DOI: 10.5433/1679-0359.2011v32n3p995

Biologic dinitrogen fixation and nutrient cycling in cover crops and their effect on organic Conilon coffee

Fixação biológica de nitrogênio e ciclagem de nutrientes por plantas de cobertura e seus efeitos sobre café Conilon orgânico

Fábio Luiz Partelli[1]*; Henrique Duarte Vieira[2]; Enderson Petrônio de Brito Ferreira[3]; Alexandre Pio Viana[2]; José Antonio Azevedo Espindola[4]; Segundo Urquiaga[4]; Robert Michael Boddey[4]

Abstract

Notwithstanding its relevance, studies regarding nutrient cycling and biological dinitrogen fixation in Conilon coffee (*Coffee canephora* cv. Conilon) associated with cover plants are very scarce. Aiming to evaluate the contribution of cover crops for organic conilon production, a field experiment was carried out consisting of *Pennisetum glaucum*, and legume species *Canavalia ensiformis, Mucuna deeringiana* and *Cajanus cajan* (inoculated and non inoculated) cultivated between coffee trees, and spontaneous vegetation as cover crops. The experiment was carried out in Espírito Santo State- Brazil, in a 6.5 years old coffee crop production system. Chemical analyses of soil and vegetative parts of spontaneous and cover crops, as well as coffee leaf nutrients concentration were performed. Biological Nitrogen Fixation (BNF) was determined by the natural abundance method. BNF contributed with about 80% of the nitrogen accumulated by the leguminous plants, corresponding to 27 - 35 kg of N ha^{-1}. Concentration and accumulation of nutrients varied among cover crops. *Rhizobium* inoculation did not influence nutrient cycling or BNF. Legume plants partially supplied the nitrogen requirements of Conilon coffee. No significant effect of the treatments was observed on the nutrient concentration of Conilon coffee or on plant growth.
Key words: *Coffea canephora*, nutrient recycling, biological dinitrogen fixation, sustainable agriculture, isotopic composition

Resumo

Apesar de sua relevância, estudos sobre a ciclagem de nutrientes e fixação biológica do nitrogênio (FBN) em café Conilon (*Coffea canephora* cv. Conilon), associadas com plantas de cobertura, são escassos. Objetivou-se, com este trabalho avaliar a ciclagem de nutrientes, a FBN e o efeito que plantas de cobertura podem causar em lavoura de *C. canephora* cv. Conilon, sob manejo orgânico. O experimento foi conduzido no Estado do Espírito Santo - Brasil, em uma lavoura de café sob manejo orgânico, com 6,5 anos. Os tratamentos consistiram de testemunha (ausência de plantas de cobertura), *Pennisetum glaucum* e as leguminosas *Canavalia ensiformis, Mucuna deeringiana* e *Cajanus cajan*, com e sem inoculação de rizóbio específico. Efetuaram-se análises químicas de solo e da parte aérea das plantas espontâneas e de cobertura, bem como as concentrações foliares de nutrientes do cafeeiro.

[1] D.Sc., Prof. da Universidade Federal do Espírito Santo, UFES, Centro Universitário Norte do Espírito Santo. Rodovia BR 101 Norte, Km. 60, Bairro Litorâneo, 29932-540, São Mateus, ES. E-mail: partelli@yahoo.com.br

[2] D.Sc., Profs. da Universidade Estadual do Norte Fluminense Darcy Ribeiro, UENF, Campos dos Goytacazes, RJ. E-mail: henrique@uenf.br; pirapora@uenf.br

[3] D.Sc., Pesq. da Embrapa Arroz e Feijão, Santo Antônio de Goiás, GO. E-mail: enderson@cnpaf.embrapa.br

[4] D.Sc., Pesqs. da Embrapa Agrobiologia. Seropédica, RJ. E-mail: jose@cnpab.embrapa.br; urquiaga@cnpab.embrapa.br; bob@cnpab.embrapa.br

* Autor para correspondência

Recebido para publicação 15/11/2010 Aprovado em 17/06/2011

A contribuição da FBN foi determinada pela técnica da abundância natural. A FBN contribuiu com cerca de 80% do N acumulado pelas leguminosas, correspondendo a 27 - 35 kg N ha^{-1}. A concentração e acúmulo de nutrientes variaram entre as plantas de cobertura. A inoculação com rizóbio não influenciou ciclagem de nutrientes e a FBN. As leguminosas forneceram parte do N necessário para o cafeeiro. Não houve efeito significativo dos tratamentos sobre a concentração de nutrientes e o crescimento do cafeeiro.
Palavras-chave: *Coffea canephora*, reciclagem de nutrientes, fixação biológica de nitrogênio, agricultura sustentável, composição isotópica

Introduction

World coffee production in recent years has been superior to 6,600 million kg, mostly in developing countries, and Brazil is the largest world producer and exporter, supplying the total national demand, providing local employment and promoting rural development. In 2010, the national coffee production was 2,886 million kg, on a cultivated area of 2.3 million ha, with a total of 6.4 billions coffee trees (CONAB, 2011).

The adoption of soil and environmental conservative practices, such as the use of cover crops, play an important role on the establishment of the agrosystem sustainability. However that practice would only be acceptable if the use of such crops favor density and diversity of soil microorganisms, which promote nutrients cycling, contributing specially to the two soil N and P balance (OGOKE et al., 2009). Moreover, cover plants may improve soil structure (CARVALHO et al. 2004) and reduce infestation by spontaneous plants in various crops (PARTELLI et al., 2010).

Therefore, it is imperative to choose cover plants with high biomass production capability and well adapted to specific soil and climatic conditions. Among the desired characteristics, cover crop species should show a good tolerance to drought, low nutritional requirements, fast growth and good soil cover capability. The use of leguminous plants as cover crops promotes the biological nitrogen fixation (BNF) in association with specific bacteria (OLIVEIRA et al., 2002a; RICCI et al., 2005; PIETSCH; FRIEDEL; FREYER, 2007).

The reduction of the molecular nitrogen from the air (N_2) to a pair of ammonium molecules (2 NH_3), known as biological dinitrogen fixation (BNF) is mediated by the nitrogenase enzyme. The BNF occurs through a serie of biological processes, which catalyze the biological reduction of N_2 (SANTOS et al., 2007), by the expenditure of renewable energy (HOWARD; REES, 1996) derived from plant photosynthesis.

On the other hand, atmospheric N_2 may be reduced by industrial processes, which show low efficiency and require high inputs of fossil energy. According to Reich et al. (2001), world's nitrogen fixation by the industry is far above 80 million tons a year, while the BNF contributes with about 170 million tons annually (EPSTEIN; BLOOM, 2006). These findings may be influenced by the inoculation of the appropriate bacterial strains among other factors (OLIVEIRA et al., 2002a). Based on such considerations, the improvement of soil fertility and the replacement of N using N_2 fixing plants may result in satisfactory yields with low production costs and high sustainability.

Notwithstanding its relevance, studies regarding nutrient cycling and BNF in cover plants associated with Conilon coffee (*Coffee canephora* cv. Conilon) are very scarce. Moreover, chemical fertilizers and liming of acidic soils responds for 30% of the total coffee production costs. In Brazil, the total coffee production area requires approximately 200 thousand tons of nitrogen fertilizers per year, corresponding to an estimated cost of 200 million dollars (EMBRAPA, 2004). Therefore, the objective of this work was to evaluate the nutrient cycling, BNF promoted by cover crops on *C. canephora* cv. Conilon under organic management.

Material and Methods

The experiment was carried out in Espírito Santo State, Brazil. The experimental area was located at 80 m above sea level and geographical coordinates of 18o South latitude and 40o West longitude. The climate is typical tropical, with hot humid summers and dry winters. The average annual precipitation is around 1,200 mm with temperatures ranging from 12 ºC to 34 ºC (INCAPER, 2007). During the experimental period, part of the weather data were similar to the historical tendency, however low precipitation was observed in January and February, while in March precipitation was higher than the historical average. The experimental area was established in a 6.5 years old *C. canephora* cv. Conilon crop with 2.0 x 1.5 m spacing and cropped according to Brazilian organic production laws. In 2005 the plot was fertilized with 80 g of natural phosphate and 2 kg of compost per plant, corresponding to 300 kg and 10 m^3 ha^{-1}, respectively.

Soil chemical and texture analyses were performed according to Silva (1999) in four replicates. Analyses results showed a sandy-clay texture, with 79% sand, 5% silt and 16% clay. pH = 4.82; organic matter = 1.07 dag kg^{-1} and P = 4.52 (by Mehlich 1 extractor); K = 40.0; Zn = 2.48; Fe = 51.6; Mn = 17.5; Cu = 0.15 and B = 0.16 mg kg^{-1}. Concentration of Ca, Mg, Al, H+Al, sum of bases and cation-exchange capacity at pH=7 were 1.91; 0.55; 0.08; 2.86; 2.62; and 4.49 $cmol_c$ dm^{-3} and base saturation = 58.4%. Coffee leaves showed 25.0; 0.70; 13.4; 1.50; 0.43 and 0.14 g kg^{-1} of N, P, K, Ca, Mg and S, respectively and 9.7; 98.0; 25.6; 24.4; and 51.0 mg kg^{-1} of Zn, Fe, Mn, Cu and B.

Plots were composed by 3 lines of coffee with 7 m each, with 42 m^2 of total area. The statistical design was a randomized block in a factorial arrangement with additional treatments with four replicates. Treatments consisted of combination of spontaneous plants grown between rows, pearl millet (*Pennisetum glaucum* cv. ENA 1) and the legumes jack-bean (*Canavalia ensiformis*), velvet bean (*Mucuna deeringiana*) and pigeon pea (*Cajanus cajan*) with or without specific rhizobium for each species.

Millet and Leguminosae plants were established in October 2005, with 0.4 m spacing between plants in a single row between coffee rows. Twenty grams of natural phosphate per plant were added (equivalent to 250 kg ha^{-1}). Legumes seed inoculation was performed with rhizobium humid peat-inoculant at sowing. Two to six seeds were sowed for each legume and 20 seed for Pearl millet.

For mass weight determination, spontaneous plants were sampled 76 days after cover crops sowing, into a rectangle equivalent to 0.5 m^2 (1.0 x 0.5 m), near the third coffee plant, and between the Leguminous, millet and the coffee rows. Pearl millet, jack-bean and velvet bean were harvested at 76 days after sowing, when flowering; pigeon pea was harvested at 170 days after sowing, when flowering. The harvest of Pearl millet and Leguminous plants was performed on a useful area of 11 m^2. To determine dry mass, nutrients concentration and accumulation, and nitrogen contents biologically fixed by the leguminous plants 300 g of fresh mass was collected and dried in oven at 65 ºC from each cover crop and spontaneous plants.

Nutrient accumulation on leaves was obtained multiplying the entire dry mass of shoot plants by nutrient concentration. Contribution of the BNF by the legume plants was calculated by the natural abundance of ^{15}N ($\delta^{15}N$) method, according to Boddey et al. (2000), using a mass spectrometer Finniga MAT, model Delta Plus, Germany. The BNF contribution was calculated by the equation % BNF = 100($\delta^{15}N$ control plant - $\delta^{15}N$ fixing plant) / ($\delta^{15}N$ control plant - B); B is the proportion of ^{15}N in the fixing plant grown under total dependence of biological N_2 fixation; the value of -1 was used for Jack-bean and Velvet bean and -0,9 for Pigeon pea (BODDEY et al., 2000).

As a reference of non fixing plants (control plant), pearl millet (*Penisetum glaucum*), hairy beggarticks (*Bidens subalternans*) and coastal sand paspalum (*Paspalum maritimum*) were used, which were harvested in the interlines of the coffee plants.

The length of plagiotropic and orthotropic branches (young branches containing one or two pairs of leaves) located in the upper middle third of the plants were evaluated. Concentration of N, P and K in Conilon coffee leaves were also evaluated on different harvesting times throughout the experimental period: at the beginning, 76 days after sowing (first cover plants and spontaneous plants cutting), 170 days (pigeon pea cutting) and at 257 days after sowing date. Leaves were collected from the 3rd and/or 4th leaf node of the plagiotropic branches, starting from the apex, on branches located on the upper middle third of the plant. Data were submitted to the analysis of variance, considering the mathematical model used in experimental design, as previously described and mean comparisons were performed at the 5% level of probability by the Tukey test.

Results and Discussion

The analysis of variance showed that inoculation with rhizobium strains promoted a positive and significant effect on iron concentration only on leaves of Jack-bean (data not shown), in which inoculated plants showed about 23% more iron in the shoots as compared with non inoculated plants. For this reason, the mean tests for nutrient concentration among cover crops were performed using the mean values of the inoculated and non inoculated plants. For most of the studied nutrients, velvet bean showed greater concentration of them as compared with pigeon pea and pearl millet. Similar tendency was also observed for jack bean; however, with less pronounced effect (Table 1).

Table 1. Nutrient concentration in the shoots of cover plants.

Treatment	N	P	K	Ca	Mg	S	Zn	Fe	Mn	Cu	B
-			(g kg^{-1})						(mg kg^{-1})		
Spontaneous*	17.5bc	9.08a	22.8a	14.8ab	4.18a	0.95a	50.7ab	82.9a	80.3a	12.4a	22.1a
Pearl millet	12.1c	7.65ab	13.6b	5.30c	3.10b	1.28a	60.3a	46.3b	21.7b	4.98b	5.97c
Jack-bean	29.1a	8.34ab	13.0b	17.9a	2.66bc	0.96a	27.3d	55.8b	28.6b	5.38b	19.6a
Velvet bean	32.6a	10.2a	13.5b	13.4b	2.28c	1.26a	39.4bc	81.2a	66.1a	14.0a	20.3a
Pigeon pea	19.0b	5.39b	5.59c	7.19c	1.98c	0.46b	20.3d	51.7b	26.1b	6.53b	13.4b
CV (%)	12.81	18.08	20.72	11.18	13.08	18.07	15.01	11.53	34.81	24.07	9.06

* without Leguminous and Pearl millet plants.
Mean values followed by the same or none letter within the column are not statistically different by the Tukey test at 5% probability.
CV = coefficient of variance.

Pigeon pea dry biomass production was higher than the other cover crops (Table 2), when its maximum potential was considered. This may be explained by the fact that this species shows the longest vegetative period (170 days after sowing) when compared to all other cover crops. Nevertheless, at the other cover crops harvesting time (76 days after sowing), Pigeon pea showed the lowest biomass content among the legume plants, with only 397 kg ha^{-1} (Data not shown).

It was observed that total N cycling was higher in treatments with legumes when compared to treatments without cover plants and with Pearl millet, corroborating results obtained by Castro et al. (2004).

Generally, legume plants have high nitrogen contents in their tissues, resulting in significant N contribution for the soil-plant system after cover crops were cut (CASTRO et al., 2004; OGOKE et al., 2009). Besides, legume plants may improve both nitrogen fertilization and decrease of nitrogen losses in the soil-plant system, also promoting the improvement of soil fertility (CARVALHO et al., 2004).

For the other quantified nutrients, the concentrations found in spontaneous plants were generally higher (K and Mg) or similar to the other nutrients found in some cover plants, showing a good capability of spontaneous plants to cycle nutrients.

However, there are some research results showing that after three years of continuous legume cropping, some soil characteristics associated to organic carbon, to soil density, to soil porosity and to soil aggregates stability remained unaffected (NASCIMENTO et al., 2005), as well as to crop yield (CARVALHO et al., 2004). When all plant biomass and bases content returns to the soil, the acidity previously produced during the nitrogen assimilation, that may help P absorption, is neutralized during final biomass decomposition without modifications on soil pH (NYATSANGA; PIERRE, 1973).

A previous work by Colozzi Filho and Cardoso (2000), showed that Sunn hemp (*Crotalaria juncea*) between lines of *C. arabica*, increased the arbuscular mycorrhizal spores concentration in the coffee tree rhyzosphere, demonstrating that this agricultural practice may favor the presence of microorganism associated with coffee roots, favoring the increase of nutrient absorption like Phosphorous and Carbon (MARSCHNER; DELL, 1994). Besides, some studies have shown that association between arbuscular mycorrhiza and leguminous *Rhizobium* may improve biological N_2 fixation (CHALK et al., 2006).

The accumulation of nutrients in cover and spontaneous plants tissues (Table 2) shows the potential of these species for nutrient recycling, making these nutrients available for the coffee plant after cutting and decomposition. Similar results were observed by Oliveira et al. (2002b), for different cover crops species alone and in association with other crops.

Pearl millet has a great potential for biomass production (BOER et al., 2007). Nevertheless, in this work it showed low production of dry biomass (Table 2). This fact may be related with the sensitivity of this specie to low irradiances resulting from coffee shading, since Pearl millet has a C4 metabolism, needing high luminescence to develop its whole growth potential.

Table 2. Dry mass productivity (DM) and nutrient accumulation in the shoots of cover plants (CP) at the beginning of flowering, and from spontaneous plants sampled among the Leguminous and millet plants (SP) at the moment of cutting and addition of the stubble to the soil.

Treatment	DM (kg ha^{-1})		N (kg ha^{-1})		P (kg ha^{-1})		K (kg ha^{-1})		Ca (kg ha^{-1})		Mg (kg ha^{-1})	
-	CP*	SP	CP	SP	CP	SP	CP	SP	CP	SP	CP	SP
Spontaneous	1217b	1217a	21.2bc	21.2a	10.7ab	10.7a	27a	27a	18.0a	18.0a	5.2a	5.2a
Pearl millet	704b	271b	8.4c	4.7b	5.31b	2.5b	9.5b	6.2b	3.7b	4.0b	2.2b	1.1b
Jack-bean	1343b	413ab	39.3ab	7.2ab	11.2ab	3.8ab	17ab	9.4ab	23.9a	6.2ab	3.7ab	1.7ab
Velvet bean	1047b	440ab	33.9ab	7.7ab	10.6ab	4.0ab	14b	10ab	14.1ab	6.6ab	2.4b	1.9ab
Pigeon pea	2408a	679ab	44.7a	11.9ab	12.6a	6.2ab	14b	16ab	17.5a	10.1ab	4.7ab	2.9ab
CV (%)	32.0	72.9	32.2	74.6	29.8	73.3	36.9	72.2	36.7	73.6	34.5	74.7
Treatment	**S (kg ha^{-1})**		**Zn (g ha^{-1})**		**Fe (g ha^{-1})**		**Mn (g ha^{-1})**		**Cu (g ha^{-1})**		**B (g ha^{-1})**	
-	CP	SP	CP	SP	CP	SP	CP	SP	CP	SP	CP	SP
Spontaneous	1.1a	1.1a	61.8a	61.8a	101a	101a	94a	94a	14.9a	14.9a	27.7a	27.7a
Pearl millet	0.9a	0.3b	41.9a	13.7b	32.7b	22.4b	16c	22b	3.4b	3.4b	4.3b	6.0b
Jack-bean	1.3a	0.4ab	36.2a	20.9ab	74.0ab	34.2ab	38bc	33ab	7.2ab	5.1ab	26.2a	9.1ab
Velvet bean	1.3a	0.4ab	40.5a	22.3ab	85.8ab	36.5ab	68ab	35b	14.5a	5.4ab	21.2ab	9.7ab
Pigeon pea	1.1a	0.7ab	48.7a	34.4ab	124a	56.3ab	62ab	54ab	15.7a	8.4ab	32.4a	15.0ab

* In January Pigeon pea had only 397 kg ha^{-1}.
Mean values followed by the same or none letter within the column are not statistically different by the Tukey test at 5% probability.

Cover plants influenced spontaneous plants control (Table 2), corroborating results reported by Partelli et al. (2010). It is possible that this interference occurs due to a physical effect, making difficult the capture of light photons by the spontaneous plants, with the consequent detrimental effect on plant growth due to energy limitations and competition for water and nutrients (SAGE; KUBIEN, 2003). Besides, these leguminous plants have a fast initial growth and high biomass production, promoting high pressure over spontaneous plants and consequently exerting a better control. Some nutrients cycling in spontaneous plants were similar to cover plants (Table 2). According to Meda et al. (2002), spontaneous plants play an important role on nutrient cycling, limestone incorporation and soil protection against erosion. However they may compete with the cultivated crop for limiting resources (water, light and nutrients), through direct competition or secretion of allelopathic substances (KONG; WANG; XU, 2007), affecting coffee trees development (RONCHI; SILVA, 2006).

Natural abundance values of ^{15}N observed for legume plants were significantly lower than in the control (non fixing plants) used as reference (6.986 ‰), showing a great contribution of BNF in nitrogen nutrition by the leguminous plants studied; however, no significant difference was observed among leguminous plants, as well as, N content and concentration derived from BNF among leguminous plants did not show significant differences (Table 3). BNF contributed with 27 - 34 kg N ha^{-1} for the production system, without taking in account the roots contribution, which was not harvested. However its contribution for the N supply is considered very modest. These results can be partially explained by the low density of sowing used for the cover crops.

Table 3. Values of natural abundance of ^{15}N ($\delta^{15}N$) from leguminous plants used on green manure and contribution of nitrogen biological fixation (BNF).

Treatment	**$\delta^{15}N$ (‰)**	**BNF (%)**	**BNF (kg ha^{-1})**
Jack-bean	0.4683	81.37	31.82
Velvet bean	0.5100	80.51	27.43
Pigeon pea	0.8367	77.73	34.87
CV (%)	51.18	5.25	27.17

The value of $\delta^{15}N$ from non-fixing N_2 plant was 6.9867±0.8534 (‰) using 3 replications of each specie: *Pennisetum glaucum, Bidens subalternans* e *Paspalum maritimum*, as well as for Leguminous plants.
Mean values followed by the same or none letter within the column are not statistically different by the Tukey test at 5% probability.
CV = coefficient of variance.

Approximately 80% of the total accumulated N came from BNF (Table 3). Similar results were observed by Ramos et al. (2001), Alves et al. (2006), but lower values were observed by Castro et al. (2004). The contribution of BNF depends on plant biomass production, and may be underestimated due to the losses of N fixed transferred to the soil or to other plants on the associated crop system (SIERRA; NIGRE, 2006).

The high values of BNF observed may be related to low N content from the soil (not fertilised with industrial or soluble products and low content of organic matter) and the excellent availability of water from rain and irrigation during plants development. All these factors favor BNF (RAMOS et al., 2001; PIETSCH; FRIEDEL; FREYER, 2007).

The percentage of N from BNF is controversial. According to literature, Sunn hemp (*C. juncea*), has an average of 53% (CASTRO et al., 2004) to 70% (RAMOS et al., 2001) of N derived from BNF. For soybean, BNF was superior to 83% (ALVES et al., 2006). Pietsch, Friedel e Freyer (2007) observed that *Medicago sativa* incorporated 89-125 kg ha^{-1} of N to soil, with 27 to 33% originated from BNF. According to Ramos et al. (2001), for *Mucuna aterrima* the BNFamounts 40% of the N disposed in the soil. Ovalle et al. (2006) observed values of 74-94% for nitrogen from BNF in forage legume plants.

Seed inoculation with adequate fixing bacterial strains in legumes may promote an increase of 30% on BNF when compared with species not inoculated (OLIVEIRA et al., 2002a). That may be related to the natural presence of rhizobia in the experimental area, once in preceding years the area was already cultivated with legume plants.

Effect of the treatments on the growth of plagiotropic and orthotropic coffee branches was not observed (Table 4), corroborating results observed by Ricci et al. (2005). Thus, it is possible to state that variation on coffee foliar N, P and K concentrations occurred in different periods. This might be associated to coffee growth habit, showing variation on foliar nutrient concentrations according to season (PARTELLI et al., 2007). However, evaluation into the same period showed no differences among treatments (Table 5). According to Bergo et al. (2006) the efficiency of cover crops associated with *C. canephora* depends on the species, spacing, crop age and other factors that might influence the behavior of green manure.

Table 4. Vegetative growth (cm) of plagiotropic and orthotropic coffee branches in different periods: 76, 170 and 257 days after sowing Leguminosae and millet plants.

Treatment	Plagiotropic branches			Orthotropic branches		
-	0-76 days	76-94 days	94- 257 days	0-76 days	76-94 days	94- 257 days
Spontaneous	14.3	9.55	26.0	7.15	6.13	15.2
Pearl millet	14.5	10.6	26.6	7.58	8.20	18.0
Jack-bean	11.1	10.8	24.9	5.53	8.16	17.2
Velvet bean	12.5	8.74	23.1	7.26	7.43	17.6
Pigeon pea	16.9	11.2	30.4	7.86	6.54	16.6
CV (%)	24.73	37.58	23.01	23.35	34.71	23.35

Mean values followed by the same or none letter within the column are not statistically different by the Tukey test at 5% probability.
CV = coefficient of variance.

Table 5. Coffee leaf concentration of N, P and K in different periods: beggining of the experiment (N0, P0 and K0), first plant cutting (N1, P1 and K1), Pigeon pea cutting (N2, P2 and K2) period and after dry matter decomposition (N3, P3 and K3).

Treatment	N0	N1	N2	N3	P0	P1	P2	P3	K0	K1	K2	K3
-					………..…………… ($g\ ha^{-1}$) ……...……..……							
Control	25ab	28.3a	28.6a	21.8b	0.7c	1.42a	1.36a	1.06b	13.4b	20.0a	18.3ab	16.7ab
Pearl millet	25b	30.9a	29.9a	23.2b	0.7c	1.46a	1.35ab	1.06b	13.4a	18.1a	15.83a	14.4a
Jack-bean	25bc	29.1a	28.8ab	23.8c	0.7c	1.40a	1.24a	1.07b	13.4b	20.0a	16.8ab	16.9ab
Velvet bean	25bc	29.2a	28.4ab	22.8c	0.7c	1.43a	1.34ab	1.06b	13.4b	19.4a	16.9ab	16.7ab
Pigeon pea	25b	30.4a	27.3ab	24.4b	0.7c	1.40a	1.24ab	1.08b	13.4b	19.4a	15.5ab	16.7ab
CV (%)		6.52				8.98				14.55		

There were no differences on the foliar nutrient concentration within the column.
Mean values followed by the same or none letter within the column are not statistically different by the Tukey test at 5% probability.

Because of the carbon/nitrogen ratio and lignin and hemicelulose content, nutrient liberation from spontaneous and cover plants tissues to the cultivated plants occurs as a function of some factors, like the season (dry or humid) and legume plant species (ESPINDOLA et al., 2006). Nutrient decomposition may be higher than 50% for most nutrients on the first 30 days after cutting (BOER et al., 2007; FERREIRA et al., 2011). Therefore, cover crops submitted to cutting at the end of December supplied nutrients, even N from BNF in January and February, period of coffee bean production when the plant has a great need for nutrients. Pigeon pea supplied nutrients after July, two months before Conilon coffee blossom, an important period considering that green manure fertilization could begin before flowering (MALAVOLTA et al., 2002; MATOS et al., 2011).

Conclusion

Biological Nitrogen Fixation contributed with about 80% of the total nitrogen accumulated by the legume plants, equivalent to 27 – 35 kg N ha^{-1} as a function of the produced dry biomass; therefore, legume plants may supply part of the Conilon coffee nitrogen needs.

Concentration and accumulation of nutrients varied among cover crops.

During all the experimental period, no significant effect of the treatments on nutrient concentration and growth of Conilon coffee were observed.

Acknowledgements

The authors would like to thank the Fundação de Amparo à Pesquisa do Estado do Rio de Janeiro (FAPERJ), the CNPq for the fellow conceded to researchers, the Universidade Estadual do Norte Fluminense Darcy Ribeiro (UENF), the Instituto Capixaba de Pesquisa e Assistência Técnica (INCAPER), the EMBRAPA Agrobiology, for the support on the ^{15}N analysis and Mr. Jaudemir Bettim, for the experimental area.

References

ALVES, B. J. R.; ZOTARELLI, L.; FERNANDES, F. M.; HECKLER, J. C.; MACEDO, R. A. T.; BODDEY, R. M.; JANTALIA, C. P.; URQUIAGA, S. Fixação biológica de nitrogênio e fertilizantes nitrogenados no balanço de nitrogênio em soja, milho e algodão. *Pesquisa Agropecuária Brasileira*, Brasília, v. 41, n. 3, p. 449-456, 2006.

BERGO, C. L.; PACHECO, E. P.; MENDONÇA, H. A.; MARINHO, J. T. S. Avaliação de espécies leguminosas na formação de cafezais no segmento da agricultura familiar no Acre. *Acta Amazônica*, Manaus, v. 36, n. 1, p. 19-24, 2006.

BODDEY, R. M.; PEOPLES, M. B.; PALMER, B.; DART, P. J. Use of 15N natural abundance technique to quantify biological nitrogen fixation by woody perennials. *Nutrient Cycling il Agroecosystems*, Dordrecht, v. 57, n. 3, p. 235-270, 2000.

BOER, C. A.; ASSIS, R. L.; SILVA, G. P.; BRAZ, A. J. B. P.; BARROSO, A. L. L.; CARGNELUTTI FILHO, A.; PIRES, F. R. Ciclagem de nutrientes por plantas de cobertura na entressafra em um solo de Cerrado. *Pesquisa Agropecuária Brasileira*, Brasília, v. 42, n. 5, p. 1269-1276, 2007.

CARVALHO, M. A. C.; ATHAYDE, L. F.; SORATTO, R. P.; ALVES, M. C.; SÁ, M. E. Adubação verde e sistemas de manejo do solo na produtividade do algodoeiro. *Pesquisa Agropecuária Brasileira*, Brasília, v. 39, n. 12, p. 1205-1211, 2004.

CASTRO, C. M.; ALVES, B. J. R.; ALMEIDA, D. L.; RIBEIRO, R. L. D. Adubação verde como fonte de nitrogênio para a cultura da berinjela em sistema orgânico. *Pesquisa Agropecuária Brasileira*, Brasília, v. 39, n. 8, p. 779-785, 2004.

CHALK, P. M.; SOUZA, R. F.; URQUIAGA, S.; ALVES, B. J. R.; BODDEY, R. M. The role of arbuscular mycorrhiza in legume symbiotic performance. *Soil Biology and Biochemistry*, Brisbane, v. 38, n. 9, p. 2944-2951, 2006.

COLOZZI FILHO, A.; CARDOSO, E. J. B. N. Detecção de fungos micorrízicos arbusculares em raízes de cafeeiro e de crotalária cultivada na entrelinha. *Pesquisa Agropecuária Brasileira*, Brasília, v. 35, n. 10, p. 2033-2042, 2000.

COMPANHIA NACIONAL DE ABASTECIMENTO

- CONAB. *Café:* segunda estimativa maio 2011. Disponível em: <http://www.conab.gov.br/>. Acesso em: 07 jun. 2011.

EMPRESA BRASILEIRA DE PESQUISA AGROPECUÁRIA - EMBRAPA. *Consórcio Brasileiro de pesquisas e desenvolvimento do café*. Brasília: EMBRAPA, 2004, 147 p.

EPSTEIN, E.; BLOOM, A. J. *Nutrição mineral de plantas:* princípios e perspectivas. 2. ed. Londrina: Planta, 2006. 403 p.

ESPINDOLA, J. A. A.; GUERRA, J. G. M.; ALMEIDA, D. L.; TEIXEIRA, M. G.; URQUIAGA, S. Decomposição e liberação de nutrientes acumulados em leguminosas herbáceas perenes consorciadas com bananeira. *Revista Brasileira de Ciência do Solo*, Viçosa, v. 30, n. 2, p. 321-328, 2006.

FERREIRA. E. P. B.; STONE, L. F.; PARTELLI, F. L. DIDONET, A. G. Produtividade do feijoeiro comum influenciada por plantas de cobertura e sistemas de manejo do solo. *Revista Brasileira de Engenharia Agrícola e Ambiental*, Campina Grande, v. 15, n. 7, p. 695-701, 2011.

HOWARD, J. B.; REES, D. C. Structural basis of biological nitrogen fixation. *Chemistry*, Boulder, v. 96, n. 7, p. 2965-2982, 1996.

INSTITUTO CAPIXABA DE PESQUISA, ASSISTÊNCIA TÉCNICA E EXTENSÃO RURAL - INCAPER. *Caracterização climática do município de Jaguaré*. 2007. Disponível em: <http://siag.incaper.es.gov.br/jaguare_carac.htm>. Acesso em: 02 nov. 2010.

KONG, C-H.; WANG, P.; XU, X-H. Allelopathic interference of Ambrosia trifida with wheat (*Triticum aestivum*). *Agriculture, Ecosystems and Environment*, Zürich, v. 119, n. 3/4, p. 416-420, 2007.

MALAVOLTA, E.; FAVARIN, J. L.; MALAVOLTA, M.; CABRAL, C. P.; HEINRICHS, R.; SILVEIRA, J. S. M. Repartição de nutrientes nos ramos, folhas e flores do cafeeiro. *Pesquisa Agropecuária Brasileira*, Brasília, v. 37, n. 7, p. 1017-1022, 2002.

MARSCHNER, H.; DELL, B. Nutrient uptake in mycorrhizal symbiosis. *Plant and Soil*, Crawley, v. 159, n. 1, p. 89-102, 1994.

MATOS, E. S.; CARDOSO, I. M.; SOUTO, R. L.; LIMA, P. C.; MENDONÇA, E. S. Characteristics, Residue Decomposition, and Carbon Mineralization of Leguminous and Spontaneous Plants in Coffee Systems. *Communications in Soil Science and Plant Analysis*, Georgia, v. 42, n. 5, p. 489-502, 2011.

MEDA, A. R.; PAVAN, M. A.; MIYAZAWA, M.; CASSIOLATO, M. E. Plantas invasoras para melhorar a eficiência da calagem na correção da acidez subsuperficial do solo. *Revista Brasileira de Ciência do Solo*, Viçosa, v. 26, n. 3, p. 647-654, 2002.

NASCIMENTO, J. T.; SILVA, I. F.; SANTIAGO, R. D.; SILVA NETO, L. F. Efeito de leguminosas nos atributos físicos e carbono orgânico de um Luvissolo. *Revista Brasileira de Ciência do Solo*, Viçosa, v. 29, n. 5, p. 825-831, 2005.

NYATSANGA, T.; PIERRE, W. H. Effect on nitrogen fixation by legumes on soil acidity. *Agronomy Journal*, Madison, v. 65, p. 936-940, 1973.

OGOKE, I. J.; IBEAWUCHI, I. I.; NGWUTA, A. A.; TOM, C. T.; ONWEREMADU, E. U. Legumes in the cropping systems of Southeastern Nigeria. *Journal of Sustainable Agriculture*, California, v. 33, n. 8, p. 823-834, 2009.

OLIVEIRA, A. L. M.; URQUIAGA, S.; DOBEREINER, J.; BALDANI, J. I. The effect of inoculating endophytic N_2-fixing bacteria on micropropagated sugarcane plants. *Plant and Soil*, Crawley, v. 242, n. 2, p. 205-215, 2002a.

OLIVEIRA, T. K.; CARVALHO, G. J.; MORAES, R. N. S. Plantas de cobertura e seus efeitos sobre o feijoeiro em plantio direto. *Pesquisa Agropecuária Brasileira*, Brasília, v. 37, n. 10, p. 1079-1087, 2002b.

OVALLE, C.; URQUIAGA, S.; DEL POZO, A.; ZAGAL, E.; ARREDONDO, S. Nitrogen fixation in six forage legumes in Mediterranean central Chile. *Acta Agriculturae Scandinavica Section B - Plant Soil Science*, Stockholm, v. 56, n. 4, p. 277-283, 2006.

PARTELLI, F. L.; VIEIRA, H. D.; CARVALHO, V. B.; MOURÃO FILHO, F. A. A. Diagnosis and recommendation integrated system norms, sufficiency range, and nutritional evaluation of Arabian coffee in two sampling periods. *Journal of Plant Nutrition*, Athens, v. 30, n. 10, p. 1651-1667, 2007.

PARTELLI, F. L.; VIEIRA, H. D.; FREITAS, S. P.; ESPINDOLA, J. A. A. Aspectos fitossociológicos e manejo de plantas espontâneas utilizando espécies de cobertura em cafeeiro Conilon orgânico. *Semina: Ciências Agrárias*, Londrina, v. 31, n. 3, p. 605-618, 2010.

PIETSCH, G.; FRIEDEL, J. K.; FREYER, B. Lucerne management in an organic farming system under dry site conditions. *Field Crops Research*, Warwick, v. 102, n. 2, p. 104-118, 2007.

RAMOS, M. G.; VILLATORO, M. A. A.; URQUIAGA, S.; ALVES, B. J. R.; BODDEY, R. M. Quantification of the contribution of biological nitrogen fixation to tropical green manure crops and the residual benefit to a subsequent maize crop using ^{15}N-isotope techniques. *Journal Biotechnology*, Bielefeld, v. 91, n. 2/3, p. 105-115, 2001.

REICH, P. B.; KNOPS, J.; TILMAN, D.; CRAINE, J.; ELLSWORTH, D.; TJOELKER, M.; LEE, T.; WEDINK, D.; NAEEM, S.; BAHAUDDIN, D.; GEORGE, D.; HENDREY, G.; JOSE, S.; WRAGE, K.; GOTH, J.; BENGSTON, W. Plant diversity enhances ecosystem responses to elevated CO_2 and nitrogen deposition. *Nature*, London, v. 410, n. 6830, p. 809-812, 2001.

RICCI, M. S. F.; ALVES, B. J. R.; MIRANDA, S. C.; OLIVEIRA, F. F. Growth rate and nutritional status of an organic coffee cropping system. *Scientia Agricola*, Piracicaba, v. 62, n. 2, p. 138-144, 2005.

RONCHI, C. P.; SILVA, A. A. Effects of weed species competition on the growth of young coffee plants. *Planta Daninha*, Viçosa, v. 24, n. 2, p. 415-423, 2006.

SAGE, R. F.; KUBIEN, D. S. Quo vadis C_4? An ecophysiological perspective on global change and the future of C_4 plants. *Photosynthesis Research*, New York, v. 77, n. 2/3, p. 209-225, 2003.

SANTOS, P. C.; MAYER, S. M.; BARNEY, B. M.; SEEFELDT, L. C.; DEAN, D. R. Alkyne substrate interaction within the nitrogenase MoFe protein. *Journal of Inorganic Biochemistry*, Columbia, v. 101, n. 11-12, p. 1642-1648, 2007.

SIERRA, J.; NYGREN, P. Transfer of N fixed by a legume tree to the associated grass in a tropical silvopastoral system. *Soil Biology & Biochemistry*, Oxford, v. 38, n. 7, p. 1893-1903, 2006.

SILVA, F. C. *Manual de análises químicas de solos, plantas e fertilizantes*. Brasília: EMBRAPA, 1999. 370 p.