

Soil Moisture and Temperature Effects on Nitrogen Release from Organic Nitrogen Sources

S. Agehara* and D. D. Warncke

ABSTRACT

Nitrogen release from organic N sources is controlled by the soil environment. Soil incubation was conducted to evaluate the effects of soil moisture (50, 70, and 90% of water holding capacity) and temperature (15/10, 20/15, and 25/20°C [14/10 h]) on N release from four organic N sources. Differential N release kinetics of the N sources were determined by measuring ammonium- and nitrate-N contents periodically over 12 wk. Net N released, as a percentage of organic N, was greatest in the order: urea (91–96%) > blood meal (BM) (56–61%) > alfalfa pellets (AP) (41–52%) > partially composted chicken manure (CM) (37–45%). Increasing soil moisture increased net N released from AP and CM by 12 and 21%, respectively, but did not significantly affect net N released from urea and BM. Increasing temperature increased net N released from AP, BM, and CM by 25, 10, and 13%, respectively, but did not significantly affect net N released from urea. The results indicate that soil moisture and temperature influence N availability from organic N materials differently depending on source of N. In greenhouse production systems, where irrigation and temperature can be controlled, fertilizer management that considers both source of N and soil environment may improve the effectiveness of organic N materials.

NITROGEN AVAILABILITY from applied N sources must be known for efficient management of N inputs. Because N release process of organic N sources involves a biological decomposition, the N availability is controlled by chemical composition (Fox et al., 1990; Ajwa et al., 1998; Rowell et al., 2001; Kumar and Goh, 2003) and soil environment (Sims, 1986; Vigil and Kissel, 1995; Seneviratne et al., 1998; Whalen et al., 2001; Cookson et al., 2002). Thus, it is difficult to predict the pattern and amount of available N released from organic N sources during the growing season. Sufficient N is necessary for optimum crop production, whereas excessive fertilization will increase the cost of production and the risk of nitrate leaching. From a management standpoint, it is important to understand how chemical composition and soil environment influence availability of N from organic N sources.

Availability of N from organic N sources varies considerably. For example, urea [CO(NH₂)₂], a synthetic organic N fertilizer, is rapidly hydrolyzed to ammonium (NH₄⁺) by the enzyme urease after being applied to soil. MacLean and McRae (1987) found rapid hydrolysis

rates of over 90% within 5 d at temperatures between 9 and 18°C in an acid podzolic soil. Tomar and Soper (1981) reported that urea hydrolyzed after 4 wk of incubation averaged 83% in 11 different soils. Unlike urea, natural organic materials, such as animal manures and plant residues, are mineralized slowly to NH₄⁺. Chae and Tabatabai (1986) reported that the percentage of total N mineralized from cow, hog, and chicken manure averaged 35, 39, and 53%, respectively, in five different soils over 26 wk of incubation. Li and Mahler (1995) reported that when ground alfalfa, spring pea, and winter wheat were incorporated with soil at the rate of 2% (w/w), the percentage of total N mineralized was 31, 23, and 16%, respectively, after 20 wk of incubation. Ciavatta et al. (1997) found that 75% of the total N in BM was mineralized after a 120-d incubation. The variation in N availability among different types of animal manures and plant residues has been attributed to the chemical composition, such as total N content (Fox et al., 1990; Constantinides and Fownes, 1994; Aulakh et al., 2000), C/N ratio (Aulakh et al., 2000; Trinsoutrot et al., 2000; Rowell et al., 2001), lignin/N ratio (Melillo et al., 1982; Constantinides and Fownes, 1994; Kumar and Goh, 2003), and polyphenol content (Fox et al., 1990; Palm and Sanchez, 1991; Constantinides and Fownes, 1994).

Soil moisture and temperature are the major environmental factors affecting N availability from organic N sources. Because urea is readily soluble in water, urea hydrolysis is largely dependent on diffusion of dissolved urea in soil (Sadeghi et al., 1989). Urease activity is generally highest near field capacity and declines as soil moisture decreases (Vlek and Carter, 1983; Sahrawat, 1984). Urea hydrolysis is also accelerated with increasing temperature as urea diffusion rate in soil is positively correlated with temperature (Pang et al., 1977; Sadeghi et al., 1988). Urease activity increases in relation to temperature with maximum urea hydrolysis occurring between 60 and 70°C (Overrein and Moe, 1967; Sahrawat, 1984; Moyo et al., 1989; Lai and Tabatabai, 1992). MacLean and McRae (1987) reported that 52, 67, 80, and 93% of urea was hydrolyzed at 4, 9, 13, and 18°C, respectively, after 3 d of incubation.

Mineralization of natural organic materials is mediated by heterotrophic bacteria and fungi. Soil moisture regulates oxygen diffusion in soil with maximum aerobic microbial activity occurring at soil moisture levels between 50 and 70% of water holding capacity (WHC) (Linn and Doran, 1984; Franzluebbers, 1999). On the other hand, low soil moisture inhibits microbial activity by reducing diffusion of soluble substrates (Griffin, 1981;

S. Agehara and D.D. Warncke, Dep. of Crop and Soil Sciences, Michigan State Univ., East Lansing, MI 48824-1325; S. Agehara currently at Texas Agricultural Experiment Station, Texas A&M Univ., 1619 Garner Field Rd., Uvalde, TX 78801. Received 22 Nov. 2004. *Corresponding author (ageharas@hotmail.com).

Published in Soil Sci. Soc. Am. J. 69:1844–1855 (2005).
Nutrient Management & Soil & Plant Analysis
doi:10.2136/sssaj2004.0361

© Soil Science Society of America
677 S. Segoe Rd., Madison, WI 53711 USA

Abbreviations: AP, alfalfa pellets; BM, blood meal; CEC, cation exchange capacity; CM, partially composted chicken manure; OM, organic matter; SAS, Statistical Analysis System; TKN, total Kjeldahl-nitrogen; WHC, water holding capacity.

Schjønning et al., 2003), microbial mobility (Killham et al., 1993), and intracellular water potential (Csonka, 1989; Stark and Firestone, 1995). Doel et al. (1990) incubated soil amended with white lupin at -0.30 , -0.03 , and -0.01 MPa for 198 d. Although immobilization was not overcome at -0.30 MPa throughout incubation, net mineralization occurred at -0.03 and -0.01 MPa after 187 and 168 d of incubation, respectively. De Neve and Hofman (2002) reported that net mineralized N from fresh carrot leaves increased with increasing soil moisture from 18 to 45% WHC, but was constant up to 60% WHC after 98 d of incubation. Similarly, increasing temperature enhances mineralization by stimulating microbial activity and accelerating diffusion of soluble substrates in soil (Nicolardot et al., 1994; MacDonald et al., 1995; Zak et al., 1999). An increase in temperature also induces a shift in the composition of microbial communities (Richards et al., 1985; Carreiro and Koske, 1992), which parallels an increase in microbial activity (Zogg et al., 1997). Griffin and Honeycutt (2000) incubated soil amended with dairy, poultry, and swine manures for 112 d, and found that increasing temperature from 10 to 24°C significantly accelerated the mineralization rate. Cookson et al. (2002) reported that 22, 33, 41, and 60% of the total N in clover residues was mineralized at 2, 5, 10, and 15°C, respectively, after 160 d of incubation.

Although many studies have evaluated the relationships between N availability and chemical composition, soil moisture, or temperature for various organic N sources, few have focused on an interaction between source of N and soil environment. Soil moisture and temperature may influence N availability from organic N sources differently depending on chemical composition. Such information will be valuable to making decisions for the efficient use of organic N sources. In greenhouse production systems, use of organic N sources is currently limited, but interest in their use is increasing. When growers choose to use organic N sources, determination of the most appropriate rate and timing of fertilization may be possible, especially under controlled irrigation and temperature conditions.

Four organic N sources in this study, including urea, AP, BM, and CM, were chosen because of wide variation in their chemical composition. The objectives of this study were to: (i) examine the effects of soil moisture and temperature on N release from the organic N sources, and (ii) determine if soil moisture and temperature effects on N release vary among the organic N sources.

MATERIALS AND METHODS

Soil

The soil used in this study was a Granby sandy clay loam (sandy, mixed, mesic Typic Endoaquolls). Approximately 20 kg of surface soil (15 cm) was collected from the Michigan State University Horticulture Teaching and Research Center in East Lansing MI, in May 2002 and August 2003. The soil was passed through a 5-mm sieve, thoroughly mixed to ensure uniformity, and stored in a covered plastic container under field moisture condition (10% w/w) at room temperature (20–23°C) until

Table 1. Chemical and physical properties of soils used in this study.

Property	Soil (2002) [†]	Soil (2003) [‡]
pH	5.7	5.8
SMP buffer pH	6.4	6.5
Sand, %	54.7	54.7
Silt, %	17.4	15.4
Clay, %	27.8	29.8
CEC, cmol kg ⁻¹	11.3	11.1
OM content, g kg ⁻¹	36.0	34.5
Organic C, g kg ⁻¹	20.9	20.0
Total N, g kg ⁻¹	4.4	4.1
NH ₄ ⁺ -N, mg kg ⁻¹ §	5.4	5.1
NO ₃ ⁻ -N, mg kg ⁻¹ §	22.0	17.8
Bray P, mg kg ⁻¹	206	175
Exchangeable K, mg kg ⁻¹	133	198
Exchangeable Ca, mg kg ⁻¹	546	627
Exchangeable Mg, mg kg ⁻¹	176	99

[†] Soil (2002) was used in the soil moisture and temperature studies performed in 2002.

[‡] Soil (2003) was used in the temperature study performed in 2003.

§ 1 M KCl extractable.

the incubation was initiated to minimize disturbance of the microbial population (Primer and Bartha, 1972; Honeycutt, 1999).

The chemical and physical properties of the soil are listed in Table 1. Soil samples for analysis were, unless otherwise noted, immediately dried at 38°C for 48 h and ground to pass through a 2-mm sieve. Soil pH and SMP buffer pH were measured with a combination reference/glass pH electrode using 5 g of soil, 5 mL of water, and 10 mL of SMP buffer (Watson and Brown, 1998). Total Kjeldahl-N (TKN) content was determined by the micro-Kjeldahl digestion procedure (Bremner and Mulvaney, 1982) followed by colorimetric determination using a Lachat rapid flow injection autoanalyzer (Lachat Instruments, Milwaukee, WI). Ammonium (NH₄⁺) and nitrate (NO₃⁻) N were determined by the ammonia-salicylate method and cadmium reduction method, respectively, using a Lachat rapid flow injection autoanalyzer following extraction with 1 M KCl. The extracts were filtered through #2 Whatman filter paper previously washed with 1 M KCl. No attempt was made to measure nitrite (NO₂⁻) N separately in the extracts, but NO₂⁻-N was measured with the NO₃⁻-N. Since TKN does not account for NO₃⁻-N, total N was calculated as the sum of TKN and NO₃⁻-N. Available P was determined colorimetrically by the Bray and Kurtz P-1 method (Frank et al., 1998). Exchangeable K, Ca, and Mg were extracted with 1 M NH₄OAc and measured by an atomic absorption spectrometer (Warncke and Brown, 1998). Cation exchange capacity (CEC) was estimated by summing the exchangeable acidity, which was obtained from SMP buffer pH measurement, and the exchangeable bases (K, Ca, and Mg) (Warncke and Brown, 1998). Soil samples for C analysis were dried at 105°C for 48 h and ground to pass through a 0.15-mm sieve. Organic C content was determined by dry combustion using a Leco carbon analyzer (Leco Corp., St. Joseph, MI). Organic matter (OM) content was estimated by multiplying the organic C content by 1.72 (Combs and Nathan, 1998). The unground soil samples dried at 38°C for 48 h and passed through a 2-mm sieve were used for particle-size analysis. Soil particle-size distribution was estimated by the hydrometer method (Gee and Bauder, 1986). For determination of WHC, soil contained in a 15-cm deep pot was saturated by setting pots in water. The covered pots were allowed to drain for 48 h. The water content of soil in the 3- to 7-cm depth was considered 100% WHC. Soil gravimetric water content was determined by oven drying samples at 105°C for 24 h. All soil analyses were performed in duplicate or triplicate.

Table 2. Chemical characteristics of four organic N sources used in this study.†

N source‡	Moisture	pH	Total C	Total N	C/N	Lignin	Lignin/N	NH ₄ ⁺	NO ₃ ⁻	P	K	Ca	Mg
	%		%	%		%		— g N kg ⁻¹ —			%		
Urea	1.41	6.99	19.7	46.00	0.43	—	—	—	—	—	—	—	—
AP	4.46	5.73	39.3	3.60	11.13	9.28	2.63	0.49	0.15	0.58	4.55	1.69	0.16
BM	5.01	7.30	41.3	12.65	3.27	—	—	0.15	0.02	0.51	0.64	1.12	0.11
CM	7.92	8.69	26.6	3.80	7.22	2.29	0.62	1.02	0.15	2.71	3.30	14.42	0.80

† All values are expressed on a dry basis (105°C). Organic N was used to calculate C/N and lignin/N ratios.

‡ AP = alfalfa pellets; BM = blood meal; CM = partially composted chicken manure.

Nitrogen Sources

Urea [CO(NH₂)₂] was used as a synthetic organic N fertilizer. Three natural organic materials were used. Alfalfa pellets (AP), which were obtained from Bradfield Industries, Inc. (Springfield, MI), are alfalfa-based fertilizers blended with animal protein, natural sulfate of potash, and molasses. Blood meal (BM) was obtained from Glorious Gardens Blood Meal Growing Markets, Inc. (West Des Moines, IA). Partially composted chicken manure (CM) was obtained from Herbruck's Poultry Ranch, Inc. (Saranac, MI).

The chemical properties of these N sources are listed in Table 2. To facilitate uniform distribution in the soil, urea was ground to be in powder form, and AP and CM were ground to pass through a 1-mm sieve before use. Since BM was originally powdered, it was used without grinding. The pH (5 g N material:10 mL water), total C, total N, NH₄⁺-N, and NO₃⁻-N were determined by the same procedures used for soil. Lignin content was determined by the acid detergent fiber method (Goering and Van Soest, 1970). Total P, K, Ca, and Mg contents were measured by a direct current plasma atomic emission spectrophotometer following dry ashing at 500°C and digestion with 3 M HNO₃ containing 1000 ppm of LiCl. Dry matter weight was determined by oven drying samples at 105°C for 24 h. All analyses were performed in duplicate.

Experimental Design

The N release kinetics of the organic N sources were determined at different soil moistures and temperatures. The soil moisture study was conducted during June to August 2002 using the soil collected in May 2002. The experimental design was a completely randomized design with three replications. Treatments consisted of a factorial combination of three soil moisture levels (50, 70, and 90% WHC), five N sources (control, urea, AP, BM, and CM), and six incubation times (0, 1, 2, 4, 8, and 12 wk).

The experimental design for the temperature study was a split-plot design with temperature designated as a main-plot. Treatments consisted of a factorial combination of three temperature levels [15/10, 20/15, and 25/20°C day/night (14/10 h)], five N sources (control, urea, AP, BM, and CM), and six incubation times (0, 1, 2, 4, 8, and 12 wk). The temperature levels were assigned randomly to three incubation chambers and the experiment was conducted in two runs (during June to August 2002 with the soil collected in May 2002 and August to October 2003 with the soil collected in August 2003), which provided two replications for temperature effect. Within each temperature, N source and incubation time were designated as subplots, and each combination of subplot factors was assigned randomly to three experimental flasks.

Soil Incubation

Twenty grams dry weight equivalent of moist soil (10% w/w) was placed in 125 mL Erlenmeyer flasks. Urea, AP, BM, and CM were mixed with the soil at the rate of 63, 100, 92, and 150 mg N kg⁻¹ soil (oven dry basis), respectively. The applica-

tion rates were calculated to provide approximately equal amounts of 60 mg available N kg⁻¹ soil using Eq. [1].

$$\text{Estimated available } N = N_i + fN_o \quad [1]$$

where N_i is the inorganic N (NH₄⁺-N + NO₃⁻-N) content, N_o is the organic N (total N - inorganic N) content, and f is the proportion of organic N fraction expected to be mineralized during the incubation (Griffin and Honeycutt, 2000). Coefficient f of 0.95, 0.59, 0.65, and 0.39 was applied for urea, AP, BM, and CM, respectively, based on a previous 2-wk soil incubation experiment that used the same procedure as in this study.

In the soil moisture study, the samples were treated with distilled water to provide 50, 70, and 90% WHC, and were randomly placed in an incubation chamber set at 20/15°C day/night (14/10 h). In the temperature study, the samples were treated with distilled water to provide 70% WHC, and were randomly placed in the incubation chambers set at 15/10, 20/15, and 25/20°C day/night (14/10 h).

Incubation was performed in dark condition. The flasks were covered with parafilm to retard moisture loss from the samples. Soil moisture was adjusted every week by weighing the samples and adding the required amount of distilled water when the loss was greater than 0.05 g. Soil samples with no N source were incubated as a control to estimate soil N mineralization and subtract from the treatments to be able to estimate the rate of N release from the organic N sources.

Inorganic Nitrogen Analysis and Calculations for Nitrogen Release

Initial NH₄⁺- and NO₃⁻-N contents were determined in samples extracted with 50 mL of 1 M KCl added directly to the flask immediately after incorporation of each N source. After 1, 2, 4, 8, and 12 wk of incubation, samples were removed from the growth chambers and extracted with 50 mL of 1 M KCl for determination of NH₄⁺- and NO₃⁻-N contents. The NO₂⁻-N concentration was assumed to be insignificant; however, any NO₂⁻-N present was included in the mineralized N pool labeled as NO₃⁻-N. The extracts were analyzed within a week after extraction, otherwise they were stored at -20°C until the analysis was performed.

The cumulative amount of N mineralized from soil OM at time t , (N_{\min})_{control}, was calculated from Eq. [2].

$$(N_{\min})_{\text{control}} = N_i (\text{control})_t - N_i (\text{control})_{t=0} \quad [2]$$

All numbers are in the units mg N kg⁻¹ soil.

The cumulative amount of N released from an applied N source at time t , (N_{rel})_{N source}, was calculated from Eq. [3].

$$(N_{\text{rel}})_{\text{N source}} = N_i (\text{N-treated soil})_t - N_i (\text{control})_t - N_i (\text{N source}) \quad [3]$$

All numbers are in the units mg N kg⁻¹ soil. The cumulative amount of NH₄⁺-N or NO₃⁻-N released from an applied N source at time t was calculated in a same manner.

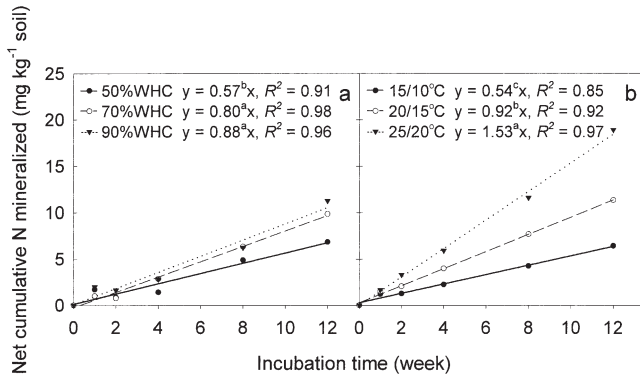


Fig. 1. Net cumulative N mineralized from soil organic matter at different soil moistures (a) and temperatures (b). Slopes in regression lines followed by the same letter are not significantly different at $\alpha = 0.05$.

The percentage of organic N released from an applied N source at time t , $(\%N_{rel})_{N\ source}$, was calculated from Eq. [4].

$$(\%N_{rel})_{N\ source} = [(N_{rel})_{N\ source}/N_o (N\ source)] \times 100 \quad [4]$$

Statistical Analysis and Nitrogen Release Models

A three-way analysis of variance (ANOVA) was conducted to test significant differences in treatment effects and interactions using the MIXED procedure of Statistical Analysis System (SAS) (SAS Institute, 1990). Data from all individual flasks were used in ANOVA. The assumption of homogeneity of variance was tested with Levene's test at $\alpha = 0.05$. The REPEATED/GROUP was used to account for unequal variances. When statistically significant differences existed according to ANOVA ($P < 0.05$), treatment means were separated using the PDIFF option of LSMEANS, and then compared using pairwise t test at $\alpha = 0.05$.

Soil N mineralization was fit to a zero-order model using the linear regression procedure REG (SAS Institute, 1990) as follows:

$$N_{min} = kt \quad [5]$$

where N_{min} (mg N kg⁻¹) is the cumulative N mineralized from soil OM at time t , and k (mg N kg⁻¹ wk⁻¹) is the zero-order rate constant. Significant differences among slopes, k , were tested with orthogonal contrasts.

Nitrogen release from the organic N sources was fit to a first-order model (Stanford and Smith, 1972) using the non-linear curve-fitting procedure NLIN (SAS Institute, 1990) as follows:

$$N_{rel} = N_0[1 - \exp(-k_0t)] \quad [6]$$

where N_{rel} (mg N kg⁻¹) is the cumulative N released from an applied N source at time t , N_0 (mg N kg⁻¹) is the size of potentially mineralizable N, \exp is the exponential constant with numerical value $\cong 2.718$, and k_0 (wk⁻¹) is the first-order rate constant. The N_0 and k_0 values were deemed significantly different ($\alpha = 0.05$) if the 95% confidence intervals did not overlap.

All equations were fit using all data points, although only mean values are shown in the figures below.

RESULTS AND DISCUSSION

Patterns of Nitrogen Release

Three different patterns of N release were shown in both soil moisture and temperature studies. In the

Table 3. Net cumulative N released, as a percentage of organic N, from four organic N sources incubated in soil at different soil moistures.†

N source	Soil moisture	Incubation time (wk)				
		1	2	4	8	12
	% WHC	%				
Urea	50	79.6b	86.3a	83.7b	87.8a	91.2a
	70	82.0b	85.7a	88.4a	89.7a	91.7a
	90	93.4a	84.6a	92.1a	88.8a	95.5a
	Mean	85.0	85.5	88.0	88.8	92.8
AP	50	15.3e	25.5d	30.4e	34.8d	41.1d
	70	15.9e	22.4de	29.5e	40.0c	44.0cd
	90	14.0e	20.6e	30.9e	40.7c	45.9c
	Mean	15.1	22.8	30.3	38.5	43.7
BM	50	23.1d	32.7c	40.2d	48.5b	57.4b
	70	28.5c	39.5b	40.7cd	49.5b	57.4b
	90	31.0c	38.8b	43.9c	51.1b	56.9b
	Mean	27.5	37.0	41.6	49.7	57.3
CM	50	22.3d	32.8c	30.7de	35.3cd	36.7d
	70	26.0cd	30.7cd	28.8e	41.2cd	41.3cd
	90	26.1cd	25.0d	37.8d	40.2cd	44.3c
	Mean	24.8	29.5	32.4	38.9	40.7

† Means in a column followed by the same letter are not significantly different according to t test at $\alpha = 0.05$.

control, cumulative mineralized N was linearly correlated with time; the R^2 values were greater than 0.85 at all soil moisture and temperature levels (Fig. 1). Soil N mineralization proceeded slowly throughout incubation, and only a small portion of soil organic N ($< 0.5\%$) was mineralized during the incubation.

In the urea treatment, a rapid N release occurred with over 75% of urea hydrolyzed in the first week (Table 3 and 4). The rate of hydrolysis in Weeks 2 through 12 was very slow. Over 90% of urea was hydrolyzed at the end of incubation (Table 3 and 4). This rapid N release pattern demonstrated the typical urea hydrolysis reported in previous studies (Tomar and Soper, 1981; Vlek and Carter, 1983; MacLean and McRae, 1987).

In the AP, BM, and CM treatments, N release showed two distinct phases during incubation; a rapid phase in the first 2 wk followed by a slow phase in Weeks 2

Table 4. Net cumulative N released, as a percentage of organic N, from four organic N sources incubated in soil at different temperatures.†

N source	Temperature	Incubation time (wk)				
		1	2	4	8	12
	°C (14/10 h)	%				
Urea	15/10	75.1b	86.6b	89.5a	90.9a	93.2a
	20/15	86.6a	90.1ab	91.0a	91.5a	92.6a
	25/20	89.5a	92.1a	92.1a	90.0a	92.9a
	Mean	83.7	89.6	90.9	90.8	92.9
AP	15/10	11.1g	17.3i	24.1g	33.3h	42.1de
	20/15	14.8fg	23.7h	30.4f	40.5fg	45.9d
	25/20	21.5e	29.8f	39.3de	47.6de	52.4c
	Mean	15.8	23.6	31.2	40.5	46.8
BM	15/10	18.2ef	36.7de	41.9cd	49.4cd	55.5c
	20/15	29.9c	40.6d	45.6c	53.9c	60.2b
	25/20	31.2c	45.9c	56.2b	59.2b	61.0b
	Mean	26.4	41.1	47.9	54.2	58.9
CM	15/10	21.7de	24.6gh	29.3f	36.0gh	39.7e
	20/15	27.3cd	30.7f	33.9ef	42.4ef	43.4d
	25/20	29.5c	32.2ef	41.2cd	44.7def	45.0d
	Mean	26.2	29.2	34.8	41.0	42.7

† Means in a column followed by the same letter are not significantly different according to t test at $\alpha = 0.05$.

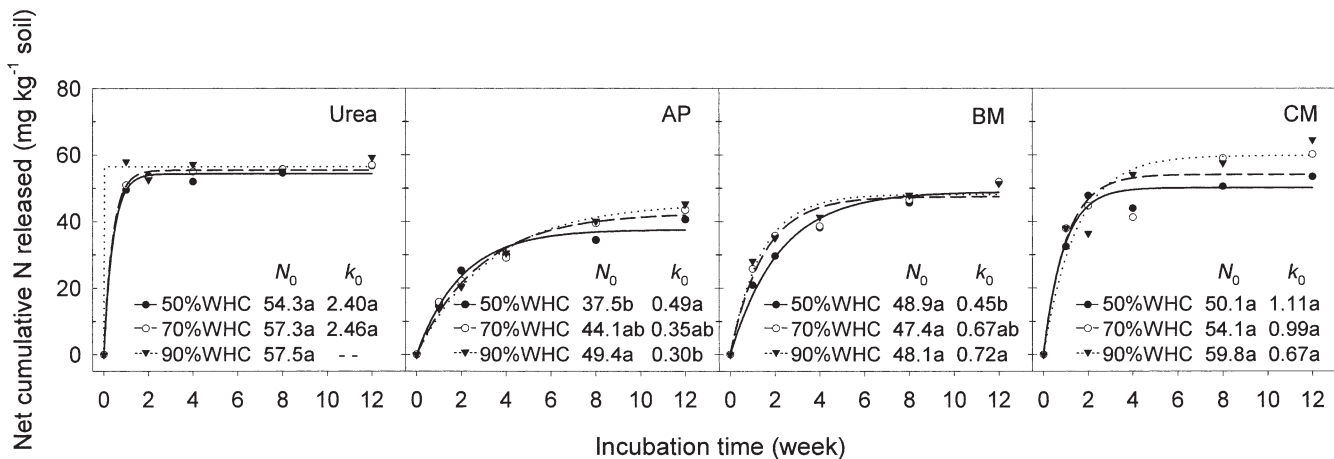


Fig. 2. Net cumulative N released from four organic N sources at different soil moistures. The N_0 or k_0 values followed by the same letter are not significantly different at $\alpha = 0.05$. AP, alfalfa pellets; BM, blood meal; CM, partially composted chicken manure.

through 12 (Fig. 2 and 3). This indicates that the organic N fraction in the natural organic materials is composed mainly of unstable forms that are readily mineralizable and stable forms that are more resistant to mineralization. The mineralization rate in the rapid and slow phases varied among N sources. Although mineralization occurred most rapidly in the first week, mineralization rate was slower with AP than with BM and CM (Table 3 and 4). During the slow mineralization phase, AP and BM showed a steady N release until the end of incubation, whereas CM showed a relatively slow N release with a plateaued phase in Weeks 8 through 12 (Table 3 and 4). Net N released from AP, BM, and CM averaged 43.7, 57.3, and 40.7% in the soil moisture study, respectively (Table 3), and 46.8, 58.9, and 42.7% in the temperature study, respectively (Table 4).

Differences in N release pattern among AP, BM, and CM could be explained by their chemical composition. First, C/N ratio has been identified as a good indicator of N availability among various organic N sources (Mtambanengwe and Kirchmann, 1995; Aulakh et al., 2000; Trinsoutrot et al., 2000; Rowell et al., 2001). The high N supplying capacity of BM was indicated by its narrow C/N ratio (3.27) compared with AP (11.13) and

CM (7.22) (Table 2). Second, it is suggested that N in lignin fraction is resistant to mineralization as lignin/N ratio is negatively correlated with mineralization rate (Melillo et al., 1982; Constantinides and Fownes, 1994; Kumar and Goh, 2003). The lignin/N ratio was higher with AP (2.63) than with CM (0.62), whereas BM, being an animal tissue, does not contain lignin (Table 2). This explains the slow N release from AP in the initial phase of incubation. On the other hand, the majority of N in BM is present in protein (Ciavatta et al., 1997), which is apparently more readily mineralized. It is also recognized that chicken manures contain urea and uric acid that are readily mineralizable (Gordillo and Cabrera, 1997; Havlin et al., 1999; Qafoku et al., 2001). However, neither C/N ratio nor lignin/N ratio could explain the nearly same N supplying capacity of AP and CM. This was probably due to the stabilization of OM in CM by the composting process (Hsu and Lo, 1999; Eghball, 2000; Hartz et al., 2000).

In addition to chemical composition, particle size plays an important role in N mineralization as it affects the surface area of N source and contact with microorganisms. Since BM was originally powdered, its fine particle

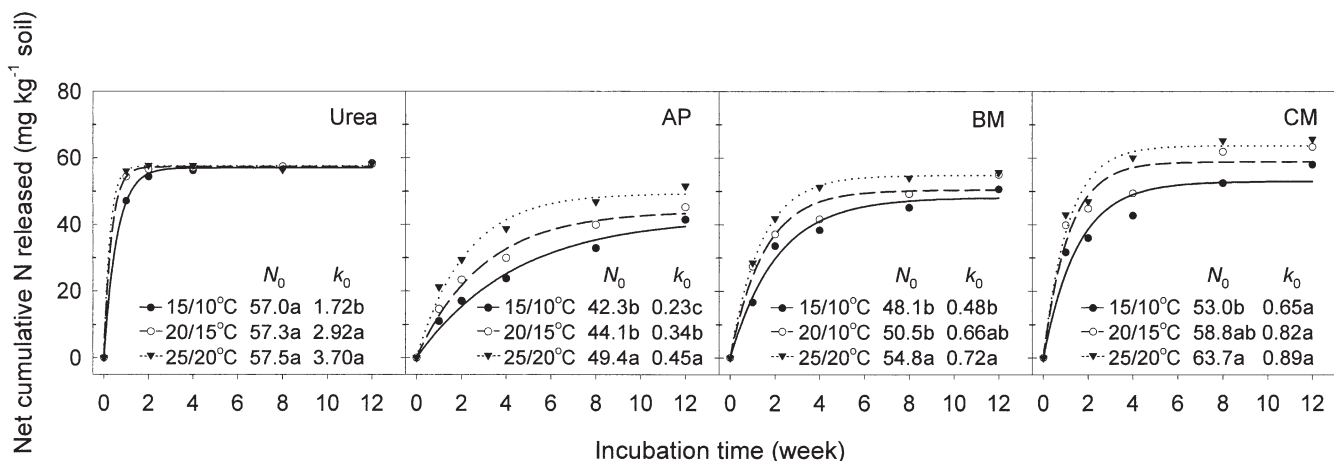


Fig. 3. Net cumulative N released from four organic N sources at different temperatures. The N_0 or k_0 values followed by the same letter are not significantly different at $\alpha = 0.05$. AP, alfalfa pellets; BM, blood meal; CM, partially composted chicken manure.

size may contribute to the more rapid N release from BM compared with AP and CM.

Soil Moisture Effects on Nitrogen Release

In the soil moisture study, net cumulative N released was significantly affected by N source, soil moisture, incubation time, and all interactions ($P < 0.05$, data not shown) except N source \times soil moisture interaction based on ANOVA.

In the control, as mineralization proceeded after Week 4, increasing soil moisture significantly enhanced mineralization (Table 5). Soil moisture regulates oxygen diffusion in soil and maximum aerobic microbial activity occurs between 50 and 70% WHC (Linn and Doran, 1984; Franzluebbers, 1999). In general, maximum mineralization of soil OM occurs in the same range, but some studies (Hopmans et al., 1980; Goncalves and Carlyle, 1994) have suggested that the range could be up to 100% WHC. The mineralization kinetics was best described by a linear function, and the mineralization rate according to a zero-order model was 0.567, 0.798, and 0.880 mg N kg⁻¹ wk⁻¹ at 50, 70, and 90% WHC, respectively (Fig. 1a). Although the difference was not statistically significant between 70 and 90% WHC, soil N mineralization was most enhanced at 90% WHC. Slow mineralization rate at 50% WHC can be explained by a decline in microbial activity resulting from limited diffusion of soluble substrates to microbes (Griffin, 1981; Schjønning et al., 2003) or reduced microbial mobility that limited access to substrates (Killham et al., 1993). The net amount of N mineralized was 6.9, 9.9, and 11.3 mg N kg⁻¹ at 50, 70, and 90% WHC, respectively (Table 5). These values demonstrate a high, although not rapid, contribution of soil OM to crop production. Taking this into account is important for making the appropriate N recommendation. Moreover, soil moisture is clearly a factor to consider when estimating N supplied from soil OM during the growing season.

Although urea hydrolysis was somewhat suppressed at low soil moisture in the first week, it was significantly more rapid (>80%) than mineralization of the natural organic materials regardless of soil moisture (Table 3). It is recognized that urea hydrolysis is enhanced with increasing soil moisture, but optimal soil moisture for maximum urease activity varies among previous studies. For example, Sahrawat (1984) found that urease activity increased as soil moisture increased from an air-dried state to field capacity in two semiarid soils (Alfisol and Vertisol). On the other hand, Dalal (1975) reported that maximum urea hydrolysis occurred at 50% WHC and further increases in soil moisture reduced urease activity in Trinidad soils. Results in this study agreed with those reported by Sahrawat (1984) to the extent that urea hydrolysis increased with increasing soil moisture to near field capacity (90% WHC). The inhibition of urea hydrolysis at low soil moisture may have been due to reduced urease activity or reduced diffusion of urea throughout the soil thus limiting urea-urease contact. During the 12-wk incubation, hydrolysis of residual urea was almost complete, and there was no significant difference in cumulative N released among soil moisture levels in Weeks 8 through 12 (Table 3). At the end of incubation, net N released ranged from 91.2 to 95.5% (Table 3). For the range tested in this study, soil moisture had a small influence on urea hydrolysis throughout incubation.

Unlike urea hydrolysis, mineralization of the natural organic materials exhibited apparent responses to soil moisture. Among these N sources, AP and CM showed similar patterns, whereas BM was quite different (Fig. 2). During the first 2 wk, mineralization of AP and CM was not enhanced with increasing soil moisture, but instead was reduced significantly at 90% WHC at Week 2 (Table 3). From Weeks 2 to 12, mineralization of AP and CM was enhanced in relation to soil moisture (Table 3). At the end of incubation, increasing soil mois-

Table 5. Net cumulative NH₄⁺- and NO₃⁻-N released from four organic N sources incubated in soil at different soil moistures.†

N source	Soil moisture % WHC	Incubation time (wk)									
		1		2		4		8		12	
		NH ₄ ⁺	NO ₃ ⁻	NH ₄ ⁺	NO ₃ ⁻	NH ₄ ⁺	NO ₃ ⁻	NH ₄ ⁺	NO ₃ ⁻	NH ₄ ⁺	NO ₃ ⁻
mg N kg ⁻¹ soil											
Control	50	0.7f	1.0i	0.6b	0.3i	0.1a	1.3h	0.7a	4.2i	0.0a	6.9i
	70	0.1f	0.9i	0.4b	0.4i	0.2ab	2.7g	0.5a	5.9h	0.0a	9.9h
	90	1.1ef	0.9i	0.4b	1.2h	0.4abc	2.4g	0.0b	6.3h	0.0a	11.3g
Urea	50	28.3a	21.2d	12.6a	41.0bc	-0.6de	52.6bc	-0.1bc	54.6c	-0.5d	57.2c
	70	22.9b	28.0bc	-0.4b	53.6a	-0.7def	55.6ab	-0.1bc	55.8bc	-0.5d	57.4c
	90	21.9b	36.1a	-0.5b	53.0a	-0.9ef	58.1a	-0.5bcde	55.6bc	-0.5d	59.8bc
AP	50	1.6ef	13.5fgh	-0.4b	25.6f	-0.4bcde	30.4f	-0.6cde	34.9g	-1.0e	41.6f
	70	1.7ef	14.0fg	-1.2b	23.3fg	-1.2f	30.2f	-0.7de	40.2f	-1.0e	44.4ef
	90	1.7ef	12.1gh	-0.9b	21.3g	-0.8ef	31.4f	-1.0e	41.1f	-1.0e	46.3e
BM	50	9.9d	11.0h	0.0b	29.6e	-0.4bcde	38.4e	-0.5bcde	46.1e	-0.2c	52.1d
	70	11.2cd	14.5fg	0.0b	35.7d	-0.5cde	39.0de	0.0b	46.5e	-0.1b	52.0d
	90	12.9c	15.2ef	-0.3b	35.3d	-0.2abcd	41.6d	-0.2bcd	48.2de	-0.2c	51.7d
CM	50	9.7d	22.7cde	-4.8c	52.6a	-5.5g	49.4bc	-5.2f	55.7bcd	-5.6f	59.0bcd
	70	5.3e	32.6abc	-4.4c	49.1ab	-5.2g	46.4cd	-5.5f	64.4a	-5.6f	65.7ab
	90	2.7ef	35.3ab	-5.4c	41.8bcd	-5.7g	59.8ab	-5.6f	63.1ab	-5.6f	70.1a

† Means in a column followed by the same letter are not significantly different according to *t* test at $\alpha = 0.05$. Initial NH₄⁺- and NO₃⁻-N contents in the treated soils (mg N kg⁻¹ soil) were: control 0.0 NH₄⁺-N, 22.0 NO₃⁻-N; urea 0.5 NH₄⁺-N, 22.0 NO₃⁻-N; AP 1.0 NH₄⁺-N, 22.1 NO₃⁻-N; BM 0.3 NH₄⁺-N, 22.9 NO₃⁻-N; CM 5.6 NH₄⁺-N, 22.3 NO₃⁻-N.

ture from 50 to 90% WHC significantly increased net N mineralized from AP and CM by 12 and 21% (4.7 and 11.1 mg kg⁻¹), respectively (Table 5). The occurrence of both reduced and enhanced mineralization at high soil moisture has been reported in several other studies. Doel et al. (1990) incubated soil amended with white lupin at -0.30, -0.03, and -0.01 MPa for 198 d. Although initial immobilization was least at -0.30 MPa, greater net mineralization occurred at -0.03 and -0.01 MPa than at -0.30 MPa. This was likely due to microbial activity being inhibited at low soil moisture throughout incubation. De Neve and Hofman (2002) conducted a soil incubation study with fresh carrot leaves at soil moistures ranging from 18 to 60% WHC. The mineralization rates at 45 to 60% WHC were relatively low in Days 0 through 16 but were high in Days 63 through 98 compared with those at 18 to 36% WHC. Since the added residues had a high water content (87% of fresh matter), the authors concluded that rewetting dry soil to 45 to 60% WHC provided excessive water in the vicinity of the residues and inhibited microbial activity during the initial phase of incubation. As soil moisture was distributed away from the residues, mineralization was enhanced at high soil moisture. However, those explanations do not apply to this study, which did not show immobilization and used moist soil and dried N sources. The explanation for our results may be that microbial communities responsible for the initial mineralization of readily mineralizable N and for the following mineralization of resistant N are different and are affected differently by soil moisture.

The negative effect of soil moisture on mineralization was not observed for BM. In the first week, the cumulative amount of N mineralized from BM showed an increase of 7.2 mg kg⁻¹ at 90% WHC compared with 50% WHC (Table 5), which was the greatest increase throughout incubation. The positive effect of soil moisture on mineralization became gradually less noticeable with incubation time, and there was no significant difference in cumulative N released among soil moisture levels after Week 8 (Table 3). This suggests that stable N in BM, which was mineralized in the late phase of incubation, was not affected by soil moisture. At the end of incubation, net N released for BM ranged from 56.9 to 57.4% (Table 3), demonstrating a significantly higher N supplying capacity compared with AP and CM, regardless of soil moisture. Since the percentage of N mineralized for AP and BM was less than estimated from the preliminary study, more N was mineralized from CM than from the other materials (Table 5).

Figure 2 illustrates the N release kinetics of the organic N sources using a first-order model. The rate constant (k_0) and the size of mineralizable N pool (N_0) reflected the effects of soil moisture on N release discussed above. Although the differences were not statistically significant for CM, the trend was for a decrease in k_0 with increasing soil moisture with AP and CM, whereas k_0 of BM increased in relation to soil moisture (Fig. 2). The trends indicate that increasing soil moisture slowed mineralization of AP and CM but accelerated mineralization of BM. The k_0 value for urea could not

be calculated at 90% WHC because the results did not fit a first-order model. Since this model was originally proposed for soil N mineralization (Stanford and Smith, 1972), the lack of fit for rapid urea hydrolysis may not be surprising. The N_0 values for urea and BM were constant across soil moisture levels, whereas those for AP and CM showed a trend for an increase in relation to soil moisture (Fig. 2). Water stress tends to reduce microbial diversity, favoring the microbes best adapted to coping with the stress (Atlas, 1984; Botter, 1985; Schimel et al., 1999). Thus, the increases in N_0 may be related to changes in composition of microbial community, such that the microbial communities favored at high soil moisture have ability to metabolize substrates that are not utilized at lower soil moistures.

The N sources used in this study demonstrated contrasting results in their N release patterns at different soil moistures. First, urea showed the least apparent responses to soil moisture of all N sources used in this study. This was likely due to the rapid hydrolysis of urea that diminished the effects of soil moisture before the first measurement was made. Second, mineralization of AP and CM was enhanced by high soil moisture during the late phase of incubation, whereas that of BM was enhanced during the initial phase of incubation. This clearly indicates that soil moisture effects on N mineralization vary with chemical composition of N sources. Incubation time is important for determination of soil moisture effects on N availability from the organic N sources.

Soil Moisture Effects on Nitrification

Nitrification, as measured by NO₃⁻-N production, occurred successively in all treatments (Table 5). Nitrification was significantly affected by N source, soil moisture, and incubation time based on ANOVA ($P < 0.05$, data not shown).

The NH₄⁺-N and NO₃⁻-N contents in the control reflected the slow mineralization of soil OM and subsequent nitrification. Whereas the NH₄⁺-N content remained very low (<2.0 mg kg⁻¹) throughout incubation, the NO₃⁻-N content increased slowly with incubation time (Table 5). Increasing soil moisture significantly increased NO₃⁻-N production in Weeks 4 through 12 (Table 5). It is apparent that the limited NH₄⁺ supply reduced NO₃⁻-N production at low soil moisture.

Addition of the organic N sources resulted in significant increases in both NH₄⁺-N and NO₃⁻-N contents in the first week (Table 5). It has been reported that active nitrification of added NH₄⁺ starts with a time lag of 4 to 10 d following rewetting of dry soil in several incubation studies (MacLean and McRae, 1987; Mulvaney et al., 1997; Williams et al., 1998). The occurrence of the lag phase of NO₃⁻ accumulation was not apparent in this study. The soil was stored under field moisture condition (10% w/w) at room temperature (20–23°C) until used, thereby likely maintaining a high population of nitrifying bacteria. The NH₄⁺-N content in the first week was closely associated with the amount of N mineralized. For example, the urea-treated soil showed a sig-

nificantly greater NH_4^+ -N content than the soils treated with the natural organic materials at all soil moisture levels, due to the rapid hydrolysis of urea (Table 5). Conversely, the AP-treated soil, with an initially slow mineralization rate, showed the lowest NH_4^+ -N content, with more than 90% of inorganic N recovered in NO_3^- form (Table 5). Apparently the NH_4^+ was nitrified to NO_3^- as quickly as it was mineralized. Similarly, the NO_3^- -N content in the first week was significantly higher in the urea- and CM-treated soils, which mineralized a greater amount of N than AP and BM (Table 5). Malhi and McGill (1982) reported that increasing NH_4^+ supply up to 200 mg kg^{-1} enhanced nitrification. The NO_3^- -N production in the first week was positively correlated ($R^2 = 0.72$, $P < 0.001$, data not shown) with NH_4^+ supply (initial NH_4^+ + mineralized N). Hence, high NO_3^- -N production from urea and CM can be explained in part by high rate of NH_4^+ formation.

Increasing soil moisture from 50 to 90% WHC significantly increased the NO_3^- -N contents in the urea-, BM-, and CM-treated soils in the first week. Low soil moisture inhibits activity of nitrifying bacteria by reducing substrate (NH_4^+) diffusion and intracellular water potential. Stark and Firestone (1995) reported that diffusional limitation of substrate is the major limiting factor at greater than -0.6 MPa, but adverse physiological effects associated with cell dehydration are more inhibiting nitrification at less than -0.6 MPa. Since 50% WHC is considered to be greater than -0.6 MPa for the soil used in this study, the reduced nitrification was mainly attributed to the diffusional limitation of substrate. In addition, in the urea- and BM-treated soils increasing soil moisture enhanced NH_4^+ formation, partly accounting for the increased NO_3^- -N production at higher soil moistures (Malhi and McGill, 1982). However, NO_3^- -N production in the AP-treated soil was not related to soil moisture (Table 5). This was likely due to the slow mineralization of AP regardless of soil moisture.

At Week 2, continued nitrification in the N-treated soils was indicated by both NH_4^+ -N disappearance and subsequent NO_3^- -N production. In the urea-treated soil, although the NH_4^+ -N contents at 70 and 90% WHC were <1.0 mg kg^{-1} , a significantly large amount of N (12.6 mg kg^{-1}) still remained as NH_4^+ at 50% WHC (Table 5). However, the cumulative amount of N released (NH_4^+ + NO_3^-) in the urea-treated soil did not differ among soil moisture levels (Table 4), suggesting that nitrification was more likely to be inhibited than urea hydrolysis at low soil moisture. The NH_4^+ -N contents in the soils treated with the natural organic materials were very low (<1.0 mg kg^{-1}) at all soil moisture levels (Table 5). It was particularly notable that, although urea and CM released almost equal amounts of N, nitrification in the CM-treated soil was almost complete even at 50% WHC (Table 5). This suggests that activity of nitrifying bacteria was stimulated in the CM-treated soil. In addition to soil moisture, soil pH also influences nitrification. Nitrification occurs over a wide range in pH (4.5–10), but the optimum pH is 8.5 and lowering pH decreases nitrification rate (Montagnini et al., 1989; Paavolainen and Smolander, 1997; Ste-

Marie and Paré, 1999; Havlin et al., 1999). Addition of urea and CM can be expected to cause different changes in soil pH. First, although urea application raises soil pH temporarily, it ultimately lowers soil pH below the original value (Martikainen, 1985; Mulvaney et al., 1997). This is caused by the hydrolysis of urea, which neutralizes H^+ when releasing 2 NH_4^+ , but subsequent nitrification of NH_4^+ produces 2 H^+ (Havlin et al., 1999). In contrast to urea, chicken manures are effective in raising soil pH because the manures generally contain high calcium carbonate (Hue, 1992; Kingery et al., 1993; Mokolobate and Haynes, 2002). The pH and Ca content were highest in CM (Table 2), indicating its high ability to neutralize soil acidity. Therefore, it could be expected that addition of CM raised soil pH, thereby stimulating the activity of nitrifying bacteria.

After Week 4, the NH_4^+ -N content was very low (<1.0 mg kg^{-1}), whereas the NO_3^- -N content increased slowly in all treatments (Table 5). It seems that newly released NH_4^+ from the organic N sources was quickly nitrified after their N release rates slowed down. The differences in NO_3^- -N production among treatments in Weeks 4 through 12 were attributed to the differences in NH_4^+ released from the organic N sources.

Temperature Effects on Nitrogen Release

In the temperature study, net cumulative N released was significantly affected by N source, temperature, incubation time, and all interactions based on ANOVA ($P < 0.05$, data not shown). Increasing temperature enhanced N release from all N sources used in this study, but the magnitude of the response to temperature varied among source of N and incubation time.

In the control, mineralization of soil OM was enhanced with increasing temperature throughout incubation (Fig. 1b). The temperature coefficient, Q_{10} , of approximately 2 over the range 5 to 35°C, is generally accepted to describe the relationship between soil N mineralization and temperature (Campbell and Biederbeck, 1972; Stanford et al., 1973; Sierra, 1997; Kätterer et al., 1998). That is, a two-fold increase in mineralization rate is associated with a shift of 10°C. In this study, the mineralization kinetics was best described by a linear function, and the mineralization rate according to a zero-order model was 0.54, 0.95, and 1.53 mg N kg^{-1} wk^{-1} at 15/10, 20/15, and 25/20°C, respectively (Fig. 1b). A 10°C increase resulted in a threefold increase in the mineralization rate. The net amount of N mineralized after 12 wk was 6.4, 11.3, and 18.9 mg N kg^{-1} at 15/10, 20/15, and 25/20°C, respectively (Table 6), demonstrating significant increases in the pool size of mineralizable N. Increases in temperature induce a shift in the composition of microbial communities (Richards et al., 1985; Carreiro and Koske, 1992). Zogg et al. (1997) found that the shift in microbial community composition paralleled an increase in microbial respiration at temperatures between 5 and 25°C. Thus, the increase in the net mineralized N by high temperature was likely due to microbial communities favored at high temperature metabolizing substrates that were not utilized at lower temperatures.

Table 6. Net cumulative NH_4^+ - and NO_3^- -N released from four organic N sources incubated in soil at different temperatures.†

N source	Temperature °C (14/10 h)	Incubation time (wk)									
		1		2		4		8		12	
		NH_4^+	NO_3^-	NH_4^+	NO_3^-	NH_4^+	NO_3^-	NH_4^+	NO_3^-	NH_4^+	NO_3^-
		mg N kg ⁻¹ soil									
Control	15/10	0.4g	0.8f	0.3c	1.0i	0.2a	2.0h	0.4a	3.8h	0.1a	6.3j
	20/15	0.0g	1.5f	0.2c	1.9i	0.1a	3.9g	0.4a	7.2g	0.2a	11.1i
	25/20	0.2g	1.5f	0.1c	3.2i	0.1a	5.8g	0.2a	11.4f	0.2a	18.7h
Urea	15/10	33.8a	13.3d	7.8a	46.5c	-0.6a	56.7b	0.0a	56.9b	-0.4a	58.8cd
	20/15	20.6b	33.7b	-0.3c	56.7a	-0.4a	57.4b	-0.1a	57.5b	-0.2a	58.3cd
	25/20	6.4de	49.7a	-0.2c	58.0a	-0.2a	57.9b	-0.2a	56.6b	-0.5a	58.8cd
AP	15/10	2.8fg	8.1ef	-0.7c	17.8h	-1.1b	24.9f	-0.8b	33.7f	-1.1a	42.6g
	20/15	0.9g	13.7d	-1.1c	24.4g	-1.1b	31.0e	-0.8b	40.7e	-0.7b	45.9g
	25/20	0.4g	20.7c	-1.0c	30.5f	-0.9b	39.6d	-0.9b	47.8cd	-1.1b	52.7ef
BM	15/10	9.4cd	7.2f	5.1b	28.5f	-0.1a	38.4d	0.0a	45.1d	0.1a	50.5f
	20/15	10.8c	16.5d	0.1c	36.9e	-0.2a	41.8d	-0.2a	49.4c	-0.2a	55.1de
	25/20	3.9efg	24.6c	-0.1c	41.9d	0.0a	51.3c	0.0a	54.1b	-0.1a	55.7de
CM	15/10	18.0b	13.6de	-4.9d	40.8de	-6.4c	49.1c	-5.7c	58.2b	-6.0c	64.0bc
	20/15	6.1def	33.7b	-5.4d	50.2bc	-6.1c	55.5bc	-5.9c	67.8a	-6.4c	69.7ab
	25/20	-3.8h	46.8a	-6.1d	53.1ab	-6.3c	66.4a	-5.8c	71.1a	-6.1c	71.8ab

† Means in a column followed by the same letter are not significantly different according to *t* test at $\alpha = 0.05$. Initial NH_4^+ - and NO_3^- -N contents in the treated soils (mg N kg⁻¹ soil) were: control 0.2 NH_4^+ -N, 19.9 NO_3^- -N; urea 0.6 NH_4^+ -N, 19.9 NO_3^- -N; AP 1.3 NH_4^+ -N, 20.3 NO_3^- -N; BM 0.4 NH_4^+ -N, 20.4 NO_3^- -N; CM 6.6 NH_4^+ -N, 20.6 NO_3^- -N.

Temperature is clearly a factor to consider when estimating N supplied from soil OM during the growing season.

Although urea hydrolysis was somewhat suppressed at 15/10°C in the first 2 wk, it was significantly more rapid (>75%) than mineralization of the natural organic materials regardless of temperature (Table 4). Hydrolysis of residual urea was almost complete by the end of Week 4. Inhibited urease activity (Overrein and Moe, 1967; Sahrawat, 1984; Moyo et al., 1989; Lai and Tabatabai, 1992) and slow urea diffusion (Pang et al., 1977; Sadeghi et al., 1988) seems to retard the hydrolysis of urea at 15/10°C. In Weeks 4–12, there was no significant difference in cumulative N released among temperature levels. At the end of incubation, net N released ranged from 92.6 to 93.2% (Table 4). For the range tested in this study, temperature had a small influence on urea hydrolysis throughout incubation. The results were consistent with those reported by MacLean and McRae (1987). In their soil incubation study, the rate of urea hydrolysis was proportional to temperature over the range of 9 to 18°C in the first 3 d but showed no difference among temperature levels after 5 d due to rapid hydrolysis of urea.

Mineralization of the natural organic materials was significantly enhanced with increasing temperature throughout the incubation period. Maximum differences in the cumulative amount of mineralized N between temperatures 15/10 and 25/20°C were observed during the first 4 wk. At Week 4, the differences were 14.9, 13.0, and 17.4 mg kg⁻¹ for AP, BM, and CM, respectively (Table 6). From Week 4 to 12, AP showed a relatively constant increase in cumulative N released with increasing temperature, but BM and CM showed a limited response (Table 4). Mineralization of BM and CM at 25/20°C slowed down after Week 4, with less than 5% of organic N mineralized in Weeks 4 through 12 (Table 4). At the end of incubation, increasing temperature from 15/10 to 25/20°C significantly increased net N min-

eralized from AP, BM, and CM by 25, 10, and 13% (10.1, 5.0, and 7.7 mg kg⁻¹), respectively (Table 6). This suggests that mineralization of stable N in AP was more influenced by temperature than BM and CM. In fact, net N released was significantly higher with AP than with CM at 25/20°C, but there was no significant difference at lower temperatures (Table 4). As in the soil moisture study, since the percentage N mineralized for AP and BM was less than estimated from the preliminary study, more N was mineralized from CM than from the other materials (Table 6).

Figure 3 illustrates the N release kinetics of the organic N sources using a first-order model. The k_0 and N_0 values reflected the effects of temperature on N release discussed above. Although the differences were not statistically significant for CM, the trend was for an increase in k_0 with increasing temperature. Except for urea, N_0 increased in relation to temperature. Increases in k_0 may be explained by stimulated microbial activity or accelerated diffusion at high temperature (Nicolardot et al., 1994; MacDonald et al., 1995; Zak et al., 1999). As discussed earlier for soil N mineralization, increases in N_0 may be related to a shift in the composition of microbial communities, such that the microbial communities favored at high temperature have the ability to metabolize substrates that are not utilized at lower temperatures (Zogg et al., 1997).

The N sources used in this study demonstrated contrasting results in their N release patterns at different temperatures. First, increasing temperature increased net N released from the natural organic materials, but did not affect net N released from urea. Considering temperature is important for making appropriate N recommendations when using the natural organic materials. For example, more N may have to be applied in a cool climatic condition because less N can be expected to be released. Second, mineralization of AP responded to temperature to a greater degree than that of BM and CM. Similar results have been reported in previous

studies. Cookson et al. (2002) reported that 22, 33, 41, and 60% of the total N in clover residues was mineralized at 2, 5, 10, and 15°C, respectively, after 160-d incubation. Griffin and Honeycutt (2000) incubated soil amended with dairy, poultry, and swine manures for 112 d, and found that increasing temperature from 10 to 24°C accelerated the mineralization rate, but did not affect the net mineralized N after 28 d. Although these results were not comparable due to different experimental conditions, our results clearly indicate that temperature effects on N mineralization vary with chemical composition of N sources.

Temperature Effects on Nitrification

Nitrification, as measured by NO_3^- -N production, occurred successively in all treatments (Table 6). Nitrification was significantly affected by N source, temperature, and incubation time based on ANOVA ($P < 0.05$, data not shown).

The NH_4^+ -N and NO_3^- -N contents in the control reflected the slow mineralization of soil OM and subsequent nitrification. Whereas the NH_4^+ -N content remained very low ($<1.0 \text{ mg kg}^{-1}$) throughout incubation, the NO_3^- -N content increased slowly with incubation time (Table 6). The NO_3^- -N production was proportional to temperature throughout incubation (Table 6). It is apparent that the limited NH_4^+ supply reduced NO_3^- -N production at low temperature.

Considerable nitrification occurred in all N-treated soils throughout incubation as a significantly higher NO_3^- -N content was recovered compared with the control (Table 6). The urea-treated soil showed the highest NH_4^+ -N content at all temperature levels due to rapid hydrolysis of urea (Table 6). Conversely, the AP-treated soil, with an initially slow mineralization rate, showed very low NH_4^+ -N contents ranging from 0.4 to 2.8 mg kg^{-1} (Table 6). Apparently NH_4^+ was nitrified as quickly as it was mineralized. The NO_3^- -N content was significantly higher in the urea and CM-treated soils, which mineralized greater amounts of N than AP and BM (Table 6). Increasing temperature stimulates the activity of nitrifying bacteria with maximum nitrification occurring between 25 and 35°C (Justice and Smith, 1962; Kowalenko and Cameron, 1976; Malhi and McGill, 1981; Breuer et al., 2002). The NH_4^+ -N content was inversely and the NO_3^- -N content was directly correlated with temperature (Table 6), indicating that activity of nitrifying bacteria was stimulated at higher temperatures. In addition, enhanced nitrification at high temperature can be explained in part by a high rate of NH_4^+ formation (Malhi and McGill, 1982).

At Week 2, continued nitrification in the N-treated soils was indicated by both NH_4^+ -N disappearance and subsequent NO_3^- -N production (Table 6). In the urea-treated soil, although the NH_4^+ -N contents at 20/15 and 25/20°C were nearly zero, a significantly large amount of N (7.8 mg kg^{-1}) still remained as NH_4^+ at 15/10°C (Table 6). However, the difference in the cumulative amount of N released ($\text{NH}_4^+ + \text{NO}_3^-$) between temperatures 15/10 and 25/20°C was relatively small (Table 6),

suggesting that nitrification was more likely to be limited than urea hydrolysis at low temperature. Among the soils treated with the natural organic materials, the BM-treated soil showed a significantly higher NH_4^+ -N content at 15/10°C (5.1 mg kg^{-1}), whereas the AP- and CM-treated soils showed very low NH_4^+ -N contents at all temperature levels (Table 6). All formed NH_4^+ -N was completely nitrified in the AP- and CM-treated soils.

After Week 4, the NH_4^+ -N content was very low ($<1.0 \text{ mg kg}^{-1}$), whereas the NO_3^- -N content increased slowly in all treatments (Table 6). It seems that newly released NH_4^+ from the organic N sources was quickly nitrified after their N release rates slowed down. Differences in NO_3^- -N production among treatments in Weeks 4 through 12 were attributed to the differences in NH_4^+ released from the organic N sources.

CONCLUSIONS

Four organic N sources used in this study had different characteristics in chemical composition and varied in N release pattern and N supplying capacity. Soil moisture and temperature influenced N availability from the organic N sources differently depending on source of N and time. These interactions must be considered to determine an appropriate rate and timing of fertilization for efficient use of N inputs, especially in greenhouse production systems when controlling irrigation or temperature.

Testing both NH_4^+ - and NO_3^- -N is necessary to estimate N availability from the organic N sources in an initial period of growing season. At high soil moisture or temperature conditions, a high concentration of NO_3^- -N accumulated in the soil immediately after urea or CM application may be a concern for nitrate leaching.

It appears that differences in N release response to soil moisture or temperature among N sources and time are related to chemical composition of the organic N sources applied. Further research on which chemical compositions are more or less responsive to the effects of soil moisture and temperature is needed to better understand availability of N from organic N sources.

ACKNOWLEDGMENTS

The authors express their appreciation to Gary Zehr, John Dahl, and Vicki Smith for assistance with sample collection and analysis. Appreciation is extended to Dr. John Biernbaum, Dr. Sieglinde Snapp, and Jeanette Makries for reviewing the manuscript and providing useful comments. The financial support from the USDA, sustainable agriculture special research grant, is gratefully acknowledged.

REFERENCES

- Ajwa, H.A., C.W. Rice, and D. Sotomayor. 1998. Carbon and nitrogen mineralization in tallgrass prairie and agricultural soil profiles. *Soil Sci. Soc. Am. J.* 62:942-951.
- Atlas, R.M. 1984. Use of microbial diversity measurements to assess environmental stress. p. 540-545. *In* M.J. Klug and C.A. Reddy (ed.) *Current perspectives in microbial ecology*. Amer. Soc. Microb., Washington, DC.
- Aulakh, M.S., T.S. Khera, and J.W. Doran. 2000. Mineralization and denitrification in upland, nearly saturated and flooded subtropical

- soil. II. Effect of organic manures varying in N content and C:N ratio. *Biol. Fertil. Soils* 31:168–174.
- Botter, P. 1985. Response of microbial biomass to alternate moist and dry conditions in a soil incubated with ¹⁴C- and ¹⁵N-labeled plant material. *Soil Biol. Biochem.* 17:329–337.
- Bremner, J.M., and C.S. Mulvaney. 1982. Nitrogen—Total. p. 595–624. *In* A.L. Page et al. (ed.) *Methods of soil analysis*. Part 2. 2nd ed. Agron. Monogr. 9. ASA and SSSA, Madison, WI.
- Breuer, L., R. Kiese, and K. Butterbach-Bahl. 2002. Temperature and moisture effects on nitrification rates in tropical rain-forest soils. *Soil Sci. Soc. Am. J.* 66:834–844.
- Campbell, C.A., and V.O. Biederbeck. 1972. Influence of fluctuating temperatures and constant soil moistures on nitrogen changes in amended and unamended loam. *Can. J. Soil Sci.* 52:323–336.
- Carreiro, M.M., and R.E. Koske. 1992. Room-temperature isolations can bias against selection of low-temperature micofungi in temperate forest soils. *Mycologia* 84:886–900.
- Chae, Y.M., and M.A. Tabatabai. 1986. Mineralization of nitrogen in soils amended with organic wastes. *J. Environ. Qual.* 15:193–198.
- Ciavatta, C., M. Govi, L. Sitti, and C. Gessa. 1997. Influence of blood meal organic fertilizer on soil organic matter: A laboratory study. *J. Plant Nutr.* 20:1573–1591.
- Combs, S.M., and M.V. Nathan. 1998. Soil organic matter. p. 53–58. *In* J.R. Brown (ed.) *Recommended chemical soil test procedures for the north central region*. North Central Regional Research Publication No. 221 (revised). Missouri Agri. Exp. Stn., Columbia, MO.
- Constantinides, M., and J.H. Fownes. 1994. Nitrogen mineralization from leaves and litter of tropical plants: Relationship to nitrogen, lignin and soluble polyphenol concentrations. *Soil Biol. Biochem.* 26:49–55.
- Cookson, W.R., I.S. Cornforth, and J.S. Rowarth. 2002. Winter soil temperature (2–15°C) effects on nitrogen transformations in clover green manure amended or unamended soils; a laboratory and field study. *Soil Biol. Biochem.* 34:1401–1415.
- Csonka, L.N. 1989. Physiological and genetic responses of bacteria to osmotic stress. *Microbiol. Rev.* 52:121–147.
- Dalal, R.C. 1975. Urease activity in some Trinidad soils. *Soil Biol. Biochem.* 7:5–8.
- De Neve, S., and G. Hofman. 2002. Quantifying soil water effects on nitrogen mineralization from soil organic matter and from fresh crop residues. *Biol. Fertil. Soils* 35:379–386.
- Doel, D.S., C.W. Honeycutt, and W.A. Halteman. 1990. Soil water effects on the use of heat units to predict crop residue carbon and nitrogen mineralization. *Biol. Fertil. Soils* 10:102–106.
- Eghball, B. 2000. Nitrogen mineralization from field-applied beef cattle feedlot manure or compost. *Soil Sci. Soc. Am. J.* 64:2024–2030.
- Fox, R.H., R.J.K. Myers, and I. Vallis. 1990. The nitrogen mineralization rate of legume residues in soil as influenced by their polyphenol, lignin and nitrogen contents. *Plant Soil* 129:251–259.
- Frank, K., D. Beegle, and J. Denning. 1998. Phosphorus. p. 21–26. *In* J.R. Brown (ed.) *Recommended chemical soil test procedures for the north central region*. North Central Regional Research Publication No. 221 (revised). Missouri Agri. Exp. Stn., Columbia, MO.
- Franzuebbers, A.J. 1999. Microbial activity in response to water-filled pore space of variably eroded southern Piedmont soils. *Appl. Soil Ecol.* 11:91–101.
- Gee, G.W., and J.W. Bauder. 1986. Particle-size analysis. p. 383–411. *In* A. Klute (ed.) *Methods of soil analysis*. Part 1. 2nd ed. Agron. Monogr. 9. ASA and SSSA, Madison, WI.
- Goering, H.K., and P.J. Van Soest. 1970. Forage fiber analyses (apparatus, reagents, procedures, and some applications). USDA Agric. Handbook No. 379. U.S. Gov. Print. Office, Washington, DC.
- Goncalves, J.L.M., and J.C. Carlyle. 1994. Modelling the influence of moisture and temperature on net nitrogen mineralization in a forested sandy soil. *Soil Biol. Biochem.* 26:1557–1564.
- Gordillo, R.M., and M.L. Cabrera. 1997. Mineralizable nitrogen in broiler litter: I. Effect of selected litter chemical characteristics. *J. Environ. Qual.* 26:1672–1679.
- Griffin, D.M. 1981. Water potential as a selective factor in the microbial ecology of soils. p. 141–151. *In* L.F. Elliot et al. (ed.) *Water potential relations in soil microbiology*. SSSA Spec. Publ. 9. SSSA, Madison, WI.
- Griffin, T.S., and C.W. Honeycutt. 2000. Using growing degree days to predict nitrogen availability from livestock manures. *Soil Sci. Soc. Am. J.* 64:1876–1882.
- Hartz, T.K., J.P. Mitchell, and C. Giannini. 2000. Nitrogen and carbon mineralization dynamics of manures and composts. *HortScience* 35:209–212.
- Havlin, J.L., J.D. Beaton, S.L. Tisdale, and W.L. Nelson. 1999. *Soil fertility and fertilizers: An introduction to nutrient management*. 6th ed. Prentice Hall, Upper Saddle River, NJ.
- Honeycutt, C.W. 1999. Nitrogen mineralization from soil organic matter and crop residues: Field validation of laboratory predictions. *Soil Sci. Soc. Am. J.* 63:134–141.
- Hopmans, P., D.W. Flinn, and P.W. Farrell. 1980. Nitrogen mineralisation in a sandy soil under native eucalypt forest and exotic pine plantations in relation to moisture content. *Commun. Soil Sci. Plant Anal.* 11:71–79.
- Hsu, J., and S. Lo. 1999. Chemical and spectroscopic analysis of organic matter transformation during composting of pig manure. *Environ. Pollut.* 104:189–196.
- Hue, N.V. 1992. Correcting soil acidity of a highly weathered Ultisol with chicken manure and sewage sludge. *Commun. Soil Sci. Plant Anal.* 23:241–264.
- Justice, J.K., and R.L. Smith. 1962. Nitrification of ammonium sulfate in a calcareous soil as influenced by combinations of moisture, temperature, and levels of added nitrogen. *Soil Sci. Soc. Am. Proc.* 26: 246–250.
- Kätterer, T., M. Reichstein, O. Andrén, and A. Lomander. 1998. Temperature dependence of organic matter decomposition: A critical review using literature data analysed with different models. *Biol. Fertil. Soils* 27:258–262.
- Killham, K., A. Amato, and J.N. Ladd. 1993. Effect of substrate location in soil and soil pore-water regime on carbon turnover. *Soil Biol. Biochem.* 25:57–62.
- Kingery, W.L., C.W. Wood, D.P. Delaney, J.C. Williams, G.L. Mullins, and E. van Santen. 1993. Implications of long-term land applications of poultry litter on tall fescue pastures. *J. Prod. Agric.* 6:390–395.
- Kowalenko, C.G., and D.R. Cameron. 1976. Nitrogen transformations in an incubated soil as affected by combinations of moisture content and temperature and adsorption-fixation of ammonium. *Can. J. Soil Sci.* 56:63–77.
- Kumar, K., and K.M. Goh. 2003. Nitrogen release from crop residues and organic amendments as affected by biochemical composition. *Commun. Soil Sci. Plant Anal.* 34:2441–2460.
- Lai, C.M., and M.A. Tabatabai. 1992. Kinetic parameters of immobilized urease. *Soil Biol. Biochem.* 24:225–228.
- Li, G.C., and R.L. Mahler. 1995. Effect of plant material parameters on nitrogen mineralization in a Mollisol. *Commun. Soil Sci. Plant Anal.* 26:1905–1919.
- Linn, D.M., and J.W. Doran. 1984. Effect of water-filled pore space on carbon dioxide and nitrous oxide production in tilled and nontilled soils. *Soil Sci. Soc. Am. J.* 48:1267–1272.
- MacDonald, N.W., D.R. Zak, and K.S. Pregitzer. 1995. Temperature effects on kinetics of microbial respiration and net nitrogen and sulfur mineralization. *Soil Sci. Soc. Am. J.* 59:233–240.
- MacLean, A.A., and K.B. McRae. 1987. Rate of hydrolysis and nitrification of urea and implications of its use in potato production. *Can. J. Soil Sci.* 67:679–686.
- Malhi, S.S., and W.B. McGill. 1982. Nitrification in three Alberta soils: Effect of temperature, moisture and substrate concentration. *Soil Biol. Biochem.* 14:393–399.
- Martikainen, P.J. 1985. Nitrification in forest soil of different pH as affected by urea, ammonium sulphate and potassium sulphate. *Soil Biol. Biochem.* 17:363–367.
- Melillo, J.M., J.D. Aber, and J.F. Muratore. 1982. Nitrogen and lignin control of hardwood leaf litter decomposition dynamics. *Ecology* 63:621–626.
- Mokolobate, M.S., and R.J. Haynes. 2002. Comparative liming effect of four organic residues applied to an acid soil. *Biol. Fertil. Soils* 35:79–85.
- Montagnini, F., B. Haines, and W. Swank. 1989. Factors controlling nitrification in soils of early successional and oak/hickory forests in the southern Appalachians. *For. Ecol. Manage.* 26:77–94.

- Moyo, C.C., D.E. Kissel, and M.L. Cabrera. 1989. Temperature effects on soil urease activity. *Soil Biol. Biochem.* 21:935-938.
- Mtambanengwe, F., and H. Kirchmann. 1995. Litter from a tropical savanna woodland (Mimbo): Chemical composition and C and N mineralization. *Soil Biol. Biochem.* 27:1639-1651.
- Mulvaney, R.L., S.A. Khan, and C.S. Mulvaney. 1997. Nitrogen fertilizers promote denitrification. *Biol. Fertil. Soils* 24:211-220.
- Nicolardot, B., G. Fauvet, and D. Cheneby. 1994. Carbon and nitrogen cycling through soil microbial biomass at various temperatures. *Soil Biol. Biochem.* 26:253-261.
- Overrein, L.N., and P.G. Moe. 1967. Factors affecting urea hydrolysis and ammonia volatilization in soil. *Soil Sci. Am. Proc.* 31: 57-61.
- Paavolainen, L., and A. Smolander. 1997. Nitrification and denitrification in soil from a clear-cut Norway spruce (*Picea abies*) stand. *Soil Biol. Biochem.* 30:775-781.
- Palm, C.A., and P.A. Sanchez. 1991. Nitrogen release from the leaves of some tropical legumes as affected by their lignin and polyphenolic contents. *Soil Biol. Biochem.* 23:83-88.
- Pang, P.C.K., C.M. Cho, and R.A. Hedlin. 1977. Distribution and transformation of band-applied urea in soil following incubation under isothermal and temperature gradient conditions. *Can. J. Soil Sci.* 57:409-416.
- Pramer, D., and R. Bartha. 1972. Preparation and processing of soil samples for biodegradation studies. *Environ. Lett.* 2:217-224.
- Qafoku, O.S., M.L. Cabrera, W.R. Windham, and N.S. Hill. 2001. Rapid methods to determine potentially mineralizable nitrogen in broiler litter. *J. Environ. Qual.* 30:217-221.
- Richards, B.N., J.E.N. Smith, G.J. White, and J.L. Charley. 1985. Mineralization of soil nitrogen in three forest communities from the New England region of New South Wales. *Aust. J. Ecol.* 10: 429-441.
- Rowell, D.M., C.E. Prescott, and C.M. Preston. 2001. Decomposition and nitrogen mineralization from biosolids and other organic materials: Relationship with initial chemistry. *J. Environ. Qual.* 30:1401-1410.
- Sadeghi, A.M., D.E. Kissel, and M.L. Cabrera. 1988. Temperature effects on urea diffusion coefficients and urea movement in soil. *Soil Sci. Soc. Am. J.* 52:46-49.
- Sadeghi, A.M., D.E. Kissel, and M.L. Cabrera. 1989. Estimating molecular diffusion coefficients of urea in unsaturated soil. *Soil Sci. Soc. Am. J.* 53:15-18.
- Sahrawat, K.L. 1984. Effects of temperature and moisture on urease activity in semi-arid tropical soils. *Plant Soil* 78:401-408.
- SAS Institute. 1990. SAS user's guide. Version 6 ed. SAS Inst., Cary, NC.
- Schimmel, J.P., J.M. Gullledge, J.S. Clein-Curley, J.E. Lindstrom, and J.F. Braddock. 1999. Moisture effects on microbial activity and community structure in decomposing birch litter in the Alaskan taiga. *Soil Biol. Biochem.* 31:831-838.
- Schjønning, P., I.K. Thomsen, P. Moldrup, and B.T. Christensen. 2003. Linking soil microbial activity to water- and air-phase contents and diffusivities. *Soil Sci. Soc. Am. J.* 67:156-165.
- Seneviratne, G., L.H.J. Van Holm, and S.A. Kulasoorya. 1998. Quality of different mulch materials and their decomposition and N release under low moisture regimes. *Biol. Fertil. Soils* 26:136-140.
- Sierra, J. 1997. Temperature and soil moisture dependence of N mineralization in intact soil cores. *Soil Biol. Biochem.* 29:1557-1563.
- Sims, J.T. 1986. Nitrogen transformations in a poultry manure amended soil: Temperature and moisture effects. *J. Environ. Qual.* 15:59-63.
- Stanford, G., M.H. Frere, and D.H. Schwaninger. 1973. Temperature coefficient of soil nitrogen mineralization. *Soil Sci.* 115:321-323.
- Stanford, G., and S.J. Smith. 1972. Nitrogen mineralization potentials of soils. *Soil Sci. Soc. Am. Proc.* 36: 465-472.
- Stark, J.M., and M.K. Firestone. 1995. Mechanisms for soil moisture effects on activity of nitrifying bacteria. *Appl. Environ. Microbiol.* 61: 218-221.
- Ste-Marie, C., and D. Paré. 1999. Soil, pH and N availability effects on net nitrification in the forest floors of a range of boreal forest stands. *Soil Biol. Biochem.* 31:1579-1589.
- Tomar, J.S., and R.J. Soper. 1981. An incubation study of nitrogen added as urea to several Manitoba soils with particular reference to retention of nitrogen. *Can. J. Soil Sci.* 61:1-10.
- Trinsoutrot, I., S. Recous, B. Bentz, M. Linères, D. Chèneby, and B. Nicolardot. 2000. Biochemical quality of crop residues and carbon and nitrogen mineralization kinetics under nonlimiting nitrogen conditions. *Soil Sci. Soc. Am. J.* 64:918-926.
- Vigil, M.F., and D.E. Kissel. 1995. Rate of nitrogen mineralization from incorporated crop residues as influenced by temperature. *Soil Sci. Soc. Am. J.* 59:1636-1644.
- Vlek, P.L.G., and M.F. Carter. 1983. The effect of soil environment and fertilizer modification on the rate of urea hydrolysis. *Soil Sci.* 136:56-63.
- Warncke, D.D., and J.R. Brown. 1998. Potassium and other basic cations. p. 31-33. *In* J.R. Brown (ed.) Recommended chemical soil test procedures for the north central region. North Central Regional Research Publication No. 221 (revised). Missouri Agri. Exp. Stn., Columbia, MO.
- Watson, M.E., and J.R. Brown. 1998. pH and lime requirement. p. 13-16. *In* J.R. Brown (ed.) Recommended chemical soil test procedures for the north central region. North Central Regional Research Publication No. 221 (revised). Missouri Agri. Exp. Stn., Columbia, MO.
- Whalen, J.K., C. Chang, and B.M. Olson. 2001. Nitrogen and phosphorus mineralization potentials of soils receiving repeated annual cattle manure applications. *Biol. Fertil. Soils* 34:334-341.
- Williams, P.H., S.C. Jarvis, and E. Dixon. 1998. Emission of nitric oxide and nitrous oxide from soil under field and laboratory conditions. *Soil Biol. Biochem.* 14:1885-1893.
- Zak, D.R., W.E. Holmes, N.W. MacDonald, and K.S. Pregitzer. 1999. Soil temperature, matric potential, and the kinetics of microbial respiration and nitrogen mineralization. *Soil Sci. Soc. Am. J.* 63: 575-584.
- Zogg, G.P., D.R. Zak, D.B. Ringelberg, N.W. MacDonald, K.S. Pregitzer, and C. White. 1997. Compositional and functional shifts in microbial communities due to soil warming. *Soil Sci. Soc. Am. J.* 61:475-481.