



Soil N₂O emissions from long-term agroecosystems: Interactive effects of rainfall seasonality and crop rotation in the Brazilian Cerrado



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ABSTRACT

In its natural state, the Cerrado biome is a mitigator of soil emissions of nitrous oxide (N₂O). However, the integration of this biome in agricultural activities induced changes in nitrogen (N) dynamics, consequently increasing N₂O emissions to the atmosphere. For one year, N₂O emissions were evaluated under interactive effects of rainfall seasonality and crop rotation in 19-year-old agricultural ecosystems in the Cerrado. The agricultural systems included: (I) no-tillage soybean in the main and sorghum in the late growing season (NTR1); (II) no-tillage maize in the main and pigeon pea in the late growing season (NTR2); (III) soybean in the main and fallow in the late growing season under conventional tillage (CT); (IV) and native Cerrado (NC), as a reference environment. Measurements in a closed static chamber were carried out from October 2013 to September 2014 to determine the fluxes by gas chromatography. The N₂O fluxes were related to the following soil and climate variables: nitrate (NO₃⁻), ammonium (NH₄⁺), soil temperature (Soil temp.), and water-filled pore space (WFPS). The annual N₂O average fluxes of the agroecosystems ranged from zero to 266 μg m⁻² h⁻¹. Fluxes were lowest in the native Cerrado, and in certain periods of the year, especially in the dry season, inflows were observed. The total annual cumulative fluxes from CT, NTR1 and NTR2 were: 1.36; 1.00 and 0.70 kg N₂O ha⁻¹, respectively. In NC, the annual cumulative total was 0.27 kg N₂O ha⁻¹. Under CT, N₂O peaks were highest in the dry period, especially after soybean harvest, from fallow soil. Of the total cumulative emissions in CT, 50% were accumulated during the dry season and 75% during the fallow period, indicating that for the Cerrado with rainfall seasonality, monoculture soybean followed by fallow soil is not an appropriate crop rotation sequence. Among the different tillage systems, NTR2 had the lowest cumulative N₂O emissions. This crop rotation is therefore indicated as the most efficient to mitigate N₂O, with emission peaks not exceeding 100 μg m⁻² h⁻¹, while in NTR1, emissions in the rainy season reached almost 270 μg m⁻² h⁻¹.

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1. Introduction

The Cerrado (Brazilian savanna) covers more than 2 million km², equivalent to 24% of the national territory. Due to the outstanding species richness, this biome is considered as one of 34 global “hotspots of biodiversity” (Bustamante et al., 2012). Over the past four decades, nearly one million km², or 50% of the total Cerrado area, were converted into agricultural areas, mainly

between 1990 and 2011 (Lapola et al., 2014; Bustamante et al., 2014). Approximately 60% of soybean and 48% of maize in Brazil are produced in agricultural areas in the Cerrado (Conab, 2015).

The rapid agricultural expansion in the Cerrado region has led to substantial changes in the biogeochemical cycles (Cruvinel et al., 2011). Among the changes already observed, resulting from chemical, physical and biological disturbances of the soil, changes in the nitrogen (N) dynamics are particularly relevant (Bustamante et al., 2012). One of the consequences of anthropogenic interference with the N dynamics, is the noticeable alteration in nitrous oxide (N₂O) emissions, one of the gases related to global climate change. The complex interactions between some practices of

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management systems and soil-climatic factors affect the rates of organic matter mineralization and can increase N₂O emissions from agricultural soils to the atmosphere (Jerecki and Lal, 2006; Ussiri et al., 2009; Huang et al., 2015).

Recent research reports indicate that Cerrado under native vegetation is a biome that naturally mitigates N₂O emissions (Martins et al., 2015). Explanations for this behavior of the Cerrado are the good drainability and aeration of the soil (Martins et al., 2015), the composition and abundance of denitrifying microbial communities (Lammel et al., 2015), the high soil acidity, and the occurrence of dry spells during the growing season, aside from a well-defined dry season (Davidson et al., 2001). The Latosols of the Cerrado generally have lower N levels and nitrification rates, which also contributes to the low N₂O fluxes of these soils (Nardoto and Bustamante 2003; Carvalho et al., 2006; Chapuis-Lardy et al., 2007; Cruvinel et al., 2011; Martins et al., 2015). However, as already mentioned, soil managements can change the soil properties and gas exchange of ecosystems drastically (Castaldi et al., 2006).

Among the agricultural practices that affect N₂O emissions, soil tillage is already well-known, altering the structure and aeration as well as oxygen concentration (Butterbach-Bahl et al., 2013), the deposition and incorporation of organic residues (Ball et al., 2014), which accelerate the decomposition and N mineralization processes, according to the chemical composition of plant residues (Carvalho et al., 2012). In addition, variables such as the phenological stage of the crop (Hayashi et al., 2015), acidity and fertility levels, air and soil temperature (Butterbach-Bahl et al., 2013), the application of mineral nitrogen fertilizers, and soil moisture also affect N₂O emissions (Martins et al., 2015; Soares et al., 2015; Pimentel et al., 2015). This influence is a result of the changes in nitrification and denitrification reactions, responsible for N₂O formation in soils (Tatti et al., 2014). In Latin America, Brazil is the largest emitter of this gas (Bustamante et al., 2014) and the main source of Brazilian N₂O emissions are agricultural soils, which account for 64% of the direct total emissions (MCTI, 2014).

Despite the large number of studies in recent years on the effects of management systems on soil N₂O emissions, research results about the emission/mitigation potential of no-tillage (NT) and conventional (CT) systems are still divergent (Abdalla et al., 2014). Some studies report higher emissions from NT than CT (Liu et al., 2007; Escobar et al., 2010; Siqueira Neto et al., 2011), while others highlight the mitigating potential of the former (Boddey et al., 2010; Alves et al., 2010). In NT systems, crop rotations with deposition or incorporation of organic residues (legumes/grasses), under ideal conditions of soil moisture and temperature and the formation of soluble organic compounds in different periods are considered promising for a reduction in N₂O emissions from agricultural soils (Dyer et al., 2012; Abdalla et al., 2014; Huang et al., 2015). However, the understanding of the interaction between NT practices with grass- and legume-based crop rotations, as observed in southern Brazil (Bayer et al., 2014) and abroad (Liu et al., 2014), is complex and poorly understood, mainly because this interaction is controlled by environmental factors (Escobar et al., 2010; Pimentel et al., 2015).

In this way, since most studies on the combined effects of the cited practices on soil N₂O emissions were carried out under other soil and climatic conditions, mainly in humid climate regions and for only one growing season (Chen et al., 2008), little is known about these interactive effects under rainfed conditions in the Brazilian Cerrado. Additionally, the monitoring of a complete crop rotation cycle is also important to determine the quantitative response of each management systems in terms of cumulative N₂O emission per grain produced as well as the partial global warming potential (pGWP) (Pramanik et al., 2014; Bayer et al., 2015). According to the latest Brazilian Panel on Climate Change (MCTI, 2014), the available N₂O emission data are still insufficient to allow

a low-uncertainty determination of emissions from agricultural systems, due to the wide diversity of environments in Brazil.

Aside from these effects of soil management, the duration of the rainy season has a strong influence, which lasts for six months, from October to March in the Cerrado, when 90% of the precipitation falls (Klink and Machado, 2005). The Cerrado has well-defined seasons, which influence nitrification and denitrification reactions, mainly when sporadic rainfalls occur that rewet the soil after dry spells. The variability of rainfall in an agricultural year may affect N₂O emissions by stimulating the mineralization of soil organic matter, promoting nitrate accumulation in dry periods and favoring N₂O emissions in the rainy season (Liu et al., 2014).

This effect of rainfall seasonality of the Cerrado on N₂O emissions has already been described by other authors (Alves et al., 2010; Martins et al., 2015), although in an isolated manner, without taking the interactive effects of seasons, management systems and crop rotations of long-term agroecosystems into consideration. Interactions which can lead to N₂O peaks of different magnitudes in agroecosystems, are complex to understand and can vary greatly, depending on the season and period (Sommer et al., 2015). Therefore, our objective was to evaluate N₂O emissions for one year under the interactive influence of seasonal rainfall and crop rotation in 19-year-old agricultural ecosystems in the Cerrado.

2. Material and methods

2.1. Local climate and soil characteristics

The study was conducted for one year, from October 2013 to September 2014, in the experimental area of Embrapa Cerrados in the municipality of Planaltina, DF, Brazil (15°33'33.99" S, 47°44'12.32" W, altitude 1035 m asl). The climate is seasonal, classified as tropical rainy Aw (Köppen), with two well-defined seasons: rainy summers, from October to March, corresponding to the rainy season, and dry winters, from April to September, corresponding to the dry period. The interval from October to March was considered as rainy season, since in the mean, 90% of the rainfall is concentrated in this period (Silva et al., 2014), and the other months as dry season. The mean annual rainfall (1974–2003) in Planaltina was 1346 mm, the air temperature between 16.5 °C and 27.7 °C and relative air humidity between 37.6% and 97.7% (Silva et al., 2014), according to the Climatological Standard Normals (Fig. 1a). The rainfall, mean air temperature and monthly relative air humidity of the study period are listed in Fig. 1b.

The soil of the experimental area was classified as a clayey Oxisol (Typic Haplustox) (Soil Survey Staff, 2006). The soil chemical and physical properties (0–20 cm) are shown in Table 1. According to the description of the mineralogical composition of Reatto et al. (2007), the diagnostic horizon consists of: kaolinite (320 g kg⁻¹), gibbsite (496 g kg⁻¹), hematite (142 g kg⁻¹), and goethite (42 g kg⁻¹).

2.2. History and description of the experiment

The long-term experiment for this study had been initiated 19 years earlier, in 1996. The plots (22 m × 18 m) were arranged in a randomized block design, with three replications. After cutting the natural Cerrado vegetation in 1995/1996, the soil of all plots was tilled with a disk plow for liming and with a moldboard plow for incorporation of organic residues into deeper layers; differences between the management systems established on this soil were only detected five years after tillage. For this study, two management systems (no-tillage, and conventional tillage) and a reference treatment in the area of dense Cerrado *sensu stricto* were selected (Table 2).

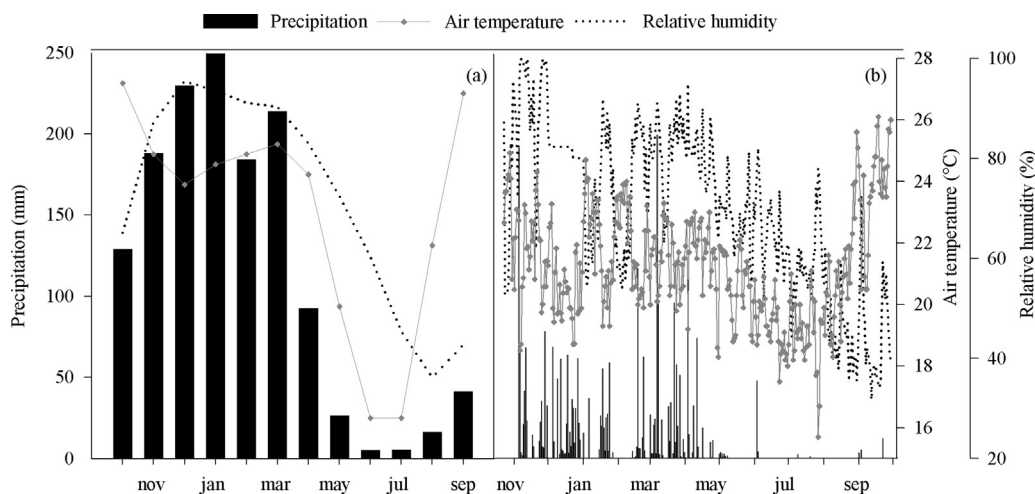


Fig. 1. Rainfall, air temperature and relative humidity in the experimental area from 1974 to 2003 (a) and during the evaluation period (b).

2.3. Measurements of nitrous oxide fluxes

The N_2O fluxes were measured for one year by the static closed chamber method (Mosier et al., 1998) from October 23, 2013, to September 29, 2014. The total N_2O fluxes were determined in 88 evaluation events, 22 of which occurred between October and December 2013, and 66 between January and September 2014.

The periodicity of collection interval was determined in function of events that are known to intensify N_2O fluxes, accordingly to Wang et al. (2011). Samples were collected twice and three times a week during the dry (April to September) and wet seasons (October to March), respectively. Regardless of the season, daily samples were taken for at least three consecutive days after rainfall or soil tillage, seeding/planting, harvest, and nitrogen fertilization.

Three static chambers were installed per plot, at a random distance from each other in the rows, and were left there for about 25 days after crop emergence and later moved to the inter-rows, due to crop growth. Each static chamber consisted of a metal base (0.38 m × 0.58 m) inserted into the soil, and an upper part of PVC with height of 9.5 cm, coated with a thermal aluminum blanket, which together with the metal base sealed the space covered by the chamber, thus forming a microenvironment, where gases are accumulated for later collection and determination. A hole was drilled in the middle of each chamber, sealed with silicone, and a rubber hose connected to a three-way valve for gas exit control was

inserted. A digital thermometer was also coupled to one of the three chambers to monitor the temperature within the chambers. Another digital thermometer was inserted into the soil to determine the soil temperature at a depth of 5 cm at the times of gas sampling. These samples were taken between 09:00 h and 11:00 h, following the recommendation of Alves et al. (2012) to best represent the daily mean flux. From within the chamber, air samples were collected 0, 15 and 30 min after closing the chamber, with 60 mL polypropylene syringes equipped with three-way valves, although samples of only 25 mL gas were collected. The vials were immediately placed in a cooler box, transported to the laboratory and stored in a refrigerated environment at 18 °C with septa face down and submerged in a sufficient distilled water layer to completely cover the aluminum seal, with maximum wait time of one to two days. Additionally, as a reference of the atmospheric air standard, one sample per block was collected.

The N_2O concentration in the air contained in the vials was determined by gas chromatography (Trace Ultra GC oven – Thermo Scientific; Milano, Italy), with a pre-column Hayesep Q[®] (Restek), an analytical column Hayesep Q[®] (Restek) and an electron capture detector. The gas chromatograph was calibrated for N_2O at four levels (concentrations of 200, 600, 1000, and 1500 ppb N_2O). Estimated limit of detection was 51 ppb and estimated limit of quantification was 154 ppb. The N_2O (FN_2O) fluxes were measured by the linear variation of gas concentration in relation to the incubation time in the sampling chambers, and calculated by Eq. (1), as proposed by Steudler et al. (1989):

$$FN_2O = (\delta C / \delta t) \times (V/A) \times m / V_m \quad (1)$$

Where FN_2O is the N flow in the form of N_2O ($\mu\text{g N-N}_2\text{O m}^{-2} \text{h}^{-1}$); $\delta C / \delta t$ is the change in N_2O concentration in the chamber during the incubation time interval, in $\text{nmol/h} \times \text{L (air)}$; V and A are respectively the chamber volume and the soil area covered by the chamber (m^2); m is the molecular weight of N_2O and V_m is the molar volume of the gas at the sampling temperature.

2.4. Soil and climate variables

At the time of gas collection, the soil was also sampled systematically for determination of NO_3^- and NH_4^+ in the soil, at a depth of 0–5 cm using a 7 cm diameter auger, at six points in-between the crop rows, to form a composite sample. An aliquot was taken from each soil sample to determine soil moisture. 15 g

Table 1

Soil chemical and physical properties of the experimental area under different management systems and native Cerrado in 2013, prior to the measurements.

Soil properties ^a	NTR1	NTR2	CT	Native Cerrado
Organic matter (g kg^{-1})	30.0	30.0	30.0	34.0
pH (H_2O)	5.5	5.0	5.5	4.7
Al^{3+} ($\text{cmol}_c \text{kg}^{-1}$)	0.4	0.4	0.2	1.4
$\text{H} + \text{Al}$ ($\text{cmol}_c \text{kg}^{-1}$)	6.4	6.6	6.6	9.9
Ca^{2+} ($\text{cmol}_c \text{kg}^{-1}$)	2.6	2.5	2.3	0.1
Mg^{2+} ($\text{cmol}_c \text{kg}^{-1}$)	1.1	0.8	0.9	0.1
P (mg dm^{-3})	17.1	20.5	16.1	1.3
K ⁺ (mg dm^{-3})	161.8	93.8	153.1	39.1
Soil density (g cm^{-3})	1.2	1.2	1.2	1.1
Clay (g kg^{-1})	468	475	483	508
Silt (g kg^{-1})	95	55	80	89
Sand (g kg^{-1})	437	470	437	403

^a Mean value in the 0–20 cm layer; NTR1 and NTR2=no-tillage system; CT=conventional tillage.

Table 2
Description of soil management systems studied and summary of the history of crop sequence on the experimental plots.

Management systems	Symbol	Description ^a
Conventional tillage with the use of heavy disking and biennial legume-grass rotation	CT	Soil tillage with heavy disking and planting of legumes only in the first two years. Later, biennial grass-legume rotation. On 10/20/2013, after soil tillage, super-early soybean BRS 6780 (≈ 100 days) was planted in 0.45 m spacing (main crop). Application of 400 kg ha ⁻¹ N-P-K fertilizer mixture (0-20-20) and seed treatment (3 mL kg ⁻¹ fungicide VITAX tyram [®] and 3 mL kg ⁻¹ insecticidal Standak [®]) and seed inoculation with <i>Bradyrhizobium japonicum</i> (strains CPAC 7 and CPAC 15). Pre-emergence herbicide Dual Gold [®] was also applied (2 L ha ⁻¹) to the soil. After soybean harvest (01/29/2014), the area was left fallow until the next crop was planted.
No-tillage with rotations biennial legume-grass and second alternate crops of grasses, legumes.	NTR1	Tillage with disk plow in the first two years and moldboard plow in the following two years. From the fifth year onwards, the no-tillage system was introduced, with biennial rotation combined with cropping sequences. In October 2013, super-early soybean (≈ 100 days) was planted as main crop, at a row spacing of 0.45 m, with the same fertilization and inoculation as applied in the CT system. After the soybean harvest on 29/01/2013, the crop residues were left on the ground. On 02/10/2014, sorghum was planted as late-season crop, at a row spacing of 0.50 m, with base fertilization of 300 kg ha ⁻¹ of the N-P-K mixture (4-30-16). A topdressing of 50 kg ha ⁻¹ N (urea) was applied on 03/09/2014. Sorghum was harvested on 06/09/2014.
No-tillage with biennial grass-legume rotations and alternating legume and grass crops.	NTR2	Tillage with a disk plow in the first two years and moldboard plow in the following two years. From the fifth year, the no-tillage management was introduced, with biennial rotation combined with cropping sequences. On 10/22/2013 a maize hybrid (main crop) as planted at a row spacing of 0.70 m. Fertilization consisted of 350 kg ha ⁻¹ of N-P-K (4-30-16) at planting and two topdressings of 70 kg ha ⁻¹ N (urea) applied 20 and 47 days after planting. After the maize harvest (03/10/2014), leaving crop residues on the soil, pigeon pea (late-season crop) was planted at a row spacing of 0.50 m on 03/12/2014 and harvested on 06/09/2014.
Native Cerrado	--	Area of dense Cerrado <i>sensu stricto</i> adjacent to the experimental area, used as a reference environment.

^a The legume and grass species, respectively, in both no-tillage treatments in the summer, were soybean and maize and as second crop in the rotations were pigeon pea (*Cajanus cajan*) after maize and sorghum BRS332 (*Sorghum bicolor* (L.) Moench) after soybean.

of the fresh soil were used for determine the content of soil mineral N, in the forms of NO₃⁻ and NH₄⁺, which was extracted with 50 mL of 2 mol L⁻¹ KCl, followed by distillation by the Kjeldahl method.

The variables mean air temperature and rainfall were recorded by an automatic weather station (Campbell Scientific) installed in the experimental area. Soil density and particles were also measured in the plots by the methods of volumetric cylinders and volumetric flask, respectively. The gravimetric soil moisture was calculated for a soil subsample by oven-drying at 105 °C for 48 h. Based on the results of soil moisture, bulk density and soil particles, the water-filled pore space (WFPS%) at each evaluation was calculated to determine the level of anoxia in the 0–5 cm layer, by equation 3:

$$WFPS = (\theta \times (BD/WD) \times 100) / [1 - (BD/PD)]$$

where: WFPS is the water-filled pore space (%); θ – gravimetric water content (g g⁻¹); BD – bulk density (g cm⁻³); WD – water density (1.0 g cm⁻³); and PD – particle density (2.65 g cm⁻³).

2.5. Statistical analysis

The data for N₂O fluxes, NO₃⁻, NH₄⁺, soil temperature, and cumulative N₂O were subjected to analysis of variance (ANOVA) and the means compared by Tukey's HSD test ($p=0.05$). Data of cumulative N₂O and soil variables were subjected to multivariate analysis (principal component analysis, PCA) to evaluate the key factors (management practices and soil variables) of soil N₂O fluxes. Only correlation coefficients (r) above 0.50 between variables and ordination axes were considered significant.

3. Results

3.1. Rainfall seasonality during crop development in the cerrado

The total precipitation from October 2013 to September 2014 was 1258 mm (Fig. 2). Of this total, 88% of the rain (1104 mm) fell in the rainy season and 12% during the dry season and in sporadic rainfalls (Fig. 2).

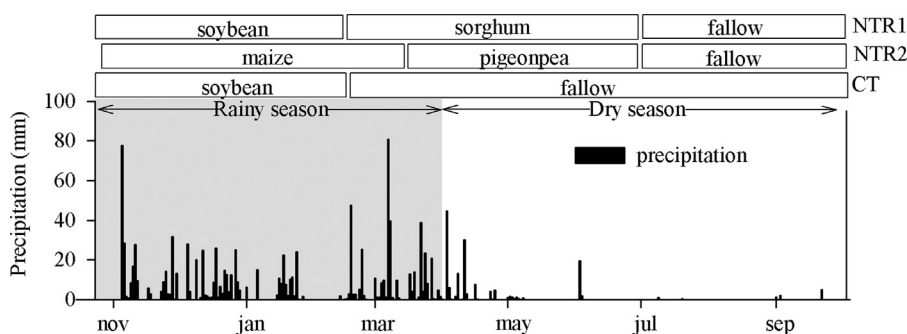


Fig. 2. Total rainfall in the experimental area in the rainy and dry seasons, during the agricultural year 2013/2014, Planaltina, DF, Brazil. NTR1 = no-tillage system with soybean and rotation with BRS332 sorghum (*Sorghum bicolor* (L.) Moench); NTR2 = no-tillage system with maize and rotation with pigeon pea (*Cajanus cajan*); CT = conventional tillage with soybean and subsequent fallow.

During the rainy season, irregularities in the total rainfall distribution were observed, with dry spells lasting longer than four days in November, January and February (Fig. 2). In February, a spell of over 17 days was observed, with no more than 3 mm rainfall.

During the crop cycles of soybean, maize, sorghum, and pigeon pea, the total rainfall was 713 mm; 959 mm; 535 mm; and 280 mm, respectively.

3.2. Daily and seasonal N₂O emissions and soil and climate variables in the cerrado agroecosystems

The Cerrado had the lowest emission peaks of soil N₂O, and 56% of the total of peaks of this system were below 5 μg m⁻² h⁻¹, with

influxes in some measurements. In this system, no significant differences in soil N₂O emissions caused by rainfall seasonality were detected in the study period (Fig. 3a).

Distinct from the Cerrado, the daily and seasonal soil N₂O emissions from the agroecosystems varied (Fig. 3a). The fluxes observed in the management systems ranged from 266 μg m⁻² h⁻¹ in the rainy season to minimum values of 0.0 μg m⁻² h⁻¹ in the dry season (Fig. 3a). The soil N₂O fluxes from the management systems increased particularly after N fertilization and WFPS in the rainy season. In the dry season, the occurrence of occasional rainfalls (>20 mm in June) promoted soil rewetting (Fig. 3b) after prolonged intervals without rain, which favored N₂O emissions associated with an increase in nitrate levels and soil temperature (Fig. 3a, c and e).

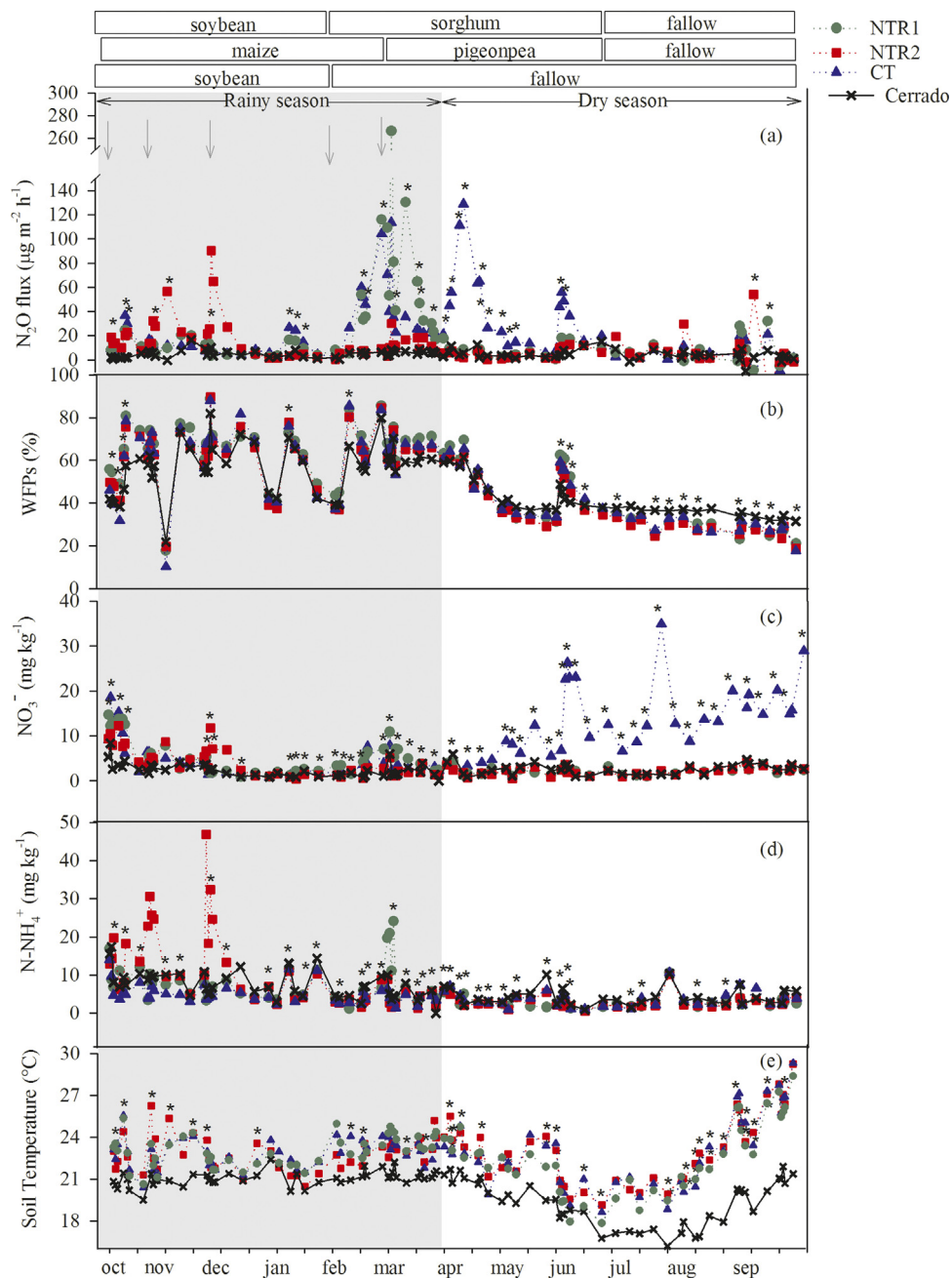


Fig. 3. Daily flows of nitrous oxide (a), water-filled pore space – WFPS (b), nitrate – NO₃⁻ (c), ammonium NH₄⁺ (d), and soil temperature (e) in the agricultural year 2013/2014 under no-tillage systems (NTR1 and NTR2), conventional (CT) and native Cerrado Oxisol, in the rainy and dry season, Planaltina, DF, Brazil. Arrows indicate nitrogenous fertilizer applications. * Significant difference by the Tukey test (P <0.05) between management systems in the study period.

Nine days after the first N topdressing of maize (11/11/2013) in NTR2, the fluxes reached $56 \mu\text{g m}^{-2} \text{h}^{-1}$. In the same period, the systems NTR1 and CT with soybean emitted $9 \mu\text{g m}^{-2} \text{h}^{-1}$, respectively, while a value of $0.0 \mu\text{g m}^{-2} \text{h}^{-1}$ was measured in the native Cerrado. In other words, N topdressing of maize induced 6.2 times higher emissions than that of soybean. After the second N topdressing of maize (12/08/2013), increasing fluxes from NTR2 occurred four days after the second N topdressing, when a peak of $90.5 \mu\text{g m}^{-2} \text{h}^{-1}$ was observed, while the values were below $4 \mu\text{g m}^{-2} \text{h}^{-1}$ in the other agricultural systems. Also after the second topdressing, fluxes from maize in NTR2 were 22.5 higher than from the other treatments. In the interval between the second N fertilization and the peak emission of $90.5 \mu\text{g m}^{-2} \text{h}^{-1}$, 46 mm of rainfall was recorded. The day before this field evaluation, 24 mm rainfall and WFPS close to 90% were reported, resulting in anaerobic conditions and promoting denitrification (Fig. 3b).

On the third day after maize harvest, and two days after planting pigeon pea in NTR2, a peak emission of $30 \mu\text{g m}^{-2} \text{h}^{-1}$ was measured, however throughout the crop cycle, the emission peaks were no higher than $19 \mu\text{g m}^{-2} \text{h}^{-1}$ (Fig. 3a). In this treatment, two peaks of 30 and $54 \mu\text{g m}^{-2} \text{h}^{-1}$ occurred in the second growing season on, respectively, 08/05/2014 and 09/09/2014.

With regard to the flow dynamics in the management systems NTR1 and CT during the soybean growth cycle, N_2O fluxes also were also highest after events such as planting and harvesting. The highest N_2O flux during early soybean development in CT occurred 10 days after planting, on 10/30/2013, reaching a value of $36 \mu\text{g m}^{-2} \text{h}^{-1}$, i.e., 18 times higher than emissions from native Cerrado ($2 \mu\text{g m}^{-2} \text{h}^{-1}$). On January 20 and 23, 2014, N_2O peaks were recorded only in the management systems with soybean (NTR1 and CT). The NTR1 on the above two days emitted a mean flux of $15 \mu\text{g m}^{-2} \text{h}^{-1}$ and CT a mean of $25 \mu\text{g m}^{-2} \text{h}^{-1}$, i.e., 40% higher than from NTR1. However, emissions were most significantly affected by the soybean residues, as can be seen in CT and NTR1 in the months after harvest on 01/29/2014 (Fig. 3a).

During the fallow period of the CT system after soybean harvest, N_2O peaks were observed with increased emissions between February and June. In the months after soybean harvest, mean fluxes of 60, 113, 128, 23 and $56 \mu\text{g m}^{-2} \text{h}^{-1}$ were measured in February, March, April, May and June, respectively. After 38 days, the fluxes from soybean increased from $4 \mu\text{g m}^{-2} \text{h}^{-1}$, on 02/03/2014, with WFPS of 42%, to $113 \mu\text{g m}^{-2} \text{h}^{-1}$ on 03/12/2014, with WFPS of 67% ($P < 0.05$). In the same period, regular rains were measured in six days, with a total of 136 mm. The effect of soil rewetting on 04/16/2014 (rainfall > 7 mm) was also observed for CT, followed by a peak of $128 \mu\text{g m}^{-2} \text{h}^{-1}$ on the next day. The effect of rewetting was also observed on 06/05/2014 when 21.2 mm rainfall during the previous two days resulted in a peak of $56 \mu\text{g m}^{-2} \text{h}^{-1}$, which probably contributed to denitrification of the N stored in the form of NO_3^- , found at levels exceeding 25 mg kg^{-1} (Fig. 3a and c). At the end of the dry season, the NO_3^- content of soil under no-tillage (NTR1 and NTR2) and native Cerrado did not exceed 2.7 mg kg^{-1} , while levels of 29 mg kg^{-1} were observed in CT in the same period (Fig. 3c).

After soybean harvest, N_2O emissions were higher from NTR1 than CT. After this harvest, emissions from NTR1 were $4 \mu\text{g m}^{-2} \text{h}^{-1}$ on 02/03/2014 and increased to $266 \mu\text{g m}^{-2} \text{h}^{-1}$ on 03/12/2014 three days after sorghum topdressing, when increases in soil NO_3^- content from 7 mg kg^{-1} to 11 mg kg^{-1} , and in NH_4^+ content from 6 mg kg^{-1} to 20 mg kg^{-1} were also identified (Fig. 3c and d). These mineral N values observed in NTR1 system were 45% and 40% higher than those determined in Cerrado and CT soil, respectively, on the same day. Excluding the effect of N fertilization, applied to sorghum in NTR1, the N_2O fluxes from the other agroecosystems varied from 33 to $115 \mu\text{g m}^{-2} \text{h}^{-1}$ and from -9 to $16 \mu\text{g m}^{-2} \text{h}^{-1}$ in the Cerrado.

3.2.1. Seasonal variations of WFPS, mineral nitrogen (NO_3^- and NH_4^+) and soil temperature

The differences in WFPS between the systems studied were significant in 22% of the measurements ($P < 0.05$). During the rainy season, the WFPS of the agroecosystems reached 90% in November. However, WFPS of less than 30% during dry spells during the crop growing season were also recorded (Fig. 3b). In the dry season, the lowest WFPS percentage was 18% (Fig. 3b). In early June after 19 mm of rainfall, WFPS reached more than 60%, and was maintained for 22 days. At the end of September, WFPS values of up to 18% were observed in the dry period in CT only, while the mean was 20% in the other systems (NTR1 and NTR2).

In the rainy season, small variations of soil NO_3^- occurred after N fertilization of maize in NTR2, reaching levels of up to $12 \text{ mg NO}_3^- \text{ kg}^{-1}$ on 12/12/2013, while in the other management systems, the levels were below 2 mg kg^{-1} in the same period. In general, the soil NO_3^- content varied from 0 to 35 mg kg^{-1} between the agroecosystems and native Cerrado. The levels and variations of NO_3^- were highest in CT in the dry period (Fig. 3c). Despite high levels of soil NO_3^- in CT in the dry season, the N_2O peaks observed during this period were restricted to the incidence of minor rainfalls (< 19 mm) which occurred occasionally in June. From July onwards, in spite of the increase in soil NO_3^- concentrations in CT, which lasted until September, no new N_2O emission peaks were observed (Fig. 3a, c). For the native Cerrado, the soil NO_3^- levels remained relatively low and constant, regardless of the seasonal period, with a similar mean content in the rainy as in the dry season (2.5 mg kg^{-1}) (Fig. 3c).

For the soil NH_4^+ content, in the systems studied, differences were observed in 60% of the data ($P < 0.05$). Unlike NO_3^- , the highest levels and variations of soil NH_4^+ occurred in the rainy season (Fig. 3d). In NTR2, levels of up to $46.8 \text{ mg NH}_4^+ \text{ kg}^{-1}$ were observed in December (Fig. 3d). In NTR1 in March, an increase in soil NH_4^+ levels was also observed. Except for the days just after of N fertilization events in all agroecosystems, NH_4^+ values between 0.5 and 16 mg kg^{-1} were observed. In native Cerrado, the soil NH_4^+ content was 30 and 56% higher than NO_3^- levels in the rainy and dry seasons, respectively (Fig. 3d).

Soil temperature ranged from a minimum of 17°C in the native Cerrado to a maximum of 29°C in CT (Fig. 3e). In the rainy season, soil temperatures in March were highest in the agricultural areas, averaging 24°C in NTR1, or 3°C above the mean of this month in the native Cerrado, while the other systems (NTR2 and CT) averaged 23°C . In the dry season, soil temperatures were lowest in July in all management systems and highest in September (Fig. 3e). In general, the soil temperature of native Cerrado was lower ($1-6^\circ\text{C}$) than in the agricultural systems. In July and August, no significant differences in N_2O peaks were observed, and soil temperatures of maximally 21°C (Fig. 3e). Associated with N_2O peaks, soil temperatures above 23°C and 19°C were recorded in the wet and dry seasons, respectively (Fig. 3a and e). In the last assessments at the end of the dry season, soil temperatures were 4°C lower in native Cerrado than in the other management systems (mean of 29°C) (Fig. 3d). As of August, progressive increases in soil temperature were observed in all evaluated systems, and increases in NO_3^- contents in CT only.

3.3. Effects of rainfall seasonality and crop rotation on cumulative N_2O emissions from agroecosystems

The cumulative soil N_2O emissions were influenced by the seasonality of rainfall, management systems, crop rotation, as well as by the interactive effects between these factors (Fig. 4). The annual cumulative N_2O emissions were ranked in decreasing order: CT (1.36 kg ha^{-1}) = NTR1 (1.0 kg ha^{-1}) \geq NTR2 (0.70 kg ha^{-1}) \geq Cerrado (0.28 kg ha^{-1}).

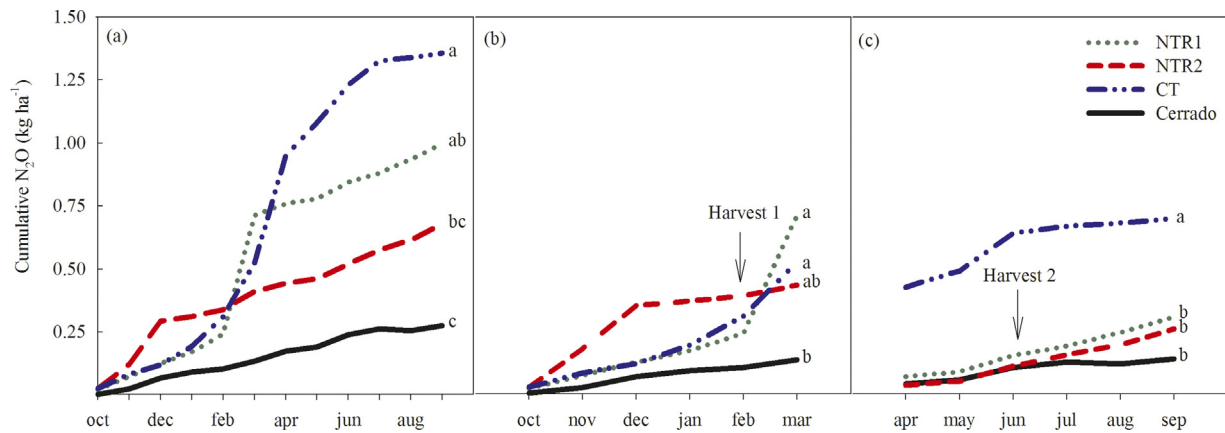


Fig. 4. Annual cumulative soil emissions of nitrous oxide (a), in the rainy (b) and the dry season (c) under no-tillage (NTR1 and NTR2), conventional tillage (CT) and native Cerrado, Planaltina, DF, Brazil. NTR1 = no-tillage system with soybean and rotation with BRS332 sorghum (*Sorghum bicolor* (L.) Moench); NTR2 = no-tillage system with maize and rotation with pigeon pea (*Cajanus cajan*); CT = conventional tillage system with soybean and late-season fallow. Arrows indicate harvests of legumes, soybean (Harvest 1) and pigeon pea (Harvest 2). Treatments followed by the same letter do not differ statistically by Tukey's test at $P < 0.05$.

During the rainy season, N_2O emissions from the agroecosystems were similar to each other and higher than those from the native Cerrado, except for NTR2 ($P < 0.05$). In the dry period, CT promoted higher cumulative N_2O emissions than the other systems ($P < 0.05$), with no differences between the native Cerrado and no-tillage systems (NTR1 and NTR2). Of the total cumulative emissions from CT, approximately 50% occurred in the dry season and 75% was accumulated during the fallow period.

Increments in cumulative N_2O after harvesting the first and second crops were observed in the agricultural ecosystems (Fig. 4a, b and c). For the first harvest, the cumulative N_2O emissions from the agroecosystems ranged from 0.23 to 0.48 $kg\ ha^{-1}$. In NTR2, maize (main crop) accumulated the highest N_2O emissions (0.48 $kg\ ha^{-1}$), i.e., twice as high as emissions from the systems NTR1 and CT. For the late-season crops in NTR2, pigeon pea accumulated only 0.15 $kg\ ha^{-1}$, while sorghum in NTR1 accumulated 0.81 $kg\ ha^{-1}\ N_2O$.

3.4. Relations between soil N_2O fluxes and soil and climate variables of management systems due to the rainfall seasonality

Two principal components were generated (PC1 and PC2) as tools for the distinction of management systems, considering all

variables together (NO_3^- , NH_4^+ , WFPS, soil temperature, and N_2O emission), for the rainy (Fig. 5a) and dry season (Fig. 5b). The distribution of selected variables showed a cumulative variance of 56.16% and 53.04% for the sum of the principal components PC1 and PC2 for the rainy and dry season, respectively.

In the rainy season, the first principal component (PC1) had the highest correlation with N_2O emission (0.59), NO_3^- content (0.74), NH_4^+ (0.48), and with soil temperature (0.59) (Table 3). For the same period, the principal component 2 (PC2) represented the variability of WFPS better, with a ratio of 0.84. In this context, two groups of positive correlations were formed in the rainy season. One group comprised N_2O emission and NH_4^+ and the other group soil temperature and NO_3^- (Fig. 5a). However, the two groups formed during the rainy season were independent of WFPS, with a low correlation.

For the dry season, the first principal component (PC1) had the highest positive correlation with N_2O emission and WFPS, with correlation coefficients (r) of 0.71 and 0.83, respectively (Table 3). For the second principal component (PC2) in this season, a positive relation of NO_3^- (0.63), NH_4^+ (0.70) and soil temperature (0.49) was observed. Thus, contrary to observations for the rainy season, N_2O emission and WFPS were strongly correlated in the dry season,

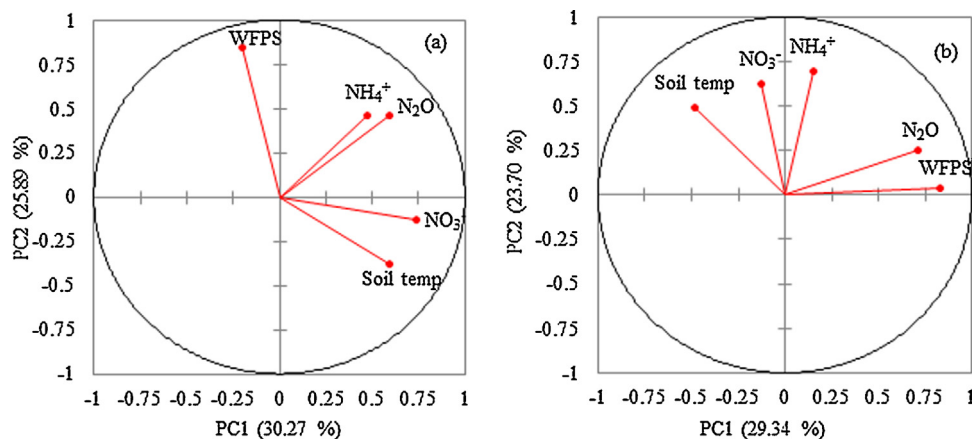


Fig. 5. Principal component analysis (PCA) in the rainy season (a) and dry season (b) for N_2O fluxes from soil and soil-climate variables: nitrate – NO_3^- , ammonium – NH_4^+ , water-filled pore space – WFPS and soil temperature (soil temp.) under different management systems and native Cerrado during the six months of each season.

Table 3

Correlation coefficients between the soil and climate variables and the principal components (PC1 and PC2), in relation to the rainfall seasonality (rainy and dry periods).

Variables	Rainy season		Dry season	
	PC1	PC2	PC1	PC2
Eigen value	1.51	1.29	1.47	1.18
% Explained	30.27	25.89	29.34	23.70
N ₂ O	0.59	0.46	0.71	0.25
NO ₃ ⁻	0.74	-0.13	-0.13	0.63
NH ₄ ⁺	0.48	0.46	0.16	0.70
WFPS	-0.20	0.84	0.83	0.04
Soil Temp.	0.59	-0.38	-0.49	0.49

forming one group, and the NO₃⁻ and NH₄⁺ contents, together with the soil temperature, another group (Fig. 5b).

4. Discussion

4.1. Effects of rainfall seasonality, tillage systems and crop rotation on N₂O fluxes

For the native Cerrado, the mean annual flow of 5 μg N₂O m⁻² h⁻¹, observed in this study, was far lower than the mean emission of 13 μg N₂O m⁻² h⁻¹ observed in other savannas and hot climate ecosystems (Castaldi et al., 2006). In general, Cerrado ecosystems are conservative systems in terms of N, which limits the supply of this nutrient and losses by decomposition (Bustamante et al., 2009). Furthermore, characteristics of the Cerrado such as high C/N ratio (~60) in the vegetation, predominance of NH₄⁺ in relation to NO₃⁻ are factors that contribute to keep the quantity of N low in the system (Bustamante et al., 2012). These factors combined with high porosity and hydraulic conductivity may limit the N₂O formation processes (Martins et al., 2015). In areas under natural forests such as the Amazon, the N₂O fluxes increase in the rainy and decrease in the dry season and may be five times higher (Corre et al., 2014; Bai et al., 2014). In tropical forests in north-eastern Australia, peaks of up to 242 μg N₂O m⁻² h⁻¹ were observed during the rainy season, while in the dry period, the values were below 20 μg N₂O m⁻² h⁻¹ (Kiese and Butterbach-Bahl, 2002). In general, in native forests with well-defined seasonal periods, there is a balance between N inputs and outputs by the decomposition of organic material (Bustamante et al., 2009), which can make the variations in N₂O emissions more dependent on environmental variables.

However, agriculture is admittedly the major driver of global N₂O emissions, especially when using nitrogen fertilization (Baumert et al., 2005). In this study, the daily and seasonal variations in agroecosystems were greater in relation to N₂O emissions from native Cerrado. Among the systems studied, N₂O emission rates ranged from 0.0 μg m⁻² h⁻¹ to a maximum of 266 μg m⁻² h⁻¹ (Fig. 3a). These values were similar to those reported in several studies (~3.5 to 357 μg m⁻² h⁻¹) carried out in Brazil (Metay et al., 2007; Jantalia et al., 2008; Gomes et al., 2009; Bayer et al., 2015; Martins et al., 2015).

During the rainy season, the highest N₂O peaks were observed after applications of N fertilizer to cereal (maize and sorghum), while in the case of legumes (soybean), which did not receive nitrogen fertilizer, emissions were highest immediately after crop harvest, when the N-rich crop residues were used by microorganisms, with subsequent N₂O release to the atmosphere (Fig. 3a, c and d). Firstly, the organic substrate is decomposed and releases N in mineral form for plant uptake. In the case of CT however, no second crop was implemented after soybean, which is a legume with a low C: N ratio and readily available symbiotic nodules for decomposition after senescence, so in the case of NTR1,

sorghum had been fertilized with a readily available N source. Seemingly, an excess of available N in mineral and organic form, along with the environmental conditions of WFPS of 35–60% induces decomposition and denitrification processes with reduction of nitrogenous compounds to gaseous N forms, including N₂O (Khalil and Baggs, 2005). Close to soybean harvest, the volume of senescent leaves and crop residues on the ground is large and in addition, the proper senescence of root nodules may also have contributed to the short-term effect with higher N₂O peaks in the CT system. Similar results with short-term effects were described by Dyer et al. (2012). However, Bayer et al. (2015) concluded that the rapid decomposition of plant residues did not affect N₂O emissions from the CT system, and explained that the crop rotation effect may have promoted a dilution of N mineralization in the topsoil.

The highest emissions in the two months after soybean harvest, about 1.3 times higher from NTR1 than from CT, may be related to the higher concentrations of labile C, a product of nodular senescence, which is consumed and used as growth substrate of microbial populations, thus favoring nitrifying and denitrifying soil microorganisms (Dyer et al., 2012). In crop rotations under subtropical conditions, Bayer et al. (2015) found no effect of tillage and conventional systems on N₂O emissions in the post-harvest period.

Long-term management systems affect N₂O emissions with short and long-term effects. For example, the organic residues deposited on the soil promote increases in emission rates in the short term. Later, probably when the most labile fraction of the residues was already decomposed (Baggs et al., 2003), emissions return to lower values (Fig. 3a). A rapid decline in emissions after N fertilization was also observed in several field studies (Martins et al., 2015; Aini et al., 2015). This may be a consequence of the high plant N demand, thus reducing the chances of mineral N losses. However, different crop rotations promote increases in certain soil properties over the years, e.g., in C and N soil, stocks, as reported by Baggs et al. (2006) and Gomes et al. (2009) in tropical and subtropical regions, respectively.

4.2. Soil-climate variables

The temporal dynamics of N₂O emissions can be attributed to differences between management systems and their soil and climate conditions. In the rainy season, the WFPS varied between 50 and 70%, with high daily variations, so that both the nitrification and denitrification may have controlled the N₂O emissions. A similar situation was observed by Ball et al. (2014) in a two-year study with crop rotation. The WFPS controls N₂O emissions by its effect on the nitrification and denitrification processes and gas transmission into the soil (Neill et al., 2005). Many studies emphasized that soil moisture, expressed by WFPS, the soil temperature and mineral N content, are the key variables that define N₂O emissions (Ball et al., 2014; Bayer et al., 2015).

In this study, the correlation between N₂O fluxes and rainfall can be explained by the short time within which water reaches the soil and fills the voids, creating anaerobic conditions that stimulate denitrifying bacteria (Butterbach-Bahl et al., 2013). As the N₂O diffusivity is low in soils with a high volume of micropores (Eickenscheidt and Brumme, 2013), a characteristic property of loamy soils, peak emissions are recorded only a few hours or days after a rainfall (Dick et al., 2001). The control mechanisms of these flows in the dry season are not completely understood yet; the dry soil conditions possibly improve N₂O diffusion from the atmosphere into the soil under limited availability of inorganic N, and atmospheric N₂O is reduced to N₂ by denitrifying bacteria (Dijkstra et al., 2013), thus reducing N₂O emissions in the dry period.

Observing the effect of seasonality (Fig. 2), some N₂O fluxes in the soil occurred in the dry season, a result that may be a consequence of the effect that takes place in drying/rewetting events, in which rewetting after long drought promotes high rates of plant decomposition and rapid soil mineralization, with declining intensity over time after rewetting (Jarvis et al., 2007). This behavior is characteristic of the region, with occasional rains throughout a long drought period of up to 5 months, promoting an intense response in microbial activity. The addition of water rapidly increases the population and metabolism of microorganisms in the soil (Jarvis et al., 2007), and can explain the relationship between rewetting and soil N₂O emissions. Similarly, Pelster et al. (2011) studied alfalfa and soybean in no-tillage and conventional systems, and observed highest N₂O peaks after soil rewetting.

The low soil N₂O fluxes from soil under natural Cerrado vegetation recorded in this study, regardless of the seasonal period, can also be explained by soil properties such as high soil drainability, high acidity (Castaldi et al., 2006; Martins et al., 2015) and low N availability (Bustamante et al., 2009). In fact, the variations in NO₃⁻ and NH₄⁺ contents in the native Cerrado were low (<10 mg kg⁻¹) during most of the study period, which can explain these low N₂O emissions (Fig. 3a, c and d). In the first week of June, N₂O fluxes from CT were higher than from the other systems, which was associated with the high NO₃⁻ contents (around 26 mg kg⁻¹) in the presence of water in the soil (WFPS around 59%). In this situation, the conditions for the occurrence of denitrification were ideal, since values above 10 mg NO₃⁻ kg⁻¹ were reported to inhibit the conversion of N₂O to N₂, since NO₃⁻ is preferred over N₂O as electron acceptor (Chapuis-Lardy et al., 2007).

The significant differences between management systems in 66% of soil temperature data observed in this study are similar to findings of other authors, who reported direct effects of soil temperature on N₂O emissions (Li et al., 2013). However, it is worth mentioning that the emissions are the result of combined effects of soil temperature with other variables such as WFPS and mineral N. Since the period of higher temperatures (T > 25 °C) usually coincides with events such as planting and fertilization, Chirinda et al. (2010) emphasized a possible masking of the isolated effect of temperature.

4.3. Interactive effect of rainfall seasonality with soil N₂O emissions and soil and climatic variables

The evaluations of daily fluxes showed significant differences between agroecosystems and that the highest emissions were not the result of a single influence but from interrelated effects.

The relations established between the variables soil temperature, WFPS and NO₃⁻ with soil N₂O flux in the two seasons indicated that N₂O production was stimulated mainly by the high WFPS values after soil moisture increase (Zhu et al., 2013). Normally, an increase in WFPS also leads to an increased consumption of soil O₂ by the microbial activity, with oxidation of labile organic carbon and consequent formation of anaerobic sites (Khalil and Baggs, 2005). Higher N₂O emissions were observed by Pimentel et al. (2015), with WFPS of at least 70% under laboratory conditions. In a field experiment, Escobar et al. (2010) obtained similar results under subtropical conditions, and Martins et al. (2015) under tropical conditions in the Brazilian Cerrado.

Another noteworthy factor, common to both seasons studied, suggesting an interference with the occurrence of major N₂O fluxes, is the contribution of high soil temperatures along with increasing soil moisture. Liu et al. (2013) explained the weak relationship between N₂O emissions and soil temperature by changes caused by low soil moisture, reducing WFPS by 50%. In

soils with 70–90% WFPS, soil N₂O emissions result mostly from denitrification (Granli and Bockman, 1994; Davidson et al., 2000). Likewise, in the dry seasons, soil rewetting at ideal concentrations of mineralized compounds accelerates the microbial activities and will therefore also increase N₂O emissions (Davidson et al., 1992).

The installation time of management systems in tropical climates should also be considered to validate the N₂O emission data. Studies by Six et al. (2004) in humid climates indicate that N₂O emissions are lower from long-term (>10 years) no-tillage areas. Under tropical conditions, differences were observed between the no-tillage systems (NTR1 and NTR2), related to the species of crop rotation used in each tillage system, reflecting the short-term emissions in response to the management. However, in the conventional system with legume monocultures, variations were wide and fluxes higher, particularly at higher soil moisture.

5. Conclusions

Our results indicate that under natural conditions, the Cerrado is a conservative system in terms of N, since the soil N₂O emissions were not affected by seasonal differences or by rain events. In the agroecosystems studied, the cumulative soil N₂O emissions were influenced by rainfall seasonality, management systems, crop rotation, as well as by the interaction between these factors. The integrated NT systems, compared to CT under soybean monoculture, contributed to mitigate N₂O emissions. The definition of NT as a mitigating system of N₂O emissions depends on the species used for crop rotation. The crop rotation used in NTR2 (maize-pigeon pea) was most efficient, with lower emission peaks than in NTR1 (soybean-sorghum). Therefore, the definition of a system for N₂O flux reduction should be based on the crop type (grass/legume) for crop rotation and the maximized use of available N in the system.

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