



Fertilizer nitrogen in fertigated coffee crop: Absorption changes in plant compartments over time

Isabeli P. Bruno^a, Murray J. Unkovich^b, Rafael P. Bortolotto^a, Osny O.S. Bacchi^{c,1},
Durval Dourado-Neto^a, Klaus Reichardt^{c,*}

^a Department of Crop Production, University of São Paulo, Escola Superior de Agricultura “Luiz de Queiroz”, Av. Pádua Dias, n° 11, C.P. 09, CEP 13418-900 Piracicaba, SP, Brazil

^b School of Agriculture, Food and Wine, The University of Adelaide, Waite Campus, PMB 1, 5074 Glen Osmond, SA, Australia

^c Laboratory of Soil Physics, University of São Paulo, Center for Nuclear Energy in Agriculture, Av. Centenário, n° 303, C.P. 96, CEP 13416-000 Piracicaba, SP, Brazil

ARTICLE INFO

Article history:

Received 6 April 2011

Received in revised form 13 July 2011

Accepted 14 July 2011

Keywords:

¹⁵N

Nitrogen management

Cerrado

Woody perennial crop

ABSTRACT

Nitrogen (N) dynamics in plants during their development in agricultural crops has to be well understood in order to design management practices that lead to maximum productivity with minimum N loss from the system. In a labeled fertilizer field study ¹⁵N accumulation in different plant parts of mature coffee was observed over time. The objective was to ascertain the time of greatest crop N demand as a scientific basis for designing fertigation schedules. Coffee plantations of central Brazil are routinely fertigated only with extremely high applications of N. Good coffee bean production should be sustainable by applying lower N quantities at those frequencies designated by additional scientific criteria. The experiment was carried out over a complete coffee cropping cycle (2008/2009) in a field of low soil fertility in the Brazilian savanna “cerrado”. Rates of 0, 200, 400, 600 and 800 kg N ha⁻¹ year⁻¹ as ¹⁵N-labeled urea were applied via fertigation, divided equally over the year into 26 portions, distributed every 14 days. Changes of N absorption in various plant compartments indicated that fertilizer use could be improved if a lower rate is applied only up to the beginning of fruit maturation, focusing on the stage before fruit filling. This specific stage was found to be the period of greatest N consumption by leaf and fruit. ¹⁵N absorption data showed that it is possible to decrease the routine fertilization rate of 600 to a much lower value, of order of 200 kg N ha⁻¹ without decreasing the production of coffee beans.

© 2011 Elsevier B.V. All rights reserved.

1. Introduction

Nitrogen (N) absorption and assimilation is fundamental for plant growth and development. Because N demand is highest among soil nutrients, it potentially limits crop yields. In agriculture, achieving maximum productivity with reduced inputs of N fertilizer is a major challenge. The ideal N fertilizer distribution, strongly linked to the specific stage of plant N demand, is one of the knowledge gaps for several crops (Lea and Morot-Gaudry, 2001; Lobell, 2007). In recent years with the emphasis shifting from increasing yield to the development of more sustainable agricultural practices, farmers strive to make more efficient use of fertilizers in order to reduce production costs and environmental impacts. Lawlor et al. (2001) states that the optimum use of nitrogen should aim to

achieve maximum production per unit of applied N, which certainly depends on the knowledge of plant requirements during various growth stages. N supply affects grain production in cereals and when applied in large N amounts, yield decreases due to the stimulus to the growth of vegetative parts (Lawlor, 2002). Hence at critical periods, it is advantageous to manipulate the supply of N to the plant in relation to carbon assimilation. However, for woody perennial crops changes in nutrient uptake dynamics as related to crop growth patterns are not well documented. For these crops the challenge of finding the appropriate rate and major N demand stages is even greater because the need of N varies significantly with phenological stages occurring differently during each crop cycle year after year.

During the last two decades coffee cultivation in Brazil was extended from traditional coffee growing areas to the savanna or “cerrado” plains as those of the Bahia state. Coffee growers in this region have used N rates (of the order of 600–800 kg N ha⁻¹ year⁻¹) far above those applied in other coffee producing areas (150–450 kg N ha⁻¹ year⁻¹) (FAO, 2010). With no scientific basis for these excessively high N application rates, losses to nitrophication are likely to be occurring (Milroy et al., 2008; Sitthaphanhit et al., 2009). Agricultural systems receiving

* Corresponding author. Tel.: +55 19 3429 4715.

E-mail addresses: isabelibruno@gmail.com (I.P. Bruno), murray.unkovich@adelaide.edu.au (M.J. Unkovich), rpbortolotto@yahoo.com.br (R.P. Bortolotto), osny@cena.usp.br (O.O.S. Bacchi), dourado@esalq.usp.br (D. Dourado-Neto), klaus@cena.usp.br (K. Reichardt).

¹ Tel.: +55 19 3429 4715.

large quantities of N that are out of phase with the greatest nutrient demand are inefficient because the nutrient storage capacity of the plant is limited. With rates of protein synthesis and carbon assimilation – the largest N consumers – being relatively slow, N within soil and plants remains vulnerable to losses (Lawlor et al., 2001).

With a relatively good supply of water, great proportions of the predominately flat regions of cerrado are perfect for irrigation—especially central pivot sprinkler systems. Fertigation, an increasingly employed agricultural practice, allows sequential, simultaneous applications of water and nutrients at any rate and frequency (number of splittings) that could reduce the waste of fertilizer. However, the use of rates much higher than those applied in traditional growing areas and the large water volumes applied by irrigation may lead to excessive losses, which are problematic from both economic and environmental points of view. Although farmers perform fertigation every 14 days, each one with small quantities of fertilizer, there is no attention paid to the temporal variation in N demand by the coffee plant. The fertilizer allocation is evenly split across the number of applications and not based on actual crop demand.

Nitrogen-15 labeled fertilizer is invaluable for studying the efficiency of N use in agroecosystems because it allows improved fertilizer application practices based on a more comprehensive understanding of the N dynamics of plant uptake as a function of time (Hardarson, 1990; Reichardt and Bacchi, 2004). Owing to the large size of woody tree crops like pear, peach, citrus and coffee, the investigation of N absorption and translocation is often hindered by experimental difficulties related to plant sampling methodologies. Because of these difficulties, a limited number of reports related to N uptake in the field appear in the literature. In addition to the complexity of non-destructive measurement of dry matter accumulation, working with trees having large volumes of dry matter requires the use of large amounts of label that make experiments very costly. To avoid such problems, Boaretto et al. (1999) and Natale and Marchal (2002) in Brazil and Lea-Cox et al. (2001) in USA, studied citrus plants at early growth stages under controlled conditions, and Bustamante et al. (1997) made similar studies on coffee seedlings in Cuba. Quiñones et al. (2003, 2005, 2007) included irrigation in their studies using lysimeters of 3.4 m³ with adult orange trees in Spain. Recently, Fenilli et al. (2007, 2008) and Reichardt et al. (2009) presented a large scale field study on the use of ¹⁵N labeled fertilizer applied to a coffee crop grown in a traditional coffee growing area in Brazil. Their complete N balance, established for relatively low N application rates (280 and 350 kg N ha⁻¹ to first and second years, respectively), shows minimal N leaching losses. However, because they used large sampling intervals on young 3–5-year-old trees, no detailed trends of N absorption and assimilation could be followed. Despite the great value of these cited studies, there have been little reports about N absorption changes and utilization by different parts of perennial woody plants during one crop cycle, which undoubtedly is important for fertilizer management, particularly using split applications of fertilizer for more efficiency.

If the time of greatest N demand of adult coffee plants is known, the total amount of N applied by fertigation should be lower than that conventionally applied to maintain desired levels of crop productivity and less N could be wasted to the environment. With this assertion in mind, the objective of this study is to analyze the changes in N absorption throughout the entire coffee tree including its different plant parts during an annual growing cycle to identify the phenological phase of greatest N plant need and finding the optimal, most efficient fertilizer N rate.

Table 1
Chemical characteristics of the Typic Hapludox for different layers.

Depth (m)	pH ^a	OM ^b (g dm ⁻³)	P ^c (mg dm ⁻³)	S ^d (mg dm ⁻³)	K ^e (mmol _c dm ⁻³)	Ca ^c (mmol _c dm ⁻³)	Mg ^c (mmol _c dm ⁻³)	Al ^e (mmol _c dm ⁻³)	H + Al ^f (mmol _c dm ⁻³)	SB ^g (mmol _c dm ⁻³)	CEC ^h (mmol _c dm ⁻³)	V ⁱ (%)	M ^j (%)	N-Total ^k (mg kg ⁻¹)
0–0.2	4.7	25	114	10	2	23	9	3	31	34	65	52	8	1080
0.2–0.4	3.6	20	40	21	1	5	3	9	34	9	43	21	50	620
0.4–0.6	3.8	16	5	60	0.8	4	2	9	31	6.8	37.8	18	57	532
0.6–0.8	3.6	14	1	72	0.8	3	1	9	31	4.8	35.8	13	65	520
0.8–1.0	3.8	14	1	96	0.8	2	1	10	31	3.8	34.8	11	72	505

^a Active acidity by CaCl₂ (0.01 mol l⁻¹) method.

^b Organic matter by colorimetry.

^c Phosphorus, potassium, calcium and magnesium by ion exchange resin method.

^d Sulfur by turbidimetry method.

^e Exchangeable aluminum by titrimetric method (1 mol l⁻¹).

^f Potential acidity by pH SMP method.

^g Sum of bases.

^h Cation exchange capacity.

ⁱ Base saturation (100 × SB/CEC).

^j Aluminum saturation (100 × Al³⁺/Effective CEC).

^k Kjeldahl method (Rajj et al., 2001).

2. Materials and methods

2.1. Description of the experimental area

The experiment was carried from August 1, 2008 to July 24, 2009, at a commercial coffee farm (Fazenda Morena), Barreiras, BA, Brazil, 11°46′00″S and 45°43′32″W; 740 m above sea level. The area is located in the central highlands of Brazil, previously covered by savannah, locally called “cerrado”, with almost 0% slope and good availability of fresh irrigation water. The soil is sandy, 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 75% sand, 3% silt and 22% clay, and an available water capacity of 85 mm in the top 1 m layer. The climate is tropical sub-humid (Aw) according to Koppen (1931) with yearly rainfall ranging from 800 to 1800 mm concentrated from October to April, with a dry season that demands irrigation, and a yearly average air temperature of 25 °C.

A detailed description of the main climate parameters is presented in Table 2.

Coffee plants (*Coffea arabica* L.) were 7–8 years old, well established and at full maturity, planted at a row spacing of 3.8 m and 0.5 m between plants, in a circular arrangement to allow central pivot irrigation/fertigation. The root system mostly explored the 0–0.6 m layer, with very few roots reaching the 1.0 m depth. Fig. 1 shows the distribution of the experimental plots along the 4th circle, each plot consisting of three labeled plants having on both sides at least four border plants. This arrangement assumes that one labeled plant on each side of the central labeled plant is sufficient to assure that the other non-labeled border plants do not influence the N absorption by the central plant. Errors caused by this design are included in the non-evaluated parts of the system, e.g. soil, litter, and various losses.

Irrigation was applied according to normal farm management practice, operating continuously except during harvest at a rate of 3–4 mm day⁻¹ every second day, including nutrient applications of N, P and K, at rates of 600, 16 and 420 kg ha⁻¹ year⁻¹, respectively; micronutrients (B, Mn, Cu and Ni). Glyphosate was used for inter-row weed cleaning. The soil pH was adjusted with applications of 3000 kg ha⁻¹ year⁻¹ of lime. Gypsum applications (400 kg ha⁻¹ year⁻¹) provided S and also rendered Al less available at greater soil depths. Applications of chicken manure (65 kg N ha⁻¹ year⁻¹) and coffee bean husks (40 kg N ha⁻¹ year⁻¹) were also made. Fertigation is traditionally performed in the farm using low energy precision application (LEPA) sprinklers that concentrate the application over the plant, avoiding the interrow. Using 14-day intervals throughout each year N has been applied at rate of 600 kg ha⁻¹ year⁻¹ during the last 7 years. During the experimental period the application of unlabeled urea by LEPA sprinklers was interrupted each time on line number 4 (Fig. 1) of the experimental set-up to avoid the overlapping of the unlabeled farm fertilizer with the experimental ¹⁵N labeled urea. All other N additions mentioned above, like chicken manure and coffee bean husks, were considered in this study as other sources of N, to distinguish them from the labeled fertilizer.

Treatments consisted of ¹⁵N labeled urea enriched to 1.4 atom%, at N rates of: N₀, no urea; N₂₀₀, 200; N₄₀₀, 400; N₆₀₀, 600; N₈₀₀, 800 kg N ha⁻¹, all with 4 replicates. These rates were divided equally into 26 applications made every 14 days during the full coffee crop cycle. The application of the labeled urea was performed manually, simulating the LEPA sprinklers using a watering can, distributing the solution over the leaves. Similar to the LEPA sprinkler application, part of the fertilizer solution reached soil surface, so that N absorption occurred via leaf and soil. This manual N application avoids drift of material to neighboring trees that commonly occurs in overhead irrigation systems. The fertilizer was diluted in

an amount of water corresponding to an irrigation of 4 mm. Border plants, were fertilized by broadcasting non-enriched solid urea on soil surface, thus avoiding the mixing of the labeled and unlabeled fertilizers.

2.2. Sampling

Sampling was constrained by: (i) labeled plants could not be sacrificed until the end of the experiment; (ii) the commercial farm did not allow the sacrifice of a great number of producing trees even outside the experimental line; (iii) distance and travel cost from the farm to the laboratory (1600 km) limited the number of visits to the experimental site. Sampling dates are counted as days after beginning (DAB), starting on August 1, 2008 = 0 DAB up to July 24, 2009 = 356 DAB. Mainly due to constraints (i) and (ii), dry matter (DM) evaluations of whole plants (trees) were performed in neighbor row plants of similar size, at 13, 181 and 265 DAB. For the same purpose, at the end of the experiment (356 DAB) whole labeled plants were harvested in the experimental circle number 4. Harvested trees were divided into different plant parts (leaves, branches, stem, fruits and roots) for which nitrogen concentration (%N), ¹⁵N abundance (%¹⁵N) and dry matter (DM) were determined separately.

On labeled trees, leaves were collected at 28 day intervals except for first and last dates, viz., 13, 41, 69, 97, 125, 153, 181, 209, 237, 265, 293, 321, 349, and 356 DAB. Eighteen leaves were chosen randomly from the upper, medium and lower thirds of each ¹⁵N labeled central plant to evaluate N concentration and ¹⁵N abundance. They were collected close to fruits, on plagiotropic branches to avoid old leaves susceptible to senescence. Fruits appeared after 69 DAB and their sampling started on 181 DAB, and thereafter continued to be sampled on the same days as leaves, collecting cherries from the same branch each leaf was taken. Roots, branches and stem were sampled for N concentration and ¹⁵N enrichment only at the end of the experiment (356 DAB) when labeled plants were destructively sampled. Roots were collected in 1 m deep pits and separated with the help of a water jet.

For DM, N concentration and ¹⁵N abundance determinations all plant parts were chopped, air dried for more than three days and weighed after 1 week in a ventilated oven at 60 °C.

For %N and %¹⁵N analyses, dry samples were ground in a ‘Wiley’ type grinder and homogenized to take representative sub-samples of approximately 10 µg that were introduced in metal capsules for analysis in an automated continuous flow isotope ratio mass spectrometer (model ANCA-SL, Europa Scientific).

Because sampling was made mostly on leaves, data of %N and %¹⁵N for other compartments for intermediate dates were calculated using conversion factors based on weighed mass averages using data of the 356 DAB samples, when whole plants were collected for analyses. This procedure assumes that the proportions of the weighted averages of %N and %¹⁵N of a given compartment, between 356 DAB and any other DAB are constant, a limitation that most likely introduces some error but that could not be avoided.

To calculate the coffee bean production at 356 DAB entire cherries were treated as traditionally made in farms, being first cleaned and then placed to dry spread in thin layers on patios, exposed to sunshine, resulting in dry cherries that after the peeling process are called green coffee beans, their weight evaluated as DM (kg ha⁻¹).

2.3. Calculations

For the calculations of plant N, three containers (*C_i*) were considered: *C₁* = whole plant (stem, branch, leaf, fruit and root); *C₂* = leaf; *C₃* = fruit. For these compartments *C_i*, on dates DAB_{*j*} we considered:

Table 2
Monthly averages of climatologic data from August 2008 to July 2009 for Fazenda Morena: air temperature (T), air relative humidity (RH), net radiation (Rn), wind speed (W), rainfall (P), irrigation and rainfall ($P+I$), drainage below 1.0 m depth (Q), and evapotranspiration by Penman-Montheith.

Month/year	T (°C)	RH (%)	Rn (MJ m ⁻² day ⁻¹)	W (m s ⁻¹)	P (mm)	$P+I$ (mm)	Q (mm)	ETR (mm)
Aug-08	24.0	42.4	10.2	1.6	0.0	118.0	8.0	136.0
Sep-08	26.4	49.5	10.4	1.9	31.5	160.0	0.0	135.0
Oct-08	28.5	36.1	11.8	2.2	0.0	140.0	0.0	178.0
Nov-08	26.8	70.8	7.8	1.5	314.5	384.0	245.0	100.0
Dec-08	25.3	76.3	9.7	1.4	195.0	217.0	132.0	102.0
Jan-09	25.4	76.6	10.7	1.3	230.0	256.0	140.0	111.0
Feb-09	25.5	77.5	10.2	1.1	185.5	197.0	89.0	96.0
Mar-09	25.7	76.3	9.7	1.0	350.5	358.0	258.0	100.0
Apr-09	24.7	83.5	7.9	0.9	108.5	135.0	57.0	78.0
May-09	23.7	78.1	8.0	1.0	67.0	115.0	59.0	78.0
Jun-09	22.6	76.2	8.4	0.9	52.5	99.0	22.0	79.0
Jul-09	22.2	69.9	9.3	1.1	0.0	52.0	0.0	78.0
Annual	25.1	67.8	9.5	1.3	1535.0	2232.0	1010.0	1271.0

(a) Total nitrogen quantity $TNQ_{i,j}$ (g plant⁻¹ or kg ha⁻¹)

$$TNQ_{i,j} = \frac{DM_{i,j} \times \%N_{i,j}}{100} \quad (1)$$

(b) Quantity of N derived from fertilizer $QNdff$ (g plant⁻¹ or kg ha⁻¹)

$$QNdff_{i,j} = TNQ_{i,j} \left[\frac{^{15}N_{i,j} - ^{15}N_{nat}}{^{15}N_{fert} - ^{15}N_{nat}} \right] \quad (2)$$

where $^{15}N_{nat}$ is the natural abundance of ^{15}N in C_i taken as 0.365 for all compartments and $^{15}N_{fert}$ is the fertilizer abundance, 1.4%.

(c) N absorption efficiency or fertilizer N recovery $R_{i,j}$ (%)

$$R_{i,j} = \left[\frac{QNdff_{i,j}}{N_{Rate}} \right] \times 100 \quad (3)$$

For compartment C_1 , the calculations of TNQ and $QNdff$ were made summing the estimates for leaf, branch, stem, fruit and root.

2.4. Statistical analyses

Data were submitted to Repeated Measures Analysis of Variance, using the Statistica program (Statsoft Inc., 2004) with the application of the Tukey test at the 5% probability level. The graphs were made using the Sigma Plot program (Systat Software Inc., 2006).

3. Results

Mature coffee trees accumulate DM mainly by new leaves and fruit during one growing cycle. The annual percentage increase in DM of the whole plant is therefore relatively small. Therefore, because DM was sampled only at four dates in relatively short intervals, intermediate data were achieved by linear interpolation. Leaf dry matter increased from 5000 to 7800 kg ha⁻¹ (265 DAB) and after that decreased until 4400 kg ha⁻¹ (356 DAB). As already mentioned, stem (mean value of 15000 kg ha⁻¹), branch (mean 6300 kg ha⁻¹) and root (mean value of 8500 kg ha⁻¹) dry matters had no significant changes during the year of evaluation (data not shown). The overall average root DM of 8500 kg ha⁻¹ was much lower than the 27,100 kg ha⁻¹ of dry matter of the vegetative above ground portion, showing a shoot:root ratio of 3.2:1. The average fruit DM for all treatments increased from a negligible quantity at 69 DAB to

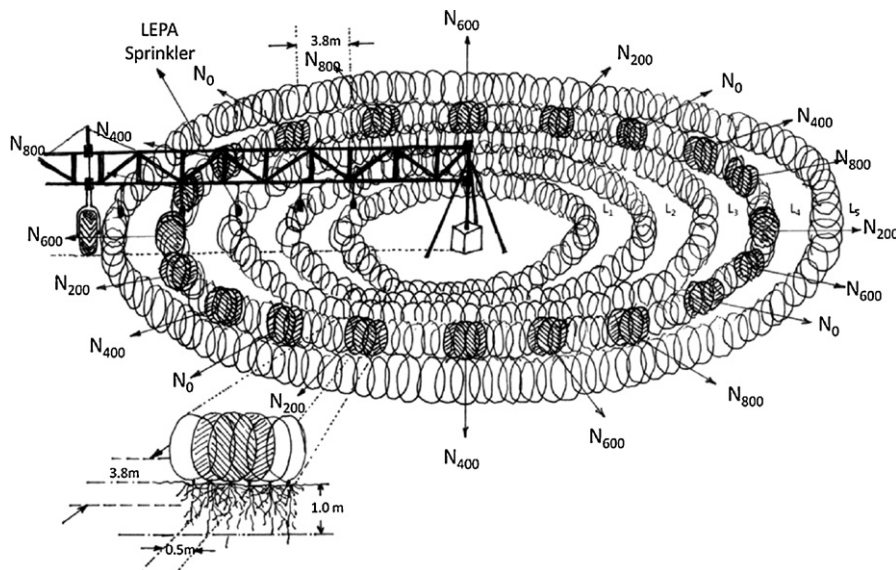


Fig. 1. Schematic presentation of the field with the experimental circle (L_4), showing: (i) random distribution of plots (3 labeled trees each, with different nitrogen rates: N_0 = without nitrogen fertilizer; N_{200} = 200; N_{400} = 400; N_{600} = 600; N_{800} = 800 kg ha⁻¹ year⁻¹ of N); (ii) the distance between rows and between coffee plants (3.8 m × 0.5 m); (iii) the effective depth of the root system (1.0 m). L_i = lines.

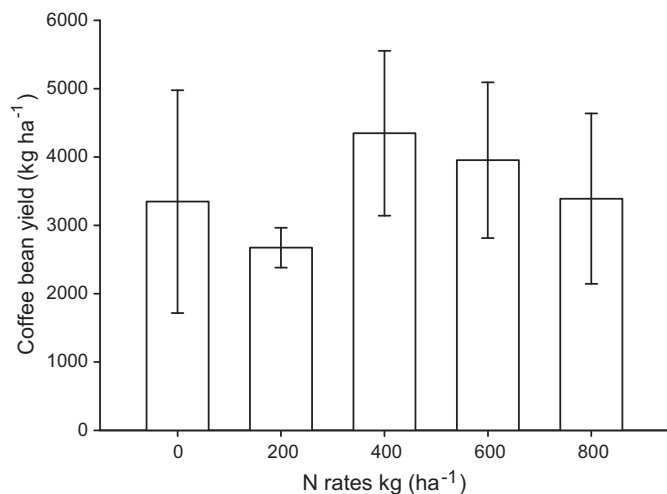


Fig. 2. Green coffee bean yield (kg ha^{-1}) for the 2009 and 2010 harvests. Bars represent \pm one standard deviation.

7774 kg ha^{-1} ($1477 \text{ g plant}^{-1}$) of dry cherries at harvest (DAB 356), following a logarithmic pattern. At harvest, due to the high coefficients of variation, no statistic difference could be detected between fertilizer rate treatments in relation to green bean yield (Fig. 2). This suggests that the four replicates for fruit yield were not enough to differentiate treatments. Although the 95% confidence interval of the N_{200} treatment lies outside that of the N_{400} treatment and essentially also outside that for the N_{600} treatment, added to the fact that the average yield of N_0 was higher than that N_{200} makes us to consider that the N_{200} average is by chance too low. Therefore, based on the statistical analysis, we consider no difference between treatments.

Total nitrogen of the whole plant, leaf and fruit (Fig. 3) had almost no significant difference among treatments during the year-long growth.

In general terms the whole plant TNQ increased during the whole crop cycle, with few decreases during the initiation of the cycle and at the end (349 DAB) increasing again at harvest time, when values were around 800 kg N ha^{-1} regardless of treatment (Fig. 3A). This is in agreement with the above-mentioned fact that these mature trees present a small yearly DM increase, and that stem, branch and root DM values mask those of leaf and fruit.

Leaf TNQ decreased slowly from 13 to 69 DAB, and subsequently tended to increase until 265 DAB for all rates. After this date leaf total N decreased again reaching values close to those at the beginning of the growth cycle, indicating a cyclic pattern. The TNQ decrease in time is due to the DM drop after 265 DAB, since the leaf %N (data not shown) changed very little during the cycle. Leaf showed differences in total N among rates, and the coffee plants receiving $800 \text{ kg N ha}^{-1} \text{ year}^{-1}$ had significantly higher N accumulation than the other treatments (Fig. 3B). The N concentration in leaf had means of 31, 32, 32 and 34 g kg^{-1} , respectively, for plants exposed to rates of 200, 400, 600 and 800 kg N ha^{-1} during the year.

Fruit TNQ increased since evaluation began and had its peak at 321 DAB, decreasing thereafter and raising again up to the previous values (Fig. 3C). There was no significant difference between treatments for fruit TNQ, which showed values around 170 kg N ha^{-1} at the end of the season, indicating that approximately 22% of N in the whole coffee plant is exported through the harvest regardless of the amount of applied fertilizer N.

The whole plant QN_{dff} (Fig. 4A) is the sum of stem, branch, root (partial data not shown), leaf (Fig. 4B) and fruit (Fig. 4C), differed among treatments. The whole plant QN_{dff} values seem fairly high, however, the N from soil and other sources (QN_{dfs}) repre-

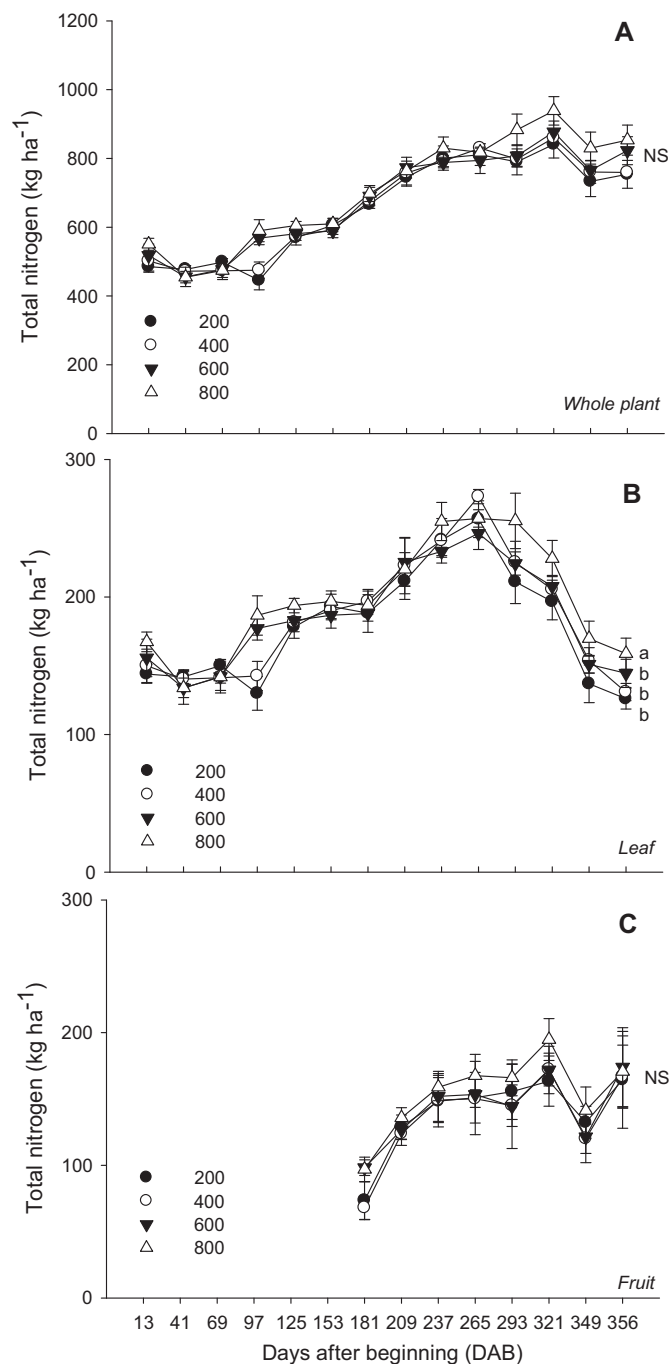


Fig. 3. Time course of the total N quantity (TNQ) in the whole plant (A), leaf (B) and fruit (C) of fertigated coffee plants from 13 to 356 days after beginning (DAB), with different N rates (200; 400; 600; 800 kg ha^{-1} of N). Lines followed by the same lower case letter are not significantly different. NS means statistically no significant ($P < 0.05$).

sents at least 40% of the total N acquired by plants. Whole plant and leaf QN_{dff} presented significant differences in relation to treatments, and the peak for leaf (Fig. 4B) corresponding to N_{800} , is 139 kg N ha^{-1} at 293 DAB (fruit maturation phase), and for N_{200} , is 68 kg N ha^{-1} at DAB 237 (fruit filling phase). Fruit N_{dff} (data not shown) and fruit QN_{dff} (Fig. 4C), although with a smaller amount of N derived from fertilizer, followed the same pattern as that for leaf, increasing up to harvest, but not showing differences between N_{200} and N_{400} , and between N_{600} and N_{800} .

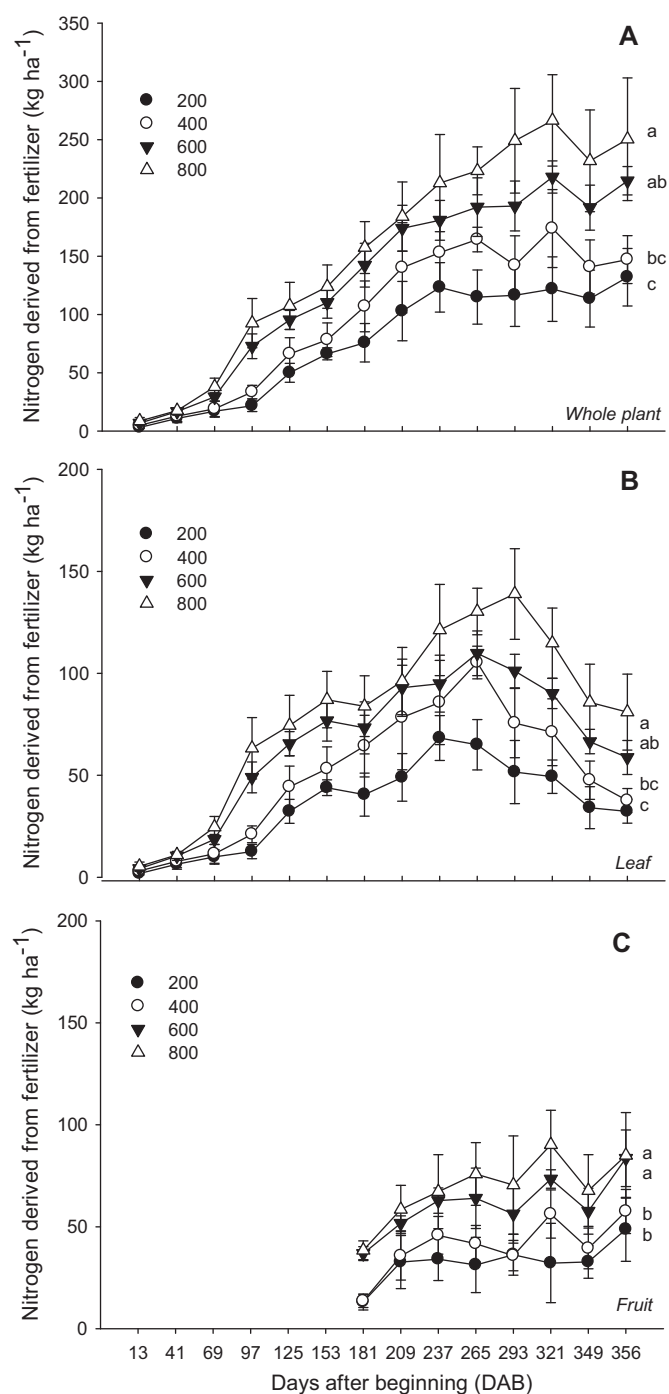


Fig. 4. Quantity of N derived from fertilizer (N_{dff}) in the whole plant (A), leaf (B) and fruit (C) of fertigated coffee plants from 13 to 356 days after beginning (DAB) with different N rates (200; 400; 600; 800 kg ha^{-1} of N). Lines followed by the same low case letter are not significantly different ($P < 0.05$).

Plants subjected to lower rates of N fertilizer were those with the highest percentage of N recovered from the fertilizer input by whole plant, leaf and fruit (Fig. 5). The highest values of whole plant N recovery appear for all treatments during fruit filling from 209 to 237 DAB, except for N_{800} that was at 97 DAB, the baby fruit stage. Fruit N recovery (Fig. 5C) increased from 181 to 237 DAB, and thereafter decreased slightly with an unexpected increase at harvest. There was no difference among treatments with exception of N_{200} .

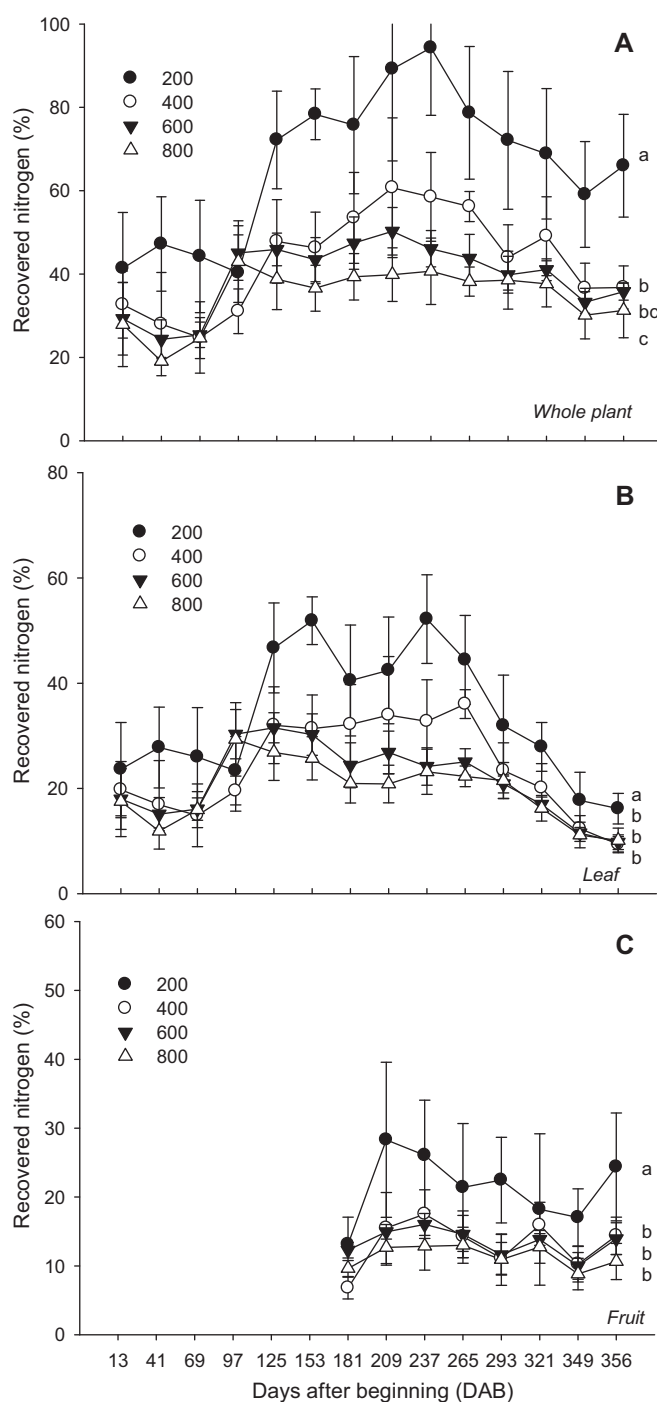


Fig. 5. Percent of the applied N recovered (R , %) by the whole plant (A), leaf (B) and fruit (C) of fertigated coffee plants from 13 to 356 days after beginning (DAB), with different N rates (200; 400; 600; 800 kg ha^{-1} of N). Lines followed by the same low case letter are not significantly different ($P < 0.05$).

4. Discussion

This experiment assessed the N absorption changes in plant compartments over time for a coffee crop grown in the Brazilian “cerrado” that was previously cultivated under high N fertilizer rates. The aim was to find out the physiological stage of greatest N need by the plant, in order to be able to suggest a more rational N fertilizer rate application interval and to show that the N requirement of adult coffee plants is less than the high rate conventionally used in fertigation. It is important to bear in mind that for woody

perennials, there is a change over the years in the amount of N required to achieve an adequate yield, and so this discussion initially applies only for so-called adult plants having an age of at least 7 years.

About 76% of the DM of the coffee plants was aboveground, similar to 6-year-old citrus biomass that presented 70% of its dry matter in the shoot (Mattos et al., 2003), but very different from results for young coffee plants, which have only 43% of their DM aboveground (Reichardt et al., 2009). The root system DM was much less than shoot DM, and was very low in relation to expected values for adult coffee plants in Brazilian plantations, presenting the same weight that was found by Costa (2006) for 3-year-old plants. Dafert and Braga (1917), cited in Franco and Inforzato (1946), report a ten times larger root system for 20-year-old coffee plants reaching down to the 0.95 m depth and Franco and Inforzato (1946), report an eight times larger root system for 10-year-old coffee plants. The reason for this discrepancy should mainly be due to coffee grown in different localities and under different managements primarily adopted in the past (Rena and Maestri, 1986). Nevertheless, our root system was not well developed and displayed a sharp reduction at the 1.0-m depth. Most of it occurred in the upper layers (0–0.6 m) where nutrients were found in excess and where water was added in a continuous and abundant way. From 0.2 m downwards pH decreases, restricting the availability of Ca and Mg which negatively affects root development (Marschner, 1995; Raij et al., 1997). The shallow root system explores a smaller soil volume increasing the chances of N losses through leaching or denitrification, although not affecting green coffee yield. This information suggests that the current practice of applying N at rates above $200 \text{ kg ha}^{-1} \text{ year}^{-1}$ may be excessive.

Leaf dry matter increased until the end of the fruit-filling phase. Subsequently, it decreased due to photosynthate loss from leaf to the strong drain, the fruit.

Green coffee production did not respond to different fertilizer rates (Fig. 2), probably due to the fact that plants were already 8 years old and were fertilized during the previous 7 years at the high rate of 600 kg N ha^{-1} . Presumably plants had already a sufficient N reserve not to respond to the lower or higher rates in terms of yield. The similar green coffee yield for all rates may indicate a remobilization of the accumulated N of past years from leaf, branch and root to fruit, as suggested by Lima-Filho and Malavolta (2003). This fact could indicate that adult coffee plants are fertigated at rates above plant requirement and that the rate of 600 kg ha^{-1} may be reduced without production losses, at least in the short term. Moreover, the rate of 800 kg ha^{-1} did not lead to a yield increase. Sanzonowicz et al. (2003) obtained similar results after a 4-year study in trimmed adult coffee plants. Lawlor et al. (2001) states that the greatest responses to N supply occur in places with N limitation, and where the N supply exceeds plant requirement so that no further dry matter is produced per unit additional N. The nutrient can be accumulated in unproductive components, and used later when N is scarce.

Total N quantity in the whole plant increased for each rate and showed a tendency to decrease during later phases (Fig. 3A), probably due to DM decrease and N losses from the plant to the environment. The coffee plant loses a good part of its leaves, which are yearly replaced by branch regrowth. These new branches are responsible for coffee bean production in future years.

The higher quantity of N derived from fertilizer followed the rate of applied N in the whole plant, leaf and fruit. The whole coffee plant absorbed at harvest time from 130 to 250 kg N ha^{-1} from fertilizer for rates of $200\text{--}800 \text{ kg N ha}^{-1}$. Considering the efficiency as the unit of N absorbed per unit of N applied, the efficiency of the rate 200 kg ha^{-1} is twice the value found for 800 kg ha^{-1} , confirming that N efficiency is greater with a lower N rate and declining substantially with increasing rates (Fig. 4A).

The peaks of fertilizer N recovery in whole plants (Fig. 5A) were high, mainly for N_{200} that absorbed up to 94% of the applied N up to 237 DAB. For the other treatments peaks declined with increasing N rates as follows: 61% for N_{400} , 50% for N_{600} , and the lowest value of 43% for N_{800} . The value of $R = 94\%$ for N_{200} is far above all reports found in the literature, e.g. Quiñones et al. (2005) with around 70% for citrus. This result has to be analyzed with caution because it is an intermediate value (at 237 DAB), affected by all restrictions mentioned above, mainly DM measurements and interpolations of $\%N$ and ^{15}N data. Its standard deviation is very high so that the minimum value is 78%, and maximum 110%. Data for 356 DAB, when whole plants were collected, are much more consistent. The large value of $QNdff_1$ (Fig. 4A) and the low recovery of N in the whole plant (Fig. 5A) indicates a luxurious N absorption for the highest rate. The peak time was for the fruit filling stage for all treatments, except for N_{800} that had its peak at the baby fruit phase showing that for high rates the N absorption pattern changes. Despite continuous fertilizer application, around 209 DAB, the N recovery for N_{800} changed very little, and for the other rates decreased, indicating a possible increment of N losses to the environment. A similar behavior was also reported by Lea-Cox et al. (2001) for 4-year-old citrus.

The N recovery for the whole plant at harvest time was 66% for N_{200} , 37% for N_{400} , 36% for N_{600} and 31% for N_{800} . This recovery of adult coffee plants was higher than for younger plants grown in a traditional planting field, in which it was only 34% (Reichardt et al., 2009), with exception to N_{800} , and even much higher when comparing with other perennial crops in the field. Young pear trees recovered only 6.3% (Neto et al., 2006), adult peach Trees 8.3–12.7% (Nario et al., 2003), and adult citrus Trees 25.5% of the applied N (Mattos et al., 2003). The N recovery of our coffee plant is near the highest values reported for similar tree crops growing under controlled environment, e.g. 8-year-old citrus that recovered 63–75% of the applied N (Quiñones et al., 2003, 2005, 2007).

According to Raij et al. (1997), the N concentration range considered adequate for coffee leaves varies during all development stages from 26 to 32 g kg^{-1} . The values obtained in this study (data not shown) are within this range, with exception to the rate of $800 \text{ kg N ha}^{-1} \text{ year}^{-1}$ that had a higher level, reaching a maximum of 36 g kg^{-1} at the beginning of the harvest time. As a result the total quantity of N in leaves did not differ among the three lower rates (Fig. 3B) during the whole growing cycle, irrespective to the development stage.

The total quantity of N fluctuation in leaves (Fig. 3B) was very large during the crop cycle due to: (1) a translocation of N from leaf to fruit during the expansion phase of fruit and from old to young leaf which are produced constantly (Amaral et al., 2001; Taiz and Zeiger, 2004), and (2) the drop of old leaf during the fruit maturation stage, i.e., after 265 DAB. Carelli et al. (2006), reviewing the N metabolism in coffee plants, found reports observing a greater nitrate absorption before anthesis (DAB 41) and at the beginning of fruit maturation (DAB 293), corresponding to the period of largest N drain to flower and fruit. Our data show this only for fruit maturation since in this period leaf TNQ was greater indicating N mobilization from leaves, for translocation to fruit at maturation, which has increased fruit TNQ (Fig. 3C) throughout the cycle.

The time course of leaf $QNdff$ (Fig. 4B) follows a behavior of a sharp initial increase and a later leveling-off. The curves present a closely sigmoidal behavior, reaching a maximum between 237 and 293 DAB. For all rates, except N_{800} , leaf $QNdff$ peaked around the fruit filling stage (237–265 DAB), indicating that during this period there is possibly a higher N uptake by leaf for subsequent translocation to fruit. This confirms the proposal made by Da Matta et al. (1999) in a study on nitrate concentration in coffee leaves from a traditional coffee production region. These results confirm that N fertilizer applications might be better targeted up to this point, and

not spread throughout crop growth as is the current practice. However, the problem of minimizing leaching losses has also to be taken into account, because the maximum N demand peak falls within the rainy period, with the maximum drainage losses (Table 2). These losses will, however, be lower for lower application rates.

Leaf fertilizer N recovery followed the inverse sequence of applied N rates: $N_{200} > N_{400} > N_{600} > N_{800}$ (Fig. 5B), and a highlight is given to N_{200} that reached a maximum of 52% while for the other treatments it oscillated around 22%. After 265 DAB plants were in the fruit maturation stage and leaf N recovery fell for all treatments. These results indicate a lower fertilizer N absorption once the proportion absorption/application decreases up to harvest, and that during the fruit maturation stage leaves translocate N to the strongest sink. These results are in agreement with Valarini et al. (2005) who analyzed only the N concentration in adult coffee plant leaves, but differ from pears that present greater remobilization of N from leaves to flowers (Tagliavini et al., 1997). The period of greatest N fertilizer absorption by leaves was that of fruit filling for N_{200} (52%) and N_{400} (36%), while for N_{600} (32%) and N_{800} (29%) the phase was fruit expansion (Fig. 5B). Fenilli et al. (2007) presented similar results for N_{200} and N_{400} , when during the fruit filling phase coffee plants absorbed 71% of the 280 kg ha^{-1} and 36% of the 350 kg ha^{-1} applied during the first and second years, respectively. These results also indicate that the time interval of the N rate should favor the period immediately prior to the fruit filling and expansion phases. The decrease of leaf N recovery (Fig. 5B) and leaf TNQ (Fig. 3B) at the end of the maturation period can also be explained by leaf senescence and gaseous N losses to the atmosphere as ammonia (Farquhar et al., 1980; Hörtensteiner and Feller, 2002).

For all tested N rates, the amount of total N in fruit at the end of the cycle was of the order of 170 kg N ha^{-1} (Fig. 3C), a value slightly below the most efficient application rate in terms of recovery, 200 kg N ha^{-1} , showing that this rate replaces fruit N export, which is the main export of this crop.

Fruit QN_{dff} increased over the cycle with a peak at maturation (Fig. 4C), showing a resembling behavior in relation to the whole plant QN_{dff} . During fruit growth and development the N requirement increases, plants need more N which should be supplied during the previous stage, i.e., beginning of fruit setting. Malavolta et al. (2002) studying only flower, leaf and branch report that for the cultivars Yellow Catuaf and Mundo Novo, flowers contained 20% of the N, so that the N supply at flowering just before fruit formation is a good choice.

Fruit N recovery (Fig. 5C) increased from 181 to 209 DAB and thereafter decreased corroborating the hypothesis that the application of N should happen up to this phase. Only the more efficient N_{200} for leaf differed significantly from all other treatments, as happened with the N fertilizer recovery by leaf (Fig. 5B). This can be interpreted as a response to the high plant demand faced by a very low application rate, for both compartments with reflection in the whole plant N fertilizer recovery (Fig. 5A). The decrease of fruit N recovery from 209 DAB on, mainly for the 200 kg ha^{-1} rate also implies that the N of the late applications of urea were not so well availed, contributing to greater losses and confirming that greater amounts of fertilizer should be made available before fruit formation. Regarding fruit, it can be stated that adult fertigated coffee plants that previously received high quantities of N, the rate can be lowered.

Data of N absorption measured over time during one production cycle of fertigated coffee indicate that the fertilizer N supply should not be made continuously, but preferentially before N demand peaks. The demand peak occurs on the final fruit-filling stage, corresponding to the natural leaf senescence and an intense N remobilization from leaf to fruit, so that the major quantity of N should be applied before this stage. The fertilizer fruit N recovery by the coffee trees submitted to $200 \text{ kg N ha}^{-1} \text{ year}^{-1}$ having been the

highest and very close to crop's export, persuades us to recommend a rate of this order of magnitude for the present conditions.

Acknowledgements

We wish to express sincere thanks to the São Paulo Research Foundation (FAPESP) and to the Brazilian Research Council (CNPq) for the scholarships and funding provided for this study, and also to Herman Burema, Wesley Vieira (Fazenda Morena) and Edmilson Figueredo (Bahia Foundation) for field support. We are also grateful to The Crawford Fund for partial support to the senior author during the writing up of this work.

References

- Amaral, J.A.T., Da Matta, F.M., Rena, A.B., 2001. Effects of fruiting on the growth of arabica coffee trees as related to carbohydrate and nitrogen status and to nitrate reductase activity. *Braz. J. Plant Physiol.* 13, 66–74.
- Boaretto, A.E., Schiavinato-Neto, P., Muraoka, T., Oliveira, M.W., Trivelin, P.C.O., 1999. Foliar nitrogen supply to young citrus plants. *Sci. Agric.* 56, 621–626.
- Bustamante, C., Ochoa, M., Rodriguez, M.I., 1997. Balance of three N fertilizers sources in a Cuban Oxisol planted with *Coffea arabica* L. *Tropicultura* 15, 169–172.
- Carelli, M.L.C., Fahl, J.I., Ramalho, J.D.C., 2006. Aspects of nitrogen metabolism in coffee plants. *Braz. J. Plant Physiol.* 18, 9–21.
- Costa, F.M.P., 2006. Growth and development of the coffee plant related to nitrogen fertilization. Dissertation, University of São Paulo, Piracicaba.
- Da Matta, F.M., Amaral, J.A.T., Rena, A.B., 1999. Growth periodicity in trees of *Coffea arabica* L. in relation to nitrogen supply and nitrate reductase activity. *Field Crop Res.* 60, 223–229.
- EMBRAPA (Empresa Brasileira de Pesquisa Agropecuária), 2006. Brazilian Soil Classification System, second ed. Embrapa Solos, Rio de Janeiro.
- FAO, 2010. Food and Agriculture Organization, Fertistat: Fertilizer Use Statistic (2010), <http://www.fao.org/ag/agl/fertistat/fst.fubc.en.asp> (Accessed 11.22.10).
- Farquhar, G.D., Firth, P.M., Wetselaar, R., Weir, B., 1980. On the gaseous exchange of ammonia between leaves and environment—determination of the ammonia compensation point. *Plant Physiol.* 66, 710–714.
- Fenilli, T.A.B., Reichardt, K., Dourado-Neto, D., Trivelin, P.C.O., Favarin, J.L., Costa, F.M.P., Bacchi, O.O.S., 2007. Growth, development, and fertilizer (15) N recovery by the coffee plant. *Sci. Agric.* 64, 541–547.
- Fenilli, T.A.B., Reichardt, K., Favarin, J.L., Bacchi, O.O.S., Silva, A.L., Timm, L.C., 2008. Fertilizer (15) N balance in a coffee cropping system: a case study in Brazil. *Rev. Bras. Cienc. Solo.* 32, 1459–1469.
- Franco, C.M., Inforzato, R., 1946. The coffee root system in the main soil types from São Paulo State. *Bragantia* 6, 443–478.
- Hardarson, G., 1990. Use of Nuclear techniques in studies of soil-plant relationships. Training Course Series 2, IAEA, Vienna.
- Hörtensteiner, S., Feller, U., 2002. Nitrogen metabolism and remobilization during senescence. *J. Exp. Bot.* 53, 927–937.
- Köppen, W., 1931. *Grundriss der Klimakunde*. Walter D E Guyter & Co., Berlin.
- Lawlor, D.W., 2002. Carbon and nitrogen assimilation in relation to yield: mechanisms are the key to understanding production systems. *J. Exp. Bot., Inorganic Nitrogen Assimilation Special Issue* 53, 773–787.
- Lawlor, D.W., Lemaire, G., Gastal, F., 2001. Nitrogen, plant growth and crop yield. In: Lea, P.J., Morot-Gaudry, J.F. (Eds.), *Plant Nitrogen*. Springer-Verlag, Berlin, pp. 343–367.
- Lea, P.J., Morot-Gaudry, J.F. (Eds.), 2001. *Plant Nitrogen*. Springer-Verlag, Berlin.
- Lea-Cox, J., Syvertsen, J., Graetz, D., 2001. Springtime (15) nitrogen uptake, partitioning, and leaching losses from young bearing Citrus trees of differing nitrogen status. *J. Am. Soc. Hort. Sci.* 126, 242–251.
- Lima-Filho, O.F., Malavolta, E., 2003. Studies on mineral nutrition of the coffee plant (*Coffea arabica* L. cv. Catuaf Vermelho). LXIV. Remobilization and re-utilization of nitrogen and potassium by normal and deficient plants. *Braz. J. Biol.* 63, 481–490.
- Lobell, D.B., 2007. The cost of uncertainty for nitrogen fertilizer management: a sensitivity analysis. *Field Crop Res.* 100, 210–217.
- Malavolta, E., Favarin, J.L., Malavolta, M., Cabral, C.P., Heinrichs, R., Silveira, J.S.M., 2002. Nutrients repartition in the coffee branches, leaves and flowers. *Pesqui. Agropecu. Bras.* 37, 1017–1022.
- Marschner, H., 1995. *Mineral Nutrition of Higher Plants*, 2nd ed. Academic Press, London.
- Mattos, D., Graetz, D., Alva, A., 2003. Biomass distribution and 15(N) partitioning in citrus trees on a sandy entisol. *Soil Sci. Soc. Am. J.* 67, 555–563.
- Milroy, S.P., Asseng, S., Poole, M.L., 2008. Systems analysis of wheat production on low water-holding soils in a Mediterranean-type environment. II. Drainage and nitrate leaching. *Field Crop Res.* 107, 211–220.
- Nario, A., Pino, I., Zapata, F., Albornoz, M.P., Baherle, P., 2003. Nitrogen (15N) fertilizer use efficiency in peach (*Prunus persica* L.) cv. Goldencrest trees in Chile. *Sci. Hortic.* 97, 279–287.
- Natale, W., Marchal, J., 2002. Absorption and distribution of (15) N nitrogen in citrus mitis Bl. *Rev. Bras. Frutic.* 24, 183–188.

- Neto, C., Carranca, C., Varennes, A., Oliveira, C., Clemente, J., Sobreiro, J., 2006. Nitrogen use efficiency of drip-irrigated rocha pear trees. *Acta Hort.* (ISHS) 721, 337–341.
- Quiñones, A., Banuls, J., Millo, E., Legaz, F., 2003. Effects of N-15 application frequency on nitrogen uptake efficiency in Citrus trees. *J. Plant Physiol.* 160, 1429–1434.
- Quiñones, A., Banuls, J., Primo-Millo, E., Legaz, F., 2005. Recovery of the (15)N labeled fertilizer in citrus trees in relation with timing of application and irrigation system. *Plant Soil* 268, 367–376.
- Quiñones, A., Martinez-Alcantara, B., Legaz, F., 2007. Influence of irrigation system and fertilization management on seasonal distribution of N in the soil profile and on N-uptake by citrus trees. *Agric. Ecosyst. Environ.* 122, 399–409.
- Raij, B.van, Andrade, J.C., Cantarella, H., Quaggio, J.A., 2001. Chemical Analysis for Evaluation of Tropical Soil Fertility. Instituto Agronômico, Campinas.
- Raij, B.van, Cantarella, H., Quaggio, J.A., 1997. Recommendations for Fertilization and Liming For the State of São Paulo. Instituto Agronômico, Campinas.
- Reichardt, K., Da Silva, A.L., Fenilli, T.A.B., Timm, L.C., Bruno, I.P., Volpe, C.A., 2009. Relation between nitrogen fertilization and water soil conditions for a coffee plantation from Piracicaba, SP. *Coffee Sci.* 4, 41–55.
- Reichardt, K., Bacchi, O.O.S., 2004. Isotopes in soil and plant investigations. In: Hillel, D. (Ed.), *Encyclopedia of Soils and the Environment*, vol. 1. Elsevier, Amsterdam, pp. 280–284.
- Rena, A.B., Maestri, M., 1986. Coffee physiology. In: Rena, A.B., Malavolta, E., Rocha, M., Yamada, T. (Eds.), *Coffee Crop: Factor That Affect Productivity*. Potafos, Piracicaba, pp. 13–86.
- Sanzonowicz, C., Toledo, P.M.R., Sampaio, J.B.R., Guerra, A.F., Silva, D.T.M., 2003. Nitrogen fertilization in trimmed coffee in a Savannah Oxisol. *Research and Development Bulletin* 104, Embrapa, Planaltina.
- Sitthaphanit, S., Limpinuntana, V., Toomsan, B., Panchaban, S., Bell, R.W., 2009. Fertilizer strategies for improved nutrient use efficiency on sandy soils in high rainfall regimes. *Nutr. Cycl. Agroecosyst.* 85, 123–139.
- Soil Survey Staff, 2010. *Keys to Soil Taxonomy*, 11th ed. USDA-Natural Resources Conservation Service, Washington.
- Statsoft Inc., 2004. *Statistica v 8.0*. Statsoft Inc., Tulsa.
- Systat Software Inc., 2006. *Sigma Plot v 10.0*. Systat Software Inc, San Jose.
- Tagliavini, M., Quartieri, M., Millard, P., 1997. Remobilised nitrogen and root uptake of nitrate for spring leaf growth, flowers and developing fruits of pear (*Pyrus communis* L.) trees. *Plant Soil* 195, 137–142.
- Taiz, L., Zeiger, E., 2004. *Plant Physiology*, 3rd ed. Artmed, Porto Alegre.
- Valarini, V., Bataglia, O.C., Fazuoli, L.C., 2005. Macronutrients in leaves and fruits of dwarf arabica coffee cultivar. *Bragantia* 64, 661–672.