DIVISÃO 3 - USO E MANEJO DO SOLO

Comissão 3.1 - Fertilidade do solo e nutrição de plantas

EFFECT OF ^{15}N-LABELED HAIRY VETCH AND NITROGEN FERTILIZATION ON MAIZE NUTRITION AND YIELD UNDER NO-TILLAGE[1]

José Alan de Almeida Acosta[2], Telmo Jorge Carneiro Amado[3], Andreas de Neergaard[4], Mads Vinther[4], Leandro Souza da Silva[3] & Rodrigo da Silveira Nicoloso[5]

SUMMARY

This study evaluated the effect of hairy vetch (*Vicia villosa* Roth) as cover crop on maize nutrition and yield under no tillage using isotope techniques. For this purpose, three experiments were carried out: 1) quantification of biological nitrogen fixation (BNF) in hairy vetch; 2) estimation of the N release rate from hairy vetch residues on the soil surface; 3) quantification of ^{15}N recovery by maize from labeled hairy vetch under three rates of mineral N fertilization. This two-year field experiment was conducted on a sandy Acrisol (FAO soil classification) or Argissolo Vermelho distrófico arênico (Brazilian Soil Classification), at a mean annual temperature of 18 ºC and mean annual rainfall of 1686 mm. The experiment was arranged in a double split-plot factorial design with three replications. Two levels of hairy vetch residue (50 and 100 % of the aboveground biomass production) were distributed on the surface of the main plots (5 x 12 m). Maize in the sub-plots (5 x 4 m) was fertilized with three N rates (0, 60, and 120 kg ha^{-1} N), with urea as N source. The hairy vetch-derived N recovered by maize was evaluated in microplots (1.8 x 2.2 m). The BFN of hairy vetch was on average 72.4 %, which represents an annual input of 130 kg ha^{-1} of atmospheric N. The N release from hairy vetch residues was fast, with a release of about 90 % of total N within the first four weeks after cover crop management and soil residue application. The recovery of hairy vetch ^{15}N by maize was low, with an average of 12.3 % at harvest. Although hairy vetch was not directly the main source of maize N nutrition, the crop yield reached

8.2 Mg ha⁻¹, without mineral fertilization. There was an apparent synergism between hairy vetch residue application and the mineral N fertilization rate of 60 kg ha⁻¹, confirming the benefits of the combination of organic and inorganic N sources for maize under no tillage.

Index terms: Soil conservation, ^{15}N recovery, cover crops, N uptake.

RESUMO: *CONTRIBUIÇÃO DA ERVILHACA MARCADA COM [15]N E DA ADUBAÇÃO NITROGENADA NA NUTRIÇÃO E PRODUTIVIDADE DO MILHO NO SISTEMA PLANTIO DIRETO*

Neste trabalho foi avaliada, por meio de técnicas isotópicas, a contribuição da ervilhaca (Vicia villosa Roth) utilizada como cultura de cobertura na nutrição e produtividade do milho sob sistema plantio direto. Para isso, três estudos foram conduzidos: estimativa da fixação biológica de nitrogênio (FBN) na ervilhaca; avaliação da taxa de liberação de nitrogênio (N) da fitomassa da ervilhaca quando mantida na superfície do solo; e estimativa do ^{15}N recuperado pelo milho da ervilhaca isotopicamente marcada sob três taxas de fertilização mineral do milho. O experimento foi conduzido durante dois anos. O solo é um Argissolo Vermelho distrófico no Sistema Brasileiro de Classificação de Solos (Acrisol no sistema da FAO). As parcelas principais mediam 5 x 12 m e tinham dois níveis de aporte de resíduos de ervilhaca (50 e 100 % da produção da parte aérea de fitomassa). As subparcelas mediam 1,8 x 2,2 m. A FBN da leguminosa empregada como cultura de cobertura foi, em média, de 72,4 %, com aporte anual de 130 kg ha⁻¹ de N atmosférico. A liberação do N da fitomassa da ervilhaca foi rápida, com aproximadamente 90 % do total liberado ocorrendo nas primeiras quatro semanas após o corte e deposição dos resíduos. A recuperação do ^{15}N da ervilhaca pelo milho foi baixa: em média, 12,3 %. Embora a ervilhaca não tenha sido diretamente a principal fonte nitrogenada do milho, a produtividade obtida, sem adubação mineral, alcançou 8,2 Mg ha⁻¹. Foi observado aparente sinergismo entre o aporte de ervilhaca e a dose de 60 kg ha⁻¹ de adubação mineral, confirmando os benefícios da integração da adubação orgânica com a inorgânica como fontes de N no plantio direto de milho.

Termos de indexação: conservação do solo, ^{15}N recuperado, culturas de cobertura, N absorvido.

INTRODUCTION

Cover crops are planted to enhance soil fertility and quality and to increase the yield of the following cash crop (Santi et al., 2003). Before the green revolution, cover crops represented one of the most traditional farming techniques. Until the first half of the 20th century, leguminous green manure was the main N source for grain crops (Reeves, 1994). However, after the Second World War there was a drastic shift due to the increased availability of mineral N obtained by the Haber-Bosch process. By this industrial process, nitrogen (N) from air and hydrogen (H) from water are combined under high pressure (35–100 MPa) and temperature (300–400 ºC), resulting in ammonia and other compounds such as urea. However, the energetic cost of the Haber-Bosch process is high, with a demand of about 1.3 Mg of fossil fuel to produce 1 Mg of N (Marin et al., 2009).

The use of mineral N fertilizers has been identified as a driving factor for increasing the grain yield as demanded by the exponential growth of the world population. On the other hand, the intensive use of mineral N fertilization for grain crops has been associated with the contamination of natural resources. Consequently, environmentally friendly husbandry practices have been investigated in the last decades, especially the use of legume cover crops to substitute mineral N fertilization partially or even completely (Amado et al., 2002; Seo et al., 2006).

Studies have confirmed the efficiency of cover crops in increasing maize yield in no-till systems in Southern Brazil (Amado et al., 2000; Aita et al., 2001). Hairy vetch is a particularly important cover crop, due its efficiency in biological nitrogen fixation (BNF), providing N-rich crop residues that improve soil fertility and reduce erosion (Ebelhar et al., 1984; Aita et al., 2001). Therefore, the main effect of hairy vetch on maize yield has been associated to an increased soil N availability (Kramer et al., 2002). Howerver, studies with labeled hairy vetch showed that soil is the main destination of legume-derived N, rather than

the direct crop uptake (Harris & Hesterman, 1990; Scivittaro et al., 2003; Seo et al., 2006).

The combination of the legume hairy vetch with complementary mineral N fertilization seems to be a promising strategy for maize nutrition which could save inputs, ensure competitive grain yields and decrease environmental risks (Scivittaro et al., 2003; Seo et al., 2006). The synergic interaction of these N sources could also induce an additional increase in maize yield, since the temporal pattern of N release from cover crop residues and by mineral fertilization is not the same (Kramer et al., 2002; Hadas et al., 2002). In tropical and subtropical climate, the combination of slow and fast N-release sources could enhance the maize N uptake by improving the synchronism between nutrient supply and plant demand compared to the single use of N source (Aita et al., 2001).

The efficiency of organic maize N fertilization depends on: 1) the yield of the legume cover crop and the nutrient content of the residues, 2) the efficiency of the BNF process of the legume species and 3) the synchronism between N release from legume residues and maize N demand (Amado et al., 2000; Aita & Giacomini, 2003). These biological processes are determined by climate conditions and therefore require site-specific assessment. The objective of this study was to evaluate the efficiency of hairy vetch and its interaction with mineral N fertilization in maize nutrition and crop yield under conservation tillage using isotope techniques in Southern Brazil.

MATERIAL AND METHODS

The experiment was carried out from May 2003 to March 2005 at an experimental station of the Federal University of Santa Maria – UFSM, in Santa Maria, State of Rio Grande do Sul, Brazil (29 º 43 ' S; 53 º 42 ' W). The soil was a sandy Acrisol (FAO soil classification) or Argissolo Vermelho distrófico arênico (Brazilian Soil Classification) (Embrapa, 2006). At the beginning of the experiment, the soil chemical properties in the 0–0.20 m soil layer were determined as follows: clay: 160 g kg^{-1}; soil organic matter: 1.1 %; pH H$_2$0: 4.9; P (Mehlich-1): 27 mg dm^{-3}; cation exchange capacity (CEC): 4.2 cmol$_c$ dm^{-3}; K (Mehlich-1): 0.07 cmol$_c$ dm^{-3}; Ca: 1.5 cmol$_c$ dm^{-3}; Mg: 0.4 cmol$_c$ dm^{-3}; and Al: 0.3 cmol$_c$ dm^{-3}. The mean monthly temperature ranged from 9.3 to 31.8 ºC and the mean annual temperature was 19.3 ºC. The mean annual rainfall (1961-1990) was 1,686 mm, according to Cogo et al. (2006). The climate was classified as subtropical humid Cfa (Köppen classification).

At the end of the growing season in 2003 and 2004, five hairy vetch (*Vicia villosa* Roth) plant samples were collected to quantify aboveground biomass. The cover crops black oat (*Avena strigosa* Schreb) and forage radish (*Raphanus sativus* var. *oleiferus* Metzg.) were also sampled as reference in adjacent areas, to determine BNF in hairy vetch by the natural abundance technique. The plant samples were dried at 65 ºC to constant weight for determination of dry-matter biomass production. Then the samples were ground and sieved through 0.5 mm mesh. Carbon and N contents and ^{15}N abundance were determined by the mass spectrometer ANCA-CL (Europa Scientific Ltd, UK, 1996) at the Faculty of Life Sciences, University of Copenhagen, Denmark.

Mesh bags (0.18 x 0.19 mm, mesh size 0.5 mm) were used to evaluate the decomposition rate of hairy vetch aboveground residues during two years. Forty mesh bags were filled with 100 g of manually chopped hairy vetch residues and placed on the soil surface. The mesh bags were collected and analyzed after the 1st, 2nd, 3rd, 4th, 6th, 8th, and 13th week after maize sowing, with five replicates at each sampling. The samples were cleaned (soil free), dried, weighed, ground, and analyzed for C and N content by wet combustion and micro-Kjeldahl, respectively, following the Nelson & Sommers (1986) and Tedesco et al. (1995) methods.

To label hairy vetch with ^{15}N, the crop was grown in September 2003 on a 20 m^2 plot adjacent to the experimental area. First, 0.705 kg of 10 % ^{15}N-enriched (NH$_4$)$_2$SO$_4$ was dissolved in water and sprayed over 20 kg of dry clean sand. Then, the hairy vetch area was divided in 20 squares of 1.0 m^2. Finally, 1.0 kg of the ^{15}N fertilizer-enriched sand was evenly distributed on each square meter of the plot, equivalent to 78 kg ha^{-1} of total N. At the time of fertilization, hairy vetch was in an early development growth stage. After four weeks, the aboveground biomass was harvested, air-dried to constant weight and stored for later use. In the following year, hairy vetch was sown on the same plot, though without ^{15}N fertilization, i.e., the ^{15}N-labelling of the second year consisted of the residual effect of fertilization of the previous year.

The experiment was arranged in a double split-plot factorial design with three replications. In the main plots (5 x 12 m) two levels of hairy vetch biomass residue (50 and 100 % of the aboveground biomass production) were distributed on the soil surface. In the sub-plots (5 x 4 m) maize was fertilized with three N rates (0, 60, and 120 kg ha^{-1} N), with urea as N source. The hairy vetch-derived N, recovered by maize, was evaluated in microplots (1.8 x 2.2 m) located in the center of the sub-plots. The non-labeled hairy vetch aboveground biomass was completely removed from the microplots and replaced with ^{15}N-labeled hairy vetch aboveground residues. The amount of hairy vetch biomass placed on each plot was 1,800 and 3,600 kg ha^{-1} in 2003, and 2,305 and 4,610 kg ha^{-1} in 2004, according to 50 and 100 % of the dry-matter biomass production of each year.

In October, the maize hybrid AG 8021 was sown. The density was adjusted to 60,000 plants ha^{-1} by

manual thinning. Phosphorus and potassium was fertilized as 60 and 40 kg ha^{-1} of P_2O_5 and K_2O, respectively. Irrigation was applied when necessary. N fertilization was split in three applications: first at sowing (base) and two topdressings 30 and 45 days after maize emergence (DAE). Four maize plants were collected from each microplot 23, 52, 82, and 141 DAE in 2003, and 27, 47, 75, and 128 DAE in 2004 to evaluate ^{15}N recovery by maize. The maize samples were dried at 65 °C to constant weight to determine dry-matter yield. The two central maize rows of each microplot were collected at maize harvest and the cobs separated from the rest of the plants. The maize yield data were adjusted to 13 % moisture. Total N and ^{15}N plant uptake were determined as previously described.

The following formulas were used, as proposed by Giller & Wilson (1993):

$$\text{Natm (\%)} = \frac{(\% \ ^{15}N \ \text{ref.} - \% \ ^{15}N \ \text{hairy vetch}) \times 100}{(\% \ ^{15}N \ \text{ref.} - \% \ ^{15}N \ \text{atm})} \quad (1)$$

where Natm = Atmospheric-derived N; atm = 0.3663 % (^{15}N atmospheric standard); ref = Reference crops (black oat and forage radish); hairy vetch = biological nitrogen-fixing plant.

$$\text{Nhv (\%)} = \frac{(\% ^{15}N\text{-labeled maize excess} - \% ^{15}N \ \text{not labeled maize}) \times 100}{(\% \ ^{15}N\text{-labeled hairy vetch excess})} \quad (2)$$

where Nhv = nitrogen from hairy vetch.

The hairy vetch residue decomposition and N release rates were adjusted to a double-exponential model. The results were subjected to ANOVA by the F test and the means compared by Duncan´s test (p ≤ 0.05), using SAS statistical package, version 5.0 (SAS, 2004).

RESULTS AND DISCUSSION

The potential of hairy vetch as maize N source is directly linked to the efficiency of BNF, the biomass production and the decomposition pattern of the crop residue. In this study, BNF accounted for 66.6 - 78.2 % of the total N accumulated in hairy vetch biomass in 2003 and 2004, respectively, averaging 72.4 % in both years. On average, 184 kg ha^{-1} yr^{-1} N was accumulated in the hairy vetch aboveground biomass, of which around 130 kg ha^{-1} N was biologically fixed from the atmosphere (Table 1). Rochester & Peoples (2005) found higher BNF in hairy vetch than in this study, contributing to 90 % of the total N in the biomass. Giller & Wilson (1993) and Seo et al. (2006) reported hairy vetch N accumulation and biomass production ranging from 100 to 200 kg ha^{-1} N and from 2.5 to 5.0 Mg ha^{-1}, respectively, similar to the values found in the present study.

The hairy vetch residue decomposition and N release rates estimated by the remaining N content in the biomass were similar in both years investigated (Figure 2). Hadas et al. (2002) found two distinct decay constant rates of hairy vetch residue decomposition in a study carried out in California. The first decay constant was very high (0.4 day^{-1}) and was associated to the labile biomass fraction of hairy vetch, and the second was very low (0.008 day^{-1}), corresponding to the recalcitrant fraction of the biomass. In this study, two distinct decay rate constants were found, ranging from 0.12 to 0.32 day^{-1} for the labile fraction and from 0.005 to 0.007 day^{-1} for the recalcitrant fraction (Figure 1).

In the first year, the amount of N released from hairy vetch residues was about 74 % higher than in the second. This result was associated with the difference in the amount of accumulated N and the N concentration in the hairy vetch biomass between the years (Table 1). Therefore, N release from hairy vetch reached 75 and 50 kg ha^{-1} N in 2003 and 2004, respectively. In both years, about 90 % of total N was released in the first 30 days of residue decomposition (Figure 2b). The fast N release from hairy vetch aboveground biomass had been observed in other residue decomposition studies under conservation tillage systems in Southern Brazil (Amado et al., 2002; Aita & Giacomini, 2003).

Maize has a limited capacity of N uptake in the early growth stages. Consequently, the fast N release from hairy vetch in warm and wet environments leads to a sharp increase of soil mineral N content (Acosta, 2009) which could lead to N leaching if associated to

Table 1. Efficiency of biological nitrogen fixation (BNF) of hairy vetch and isotopic properties of the ^{15}N labeled legume residues in 2003 and 2004

BNF	2003	2004
N content (%)	4.34 $^{\pm 0.38 (3)}$	2.87 $^{\pm 0.19 (3)}$
^{15}N enrichment (atom %)	0.370 $^{\pm 0.03}$	0.367 $^{\pm 0.012}$
C/N ratio	10.6 $^{\pm 0.8}$	15.2 $^{\pm 0.89}$
Natm (%) [1]	66.6 $^{\pm 12.4}$	78.2 $^{\pm 6.6}$
Labeled legume residue properties		
Dry matter production (kg ha^{-1})	5400	4610
N content (%)	4.14	3.13
N in aboveground biomass (kg ha^{-1})	223.3	144.4
C/N ratio	11:1	14:1
^{15}N enrichment (atom %)	1.830	0.418
^{15}N excess (atom %) [2]	1.463	0.052
^{15}N excess (kg ha^{-1})	3.270	0.075

[1] Natm = atmospheric-derived N. Mean of reference plants (black oat and forage radish). [2] In relation to the natural atmospheric ^{15}N abundance of 0.3663 %. [3] Standard deviation (n = 5).
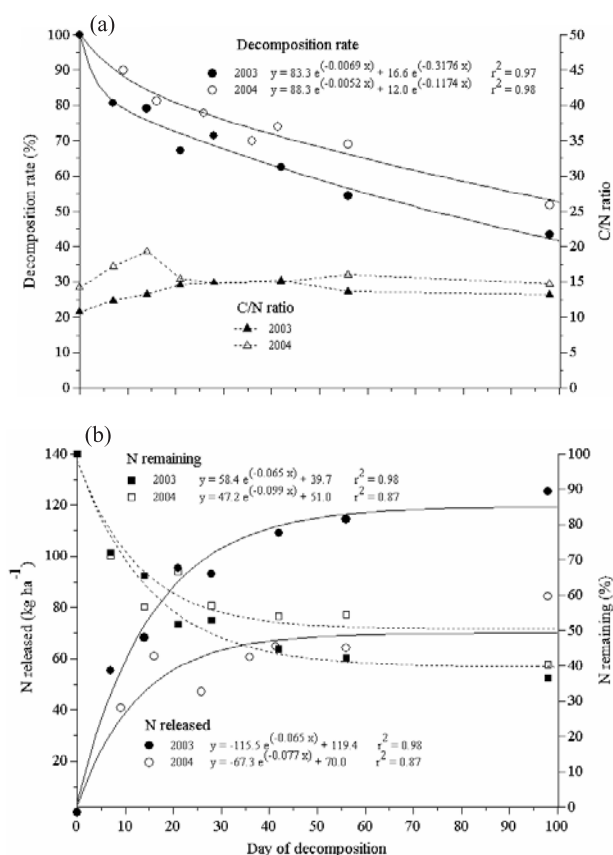
Figure 1. Decomposition rate of hairy vetch residues and C/N ratio (a), released N and remaining N percentage in residues (b) after 100 days of decomposition in 2003 and 2004. These legume residues were not labeled.

intense rainfall (Mai et al., 2003). In response, management strategies have been suggested avoiding fallow periods by planting maize immediately after cutting hairy vetch (Aita et al., 2001), reducing fertilization rates of base and topdressing mineral N (CQFSRS/SC, 2004) and more recently, the use of slow-release N sources (Kramer et al., 2002).

Maize ^{15}N recovery from hairy vetch-derived N was influenced by the amount of residue input, mineral N application rate and maize growth stage, with significant variations between years (Figure 2). The ^{15}N recovery rate in maize was highest at silking, with 26 % when associated to 60 kg ha^{-1} N mineral fertilization in 2004 compared to only 3 % in the control treatment (without mineral N fertilization), both in the case of input of 100 % hairy vetch biomass. In a review, Seo et al. (2006) reported an average of 15 % [15]N recovery by maize. In this study maize recovered on average 12.3 % [15]N at silking. The general trend of low recovery of hairy vetch-N by maize in this experiment could be associated partially to a poor synchronism between the fast N release from the legume residues after cutting (Figure 1) and the low maize N demand in the early growth stages (Table 2). To illustrate this fact, 50 days after hairy vetch

cutting, the amount of N released from the residues ranged from 69 to 114 kg N ha^{-1} (Figure 1) while the maize uptake ranged from only 2 to 6 kg ha^{-1} N (Figure 2).

The larger N recovery by maize at silking was associated with the higher maize demand for water and N (Fancelli & Dourado Neto, 2004). At maize silking in 2004, there was a significant interaction effect of mineral N fertilization rate with the residue input level on hairy vetch-derived N recovery by maize. In the treatment with 60 kg mineral N ha^{-1} and high residue input level (100 % of aboveground vetch production) higher absolute and relative maize ^{15}N recovery (39 kg ha^{-1} N and 26 %, respectively) was observed. At maturity , this interaction effect was only significant in 2003, when ^{15}N recovery by maize was 18 and 27 kg ha^{-1} N for the 50 and 100 % residue input levels, respectively (Figure 2).

Considering that in 2003 about 120 kg N ha^{-1} were released 100 days after cutting hairy vetch (Figure 1b) and that BNF was 66.6 % (Table 1), the relative N recovery at maize silking under mineral N rates was 8 and 14 % for the 100 and 50 % residue input levels, respectively (Figure 2). In 2004, the legume biomass release was 70 kg N ha^{-1} and the BNF was 78.2 %, the relative maize N recovery ranged from 10 to 19 % in the 50 % residue input treatment, and from 17 to 56 % for 100 % residue input. The highest legume cover crop derived-^{15}N recovery by maize was observed under normal residue input and 60 kg ha^{-1} N mineral fertilization rate treatment in 2004. Therefore, in this study the hairy vetch-derived N had two main destinations: maize plant nutrition and the soil N cycle through mineralization and immobilization processes. Previously, Scivittaro et al. (2003) and Seo et al. (2006) reported that the soil was the main destination of up to 55 % of the hairy vetch-derived N or up to twice the amount of N recovered by maize. Varco et al. (1993) explained these results by the low C/N ratio of hairy vetch residues, promoting high biological activity and rapid N release to soil. Crop residues maintained on the soil surface under no tillage could increase the N immobilization potential, which could decrease soil N availability temporarily. This effect could be positive from an environmental point of view since it contributes to reducing N losses by leaching under wet climate regimes in a period of low nutrient plant demand (Urquiaga et al., 1990).

Most of the ^{15}N taken up during the vegetative stage was translocated to maize grains in the reproductive stage (Figure 2), accounting for more than 75 % of the total ^{15}N uptake in both evaluation years. These results confim the high plant N mobility in maize crop (Uhart & Andrade, 1996).

The effect of mineral N fertilization on maize yields was only significant under low hairy vetch residue input (50 %) in both years (Table 2). However, the plant N content and total N accumulated in maize in
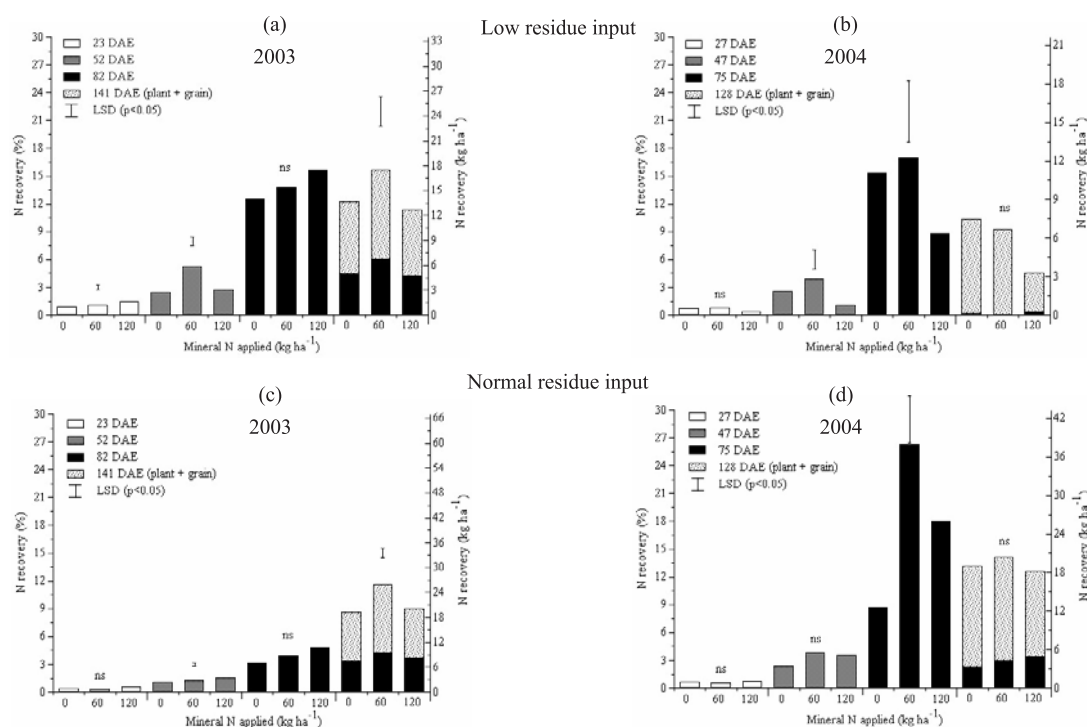
Figure 2. ^{15}N recovery by maize plants, absolute and relative to the amount of ^{15}N in the labeled hairy vetch residues at different maize stages as a function of low residue (a, b) and normal residue input levels (c, d) in 2003 and 2004. DAE= days after maize emergence. Vertical bars are the minimum significant difference by Duncan's test (p ≤ 0.05). ns = not significant.

the 100 % residue input treatment increased with mineral N fertilization, suggesting luxury consumption of N. Previously, Below (2002) had reported maize yields of up to 6.0 Mg ha^{-1} without mineral N fertilization in succession to hairy vetch and Mischler et al. (2010) reported maize grain yields of up to 9.6 Mg ha^{-1} after hairy vetch under favorable climatic and soil conditions. In this study, maize grain yield without mineral N fertilization ranged from 7.27 to 7.75 Mg ha^{-1} under limited residue input (50 %), and from 7.94 to 8.16 Mg ha^{-1} under high residue input (100 %). The maize grain yield was highest (10.24 Mg ha^{-1}) in the treatment combining 120 kg ha^{-1} mineral N with low hairy vetch residue input (50 %) where the maize plants accumulated 167.7 kg ha^{-1} N in aboveground biomass.

Taking environmental and economic aspects into account, the use of an intermediate mineral N fertilization rate (60 kg ha^{-1}) combined with hairy vetch residue input was an efficient strategy to improve plant nutrition, achieve competitive maize grain yields (> 7.0 Mg ha^{-1}), reduce mineral fertilizer input, and decrease the environmental impact (Kramer et al., 2002). Seo et al. (2006) concluded that the integrated use of organic and mineral N sources seems to be a step towards sustainability since the legume derived-N could replenish the soil reserves while the mineral fertilizer N could meet the plant demand. This study did not investigate the additional beneficial effects of hairy vetch as soil cover, or longer-term effects of FBN

on soil fertility. By the design of this study it was impossible to verify the fate of the residual N that was not recovered by maize, whether it was leached from or immobilized in the soil.

The higher mineral N fertilization rate decreased the effect of maize isotopic labeling such as the ^{15}N atom % and ^{15}N excess. The total N uptake by maize was increased under mineral N fertilization, causing a ^{15}N dilution effect, as previously reported by Hadas et al. (2002). According to Kramer et al. (2002), maize plants preferentially absorb mineral N before the hairy vetch-derived N, resulting in ^{15}N dilution.

The recovery by maize of second year hairy vetch-derived N ranged from 2.3 to 3.8 % or 2.5 to 6.5 kg ha^{-1} N, averaging 2.9 % in 2004 (on the plots where vetch residue was applied in 2003) (Table 3). Seo et al. (2006) also reported N recovery from hairy vetch by maize grains ranging from 2 to 4 % in the second year of their study. In the first year, the hairy vetch-derived N recovery in our study ranged from 10.1 for low (50 %) to 9.8 % for normal (100 %) residue input treatments, respectively, or 7.4 to 14.3 kg ha^{-1} N, for all mineral N fertilization rates and years of evaluation (Table 2). In conclusion, the hairy vetch-derived N in the second year (residual effect) was only 1/3 of the first year. This result suggests that the soil pool where the ^{15}N is stored could have a long half-life, as presumed by Seo et al. (2006), or that N losses due to leaching under wet subtropical climate conditions (> 1500 mm yr^{-1}) could be high.

Table 2. Maize grain yield and isotopic properties of the ^{15}N recovery in maize grains (first year) as a function of mineral N fertilization rates and levels of hairy vetch residue input in 2003 and 2004

Assessed caracteristics	Mineral N rate (kg ha^{-1}) - 2003			Mineral N rate (kg ha^{-1}) - 2004		
	0	60	120	0	60	120
	Low residue input (50 %)[1]					
Grain yield (Mg ha^{-1})	7.27 b [4]	8.18 ab	9.22 a	7.75 b	9.01ab	10.24 a
N (%)	1.14 b	1.25 ab	1.40 a	1.42 b	1.33 c	1.64 a
N (kg ha^{-1})	82.5 b	103.0 ab	129.0 a	109.8 b	119.6 b	167.7 a
^{15}N enrichment (atom %)	0.528 a	0.525 a	0.467 b	0.372 a	0.371 a	0.370 b
^{15}N excess (atom %)	0.161 a	0.159 a	0.100 b	0.006	0.005 b	0.004 c
Nhv (%)[3]	10.5 a	10.5 a	6.2 b	6.6 a	5.6 a	1.8 b
Nhv (kg ha^{-1})	8.7 ns	10.8	8.0	7.4 a	6.7 a	3.0 b
N recovery (%)	11.7 ns	14.6	10.7	10.2 a	9.3 a	4.2 b
	Normal residue input (100 %)[2]					
Grain yield (Mg ha^{-1})	7.94 ns[4]	9.20	9.47	8.16 ns	8.95	8.83
N (%)	1.21 ab	1.12 b	1.41 a	1.45 ns	1.74	1.73
N (kg ha^{-1})	96.1 b	103.2 b	133.2 a	119.5 ns	155.4	152.9
^{15}N enrichment (atom %)	0.555 b	0.607 a	0.510 c	0.375 ns	0.374	0.373
^{15}N excess (atom %)	0.189 b	0.241 a	0.143 c	0.009 ns	0.0072	0.007
Nhv (%)[3]	12.4 b	16.1 a	9.1 c	13.1 ns	10.2	8.7
Nhv (kg ha^{-1})	11.8 b	16.5 a	12.0 b	15.8 ns	16.2	13.3
N recovery (%)	7.9 b	11.1 a	8.1 b	11.0 ns	11.2	9.2

[1] Low residue input = 50 % of the hairy vetch dry matter production; labeled residues. [2] Normal residue input = 100% of the hairy vetch dry matter production; labeled residues. [3] Nhv = hairy vetch-derived N. [4] Values followed by the same letter at the same residue level and in the same evaluation year were not significantly different by Duncan´s test (p ≤ 0.05).

Table 3. Maize grain yield and isotopic properties of ^{15}N recovered by maize grains in 2004 (second year) as a function of input levels of labeled hairy vetch residues on the microplots in 2003 (residual effect)

Assessed caracteristics	Mineral N rate (kg ha^{-1})					
	0	60	120	0	60	120
	―― Low residue input (50 %)[1] ――			―― Normal residue input (100 %)[2] ――		
Grain yield (Mg ha^{-1})	8.64 ns[4]	7.99	9.20	9.09 ns	9.22	9.50
N (%)	1.33 b	1.66 a	1.46 ab	1.40 ns	1.36	1.54
N (kg ha^{-1})	114.3 ns	132.9	133.7	128.3 ns	125.5	146.2
^{15}N enrichment (atom %)	0.423 a	0.410 b	0.397 c	0.435 ns	0.443	0.422
^{15}N excess (atom %)	0.056 a	0.044 b	0.031 c	0.068 ns	0.077	0.055
Nhv (%)[3]	3.7 a	2.9 b	1.95 c	4.5 ns	5.1	3.6
Nhv (kg ha^{-1})	4.3 a	3.8 a	2.5 b	5.8 ns	6.5	5.3
N recovery (%)	3.8 a	3.4 a	2.3 c	2.6 ns	2.9	2.4

[1] Low residue input = 50% of the hairy vetch dry matter production, residues were not labeled in 2004. [2] Normal residue input = 100% of the hairy vetch dry matter production, residues were not labeled in 2004. [3] Nhv = hairy vetch-derived N. [4] Values followed by the same letter at the same residue level and in the same evaluation year were not significantly different by Duncan's test (p ≤ 0.05).

The limited hairy vetch-derived N recovery even at silking and grain filling maize growth stages indicate that most of the N was taken up from non-labeled sources, i.e., from the soil (Scivittaro et al., 2003) and mineral N fertilizer. Seo et al. (2006) reported that 53, 29, and 18 % of the N was taken up by maize from soil, mineral fertilizer and hairy vetch, respectively, in an experiment in which both legume residues and fertilizers had been labeled. The limited direct role of hairy vetch in maize plant nutrition contrasts with the positive effect on maize yield, reaching 80 % of the maximum productivity of the experiment without mineral N fertilization. A possible explanation for these divergent results could be that the N release from hairy vetch residues stimulates the soil inorganic N recycling. In this case, part of the N released from hairy vetch residues could be replaced by the non-labeled soil N. Sometimes this process occurs with a concomitant effect on the soil N mineralization rate ("priming effect"), sometimes without this effect, but always maintaining high soil N availability to the plants resulting in low ^{15}N recovery. In view of these processes, Hadas et al. (2002) claimed that evaluations based on the ^{15}N

labeling of hairy vetch underestimates the real value of N recovery and therefore of legume cover crop fertilization.

The adequate N availability in the early phenological stages of maize after hairy vetch is an important growth stimulus for the maize root system and increases photosynthesis rates and photosynthate accumulation (Uhart & Andrade, 1995). Fancelli & Dourado Neto (2004) reported that important components of maize grain yield such as number of rows per ear and number of grains per rows are prematurely defined (until the emission of the fourth leaf). Therefore, under the warm and wet subtropical climate -cover crops such as hairy vetch preceding maize could stimulate plant growth for high grain yields.

CONCLUSIONS

1. Biological nitrogen fixation was responsible for more than two thirds of the total nitrogen accumulated in the hairy vetch above-ground biomass. Nitrogen release from hairy vetch biomass under the warm and wet subtropical climate regime was fast, even when the residues were maintained on the soil surface.

2. The recovery of maize plants of hairy vetch-derived N was low. Legume derived-N was therefore not the main responsible N source for maize nutrition due to its rapid release and interaction with the soil N cycle. Nevertheless, maize produced high yields in succession to hairy vetch, without mineral N fertilization.

3. The combined use of hairy vetch as cover crop (organic source) with supplements of mineral N fertilization (mineral source) was an efficient strategy for improved plant nutrition, grain yield and environment protection in no-tillage maize in Southern Brazil.

ACKNOWLEDGEMENTS

LITERATURE CITED

ACOSTA, J.A.A. Dinâmica do nitrogênio sob sistema plantio direto e parâmetros para o manejo da adubação nitrogenada no milho. Santa Maria, Universidade Federal de Santa Maria, 2009. 200p. (Tese de Doutorado)

AITA, C.; BASSO, C.J.; CERETTA, C.A.; GONÇALVES, C.N. & DA ROS, C.O. Plantas de cobertura de solo como fonte de nitrogênio ao milho. R. Bras. Ci. Solo, 25:157-165, 2001.

AITA, C. & GIACOMINI, S.J. Decomposição e liberação de nitrogênio de resíduos culturais de plantas de cobertura de solo solteiras e consorciadas. R. Bras. Ci. Solo, 27:601-612, 2003.

AMADO, T.J.C.; MIELNICZUK, J. & FERNANDES, S.B.V. Leguminosas e adubação mineral como fontes de nitrogênio para o milho em sistemas de preparo do solo. R. Bras. Ci. Solo, 24:179-189, 2000.

AMADO, T.J.C.; MIELNICZUK, J. & AITA, C. Recomendação de adubação nitrogenada para o milho no RS e SC adaptada ao uso de culturas de cobertura do solo, sob sistema plantio direto. R. Bras. Ci. Solo, 26:241-248, 2002.

BELOW, F.E. Fisiologia, nutrição e adubação nitrogenada do milho. Inf. Agron. Potafos, 99:7-12, 2002.

COGO, C.M.; ELTZ, F.L.F. & CASSOL, E.A. Erosividade das chuvas em Santa Maria, RS, determinada pelo índice EI_{30}. R. Bras. Agrometeorol., 14:1-11, 2006.

COMISSÃO DE QUÍMICA E FERTILIDADE DO SOLO - CQFS/RS-SC. Manual de adubação e calagem para os Estados do Rio Grande do Sul e de Santa Catarina. 10.ed. Porto Alegre, SBCS/Núcleo Regional Sul/UFRGS, 2004. 400p.

EBELHAR, S.A.; FRYE, W.W. & BLEVINS, R.L. Nitrogen from legume cover crops for no-till corn. Agron. J., 76:51-55, 1984.

EMPRESA BRASILEIRA DE PESQUISA AGROPECUÁRIA - EMBRAPA - Centro Nacional de Pesquisas de Solos. Sistema brasileiro de classificação de solos. Rio de Janeiro, 2006. 412p.

FANCELLI, A.L. & DOURADO NETO, D. Produção de milho. Guaíba, Agropecuária, 2004. 360p.

GILLER, K.E. & WILSON, K.J. Nitrogen fixation in tropical cropping systems. Wallingford, CAB International, 1993. 313p.

HADAS, A.; DOANE, T.A.; KRAMER, A.W.; van KESSEL, C. & HORWATH, W.R. Modelling the turnover of 15N-labelled fertilizer and cover crop in soil and its recovery by maize. Eur. J. Soil Sci., 53:541-552, 2002.

HARRIS, G.H. & HESTERMAN, O.B. Quantifying the nitrogen contribution from alfalfa to soil and two succeeding crops using [15]N. Agron. J., 82:129-134, 1990.

KRAMER, A.W.; DOANE, T.A. & HORWATH, W.R. & van KESSEL, C. Short-term nitrogen-15 recovery vs. long-term total soil N gains in conventional and alternative cropping systems. Soil Biol. Biochem., 34:43-50, 2002.

MAI, M.E.M.; CERETTA, C.A.; BASSO, C.J.; SILVEIRA, M.J.; PAVINATO, A. & PAVINATO, P.S. Manejo da adubação nitrogenada na sucessão aveia-preta/milho no sistema plantio direto. Pesq. Agropec. Bras., 38:125-131, 2003.

MARIN, V.A.; BALDANI, V.L.D.; TEIXEIRA, K.R.S. & BALDANI, J.I. Fixação biológica de nitrogênio: Bactérias fixadoras de nitrogênio de importância para a agricultura tropical. Disponível em: <http://www.cnpab.embrapa.br/publicacoes>. Acesso em: 20 fev. de 2009.

MISCHLER, R.; DUIKER, S.W.; CURRAN, W.S. & WILSON, D. Hairy vetch management for no-till organic corn production. Agron. J.,102:355-362, 2010.

NELSON, D.W. & SOMMERS, L.E. Total carbon, organic carbon and organic matter. In: PAGE, A.L.; MILLER, R.H. & KENNEY, D.R., eds. Methods of soil analysis: Chemical and microbiological properties. Madison, American Society of Agronomy, 1986. Part 2. p.539-579.

REEVES, D.W. Cover crops and rotations. In: HATFIELD, J.L. & STEWART, B.A., eds. Crops residue management. Advances in soil science. Boca Raton, CRC Press, 1994. p.125-172.

ROCHESTER, I. & PEOPLES, M. Growing vetches (*Vicia villosa* Roth) in irrigated cotton systems: Inputs of fixed N, N fertiliser savings and cotton productivity. Plant Soil, 271:251-264, 2005.

SANTI, A.; AMADO, T.J.C. & ACOSTA, J.A.A. Black oat biomass and nutrient cycling as affected by nitrogen fertilization in soil under no-tillage. R. Bras. Ci. Solo, 27:1075-1083, 2003.

SAS Institute. SAS user's guide: statistic. Cary, 1990. 846p.

SCIVITTARO, W.B.; MURAOKA, T.; BOARETTO, A.E. & TRIVELIN, P.C.O. Transformações do nitrogênio proveniente de mucuna-preta e uréia utilizados como adubo na cultura do milho. Pesq. Agropec. Bras., 38:1427-1433, 2003.

SEO, J.; MEISINGER, J.J. & LEE, H. Recovery of nitrogen-15-labeled hairy vetch and fertilizer applied to corn. Agron. J., 98:245-254, 2006.

TEDESCO, M.J.; GIANELLO, C.; BISSANI, C.A.; BOHNEN, H. & VOLKWEISS, S.J. Análise de solo, plantas e outros materiais. 2.ed. Porto Alegre, Universidade Federal do Rio Grande do Sul, 1995. 174p. (Boletim Técnico de Solos, 5)

UHART, S.A. & ANDRADE, F.H. Nitrogen deficiency in maize. I. Effects on crop growth, development, dry matter partitioning, and kernel set. Crop Sci., 35:1376-1383, 1995.

URQUIAGA, S.; BODDEY, R.M. & ALVES, B.J.R. Dinâmica de nitrogênio no solo. In: SIMPÓSIO BRASILEIRO SOBRE NITROGÊNIO EM PLANTAS, 1., Itaguaí, 1990. Anais. Itaguaí, Universidade Federal Rural do Rio de Janeiro, 1990. p.181-251.

VARCO, J.J.; FRYE, W.W.; SMITH, M.S. & MACKWON, C.T. Tillage effects on nitrogen recovery by corn from a nitrogen-15 labeled legume cover crop. Soil Sci. Soc. Am. J., 57:750-756, 1993.