GROWTH, DEVELOPMENT, AND FERTILIZER-15N RECOVERY BY THE COFFEE PLANT

Tatiele Anete Bergamo Fenilli[1]; Klaus Reichardt[2]*; Durval Dourado-Neto[3]; Paulo César Ocheuze Trivelin[4]; José Laercio Favarin[3]; Flávio Murilo Pereira da Costa[5]; Osny Oliveira Santos Bacchi[2]

[1]*Fundação Universidade Regional de Blumenau (FURB) - Depto. de Engenharia Florestal, Campus II, R. São Paulo, 3250 - Itoupava Seca, 89030-000 - Blumenau, SC - Brasil.*
[2]*USP/CENA - Lab. Física do Solo, C.P. 96 - 13400-970 - Piracicaba, SP - Brasil*
[3]*USP/ESALQ - Depto de Produção Vegetal, C.P. 09 - 13418-970 - Piracicaba, SP - Brasil.*
[4]*USP/CENA - Lab. Isótopos Estáveis.*
[5]*IICA-Agrometeorologista/SEAF, Ministério do Desenvolvimento Agrário - SBN-Q1 Ed. Palácio do Desenvolvimento-Sala 604, 70057-900 - Brasília, DF - Brasil.*
Corresponding author<klaus@cena.usp.br>

ABSTRACT: The relationship between growth and fertilizer nitrogen recovery by perenial crops such as coffee is poorly understood and improved understanding of such relations is important for the establishment of rational crop management practices. In order to characterize the growth of a typical coffee crop in Brazil and quantify the recovery of ^{15}N labeled ammonium sulfate, and improve information for fertilizer management practices this study presents results for two consecutive cropping years, fertilized with 280 and 350 kg ha^{-1} of N, respectively, applied in four splittings, using five replicates. Shoot dry matter accumulation was evaluated every 60 days, separating plants into branches, leaves and fruits. Labeled sub-plots were used to evaluate N-total and ^{15}N abundance by mass spectrometry. During the first year the aerial part reached a recovery of 71% of the fertilizer N applied up to February, but this value was reduced to 34% at harvest and 19% at the beginning of the next flowering period due to leaf fall and fruit export. For the second year the aerial part absorbed 36% of the fertilizer N up to March, 47% up to harvest and 19% up to the beginning of the next flowering period. The splitting into four applications of the used fertilizer rates was adequate for the requirements of the crop at these growth stages of the coffee crop.
Key words: *Coffea arabica* L., stable isotope, nitrogen, dry matter accumulation, ^{15}N

CRESCIMENTO, DESENVOLVIMENTO E RECUPERAÇÃO DO 15N DO FERTILIZANTE EM CAFEEIRO

RESUMO: A relação entre o crescimento e a recuperação do N do fertilizante aplicado a culturas perenes como a do café não é bem conhecida e é um passo chave para o estabelecimento de práticas culturais racionais. Para caracterizar o crescimento de uma cultura típica de café do Brasil, quantificar a recuperação do sulfato de amônio marcado com ^{15}N e contribuir para a melhoria das práticas de manejo de fertilizantes, este estudo apresenta dados para dois anos agrícolas consecutivos, com fertilização de 280 e 350 kg ha^{-1} de N, respectivamente, aplicados em quatro vezes, utilizando cinco repetições. O acúmulo de matéria seca pela parte aérea foi medido a cada 60 dias separando as diferentes partes da planta em ramos, folhas e frutos. Subparcelas marcadas com ^{15}N foram utilizadas para determinar as concentrações de ^{15}N e N-total por espectrometria de massa. A parte aérea da planta durante o 1° ano chegou a absorver 71% do N do adubo aplicado até fevereiro, mas esta porcentagem se reduziu até 34% na época da colheita e até 19% no início da nova floração. No 2° ano a parte aérea chegou a absorver 36% do N do adubo até março, 47% na colheita e 19% no início da próxima floração. As doses aplicadas (280 kg ha^{-1} de N no 1° ano e 350 kg ha^{-1} no 2° ano, ambos parcelados em quatro vezes) foram adequadas neste estágio de crescimento da cultura.
Palavras-chave: *Coffea arabica* L, isótopo estável, nitrogênio, acumulação de matéria seca, ^{15}N

INTRODUCTION

Coffee (*Coffea arabica* L.) is one of the most important crops of Brazilian agribusiness and despite the fact that its management practices are at a well defined level several aspects of its cropping still need to be clarified, one of them being the nitrogen fertilization as related to growth and development. Nitrogen is the nutrient of greatest demand by this crop,

and is second (after Potassium) in amounts translocated to fruits, therefore being the most exported one (Catani & Moraes, 1958; Malavolta, 1993). The lack of N mostly limits its growth and productivity, therefore the application of N as fertilizer represents a great share in the total production cost (Vaast et al., 1998). High rates of 450 kg ha^{-1} of N or more have been recommended per agricultural year to be applied during the rainy season (Raij et al., 1996).

The best use of fertilizer by the coffee plant, mainly nitrogen, is related to the appropriate choice of rates and splittings (Martins, 1981; Viana, 1980) and, mainly, timing (Küpper, 1976; Moraes & Catani, 1964; Viana, 1980; Malavolta, 1986). A positive effect of NPK fertilization (Viana et al., 1987; Raij et al., 1996; Sanzonowics et al., 2001)is found in several reports, however, other demonstrate little or no response to the application of these nutrients (Gallo et al., 1999; Garcia, 1999).

The use of stable isotopes in the form of ^{15}N labeled fertilizer is an excellent tool for such investigation (Reichardt & Bacchi, 2004). Several studies have been carried out on Citrus using the ^{15}N as a tracer to understand the plant demand on N during different phenologic stages, as well as its distribution within the plant (Wallace et al., 1954; Legaz et al., 1982; 1995; Feigenbaum et al., 1987; Lea-Cox et al., 2001). In Brazil, in relation to perenial crops, of special interest are the reports of Boaretto et al. (1999a; 1999b) and Fenilli et al. (2004). For the coffee crop there is little reference to ^{15}N studies (Bustamante et al., 1997; Snoeck & Domenach, 1999; Snoeck et al., 1998). Recently, Lima Filho & Malavolta (2003) studied the remobilization of N e K in normal and deficient coffee plants.

Using the ^{15}N technology, the present study intends to clarify several points related to the behavior of the nitrogen in coffee crops and the improvement of fertilizer management practices.

MATERIAL AND METHODS

Field studies were conducted from 2003 to 2005 in Piracicaba, SP, Brazil (22°42' S, 47°38' W, 580 m) on a Typic Rhodudalfs (USDA, 1975), named Nitossolo Vermelho Eutroférrico, according to Brazilian classification system (Embrapa, 2006). The area presents an average slope of 9.2 ± 0.3% and average initial soil test levels performed during August 2003 for the 0-0.2 m surface layer were: 5.3 pH in water, 21.2 g kg^{-1} organic matter, 5.5 g kg^{-1} available P, 2.9, 19.9, 13.7, 20.5 mmol$_c$ kg^{-1} exchangeable K, Ca, Mg and (H + Al), respectively (Raij et al., 2001), 64% base saturation, 255; 309 and 436 g kg^{-1} of sand, silt and clay, respectively, and 1460 kg m^{-3} bulk density.

The climate is of the type "tropical highland" (Cwa) according to Köppen´s classification (Köppen, 1931), very adequate for coffee cultivation under plain sunshine conditions. Long term annual averages of air temperature, rainfall and relative humidity are: 21.1° C; 1,257 mm, and 74%, respectively. The rainy season extends from October to March and the dry season from July to September. More details on water balance components that prevailed during the experimental period can be found elsewhere (Silva et al., 2006)

Coffee seedlings of the variety "Catuaí vermelho IAC-44" were planted in rows along contour lines in May 2001 with a row spacing of 1.75 m and 0.75 m between plants, with a population of 7,620 plants per ha. Coffee is a perennial crop which starts producing beans in the third year. In the region in question, the coffee is grown mainly under full sunshine, and each crop cycle begins with flowering at the end of the cold and dry season, as a consequence of the first significant rain, which occurs in the Piracicaba region during August-September. Fruit setting, grain filling and maturation take 9-10 months so that harvest is made between May and June. Therefore our fertilizer trial started on September 1st, 2003 when plants were 1.2 m tall, and continued during two years, until August 30, 2005.

Within an area of 0.2 ha of the established coffee crop five plots (replicates) of about 120 plants each (Fenilli, 2006) were randomly selected to receive the fertilizer N rates: 280 kg ha^{-1} of N in 2003/2004 and 350 kg ha^{-1} of N in 2004/2005, applied each year as ammonium sulfate, split into four applications: September 1st and 60, 105 and 150 days after. These rates were chosen based on soil fertility analysis, the age of the plants and their expected productivity (Raij et al., 1996). Within each of these plots, sub-plots of 3.94 m^2 with three plants of one row were depicked for the N labeled fertilizer study. The ammonium sulfate was enriched at 2.072 ± 0.001 atom percent for both year's applications. The central one of the labeled plants was used for N total and N abundance evaluations, collecting only one full branch (out of more than 50 branches at the beginning) at each sampling time, taken as representative for these concentration intensive variable measurements. This minimized sampling interference on the growth and development of the labeled plant, which could not be sacrified. Samples were oven dried at 65°C and finely ground. Sub-samples were used for total N and N abundance evaluations by mass spectrometry in an automated continuous flow Mass Spectrometer, Model ANCA-SL (Europa Scientific) as described by Mulvaney (1993) and Barrie & Prosser (1996).

For total shoot dry matter DM (extensive variable) one whole plant per replicate was harvested (sacrificed) outside each isotope sub-plot, chosen as being as similar as possible in shape and size in relation to the isotopic plant. They were dissected into parts as follows: central stem or orthotropic branch (OB); productive plagiotropic branches (PB); leaves of productive branches (LPB); vegetative plagiotropic branches (VB); leaves of vegetative branches (LVB); and fruits (FR) (beans). The growth and development of these parts were followed every 60 days during the whole experimental period (Pereira da Costa, 2006). This partitioning of the plant was made due to the differences in total-N content (C_N) and ^{15}N abundance (A_N) of plant parts. The beginning of the evaluations started at 8:00 am of September 1, 2003 and dates after this are numbered as days after beginning (DAB), so that each experimental day starts at 8:00 am and ends the day after at 8:00 am.

In order to follow continuously the total DM of the plants as a function of time (DAB), data were adjusted to the four parameter sigmoidal model represented by the equation:

$$DM = a + \frac{b}{1 + e^{-\left(\frac{t-c}{d}\right)}} \quad (1)$$

Dry matter (DM), nitrogen concentration in dry matter (C_N), and isotopic abundance (A_N) data of the different parts and of the whole shoot allowed the calculation at any DAB of:

1) total accumulated nitrogen (NA) (g per plant or kg ha^{-1}):

$$NA = DM \times \frac{C_N}{100} \quad (2)$$

2) the N derived from the applied fertilizer (Ndff) (%):

$$Ndff = \left(\frac{A_N - A_{N_c}}{A_{N_f} - A_{N_c}}\right) \times 100 \quad (3)$$

where A_{N_c} is the natural isotopic ^{15}N abundance of the plant (0.367) and A_{N_f} the enriched abundance of the fertilizer;

3) the quantity of N derived from fertilizer (QNdff) (g per plant or kg ha^{-1}):

$$QNdff = \left(\frac{Ndff}{100}\right) \times NA \quad (4)$$

4) the fertilizer recovery by the plant (R):

$$R = \left(\frac{QNdff}{Q_{N_f}}\right) \times 100 \quad (5)$$

where Q_{Nf} is the amount of applied fertilizer.

Data were analysed using descriptive statistical concepts: averages of five replicates, with respective standard errors in brackets.

RESULTS AND DISCUSSION

Dry matter of the total shoot accumulation as a function of time (Table 1), shows that during the first year, starting from the beginning of flowering (0 DAB) up to the first harvest (243 DAB), dry matter increased continuously. After harvest a decrease in DM was observed due to fruit export and fallen leafs at harvest and cold winter (366 DAB). Some leaf fall also occurred before harvest due to carbohydrate redistribution from them to fruits. The absorbed N during fall

Table 1 - Averages and standard error of total dry matter (DM); accumulated N (NA); quantity of N derived from the fertilizer (QNdff); and fertilizer recovery (R) of the aerial part of the coffee plant, as a function of time (Days After Beginning, DAB), for both agricultural years.

DAB	DM	NA	QNdff	R
	----------------------------- g per plant -----------------------------			%
63	690 (Ø77.40)	17.87 (Ø1.59)	4.06 (Ø0.46)	44.1 (Ø5.05)
126	1,105 (Ø116.33)	30.20 (Ø2.66)	13.00 (Ø0.88)	47.2 (Ø3.17)
182	1,659 (Ø140.32)	49.66 (Ø3.07)	26.20 (Ø1.08)	71.3 (Ø2.92)
243	2,222 (Ø180.79)	58.34 (Ø5.95)	15.77 (Ø2.78)	42.9 (Ø7.57)
366	1,392 (Ø100.91)	20.76 (Ø2.20)	7.60 (Ø1.23)	20.7 (Ø3.34)
430	1,767 (Ø122.68)	33.55 (Ø2.37)	14.82 (Ø1.69)	31.7 (Ø3.51)
491	2,162 (Ø115.71)	42.22 (Ø3.08)	20.27 (Ø2.42)	28.5 (Ø3.40)
548	3,014 (Ø251.76)	55.82 (Ø4.65)	29.82 (Ø3.69)	36.1 (Ø4.47)
636	3,944 (Ø272.60)	77.31 (Ø5.29)	38.48 (Ø3.61)	46.54 (Ø4.37)

and winter stays accummulated in the plant until spring when it is redistributed, being partially used for flowering and fruit development (Malavolta, 1993). When the vegetative growth is reassumed, the N content of the leaves tends to decrease, presumably due to its transfer to branches, new leaves, flower buds and fruits.

The total shoot DM accumulation from 0 to 243 DAB presented a sigmoidal behavior like annual crops, characterized by greater growth rates in the period of leaf production and fruit formation, with a slowing down towards fruit maturation and harvest. This fact can better be appraised by the excellent adjustment of Table 1 DM data to equation (1), shown in Figure 1 (A). During the same growth period of the second year (flowering to harvest, 366 to 548 DAB), the crop presented the same behavior, and Figure 1 (B) shows how well equation (1) also fitted to the experimental data of the aerial part of the coffee plant. The fittings were made through the Table Curve program, with the four parameters shown in Table (2), with R^2 coefficients of 0.9999 and 0.9954 for both years, respectively, indicating a practically perfect adjustment of the experimental data to the model. The fact that the same model (eq. 1) fitted to data of both agricultural years lead to a generalization in which both years data were jointly used to fit eq. 1 with relative values $Y = [(DM - DM_f) / (DM_{max} - DM_f)]$, where DM_f

is the dry matter at flowering (0 DAB) and DM_{max} at harvest; and relative time $X = t/t_{max}$, where t_{max} is the time at harvest (DAB). A very good adjustment ($R^2 = 0.9895$) was also here obtained, with parameters presented in Table 2.

Equation 1 also permitted the estimation of the coffee growth rates through their derivatives d(DM)/dt, also presented in Figure 1. During the first year the growth rate was positive from 0 to 172 DAB, when a maximum of 10.5 g per plant day^{-1} was reached. This corresponds to 2.39 kg ha^{-1} day^{-1} of total-N and 1.20 kg ha^{-1} day^{-1} of fertilizer-N. Under average conditions, with a constant absorption rate of 0.72 kg ha^{-1} day^{-1} of fertilizer-N, measured in this experiment, the crop would take up 194 kg ha^{-1} of fertilizer N during 9 months, which is the main growth period of the crop. This shows that the 280 kg ha^{-1} of N applied the first year were more than average plant uptake.

Despite that this soil contains about 3,082 kg ha^{-1} of N in the 0-1.0 m layer, which represents 96% in relation to the applied fertilizer N, the fertilizer-N representing only 4% of this N, is readily available reaching a recovery of 71.3% at 182 DAB, not taking into account root fertilizer N. This stresses the importance of the N fertilization for coffee. An analysis of the growth rate evolution in Figure 1A indicates that the four fertilizer splittings (0, 63, 105 e 151 DAB) were applied during the phase of increasing growth rates,
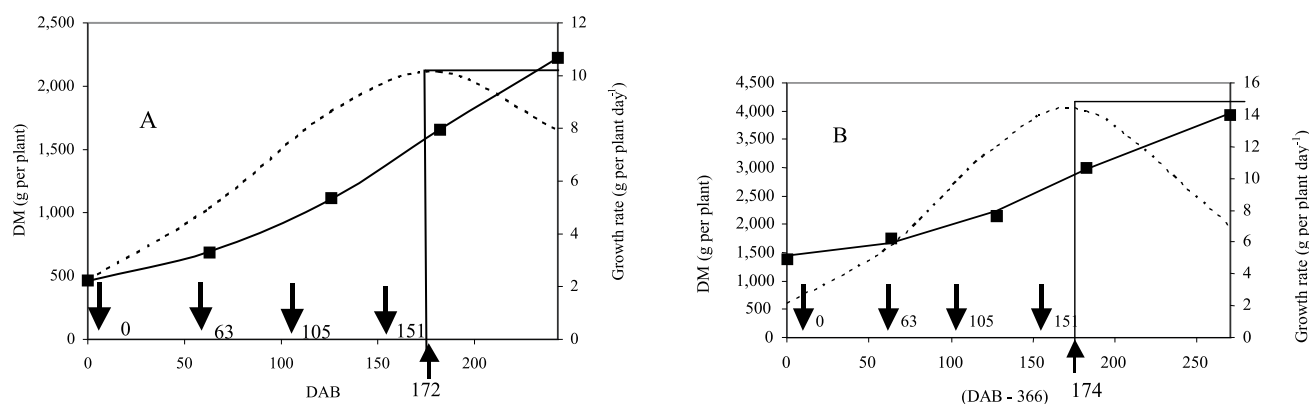

Figure 1 - Dry matter (DM) accumulation during the first year (A) and second year (B). Points are observed data; solid line follows eq. 1; and dotted line is the DM growth rate (dDM/dt). Arrows indicate dates of fertilizer splittings and date of maximum growth rate. DAB= Days after beginning. In (B), (DAB-366) was used only in order to ajust model of eq. 1.

Table 2 - Parameters for equation (1) for coffee dry matter accumulation as a function of time (DAB = days after beginning) for both years and for non-dimensional data.

Parameter	1st year (0 - 243 DAB)	2nd year (366 - 636 DAB)	1st and 2nd years (non-dimensional)
a	313.2	1326.4	- 0.0597
b	2604.5	3062.5	1.3454
c	178.0	173.8	0.6872
d	64.1	53.7	0.2385

before the maximum was reached. The choice of four splittings spaced 60, 105 and 150 days after flowering was coherent with plant needs. The most indicated phase for fertilizer application to coffee (Silva et al., 1984 and Matiello et al., 1983) should be after bloom, but before fruit filling, from October to February splitting it two to three times, since applications before blooming in September or in the grain filling stage (end of March) are less effective on productivity gain, probably due to N losses in September (low rainfall period) and March, a late period from the plant need point of view. After the last application was made (151 DAB) the growth rate started decreasing, however maintaining relatively high values up to harvest time.

The growth rate of the second year increased from 366 DAB (corresponding to 0 DAB in Figure 1B) to 540 DAB (174 DAB in Figure 1B), when it reached a maximum of 14.25 g per plant day^{-1} or 2.01 kg ha^{-1} day^{-1} of total-N and 1.05 kg ha^{-1} day^{-1} of fertilizer-N, thereafter decreasing until harvest 636 DAB. Again, four application times fell inside the period of increasing growth rates.

In relation to QNdff present in the plant (Table 1), at 63 DAB the aerial part absorbed 4.06 of the 9.19 g per plant applied at 0 DAB (1st fertilizer application), which corresponds to 44.1%. At 126 DAB the crop had already received three applications of fertilizer, that is, 27.57 g per plant, and had absorbed 13.00 g per plant or 47.2%. At 182 DAB when QNdff passed through a maximum, the four applications had already been made and the aerial part absorbed 26.2 g per plant, that is, 71.3% which is a very high proportion of the applied amount. If the QNdff of roots (5.2 g pl^{-1}) is added to this value, which is presented in more detail elsewhere (Fenilli, 2006), the plant recovery efficiency reaches 85.5%. The QNdff balance presented in Fenilli (2006) justifies this high fertilizer-N recovery showing that the other fates of the fertilizer-N, including losses, were very small. However, just before harvest (243 DAB) the QNdff was reduced to 15.77 g per plant. This reduction can be explained by translocation of N from shoot to root, losses of plant parts like leaves and fruits and N losses from the plant to the atmosphere in the form of ammonia as suggested by Trivelin et al. (2002) and Holtan-Hartwig & Bockman (1994). The remobilization of certain nutrients is an important metabolic feature during plant development, in cases of seed germination, during stress conditions within the period of vegetative growth, and during the reproductive stage as well and, in the case of perennials, before leaf fall (Marschner, 1995).

At 366 DAB the plant had recovered 7.60 g per plant from the applied 36.76 g during the first year, corresponding only to 20.7% (Table 1). This result masks the real value of 71.3% obtained for the previous sampling (243 DAB) and shows the importance of sampling times in studies of fertilizer-N accumulation in perenial crops. Samplings should be performed as many times as possible, in order not to miss important intermediate information. Other compartments of the whole soil-plant-atmosphere system, reported elsewhere (Fenilli, 2006), give a better overview of the pool sizes and processes involved in the fate of the fertilizer-N. In other situations, reports show lower recoveries, e.g. Bustamante et al. (1997) who found that coffee absorbed 35 to 46% of the fertilizer-N depending on the kind of fertilizer used as a source of N. It is important to mention that these authors considered only the aerial part of the plant, which in our case would be 42.9% in the first year.

At 430 DAB the first application of the second year had already been made (36.76 + 11.48 g per plant of fertilizer-N), and the plant presented a recovery of 31.70% of fertilizer-N. At 491 DAB, with three fertilizer applications of the second year, plant absorption was 20.27 g per plant or 28.5%. At 548 DAB, when all fertilizer splittings had already been performed, the shoot had absorbed 29.82 g per plant or 36.1%, and at harvest these numbers changed to 38.48 g per plant and 46.5%, respectively. These fertilizer-N recoveries evaluated during the second year, follow the same trend of those of the first year and the same comments made for the first year are valid for the second year.

A more detailed analysis of the QNdff is in Table 3, which shows the contribution of each sampled part to the total aerial plant. From 182 to 243 DAB all parts lost fertilizer-N and the relative losses were: OB = 39%; PB = 49%; LPB = 67%; VB = 11%; LVB = 23% and FR = 49%. These losses represent N translocations among plant parts and mainly to the root system as a preparation for the next production cycle. Another part of these losses is in fallen leaves and fruits even before harvest, which build up a litter layer below canopy. Since most plant parts gained in total-N (Table 1) while loosing fertilizer-N (Table 3) it is suggested to assume that the "new N", that of the fertilizer applied during the year under consideration, is more mobile than the "old N", that already takes part of plant constitution. Among the several reserve organs of the plant, leaves and branches accumulated the largest amounts of N, but among the Young organs, fruits and branches exported most of the N from the reserves (Lima Filho & Malavolta, 2003). This "new N" dynamics could only be observed due to the frequent samplings (every 60 days), which in our case showed that at 182 DAB the aerial part contained already 71.3% of the fertilizer-N, and that this number

Table 3 - Averages and standard error of quantity of fertilizer-N (QNdff) as a function of time (DAB = days after beginning) for the different aerial parts of the coffee plant.

Agricult.year	DAB	QNdff					
		OB	PB	LPB	VB	LVB	FR
		-- g per plant --					
1st	63	0.49 (Ø0.06)	0.15 (Ø0.03)	0.51 (Ø0.18)	0.33 (Ø0.09)	2.55 (Ø0.24)	0.03 (Ø0.01)
	126	1.67 (Ø0.12)	0.64 (Ø0.07)	2.05 (Ø0.47)	0.71 (Ø0.09)	7.41 (Ø0.60)	0.52 (Ø0.28)
	182	2.68 (Ø0.17)	1.90 (Ø0.09)	6.88 (Ø0.68)	1.28 (Ø0.05)	10.96 (Ø0.87)	2.50 (Ø0.71)
	243	1.64 (Ø0.54)	0.97 (Ø0.27)	2.29 (Ø0.48)	1.14 (Ø0.55)	8.46 (Ø3.24)	1.27 (Ø0.81)
2nd	366	2.34 (Ø0.34)	1.19 (Ø0.26)	0.45 (Ø0.14)	0.73 (Ø0.10)	2.46 (Ø0.42)	0.43 (Ø0.10)
	430	3.43 (Ø0.31)	2.79 (Ø0.44)	1.16 (Ø0.29)	0.71 (Ø0.08)	5.32 (Ø0.53)	1.41 (Ø0.31)
	491	3.55 (Ø0.52)	1.89 (Ø0.26)	0.46 (Ø0.06)	0.51 (Ø0.11)	6.29 (Ø0.56)	7.56 (Ø1.05)
	548	4.16 (Ø0.78)	1.94 (Ø0.32)	0.38 (Ø0.08)	1.19 (Ø0.16)	12.35 (Ø1.96)	9.81 (Ø1.50)
	636	4.99 (Ø0.66)	2.37 (Ø0.25)	0.25 (Ø0.06)	1.21 (Ø0.20)	10.44 (Ø1.15)	19.22 (Ø3.53)

OB - orthotropic branch; PB - productive plagiotropic branches; LPB - leaves of productive branches; VB - vegetative plagiotropic branches; LVB - leaves of vegetative branches; FR - fruits. DAB = days after beginning

reached 85.5% including roots. This very high fertilizer-N recovery could even suggest the application of more than 280 kg ha^{-1} of N.

At 366 DAB the 1st application of the fertilizer of the second year was performed, using the same fertilizer with the same enrichment and, therefore, calculations of N recovery were made in a cumulative way, including the amount applied the first year. During the second year (Table 1) both total-N and fertilizer-N increased continuously. Detailed data on QNdff (Table 3) show that from 366 to 636 DAB, exception made to LPB all parts presented increases in fertilizer-N. Leaves from vegetative branches and fruits presented the highest N concentration, 27.1% and 49.9%, respectively, of the total aerial part. Since the second year was one of high productivity, translocation of fertilizer-N from all parts to fruits was expressive, explaining the bianuality of the yield of coffee crops. The high fertilizer-N in leaves from vegetative branches indicates the importance of fertilization in the preparation of the crop for next year's production, since the next flowers will bear on these branches.

CONCLUSION

The growth of the coffee plant in terms of aboveground dry matter accumulation was sigmoidal for two consecutive years, between flowering (August/September) and harvest (May/June) and could be adjusted to a four parameter sigmoidal equation using non-dimensional variables.

During the first agricultural year the aerial part of the coffee plant recovered 71% of the applied fertilizer, and this value was reduced to 34% at harvest,

and to 19% at the beginning of the new flowering period. During the second year, considering the fertilizer applied since the first year, the aerial part recovered 36% of fertilizer-N up to March, 47 at harvest and 19% at the beginning of the next flowering period.

ACKNOWLEGMENTS

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