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NITROGEN BALANCE AND FERTIGATION USE EFFICIENCY IN A FIELD COFFEE CROP

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□ *Brazil is a major world coffee producer, using increasing quantities of nitrogen (N) fertilizer as the monoculture expands across the savannas. The fate and efficiency of this fertilizer N were evaluated for one complete cropping cycle using ^{15}N tracer, permitting an N balance at harvest. Annual rates of 200, 400, 600, and 800 kg N ha^{-1} year^{-1} of ^{15}N-labeled urea and an unfertilized control were applied every 14 days via fertigation. The N concentration, percentage of N derived from fertilizer, quantity of N derived from fertilizer, and percentage of nitrogen derived from fertilizer per N rate was assessed for 8-year-old coffee trees. The most efficient N use was with 200 kg ha^{-1} year^{-1} because it presented the lowest losses and highest N recoveries in the crop. Conversely, the least sustainable rate was 800 kg ha^{-1} year^{-1}, which presented the greatest losses and the lowest N recovery in the whole plant.*

Keywords: N loss, ^{15}N, *Coffea arabica*, N recovery

INTRODUCTION

One of the major agricultural challenges is to keep increasing food production for a continuously growing population with the least inputs and low environmental impact. One contribution to this goal is through improving the efficiency of nutrient use by crop plants, particularly nitrogen (N). This will assist by reducing N losses, decreasing environmental pollution, and

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lowering unit production costs. Nitrogen use efficiency has been addressed by researchers with different approaches, depending on the purposes of the studies (see e.g. Lees et al., 2000; Hirel et al., 2007; Weih et al., 2011; Motior et al., 2011; Khalil et al., 2011), but can conveniently be assessed either as yield obtained per unit N applied or total N recovery of nitrogenous fertilizers.

To increase N-use efficiency through improved management practices it is necessary to know both the N utilization by the plant and its fate in the environment under different conditions. Complete N balances in the soil-plant system using nitrogen fifteen isotope (^{15}N) tracer techniques are invaluable tools to achieve these objectives (Ma and Dwyer, 1998; Cassman et al., 2002). Most field studies using ^{15}N-labeled fertilizer have been on annual plant systems and there is a knowledge gap for woody perennial crops, which have different physiological and nutritional needs (Marschner, 1995). Most reports dealing with perennials consist of experiments performed with seedlings, young plants or adult plants with confined roots (Boaretto et al., 1999; Bustamante et al., 1997; Lea-Cox et al., 2001; Natale and Marchal, 2002; Quiñones et al., 2007; Martínez-Alcántara et al., 2012). Very few studies have been carried out under true field conditions with mature perennial crop plants [e.g. Feigenbaum et al. (1987) and Mattos-Júnior et al. (2003) for citrus, and Nario et al. (2003) for peach]. The scarcity of studies on N absorption by woody perennials in the field using the ^{15}N isotope tracer technique is explained by the large size plants and their relatively slow growth rate, which do not lend themselves to short-term studies, related sampling and experimental costs, and the high rates of fertilizer application which make ^{15}N experiments very expensive.

An economically important woody perennial crop is coffee (*Coffea arabica* L.), for which there is little information on fertilizer N-use efficiency. One study on the fate of N in the field was carried out using young (3 to 5-year-old) coffee plants (Fenilli et al., 2008; Fenilli et al., 2007a, 2007b; Reichardt, et al. 2009). The main global coffee producer is Brazil, and new plantations are being established away from the traditional south/southeast region to the central savannah region, locally called cerrado. In these regions, the N fertilizer rates usually applied to coffee crops (600 to 800 kg ha^{-1}) are far above traditional recommendations of 150 to 450 kg ha^{-1}, with very little apparent scientific justification. These cerrado areas are very flat, with sandy soils and a readily available supply of fresh water for irrigation. Farmers employ fertigation with these high N rates divided in many portions in irrigation water, almost continuously over the whole cropping cycle. Although this N fertilizer splitting may avoid some nitrate leaching, this type of management may not provide for optimal N fertilizer use efficiency.

Nitrogen absorption changes in plant compartments over time in a fertigated coffee crop has been previously reported (Bruno et al., 2011), however this did not include the environmental fate of the applied fertilizer N. Thus the present study reports on the fate and efficiency of fertilizer N use through a complete N balance made at bean harvest. The experiment was carried out in the field, in a monoculture under conventional tillage, with mature coffee plants receiving 0, 200, 400, 600, and 800 kg ha^{-1} of N via fertigation. The goals were: i) making a fertilizer N balance in the soil-plant system for one productive cycle, with special focus on N losses by leaching; ii) examining the N-use efficiency by coffee plants as a function of the rate of N application; and iii) providing a scientific basis for improved fertilization management.

MATERIALS AND METHODS

Experimental Area

The experiment was carried for a complete coffee crop cycle in a cerrado soil, from 1 August 2008 to 24 July 2009, at a commercial coffee farm located in the central highlands of the Bahia State, Brazil, 11°46’00” S and 45°43’32” W, 740 m above sea level. The area presents with almost 0% slope and abundant fresh water for irrigation. The soil is sandy (80% of sand in the 0–0.2m layer and 70% in the 0.8–1.0m layer), classified as a “Latossolo Vermelho-Amarelo Alumínio típico” (EMBRAPA, 2006) or as a Typic Hapludox (Soil Survey Staff, 2010), of low natural fertility (Table 1) with an available water capacity of 85mm in the top 1m layer. The climate is tropical sub-humid (Aw) according to Köppen (1931), with yearly average air temperature of 25°C and, although annual rainfall ranges from 800 to 1,800 mm, there is a well-defined dry season that requires additional water supply for perennial crops, typically applied through central pivot irrigation under conventional tillage.

TABLE 1 Chemical characteristics of the Typic Hapludox soil for different layers before start of the experiment

M		g dm^{-3}	mg dm^{-3}		mmol$_c$ dm^{-3}							%	
Depth	pH^1	OM^2	P^3	S^4	K^3	Ca^3	Mg^3	Al^5	$H+Al^6$	SB^7	CEC^8	V^9	m^{10}
0–0.2	4.7	25	114	10	2	23	9	3	31	34	65	52	8
0.2–0.4	3.6	20	40	21	1	5	3	9	34	9	43	21	50
0.4–0.6	3.8	16	5	60	0.8	4	2	9	31	6.8	37.8	18	57
0.6–0.8	3.6	14	1	72	0.8	3	1	9	31	4.8	35.8	13	65
0.8–1.0	3.8	14	1	96	0.8	2	1	10	31	3.8	34.8	11	72

[1]Active acidity by $CaCl_2$ (0.01 mol l^{-1}) method. [2] Organic matter by colorimetry. [3] Phosphorus, potassium, calcium and magnesium by ion exchange resin method. [4] Sulfur by turbidimetry method. [5] Exchangeable aluminum by titrimetric method (1 mol l^{-1}). [6] Potential acidity by pH SMP method. [7] Sum of bases. [8] Cation exchange capacity. [9] Base saturation (100×SB/CEC). [10] Aluminum saturation (100×$Al_3{}^+$/effective CEC) Raij et al. 2001.

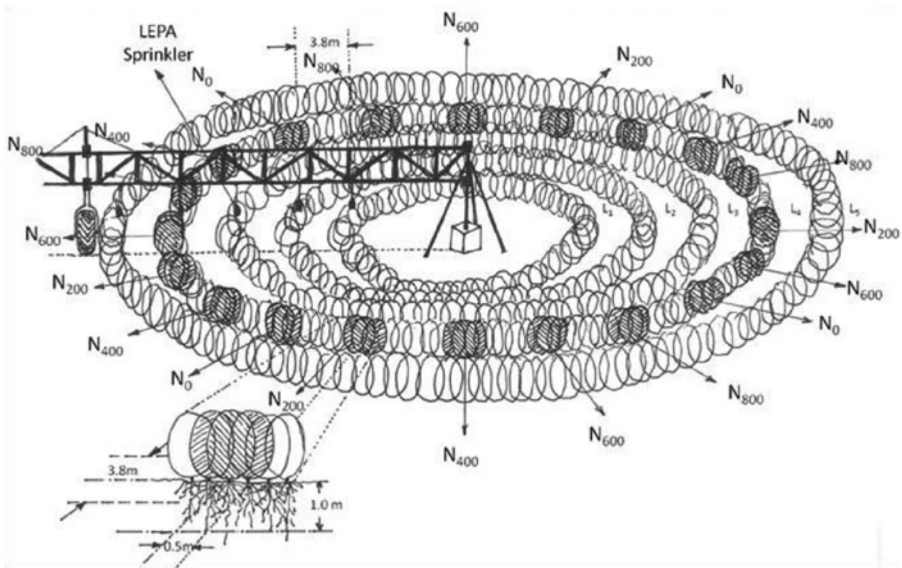

FIGURE 1 Schematic presentation of the field with the experimental circle (Line 4 = L4), showing: i) random distribution of plots (3 labeled trees each, with different nitrogen rates: N_0 = without nitrogen fertilizer; N_{200} = 200; N_{400} = 400; N_{600} = 600; N_{800} = 800 kg ha^{-1} year^{-1} of N); ii) the distance between rows and between coffee plants (3.8 × 0.5 m), and iii) depth of the root system (1.0 m). L_i = lines.

The experimental site was arranged in a commercial coffee plantation (*Coffea arabica* L.), with plants 7 to 8-years-old, well established and at full maturity, with spacing between rows of 3.8 m (circles for central pivot irrigation) and between plants 0.5 m, so that each circle formed a tier, resulting in a plant density of 5,263 plants ha^{-1}, which is considered a high density coffee plantation (Figure 1).

The experiment consisted in five N rates and four replicates, resulting in twenty plots distributed along the 4th circle (from center to the edge, L4) of the central pivot arrangement, in a completely randomized design. Each plot contained three ^{15}N labeled plants having borders of at least four non-labeled plants on each side (Figure 1). The central ^{15}N labeled plant was used for ^{15}N analysis and dry matter sampling. Further detail on the experimental design can be found in Bruno et al. (2011).

Treatments and Management

Treatments of: 0 (control), 200, 400, 600, and 800 kg ha^{-1} year^{-1} of N as ^{15}N-labeled urea, enriched to 1.035 atom excess%^{15}N, with four replicates were applied to this coffee monoculture plantation that had received 600 kg ha^{-1} year^{-1} during the preceding 7 years.

The N rates of each treatment were divided equally into 26 equal applications made every 14 days during the study period, following the management practice adopted by local coffee farmers. Fertigation is traditionally performed in the farm using low energy precision application (LEPA) sprinklers that concentrate the application over the plant, avoiding the inter-row.

During the year of the experiment, the non-labeled urea application of the farm was discontinued on L4 of the experimental pivot circle (Figure 1). The application of the labeled urea on this circle was performed manually, simulating the LEPA sprinklers with a watering can, distributing the solution over the leaves as per the usual overhead emitters. Similar to the LEPA sprinkler application, part of the fertilizer solution reached the soil surface, so that N absorption occurred via both leaf and soil. The fertilizer was diluted in an amount of water corresponding to the usual irrigation of 4 mm. Border plants were fertilized by broadcasting with non-enriched solid urea on the soil surface, thus minimizing the mixing of the labeled and unlabeled fertilizers. All other applications of phosphorus (P), potassium (K) (16 and 420 kg ha^{-1} $year^{-1}$ respectively), micronutrients [boron (B), manganese (Mn), copper (Cu), and nickel (Ni)], organic manure (105 kg N ha^{-1}) and the disease/insect/weed control remained as in all other circles, following the regular farm management. Irrigation was undertaken according to the normal farm management practice, operating continuously over the year at a rate of 3–4 mm day^{-1}, every second day, except during harvest.

Additions of N other than urea, like organic manure, N residues from previous fertilizations or natural inputs were classified in this study as ‘other sources of N’.

Sampling

The following components of the soil-plant system were considered: tree leaf, branch, stem, fruit, root, and litter; soil, drained soil solution, and other losses (non-evaluated compartments, including experimental errors). Assessment of dry matter or N concentration and ^{15}N abundance were made at coffee bean harvest on 24 July 2009, following one plant cycle of labeled fertilizer application.

Plant and Litter Dry Matter

Dry matter sampling was constrained by: i) the commercial farm allowing the sacrifice of just one mature tree per treatment, ii) the distance and travel cost from the farm to the laboratory (1600 km), and iii) the bulk of material that could be collected and transported. Therefore the leaf, branch, stem, and root dry matter were collected in only one of the four replicates of each treatment. The root system mostly explored the 0 to 0.6 m layer, with very few roots reaching 1.0 m depth. Roots for dry matter assessment were extracted using a water jet, from the 0–1.0 m soil profile, under the effective area occupied by one plant. Taking into account the spacing, each coffee plant corresponded to an area of 1.9 m^2 (3.8 m × 0.5 m). However, since plants do not fully occupy this area due to the wide inter-row, it was estimated that the effective area used by a plant was approximately 75% of this (1.425 m^2). All vegetative plant material was chopped and air dried for about 7 days, and thereafter oven dried at 60°C to a constant weight to calculate respective dry matter values in kg ha^{-1}. Fruits were collected manually on all plots (four replicates), first evaluating the fresh weight in volume (liters), and thereafter an aliquot of one liter of fruits per plot was taken for dry matter, ^{15}N and total N as for the vegetative parts.

Samples of litter for dry matter evaluation were collected under the canopy projection of the central plant for each treatment. At the beginning of the experiment all the previously accumulated litter was removed from each plot to be sure that the collected material corresponded to the contributions from the 2008/2009 cropping cycle. Litter samples were processed as for other plant material.

Soil and Leachate

Soil samples were collected for bulk density determinations in pits down to the 1.0 m depth. Undisturbed clods were carefully taken from each 0.2 m layer to evaluate soil bulk density by the paraffin clod method, to calculate the total soil dry weight. The calculation of soil dry matter in each soil layer was made using the effective area occupied by one coffee plant, as discussed above. Due to the wide inter-row, the labeled fertilizer that is applied only over the plant canopy would be dispersed in the soil during its travel to the 1.0 m depth, leaving about 25% of the soil free of fertilizer, resulting in the effective area of 1.425 m^2 $plant^{-1}$.

The amount of water draining below 1.0 m depth, in millimeters, was obtained through the water balance program developed by Rolim et al. (1998), using climatic data from an automatic weather station located near the experimental area. This calculates a water balance component called excess, which includes run-off, deep-drainage and/or capillary rise. More details of this procedure can be found in Bortolotto et al. (2011). Because the landscape of the experimental area has almost zero slope and the local water table is situated at least 15 meters below soil surface, this excess was considered as drainage. Drainage data in mm were transformed into kg ha^{-1} to be able to calculate amounts of leached N also in kg ha^{-1}.

Coffee Bean Yield Evaluation

In order to obtain the so called "green coffee" yield - a traditional productivity measure - the harvested coffee cherries, separated by treatment, were sent to the normal coffee processing operations of the farm, resulting in yield in kg ha^{-1}.

N Concentration and ^{15}N Abundance

Plant and litter. For ^{15}N abundance (^{15}N,%) and nitrogen concentration (N,%) analysis, samples were collected from the central plant of each plot (replicate) in the different compartments.

At harvest time 18 fully expanded leaf samples and 18 ripe fruits were collected per plant for all plots, 6 from each third of the plant (upper, central and lower parts). So as not to sacrifice mature plants, only one branch per replicate was also collected from all plots (central part), considering the first centimeter of the branch as representing the stem, which could not be harvested. Litter subsamples of 100 g of each plot were taken for analysis. Roots for N concentration and ^{15}N abundance assessment were obtained from soil samples extracted from auger wholes positioned 20 cm from the stem of the central plant, in 0.2 layers down to 1.0 m in each plot.

Plant parts and litter were first air dried for one week and after that kept in a ventilated oven at 60°C until constant weight. The dry samples were ground in a 'Wiley' type grinder and homogenized to take representative sub-samples of approximately 10 μg that were introduced in tin capsules. Nitrogen concentration and ^{15}N abundance were measured in an automated continuous flow isotope ratio mass spectrometer (model ANCA-SL, Europa Scientific Ltd., Cambridge, UK).

Soil and leachate. Soil samples were collected with augers, each 0.2 m layer down to 1.0 m per plot. For analysis of ^{15}N and N concentration soil samples from the same layers and treatments were combined to form composite samples, to reduce cost. For this reason, it was impossible to execute statistical analyses to ^{15}N and N concentration in the soil. The composite soil samples were air dried and ground in a ball mill to separate sub-samples of 10 μg for mass spectrometer analysis.

To calculate the N losses from fertilizer by leaching, the drained soil solution was sampled by extraction with porous ceramic cups installed at 1.0 m depth, submitted to vacuum with a manual pump. Extractions started at the beginning of the rainy season (November 2008), at least once each month through the end of experiment. Due to financial constraints, the extractors were installed only in the plots corresponding to 400 and 800 kg ha^{-1} of N. Since these losses occur over the crop cycle, the total amount of N leached by harvest time was integrated over time. Because of the low N concentration in leachate, samples of the same treatment and date (replicates) had to be combined to obtain the sufficient N for mass spectrometer analysis. Solutions of each sampling date were frozen up to analysis date. The quantity of leached N derived from fertilizer for treatments 200 and 600 kg ha^{-1} were obtained by interpolation, assumed to be zero for the treatment without N application. Equation 1 describes the curve of quantity of N derived from fertilizer in the leachate, with a coefficient of determination of 0.999 achieved by Table Curve software (Systat Software Inc, San Jose, CA, USA).

$$y^{0.5} = a + bx^{1.5} \quad (1)$$

y = quantity of N derived from fertilizer in the leachate (kg ha^{-1})
x = N rate applied (kg ha^{-1})
a = 0.35859469; b = 0.00043519

Soil solution samples were first analyzed for nitrate (NO_3^-) and ammonium (NH_4^+) by flow injection analysis (FIA). The N from sub-samples was first concentrated in glass fiber strips as described in Sørensen and Jensen (1991) and then analyzed for ^{15}N abundance and N concentration.

Other losses that were not possible to be measured during the experiment, such as volatilization and denitrification, were estimated as the bulk loss in the balance sheet, by the difference between the amount of N that was applied and that found in all other measured compartments.

Calculations and Statistical Analysis

At harvest time, for each compartment (i) the following calculations were made:

Percentage of N derived from fertilizer ($Ndff_i$,%):

$$Ndff_i = [^{15}N_{sample} - ^{15}N_{nat} / ^{15}N_{fert} - ^{15}N_{nat}].100 \quad (2)$$

where $^{15}N_{sample}$ is the ^{15}N abundance in each compartment sample, $^{15}N_{nat}$ is the natural abundance of ^{15}N, taken from the treatment without N (control) and $^{15}N_{fert}$ is the fertilizer abundance, equal to 1.035 atom excess% ^{15}N.

Quantity of N derived from fertilizer ($QNdff_i$, kg ha^{-1}):

$$QNdff_i = [DM.\%N/100].Ndff_i \quad (3)$$

where DM is the dry matter mass (kg ha^{-1}) and%N is nitrogen concentration (%). For the leaching compartment, DM in equation 3 was substituted by the amount of drained water below the root zone, transforming mm into kg ha^{-1}.

Quantity of N derived from the soil and other sources ($QNdfs_i$, kg ha^{-1}):

$$QNdfs_i = [DM.\%N/100] - QNdff_i \quad (4)$$

It has to be recognized that in the case of adult perennial crops QNdfs represents the non-labeled N in the plant accumulated during the eight years of cultivation, including the previous fertilizer applications made before start of the experiment.

Fertilizer N recovery (R_i,%):

$$R_i = [QNdff/NR].100 \quad (5)$$

where NR is the N rate (kg ha^{-1}).

The fertilizer N recovery was calculated for each compartment and for the whole plant, by adding the N recovered in each plant part, including fruit.

The agronomic efficiency index (AEI,%) was taking as green coffee yield per N fertilizer rate, calculated as follows:

$$AEI = [CY/NR].100 \quad (6)$$

where CY is the green coffee yield, kg ha^{-1}.

Data collected in replicates (plant and litter data of ^{15}N abundance, N concentration and green coffee yield) were submitted to one-way analysis of variance (ANOVA), and significant differences were identified using the Tukey test at the 5% probability level, using the Statistica program (StatSoft Inc., Tulsa, OK, USA). The quantity of N from fertilizer and fertilizer recovery data could not be submitted to statistical analysis because they include data which was not replicated for the reasons discussed before.

RESULTS

Plant and Litter

For the 8-year-old coffee plants in this experiment, no visual difference in stature was observed among the plants of the different treatments; therefore the dry matter obtained for only one plant per treatment was used to calculate amounts of N in plant compartments, and averaged across treatments. It is important to note that these dry matter results are only used for calculations in equations 3 to 5, and that the expected differences in fertilizer N uptake would essentially come from the percentage of N derived from fertilizer, calculated with four replicates. The average whole plant dry matter (shoots + roots) was a little over 42,000 kg ha^{-1} with 80% in the above ground mass. As typically observed for woody perennials, the stem contributed most to this mass, with 36% of the total, followed by root (20%), fruit (19%), branch (15%), and leaf (10%). There were no significant differences in litter dry matter between N rate treatments, which were collected in all replicates, with an overall average of 8,126 kg ha^{-1}. This supports the dry matter of the standing biomass being fairly consistent.

TABLE 2 Average and standard deviations (four replicates) of nitrogen concentration (N,%), 15 nitrogen abundance (^{15}N,%) and nitrogen derived from fertilizer (Ndff,%) for different compartments of coffee plants receiving different fertilizer rates

	Compartment/ rate (kg ha^{-1})	0	200	400	600	800
N concentration (%)	Leaf	$2.8^{c} \pm 0.2$	$2.8^{bc} \pm 0.2$	$3.0^{bc} \pm 0.1$	$3.3^{b} \pm 0.2$	$3.6^{a} \pm 0.3$
	Branch	$1.4^{ns} \pm 0.3$	$1.5^{ns} \pm 0.1$	$1.7^{ns} \pm 0.1$	$1.5^{ns} \pm 0.1$	$1.7^{ns} \pm 0.1$
	Stem	$1.1^{ns} \pm 0.1$	$1.2^{ns} \pm 0.1$	$1.0^{ns} \pm 0.3$	$1.1^{ns} \pm 0.4$	$1.3^{ns} \pm 0.2$
	Fruit	$1.7^{ns} \pm 0.4$	$2.1^{ns} \pm 0.5$	$2.1^{ns} \pm 0.3$	$2.2^{ns} \pm 0.4$	$2.2^{ns} \pm 0.3$
	Root	$1.7^{b} \pm 0.1$	$2.1^{ab} \pm 0.2$	$2.0^{ab} \pm 0.1$	$2.2^{a} \pm 0.3$	$2.0^{ab} \pm 0.1$
	Litter	$2.9^{b} \pm 0.5$	$3.2^{ab} \pm 0.2$	$3.0^{b} \pm 0.3$	$3.3^{ab} \pm 0.2$	$3.6^{a} \pm 0.2$
^{15}N abundance (%)	Leaf	$0.371^{c} \pm 0.002$	$0.633^{b} \pm 0.062$	$0.662^{b} \pm 0.043$	$0.784^{a} \pm 0.035$	$0.890^{a} \pm 0.092$
	Branch	$0.369^{c} \pm 0.001$	$0.468^{bc} \pm 0.004$	$0.498^{abc} \pm 0.041$	$0.558^{ab} \pm 0.062$	$0.617^{a} \pm 0.129$
	Stem	$0.368^{b} \pm 0.001$	$0.453^{ab} \pm 0.015$	$0.508^{a} \pm 0.012$	$0.570^{a} \pm 0.062$	$0.567^{a} \pm 0.123$
	Fruit	$0.372^{d} \pm 0.003$	$0.672^{c} \pm 0.068$	$0.730^{bc} \pm 0.100$	$0.864^{ab} \pm 0.043$	$0.877^{a} \pm 0.061$
	Root	$0.369^{b} \pm 0.001$	$0.582^{a} \pm 0.146$	$0.512^{ab} \pm 0.054$	$0.520^{ab} \pm 0.024$	$0.515^{ab} \pm 0.052$
	Litter	$0.372^{d} \pm 0.002$	$0.546^{c} \pm 0.038$	$0.609^{bc} \pm 0.041$	$0.671^{ab} \pm 0.041$	$0.720^{a} \pm 0.089$
Ndff (%)	Leaf	0^{c}	$25.4^{b} \pm 6.1$	$28.3^{b} \pm 4.2$	$40.1^{a} \pm 3.4$	$50.5^{a} \pm 9.0$
	Branch	0^{c}	$9.7^{bc} \pm 0.4$	$12.5^{abc} \pm 4.0$	$18.4^{ab} \pm 6.0$	$24.1^{a} \pm 12.5$
	Stem	0^{b}	$8.3^{ab} \pm 1.4$	$13.6^{a} \pm 1.2$	$19.6^{a} \pm 6.0$	$19.3^{a} \pm 11.9$
	Fruit	0^{d}	$29.1^{c} \pm 6.6$	$34.8^{bc} \pm 9.7$	$47.9^{ab} \pm 4.2$	$49.1^{a} \pm 6.0$
	Root	0^{b}	$20.7^{a} \pm 14.2$	$13.9^{ab} \pm 5.3$	$14.7^{ab} \pm 2.3$	$14.1^{ab} \pm 5.0$
	Litter	0^{d}	$16.9^{c} \pm 3.7$	$23.0^{bc} \pm 4.0$	$29.1^{ab} \pm 4.0$	$33.8^{a} \pm 8.7$

*Average of four replicates. Results in the line followed by the same letter are not significantly different ($P < 0.05$). ns = Not significant at 0.05 level in the same line.

Table 2 shows N concentration, ^{15}N abundance and corresponding values of percentage of N derived from fertilizer in different plant compartments. The isotopic data show that the fertilizer label of 1.035 atom excess%^{15}N was sufficient to differentiate treatments in terms of percentage of N derived from fertilizer. The N concentration in the different compartments shows a slight tendency of increase with fertilizer rate.

Generally, the quantity of N from fertilizer was much less than half of the total N accumulated in the different plant compartments (Figure 2) and the whole plant (Figure 3). This is expected for older perennial crops because they have a large legacy of N accumulated during the previous years.

Soil and Leachate

The mean soil bulk density was 1,582 kg m^{-3} across the 0–1.0m layer, resulting in an average soil dry matter of 2,254 kg per plant. As a result of composite samples of four replicates, in the 1m soil layer the N concentration and N total was around 0.02% and 2,700 kg of N ha^{-1} for all treatments. The ^{15}N abundance ranged from 0.373 to 0.449% and percentage of N derived from fertilizer ranged from 0.2 to 7.6% respectively from 200 to 800 kg ha^{-1} N rates.

Mean (composite samples) ^{15}N enrichment of soil solution, varied from 0.689 (November) to 0.981% (June) for 400 kg ha^{-1} N and 0.901 (November) to 1.058% (May) for 800 kg ha^{-1}, so that the leachate

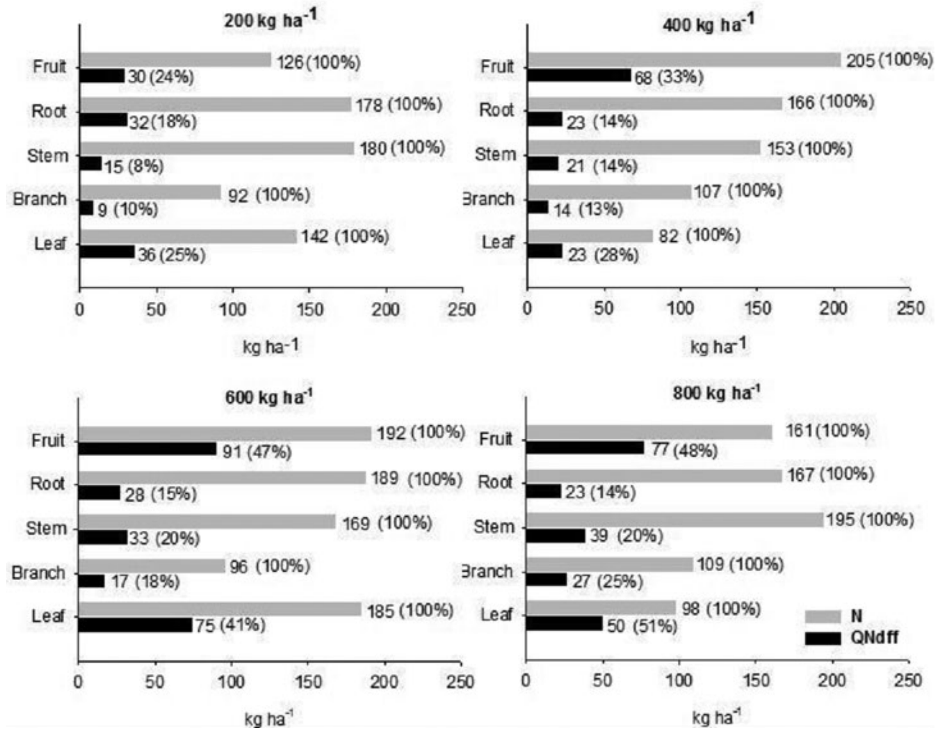

FIGURE 2 Total nitrogen accumulated quantity (N) and quantity of nitrogen derived from fertilizer (QNddf) in different compartments of the coffee plant: fruit, root system, stem, branch, and leaf.

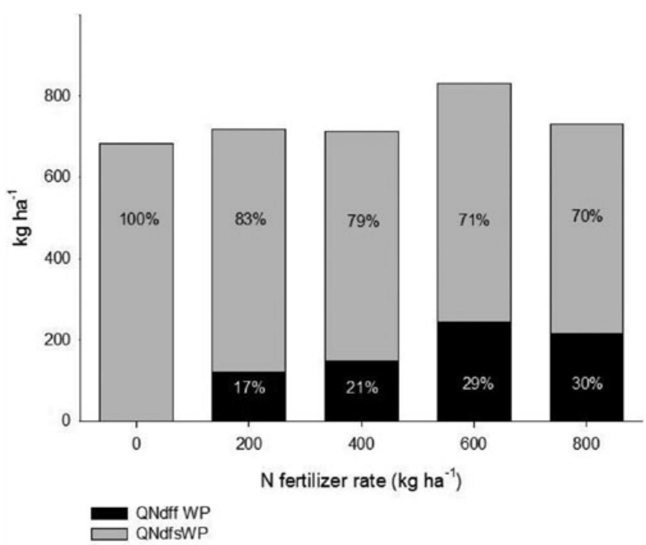

FIGURE 3 Comparison between the quantity of nitrogen derived from the fertilizer in the whole plant (QNdffWP, kg ha^{-1}) and quantity of nitrogen derived from the soil and other sources in the whole plant (QNdfsWP, kg ha^{-1}), at harvest for coffee plants receiving increasing N fertilizer rates.

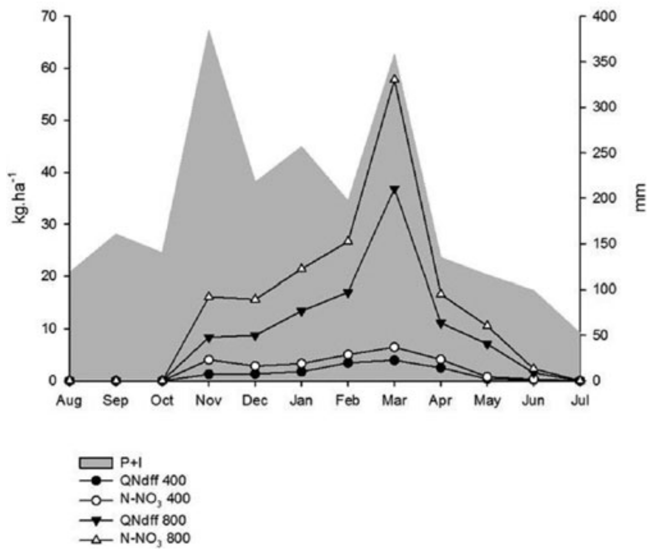

FIGURE 4 Rainfall plus irrigation (P+I, mm) during the evaluated cropping season (2008/2009) and total quantities of N-NO_3^- leached below the 1.0 m depth, for treatments 400 and 800 kg ha^{-1} and the respective quantities of the leached nitrate derived from the fertilizer (QNdff 400 and QNdff 800).

percentage of N derived from fertilizer could be reliably estimated. The integrated value of drainage over the crop cycle was 1,010 mm, corresponding to 1,440 kg of drained water per plant.

Because NH_4^+ in the soil solution extracts was below detection level after FIA analysis, all N in the samples of soil solution extracts was considered as NO_3^-, and the results thus refer to nitrate N (N-NO_3^-, kg ha^{-1}). Both the total quantity of leached N-NO_3^- and the respective quantity of N derived from fertilizer followed the water regime, rainfall plus irrigation (P+I, mm), in a more pronounced way for the 800 kg ha^{-1} treatment in comparison to the 400 kg ha^{-1} treatment (Figure 4). The leaching peak occurred in March 2009 for both treatments, when (P+I) was 358 mm. Despite the presence of the first (P+I) peak in November 2008, with a total of 384 mm, leached N-NO_3^- and quantity of N derived from fertilizer were very low because at that moment only one third of the fertilizer had been applied. The percentage of N derived from fertilizer in the total amount of N-NO_3^- leached was 55% for the treatment 400 kg ha^{-1} N and 62% for the 800 kg ha^{-1} N treatment.

Green Coffee Bean Yield and Agronomic Efficiency

Green coffee bean yield did not differ among treatments, but the nitrogen use efficiency in terms of green coffee yield per applied fertilizer N rate

TABLE 3 Green coffee bean yield (kg ha^{-1}) and the agronomic efficiency index (kg kg^{-1}) for a complete productive cycle of coffee plants (2008/2009) receiving different nitrogen rates

Nitrogen rate	Green coffee bean yield (kg ha^{-1})	Agronomic efficiency index* (kg kg^{-1})
0	$3347^{ns} \pm 1630$	0^{c}
200	$2673^{ns} \pm 292$	$13^{a} \pm 1$
400	$4347^{ns} \pm 1207$	$11^{a} \pm 3$
600	$3954^{ns} \pm 1140$	$7^{b} \pm 2$
800	$3390^{ns} \pm 1247$	$4^{b} \pm 2$

Results in the columns followed by the same letter are not significantly different ($P < 0.05$). ns = Not significant at 0.05 level in the same column. * Agronomic efficiency index corresponds to green coffee yield (kg) per applied fertilizer N (kg).

was different (agronomic efficiency index, Table 3). The lowest N rates were most efficient in nutrient use to produce coffee beans.

N Balance

For discussion purposes system compartments are aggregated to whole plant (leaf, branch, stem, fruit and root), reserve (litter and soil) and losses (leached solution and other losses).

The N balance made after one complete cycle of coffee bean production is the distribution of the N derived from the fertilizer, represented by the QNdff (kg ha^{-1}) and R (%) in the different compartments as a result of the continuous applications of ^{15}N-urea, and includes the effects of N translocated between compartments (Table 4).

TABLE 4 Balance of nitrogen in soil-plant system, with average quantity of nitrogen derived from fertilizer (QNdff) and percentage of nitrogen recovered from the total rate of fertilizer applied (R)

	200		400		600		800	
Pool/rate (kg ha^{-1})	QNdff (kg ha^{-1})	R (%)	QNdff (kg ha^{-1})	R (%)	QNdff (kg ha^{-1})	R (%)	QNdff (kg ha^{-1})	R (%)
Leaf	35.9	17.9	23.3	6.0	74.7	12.4	49.7	6.2
Branch	8.9	4.5	13.5	3.0	17.4	2.9	26.7	3.3
Stem	14.7	7.4	20.8	5.0	32.8	5.5	38.9	4.9
Root	31.7	15.9	23.2	6.0	27.8	4.6	23.5	2.9
Fruit	29.7	14.9	67.9	17.0	91.2	15.2	77.4	9.7
Whole plant sub-total	121.0	60.5	148.7	37.0	243.9	40.6	216.3	27.0
Litter	42.6	21.3	48.4	12.0	82.1	13.7	94.7	11.8
Soil	8.5	4.2	76.9	19.2	182.6	30.4	312.3	39.0
Reserve sub-total	51.0	25.5	125.3	31.2	264.7	44.1	407.1	50.9
Leaching	2.5	1.3	14.7	3.7	45.6	7.6	104.2	13.0
Other losses	25.5	12.7	111.2	28.1	45.8	7.6	72.5	9.1
Losses sub-total	28.0	14.0	126.0	31.8	91.4	15.2	176.7	22.1
Total recovery	174.5	87.3	288.8	71.9	554.2	92.4	727.5	90.9

Coffee plants that received 600 kg ha^{-1} of N accumulated most fertilizer N in leaf and fruit, whereas those that have accumulated more N (kg ha^{-1}) in stem and branch received 800 kg ha^{-1} of N.

The total fertilizer N recovery of the different compartments was greater for leaf, root and litter in plots that received 200 kg ha^{-1} of N and practically did not vary for the other rates in the other compartments.

The quantity of N derived from fertilizer in soil varied from 8.5 to 312.3 kg ha^{-1} of N, increasing according to N application rate, and the major soil recovery was in the 800 kg N ha^{-1} treatment (Table 4).

The amount of N-$NO_3{}^-$ in soil solution was 27 kg ha^{-1} under 400 kg ha^{-1} of N and 167 kg ha^{-1} under 800 kg ha^{-1} of N. The quantity of N derived from fertilizer values for the leached soil solution were measured for 400 and 800 kg ha^{-1} of N, and estimated by interpolation for 200 and 600 kg ha^{-1}. Thus treatments 800 and 200 kg ha^{-1} of N had the highest and lowest leaching, respectively (Table 4).

The quantity of N derived from fertilizer in the whole plant increased from 121 kg ha^{-1} for 200 to 244 kg ha^{-1} for 600, thereafter decreasing to 216 for 800 kg ha^{-1} of N (Table 4 and Figure 3).

The percentage of fertilizer N recovered in the plants showed a tendency of decreasing with increasing fertilizer with losses showing an inverse tendency (Table 4).

DISCUSSION

Plant and Litter

The largest N concentration occurred in the compartments of leaf and litter, with treatment 800 kg $ha^{-1}$of N significantly higher, increasing in accordance with the N fertilizer rates (Table 2). This could indicate that there is an accumulation of unused fertilizer N, since the coffee bean production did not differ among the different treatments (Table 3). Leaf and consequently litter - which consists mainly of dead leaves - are the major sinks of N during the first crop cycle after labeled fertilizer application. Root presents significant differences in N concentration only between the control and other treatments, which can denote that N fertilization contributes with the constitution of new roots, confirmed by root ^{15}N abundance and percentage of N derived from fertilizer (Table 2).

Plant ^{15}N abundance data show significant differences between the control and all fertilizer rate treatments, excepting for branches up to the 400 kg ha^{-1} rate, and stem up to the 200 kg ha^{-1} rate (Table 2). These results indicate that the choice of the fertilizer enrichment of 1.035 atom excess% ^{15}N was sufficient to differentiate the fertilizer N balance (Table 4) among the majority of the plant compartments over the one year study.

The N concentrations in leaf of the 200 and 400 kg ha^{-1} plants are considered adequate according to Malavolta (2006), while that of 600 kg ha^{-1} is high and 800 kg ha^{-1}excessive (Table 2). Larger leaf growth depends on N availability and promotes an increase in the photosynthetic leaf area that is reflected in higher productivity. However, since plants receiving 600 N kg ha^{-1} did not stand out among the other rates in terms of green coffee yield (Table 3), the possible greater leaf growth resulting from the high N rate favored vegetative but not fruit growth. Although not observed for data in the plantation of this study, excess N can reduce productivity, foster pathogens and weeds, reduce fitoalexin synthesis, and retard fruit ripening (Gallo et al., 1999; Ricci and Neves, 2004).

In adult coffee plants, the yearly increase in dry matter corresponds mainly to fruit, new leaves, branch growth, and replacement of dead roots. The present data indicate that considerable amounts of N were transferred among compartments, in addition to N from soil, since the contribution of fertilizer N was less than half the total N of the coffee plant (Figure 2 and 3). The fertilizer N represented by quantity of N derived from fertilizer is a very low part of the total N, with exception to a few compartments in the 600 and 800 kg ha^{-1} rates, which reached values around 50% (Figure 2). Fertilizer N accounted for less than 25% of the total N for stem, branch and root in all treatments, suggesting that most of their growth was sponsored by N present in the plant before the start of the experiment and the uptake of soil and residual fertilizer N from previous years. The very high quantities of N in the root neither presented differences among treatments, nor among for stem and branch. The low contribution of fertilizer N in the total composition of the root suggests a reduced production of new roots, resulting in low root dry matter or in uptake of residual N from previous years. The quantity of N derived from fertilizer in roots (Figure 2) is much lower than that presented by Fenilli et al. (2008) for 5-year-old coffee trees, which varied from 40 to 60 kg ha^{-1} of N derived from fertilizer (N rates of 280 and 350 kg ha^{-1} as ammonium sulfate), mainly due to new root growth. Fruit quantity of N derived from fertilizer was lower for 200 kg ha^{-1}, and less than one quarter of the N of this compartment had its origin in the applied urea. For the other treatments, the percentage of N derived from fertilizer in fruit increased (Figure 2), but was not reflected in increased fruit yield of the higher fertilizer rate treatments (Table 3).

Soil and Leachate

Crop uptake of N from soil was higher than from applied fertilizer (Figure 3), which is expected for annual crops, and is a result of fertilizer loss and immobilization by soil microorganisms (Cassman et al., 2002). When a mineral fertilizer is applied to a soil of low or moderate organic matter content (Table 1), immobilization by microorganisms can be significant. Adding a ^{15}N source to the non-labeled inorganic soil N, the ^{15}N is exposed to various processes besides plant absorption, depending on the environmental conditions and soil properties. The applied ^{15}N can take the place of the native ^{14}N

that would be immobilized, leaving less ^{15}N and more ^{14}N available to the plant, an effect called "pool substitution" (Powlson and Barraclough, 1993; Jenkinson et al., 1985). In coffee monocultures, much of the N immobilized by microorganisms will gradually become available to plants.

Fertilizer N can be lost after its application, with leaching being one of the major processes, following excessive rainfall or irrigation. This can potentially cause damage to the environment and also leads to economic loss. In the present study, leached soil solution drained below the 1 m depth had very little chance to be absorbed by plant roots.

Coffee Bean Yield and Agronomic Efficiency

Green coffee bean yield presented no difference among the applied N rates. In order to analyze the green coffee bean production (Table 3) one has to consider the agronomic history of the crop and coffee plant physiology. Coffee plants take two years to complete their phenological cycle, in which first appear vegetative axillary buds during the long day months. Reproductive buds are induced during the short day months, and that later become ripe, dormant, and enter anthesis. In this way, vegetative buds of the last year define – in conjunction with management and environmental factors – the next year's yield, reflecting the nutritional status of the plant when the vegetative buds were formed. These particularities influence the equality of green coffee production, but when the agronomic efficiency index (Equation 6) is observed, the nitrogen use efficiency is clearly higher when lower rates are applied.

It was expected that the agronomic efficiency index would decrease as the fertilizer rates would increase, as observed in the literature (Mattos-Júnior et al., 2003). In the present study, the highest agronomic efficiency index was observed for the lowest fertilizer rate, corresponding to one third of the rate used by farmers of the region, and showing that N fertilization could be drastically reduced without sacrificing yield, and consequently reducing the environmental pollution potential of high N applications.

N Balance

Plants receiving 600 kg ha^{-1} and 800 kg ha^{-1} had the most quantity of N derived from fertilizer in all compartments of the soil-plant system, including compartments of reserve and losses (Table 4). Even though this was not reflected in a significant increase of green coffee production (Table 3), it indicates that there has been a surfeit of N, not only in the reserve organs but also in fruit (Table 2). However, the total fertilizer N recovery was higher for plants receiving 200 kg N ha^{-1} for all plant compartments, and consequently in litter, except for fruits which had the major recovery with 400 kg ha^{-1} of N (Table 4). This low N rate may thus be providing sufficient N to balance the N export by harvest - varying from 126 to 205 kg ha^{-1} of N (Figure 2) - and leaving much less N available for leaching (Table 4). In the present study, the 200 kg ha^{-1} N treatment showed N recoveries for the whole plant consisted with adult citrus, another woody perennial, while other treatments presented lower N recovery (Table 4) (Feigenbaum et al., 1987; Quiñones et al., 2007, 2005). The 60% N recovery in the coffee plants under 200 kg ha^{-1} of N treatment (Table 4) is far above that of 33% for annual plants in a review of world wide data by Dourado-Neto et al. (2010), there presented as N-use efficiency.

Furthermore the quantity of N derived from fertilizer of whole plants averaged 183 kg N ha^{-1} across treatments, close to the yearly harvest offtake of this crop, indicating that the 200 kg ha^{-1} rate may be adequate for this coffee plantation. The percent fertilizer N recovery by the coffee plants showed a tendency to be lower with increasing rates, while the lost N had the inverse behavior, highlighting lower N losses with lower N application rates.

A fertilizer N balance study (Fenilli et al., 2008) with 3 to 5-year-old *Coffea arabica* plants carried out in Piracicaba, SP, showed that after 2 years, of the N applied as ammonium sulfate (280 kg ha^{-1} and 350 kg ha^{-1} of N in the first and second years, respectively) 19% was in the above ground vegetative part; 24% in litter; 26% exported in fruits; 1% lost by leaching; 2% lost by volatilization; 13% remained in the soil; and 6% was unaccounted for. The N recovery by fruit of these 5-year-old coffee trees was greater than that of this study, probably because of the legacy of long-term fertilizer use in the present study.

The branch and stem quantity of N derived from fertilizer increased for increasing N rates (Table 4), indicating N storage in these compartments. The same did not occur in the root system, a well-known storage compartment. The root system presented quantity of N derived from fertilizer very similar among treatments, indicating little re-growth. Root development at depth was very low, presumably due to water and nutrient excess provided on the surface though the fertigation. The high acidity and aluminum, and low amounts of calcium and magnesium below the 0-0.2m layer (Table 1) might also constrain root system formation during the studied crop cycle.

Litter is one of the main reserves of N in perennial agro-ecosystems, and its composition can be influenced by fertilizer management. Its quantity of N derived from fertilizer increased with fertilizer rate, while the N recovery in litter was highest for plants that received 200 kg N ha^{-1} (Table 4). In a coffee crop study, Reichardt et al. (2009) found 80 and 70 kg ha^{-1} of N from fertilizer in litter for the first and second years of cultivation. These results are comparable to the values for 600 and 800 kg ha^{-1}, despite the fact that their fertilizer application rates were substantially lower. In a study on adult citrus trees litter recovered 1.6 to 2.9% of the applied N at the rate of 125 g plant^{-1} year^{-1} of N (Quiñones et al., 2007, 2005), values much below those of coffee plants. Orange trees certainly translocate N from old to young leaves with a greater efficiency than coffee trees, making orange tree litter less N enriched.

The quantity of nitrogen derived from fertilizer in soil varied widely among N rates, a variation far greater than that reported by Reichardt et al. (2009). While in the present study the soil N derived from fertilizer was almost 9 to 312 kg ha^{-1} for plots that received 200 and 800 kg ha^{-1} of N, respectively, for the Reichardt et al. (2009) study the values varied from 50 to 79 kg ha^{-1} of N from fertilizer for the rates of 280 and 350 kg ha^{-1} of N. The percentage of fertilizer N in the soil after coffee harvest varied from four to almost 40% of the applied, increasing with N rate (Table 4). Comparable results were found for orange trees, with 13 to 23% of the fertilizer N remaining in the soil (Quiñones et al., 2005). Soil N can be stored as soil reserves, absorbed by the plant or lost by leaching/volatilization, with a potentially negative effect to the environment. Most soil N is in the organic form, however the soil quantity of N derived from fertilizer can be both inorganic and organic. The soil N half-life affects the reserve/loss ratio, and it can stay thousands of years in the soil protected by colloidal material (Moreira and Siqueira, 2006). The N in the soil of our study is probably also in the organic form once urea is readily transformed into nitrate which is less retained by the soil. Due to budget restrictions it was not possible to verify the proportion of inorganic/organic N in the soil, and also not its distribution along the soil profile, which would be very interesting in future studies of fertigated coffee plantations.

All mineral N in the leached solution was in the form of nitrate, mainly due to the transformations of the urea in the soil, both from the original soil organic matter or from the fertilizer. Nitrate is very mobile in the soil and when not absorbed by root, can leach down in the soil (Havlin et al., 2005; Addiscott, 2005). As expected, the total amount of N-NO_3^- and the respective quantity of N derived from fertilizer in soil solution at the 1 m depth were much greater for the soil profiles of 800 kg ha^{-1} in relation to 400 kg ha^{-1}, independent of the time of year (Figure 4). The quantity of N derived from fertilizer for 400 kg ha^{-1} was always much lower, even under potential leaching conditions. That estimated for 600 kg ha^{-1} was intermediate, so that in terms of leaching, fertilizer application rates of up to 400 kg ha^{-1} of N might be considered safe (Table 4). Since the percentage of N from fertilizer in the total amount of N- NO_3^- leached was 55 and 62% for treatments 400 and 800 kg ha^{-1} of N it can be said that, as opposed to plants, for these compartments the fertilizer N represents the largest portion. Fertigation is performed continuously over the whole year in the farms of this Brazilian region, even during rainy periods. Among factors that enhance leaching, the rate and the time of fertilizer application are the most important (Havlin et al., 2005), and in all these factors contributed to the intensity of leaching in the present study. The continuous fertilizer application without attention to periods of greater N need by the plant, and disregarding the climatic conditions are the main reasons for N leaching. In contrast to the results obtained here in central Brazil, the majority of reports state that N leaching is not a significant process under southeastern Brazilian soil and climate conditions (Urquiaga and Zapata, 2000; Cantarella, 2007), with exception to the study of Camargo (1989), which reports a leaching loss of almost 30 kg ha^{-1} of fertilizer N in a sugarcane crop that received 100 kg ha^{-1} of ^{15}N-urea grown on a sandy soil. In our study the large leaching losses occurred due to high rainfall and sandy soils combined with excessive irrigation and N fertilization.

Nitrogen export in harvested fruit is here not considered as a loss because it contributes directly to economic yield. However, the data shows that more N may be exported by fruits than is necessary for maximal bean yield. The treatment that showed fewer total losses in percentage was 200 kg N ha^{-1} (Table 4), and this rate is recommended to Brazilian cerrado coffee plantations.

Other possible N losses during the experimental period were not measured, but could include volatilization of fertilizer ammonia, denitrification and ammonia emissions via leaf, well documented by Clough et al. (2001), Hörtensteiner and Feller (2002) and Christopher and Lal (2007). Gaseous emissions by coffee crops exist, and are probably contributed to the percentage of ^{15}N unaccounted for in the present experiment. These losses have to be better investigated in future research, not only because they represent an economic loss to the farmer but also due to their global warming potential.

CONCLUSION

Adult coffee plants that received a high N fertilizer rate (600 kg N ha^{-1} $year^{-1}$) during the past eight years did not respond to increasing N fertilizer rates, as shown by green coffee bean yield. The most efficient N rate in terms of N recovery in the whole plant was 200 kg N ha^{-1} $year^{-1}$, which also presented the lowest N losses to environment. The least sustainable rate was the 800 kg N ha^{-1} $year^{-1}$, which showed the greatest losses and smallest whole plant recovery. Nitrate and N from fertilizer losses to the environment via leaching mirrored the rainfall plus irrigation and fertilizer application rates.

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