

Rate of Aerobic Nitrogen Transformations in Six Acid Climax Forest Soils and the Effect of Phosphorus and CaCO₃

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ABSTRACT. Nitrogen transformations (mineralization, nitrification, and nitrous oxide production) were evaluated in acid forest floor soils collected from six climax forest sites on Blackhawk Island, Wisconsin. Soils' acidity (CaCl₂) ranged from pH 3.9 to 5.1, and organic matter concentrations varied from 2.4 to 59.0 percent. The samples were incubated aerobically for 4 weeks at 30°C under field moist conditions. Treatments were: control; 100 mg P (as KH₂PO₄) kg⁻¹ soil; CaCO₃; and P with CaCO₃.

Nitrification of mineralized N ranged from nearly complete in the SM Alfisol to almost non-existent in the Histosol. Addition of P had little effect on ammonification or nitrification. Liming, however, greatly enhanced ammonification on nonnitrifying or slowly nitrifying soils and both ammonification and nitrification in nitrifying soils. Adding P and lime together did not affect N transformations compared to liming alone. Nitrous oxide emission rates in the soils were related to nitrification. From 0.03 to 0.3 percent of the NH₄-N nitrified was released as N₂O-N. Phos-

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phorus addition had little effect but liming increased N_2O emission rates in soils where nitrification was also enhanced. *FOREST SCI.* 31:680-684.

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THE LACK OF UNDERSTANDING of the internal N cycle in forest soils is evident from our inability to predict N availability, NO_3^- production, and N losses in these soils (Vitousek and Melillo 1979, Keeney 1980). Olson and Reiners (1983) have discussed the potential factors that affect the rate of nitrification in forest ecosystem soils. These include low pH, limiting nutrients, particularly P, inadequate low nitrifier population density, low ammonium availability due to either low mineralization rate or rapid immobilization rate, and presence of substances inhibitory to nitrification in forest floors.

We initiated a study on the effect of P addition and pH modification by liming on the mineralization, nitrification, and nitrous oxide (N_2O) production in a set of forest ecosystem soils from Blackhawk Island, Wisconsin. The soils and the vegetation at this site are characterized in Pastor and others (1984). The site was selected since a previous study had shown that the soils have a wide range in N mineralization and nitrification rates, and that the soil P status is correlated with the nitrification rate in these soils (Pastor and others 1984). As a starting point, we wanted to investigate the effects of factors, such as pH and nutrients, limiting nitrification in these soils before evaluating any possible inhibitory substances involved in control of nitrification and N_2O production. Our ultimate aim is to develop mechanistic models of factors affecting N_2O production in natural ecosystem soils so that their contribution to atmospheric N_2O and ultimately the effect of man's activities on the ozone layer can be assessed (Crutzen 1981, Goodroad and Keeney 1984a).

MATERIALS AND METHODS

The soil samples (Table 1) were obtained from study sites at Blackhawk Island described by Pastor and others (1984). The 70-ha island is located in the Wisconsin River north of Wisconsin Dells (latitude $43^{\circ}40'N$, longitude $89^{\circ}45'W$). The edaphic climax forests (>100 yr age) had never been logged. Surface (0-10 cm) soil samples (the forest floor and some mineral soil) were collected in autumn 1983. Five bulk samples were collected at each site and were combined to give a composite sample. The samples were refrigerated ($2^{\circ}C$) without amendments until used. For the analyses reported in Table 1, soil pH was measured in a soil- $CaCl_2$ (0.01 M) suspension using a 1:2 soil to $CaCl_2$ solution ratio, and NH_4-N and NO_3-N by steam distillation (Keeney and Nelson 1982). Organic matter content was determined by the method of Harris and Adams (1979).

Field moist soil samples (10 g) were placed in 125 mL Erlenmeyer flasks. The treatments were: (1) control, 1 mL of water; (2) addition of 1 mL of a solution containing 1,000 mg L^{-1} P (as KH_2PO_4); (3) addition of 0.2 g (2 percent) of reagent-grade, powdered $CaCO_3$; and (4) addition of P and $CaCO_3$. This is a 2×2 factorial with two replications. Preliminary experiments with these soils showed that addition of 2 percent $CaCO_3$ resulted in neutral pH (6.7 to 7.5) by 24 h of equilibration, and that addition of KH_2PO_4 lowered soil pH ca. 0.1 unit without $CaCO_3$ and did not affect the pH of $CaCO_3$ -amended soils. Each flask was swirled gently to mix the contents, stoppered with a rubber septum and incubated in a constant temperature chamber at $30^{\circ}C$ for 4 weeks. The flasks were aerated every 4 days, and the flask atmospheres sampled at 9, 20, and 28 days for N_2O concentration. On days 20 and 28, the atmosphere samples were obtained before aeration, and soil inorganic N was determined at 28 days. Nitrous oxide concentration was measured with a Perkin Elmer Sigma 3 gas chromatograph equipped with a high temperature electron capture detector (Goodroad and Keeney 1984b). All results are calculated on the basis of oven-dry soil.

RESULTS

The 0-10 cm sample included the forest floor in all but the disturbed RP. The SM and the RP had a mor-type humus while the remainder had mull-type humus (Table 1). The soils were relatively dry as collected, and when incubated with the additional 1 mL of water or

TABLE 1. Some characteristics of the Blackhawk Island sites and soils used in this study.

No. ^a	Site		Forest floor type	pH (CaCl ₂)	Soil water content ^c		Or- ganic matter ^c	Inorganic N ^c	
	Soil	Stand ^b			Initial	Incubated		NH ₄ -N	NO ₃ -N
					Percent		Per-	mg kg ⁻¹	
					--- oven-dry soil ---		cent	-----	
9	Histosol	Hemlock (H)	Mor	3.9	47.1	61.8	59	70	1
7	Alfisol	Sugar maple (SM)	Mull	5.1	30.5	43.6	22	5	26
6	Alfisol	Red oak (RO)	Mor	4.3	24.1	36.5	16	54	16
5	Alfisol	White oak (WO)	Mor	4.4	24.0	36.5	25	36	12
3	Spodosol	White pine (WP)	Mor	4.4	20.1	32.1	42	15	21
1	Entisol	Red pine (RP)	Disturbed	4.4	7.3	18.0	2.4	11	7

^a Numbers refer to sites in Figure 1, Pastor and others (1984).

^b Hemlock, *Tsuga canadensis* (L.) Carr.; sugar maple, *Acer saccharum* Marsh.; red oak, white oak, *Quercus rubra* L., *Q. alba* L.; red pine, white pine, *Pinus resinosa* Ait., *P. strobus* L.

^c Determined in this investigation.

P solution, the water contents were in the range of 50 to 60 percent of saturation (based on weight of H₂O held when soils were saturated with H₂O). Except for the RP, organic matter contents were high. Most samples contained considerable exchangeable NH₄.

All soil samples exhibited net N mineralization with the greatest amount of mineral N produced in the organic soil and the lowest in the low organic matter RP (Table 2). Nitrification of mineralized NH₄ ranged from nearly complete in the SM to almost non-existent in the Histosol. Addition of P did not influence the rate of mineralization or nitrification. Liming markedly increased N mineralization in all samples and greatly increased the nitrification rate in all but the SM (which was already nitrifying) and the Histosol.

TABLE 2. Mineral N produced in six forest soils treated with water, P, CaCO₃, or P + CaCO₃ after 28 days at 30°C.

Site ^a	Control			+P			+CaCO ₃			+P + CaCO ₃		
	NH ₄	NO ₃	Sum	NH ₄	NO ₃	Sum	NH ₄	NO ₃	Sum	NH ₄	NO ₃	Sum
	Mineral N (mg N kg ⁻¹ soil) ^b											
H	439 (34)	3 (0)	442 (34)	410 (12)	1 (1)	411 (12)	521 ^c (0)	5 (0)	526 ^c (0)	549 (5)	7 (0)	556 (5)
SM	1 (0)	61 (2)	62 (2)	1 (4)	46 (17)	47 (17)	-1 (1)	94 (1)	93 (1)	-1 (2)	83 (7)	82 (7)
RO	111 (20)	25 (2)	136 (21)	118 (7)	29 (4)	147 (7)	-43 (8)	251 (11)	208 (14)	-11 (17)	232 (57)	221 (59)
WO	117 (25)	14 (3)	131 (25)	113 (26)	9 (1)	122 (26)	-30 ^c (58)	259 (58)	229 ^c (58)	-22 (8)	258 (15)	236 (17)
WP	53 (3)	69 (0)	122 (3)	79 (15)	58 (3)	137 (16)	-10 (0)	182 (14)	172 (14)	-11 (1)	174 (0)	163 (1)
RP	7 (5)	17 (3)	24 (6)	8 ^c (3)	15 (3)	23 ^c (3)	-8 (1)	6 (1)	53 (1)	-8 (1)	57 (8)	49 (8)

^a See Table 1 for site descriptions.

^b Values are mineral N at end of incubation minus initial (Table 1) values. Numbers in parentheses represent standard deviations.

^c Only one value available.

TABLE 3. Ratio of N_2O -N produced to NO_3 -N produced during aerobic incubation of six forest soils treated with water only, P, $CaCO_3$, or P + $CaCO_3$ after 28 days at 30°C.

Site	N_2O -N				Ratio			
	Control	+P	+ $CaCO_3$	+P + $CaCO_3$	Control	+P	+ $CaCO_3$	+P + $CaCO_3$
	N_2O -N (μg N kg^{-1} soil)				N_2O -N/ NO_3 -N $\times 10^3$			
H	7 (2)	3 ^a	4 (1)	5 (1)	2.09 (0.61)	2.27 ^a	0.75 (.17)	0.78 (.19)
SM	19 (1)	29 (11)	29 (13)	7 ^a	.31 (.02)	0.62 (.33)	.31 (.14)	.08 ^a
RO	49 (8)	48 (1)	110 (19)	109 (6)	1.92 (.35)	1.66 (.22)	.44 (.08)	.47 (.12)
WO	36 (4)	28 (1)	109 ^a	128 (9)	2.51 (.55)	3.03 (.51)	.42 ^a	.50 (.04)
WP	56 (4)	62 (16)	96 (8)	39 ^a	.81 (.05)	1.06 (.28)	.52 (.06)	.22 ^a
RP	6 (3)	4 (1)	25 ^a	11 ^a	.36 (.16)	.29 (.05)	.40 ^a	.19 ^a

^a Only one value available.

Compared to $CaCO_3$ alone, addition of P did not markedly affect mineralization, and P had negligible effects or actually decreased somewhat the rate of nitrification.

The amount of N_2O -N evolved (Table 3) was roughly proportional to the rate of NO_3 formation, ranging from very low values in the Histosol and the non-limed RP up to 100 μg N_2O -N kg^{-1} soil with the limed RO and WO samples. It is important to note that N_2O -N comprised only a very small portion of the nitrified N in these aerobic systems.

DISCUSSION

It would appear from the results of our study that low acidity and not lack of available P is the cause for the slow rate of nitrification in these forest soils. In contrast, Pastor and others (1984) found that the nitrification rate measured in the field at these sites was correlated with the available PO_4 supply in the spring and early summer. Our findings agree with those of Wheeler and Donaldson (1983) who found that the rates of nitrification in an upland forest sere in the Arkansas Ozarks could be explained largely on the basis of soil acidity. Nommik (1978) reported that liming forest floors increased mineralization of N, and that the effect of liming was more pronounced in mor than in mull humus samples. Olson and Reiners (1983), on the other hand, indicated that increase in pH and essential nutrients did not increase nitrification in the Al horizon beneath balsam fir (*Abies balsamea* Mill.) forest floors and felt that an unidentified inhibitor was the cause of lack of nitrification. Klein and others (1983) noted high rates of nitrification in some extremely acid forest soils of the Adirondacks.

Results of our study show that an elevation of pH in these forest floor samples enhanced mineralization and nitrification in all but the Histosol. It is possible that an increase in soil pH increased the microbial population and/or the activity of the nitrifiers increased in response to higher pH. Alternatively, the toxicity of nutrients such as Al or the activity of an inhibitor could have been alleviated.

Until recently, denitrification was generally regarded as the dominant mechanism for N_2O production in soils. However, Bremner and Blackmer (1981) provide conclusive evidence that N_2O is released on aerobic incubation of soils treated with NH_4 . Several workers have found that from about 0.05 to 0.2 percent of the NH_4 nitrified is evolved as N_2O (see Goodroad and Keeney 1984b for literature review). Goodroad and Keeney (1984b) found that soil pH, temperature, and water content were interactive in affecting the amount of N_2O -N evolved during nitrification. Using an agricultural soil, they noted

highest ratios of N_2O -N/ NO_3 -N produced in an acid (pH 4.7) soil at 30°C. Under conditions where the water content was low, the ratio at pH 4.7 and 30°C was 1.9 to 2.4×10^{-3} while at pH 5.1 the corresponding value was 0.9×10^{-3} .

The ratios obtained in this study (Table 3) are generally similar to those obtained by Goodroad and Keeney (1984b). Liming of the RO and WO soils markedly decreased the ratio; nitrification was most severely limited by pH in these soils. With soils where the liming effect was not as marked, the ratio declined less or not at all on liming. We are not able to define mechanisms for the higher N_2O evolution per unit of N nitrified in the acid RO, WO, and perhaps WP soils. Possible mechanisms include chemical reaction of nitrite (Nelson 1982) or changes in the metabolism or ecology of the ammonium oxidizers (Focht and Verstraete 1977).

These data indicate that the very low N_2O emissions sometimes observed from forest soils (Goodroad and Keeney 1984a) are likely due to the limitations on nitrification in the system, and that the overall estimate of 0.1 to 0.3 percent of the N_2O produced per unit of N nitrified (Goodroad and Keeney 1984b) is valid for forest as well as agricultural soils.

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