

Nitrogen Fertilizer Effects on Nitrous Oxide Emission from Southwest Brazilian Amazon Pastures

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Abstract

Beef production is one of the most important agricultural activities in Brazil. In order to increase production without increasing deforestation, farmers are intensifying breeding and pasture improvements. The main technique for increasing pasture improvement is the application nitrogen fertilizer, but this action can result in emission of nitrous oxide (N₂O). We assessed the impact of nitrogen fertilizer application on GHG emissions and pasture yield in a pasture located at Southwest Brazilian Amazon. Agronomic recommended rates of nitrogen fertilizer (NF) and higher rates, as two times (2NF) and four times (4NF) the recommended rate were applied. A control treatment with no fertilizer was also analysed. The experiment had 30 days duration, where we observed the baseline emissions from all treatments, including control. Nitrogen fertilizer application resulted in high N₂O emissions. We found no differences between NF and 2NF treatments, but all treatments were different from control. The higher forage yield leads to low N₂O emission per kg of forage in 4NF treatment. According to our study, the best (agro-environmental benefits) practice is the application of 100 kg N ha⁻¹ (2NF treatment) in the region studied.

Keywords: Beef production; Tropical climate; N₂O; Pasture yield

Introduction

Brazil is the second largest beef exporter, responsible for 15% of worldwide production [1]. The typical system of beef production in Brazil is pasture-based, predominantly occurring on unimproved pastures. Pastures occupy three-quarters of the national agricultural area, about 180 million hectares [2]. There are projections of increased demand for beef in the order of 2.5% per year by 2017-2018. In order to meet this demand, Brazilian farmers must develop a more intensive system in order to produce more beef without increase deforestation [3]. This intensive system must have higher beef production per unit area, with low emissions of greenhouse gases (GHGs) per kg of beef produced. If farmers do not adopt sustainable options for pasture intensification, deforestation could be increased, increasing GHG emissions from the sector. The improvement of the whole system of beef production is a key component to reduce emissions from all relevant sources, including land use, land use change and livestock [4].

In order to improve the beef system, intensification methods must be applied, such as the use of nitrogen fertilizer. The main limiting-nutrients for grass growth in Brazilian conditions are phosphorus (P) and nitrogen (N). The application of nitrogen fertilizer enhances the availability of N to the plant and microorganisms, but an excess of N can result in nitrous oxide (N₂O) emissions through nitrification and denitrification processes [5]. The effect of N fertilizer on N₂O emission is well reported in the literature [6,7]. However, there are very few studies in tropical climates examining N fertilizer application in pastures [8,9] with respect to GHG emissions and they do not cover the range of edaphoclimatic conditions.

We measured the effect of N fertilizer application on soil N₂O emission. To simulate the intensification practices, we tested the effect of the application of the currently recommended levels of fertilizer on GHG emissions. There is anecdotal evidence that farmers usually apply more than the recommended rate. Therefore, we also tested higher rates of N fertilizer to verify the impacts on GHG intensities.

Materials and Methods

The experiment was carried out on a permanent pasture, covered by *Brachiaria* and was not grazed by livestock before or during the experiment. The studied site was split in 4 paddocks (0.1 ha). Plots consisted of an area fertilizer application (0.05 ha). The experiment was carried out from 09 November to 10 December of 2012 (summer) at Agropecuária Nova Vida, Rondônia state, Brazil (10°10'05"S and 62°49'27"W) under tropical climatic conditions (Aw-Köppen climatic classification). Soil is an Oxisol (Ustox), and its texture is sandy loam. Soil properties (upper 10 cm) at the start of the experiment are showed in Table 1. Meteorological data were recorded at the nearest meteorological station (rainfall and air temperature), which was within 1 km of the field site.

We studied the application of ammonium nitrate (NH₄NO₃) at rates of 0, 50, 100 and 200 kg N ha⁻¹ (treatments C, NF, 2NF and 4NF, respectively), with five replicates to each treatment, in a complete randomized block design. The fertilizer was applied in the first day of the experiment, right before the first sampling.

| Sand | Clay | Silt | pH | Bulk density | Total C | Total N |
|-------------|-------------|-------------|-------------------|-------------------|-------------------------------|-------------------------------|
| -----%----- | -----%----- | -----%----- | CaCl ₂ | g m ⁻³ | -----g kg ⁻¹ ----- | -----g kg ⁻¹ ----- |
| 65.0 | 26.7 | 8.3 | 5.1 | 1.5 | 27.3 | 3.0 |

Table 1: Soil properties (0-10 cm) at the beginning of the field experiment.

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The closed static chamber technique [10] was used to collect gas samples. At the field, unvented chambers (28 cm diameter, 13 cm height) were placed two days before the first gas sampling. The chambers were inserted to a depth of up to 3 cm to ensure an airtight seal. The volume enclosed by the chamber was approximately 11 L. At the time of sampling, lids were placed on top of the chambers and a seal was achieved via a water-filled groove on the chamber that the lid fitted in to. Gas sampling was normally carried out between 9:00 and 12:00. Samples were collected at 0, 10, 20 and 30 minutes after the chambers were closed. A 20 ml syringe was used to collect the gas samples from the chambers, which were then transferred to pre-evacuated 13 ml headspace vials using a hypodermic needle. The glass vials had a chlorobutyl rubber septum (Chromacol). The pre-evacuation was carried out using a vacuum pump.

Gas sampling was carried out daily during the first week, then twice a week for 3 weeks, and then once a week until the end of the experiment. The samples were analysed within 2 weeks of collection using gas chromatography (GC - Shimadzu 2014). The N₂O was detected with an ECD (electron capture detector).

The flux of N₂O was calculated using the linear change in the concentration as a function of the incubation time within the chamber. Gas fluxes were calculated from the time vs. concentration data using linear regression. These data were used to calculate the cumulative emissions over the experimental period by the linear interpolation of data points between two successive days and numerical integration of the area under the curve using the trapezoid rule [11]. The emission factors were calculated using the recommendations of the IPCC [12].

Designated soil sampling plots were installed adjacent to each gas chamber, which also received the same fertilizer rate. Soil (0-10 cm) was sampled on days 1, 7, 14, 21 and 28. Soil mineral N concentrations were determined by extraction with 2 M KCl with a 1:2 ratio of soil

and extractant [13]. Soil extracts were filtered and stored at 4°C. Concentrations of NH₄⁺ and NO₃⁻ in the extracts were determined by automated flow injection analysis (FIA) [14].

Gravimetric moisture contents were determined after drying at 105°C for 48 h. Grasses from each chamber were cut at 4-5 cm height at the end of the experiment. The green matter was transferred to a pre-weighed paper bag and dried at 70°C for 1 week. After that, dry matter weight was recorded.

Statistical analyses

Total GHG emissions were estimated by calculating cumulative fluxes over an experimental period of 30 days. Data were verified for normal distribution and treatment means for daily N₂O fluxes and cumulative fluxes over the period of the experiment were compared using one-way analysis of variance. To determine the statistical significance of the mean differences, Turkey tests were carried out at 0.05 probability level.

Results

The average air temperature and total precipitation were 29°C (varying from 25 to 33°C) and 250 mm (varying from 3 to 107 mm) (Figure 1). Those conditions are representative of the summer season of the southwestern part of the Brazilian Amazon. Due to the periodical rainfall, the water-filled-pore-space (WFPS) was high during all the experiment, ranging from 69 to 86% (Table 2).

Nitrous oxide emissions were variable over the study period (Figure 2). Our results showed that the application of N fertilizer increased N₂O emissions, changing the pasture status from a net sink to a net source of N₂O (Table 3). The increase in N fertilization had a high correlation with N₂O emission ($r^2=0.99$). The emission factors calculated were 0.45, 0.12 and 0.17 for NE, 2NF and 4NF treatments, respectively.

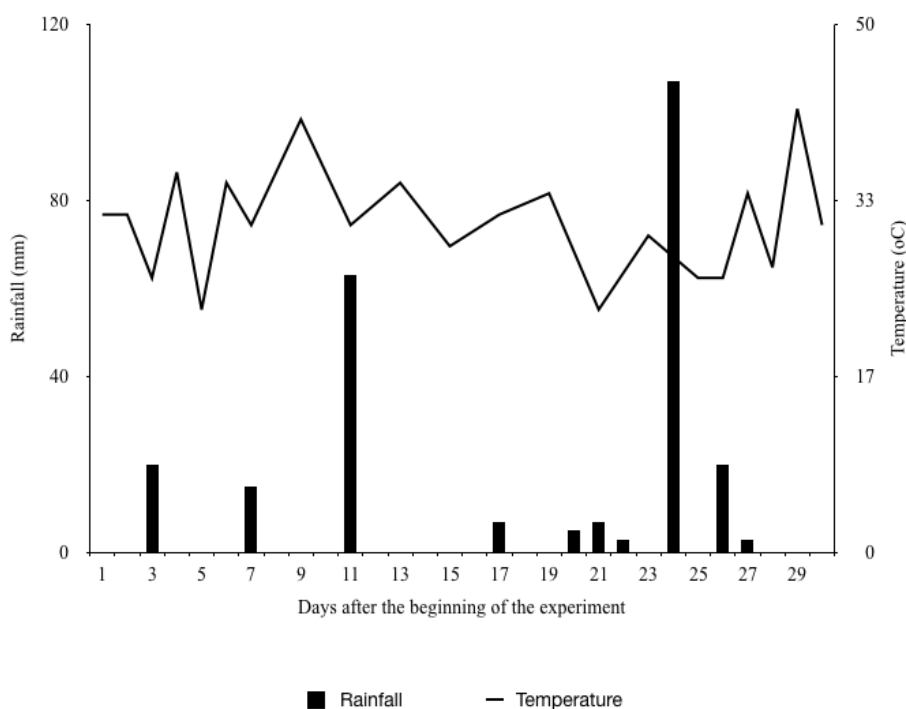


Figure 1: Climatic data at the study site (Rondônia, Southwestern Brazilian Amazon, Brazil).

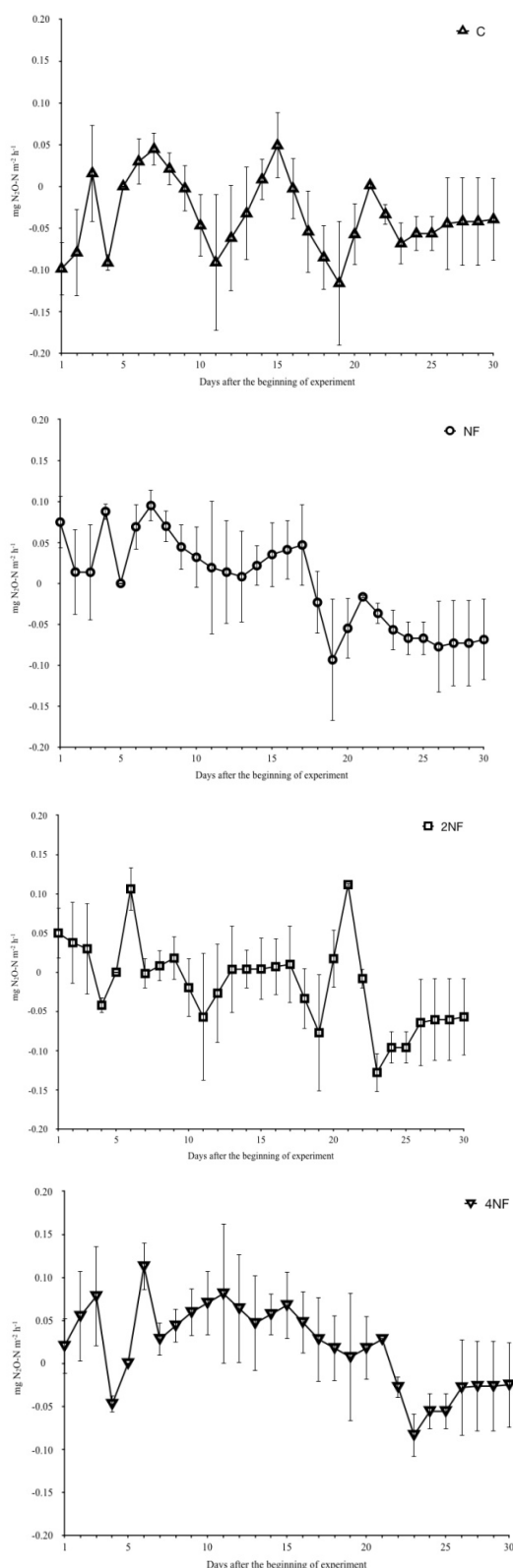


Figure 2: N₂O emissions from the studied site. C: Control; NF: application of 50 kg N ha⁻¹; 2NF: application of 100 kg N ha⁻¹; 4NF: application of 200 kg N ha⁻¹. The error bars denote the standard deviation.

Soil NH₄⁺ levels in the NF treatment increased rapidly after the application of N fertilizer, with levels different from the control (p<0.05) throughout the experiment until day 21 (Figure 3). Soil NH₄⁺ levels in the 2NF and 4NF treatments increased after day 7, and remained high until day 21 (Figure 3). Soil NO₃⁻ concentrations in all treatments were significantly higher than the control during throughout the experiment.

The forage yield increased with N fertilizer application (Table 3). While all N-treated plots had significantly higher N₂O emissions than the control, especially 4NF (Table 3), the increase in forage yield with N fertilizer lead to lower N₂O emission per kg of forage (166, 59 and 105 mg N₂O kg dry matter⁻¹ for NF, 2NF and 4NF, respectively).

Discussion

The increase in N₂O emission was expected, since the application of ammonium nitrate increases the availability of nitrate in soil (Figure 3). The available N can be quickly taken up by plants or lost as N₂O in a few days [15]. The assimilation of NH₄⁺ is energetically more efficient than NO₃⁻ [16]. *Brachiaria* grasses are well adapted to the N poor soils from Brazil. When both NH₄⁺ and NO₃⁻ are available in soil, the plant absorbs NH₄⁺ preferably, leading NO₃⁻ that can be denitrified in soil [17].

The relationship between the N₂O emission and the amount of N fertilizer applied was not linear. Other studies showed that the exponential curve fits better [18]. In our study the N₂O emission from NF and 2NF treatments did not show statistical difference. In this case was not possible to test wich curve would fit better, since there was only 2 possible points (NF × 4NF or 2NF × 4NF).

Soil moisture is a key factor for N₂O emission [19]. During this study the soil showed high WFPS (Table 2) due to the periodical rainfall, typical from the summer in the Amazon region. The temperature is also an important factor in the studied situation, since the high temperature of the tropics can stimulate N turnover, consuming O₂ and creating an anaerobic environment, ideal for the denitrification process. The interaction between soil moisture and temperature can provide a possible explanation for the negative fluxes observed, mainly in the control treatment. The increase in temperature has a positive impact in the N mineralization, combined with the ideal anaerobic environment

| | % | SD | CV |
|--------|------|----|------|
| Day 1 | 69.6 | b | 7.8 |
| Day 7 | 72.4 | b | 8.3 |
| Day 14 | 84.4 | a | 8.8 |
| Day 21 | 82.3 | a | 10.4 |
| Day 28 | 86.3 | a | 9.1 |

Table 2: Soil water filled pore space (WFPS-%) in the soil (0-10 cm) at the field experiment. SD: Standard deviation; CV: Coefficient of variation; Means followed by the same letters in columns are not statistically different (Tukey, pB 0.05).

| Treatment | N-N ₂ O | | | Yield | | |
|--------------------------|--------------------|----|-----|---------|----|-------|
| | CE | SD | CV | Average | SD | CV |
| ⁰ N (control) | -18.7 | c | 4.9 | 0.08 | d | 0.008 |
| NF | 3.9 | b | 1.2 | 0.14 | c | 0.009 |
| 2NF | -7.0 | b | 0.4 | 0.19 | b | 0.023 |
| 4NF | 15.4 | a | 6.2 | 0.32 | a | 0.025 |

Table 3: Cumulative N₂O emission from field study and the effect of nitrogen fertilizer application on pasture yield. C: Control; NF: application of 50 kg N ha⁻¹; 2NF: application of 100 kg N ha⁻¹; 4NF: application of 200 kg N ha⁻¹; CE: Cumulative emission; SD: Standard Deviation; CV: Coefficient of Variation; Means followed by the same letters in columns are not statistically different (Turkey, p ≤ 0.05).

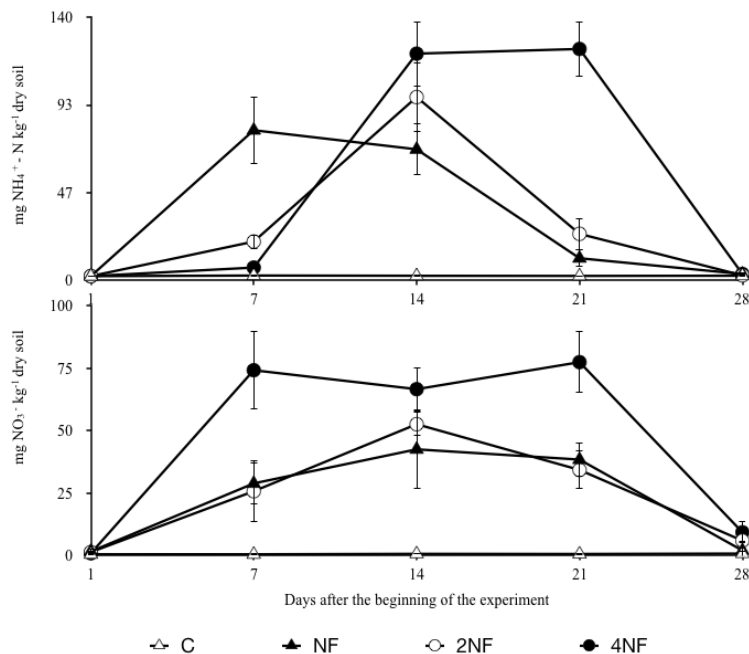


Figure 3: Ammonium (NH₄⁺) and nitrate (NO₃⁻) content in soil from the studied site. C: Control; NF: application of 50 kg N ha⁻¹; 2NF: application of 100 kg N ha⁻¹; 4NF: application of 200 kg N ha⁻¹. The error bars denote the standard deviation.

created by the high WFPS. In such situation, most of the N of the soil is consumed by microorganisms or completed denitrificated to N₂. Wetlands can act as N₂O sinks [20]. The high WFPS can contribute to complete denitrification, reducing even further the N₂O emission. One possible explanation for the negative fluxes is the use of N₂O as an final electron acceptor in the absence of other source [21]. Recently, more studies have been reported negative fluxes of N₂O [22-24] but there are no consensus of which mechanism is responsible for the N₂O consume. More studies must be done to address this gap, specially those in controlled conditions with different types of soil, moisture content and temperature, associated with a profile of the microbial communities.

The Biological Nitrification Inhibition (BNI) is other factor that can have a high impact on N₂O emission from Brazilian pastures. In pastures covered by *Brachiaria* grasses the flow of N from NH₄⁺ to NO₃⁻ is restricted by a natural root exudate (brachialactone), and NH₄⁺ accumulates in soil [17]. In such situation, the BNI keeps NH₄⁺ in the soil and the plant gradually absorbs this nutrient, while the excess is nitrified to NO₃⁻. Such process must be investigated, since *Brachiaria* is the main type of grass in Brazilian pastures.

The emission factors obtained in this study are significantly lower than the recommended by the IPCC (1%), regardless the amount of N fertilizer applied. There is a lack of studies on N₂O emissions from fertilizers in tropical pastures. The study of Morais et al. [9] was conducted in Rio de Janeiro, with a different source of N (urea) and a different type of grass (elephant grass), resulting in a higher emission factor (0.51%) than the obtained in our study. Other studies also reported that N₂O fluxes are larger when ammonium nitrate is used as an N source compared to other mineral or organic fertilizers [14,25]. Cardenas et al. [26] showed higher N₂O emissions in wetter regions of the UK. Soil temperature influences N₂O emissions, increasing the nitrification and denitrification processes [27]. These differences in N source, rainfall and temperature can significantly change the N dynamics in soil. Therefore, our recommendation is that the emission

factors for Brazilian conditions must be specific for the different sources of N, soil type and regions or biomes.

It is only recently that molecular-based analyses of microbial diversity have been combined with measurements of N₂O production and process rates [28]. There are few studies that offer a rigorous assessment of the microbial community and N₂O emissions, most of them with conflicting results [29-31]. We think that more studies concerning the microbial diversity and N₂O emission must be encouraged and performed in order to obtain a better picture of this relationship.

Although our study was a short-term (30 days), we noticed that baseline emissions were achieved after this period of time. Furthermore, usually in Brazil the N fertilizer is applied in fractions during the rainy season. This one-month result completely fits in the interval of application. Our study showed that these fractions must not be higher than 100 kg N ha⁻¹ (2NF treatment), since this dose increased the forage yield (agronomic benefits) and decreased N₂O emission (environmental benefits) (Table 3), resulting in the lowest N₂O emission per kg of Dry matter (agro-environmental benefits) in the studied area.

Conclusion

The application of N fertilizer resulted in an increase in N₂O emission under the studied conditions. Our study showed that the application of N fertilizer at doses above the recommended rate increases the N₂O emission even further. In order to improve grassland management, we advise the application of a maximum fertilizer rate of 100 kg N ha⁻¹.

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