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Utilization of nitrogen (^{15}N) from urea and green manures by rice as affected by nitrogen fertilizer rate

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The use of green manures (GMs) in combination with nitrogen (N) fertilizer application is a promising practice to improve N fertilizer management in agricultural production systems. The main objective of this study was to evaluate the N use efficiency (NUE) of rice plant, derived from GMs including sunn hemp (*Crotalaria juncea* L.), millet (*Pennisetum glaucum* L.) and urea in the greenhouse. The experimental treatments included two GMs (sunn hemp- ^{15}N and millet- ^{15}N), absence of N organic source (without GM residues in soil) and four N rates, as urea- ^{15}N (0, 28.6, 57.2 and 85.8 mg N kg⁻¹). The results showed that both rice grain and straw biomass yields under sunn hemp were greater than that of millet or without the application of GM. The NUE of rice under sunn hemp was greater than that under millet (18.9 and 7.8% under sunn hemp and millet, respectively). The urea N application rates did not affect the fertilizer NUE by rice (53.7%) with or without GMs. The NUE of GMs by rice plants ranged from 14.1% and 16.8% for root and shoot, respectively. The study showed that green manures can play an important role in enhancing soil fertility and N supply to subsequent crops.

Key words: *Oryza sativa* L., sunn hemp, millet, isotopic dilution, nitrogen mineralization.

INTRODUCTION

Rice is a staple food for more than 50% of the world population (Fageria et al., 2010). Nitrogen (N) is the main

nutrient that affects the rice yield, because it increases the percentage of filled spikelets, increases the leaf

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surface and contributes to improvement of the quality of the grain (Cazetta et al., 2008).

Brazil is the world's largest producer of upland rice, but upland rice yield is much lower than lowland-flooded rice (Fageria et al., 2010, 2011; Conab, 2015). Nitrogen is one of the most important nutrients in determining upland rice yield in Brazil, since low level of N fertilizers is used by farmers due to high cost of these fertilizers (Fageria, 2007; Fageria et al., 2010).

The efficiency of N fertilizers is usually low, with a recovery estimated, on average by the shoots of about 50% of the N applied (Dobermann, 2005; Chien et al., 2009). The importance of green manures (GMs) in crop systems is increasing in recent years due to concern for reducing chemical inputs and improving soil quality (Fageria et al., 2005). The application of N chemical fertilizer in combination with GM may be an effective method to enhance fertilizer N use efficiency and crop yield in dryland (Li et al., 2015).

In addition, the association of GM to N fertilizer is a promising approach to improve N fertilizer management, soil fertility and organic matter, reduce N losses from the readily available sources, increase the use of green manure N and yield of grain crops, and in the case of legume GM, incorporate biologically fixed N to the soil (Bordin et al., 2003; Cazetta et al., 2008; Muraoka et al., 2002; Pöttker and Roman, 1994; Scivittaro et al., 2005, 2008; Reis et al., 2008; Silva et al., 2006, 2010).

Therefore, the use of GM in combination with N fertilizers can be an important complementary strategy to reduce cost of production and to improve upland rice yield. The fate of green manure N as affected by N fertilizer in dryland soils and vice versa is poorly understood (Aulakh et al., 2000; Li et al., 2015). The use of ^{15}N tracer technique is important to evaluate the fate of N derived from N fertilizer, GM and their combination.

The objectives of this work were to evaluate (i) N use efficiency (NUE) by the rice plant derived from GMs sunn hemp (*Crotalaria juncea* L.), millet (*Pennisetum glaucum* L.) and urea (labeled with ^{15}N); (ii) the effect of N sources on rice yield; (iii) the contribution of N derived from the roots and above ground parts of GMs to rice; (iv) and to quantify the N in rice as affected by urea application time (at seeding and topdressing).

MATERIALS AND METHODS

The experiment was carried out in the greenhouse at the Center for Nuclear Energy in Agriculture (Cena/USP), state of Sao Paulo, Brazil, at 22°42'30"S, 47°38'0"W, 554 m altitude. The study was carried out with clay texture soil (440 g kg⁻¹ of clay), collected from the top 20 cm depth of a Rhodic Hapludox according to United States (2006), classified as the Brazilian soil Dystroferric Red Latosol, loamy, cerrado (savannah) phase by Embrapa (2013). The soil chemical analyses were performed according to the methods described by Raij et al. (2001), with the following characteristics: pH (CaCl₂): 4.8; total N: 1.0 g kg⁻¹; soil organic matter: 12.7 g dm⁻³; P (resin-extractable): 11.7 mgdm⁻³; S: 7.5 mgdm⁻³; K⁺: 2.0 mmol_cdm⁻³;

Ca²⁺: 20.0 mmol_c dm⁻³; Mg²⁺: 10.0 mmol_c dm⁻³; H+Al: 26.0 mmol_c dm⁻³; base sum: 32.0 mmol_c dm⁻³; cation exchange capacity: 58.0 mmol_c dm⁻³; base saturation: 55.0%.

The study was conducted in two phases. The first phase was focused on the production of sunn hemp and millet, with and without the addition of labeled ^{15}N fertilizer. In the second trial phase, rice plants were grown with combined application of GM residues unlabeled and ^{15}N -urea labeled or GM residues isotopically ^{15}N -labeled combined with urea without ^{15}N . These two phases were performed in order to distinguish the origin of N in rice plants from different sources (urea, sunn hemp or millet).

In the first phase, plastic pots lined with polyethylene bags, containing 5 kg soil, were used to produce the biomass of GMs. Twenty five pots received 12 seeds each of sunn hemp (*C. juncea*, cv. IAC KR1) or millet (*P. glaucum*, cv. ADR-500) and, five days after emergence (DAE), seedlings were thinned to five plants per pot. The isotopic labeling of the GMs was performed with a rate of 300 mg N per pot as (NH₄)₂SO₄ enriched with 10% of atoms of ^{15}N , split into three applications of 100 mg N per pot vessel at 15, 33 and 45 DAE. Plant shoots and roots were harvested at 63 DAE, rinsed with deionized water, oven-dried at 70°C, weighed, and fragmented into pieces of approximately 0.02 cm. A representative sub sample of each GM was ground to 0.149 mm in a Wiley mill, for chemical analyses. After digestion with H₂SO₄, total N was determined by the Kjeldahl method as described in Malavolta et al. (1997). Total C was analyzed by dry combustion at 1,400°C in an elemental analyzer Leco CN 2000 (Nelson and Sommers, 1982). Then, the C/N ratio was determined. The abundance of ^{15}N was analyzed in mass spectrometry (Barrie and Prosser, 1996).

In the second phase, rice plants were grown in pots with 4 kg of air dried soil each, by combining with or without N fertilizer as urea and GM residues labeled with ^{15}N . The residues were applied in proportion of 4:1, being 20 g of shoots and 5 g of root for both GMs, which were mixed to the soil 12 days before sowing the rice. The soil was watered using deionized water (0.6 μS cm⁻¹), and soil water content was kept at approximately 60% of the retention capacity.

In rice plants (cultivar IAC 202) cultivation, a completely randomized design with 12 treatments was used, in a 3x4 factorial arrangement, with three replicates. The treatments were the combination of two GMs (sunn hemp or millet) and no organic source of N (soil without GM) with four N rates as urea (0, 28.6, 57.2 and 85.8 mg N kg⁻¹).

At sowing, 15 seeds of rice were sown per pot and thinning was carried out eight DAE, keeping four seedlings. All pots received a sowing fertilization with 25.51 mg P kg⁻¹ as single superphosphate, 24.94 mg K kg⁻¹ as potassium chloride. At 10 DAE, basic nitrogen fertilization (20% of total N rate) was done, except in the control treatments (no added urea-N), and 43 DAE (maximum tillering), 80% of the rest was applied in topdressing.

Rice plant shoots were harvested at 120 DAE, after grain physiologic maturation, by cutting the plants at 1 cm above the soil surface, then, the shoots were separated into straw and grain part. Straw and grain were oven-dried at 60°C, for 72 h, weighed, and ground at 0.42 mm in a Wiley mill. The rice grain yield data were adjusted to 13% moisture. Total N was determined by Kjeldahl method, after digestion with H₂SO₄ according to the methods described by Malavolta et al. (1997). Also, total N and the abundance of ^{15}N were determined in the mass spectrometer (Isotope Ratio Mass Spectrometry, IRMS), elemental analyzer interfaced with N, Stable Isotope Laboratory in the Cena/USP (Barrie and Prosser, 1996).

The calculations of the total N accumulated, percentage (NPDFGM) and quantity (QNPDFGM) of N in rice plant derived from green manures and derived from urea (NPDF and QNPDF) and the N utilization efficiency (NUE) from green manure and urea by rice plants were performed according to IAEA (2001).

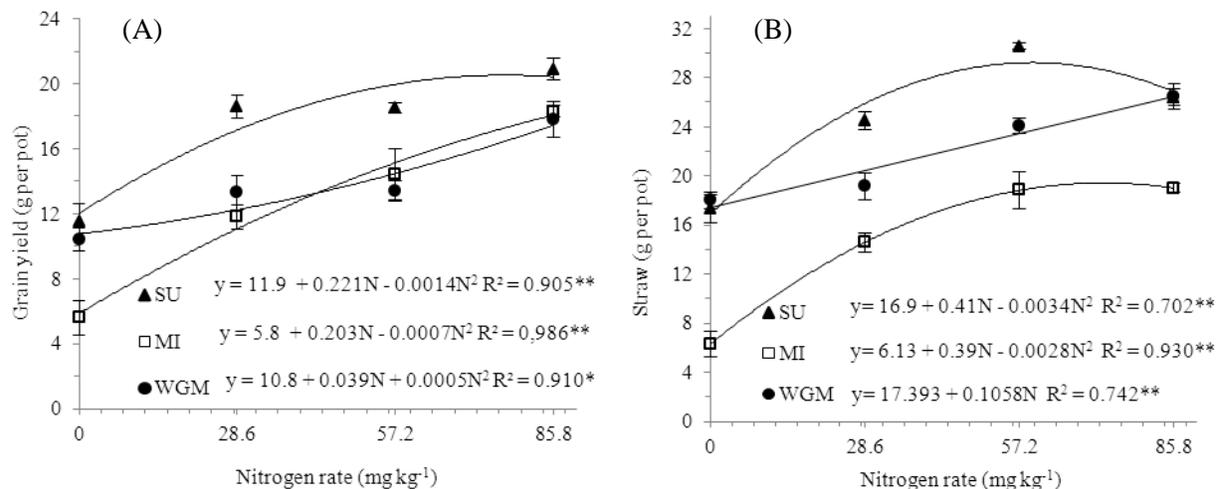


Figure 1. Rice grain yield (A) and straw biomass (B) as affected by N fertilizer rate and without application of green manure (WGM) and application of sunn hemp (SU) or millet (MI). ** and * significant by the F test at 1 and 5%, respectively.

The data obtained were subjected to analysis of variance and, when significant effects were detected at 5% probability, Tukey test at the 0.05 probability was used to compare the means. The factors were adjusted to regression equations for N rates. Linear and quadratic components were tested, and the model with larger significant degree was chosen. The statistical analyses were performed using SAS package (SAS Institute, 2001).

RESULTS AND DISCUSSION

Carbon (C) and nitrogen content in green manures

Shoot C and N content of sunn hemp were 429.6 and 24.0 g kg⁻¹, respectively; in roots, the C and N content were 339.8 and 19.2 g kg⁻¹, respectively. Similarly for millet, the C and N contents of shoot were 419.4 and 10.7 g kg⁻¹, respectively, and in the roots were 320.0 and 10.0 g kg⁻¹, respectively. The greater N content in both shoot and root of sunn hemp as compared to millet may be due to biological N fixation by sunn hemp (Muraoka et al., 2002; Scivittaro et al., 2003; Singh et al., 2004).

Upland rice grain yield and straw biomass

Both upland rice grain yield and straw biomass were significantly influenced by N-urea application rate and GM application. Grain yield increased significantly in a quadratic model with increasing N-urea rate, both with and without the application of sunn hemp or millet (Figure 1A). As compared to the millet treatments or without any GM application, the use of sunn hemp showed greater grain yield and maximum grain yield (20.5 g per pot) was obtained with N fertilizer application rate of 77.0 mg kg⁻¹. Therefore, it is suggested that the application of sunn

hemp supplied more N for rice plants, which resulted in higher grain yield with less N fertilizer. Previous studies with GM also showed higher rice grain yield with the application of N fertilizer in combination with GM when compared with the supply of individual N sources (Muraoka et al., 2002; Bordin et al., 2003; Scivittaro et al., 2003; Fageria and Santos, 2007). Biological N fixation by leguminous green manures can reduce the need for inorganic N fertilizers for the succeeding crop (Singh et al., 2004).

Without N fertilizer input (control treatment with no N fertilizer), the application of sunn hemp increased grain yields by 51 and 12%, respectively, as compared to millet and without application of GM treatments (Figure 1A). Furthermore, the application of only sunn hemp was equivalent to N-urea rate from 24.6 mg kg⁻¹. Muraoka et al. (2002) observed that the application of sunn hemp showed rice grain yield equivalent to N fertilizer rate of 40 kg ha⁻¹. In another study with maize, Silva et al. (2006) observed that the application of sunn hemp resulted in an effect equivalent to N-urea rate of 60 to 90 kg ha⁻¹. This observation suggests that other nutrients were probably released from GM simultaneously with N during the decomposition of sunn hemp residues.

On the other hand, the use of millet affected negatively upland rice yield. In the control treatment (no N fertilizer), rice grain yield was reduced by 44% as compared to without application of GM treatment and it was also lower up to N fertilizer rate of 43 mg kg⁻¹ (Figure 1A). This negative effect is related to the high C/N ratio of the GM that resulted in temporary N immobilization as also shown by Amado et al. (2002) and Silva et al. (2006).

For treatments with GM application, rice straw biomass increased significantly and quadratically with increased N application rates. The maximum straw biomass (29.2 and

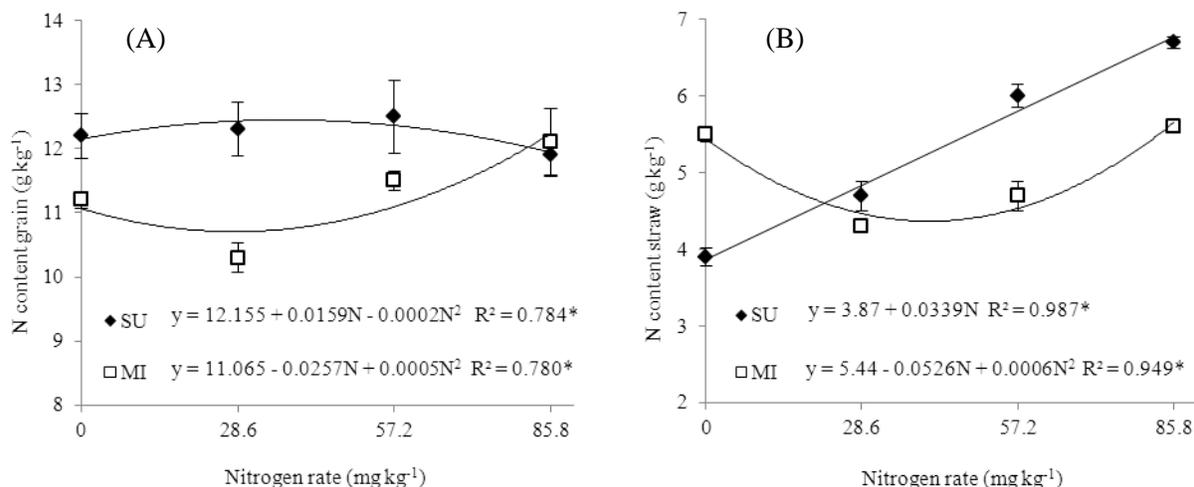


Figure 2. Nitrogen concentration in rice grain (A) and straw biomass (B) as affected by N fertilizer rate and application of green manure sunn hemp (SU) or millet (MI). * significant by the F test at 5%.

19.4 g per pot for sunn hemp and millet, respectively) were obtained with N fertilizer rates of 59.7 and 72.7 mg kg⁻¹ for sunn hemp and millet, respectively (Figure 1B). The maximum rice straw biomass for treatment with application of sunn hemp was 33.6% higher as compared to millet. Therefore, to achieve the same maximum rice straw biomass obtained in treatment with application of sunn hemp, it would be necessary to increase N fertilizer rate to about 20% in the treatment with millet (Figure 1B). Thus, these results suggest that the application of millet increase N immobilization in soil as also shown by Amado et al. (2002) and Silva et al. (2006).

Nitrogen uptake by upland rice as affected by green manures

Nitrogen concentration in rice grain was greater than that in straw (Figure 2) and showed that grain is an important sink for N. There was a significant quadratic relationship between N concentration in rice grain and N fertilizer rate both with sunn hemp and millet application (Figure 2A). For sunn hemp, the maximum N concentration in grain was 12.5 g kg⁻¹ for the N rate of 37.9 mg kg⁻¹. In treatment with application of millet, the maximum N concentration of 12.0 g kg⁻¹ was obtained with N fertilizer application rate of 85.8 mg kg⁻¹.

Nitrogen concentration in rice straw increased significantly with the application of sunn hemp and linearly with increasing N rate, ranging from 3.9 to 6.8 g kg⁻¹ (Figure 2B). With the application of millet, N concentration in straw had significant quadratic relation with N-urea rate and ranging from 4.4 to 5.6 g kg⁻¹. Under millet, N concentration in straw (ranging from 4.4 to 5.6 g kg⁻¹) showed significant quadratic association with N fertilizer application rate and decreased up to N fertilizer

rate of 41.1 mg kg⁻¹ probably because of N immobilization at low rates of N fertilizer.

Nitrogen accumulation in rice straw has significant quadratic association with N fertilizer rate under both sunn hemp and millet (Figure 3A). The application of sunn hemp showed maximum N accumulation of 182.7 mg per pot and it was equivalent to the application of 83.7 mg kg⁻¹ as N fertilizer. While the N fertilizer required for maximum N accumulation was 83.7 mg kg⁻¹ under sunn hemp, the N accumulation in rice straw was 42.4% lower when similar amount was applied with N fertilizer and millet.

Percentage of N in rice straw biomass derived from green manure (NPDFGM) decreased with increase in N fertilizer rate under both sunn hemp and millet (Figure 3B). The minimum NPDFGM (5.56%) in straw was obtained with application of millet and at N fertilizer rate of 65 mg kg⁻¹ mineral. In the control treatment with no N fertilizer application, the NPDFGM in straw was 22% greater in sunn hemp than that under millet.

The amount of N in rice straw biomass derived from green manure (QNPDFGM) was significantly ($P \leq 0.01$) and quadratically increased with increasing N fertilizer rates under both sunn hemp and millet (Figure 3C). The QNPDFGM (Figure 3C) and NUE from GM in straw were lower with the application of millet as compared to sunn hemp at all N fertilizer rates which may be due the lower N content and higher C/N ratio of millet (Figure 3D). NUE of rice straw under millet was low and ranged from 2.2 to 2.8% of the N contained in the GM (Figure 3D). With application of sunn hemp, the maximum NUE was 8.1% and it was obtained at N fertilizer of 65 mg kg⁻¹. The N fertilizer rate significantly increased N accumulation in rice grain and is in a quadratic association with application of sunn hemp (Figure 4A). The maximum N accumulation was 248.7 mg per pot for sunn hemp at N

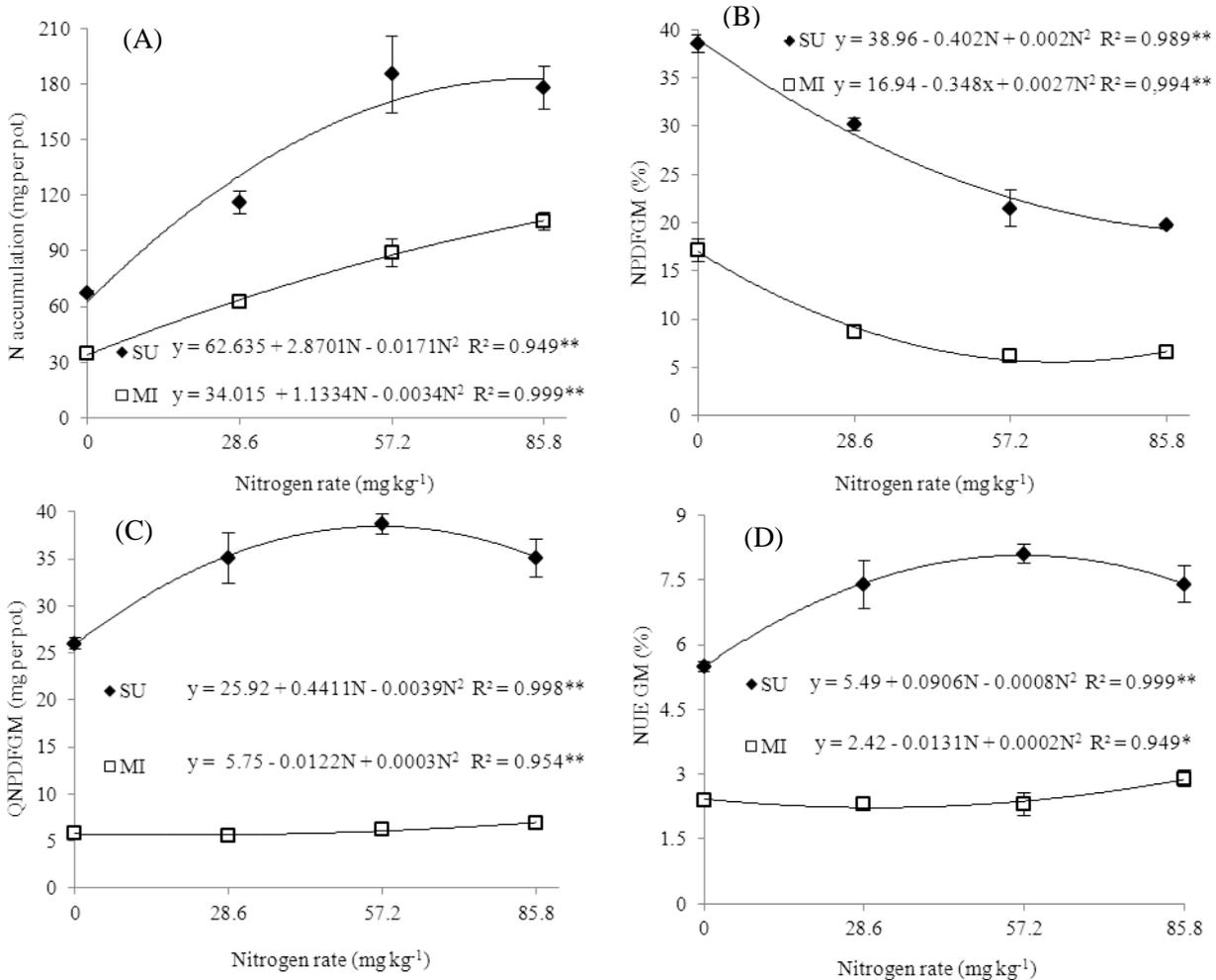


Figure 3. Nitrogen accumulation in rice straw (A), percent (NPDFGM) (B) and quantity (QNPDFGM) of N in rice straw from green manure (C) and N utilization efficiency (NUE) from sunn hemp (SU) and millet (MI) by straw rice (D), as affected by N fertilizer rate. ** and * significant by the F test at 1 and 5%, respectively.

fertilizer rate of 69.3 mg kg⁻¹. With the application of millet, N accumulation in grain increased linearly with increasing N fertilizer rate and ranged from 64.52 to 221.18 mg per pot (Figure 4A). The percentage of N in rice grain derived from green manure (NPDFGM) decreased significantly and quadratically with increasing N fertilizer rates to both sunn hemp and millet (Figure 4B). The percentage of NPDFGM with application of sunn hemp was higher as compared to millet at all N fertilizer rates. With no N fertilizer input, the application of sunn hemp was equivalent to N fertilizer rate of 23 mg kg⁻¹ as compared to same treatment with millet.

The QNPDFGM both for sunn hemp and millet (Figure 4C) and the NUE from these two GMs by rice grains (Figure 4D) were not significantly influenced by N fertilizer application rates. The average of QNPDFGM sunn hemp was 56.4 mg pot⁻¹ and it was equivalent to 11.8% of N applied as residue (476.77 mg per pot). With

application of millet, the QNPDFGM was on average 12.7 mg per pot, equivalent to a recovery of 5.3% of N applied as GM (239.26 mg per pot). Muraoka et al. (2002), using legumes sunn hemp (*Crotalaria juncea*) and velvet bean (*Mucuna aterrima*) as green manure, reported an effect equivalent to the fertilization of 40 kg ha⁻¹ of urea, indicating that these legumes are an important alternative source of N and other plant nutrients. Scivittaro et al. (2004) conducted an experiment to determine the temporal pattern of N release from velvet bean (*Mucuna aterrima*) and studied the dynamics of N contained in green manure in soil-plant system, and found that the incorporation of GM promoted increased dry matter yield and N uptake by rice plants.

As compared to millet, sunn hemp showed greater N accumulation (NA), percentage (NPDFGM) and amount (QPDFGM) of N in rice derived from green manure, NUE from green manure by upland rice (Table 1). Therefore,

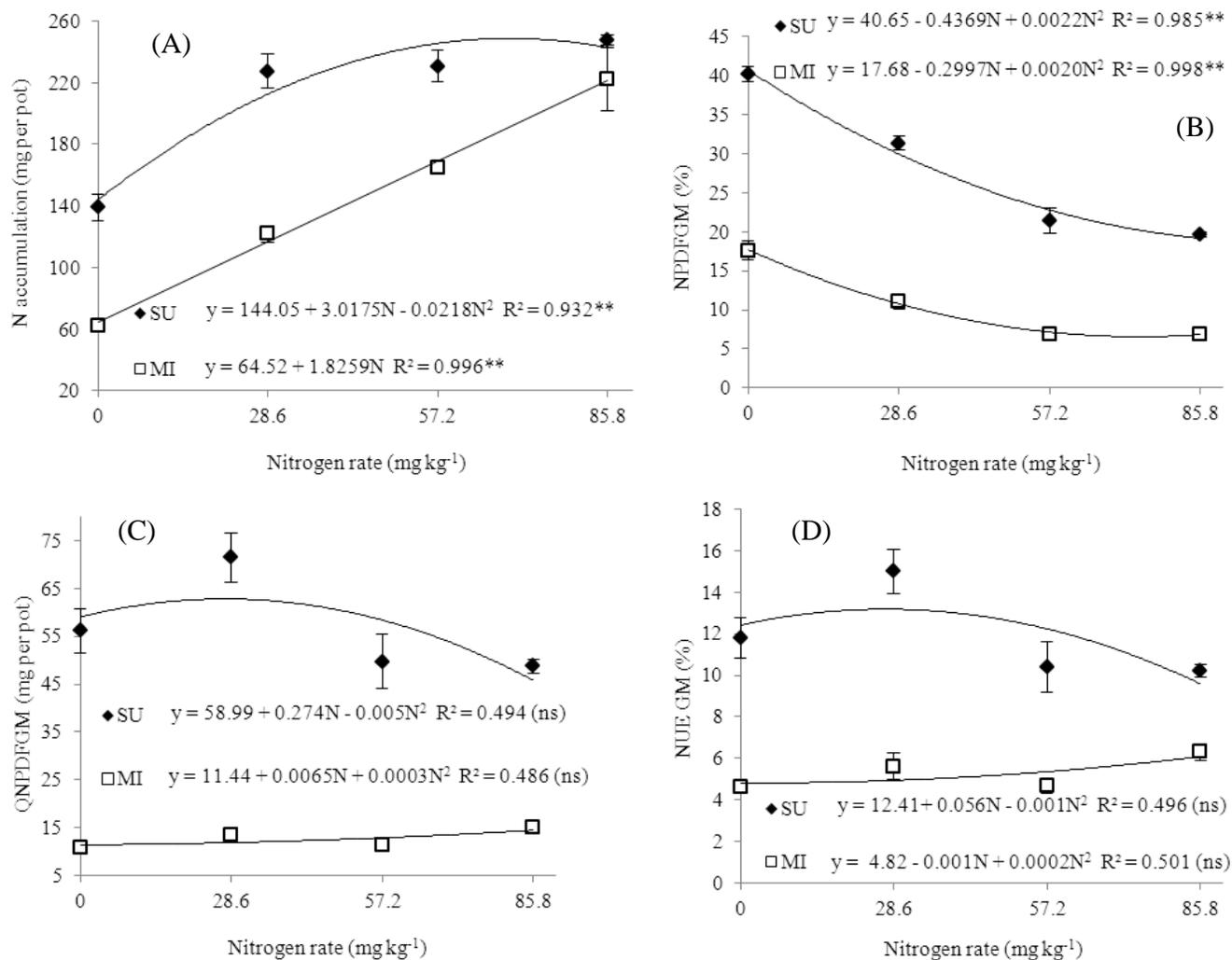


Figure 4. Nitrogen accumulation in rice grain (A), percent (NPDFGM) (B) and quantity (QNPDFGM) of nitrogen in rice grain derived from sunn hemp (SU) and millet (MI) (C) and N utilization efficiency (NUE) from green manures by the rice grain (D), as affected by N fertilizer rate. ns and ** not significant and significant by the F test at 1%, respectively.

the amount of N in rice derived from sunn hemp and the NUE from this GM were approximately five and two times greater than those with millet. This effect was observed probably due mainly to higher C/N ratio of millet as compared to sunn hemp that resulted in lower mineralization rate of the residue and probably immobilization of N under millet (Silva et al., 2006, 2010; Carvalho et al., 2011). Other studies have obtained values of NUE from sunn hemp to be between 10 and 37% for rice crop (Muraoka et al., 2002) and 8% for wheat (Araújo et al., 2005). Studies with other legumes (*Sesbania aculeata*) in rice grown area reported NUE of 19.3% (Azam, 1990) and of 19.7% for sorghum (Kurdali et al., 2007). Using GMs as N source, studies obtained values of NUE from GMs to be between 3 and 8% for rice (Muraoka et al., 2002), and 12% for corn crop (Scivittaro et al., 2003; Silva et al., 2010). In studies with three

legumes, Odhiambo (2010) showed that sunn hemp tend to release N at a faster rate, followed by lablab and velvet bean. The application of sunn hemp also showed potential to increase maize yield in smallholder farms (Odhiambo, 2011).

The low NUE from green manure observed in this study indicates that a majority of N from GM remained in the soil, probably as organic form (Azam et al., 1995, Amado et al., 2002; Muraoka et al., 2002; Ambrosano et al., 2003; Scivittaro et al., 2003). Studies reported that less than 50% of the N incorporated into soil as organic form is transformed into inorganic N by mineralization and the other part has been found to be in association with the soil microbial biomass (Mengel, 1996; Scivittaro et al., 2004).

Traditionally, both legume and non-legumes have been used as GM to develop sustainable agricultural systems

Table 1. Nitrogen accumulation (NA), quantity (QNPDFGM) and percentage (NPDFGM) of N in upland rice (straw + grain) derived from green manure and N utilization efficiency (NUE) from green manure by upland rice (straw + grain), as affected by green manure specie (sunn hemp and millet).

Green manure	NA	QNPDFGM	NPDFGM	NUE
	mg per pot		%	
Sunn hemp	347.9 ^{a*}	90.2 ^a	27.5 ^a	18.9 ^a
Millet	215.8 ^b	18.6 ^b	10.0 ^b	7.8 ^b
CV (%)	9.99	15.54	10.21	14.18

Values followed by different letters, in the same columns, do not differ by the Tukey test at 5%.

Table 2. Percentage (NPDFGM) and quantity (QNPDFGM) of N in upland rice (straw + grain) derived from root or shoot green manure and N utilization efficiency (NUE) of green manure by rice plant (straw + grain), as affected by green manure specie (sunn hemp and millet).

Green manure		Straw			Grain		
		NPDFGM	QNPDFGM	NUE	NPDFGM	NPDFGM	NUE
		%	mg per pot	%	%	mg per pot	%
Sunn hemp	Shoot	21.7 ^a	38.3 ^a	9.6 ^a	23.3 ^a	50.2 ^a	12.5 ^a
	Root	3.1 ^b	4.8 ^b	6.3 ^{ab}	3.0 ^b	7.5 ^b	9.8 ^{ab}
Millet	Shoot	5.2 ^b	8.3 ^b	4.6 ^b	5.8 ^b	12.2 ^b	6.8 ^b
	Root	2.0 ^b	3.2 ^b	5.3 ^b	2.0 ^b	4.1 ^b	6.8 ^b

Values followed by different letters, in the same columns, do not differ by the Tukey test at 5%.

due their numerous benefits which is related to improving soil fertility, water retention and reducing soil erosion (Fageria et al., 2005; Fageria, 2007). However, leguminous GMs are considered superior because substantial amount of biologically fixed N can be subsequently transferred to non-legume cereal crop under subsequent rotations (Fageria, 2007).

Nitrogen in upland rice derived from shoot and root of green manures

The highest percentage and amount of NPDFGM, both in upland rice grain or straw, was observed with application of sunn hemp shoot as compared to others treatments (Table 2). NUE of shoot and root of sunn hemp by rice (grain and straw) were similar, but NUE of sunn hemp shoot was higher as compared to the application of both shoot and root of millet.

The NUE from root (5.3%) and shoot (4.6%) of millet by rice straw was lower than N from sunn hemp shoot (9.6%) (Table 2), possibly due to slower mineralization of the millet residues with higher C/N ratio. Also, reimmobilization of millet residues mineralized N (Silva et al., 2006; Carvalho et al., 2011) may have occurred. Although, usually only the N content in shoot of GMs is considered, the roots is another important N and other nutrients source for succeeding crops (Araújo et al.,

2004; Scivittaro et al., 2004; Silva et al., 2008).

The NUE from sunn hemp shoot by rice (grain or straw) did not differ significantly from root of this legume (Table 2), and it was higher than the N recovery from millet root and shoots. The overall average NUE from GMs root by upland rice plants was 14.1%, whereas it was 16.8% for shoot.

Nitrogen use efficiency by upland rice derived from N fertilizer (urea)

Nitrogen use efficiency in rice plants (grain + straw) was not influenced by N fertilizer application rates under with or without GMs and it was on average, 53.7% (Figure 5A). Bronson et al. (2000) reported lower NUE of urea by rice crop (44%). Therefore, the NUE from urea in rice grain (34.8% on average) was not affected either by N fertilizer rate or application of different GMs (Figure 5B).

The NUE from urea in rice straw (18.9% on average) was significantly influenced by the application of GM. The application of millet reduced NUE from urea as compared to treatment without GM and with sunn hemp application (Figure 5C), probably due to N immobilization as shown by Corak et al. (1992). However, NUE did not differ significantly between treatments with application of sunn hemp and without addition of GM. The NUE from N fertilizer in rice straw was not influenced by N-urea rates.

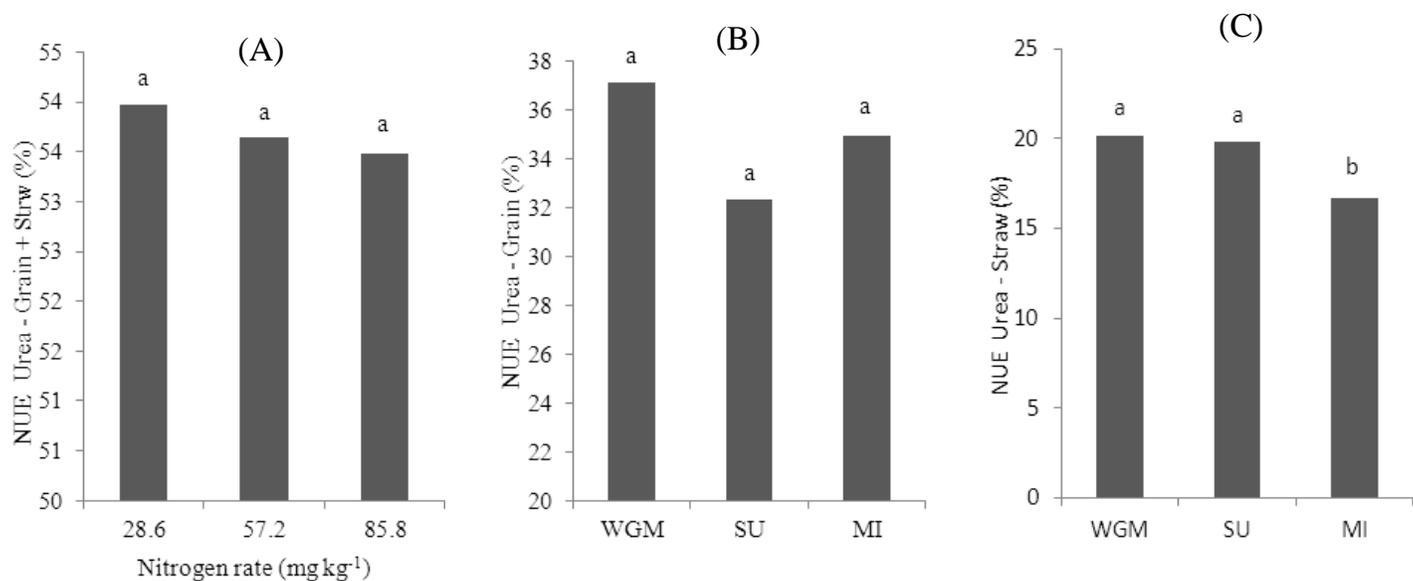


Figure 5. Nitrogen utilization efficiency (NUE) from fertilizer (urea) by upland rice: grain yield + straw biomass (A), grain yield (B) and straw biomass (C), as affected by N fertilizer rate and application of sunn hemp (SU), millet (MI) and without green manure addition (WGM). Bars followed by the same letter do not differ significantly according to Tukey test at 5%.

Table 3. Nitrogen in upland rice (straw + grain) derived from fertilizer urea (NPdff) and N utilization efficiency (NUE) as affected by N fertilizer application time, at seeding or topdressing, grown in the absence and presence of green manure (sunn hemp or millet).

Green manure	NPdff		NPdff		NUE	
	%		mg per pot		%	
	Seeding	Topdressing	Seeding	Topdressing	Seeding	Topdressing
Without GM	7.6 ^{Bab}	29.0 ^{Ab}	28.9 ^{Ba}	124.1 ^{Aa}	63.3 ^{Aa}	67.8 ^{Aa}
Sunn hemp	6.2 ^{Bb}	21.6 ^{Ac}	28.1 ^{Ba}	107.7 ^{Aa}	61.7 ^{Aa}	47.1 ^{Bb}
Millet	8.3 ^{Ba}	38.4 ^{Aa}	24.0 ^{Ba}	107.3 ^{Aa}	52.7 ^{Ab}	58.6 ^{Aa}
CV (%)	9.9	9.4	9.7	7.2	9.6	7.6
Average	7.4	29.7	27.0	113.0	59.2	57.8

Values followed by the same letters, for the same variable (lower case letter in columns and capital letter in row), do not differ by the Tukey test at 5%.

Nitrogen use efficiency as affected by time of N application (seeding and topdressing)

Percentage and amount of N in rice derived from fertilizer (NPdff) were greater when N fertilizer was applied at topdressing as compared to seeding (Table 3). In contrast, the percentage of N in rice derived from green manure (Table 1), the percentage of NPdff in upland rice (straw + grain) were greater in treatments with application of millet as compared to the sunn hemp (Table 3).

Nitrogen use efficiency of urea was not affected significantly by supply of N fertilizer at seeding or topdressing, in treatments without GM or with application of millet (Table 3). However, with application of sunn

hemp, the NUE of urea was greater when N fertilizer was applied at seeding (61.7%) as compared to topdressing (47.1%).

In this study, N recovery in rice root was not evaluated, thus the NUE of GM or urea by whole upland rice plant (root + shoot) were underestimated. Previous studies have been reported in literature that high amount of N fertilizer is present in plant roots under these production systems (Azam et al., 1995; Silva et al., 2006, 2008).

Results of this study showed that the application of GM, mainly sunn hemp, in combination with N fertilizer may be a fertilizer management practice to increase upland rice yield, improve N fertilizer efficiency and reduce inorganic N fertilizer inputs. As some of these GM are legumes, they have the potential to grow as rotational vegetation

with crops such as rice, maize and other cereal crops.

Conclusion

The introduction of sunn hemp to soils before rice crops showed greater upland rice grain yield and straw biomass as compared to millet and without application of green manure. As a legume, sunn hemp fixes atmospheric N and transfer part of this N to soil. Some of this N can be subsequently mineralized and made available to rice crops and subsequently increase yield. The NUE from sunn hemp by rice grain was higher than the NUE by the straw. The sunn hemp NUE by the rice plants was two times higher than that from the millet. The N-urea rates, with or without green manures, did not affect the NUE of fertilizer by rice plants. The NUE from shoot and root of GM by the rice plants were similar. With application of sunn hemp, the NUE from urea was observed when N fertilizer was applied at seeding as compared to topdressing. However, this effect was not observed with application of millet and without addition of GM. The study showed that the introduction of green manures, particularly legume green manures may help meet some of the N requirements of subsequent cereal crops such rice. Further field studies are important to assess the role of legume green manures in supplying N to subsequent crops.

Conflict of Interests

The authors have not declared any conflict of interests.

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