NITRIFICATION POTENTIAL IN A CERRADO SOIL

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Introduction

Nitrification, the oxidation of NH_4^+ to NO_3^-, is one of the major processes in the nitrogen cycle in natural ecosystems. The oxidation is generally mediated by two types of chemoautotrophic bacteria, one oxidizing ammonia to nitrite and the other oxidizing nitrite to nitrate. Autotrophic nitrifying bacterias are considered to be the predominant agents of nitrification in the soil ecosystem. However, the occurrence of heterotrophic nitrification (Focht and Verstraete, 1977; Stroo et al. 1986) or chemical nitrification (Barlett, 1981) has been suggested.

In cropped soils, nitrification is important for the effective utilization of nitrogen fertilizer and for the nutrition of crops. The nitrification potential of the Cerrado soils has not been extensively investigated. The purpose of this study was to evaluate the nitrification potential of one Cerrado soil.

Materials and Methods

Soil samples

Soil samples of a dark red oxisol were colected at the 0-10 cm layer of a dark red oxisol, lacated at the experimental area of the Centro de Pesquisa Agropecuária dos Cerrados, Brasília,DF, Brazil. The samples were taken from a corn field in the treatments with and without N (200kg/ha), and from a non-cultivated area. Some of the characteristics of these samples are given in Table 1. The samples were immediately sieved, mixed and used for analysis. Air drying of soil was not employed in the present study because the nitrification activity would have been lost by this treatment.

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TABLE 1 - Characteristics of soil samples.

Soil	pH(H_2O)	Total N (%)
Latosol non-cultivated soil	5.3	0.16
Latosol non-fertilized	6.4	0.11
Latosol fertilized (200 kgN of urea/ha)	6.5	0.12

Soil suspension experiment

Forty gram of moist soil samples were transferred into 500 ml Erlenmeyer flasks containing 200 ml of medium for ammonia or nitrite- oxidizing bacteria. The medium for ammonia oxidation consisted of $(NH_4)_2SO_4$ (0.2g/l of N), KH_2PO_4 (1g/l), $MgSO_4.7H_2O$ (0.1g/l), $FeSO_4.7H_2O$ (0.03g/l), $CaCO_3$ (2g/l). The medium for nitrite oxidation consisted of $NaNO_2$ (0.02g/l of N), KH2PO4 (1g/l), $MgSO_4.7H_2O$ (0.1g/l), NaC1 (0.3g/l), $FeSO_4.7H_2O$ (0.03g/l), $CaCO_3$ (2g/l). The pH of both media was adjusted to 7.0. The soil suspensions were incubated at 25°C on a reciprocal shaker operating at 110 rpm. Nitrapyrin (2-chloro-6(trichloromethyl)-pyridine), a selective inhibitor of autotrophic ammonia oxidation, or sodium chlorate, which is a specific inhibitor of autotrophic nitrite oxidation, was used to differentiate autotrophic from heterotrophic nitrification. Nitrapyrin was added at the concentration of 10 ppm before incubation. Sodium chlorate was added at the concentration of 10 mM. Samples were taken periodically for the analysis of NH_4^+-N, NO_2^--N, NO_3^--N. The experiments were performed with two replicates.

Soil incubation

Twenty gram of the soil samples in duplicate were transferred into a 100 ml conical beaker or flask. A known quantity of the $(NH_4)_2S0_4$ solution (to 20 mgN/100g of soil) was added to each sample. The beakers were covered with a thin polyethylene film to maintain the soil moisture content. Incubation was carried out in the dark at 25°C. After 12 days, the samples were extracted with 50 ml of 10% KC1 and the contents of NH_4^+-N, NO-N, NO_3^--N were determined. The initial concentration of these inorganic nitrogen compounds was determined immediately after the addition of the $(NH_4)_2SO_4$ solution. Nitrapyrin was also used. $CaCO_3$ was added to the non-cultivated soil to study the effect of neutralization on the nitrification activity in non-cultivated soil.

Chemical analysis

The concentrations of NH_4^+-N and NO_3^--N were determined by distillation of the filter extract with MgO and Devarda alloy. The NO_2^-N contents was measured by the Griess-Ilosvay method. The soil pH was determined in the soil water suspension (1/2.5 w/v). The total nitrogen content was determined by the Kjeldahl method.

Results and Discussion

Nitrite oxidation and NO_3^- production in the soil suspension culture are shown in Figure 1. The oxidation of NO_2^- was much more pronounced in the fertilized soil than in the non-fertilized and non- cultivated soils. NO_2^- oxidation during the 8 day period amounted to 21.9% of the added NO_2^- in the non-fertilized soil, 22.4% in the non-cultatived soil, and almost 100% in fertilized soil. These results show that fertilization led to an increase in the nitrite oxidation potential. Chlorate completely inhibited the nitrite oxidation in all the tested soil suspensions, indicating that nitrite oxidation was caused by the autotrophic nitrite-oxidizing bacteria.

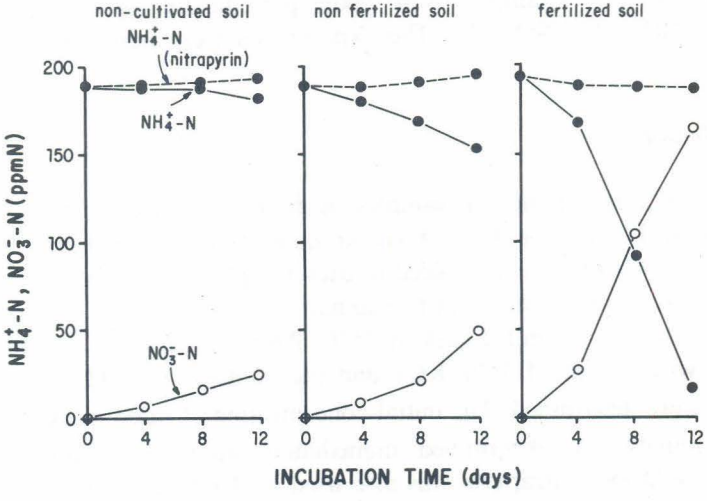

FIG 1 - Time course of NO_2^- - N oxidation in soil suspension culture.

Ammonium oxidation and NO_3^- production in the soil suspension culture are shown in Figure 2. NH_4^+ was rapidly oxidized to NO_3^- in the fertilized soil. The NH_4^+ -oxidizing activity in the two other soils was lower than that in the fertilized soil. These findings indicate that fertilization resulted in an increase in the number of ammonia-oxidizing bacteria. Nitrapyrin completely inhibited the NH_4^+ oxidation in all the tested soil suspensions, suggesting that the autotrophic ammonia-oxidizing bacteria were responsible for the nitrification in the Cerrado soils tested.

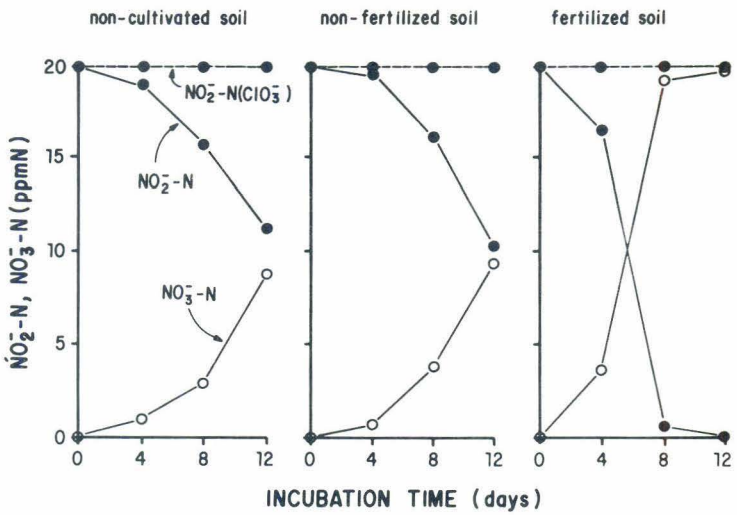

FIG. 2 - Time course of NH_4^+ -N oxidation in soil suspension culture.

The changes in the NH_4^+-N and NO_3^--N concentrations after incubation are shown in Table 2. The production of nitrate was much higher in the fertilized soil than in the non-fertilized and non-cultivated soils. During the incubation period of 12 days, 57.4% of added NH_4^+ -N was oxidized and 11 mgN of $NO_3^--N/100g$ of soil was produced. The amounts of NO_3^- producted in the non-fertilized and non-cultivated soils were 1.9 and 1.4 mg/100g, respectively. The pH of the non-cultivated soil was lower than that of the two cultivated soils. Adjusting the soil pH to about 6.5 by $CaCO_3$ ammendment did not affect its nitrification potential. Nitrapyrin completely inhibited the formation of nitrate in all the tested soils.

TABLE 2 - Change in NH_4^+ -N and NO_3^- -N levels after the addition of $(NH_4)_2SO_4$ incubation for 12 days.

Soil	Inhibitor	Inorganic N (mgN/100g of dry soil)	
		NH_4^+ -N decrease[a]	NO_3^- -N increase
non-cultivated soil	none	2.6	1.6
	nitrapyrin	-1.6	< 0.1
non-cultivated soil	none	2.5	1.2
($CaCO_3$ added)	nitrapyrin	-2.1	< 0.1
non-fertilized	none	2.8	2.3
	nitrapyrin	-0.5	< 0.1
fertilized	none	11.9	10.9
	nitrapyrin	-0.8	< 0.1

[a] Negative values indicate a net production of NH_4^+ -N during the incubation.

Autotrophic-nitrifying bacteria derive all their energy from the oxidation of NH_4^+ or NO_2^- . In natural ecosystems such as the Cerrados, the supply of NH_4^+ for the nitrifying bacteria is markedly limited. The nitrification potential is lower in acid soils than in neutral soils because autotrophic nitrifying bacteria are sensitive to acidic conditions. The nitrification activity in the Cerrado non-cultivated soil was very low. The results obtained in the present study indicated that an abundant supply of NH_4^+ led to a increase in the nitrification activity.

Conclusion

1. Fertilization with urea results in an increased the nitrification activity in the soil tested.
2. The nitrification activity in the soil tested was caused by the autotrophic nitrifying bacteria.

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