

# NITROGEN FERTILIZATION ( $^{15}\text{NH}_4\text{NO}_3$ ) OF PALISADEGRASS AND RESIDUAL EFFECT ON SUBSEQUENT NO-TILLAGE CORN<sup>(1)</sup>

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## SUMMARY

Nitrogen is required in large amounts by plants and their dynamics in corn and perennial forages intercropped is little known. This study analyzed the efficiency of nitrogen fertilization ( $^{15}\text{NH}_4\text{NO}_3$ ) applied after corn grain harvest to palisadegrass (*Brachiaria brizantha* cv. Marandu) in intercrops sown at two times, as well as the N residual effect on the subsequent corn crop. The field experiment was performed in Botucatu, São Paulo State, in southeastern Brazil, on a structured Alfisol- under no-tillage. The experiment was arranged in a randomized block design in a split plot scheme with four replications. The main plots consisted of two intercropping systems (corn and palisadegrass sown together and palisadegrass sown later, at corn top-dressing fertilization). The subplots consisted of four N rates (0, 30, 60, and 120 kg ha<sup>-1</sup> N). The subplots contained microplots, in which enriched ammonium nitrate ( $^{15}\text{NH}_4\text{NO}_3$ ) was applied at the same rates. The time of intercrop sowing affected forage dry matter production, the amount of fertilizer-derived N in and the N use efficiency by the forage plants. Nitrogen applied in autumn to palisadegrass intercropped with corn, planted either at corn sowing or at N top-dressing fertilization, increased the forage yield up to a rate of 60 kg ha<sup>-1</sup>.

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The amount of fertilizer-derived N by the forage plants and the fertilizer use efficiency by palisadegrass were highest 160 days after fertilization for both intercrop sowing times, regardless of N rates. Residual N did not affect the N nutrition of corn plants grown in succession to palisadegrass, but increased grain yield at rates of 60 and 120 kg ha<sup>-1</sup> N, when corn was grown on palisadegrass straw from the intercrop installed at corn fertilization (top-dressing). Our results indicated that the earlier intercropping allowed higher forage dry matter production. On the other hand, the later intercrop allowed a higher corn grain yield in succession to N-fertilized palisadegrass.

**Index terms:** *Zea mays*, *Brachiaria brizantha*, crop system, sustainability.

**RESUMO: ADUBAÇÃO NITROGENADA (<sup>15</sup>NH<sub>4</sub>NO<sub>3</sub>) NO CAPIM-MARANDU E EFEITO RESIDUAL NO MILHO, EM SISTEMA PLANTIO DIRETO**

O nitrogênio é requerido em grandes quantidades pelas plantas e sua dinâmica no consórcio entre milho e forrageiras tropicais perenes é pouco conhecida. Objetivou-se avaliar a eficiência de utilização da adubação nitrogenada (<sup>15</sup>NH<sub>4</sub>NO<sub>3</sub>) no capim-marandu (*Brachiaria brizantha* cv. Marandu), proveniente de duas épocas de consórcios com o milho, realizadas após a colheita da cultura granífera, bem como o efeito residual no milho cultivado em sucessão. O experimento foi conduzido na Faculdade de Ciências Agrônomicas da UNESP em Botucatu, SP, em Nitossolo Vermelho sob sistema plantio direto. O delineamento experimental foi em blocos casualizados em parcelas subdivididas. As parcelas foram constituídas pelas épocas de consórcio: milho e capim-marandu semeados simultaneamente; e capim-marandu semeado na adubação de cobertura do milho. As subparcelas foram compostas pelas doses de 30, 60 e 120 kg ha<sup>-1</sup> de N, aplicadas no capim-marandu após a colheita da cultura do milho. Nas subparcelas, foram alocadas microparcelas para aplicação de nitrato de amônio enriquecido (<sup>15</sup>NH<sub>4</sub>NO<sub>3</sub>) nessas mesmas doses. A época de implantação do consórcio influenciou a produção de massa de matéria seca da forrageira, a quantidade de N na planta proveniente do fertilizante e a eficiência de utilização do N pelas plantas forrageiras. A aplicação de N no outono no capim-marandu, implantado por meio do consórcio com o milho, tanto na semeadura quanto na adubação nitrogenada de cobertura, proporcionou aumento de produtividade da forrageira até a dose de 60 kg ha<sup>-1</sup>. Os maiores acúmulos de N e a eficiência de utilização do fertilizante pelo capim-marandu, oriundo de ambas as épocas de consórcio, ocorreram aos 160 dias após a fertilização, independentemente das doses de N. O residual das doses de N, aplicadas no capim-marandu, não interferiu na nutrição nitrogenada do milho em sucessão, porém incrementou a produtividade de grãos nas doses de 60 e 120 kg ha<sup>-1</sup> de N, quando o cereal foi cultivado sobre palhada proveniente do consórcio implantado na adubação de cobertura do milho. Com base nos resultados, pode-se inferir que a implantação antecipada do consórcio proporciona maior produtividade de massa de matéria seca da forrageira, enquanto quando implantado mais tarde propicia maior produtividade de grãos do milho em sucessão à forrageira adubada com N.

*Termos de indexação:* *Zea mays*, *Brachiaria brizantha*, sistema de produção, sustentabilidade.

## INTRODUCTION

The greatest limitation to the sustainability of no-tillage systems (NTS) in dry-winter regions is the low straw production by green manure and cover crop as well as grain crop species, during autumn/winter and winter/spring, due to unfavorable weather conditions such as low water availability and temperature (Crusciol et al., 2012). Moreover, due to the high decomposition rate of straw of grain crops, especially legumes, and the high probability of crop failure in the late summer, many areas may be left fallow for up to seven months a year with low vegetation cover, which decreases the viability and sustainability of NTS in these regions.

In order to minimize these problems, intercropped cultivation of commercial crops, such as corn (Borghi et al., 2012; Borghi et al., 2013a; Crusciol et al., 2013), soybean (Crusciol et al., 2012, 2014), rice (Carvalho et al., 2011), and sorghum (Crusciol et al., 2011; Mateus et al., 2011; Borghi et al., 2013b) are becoming more and more frequent in Brazil, especially with palisadegrass (*Brachiaria brizantha* cv. Marandu) (Kluthcouski et al., 2003) of which corn is the most widely used for grain crop (Cobucci et al., 2001; Freitas et al., 2008).

The corn plant has many advantages in competing with other species, mainly due to its elevated rate of dry matter accumulation in the early stages of development and to its high capacity of

photosynthetically-active radiation interception (Silva et al., 2004; Borghi et al., 2007). Conversely, palisadegrass has a slow initial growth, which reduces competition during early corn development (Alvarenga et al., 2006; Borghi et al., 2007; Lara Cabezas & Padua, 2007; Freitas et al., 2008; Valle & Pagliarini, 2009). However, after corn harvest (April-May), even though temperature, rainfall and photoperiod gradually decrease until the winter solstice, palisadegrass grows well because of its deep root system and considerably higher drought tolerance compared to grain-producing species. For this reason, palisadegrass fast accumulates dry mass and spreads over the ground during the off-season (Borghi et al., 2007; Garcia et al., 2008). Besides, due to its perennial habit, palisadegrass also has high regrowth ability and begins to grow quickly after the first spring rains (Valle & Pagliarini, 2009; Ferreira et al., 2010; Borghi et al., 2012; Crusciol et al., 2012; Borghi et al., 2013b).

It worth mentioning the possible competition for nitrogen (N) between intercropped species in corn-grass intercrops, which may compromise corn and/or forage yield. However, corn grain yield is not always limited by intercropping (Cobucci et al., 2001; Freitas et al., 2008; Borghi et al., 2012; Borghi et al., 2013a; Crusciol et al., 2013). It is therefore still unclear whether the amount of N fertilizer applied to corn is sufficient to provide residual N to meet the demand of the subsequent forage. The recommendations for N fertilizer published in the fertilization tables are destined to meet the needs of grain crops in monoculture, whereas to date there are still no N fertilizer recommendations for the intercropping system under study. In this sense, Lara Cabezas & Pádua (2007) observed a low amount of fertilizer-derived N in the forage plant (FDNFP), recovered during simultaneous development of the species (on average 1.5 %). Most of the total nitrogen applied was recovered by corn (50.8 % of total N), suggesting differences in N uptake by the two intercropped crops due to competition.

Additionally, the viability of N application to the forage after corn harvest as well as its effect on the subsequent crop is still questionable. Cover crop cultivation during the off-season, prior to corn crops in NTS, may be a promising alternative to nitrogen supplementation for the grain crop (Gonçalves et al., 2000; Silva et al., 2006b). The quality and amount of plant residues and the availability of mineral N in the soil solution directly influence nitrogen recovery from these residues by corn (Ceretta et al., 2002; Lara Cabezas et al., 2004; Ernani et al., 2005). Thus, Aita et al. (2006) and Silva et al. (2006a) stated the possibility of reducing the amount of mineral N applied to corn when grown after cover crops.

However, the efficiency of preceding crops as N source to subsequent crops is low, rarely exceeding 20 % in the first crop after application (Harris & Hesterman, 1990; Scivittaro et al., 2000). For Campos

(2004) and Lara-Cabezas (2011), the recovery of nitrogen applied under field conditions may vary from 30-90 % of the total N applied. These variations suggest that it is often difficult to determine the source of this nutrient in N dynamics studies in a soil-plant system (Scivittaro et al., 2000). The use of  $^{15}\text{N}$ -labeled nutrient sources allows an accurate and efficient quantification of N utilization and donor source (Muraoka et al., 2002), contributing to decision making with regard to the N management. Additionally, there is still little information about the efficiency of N fertilization on corn-forage intercropping and its effects on the subsequent crop (Lara-Cabezas, 2011). The changes caused by N fertilization in the soil-plant system should therefore be investigated, with regard to the amount of N absorbed by the crops throughout the plantation-livestock farming integration system.

Considering this, it was hypothesized that the efficiency of N utilization by forages would depend on when the intercropping is implemented. Furthermore, an increase in the N rate applied to palisadegrass, following corn harvest, would increase forage dry matter production as well as partially supply N to the subsequent corn crop.

This study evaluated the efficiency of nitrogen fertilization ( $^{15}\text{NH}_4\text{NO}_3$ ) after grain corn harvest on palisadegrass (*Brachiaria brizantha* cv. Marandu), for two intercrop sowing times, as well as the nitrogen residual effect on the subsequent corn crop.

## MATERIAL AND METHODS

### Study area

The experiment was carried out in the growing seasons of 2004/05 and 2005/06 in Botucatu, São Paulo State, in southeastern Brazil (48° 23' W, 22° 51' S, at 765 m asl). The climate is Cwa, according to the Köppen classification, i.e., tropical with dry winters and rainy/warm summers. The average annual high and low temperatures in the first year were 25.1 °C and 16 °C, respectively, and the annual pluvial precipitation was 1,330 mm. In the second year, these values were 26.5 °C, 16.5 °C, and 1,396 mm for average high temperature, average low temperature, and pluvial precipitation, respectively.

The soil of the experimental area was classified as structured Alfisol (Embrapa, 2006), managed under NTS for six years (year 1: corn/oat; year 2: soybean/corn; year 3: corn/oat; year 4: soybean/oat; year 5: corn/oat/millet; and year 6: corn intercropped with palisadegrass). The chemical properties prior to the experiment in the 0-0.20 m soil layer were: pH( $\text{CaCl}_2$ ) = 5.2; organic matter (OM) = 23 g  $\text{kg}^{-1}$ ; P (resin) = 19 mg  $\text{dm}^{-3}$ ; K = 2.5 mmol $_c$   $\text{dm}^{-3}$ ; Ca = 31 mmol $_c$   $\text{dm}^{-3}$ ; Mg = 14 mmol $_c$   $\text{dm}^{-3}$  and base saturation = 56 %.

## Experimental design and treatments

The experiment was arranged in a randomized block design in a split plot scheme with four replications. The plots consisted of two periods of intercropping: Corn + palisadegrass simultaneously sown (SS), and corn+palisadegrass sown in a row with a disk fertilizer applicator at N top-dressing fertilization (STD), after corn had reached the stage of four developed leaves, corresponding to 35 days after corn emergence. The subplots were represented by three N rates in the form of ammonium nitrate (30, 60 and 120 kg ha<sup>-1</sup> N), applied to forage after corn harvest. The evaluated plot and subplot areas were 75 and 25 m<sup>2</sup>, respectively. The hybrid corn line P30F90 and grass species *Brachiaria brizantha* Stapf cv. Marandu were used.

## N application to palisadegrass after corn harvest

Prior to N application to the subplots, corn was harvested in April 2005, with a self-propelled harvester. At this point, all experimental units were equalized at a reference height of 25 cm above the soil with a horizontal chopper (tractor), as proposed by Souza Junior (2007). After 10 days, N rates were applied to the surface in fine lines between forage rows, using non-enriched ammonium nitrate as N source. Also, two 1-m long rows were drawn in the center of each subplot, forming microplots, which received the enriched fertilizer (labeled) at the same rate as the rest of the subplot. This way, enriched ammonium nitrate solution (<sup>15</sup>NH<sub>4</sub>NO<sub>3</sub>), with 5 % (<sup>15</sup>N) isotopic abundance in the ammonium of the ammonium nitrate source only, was applied to the surface by hand.

## Forage evaluation

In order to determine forage dry matter production (FDMP), subplots were sampled 50, 160 and 198 days after fertilization (DAF). However, it should be noted that the first evaluation (50 DAF) of the STD treatment could not be carried out due to the small size of the palisade grass.

At all sampling times, a mechanical mower was used to cut the plants at 25 cm from the soil, always at random points in a 2 m<sup>2</sup> area per experimental unit and outside the <sup>15</sup>N-treated microplots. In the remaining area of each experimental unit, after the collection of the plant material from the microplots, the horizontal chopper was used to equalize and further remove the harvested material simulating grazing (Crusciol et al., 2012; Borghi et al., 2013b; Crusciol et al., 2014). However, mowing at 25 cm above the soil left large enough amounts of forage shoots to cover the soil surface completely and maintain a considerable green leaf area of the plants in all samplings (Pariz et al., 2011). At every sampling, the cut material was dried in a forced air ventilation oven at 60 °C to constant weight and then weighed, to quantify dry matter production.

In order to evaluate the following variables: amount of fertilizer-derived N in the plant (FDNFP), <sup>15</sup>NH<sub>4</sub>NO<sub>3</sub> expressed in kg ha<sup>-1</sup>, and the fertilizer use efficiency (FUE), expressed in percentage, all forage shoots of each microplot were collected and removed with a mechanical mower. Following, the collected material was analyzed in the Laboratory of Stable Isotopes, Center for Nuclear Energy in Agriculture (CENA) of the University of São Paulo (USP), in Piracicaba, Brazil, according to methods and expressions of the isotope dilution method (Trivelin et al., 1994; Oliveira et al., 2003; Lara Cabezas & Pádua, 2007). All material was analyzed with a mass spectrometer coupled to an ANCA-SL analyzer (Europa Scientific Ltda).

Total FDMP and FDNFP were determined considering the sum of all evaluation times, as well as the FUE, which represents the N recovery rate by the plant.

The amount of fertilizer-derived N in the forage plant (FDNFP) was calculated using equation 1:

$$\text{FDNFP (kg ha}^{-1}\text{)} = \left[ \% \frac{\text{FDNFP}}{100} \right] \cdot \text{TNA} \quad (1)$$

where %FDNFP = percentage of fertilizer-derived N in the plant, obtained by equation 2:

$$\% \text{TNA} = \left[ \frac{(a-c)}{(b-c)} \right] \cdot 100 \quad (2)$$

where TNA = total nitrogen accumulated (kg ha<sup>-1</sup>); a = <sup>15</sup>N abundance in plant dry matter of the microplot; b = percentage of <sup>15</sup>N in ammonium nitrate (5 %); and c = <sup>15</sup>N abundance in plant dry matter at the microplot edges.

As in ammonium nitrate only NH<sub>4</sub><sup>+</sup> was labeled (5 % of total NH<sub>4</sub><sup>+</sup> of the fertilizer ammonium nitrate), all calculations were performed considering that palisadegrass can absorb equal amounts of NO<sub>3</sub><sup>-</sup> and NH<sub>4</sub><sup>+</sup>. Thus, the FDNFP was estimated with regard to N derived from NH<sub>4</sub><sup>+</sup>. However, the data were multiplied by two, which is equivalent to the two N forms present in the fertilizer. The FDNFP results were also converted to kg ha<sup>-1</sup>.

From the results from FDNFP analyses, the FUE was calculated using equation 3:

$$\text{FUE (\%)} = \left[ \frac{\text{FDNFP}}{\text{Amount of N applied}} \right] \cdot 100 \quad (3)$$

## Implantation of corn in succession

On December 18<sup>th</sup>, 2005, 20 days after the last palisadegrass evaluation (November), the forage was desiccated with glyphosate herbicide at a rate of 1,800 g ha<sup>-1</sup> of active ingredient, applying a volume of 250 L ha<sup>-1</sup>, to sow the subsequent corn crop. The corn hybrid P30F90 was sown in December, at a density of 60,000 plants ha<sup>-1</sup>, in 0.80 m row spacing, including

the  $^{15}\text{N}$ -fertilized microplots. Corn plants emerged six days after sowing, on December 26<sup>th</sup>, 2005.

Mineral fertilization at sowing consisted of the application of 26 kg ha<sup>-1</sup> N, 90 kg ha<sup>-1</sup> P<sub>2</sub>O<sub>5</sub> and 51 kg ha<sup>-1</sup> K<sub>2</sub>O, following the recommendations of Raij et al. (1996) for corn. During the corn development period, all agricultural procedures were adopted, according to the crop requirements (Fornasieri Filho, 2007).

When corn plants reached the stage of four developed leaves (18 days after emergence), top-dressing fertilization was applied at a rate of 120 kg ha<sup>-1</sup> N, corresponding to 375 kg ha<sup>-1</sup> of the fertilizer ammonium nitrate, according to Raij et al. (1996).

### Evaluations of corn in succession to palisadegrass

For N leaf analysis, the central third of 30 leaves was sampled at the corn ear base when at least 50 % of the corn plants were in full flowering stage. Consequently, the selection was randomized, according to the methods of Cantarella et al. (1997). Leaf samples were then washed in tap water, wrapped in paper bags, dried in a forced-air circulation oven at 65 °C for 72 h, and finally ground. Nitrogen concentration in the leaves was determined according to Malavolta et al. (1997). Nitrogen was extracted with H<sub>2</sub>SO<sub>4</sub> into solution and the N concentration in this solution was determined by the Kjeldahl distiller method.

The corn was harvested mechanically within the evaluation area of each subplot 142 days after corn emergence. The harvested grains were then weighed and the grain yield per hectare (grain water content corrected to 130 g kg<sup>-1</sup>, on a wet basis) was calculated. Prior to this, all plants from the microplots were harvested along two 1-m long rows. The corn plants were separated in shoots and grains.

Finally, both FDNFP and FUE were determined for plant shoots and grains, by the methodologies described above. With these results these variables were calculated for the whole plant (plant + grains).

### Statistical analysis

Data were subjected to analysis of variance and the means compared by Tukey's test at 5 %, using the statistical program SISVAR (Ferreira, 1999).

## RESULTS AND DISCUSSION

### Evaluation of palisadegrass

In the first evaluation experiment, 50 days after fertilization (DAF) in SS, the average FDNFP was 6.3 kg ha<sup>-1</sup> and FUE 11.4 % (Table 1).

It was found that 50 DAF the N rates had no effect on forage dry matter production. However, FDNFP

was highest after applying 120 kg ha<sup>-1</sup> N compared to the other rates, and FUE was highest when the lowest N rate (30 kg ha<sup>-1</sup>) was applied. In the other evaluation periods (160 and 198 DAF) and in total, the analysis of all palisadegrass variables suggested significant interactions between factors (Table 2).

Our data show that SS generated the highest dry matter production in both evaluations (160 and 198 DAF) and in total for all N rates (Table 2). The higher forage yields under SS were proportional to the intercrop sowing time, so that under SS the forage was better developed and possibly the root system was more robust. This resulted in a greater accumulation of dry matter (DM) after corn harvest. Similar results were also obtained by other authors when forage was planted earlier (during corn sowing) (Alvarenga et al., 2006; Pariz et al., 2009; Borghi et al., 2012; Crusciol et al., 2012; Borghi et al., 2013a).

Regarding N levels, in both evaluations (160 and 198 DAF) and in the total, the 60 kg ha<sup>-1</sup> rate resulted in the highest DM increases (Table 2) for both SS and STD. There were no differences in N contents 160 DAF and in total after the 120 kg ha<sup>-1</sup> rate. According to Carad et al. (2008), within certain limits that vary according to the genetic potential of the species, increases in the plant DM yield are normal due to N fertilization.

Carad et al. (2008) observed increases in palisadegrass production only up to rates of 200 kg ha<sup>-1</sup> N with grass sowing in the beginning of the rainy season. According to Müller et al. (2002), a possible explanation for the limited response to N is that after corn harvest, in May, the weather conditions (low temperature and precipitation) were inadequate for appropriate forage development and nutrient uptake. Furthermore, according to Costa et al. (2005), the optimal temperature for palisadegrass development is between 30 and 35 °C, while it will not grow at temperatures between 10 and 15 °C. However, the forage DM production was higher 198 DAF than 160 DAF, due to the progression of palisadegrass development as well as the more favorable climatic conditions in the beginning of the rainy season (Jayme et al., 2009).

For both intercropping systems, FDNFP levels rose with increasing N rates in all evaluations and in total (Table 2). The analysis of the intercrop sowing time for each N rate revealed that 160 DAF, only 60 kg ha<sup>-1</sup> induced differences, with higher FDNFP in SS than in STD. At 198 DAF, STD provided more FDNFP than SS at all N rates. This is likely due to the greater increase of DM production by STD in relation to SS, in a comparison of the samplings 198 and 160 DAF, directly reflecting on FDNFP. However, when overall FDNFP is assessed, SS resulted in higher values at all N rates, as an effect of total forage DM production.

Thus, it may be inferred that the establishment of early forage-corn intercropping (SS) will allow a better

**Table 1. Forage dry matter production (FDMP), amount of fertilizer-derived N in the forage plant (FDNFP), and fertilizer use efficiency (FUE) by palisade grass sown in intercrops with corn at two times and fertilized with N rates (source  $^{15}\text{NH}_4\text{NO}_3$ ) applied after corn grain harvest and evaluated at three sampling times (50, 160 and 198 days after fertilization - DAF) and total**

Treatment	50 DAF <sup>(1)</sup>			160 DAF			198 DAF			Total		
	FDMP	FDNFP	FUE	FDMP	FDNFP	FUE	FDMP	FDNFP	FUE	FDMP	FDNFP	FUE
	— kg ha <sup>-1</sup> —			— kg ha <sup>-1</sup> —			— kg ha <sup>-1</sup> —			— kg ha <sup>-1</sup> —		
	%			%			%			%		
Corn + palisadegrass simultaneously sown	648	6.3	11.4	3,310 a	27.6 a	43.1 a	4,524 a	21.7 b	31.1 a	8,482 a	55.5 a	82.3 a
Corn + palisadegrass sowed at corn topdressing				2,486 b	23.7 a	37.3 a	3,804 b	26.8 a	35.6 a	6,290 b	50.5 a	70.6 b
N rate (kg ha <sup>-1</sup> )												
30	640 a	5.8 b	19.3 a	2,054 b	12.8 c	42.7 a	3,427 c	8.3 c	27.7 b	6,441 c	24.0 c	79.9 a
60	670 a	5.1 b	8.5 b	3,244 a	25.4 b	42.3 a	5,009 a	16.2 b	27.0 b	9,413 a	44.2 b	73.6 a
120	635 a	7.6 a	6.3 b	3,398 a	38.8 a	32.3 a	4,055 b	48.3 a	40.2 a	8,088 b	91.0 a	75.8 a
F probability												
Intercrop sowing time (IT)				<0.001	0.054	0.101	<0.001	<0.001	0.062	<0.001	0.065	<0.001
N rate (N)	0.384	0.032	<0.001	<0.001	<0.001	0.087	<0.001	<0.001	<0.001	<0.001	<0.001	0.074
IT × N				<0.001	0.023	0.041	<0.001	0.047	0.002	0.016	0.048	0.027

<sup>(1)</sup> DAF: days after nitrogen fertilization. Means followed by same letter in the column (lowercase) do not differ by Tukey test at 5 %.

N use by the grass, because it will be established for a longer period at the time of fertilization, compared to STD. Therefore, early-established palisadegrass is able to develop more quickly and the first grazing can be scheduled earlier. Similar results, reporting an increase in plant N accumulation as N fertilization increases, were obtained by Scivittaro et al. (2000), Silva et al. (2006a) and Lara-Cabezas (2011). According to these authors, this stimulates the root development, resulting in a better use of water and soil nutrients by the plant. However, the response to higher rates may be limited by adverse weather conditions, because the forage growth potential is higher in spring/summer than in autumn/winter (Kluthouski et al., 2003).

Regarding the FUE, which takes into account the amount of absorbed nutrient in relation to the initially applied rate, 160 DAF, an application of 120 kg ha<sup>-1</sup> resulted in a lower FUE in SS (Table 2). When intercrop sowing times were compared, differences were observed only at the 60 kg ha<sup>-1</sup> rate, with higher FUE in SS than STD. At 198 DAF, the FUE in SS was lower at 60 kg ha<sup>-1</sup>, differing from the other rates. In STD the FUE was highest at an N rate of 120 kg ha<sup>-1</sup>.

The comparison between intercrop sowing times, revealed a higher FUE in STD at 60 and 120 kg ha<sup>-1</sup> N (Table 2). Since there was no sampling 50 DAF in STD because at that time it had been sown shortly before, there was no mowing or removal of plant material above 25 cm from the soil nor N export. Therefore the soil probably contained more N fertilizer to be absorbed by the forage at the intercrop sowing time compared to SS, which affected both FDNFP and FUE.

The analysis of FUE revealed reduced values as N rates increased (from 88.7 % with 30 kg ha<sup>-1</sup> to 75.2 % with 120 kg ha<sup>-1</sup>) in SS (Table 2). The FUE was higher in STD (76.2 %) at an N rate of 120 kg ha<sup>-1</sup>. The comparison of total FUE between intercrop sowing times showed that SS led to higher percentages at 30 and 60 kg ha<sup>-1</sup> N, compared to STD. Viana et al. (2011) reported nitrogen FUE of 33, 42 and 42 % of palisadegrass DM when applying 100, 200 and 300 kg ha<sup>-1</sup> N, respectively. Silva et al. (2006b) reported that N recovery by corn was about 15 % and that these values rarely exceed 50 %. On the other hand, Lara-Cabezas (2011) added that nitrogen FUE of crops is affected by several factors and can vary from 30 to 90 %, and that the recovered nutrient could be used by the subsequent crop.

The data of FDNFP and FUE in the evaluation periods showed that the N accumulation and consequently the recovery efficiency by SS palisadegrass were highest 160 DAF (FDNFP 27.6 kg ha<sup>-1</sup> and FUE 43.1 %) (Table 1). In STD this effect was observed 198 DAF (FDNFP 26.8 kg ha<sup>-1</sup> and FUE 35.6 %).

These differences may be related to the intercrop sowing time, because even though N fertilization was carried out at the same time, plants in SS were in a more advanced developmental stage than in STD. Therefore, the higher N uptake in STD was only observed 198 DAF, when the plants were better developed. Since STD was initiated 35 days after corn emergence (N fertilization), the plants in STD probably had the same physiological age 198 DAF as in IS plants at 160 DAF.

Fagundes et al. (2005) and Alexandrino et al. (2005) emphasized that among the factors that limit

**Table 2. Interaction split between intercrop sowing times  $\times$  N rates ( $^{15}\text{NH}_4\text{NO}_3$  source) applied after corn harvest for forage dry matter production (FDMP), amount of fertilizer-derived nitrogen in the plant (FDNFP) and fertilizer use efficiency (FUE) of palisadegrass, in two evaluation periods (160 and 198 days after fertilization - DAF) and total**

Treatment	N rate (kg ha <sup>-1</sup> )		
	30	60	120
		FDMP (kg ha <sup>-1</sup> )	
		160 DAF	
Corn + palisadegrass simultaneously sown	2,240 aC	4,107 aA	3,585 aB
Corn + palisadegrass sowed at corn topdressing	1,867 bB	2,690 bA	2,902 bA
		198 DAF	
Corn + palisadegrass simultaneously sown	3,638 aC	5,660 aA	4,273 aB
Corn + palisadegrass sowed at corn topdressing	3,217 bC	4,359 bA	3,837 bB
		Total	
Corn + palisadegrass simultaneously sown	6,518 aC	10,437 aA	8,493 aB
Corn + palisadegrass sowed at corn topdressing	5,084 bB	7,049 bA	6,739 bA
		FDNFP (kg ha <sup>-1</sup> )	
		160 DAF	
Corn + palisadegrass simultaneously sown	13.9 aC	30.7 aB	38.3 aA
Corn + palisadegrass sowed at corn topdressing	11.6 aC	20.1 bB	39.4 aA
		198 DAF	
Corn + palisadegrass simultaneously sown	6.9 bC	14.0 bB	44.3 bA
Corn + palisadegrass sowed at corn topdressing	9.7 aC	18.5 aB	52.3 aA
		Total	
Corn + palisadegrass simultaneously sown	26.6 aC	49.8 aB	90.2 aA
Corn + palisadegrass sowed at corn topdressing	21.3 bC	38.6 bB	91.7 aA
		FUE (%)	
		160 DAF	
Corn + palisadegrass simultaneously sown	46.3 aA	51.2 aA	31.9 aB
Corn + palisadegrass sowed at corn topdressing	40.0 aA	33.5 bA	38.3 aA
		198 DAF	
Corn + palisadegrass simultaneously sown	33.0 aA	23.3 bB	36.9 bA
Corn + palisadegrass sowed at corn topdressing	32.3 aB	30.8 aB	43.6 aA
		Total	
Corn + palisadegrass simultaneously sown	88.7 aA	83.0 aAB	75.2 aB
Corn + palisadegrass sowed at corn topdressing	71.0 bAB	64.3 bB	76.4 aA

Means followed by the same letter in a column (lowercase) and row (uppercase) do not differ by Tukey's test at 5 %.

forage development, low nutrient availability, especially nitrogen, is one of the main aspects affecting forage yield and quality. Fagundes et al. (2005) also mentioned that the palisadegrass yield potential is a genetically determined trait. However, in order to achieve this potential the appropriate environmental conditions (temperature, humidity, light, nutrient availability) must be given. Hence, adequate timing and dosage of nitrogen applications increase the use efficiency and forage dry matter production.

According to Lara & Pedreira (2011), our knowledge on the physiological responses of forage must be deepened so that growth acceleration can be adequately exploited to generate long-lasting productivity gains.

### Residual effect of nitrogen on corn after palisadegrass

The evaluation of the subsequent corn crop reveals that, with the exception of N leaf content, there was a significant interaction between the factors for the other variables (Table 3). In general, low FDNFPs (2.94 and 2.23 kg ha<sup>-1</sup> for SS and STD, respectively), and consequently low FUE values (4.78 and 3.62 % N the SS and STD, respectively) were observed in the whole corn plant (grain + plant). Thus, of the total N applied to palisadegrass in the previous year, which continued growing throughout the autumn-winter period and was cut to form straw for NTS corn seeding, great part was recovered by the developing forage (FUE 82.3 and 70.6 % of N in SS and STD, respectively) (Table 1). On the other hand, only little amounts of the N

**Table 3. Residual effect of N fertilization ( $^{15}\text{NH}_4\text{NO}_3$ ) applied in palisadegrass detected in corn by the evaluation of fertilizer-derived N content in grain, plant, and total (grain + plant) (FDNFP), N fertilizer use efficiency (FUE), N corn leaf concentration (N), and grain yield (GY) of corn sown into straw of palisade grass sown in two intercrops at different times**

Treatment	Grain		Plant		Grain + plant		N	GY
	FDNFP	FUE	FDNFP	FUE	FDNFP	FUE		
	kg ha <sup>-1</sup> N	%	kg ha <sup>-1</sup> N	%	kg ha <sup>-1</sup> N	%	g kg <sup>-1</sup>	kg ha <sup>-1</sup>
Corn + palisadegrass simultaneously sown	1.10 a	1.74 a	1.84 a	2.87 a	2.94 a	4.78 a	30.4 a	10,544 b
Corn + palisadegrass sowed at corn topdressing	0.78 b	1.29 b	1.46 b	2.33 a	2.23 b	3.62 b	30.2 a	11,409 a
N rate (kg ha <sup>-1</sup> )								
30	0.60 c	2.00 a	1.01 c	3.11 a	1.61 c	5.37 a	30.9 a	10,792 a
60	0.86 b	1.42 b	1.69 b	2.82 a	2.52 b	4.21 b	30.0 a	11,220 a
120	1.37 a	1.13 c	2.25 a	1.87 b	3.62 a	3.01 c	30.0 a	10,916 a
F probability								
Intercrop sowing time (IT)	<0.001	<0.001	<0.001	0.153	<0.001	0.003	0.358	0.028
N rate (N)	<0.001	<0.001	<0.001	0.004	<0.001	<0.001	0.297	0.106
IT × N	0.018	0.046	0.028	0.041	0.039	0.044	0.401	0.042

Means followed by same letter in the column (lowercase) do not differ by Tukey test at 5 %.

that remained in the straw and/or soil were available to be absorbed by the corn plants sown in succession, which corroborates the data of Silva et al. (2006a). Considering each intercrop sowing time, regardless of the rate of N applied, and the total forage and whole corn plant (grain + plant), the FUE was 87.1 and 74.2 % in SS and STD, respectively (Tables 1 and 3).

From the analyses of the interactions (Table 4), higher FDNFPs were observed as the levels of N increased in both intercropping seasons and in all plant structures analyzed. A comparison between intercrop sowing times revealed that the 60 and 120 kg ha<sup>-1</sup> rates resulted in higher N amount in the SS grain (0.99 and 1.64 kg ha<sup>-1</sup> N, respectively) than that in STD (0.73 and 1.09 kg ha<sup>-1</sup> N, respectively). This is likely an effect of the larger amounts of N applied, which normally have a close relationship to the N concentrations found in the plant (Lara-Cabezas & Pádua, 2007). At a rate of 120 kg ha<sup>-1</sup>, FDNFP was higher in SS (2.55 kg ha<sup>-1</sup> N) than STD (1.94 kg ha<sup>-1</sup>) in the rest of the plant (leaves + stem + straw + cob), which directly affected the FDNFP results for the whole plant (plant + grains). The reason is that the plant without grains has a higher representation than the grains alone.

The FUE found in SS was higher at the lowest N rate (30 kg ha<sup>-1</sup>) in all plant structures and the whole plant (Table 4). These results corroborate those of Silva et al. (2006b), who evaluated the usage of nitrogen ( $^{15}\text{N}$ ) provided by millet (*Pennisetum glaucum*) and *Crotalaria* spp. by corn. Similar results were found after the evaluation of STD grains. However, when the plant (leaves + stem + straw + cob) was taken into consideration, differently from the 120 kg ha<sup>-1</sup> rate, the FUE was higher at lower N rates (30 and 60 kg ha<sup>-1</sup>), which directly influenced the results of the entire plant.

The FUE levels of corn grains were higher in SS than in STD at all N rates when the intercrop sowing time was compared (Table 4). However, this was not observed when the plant (leaves + stem + straw + cob) FUE was analyzed. Considering the whole plant (plant + grains), we found that at 30 kg ha<sup>-1</sup> N, FUE was greater in SS than in STD (6.30 % in SS and 4.45 % in STD).

The FUE results make it clear that a considerable portion of the N absorbed by the plant was translocated into the grains, in the form of protein and amide compounds (Fancelli & Dourado Neto, 2000). On average, considering only the intercrop sowing time, regardless of probable N losses from the soil and plants and independently of N rates, some 12.9 and 25.8 % of the N applied to forage as fertilizer were not recovered by the corn sown into palisadegrass straw in SS and STD, respectively. However, the overall FUE values were far below the estimated potential (Table 3). This was been observed by Silva et al. (2006a) and Fernandes (2006) who detected less than 3 % of residual N when fertilizer was applied in the previous crop year.

The low recovery and amount in the corn plant of the N applied to the growing forage as fertilizer may be related to several factors. However, it is important to highlight that the low recovery refers to the fertilizer applied to palisadegrass and not to corn, since corn was not fertilized with labeled compounds.

Nitrogen was provided as a seeding fertilizer during both years of corn sowing, at the rate of 26 kg ha<sup>-1</sup>, followed by an additional 120 kg ha<sup>-1</sup> rate in the form of ammonium nitrate at the stage of four developed leaves. Therefore, N concentrations in the corn leaves were not affected by rate, intercrop sowing time and interaction (Table 3). The observed values were



**Table 4. Interaction between intercrop sowing time  $\times$  N rates (source  $^{15}\text{NH}_4\text{NO}_3$ ) applied to palisade grass for: fertilizer-derived N in the plant (FDNFP), fertilizer use efficiency (FUE) by the grain, plant, and total (grain + plant), and grain yield (GY) of corn sown into straw of palisade grass of two intercrop sowing times**

Treatment	N rate (kg ha <sup>-1</sup> )		
	30	60	120
		FDNFP (kg ha <sup>-1</sup> )	
		Grain	
Corn + palisadegrass simultaneously sown	0.68 aC	0.99 aB	1.64 aA
Corn + palisadegrass sowed at corn topdressing	0.52 aC	0.73 bB	1.09 bA
		Plant	
Corn + palisadegrass simultaneously sown	1.21 aC	1.77 aB	2.55 aA
Corn + palisadegrass sowed at corn topdressing	0.81 bC	1.61 aB	1.94 bA
		Grain + plant	
Corn + palisadegrass simultaneously sown	1.89 aC	2.73 aB	4.19 aA
Corn + palisadegrass sowed at corn topdressing	1.33 aC	2.31 aB	3.04 bA
		FUE (%)	
		Grain	
Corn + palisadegrass simultaneously sown	2.26 aA	1.63 aB	1.34 aB
Corn + palisadegrass sowed at corn topdressing	1.75 bA	1.21 bB	0.91 bB
		Plant	
Corn + palisadegrass simultaneously sown	3.53 aA	2.95 aAB	2.13 aB
Corn + palisadegrass sowed at corn topdressing	2.70 aA	2.69 aA	1.62 aB
		Grain + plant	
Corn + palisadegrass simultaneously sown	6.30 aA	4.55 aB	3.49 aB
Corn + palisadegrass sowed at corn topdressing	4.45 bA	3.87 aA	2.53 aB
		GY (kg ha <sup>-1</sup> )	
Corn + palisadegrass simultaneously sown	11,003 aA	10,614 bA	10,016 bA
Corn + palisadegrass sowed at corn topdressing	10,582 aB	11,827 aA	11,817 aA

Means followed by the same letters in columns (lowercase) and rows (uppercase) do not differ by Tukey's test at 5 %.

within the range of 27 to 35 g kg<sup>-1</sup>, which is considered suitable for corn cultivation, according to Cantarella et al. (1997). This way, surface N fertilization (120 kg ha<sup>-1</sup>) probably provided the crop with sufficient amounts of the nutrient and also reduced the response (FUE = 3.49 and 2.53 %, respectively, for SS and STD) to the N rates applied to the forage preceding corn.

This shows that the residual N from fertilization in SS did not affect the grain yield of corn sown into palisadegrass straw (Table 4). Campos (2004) and Lara-Cabezas & Pádua (2006) observed no corn yield increases either, except in the control treatment (no N), when higher N rates were applied in monoculture or simultaneous intercropping with palisadegrass.

However, when corn was sown into STD palisadegrass straw, the residual N from fertilization at rates of 60 and 120 kg ha<sup>-1</sup> led to higher grain yield than 30 kg ha<sup>-1</sup> N (Table 4). Furthermore, when the residual N from fertilization was compared considering the straw from different intercrop sowing times, we found that corn grain yield was higher on STD straw treated with 60 and 120 kg ha<sup>-1</sup> N, statistically differing from SS. This can be explained by the lower

forage production in STD than SS (Table 3), which resulted in lower N export, with most residual N remaining in the soil.

Moreover, despite the removal of the harvested material after each evaluation, the greater forage yield induced by higher N rates (60 and 120 kg ha<sup>-1</sup>) may have increased the cycling of other non-nitrogen nutrients (Pariz et al., 2011). Chemical, physical and biological characteristics of the soil may also have been improved by the higher forage biomass (Crusciol et al., 2009), since the amount of forage left after mowing was large enough to cover the soil completely. By using this management, the amount of nutrients recycled by palisadegrass is relatively high, as exports occur only in tender leaves with significantly lower dry matter contents than shoots, while the crop residues are left in the field (Pariz et al., 2011).

In summary, we observed that N fertilization significantly affected forage dry matter production up to a rate of 60 kg ha<sup>-1</sup>, with higher values in SS than STD. Similarly, we found that fertilizer use efficiency was higher in intercrops sown earlier. It was also verified that uptake of N applied to forage was low in the subsequent corn crop. However, higher rates (60

and 120 kg N ha<sup>-1</sup>) resulted in increased grain corn yield when the intercrop was sown at top dressing. Based on our results, we can infer that earlier intercropping allowed a higher forage dry matter production. On the other hand, the establishment of intercropping at later times resulted in higher grain yield by corn in succession to nitrogen-fertilized forage.

## CONCLUSIONS

1. The intercrop sowing time significantly affected forage dry matter production, the amount of fertilizer-derived nitrogen in the forage plant and nitrogen fertilizer use efficiency by forage plants.

2. Nitrogen application in autumn on palisadegrass intercropped with corn, either at sowing or at top dressing of nitrogen fertilization, increased the forage yield up to a rate of 60 kg ha<sup>-1</sup>.

3. The amount of fertilizer-derived nitrogen in the forage plant and the fertilizer use efficiency by palisadegrass for both intercrop sowing times were highest 160 days after fertilization, regardless of N rates.

4. Residual nitrogen did not affect the N nutrition of corn plants grown in succession to palisadegrass, but increased grain yield at rates of 60 and 120 kg ha<sup>-1</sup> N, when corn was grown on straw of palisadegrass from the intercrop sown at corn top dressing fertilization.

5. Based on our results, it can be inferred that the early intercropping allowed higher forage dry matter production. On the other hand, the intercrop sown later allowed a higher corn grain yield in succession to nitrogen-fertilized palisadegrass.

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