



Efficiency of absorption, translocation, and use of nitrogen by water-stressed coffee

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ABSTRACT. Climate change and the expansion of agricultural lands to low-fertility soils have prompted studies on the effects of water and nutritional stress on nutrient uptake and use. This study aimed to evaluate the efficiency of absorption, translocation, and use of N in coffee cultivars under water stress (WS) and different N doses. The experiment was performed hydroponically and arranged in a 4 × 2 × 4 factorial randomized block design with three replications. These factors consisted of four coffee varieties, two N doses, and four levels of WS. At the end of the experiment, the dry matter production, N concentration, and N accumulation of plants, as well as nutritional efficiency indices were evaluated. The data were subjected to analysis of variance and regression. The results showed that in plants previously well-supplied with N, N stress induces greater allocation of dry matter in roots, mitigating the effects of subsequent water deficits on shoot dry matter production. Young Mundo Novo plants subjected to N stress followed by increasing water stress maintained a high utilization efficiency (UE). Young Acauã plants well-supplied with N, maintained their N utilization efficiency up to -1.6 MPa of water stress; however, when they were N starved, they underwent a drastic reduction in UE at potentials below -0.22 MPa.

Keywords: *Coffea arabica* L.; N accumulation; shoot/root ratio.

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Introduction

Extensive areas of coffee cultivation lack irrigation systems; therefore, water availability in these regions is subject to climatic variations that can negatively affect plant growth, either through high temperatures or low rainfall. The effects of changes in rainfall patterns and the occurrence of droughts, which have increased in recent decades in a pronounced and unpredictable manner (IPCC, 2021), have caused uncertainties in the coffee sector. Regionally, rainfall periods have changed, affecting frequency and intensity, with negative impacts on the water balance both by excess and deficit, increasing droughts that lead to water deficits in plants. In addition, agricultural lands are expanding to low-fertility soils, which implies greater use of N fertilizers. N availability and water deficit are limiting factors that affect agricultural production, as they affect processes related to plant growth and metabolism.

The effects of water deficit on plants in general, as well as on coffee trees, have been widely reported in the literature in terms of physiological, morphological, biochemical, and molecular aspects (Da Matta & Cochicho Ramalho, 2006; Taiz, Zeiger, Moller, & Murph, 2017; Bonomo, Oliveira, Silveira Neto, & Bonomo, 2008; Sakai, Barbosa, Silveira, & Pires, 2015). These effects influence crop growth and yield, water and nutritional relationships, photosynthesis, assimilation partitioning, respiration, and oxidative damage. Although plant responses to drought are relatively well known, information on performance in complex environments where multiple stresses arise simultaneously is limited.

In adult coffee plants, reductions in height, trunk and crown diameter, and the number of flowers and fruits are well-documented responses to water stress (Silva, Cavatte, Morais, Medina, & DaMatta, 2013). Water stress can directly and indirectly affect N absorption, which consequently affects crop growth and yield. This is associated with a reduction in the net carbon assimilation rate owing to stomatal and non-stomatal limitations, which reduce the intercellular CO₂ concentration. In addition, water scarcity can lead to nutrient deficiency because water carries nutrients from the soil to roots (DaMatta, Loos, Silva, Loureiro, & Ducatti, 2002). Martinez, Souza, Caixeta, Carvalho, and Clemente (2020) evaluated eight coffee cultivars under short-

term water deficit induced by polyethylene glycol (PEG) and found that Catuaí Amarelo, Catuaí Vermelho, and Acauã significantly increased the nitrate concentration in the roots, whereas Mundo Novo showed no significant difference in relation to the treatment without water stress.

In the absence of limiting factors, an adequate N supply promotes rapid coffee growth by increasing the number of leaves, pairs of plagiotropic branches, number of nodes per branch, and number of flowers and fruits per node, which are related to higher coffee yields (Carelli, Fahl, & Cochicho Ramalho, 2006). However, excessive N supply can severely reduce root growth (Salih, Agren, & Hallbacken, 2005). Low concentrations of nutrients in the soil induce root growth, while high concentrations of nutrients in the soil cause greater biomass allocation to shoots (Wu, Bao, Li, & Wu, 2008).

In recent decades, N has been studied as a factor that can mitigate the negative effects of water deficit on plant performance, focusing on the effects of high irradiance on photosynthesis and, more recently, on water relations, osmotic adjustment, and antioxidant enzyme activity. DaMatta et al. (2002) evaluated N levels and irrigation regimes and found that N increased cell wall rigidity and osmotic adjustment. These responses are associated with improved water extraction and reduced cell volume loss, providing some degree of drought tolerance. This study indicated that under high N conditions, *Coffea canephora* improved its drought tolerance by increasing its water absorption capacity rather than by reducing water loss via stomatal control. In contrast, the growth of young *Sophora davidii* was co-limited by water stress and N status, with a positive response to low N, leading to the conclusion that high N should be avoided in the seedling phase (Wu et al., 2008). The regulatory role of N in water stress tolerance in plants depends on the intensity of the N deficit and nutritional status. According to Zhong et al. (2017), adequate N levels support the regular growth of plants and help them resist water stress.

The study of nutritional efficiency under nutritional and water limitations can provide information for the selection of cultivars that are efficient in the use of important agricultural resources, such as N fertilizers and water, thereby improving their utilization efficiency.

Few studies have addressed the concomitant effect of water deficit and N fertilization in coffee, which prompted the investigation of the performance of these combined effects as a strategy for selecting cultivars with the potential to adapt to adverse conditions. Therefore, the aim of this study was to evaluate the N accumulation and biomass production of four Arabica coffee cultivars grown in nutrient solutions, and their nutritional efficiency indices in environments with contrasting N availability and water deficit.

Material and methods

Plant material and growing conditions

The experiment was conducted in a greenhouse at the Department of Agronomy of the Federal University of Viçosa (20°45' S, 42°15' W, 650 m altitude), Minas Gerais State, Brazil. Six-month-old coffee plants (*C. arabica* L.) of the cultivars Catuaí Amarelo 62 (CA), Catuaí Vermelho IAC44 (CV), Acauã (AC), and Mundo Novo IAC 379-19 (MN) were cultivated in a complete nutrient solution. Each experimental unit consisted of one plant and received 6.5 L of nutrient solution at concentrations that defined the N levels as high (HN: 7 mmol L⁻¹ NO₃⁻) and low (LN: 2.8 mmol L⁻¹ NO₃⁻). The concentrations of the other nutrients remained constant as follows: 1 mmol L⁻¹ P, 4.8 mmol L⁻¹ K, 1 mmol L⁻¹ Mg, 1 mmol L⁻¹ S, 2.1 mmol L⁻¹ Ca, 40 μmol Fe-EDTA, 23 μmol L⁻¹ B, 0.8 μmol L⁻¹ Cu, 12 μmol L⁻¹ Mn, 0.3 μmol L⁻¹ Mo, and 1 μmol L⁻¹ Zn. The plants were initially subjected to NO₃⁻ doses for 67 days and then to different degrees of water stress for 76 days; the experimental period lasted 143 days in total.

Water stress of 0.0, -0.4, -0.8, and -1.6 MPa was induced by 0, 172.6, 240.2, and 335.8 g L⁻¹ of PEG 6000, respectively (Villela, Doni Filho, & Sequeira, 1991). The stress was gradually induced by applying the sixth part of the PEG amount on consecutive days.

The containers with nutrient solutions were aerated during the entire experimental period. The water lost by evapotranspiration was replaced daily to maintain a constant solution volume, and the pH was maintained at 6 ± 0.2. Electrical conductivity (EC) was used as an indicator of the nutrient concentration. Thirty percent of the initial nutrient doses were replaced whenever the EC was reduced by 30%, except for N. The nitrate concentration of the solution was measured using a LAQUA Twin Nitrate Meter and 30% of the initial dose was added whenever a corresponding reduction was detected.

Nitrogen concentration in plants and nutritional efficiency indices

At the end of the experiment, the plants were collected and separated into leaves, stems (including branches), and roots. The material was washed with deionized water, placed in paper bags, and dried in a forced-air circulation oven at 70°C to a constant weight. The dry matter mass (DM) was determined for each plant organ and the total plant (TDM). The N concentration was determined by the micro Kjeldahl method in dried samples ground in a Wiley mill, sieved through a 0.841 mm mesh, and digested with sulfuric acid (Malavolta, Vitti, & Oliveira, 1997). The total N accumulation was calculated as the sum of N in the different organs, which was calculated by multiplying the nutrient concentration (g kg^{-1}) by the dry matter (kg) of each plant organ. The shoot/root ratio (S/R) was calculated by dividing the shoot dry matter production (SDM) by the root dry matter production (RDM).

The nutritional efficiency indices were calculated from the following expressions:

Absorption efficiency (AE): total N accumulated/root dry mass [g g^{-1}] (Swiader, Chyan, & Freiji, 1994).

Translocation efficiency (TE): (N accumulated in shoot/total N accumulated) \times 100 [%] (Li, McKeand, & Allen, 1991).

Use efficiency (UE): (total dry mass produced)²/total N accumulated [$\text{g}^2 \text{g}^{-1}$] (Siddiqi & Glass, 1981).

Experimental design and statistical analysis

The experimental units received a combination of three factors: two N doses (high and low), four degrees of water deficit (0, -0.4, -0.8, and -1.6 MPa), and four coffee cultivars in a factorial randomized block design with three replications.

The data were subjected to analysis of variance and regression. The means of the qualitative factors were compared using Tukey's test at 5% probability. Regression equations were fitted to the quantitative factors. The models were chosen based on the significance of the regression coefficients, coefficient of determination, and behavior of the phenomenon.

Results and discussion

This experiment simulated the conditions of seedlings newly planted in the field in rainfed orchards. In general, the seedlings are supplied with adequate nutrition in the nursery and transplanted to the field in the rainy summer (October to March). After settled they receive varying doses of top-dressing N until the end of the rainy season, and then underwent long-term water stress, coinciding with the dry winter.

Dry matter production, shoot/root ratio, and N concentration and accumulation

In general, increasing water stress caused reductions in SDM and RDM production and N accumulation in coffee plants grown under high N doses (Figure 1A, C, and E). In cultivars Mundo Novo and Catuaí Amarelo the reductions followed quadratic functions; in cultivar Catuaí Vermelho the reductions followed linear functions. However, there was no model adjustment for Acauã, suggesting the stability of these variables in water-stressed conditions.

In plants under long-term N stress (low N dose), water stress caused a linear reduction in SDM production only in Catuaí Vermelho, but for the other cultivars, it remained stable (Figure 1B). In contrast, the RDM of cultivars Catuaí Amarelo and Catuaí Vermelho decreased with an increase in water stress in a quadratic manner, while Acauã decreased linearly, and Mundo Novo showed no variation (Figure 1D). The accumulation of N in plants subjected to N restriction also showed no variation with increases in water deficit ($\hat{y} = \bar{y}$ between 0.77 and 0.87 g - Figure 1F).

The analysis of the response to N showed that under each water stress degree, SDM (Table 1) and TDM (data not shown) presented no significant response to N doses, whereas RDM and the S/R ratio were significantly affected by this factor. The average RDM production in plants grown under LN conditions was higher in the four cultivars. When low N levels were combined with increasing water stress, the differences in RDM production between the two N doses were greater.

The combined effect of the two stresses was observed in Acauã. In HN, Acauã produced 9.7, 12.5, and 11.6 g of RDM under stress degrees -0.4, -0.8, and -1.6 MPa, respectively. This response was more related to water absorption, as N would not be a limiting factor in this case. When subjected to LN, Acauã produced 23.1, 17.4,

and 14.1 g of RDM under stress degrees -0.4, -0.8, and -1.6 MPa, respectively, indicating that the increase in RDM was magnified by long-term N stress (Table 1). Pinheiro, DaMata, Chaves, and Ducatti (2005) reported that larger root systems and greater mass are related to better performance of *C. canephora* genotypes under drought conditions.

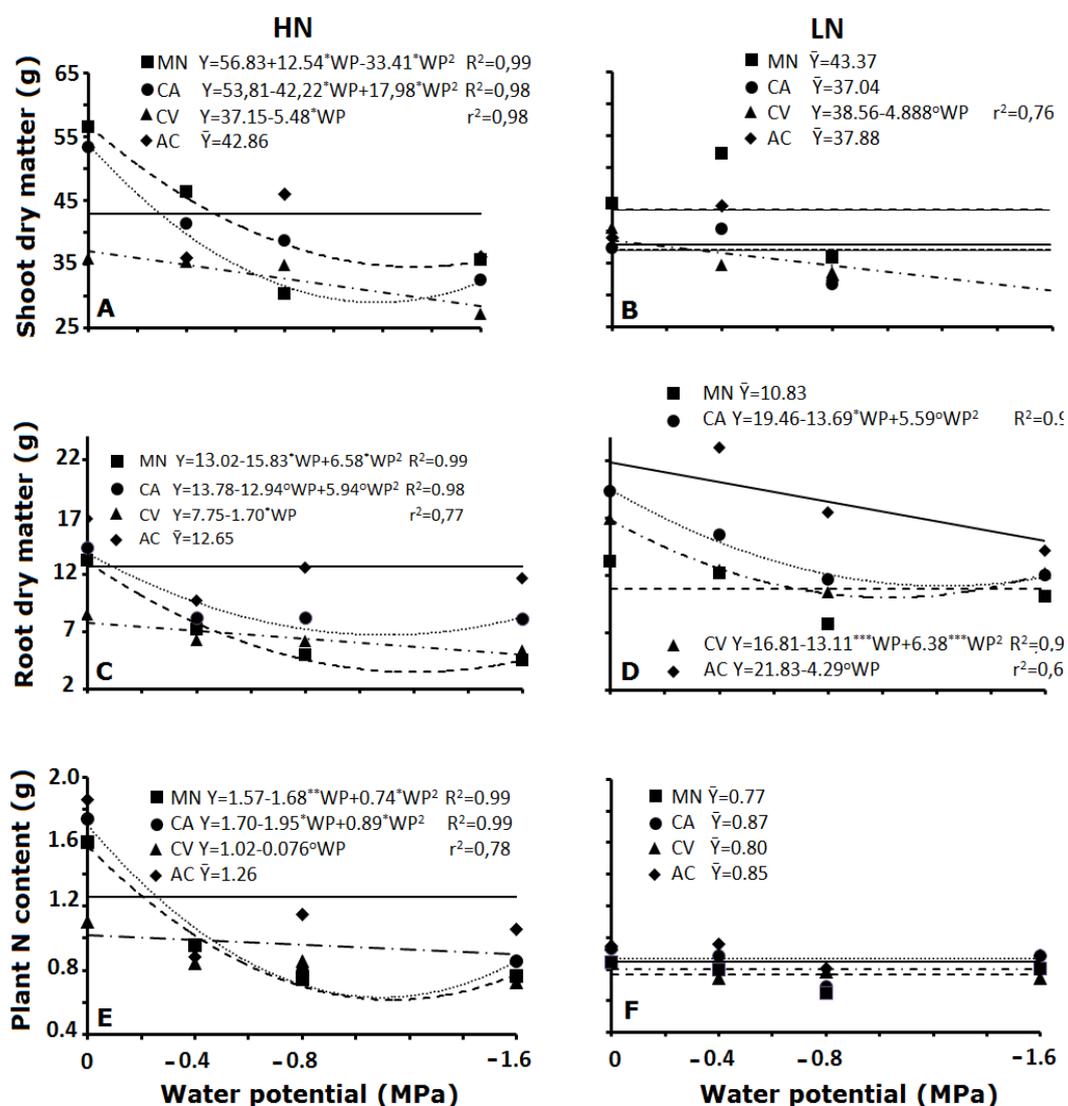


Figure 1. Shoot dry matter (A and B), root dry matter (C and D), and N accumulation (E and F) by four coffee cultivars subjected to high (HN) and low (LN) doses of nitrogen and subjected to increasing water stress. ***, **, * and $^{\circ}$ – Regression coefficients significant at 1%, 5%, 10%, and more than 10% probability, respectively.

Several authors have reported the magnitude of the effect of N deficiency on the vegetative and reproductive growth of coffee (Bruno et al., 2011; Clemente, Martinez, Alves, & Lara, 2013; Valadares et al., 2013; Colodetti, Rodrigues, Martins, & Tomaz, 2014). However, in the present study, a comparison of the dry matter production of the cultivars showed there was no reduction in the SDM production of young plants previously well supplied with N under any degree of water stress, even after 4.5 months of N deprivation (Table 1). The TDM (data not shown) followed the same response pattern as the SDM. In HN, N accumulation followed the pattern of dry matter production, that is, it decreased or remained stable with increasing water stress (Figure 1E).

In LN, the accumulation remained stable with increasing water stress (Figure 1F). At each degree of water stress, the SDM production at low N was maintained at the expense of the reduction of the N concentrations in the tissues, notably the roots (Table 2). These results suggest that coffee seedlings under good nutritional conditions can withstand post-planting N stress. The dilution of the previously absorbed and accumulated N reduces its concentration, especially in the roots, thereby preserving the shoot growth.

Table 1. Shoot and root dry matter, and shoot/root ratio (S/R) of four coffee cultivars grown at two nitrogen doses and four degrees of water deficit (Ψ_w - MPa).

Shoot dry matter (g)									
Cultivar	Ψ_w 0		Ψ_w -0.4		Ψ_w -0.8		Ψ_w -1.6		CV%
	HN	LN	HN	LN	HN	LN	HN	LN	
CV	35.8 ns	40.6 ns	35.3 ns	34.6 ns	34.9 ns	33.6 ns	27.2 ns	31.8 ns	
A	53.3 ns	39.0 ns	36.0 ns	44.0 ns	45.9 ns	32.4 ns	36.2 ns	36.1 ns	
CA	53.2 ns	37.3 ns	41.3 ns	40.3 ns	30.4 ns	31.7 ns	32.5 ns	38.8 ns	
MN	56.5 ns	44.3 ns	46.4 ns	52.2 ns	37.5 ns	35.8 ns	35.6 ns	41.2 ns	
Mean	49.7	40.3	39.7	42.8	37.2	33.4	32.9	37.0	
CV%	26.2								
Root dry matter (g)									
Cultivar	Ψ_w 0		Ψ_w -0.4		Ψ_w -0.8		Ψ_w -1.6		CV (%)
	HN	LN	HN	LN	HN	LN	HN	LN	
CV	8.4 Bb	16.8 Aa	6.8 Aa	12.5 Ba	6.2 Aa	10.5 ABa	5.3 Ab	12.1 Aa	
A	16.8 Aa	19.3 Aa	9.7 Ab	23.1 Aa	12.5 Aa	17.4 Aa	11.6 Aa	14.1 Aa	
CA	14.3 Ba	19.2 Aa	8.2 Ab	15.5 ABa	8.2 Aa	11.5 ABa	8.1 Aa	11.9 Aa	
MN	13.2 Ba	13.2 Aa	7.2 Aa	12.2 Ba	4.9 Aa	7.7 Ba	4.5 Aa	10.2 Aa	
Mean	13.2	17.2	7.9	15.8	8.0	11.8	7.4	12.1	
CV (%)	31.9								
Shoot/Root ($g\ g^{-1}$)									
Cultivar	Ψ_w 0		Ψ_w -0.4		Ψ_w -0.8		Ψ_w -1.6		CV (%)
	HN	LN	HN	LN	HN	LN	HN	LN	
CV	4.3 ns	3.1 ns	5.7 ABa	2.9 ABb	5.5 ABa	3.6 ABb	5.5 ABa	2.7 Ab	
A	3.2 ns	2.0 ns	3.8 Ba	1.9 Bb	3.7 Ba	2.0 Ba	3.1 Ca	2.7 Aa	
CA	3.7 ns	2.0 ns	5.1 ABa	2.6 ABb	4.2 Ba	2.7 ABa	4.1 BCa	3.5 Aa	
MN	4.7 ns	3.3 ns	6.8 Aa	4.4 Ab	7.7 Aa	4.8 Ab	7.3 Aa	4.4 Ab	
Mean	4.0	2.6	5.3	3.0	5.3	3.3	5.0	3.3	
CV (%)	56.0								

Within each water deficit, means followed by at least one different capital letter in the column and lower case letters in the row do not differ significantly at the 5% probability by Tukey's test. ns = non-significant.

The responses to N stress were not uniform across the cultivars. Catuaí Vermelho responded to N stress in the absence of water stress, with a significant increase in root dry matter production. This response was lost with water stresses of -0.4 and -0.8 MPa, but it was very intense at -1.6 MPa. At -0.4 MPa, Acauã and Catuaí Amarelo produced 2.3 and 1.9 times, respectively, more RDM in LN (Table 1). Increased root growth is common under nutritional limitation as a strategy to increase the absorptive surface area of the roots to meet the plant's demand. According to Föhse, Claassen, and Jungk (1988), roots can become preferential sinks for photoassimilates when nutrients, especially N, have reduced availability. Wu et al. (2008) studied the response of young plants of *Sophora davidii* to different N doses and irrigation at 80, 40, and 20% of the field capacity in a pot experiment. They verified a higher partition of the dry mass to the roots (higher root/shoot relationship) under irrigation of 40 and 20% of the field capacity and low N dose than at irrigation of 80% of the field capacity and high N dose.

Changes in the partitioning of dry matter produced under different N availabilities resulted in significant changes in the S/R ratio in the plants subjected to water stress, whereas plants without water restriction were not significantly affected by the N dose or the cultivar.

Overall, under water deficit conditions, the S/R ratio was higher in plants grown under high N doses. In Catuaí Vermelho and Mundo Novo, this pattern of response was evident at all degrees of induced water stress. In contrast, Acauã and Catuaí Amarelo showed this response pattern only at -0.4 MPa. At all degrees of water stress, Acauã presented a shoot/root ratio lower than that of the other cultivars, indicating a greater allocation of dry matter to roots. Without water stress, the S/R ratio of Acauã showed no difference from the other cultivars. Under all water stress conditions, the Mundo Novo cultivar had the highest shoot/root ratio, which did not differ from that of the Catuaí Vermelho cultivar (Table 1).

The relationship between shoot and root dry matter production (S/R) provides information regarding the relative investment of photoassimilates for growth in these compartments. A higher ratio reflects a lower investment in RDM production. Mundo Novo had significantly higher S/R values at stresses of -0.8 and -1.6 MPa. Föhse, Claassen, and Jungk (1988) stated that P-absorption-efficient plants have a high rate of nutrient absorption per unit of root mass (high influx) as well as a high shoot/root ratio. The latter condition was verified in this study. To investigate water stress, Covre, Parteli, Gontijo, and Zucoloto (2015) studied the

distribution of the root system of the cultivar Conillon (*C. canephora*) and found that non-irrigated plants had roots of greater length, volume, and surface area.

In the absence of limiting factors, adequate N promotes rapid plant growth by increasing the number of leaves and pairs of plagiotropic branches, number of nodes per branch, number of flowers, and fruits per node, which is associated with higher coffee production (Carelli et al., 2006). However, Wu et al. (2008) investigated the effects of water stress and N supply on seedlings of the tree species *S. davidii* and found that excessive N can severely reduce root growth. These authors argued that low concentrations of nutrients in the soil stimulate root growth, whereas high concentrations promote greater biomass partitioning to the shoot.

Changes in production and increases in dry matter partitioning to roots seem to be largely responsible for maintaining SDM production in plants that had undergone N stress when subjected to increasing water deficit (except for Catuaí Vermelho). In addition to the dilution of previously accumulated N, there was an increase in the production of root dry matter and a change in its partition, with a reduction in the S/R ratio in response to stress.

Leaf and root N concentrations were higher in plants cultivated under high N concentrations. At this dose, the cultivars did not differ from each other in terms of leaf N concentration under any degree of water stress. At low N, when the water stress was -0.4 MPa, Catuaí Amarelo had the highest leaf N concentration and Acauã had the lowest. Root N concentrations differed among the cultivars under high N and -0.8 and -1.6 MPa water stress; Catuaí Vermelho and Catuaí Amarelo had the highest and lowest concentration, respectively, at -0.8 MPa, while Acauã and Mundo Novo had the highest and lowest content, respectively, at -1.6 MPa (Table 2).

In plants that had not been subjected to water stress, the accumulation of N was higher when they were cultivated in HN, except for Catuaí Vermelho. However, when the plants were subjected to water stress, there was no difference between the N doses. This indicates that any degree of water stress imposed limitations on the N absorption process, since the N availability in HN was high and in a nutrient solution continuously stirred by compressed air injection, the effect of water stress on nutrient transport to the roots was not present (Figures 1E and F, Table 3). In fact, the maximum rate of nitrate absorption by these cultivars (except for Mundo Novo) was reduced at -1.5 MPa of water stress (Martinez et al., 2020).

Table 2. N concentration in leaves and roots and the total N accumulation of four coffee cultivars grown at two nitrogen doses and four degrees of water deficit (Ψ_w - MPa).

N concentration in leaves (g kg^{-1})									
Cultivar	$\Psi_w 0$		$\Psi_w -0.4$		$\Psi_w -0.8$		$\Psi_w -1.6$		LN
	HN	LN	HN	LN	HN	LN	HN	LN	
CV	30.5 Aa	22.0 Ab	24.8 Aa	21.7 ABb	25.4 Aa	20.7 Ab	27.8 Aa	22.3 Ab	
A	31.6 Aa	20.3 Ab	21.8 Aa	17.9 Bb	23.8 Aa	21.8 Aa	24.9 Aa	21.7 Ab	
CA	31.2 Aa	20.6 Ab	22.7 Aa	21.9 Aa	24.8 Aa	21.2 Ab	24.6 Aa	21.7 Ab	
MN	30.7 Aa	21.4 Ab	24.3 Aa	20.8 ABb	22.9 Aa	21.6 Aa	25.4 Aa	24.1 Aa	
Mean	31.0	21.1	23.4	20.6	24.2	21.3	25.7	22.4	
CV (%)	8.4								
N concentration in roots (g kg^{-1})									
Cultivar	$\Psi_w 0$		$\Psi_w -0.4$		$\Psi_w -0.8$		$\Psi_w -1.6$		LN
	HN	LN	HN	LN	HN	LN	HN	LN	
CV	29.5 Aa	19.5 Ab	21.9 Aa	16.6 Ab	24.1 Aa	20.0 Ab	21.8 ABa	15.6 Ab	
A	28.6 Aa	16.8 Ab	21.0 Aa	17.6 Ab	21.7 ABa	16.3 Ab	26.1 Aa	17.7 Ab	
CA	28.6 Aa	17.9 Ab	20.1 Aa	15.0 Ab	19.2 Ba	15.5 Ab	22.1 ABa	17.3 Ab	
MN	29.7 Aa	17.4 Ab	20.8 Aa	15.1 Ab	22.3 ABa	16.5 Ab	21.0 Ba	18.1 Aa	
Mean	29.1	17.9	20.9	16.1	21.8	17.1	22.7	17.2	
CV (%)	13								
N acumulation (g plant^{-1})									
Cultivar	$\Psi_w 0$		$\Psi_w -0.4$		$\Psi_w -0.8$		$\Psi_w -1.6$		LN
	HN	LN	HN	LN	HN	LN	HN	LN	
CV	1.10 Ba	0.96 Aa	0.84 ns	0.74 ns	0.86 ns	0.79 ns	0.73 ns	0.74 ns	
A	1.86 Aa	0.94 Ab	0.88 ns	0.96 ns	1.15 ns	0.80 ns	1.05 ns	0.88 ns	
CA	1.74 Aa	0.93 Ab	0.96 ns	0.89 ns	0.79 ns	0.69 ns	0.86 ns	0.88 ns	
MN	1.60 Aa	0.84 Ab	0.96 ns	0.80 ns	0.75 ns	0.65 ns	0.77 ns	0.80 ns	
Mean	1.57	0.92	0.91	0.85	0.89	0.73	0.85	0.83	
CV	23.2								

Within each water deficit, means followed by at least one different capital letter in the column and lower case letters in the row do not differ significantly at the 5% probability by Tukey's test. ns = non-significant.

Absorption efficiency

Two response trends for increasing water stress under HN were observed in the cultivars. Mundo Novo and Catuaí Vermelho increased AE with increasing water stress, as defined by linear equations, with a greater slope toward Mundo Novo (Figure 2A).

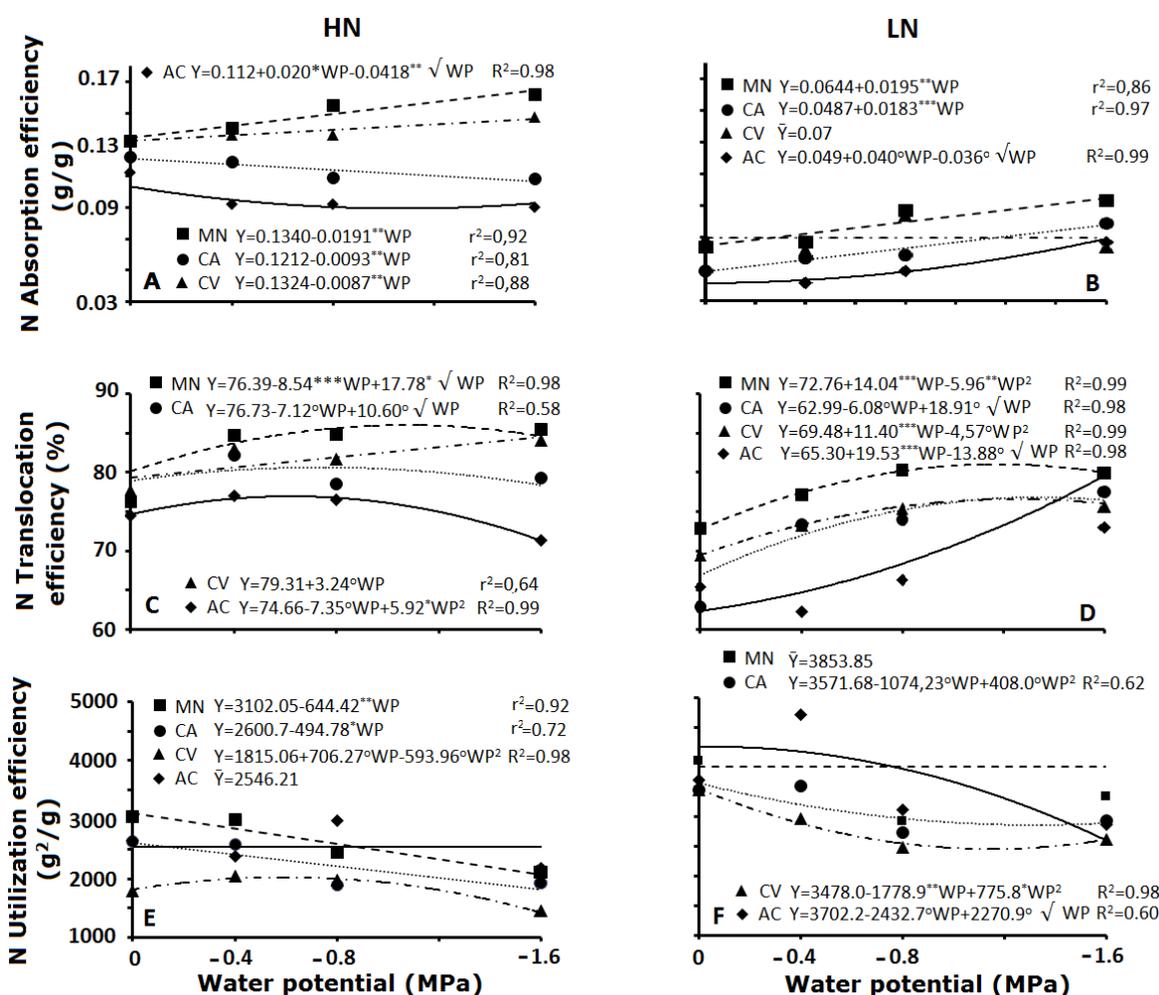


Figure 2. Absorption (A and B), translocation (C and D), and utilization (E and F) efficiency by four coffee cultivars grown with high (HN) and low (LN) nitrogen rates and subjected to water stress. ***, **, * and ° – regression coefficients significant at 1%, 5%, 10%, and more than 10% probability, respectively.

Concomitantly, the RDM of these cultivars decreased with increasing water stress (Figure 1C), suggesting an increase in absorption rates. However, a study carried out by Martinez et al. (2020) with eight coffee cultivars, including those used in this study, reported reductions in nitrate absorption rates under short-term water stress of -1.5 MPa. Mundo Novo was the only cultivar with no significant reduction in the nitrate absorption rate under these conditions. According to Glass and Siddiqi (1984), short-term responses would involve changes in the turnover rates of transport proteins, whereas long-term responses could involve an increase in the synthesis of these proteins. Martinez et al. (2020) also observed that Mundo Novo had a large increase in the expression of genes related to nitrate absorption NRT 1.2 and NRT 3.2, when subjected to -1.5 MPa of water stress; this was less intense in plants under low N conditions.

The induction of water stress negatively affected the AE of Catuaí Amarelo and Acauã, which decreased with increasing water deficit (Figure 2A). In these cultivars, root dry matter production was reduced or remained stable, and tended to decrease with increasing water stress (Figure 1C).

When subjected to LN, the cultivars Mundo Novo and Catuaí Amarelo showed a linear increase in AE, whereas Acauã had a square root response. Catuaí Vermelho maintained its AE stability under increasing water stress (Figure 2B). In the latter two cultivars, the water deficit and nutritional condition elicited a response that was of a lower magnitude than that shown by Mundo Novo and Catuaí Amarelo.

Among the cultivars, Acauã showed the lowest AE under both nutritional conditions, which indicates that its higher production of RDM (amount of tissue acquisition) under water stress (Figures 1 D, Table 1) did not result in higher N uptake. In general, under conditions of mild water deficit, the relative biomass allocation to roots increases (Hamblin Tennant, & Perry, 1991), providing an advantage by increasing the absorption capacity, but this cultivar showed no significant increase in total N accumulation.

The AE of all cultivars was higher under HN conditions at all degrees of water stress (Table 3). Consequently, it can be affirmed that N availability influences the AE. Machado et al. (2016) evaluated the nutritional efficiency of 13 coffee clones as a function of the increase in the N dose and identified increases in the absorption efficiency in most clones, demonstrating that under greater N availability, plants absorb nutrients more efficiently as long as they are not restricted by other factors.

Table 3. Efficiency of N absorption, translocation, and utilization by four coffee cultivars grown at two nitrogen doses and four degrees of water stress (Ψ_w - MPa).

Absorption efficiency $g\ g^{-1}$									
Cultivar	$\Psi_w 0$		$\Psi_w -0.4$		$\Psi_w -0.8$		$\Psi_w -1.6$		CV (%)
	HN	LN	HN	LN	HN	LN	HN	LN	
CV	0.13 Aa	0.07 Ab	0.14 Aa	0.06 Ab	0.14 ABa	0.08 Ab	0.15 ABa	0.06 Ab	
A	0.11 Aa	0.05 Ab	0.09 Aa	0.04 Ab	0.09 Ba	0.05 Ab	0.09 Ca	0.07 Aa	
CA	0.12 Aa	0.05 Ab	0.12 Aa	0.06 Ab	0.11 ABa	0.06 Ab	0.11 BCa	0.08 Aa	
MN	0.13 Aa	0.06 Ab	0.14 Aa	0.07 Ab	0.15 Aa	0.09 Ab	0.16 Aa	0.09 Ab	
Mean	0.12	0.06	0.12	0.06	0.12	0.07	0.13	0.08	
CV (%)	26.2								
Translocation efficiency (%)									
VAR	$\Psi_w 0$		$\Psi_w -0.4$		$\Psi_w -0.8$		$\Psi_w -1.6$		CV (%)
	HN	LN	HN	LN	HN	LN	HN	LN	
MN	76.3 Aa	72.8 Aa	84.7 Aa	77.2 Aa	84.8 Aa	80.3 Aa	85.4 Aa	79.9 Aa	
CV	77.7 Aa	69.5 Ab	82.9 Aa	73.2 Ab	81.7 Aa	75.4 ABa	84.0 Aa	75.6 Ab	
CA	76.5 Aa	62.9 Ab	82.2 Aa	73.3 Ab	78.5 Aa	74.0 ABa	79.3 ABa	77.5 Aa	
A	74.5 Aa	65.4 Ab	77.0 Aa	62.3 Bb	76.5 Aa	66.3 Bb	71.3 Ba	73.0 Aa	
Mean	76.2	67.6	81.7	71.5	80.4	74.0	80.0	76.5	
Utilization efficiency ($g^2\ g^{-1}$)									
VAR	$\Psi_w 0$		$\Psi_w -0.4$		$\Psi_w -0.8$		$\Psi_w -1.6$		CV (%)
	HN	LN	HN	LN	HN	LN	HN	LN	
MN	3052 Aa	3929 Aa	3007 Ab	5218 Aa	2443 ns	2923 ns	2103 ns	3545 ns	
CV	2648 Aa	3621 Aa	2372 Ab	4718 ABa	2982 ns	3121 ns	2182 ns	2878 ns	
CA	2624 Aa	3455 Aa	2575 Aa	3518 ABa	1899 ns	2740 ns	1919 ns	2936 ns	
A	1798 Ab	3449 Aa	2051 Aa	2968 Ba	1972 ns	2493 ns	1456 ns	2627 ns	
Mean	2531	3614	2501	4105	2324	2819	1915	2946	
CV (%)	30.6								

Within each water deficit, means followed by at least one different capital letter in the column and lower case letters in the row do not differ significantly at the 5% probability by Tukey's test. ns = non-significant.

DaMatta et al. (2002) investigated the effect of N rates (high and low) and two irrigation levels (high and low) and found that in the low irrigation treatment, neither the leaf water potential at pre-dawn (Ψ_{pd}) nor leaf water potential at midday (Ψ_m) were significantly altered by the N levels; however, the plants had higher relative water content at high than at low N, in a way that the water relations were, to some extent, related to N supply. Additionally, the osmotic potential was more negative in the low irrigation and high N treatment, indicating that a greater N supply can induce osmotic adjustment, which would contribute to increasing water absorption capacity and, consequently, N uptake. Rocha, Martinez, and Ribeiro (2023) reported increases in the average concentration of proline in the roots of the same cultivars with an increase in water stress under the same experimental conditions used in this study.

Under both nutritional conditions, Mundo Novo showed the most efficient N absorption under water stress, whereas Acauã was the least efficient (Figure 2A and B, Table 3). Under both nutritional conditions, there was an inverse relationship between the magnitudes of RDM and the values of AE (Figure 1C and D and Figure 2A and B).

Translocation efficiency

In HN (Figure 2C), the cultivars Mundo Novo and Catuaí Amarelo increased their TE in response to water stress according to the square root model and Acauã according to the quadratic model. The maximum TE

occurred at -1.02 MPa of stress for Mundo Novo, -0.86 MPa for Catuaí Amarelo, and -0.62 MPa for Acauã. Water potentials below these values reduced TE in these cultivars. Catuaí Vermelho showed an increase in TE with increasing water stress, fitting to a linear model (Figure 2C).

Plants increase their TE as a mechanism of adjustment to maintain shoot growth. The adjustment occurred up to the highest stress in Mundo Novo, which also exhibited the best response in terms of increasing the AE. The maximum TE was achieved with different degrees of water stress, indicating that the translocation mechanisms of the cultivars were differentially affected. It is evident that low water availability reduces nutrient transport to the shoots owing to interference with xylem transport mechanisms and reductions in transpiration flow.

Under LN conditions (Figure 2D), TE increased with water deficit and fitted square root functions for Catuaí Amarelo and Acauã, and quadratic equations for Catuaí Vermelho and Mundo Novo. The average increases of the cultivars, were 5.7, 9.4, and 13.1% at -0.4, -0.8, and -1.6 MPa of stress, respectively, compared with the control without water stress. The points of maximum efficiency as a function of the stresses were -1.24, -1.25, and -1.18 MPa for Catuaí Amarelo, Catuaí Vermelho, and Mundo Novo, respectively. The TE was lower in LN than in HN because of the lower N availability and likely because of the lower transpiration fluxes. Nevertheless, the TE in both HN and LN increased under water stress, implying that under water limitation, the cultivars improved N transport to the shoot.

Under both HN and LN conditions, Mundo Novo had the highest translocation efficiencies and Acauã had the lowest (Figure 2C and D, Table 3). In addition to being affected by transpiration rates, the translocation efficiency is also influenced by the nutritional status of the root cells and the N demand for metabolic machinery functioning, especially during photosynthesis. In addition, the efficiency of the activity of the enzymes involved in photosynthesis could require lower or higher concentrations to achieve a particular yield, which would contribute to greater or lesser nutrient transport to the leaves (Abichequer & Bohnen, 1998).

Utilization efficiency

The efficiency of utilization indicates the biomass production per unit of accumulated nutrient. The effect of water stress on this variable under each N dose treatment and cultivar is illustrated in Figure 2E and F. In HN, all cultivars, except Acauã, exhibited a reduction in UE with increasing water stress. There was no variation in the utilization efficiency of Acauã with increasing water stress. This behavior is in agreement with the stability of this cultivar in the production of SDM, RDM, and N accumulation under these nutritional conditions (Figure 1A, C, and E). Mundo Novo and Catuaí Amarelo, the most efficient cultivars under the no-stress condition, showed linear reductions in UE when subjected to prolonged and increasing water stress. Catuaí Vermelho was the least efficient among the cultivars studied and decreased in a quadratic manner after achieving maximum UE at -0.65 MPa of water stress. In LN, contrary to the result found in HN, the UE of Acauã declined sharply at water potentials below -0.22 MPa, while the UE of Mundo Novo was not affected by the increase in water stress.

Souza, Martinez, Carvalho, Loureiro, and Sturião (2020) evaluated 240-day-old plants of these same cultivars under -1.5 MPa of water stress and recorded a reduction of 56% (average of cultivars) in net photosynthesis (A) under short-term water stress. Without stress, Acauã and Mundo Novo showed the highest A, highest transpiration rate (E), and lowest water use efficiency (A/E). Short-term N stress impacted A less than water stress, which the authors attributed to the good nutritional conditions of the plants during N stress, but Acauã was the only cultivar for which photosynthesis was significantly reduced by short-term N stress. Silva et al. (2010) reported that coffee genotypes with relatively high yields subjected to water stress can maintain adequate leaf water potentials through a combination of deeper root systems and stomatal control.

In the present study, the increases in AE and TE were not sufficient to prevent the reduction in UE under water stress conditions, even when the plants received a higher N dose. The same occurred for plants that received the lower dose of the nutrient, even though under this condition, the UE increased compared to HN (Table 3). According to Bridgham, Pastor, McClaugherty, and Richardson (1995) and Silva, Furtini Neto, Vale, and Curi (1996), the utilization efficiency decreases with greater nutrient availability because the biomass production does not increase proportionally to the accumulation of the nutrient, thereby causing a decline in the use of the nutrient for biomass production.

DaMatta et al. (2002) reported that coffee under limited N supply seems to have the ability to use N more efficiently by changing its allocation to photosynthetic components as well as by increasing the activation of

Rubisco under nitrogen-limited conditions. On the other hand, the use of N previously allocated to other compartments, such as stems and branches, via retranslocation would contribute to the highest efficiencies as a response to maximize the use of the limiting nutrient.

In this study, two cultivars, Mundo Novo and Acauã, stood out. Mundo Novo showed high shoot dry matter production and high UE at the low N dose regardless of the degree of water stress, probably owing to the increase in absorption and translocation efficiencies (Figures 1B, 2B, D, and F). At the high N dose, this behavior was repeated up to -0.86 MPa of stress. At higher stresses, the UE of Mundo Novo was surpassed by that of Acauã. Under both nutritional conditions, Mundo Novo showed increased efficiency of N absorption and translocation under water stress. These adjustments led to high utilization efficiency in both nutritional conditions, notwithstanding susceptibility to water stress at high N and stability at low N doses. Therefore, Mundo Novo is suitable for environments with and without N limitation under water deficit conditions.

The shoot and root dry matter production of cultivar Acauã was not affected by water stress in the HN treatment. Under N stress, Acauã showed no significant reduction in SDM production in response to water stress, but showed a reduction in the production of RDM. Under both nutritional conditions, it presented high N accumulation, which is not explained by high absorption or translocation efficiencies, but may be related to the greater mass partition to the root system. With adequate N nutrition, Acauã had the most efficient N use under water potentials below -0.86 MPa, but under N stress, this cultivar greatly reduced its utilization efficiency in water potentials below -0.22 MPa.

Catuaí Vermelho had the lowest UE associated with the highest average foliar N concentration and the lowest dry matter production, although no significant differences were identified among cultivars. In HN, Catuaí Amarelo performed similarly to Mundo Novo in terms of SDM, RDM, and N accumulation in response to water stress. However, its UE was lower than that of Mundo Novo owing to the decrease in AE, lower TE, and lower efficiency of N utilization. Catuaí Amarelo under LN conditions showed stable SDM production, probably as a response to the increase in AE and TE, resulting in a lower percentage variation in UE. Its high N accumulation, associated with lower dry matter production, resulted in lower UE than that presented by Mundo Novo and Acauã.

Conclusion

In a nutrient solution containing polyethylene glycol (PEG), the nutritional status of N affects the response of young coffee plants to water stress. In plants previously well-supplied with N, N stress induces a greater dry matter mass allocation to roots, mitigating the effects of subsequent water deficits on shoot dry matter production. In the nutrient solution containing PEG, young plants of cultivar Mundo Novo subjected to N stress followed by increasing water stress maintained a high UE. This cultivar also stands out for its high UE in well-nourished plants, in terms of N, and water potentials above -0.86 MPa. In nutrient solutions containing PEG, young nitrogen well-nourished plants of the cultivar Acauã maintained their N use efficiency stable up to -1.6 MPa of water stress. Below -0.86 MPa, this cultivar performed better than Mundo Novo. However, under N stress, this cultivar drastically reduced its UE at potentials below -0.22 MPa. In nutrient solutions containing PEG, plants receiving high N doses had greater efficiency of nutrient absorption and translocation but lower efficiency of utilization than plants under N stress. Water stress causes reductions in N use efficiency.

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