively affecting the environment.

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Table 1. Characteristics of soil properties used in the in situ field study reported on a dry wt basis.

Soil Series	pH	CEC	Total C	Total N	C:N Ratio
		cmol kg^{-1}	-----------g kg^{-1}-----------		
Spring 2004					
Bama	6.31	5.84	4.42	0.48	9.21
Lynchburg	6.1	5.46	5.57	0.51	10.92
Goldsboro	6.24	6.09	3.77	0.41	9.2
Summer 2005					
Bama	6.26	5.7	3.77	0.39	9.67
Lynchburg	6.25	7.79	6.12	0.58	10.56
Goldsboro	6.86	5.12	4.02	0.54	7.41

Table 2. Soil physical characteristics of soils used in this study

	BD	Sand	Silt	Clay
	g cm^{-3}	----------------- %-----------------		
Bama	1.68	66.25	21.25	12.50
Lynchburg	1.64	46.25	41.25	12.50
Goldsboro	1.61	33.75	48.75	17.50

Table 3. Proportional distribution of microbial groups identified using PLFA biomarkers for the winter and summer months

Soil	Saturated n mole (%)	Unsaturated n mole (%)	Monounsaturated/ Saturated ratio	Gram-positive/ Gram-negative ratio	Fungi/Bacteria ratio	Cyclopropyl/ Monoenoic ratio
Winter						
Bama	56.09	35.00	0.39	1.86	0.19	1.60
Bama manure	54.45	36.26	0.45	1.83	0.17	1.51
Lynchburg	57.51	37.36	0.44	2.24	0.15	1.71
Lynchburg manure	53.23	37.35	0.45	1.66	0.17	1.71
Goldsboro	53.64	35.66	0.46	1.90	0.17	1.37
Goldsboro manure	54.30	36.26	0.44	1.76	0.16	1.62
Summer						
Bama	53.05	36.61	0.37	2.06	0.25	2.22
Bama manure	54.34	34.00	0.40	2.02	0.16	1.87
Lynchburg	54.32	34.45	0.41	2.07	0.15	1.81
Lynchburg manure	50.89	37.59	0.46	1.75	0.17	1.88
Goldsboro	52.05	37.12	0.43	2.04	0.22	2.24
Goldsboro manure	50.51	39.22	0.49	1.59	0.20	1.84

Table 4. Canonical discriminant analysis of PLFAs accounting for difference in the canonical discriminant varables

Canonical variable	Eigenvalue	% Variance	Cumulative % variance
1	56.7	48.22	48.22
2	29.9	25.44	73.65
3	13.1	11.11	84.79
4	6.6	5.66	90.44

Table 5. PLFAs of the first five scores accounting for the variance of the first four canoncial axes

Fatty acid	Score	Score	Specificity as a biomarker
Canonical variable 1			
16.1ω5c		0.82	Bacteia (Gram-positive and Gram-negative)
18:3ω6c		-0.43	Fungi
18:1ω 7c		0.42	Aerobic bacteria, Gram-negative
cy19:0		0.40	Anaerobes, Gram-negative bacteria
20:4ω6,9,12		0.39	Fungi
Canonical variable 2			
16:1ω7c/i15:0 2OH		-0.70	Nonspecific
18:1ω7c		-0.53	Aerobic bacteria, Gram-negative
18:0		0.50	Biomass all organisms
a15:0		-0.38	Gram positive bacteria
18:3ω6c		0.37	Fungi
Canonical variable 3			
i17:0		0.38	Gram-postive bacteria
a18:0/18:2ω6,9c		-0.42	Grampositive/ Fungi
16:1 20H		0.37	Nonspecific
cy 17:0		0.37	Gram-negative
17:0 10 methyl		0.36	Actinomycetes
Canonical variable 4			
14:0		0.47	Biomass all organisms
i15:0		0.46	Gram-postive bacteria
a15:0		0.42	Gram-postive bacteria
18:0		-0.32	Biomass all organisms
16:0 10 methyl		0.30	Actinomycetes

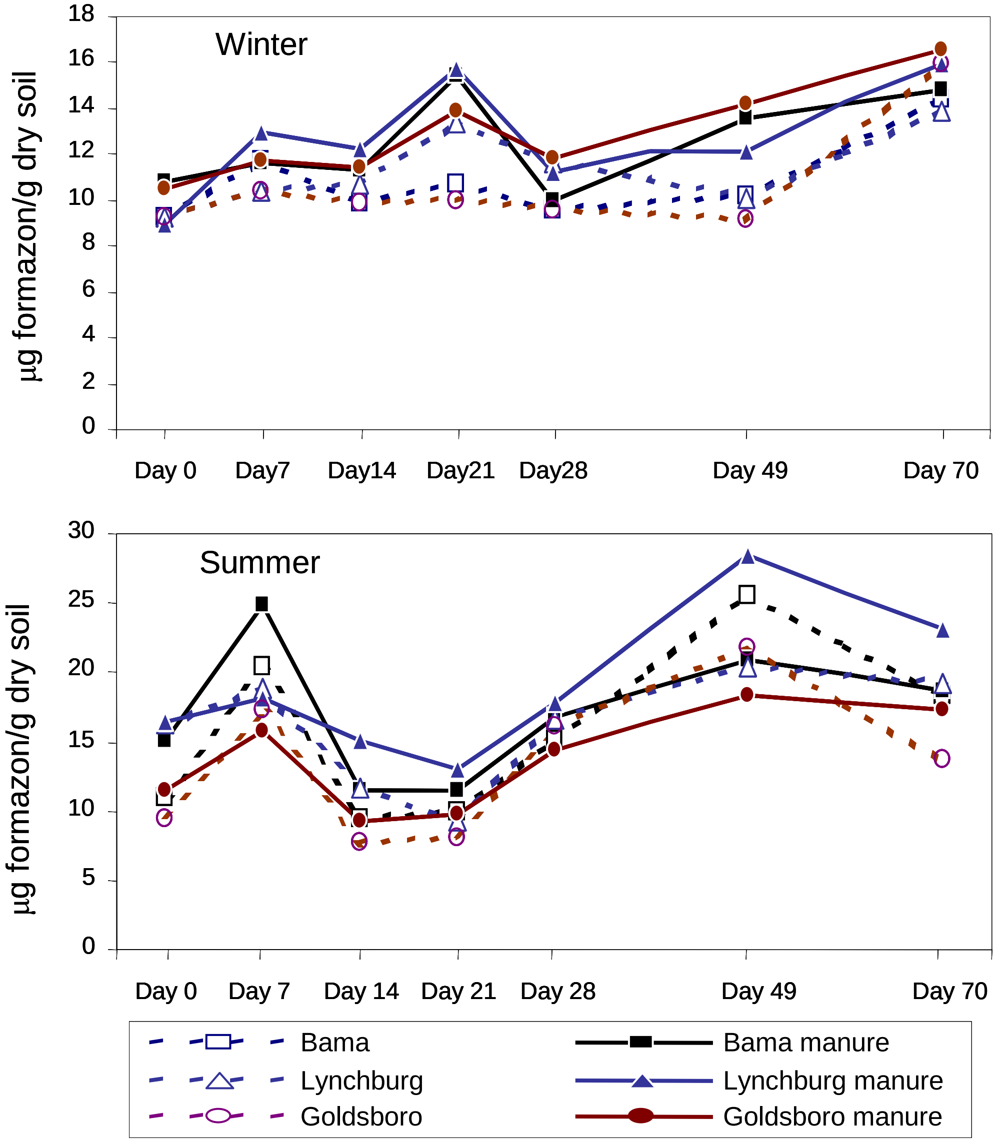

Figure 1. Dehydrogenase enzyme activity interactive effects of soil type and dairy compost manure for the summer and winter season.

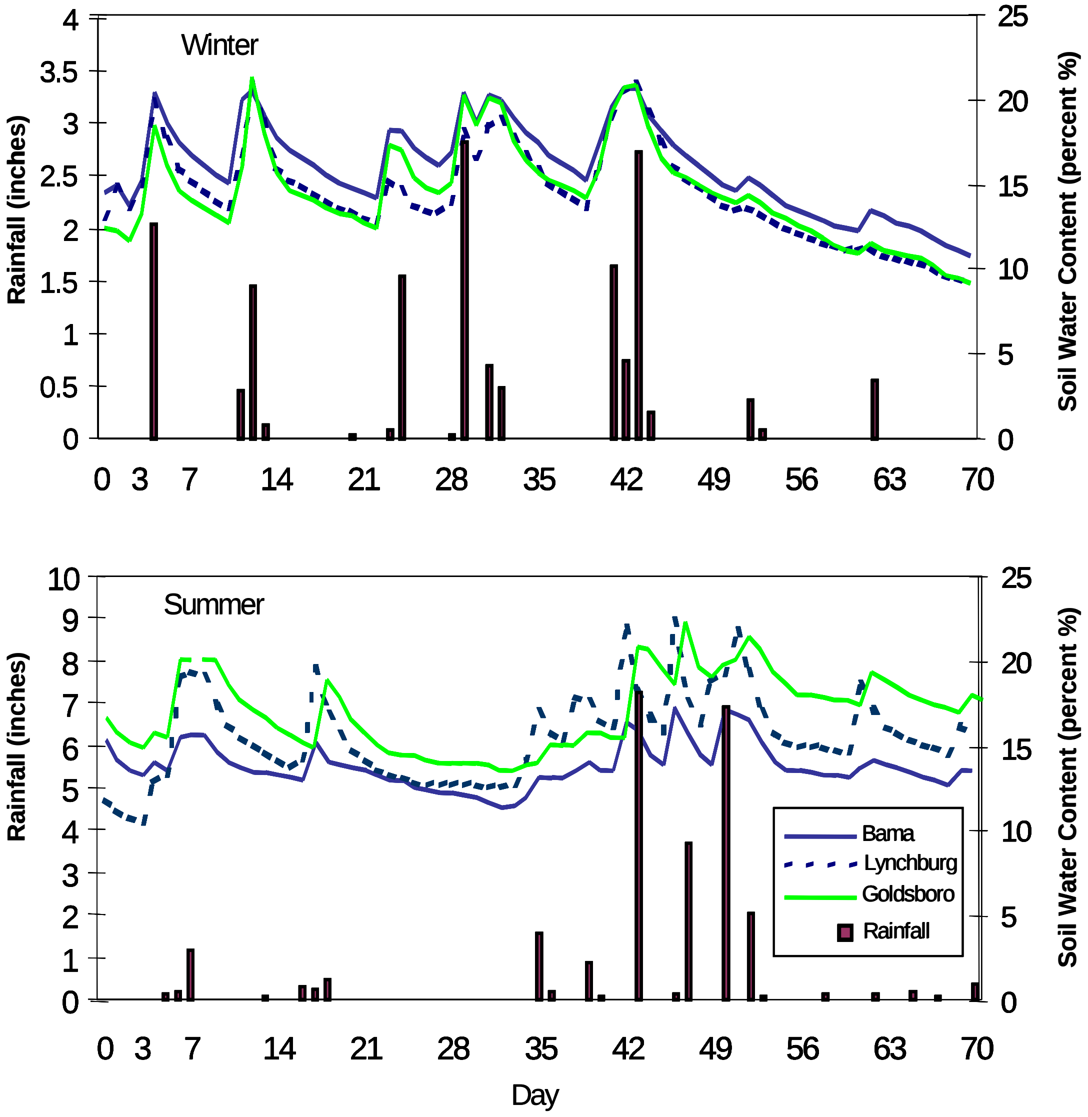

Figure 2. Seasonal mean soil moisture and average rainfall (Winter 2004 and Summer 2005) in the microplot cylinders for three soil types during the season.

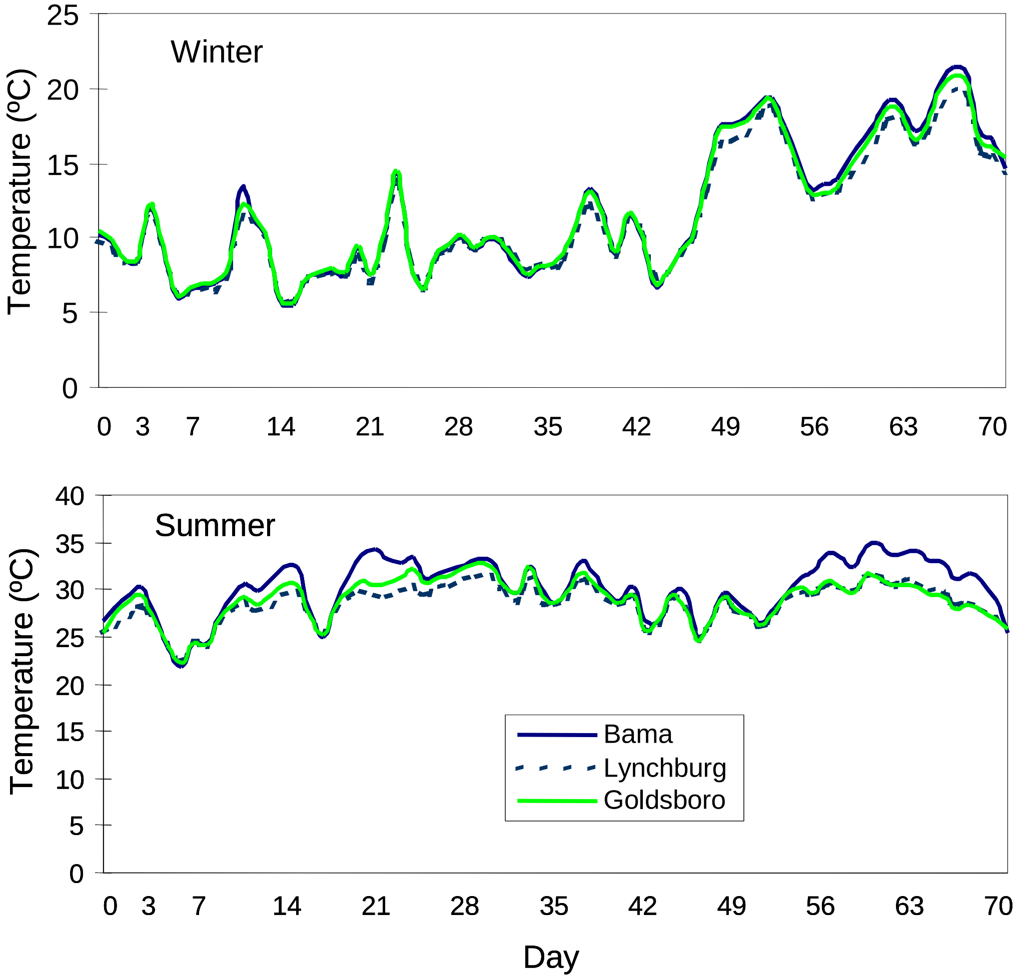

Figure 3. Seasonal mean soil temperature (Winter 2004 and Summer 2005) in the microplot cylinders for three soil types during the season.

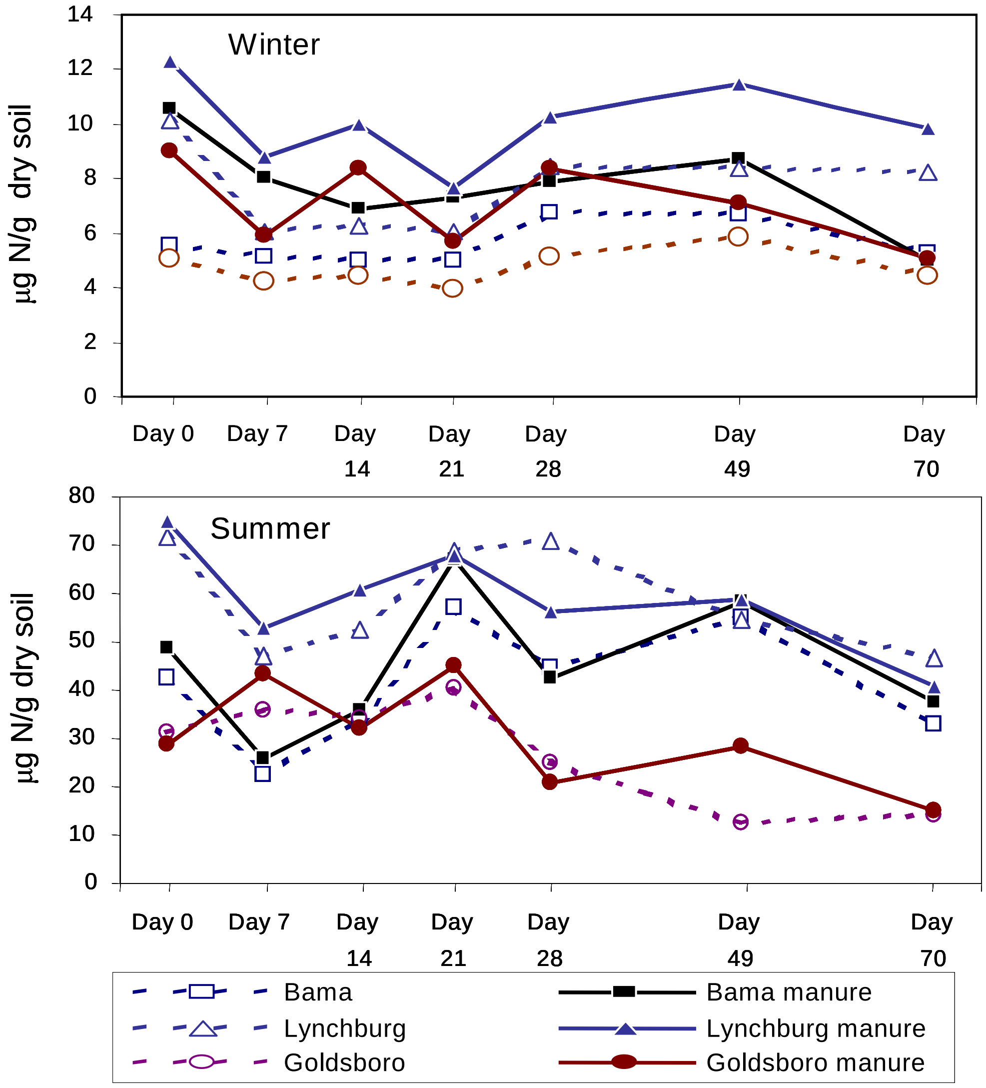

Figure 4. Microbial biomass N interactive effects of soil type and dairy compost manure for the summer and winter season.

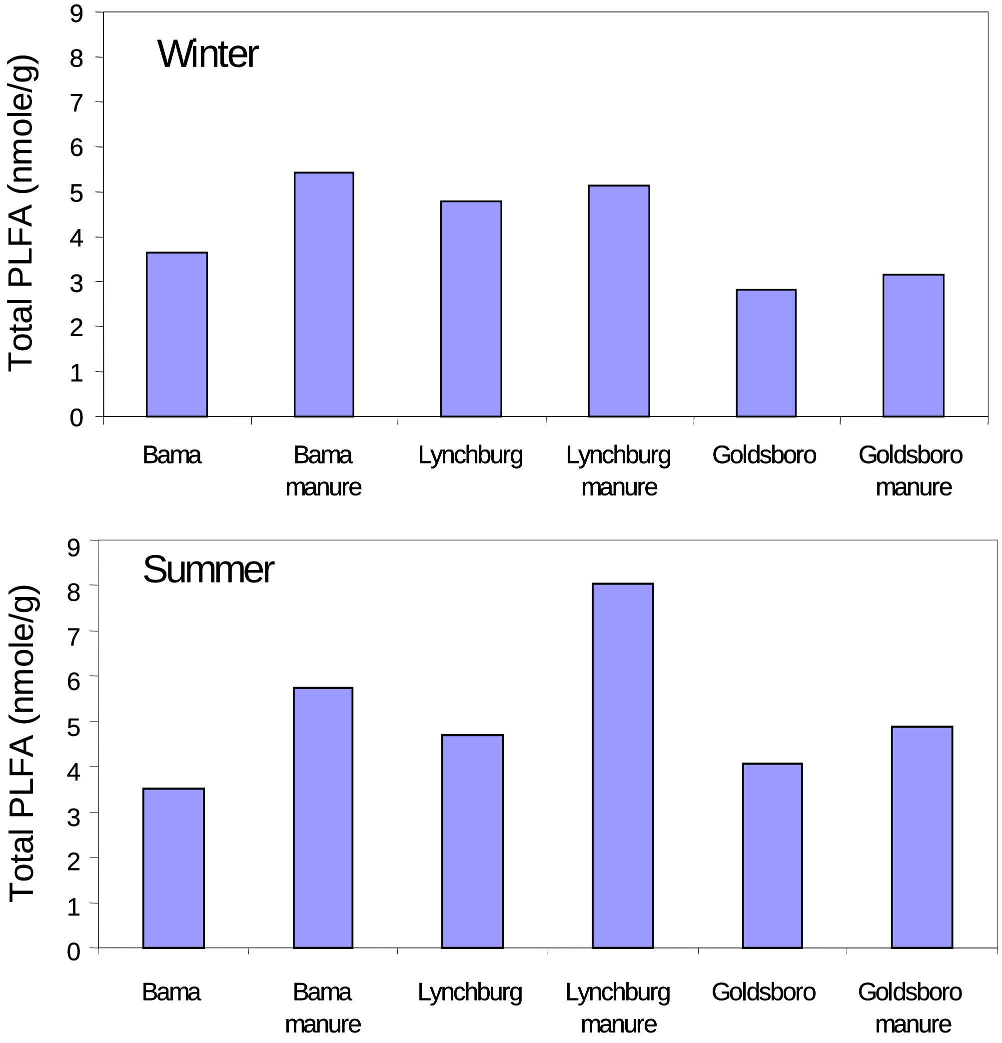

Figure 5. Total phospholipids fatty acids (PLFA) for samples collected on the last day of the incubation of the *in situ* soil cores.

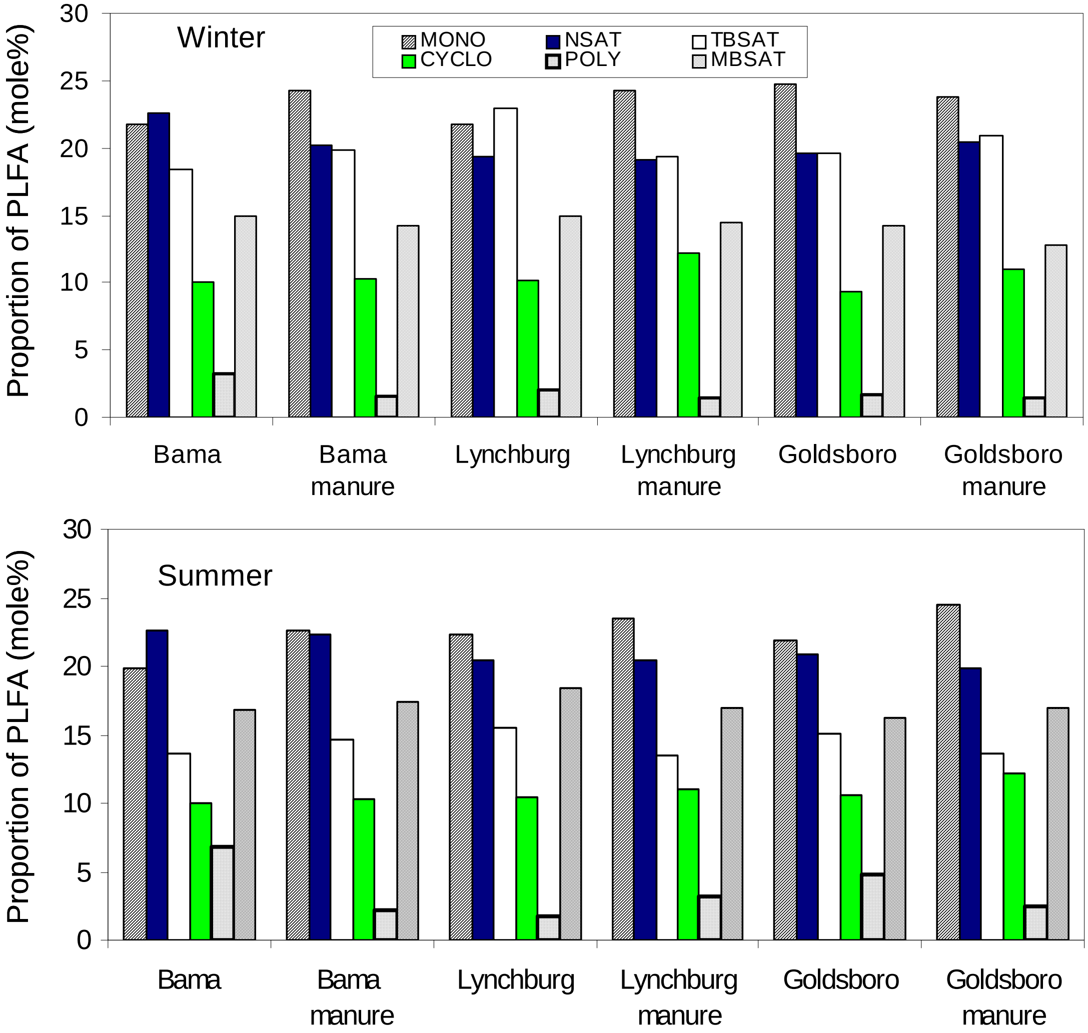

Figure 6. The relative microbial community composition of different functional groups of biomarkers for the Bama, Lynchburg and Goldsboro with and without dairy compost manure for the winter and summer months.

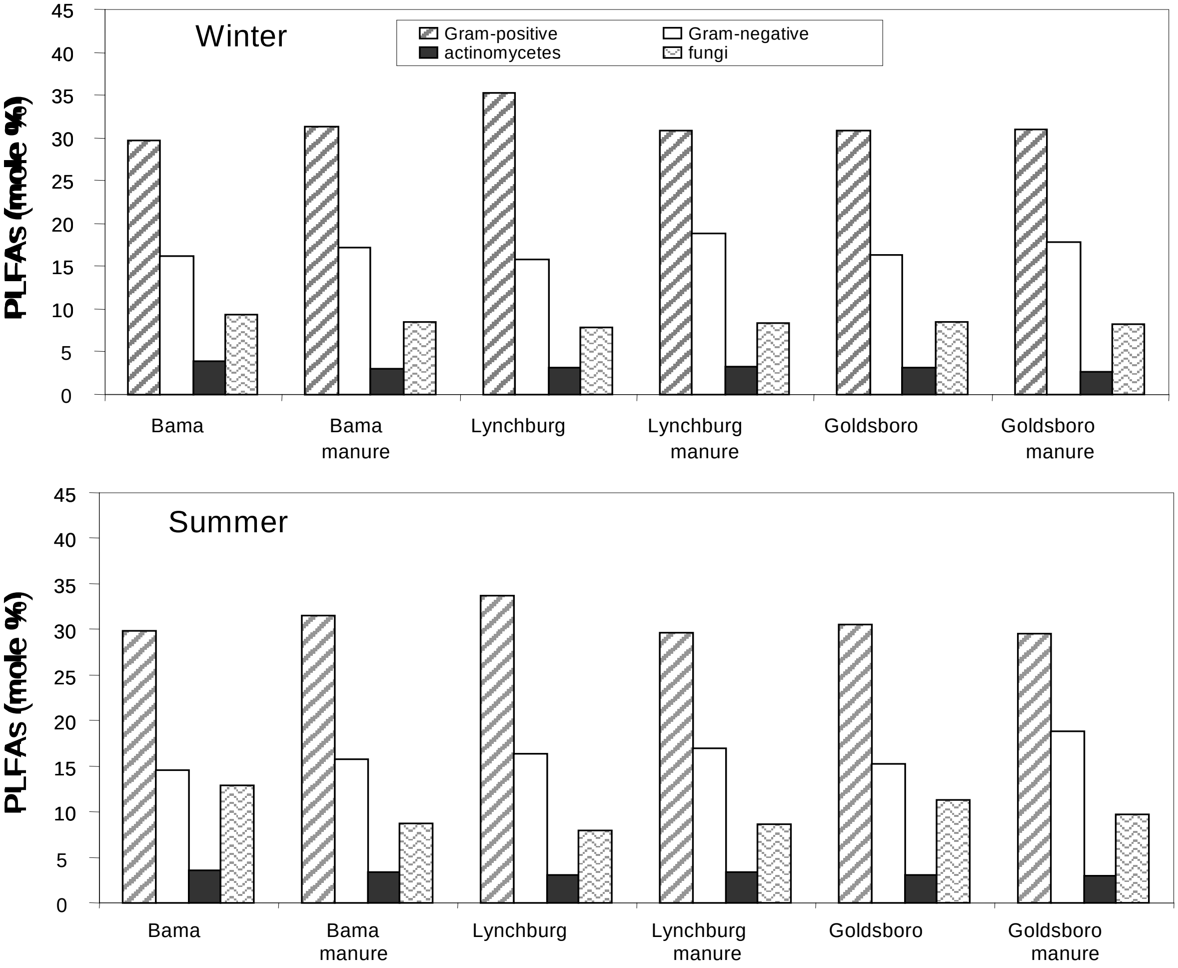

Figure 7. The relative microbial community composition of different functional groups for Gram-positive and Gram-negative bacteria, fungi, actinomycetes for the Bama, Lynchburg and Goldsboro with and without dairy compost manure for the winter and summer months.

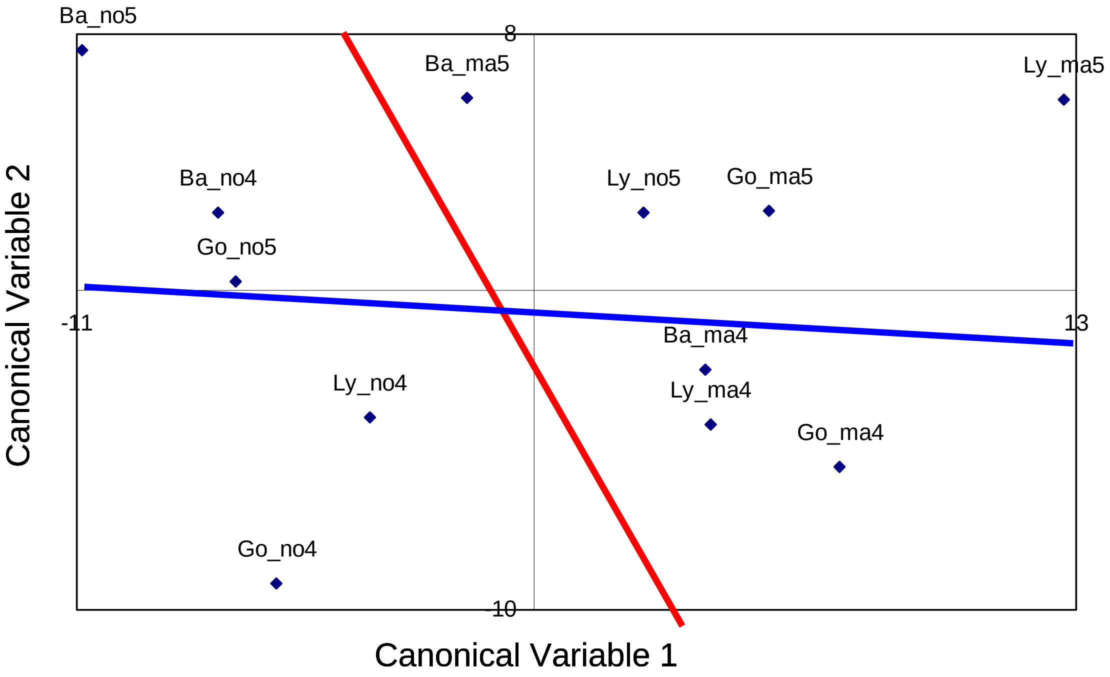

Figure 8. Canonical discriminant analysis (CDA) of phospholipid fatty acid profiles for the canonical variates (CV). Plot of ordination of CV1 against CV2 during the summer (05) and winter (04) months for the Bama (Ba), Lynchburg (Ly) and Goldsboro (Go) soil with (ma) and without dairy compost manure (no).

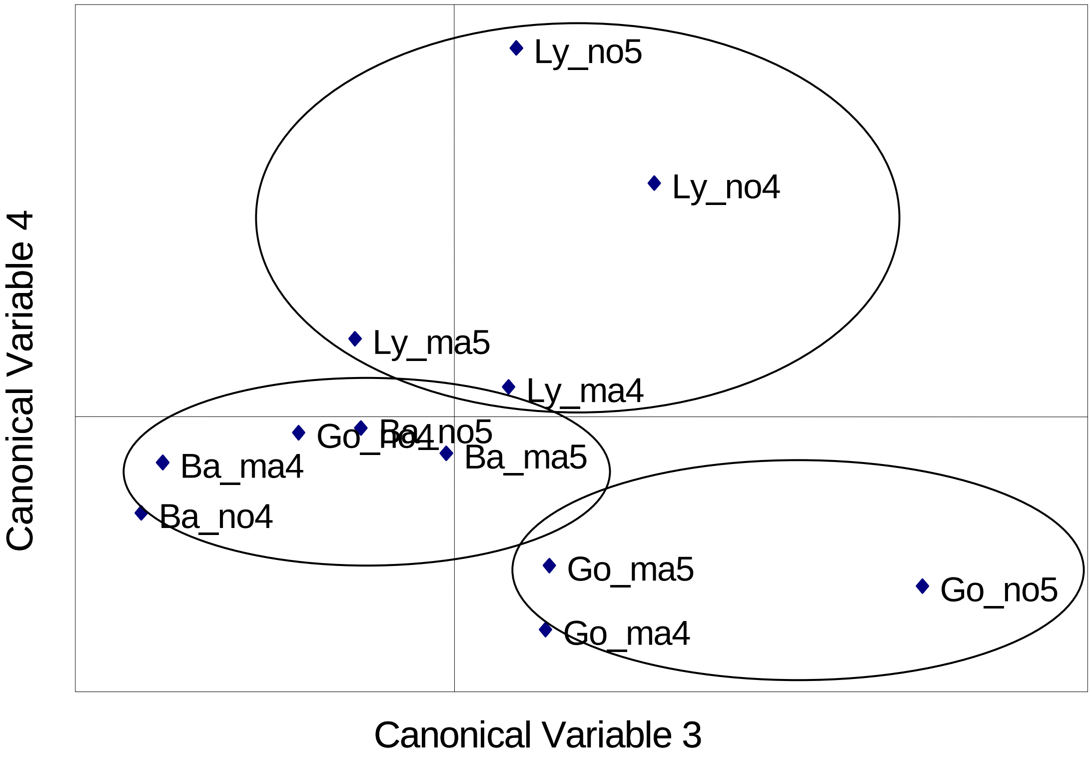

Figure 9. Canonical discriminant analysis (CDA) of phospholipid fatty acid profiles from for the canonical variates (CV). Plot of ordination of CV3 against CV4 during the summer (05) and winter (04) months for the Bama (Ba), Lynchburg (Ly) and Goldsboro (Go) soil with (ma) and without dairy compost manure (no).