LEAF CONCENTRATIONS OF NITROGEN AND PHOSPHORUS IN *Phaseolus vulgaris* L. PLANTS UNDER HIGH CO_2 CONCENTRATION AND DROUGHT STRESS

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ABSTRACT: This study aims to evaluate the leaf concentration of nitrogen and phosphorus correlated to the production of photoassimilates in beans plants (*Phaseolus vulgaris* L.) under high [CO_2] and drought stress. The experiment was conducted in Viçosa (Brazil), during the period from April to July 2009, by using open-top chambers equipped with CO_2 injection system. The drought stress was applied, through the irrigation suspension, during the period from flowering to maturation. The experimental design was randomized blocks in split-plot scheme with four replication, where the plots with plants grown in [CO_2] of 700 mg L^{-1} and [CO_2] environment of 380 mg L^{-1} and the subplots with plants with and without drought stress. The results were submitted to ANOVA and Tukey test ($p < 0.05$). In the plants under high [CO_2] with and without drought stress, the photosynthetic rate increased by 59%, while the dry matter presented an increment of 20% in the plants under high [CO_2] without drought stress. Reductions in [N] and [P] occurred in plants grown under high [CO_2], resulting in greater efficiency in nitrogen use for photosynthesis. The high [CO_2] increase only the total dry matter and not the total mass of grains. The drought stress reduces the dry matter and mass of grain, even at high [CO_2].

KEYWORDS: beans, mineral nutrition, photosynthesis, climatic changes.

NITROGÊNIO E FÓSFORO FOLIAR NO CRESCIMENTO DE PLANTAS DE FEIJÃO CULTIVADAS SOB ALTA [CO_2] E DEFICIÊNCIA HÍDRICA

RESUMO: Neste trabalho, as concentrações de nitrogênio e fósforo foliares foram analisadas e relacionadas com a produção de fotoassimilados do feijoeiro (*Phaseolus vulgaris* L.) cultivado sob alta concentração de CO_2 e deficiência hídrica. O experimento foi realizado em Viçosa-MG, entre abril e julho de 2009, utilizando-se de câmaras de topo aberto equipadas com sistema de injeção de CO_2. A deficiência hídrica foi aplicada pela supressão da irrigação, durante o período de floração à maturação. O delineamento experimental foi em blocos casualizados, tendo nas parcelas plantas cultivadas em [CO_2] a 700 ppm e [CO_2] ambiente (380 ppm), e nas subparcelas, plantas sem e com deficiência hídrica. Os resultados foram submetidos à Anova e ao teste de Tukey ($p < 0,05$). Nas plantas sob alta [CO_2] e sem e com estresse hídrico, a taxa fotossintética aumentou 59%, enquanto o crescimento analisado pela massa de matéria seca apresentou incremento de 20% nas plantas sob alta [CO_2] e sem estresse hídrico. Reduções nas [N] e [P] ocorreram nas plantas sob alta [CO_2], resultando em maior eficiência no uso do nitrogênio pela fotossíntese. A alta [CO_2] aumenta somente o total de massa seca da planta e não a massa de grãos. O estresse hídrico reduz a massa seca da planta e dos grãos, mesmo em alta [CO_2].

PALAVRAS-CHAVE: *Phaseolus vulgaris* L., nutrição mineral, fotossíntese, mudanças climáticas.

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INTRODUCTION

Recently, there has been an increasing fear how the future climate conditions may affect the productivity of crops (LUO et al., 2005; JAGGARD et al., 2007; COSTA et al., 2009). Among these futures climate conditions are the increase of CO_2 concentration and alteration on the water availability (LUO et al., 2005; JAGGARD et al., 2007).

Today, the current concentration of CO_2 ([CO_2]) in the atmosphere is not able to saturate the photosynthetic process, being a limiting factor as for the growth and productivity of many species (AINSWORTH & ROGERS, 2007). But in conditions of high [CO_2], studies indicate the possibility of changes in carbon/nitrogen relation (C:N) in plants with C3 photosynthetic metabolism (LEAKEY et al., 2009). This occurs due to high photosynthetic rates and biomass accumulation, with an average increase of 22% (SARDANS et al., 2012), in particular under nitrogen restrict availability, affecting crop productivity (SMITH et al., 2001). Despite the increase in photosynthesis and in the carbon allocation in plants exposed to high [CO_2], productivity is also associated with the increment in nutrients supply. In conditions of nutrient limitation, mainly nitrogen, leguminous plants such as common beans can be benefited at elevated [CO_2] due to nitrogen fixation. This further reduces the negative impacts of drought stress due to the fixation increase (ROGERS et al., 2009).

One should noted that due to the long time exposure to high [CO_2] during the cycle, many plants have presented significant photosynthetic down-regulation. This reduction in photosynthetic rate is induced by decrease in leaf nitrogen [N] and phosphorus [P] concentrations and stomatal conductance in response to high [CO_2] (SANZ-SÁEZ et al., 2010; LEE et al., 2011). The drought stress reduce the stomatal conductance too, besides the leaf [N] and [P], due the lower mineral absorbing. Also, the reduction in leaf [N] in plants under high [CO_2] (MIYAGI et al., 2007; GARTEN JR. et al., 2008; NIU et al., 2012), may result from physiological adjustment to increase the efficiency of nitrogen by high photosynthetic rate. This results in an increase in carbon absorbed rate per unit of leaf nitrogen and a decrease in leaf specific surface area and [N] per unit of dry matter. However, this increase in use efficiency of nitrogen by photosynthesis is also correlated with leaf phosphorus concentration (NIINEMETS et al., 1999). Reduction in leaf [N] in plants under high [CO_2] may also be related to weaker stomatal conductance (TAUB & WANG, 2008), has been observed in plants under high [CO_2] (AINSWORTH & ROGERS, 2007).

The common bean (*Phaseolus vulgaris* L.) presents a high degree of proteins, being responsible for much of the protein consumed worldwide, especially in Latin America and Africa. When grown in high [CO_2], and under potential supply of nitrogen fertilizer common beans presents increments between 40 and 80% in photosynthesis and 26 to 40% in dry matter. Leaf [N] in bean plants under high [CO_2] have shown a reduction between 31 to 35%, if compared to plants under [CO_2] ambient atmosphere. Despite these positive responses to treatment with CO_2, beans under high [CO_2] have presented photosynthetic down-regulation, especially in treatments with low nitrogen supply (JIFON & WOLFE, 2002).

It is clear that several questions remain unresolved insofar the link between common beans productivity and the [N], [P] and [CO_2] is concerned. Therefore, this study aims to evaluate the leaf concentration of nitrogen and phosphorus correlated to the production of photoassimilates in beans plants (*Phaseolus vulgaris* L.) under high [CO_2] and drought stress.

MATERIAL AND METHODS

The experiment was conducted at Lysimetric Station at the *campus* of the Federal University of Viçosa, Brazil (20° 45'S, 42° 45'W), between April and July 2009. Lysimeters with 1.0 m x 1.4 m section and 0.8 m depth were divided by a metal plate septum, to perform the experimental design in subplots. The substrate to fill the lysimeters was a clayey dystrophic Red-Yellow Latosol, with very clayey texture, in which the acidity was corrected by liming (Table 1). Two fertilizations have been done during the experiment, a planting one with NPK 8-28-16, 650 kg ha^{-1} and 300 kg ha^{-1} of magnesium sulfate, and the topdressing in the 16th day after sowing (DAS) with 200 kg ha^{-1} of urea and 250 g ha^{-1}of sodium molybdate by leaf fertilization. The bean variety that has been used was Majestic-UFV (*Phaseolus vulgaris* L. cv. BRSMG Majestoso), carioca type of short cycle. The plant spacing was 0.05 m and between rows was 0.50 m, totaling: 2 rows and 54 plants per lysimeter (plot); and one row and 27 per subplot.

TABLE 1. Analytical results of the soil substrate at 0 - 20 cm depth after correction of acidity by liming.

pH	P	K	Zn	Fe	Mn	Cu	B	V	m	Mo
H_2O	------------------------------ mg dm^{-3} ------------------------------							------- % -------		dag kg^{-1}
6.2	27.3	142.0	10.0	71.2	4.9	0.7	0.1	41.0	0.0	16.0
Ca^{2+}	Mg^{2+}	Al^{3+}	H+Al	SB	CEC(E)	CEC(T)	P-rem	Clay	Silt	Sand
---------------------------- cmol$_c$ dm^{-3}----------------------------							mg L^{-1}	-------------- % --------------		
1.7	0.6	0.0	3.8	2.7	2.7	6.5	13.3	70.0	9.0	21.0

where, SSNa - Sodium Saturation index; BS - Sum Base; CEC (E) - Effective Cation Exchange Capacity; CEC (T) - Total Cation Exchange Capacity in pH 7,0; V - Base Saturation; m - Aluminum Saturation; e P-rem - Remaining Phosphorus.

All plants were grown in open top chambers composed by mobile rectangular modules (1.8 x 1.4 x 0.9 m), where the modules was added to follow the growth of plants (1.8 x 1.4 x 0.5 m), and provided with an internal air distribution system consisting of fans and perforated PVC pipes (Figure 1). Two [CO_2] were used in chambers, 380 mg L^{-1} (ambient) and 700 mg L^{-1}. To have plants under [CO_2] of 700 mg L^{-1} a daily injection of CO_2 between 6:00 to 18:00 from the 9th and 90th DAS have been applied. The temperature inside the chambers was, on average, 2° C above the external environment. Drip tapes were used in plant irrigation in order to maintain the soil closer to its field capacity (FC = 33.62%). In plants subjected to drought stress, total irrigation suppression was done, between flowering (22nd DAS) and maturation (70th DAS), lasting 48 days. The water content in soil was monitored hourly by TDR (Time Domain Reflectometry), with probes installed at 0.20 m depth in soil and automatic.

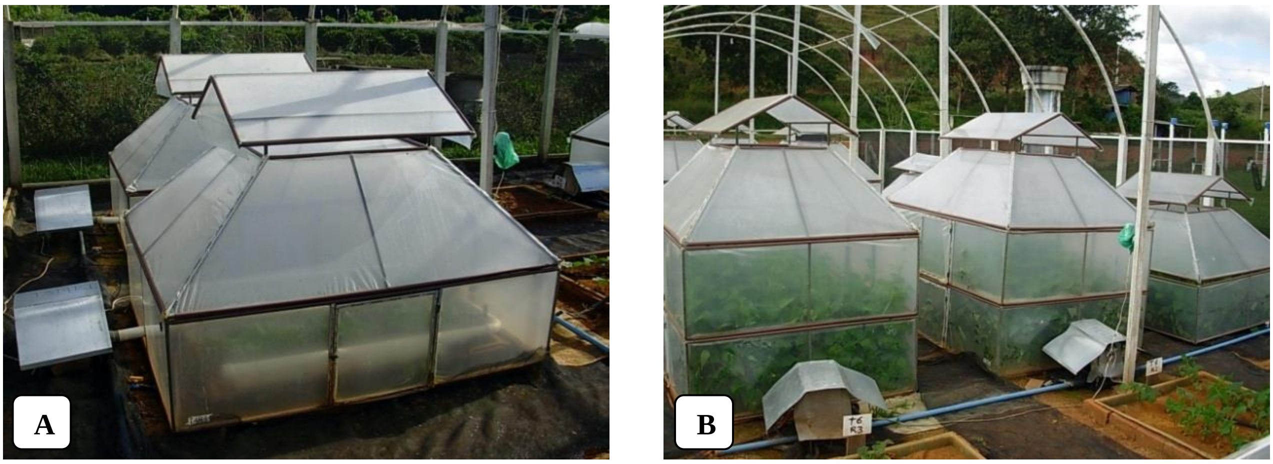

FIGURE 1. Open-top chambers composed by modules, added to follow the growth of plants: A - chambers with a module, from 9th to 40th DAS, and B - chambers with two modules, from 41st to 90th DAS.

Data collected

The data collected were: leaf nitrogen concentration ([N]); leaf phosphorus concentration ([P]), photosynthetic rate (*A*), chlorophyll total index (*CT*) and total dry matter (*DM*). The latest leaves developed were collected to determine the leaf [N] and [P], made a compose sample each sub-plot, to the leaf laboratorial analysis, as in Embrapa (2011). The photosynthetic rate was measured in the morning, between 8:00 and 11:00, with portable gas exchange meter (IRGA - Infra

Red Gas Analyzer), with external radiation source system, 1,200 µmol J m^{-2} s^{-1} and the environmental conditions (temperature, [CO_2] and relative humidity) was the same inside the chambers. The chlorophyll total index was measured with a portable chlorophyll meter. Four measurements of photosynthetic rate, chlorophyll total index and total leaf concentration of nitrogen and phosphorus were performed: 1st measurement, in the 20th DAS (vegetative stage); 2nd measurement, in the 41th DAS (pre-flowering); 3rd measurement, in the 67th DAS (grain filling); and 4th measurement, in the 82nd DAP (maturation). The efficiency use of nitrogen for photosynthesis (*AN*) was also determined, corresponding to the product of the photosynthetic rate (*A*) by nitrogen concentration ([N]), as in LEE et al. (2011). At the end of the crop cycle, in the 100th DAS, is was determinate the total mass of grains (*MG*) and total dry matter (*DT*), determined by the shoot of the plant dried in a ventilated oven at 70° C for three days.

Experimental design and statistical analysis

The experimental design was randomized blocks in split-plot scheme with four replications (blocks). The plots were the primary treatment (F), or levels of [CO_2]: F1 - plants grown under [CO_2] to 700 mg L^{-1}; and F2 - plants grown under [CO_2] ambient (380 mg L^{-1}). The subplots were the secondary treatment (S), or water availability for crop: S1 - plants grown without drought stress; and S2 - plants grown under drought stress (Figure 2). The interactions resulted in four treatments: F1S1; F1S2; F2S1 and F2S2.

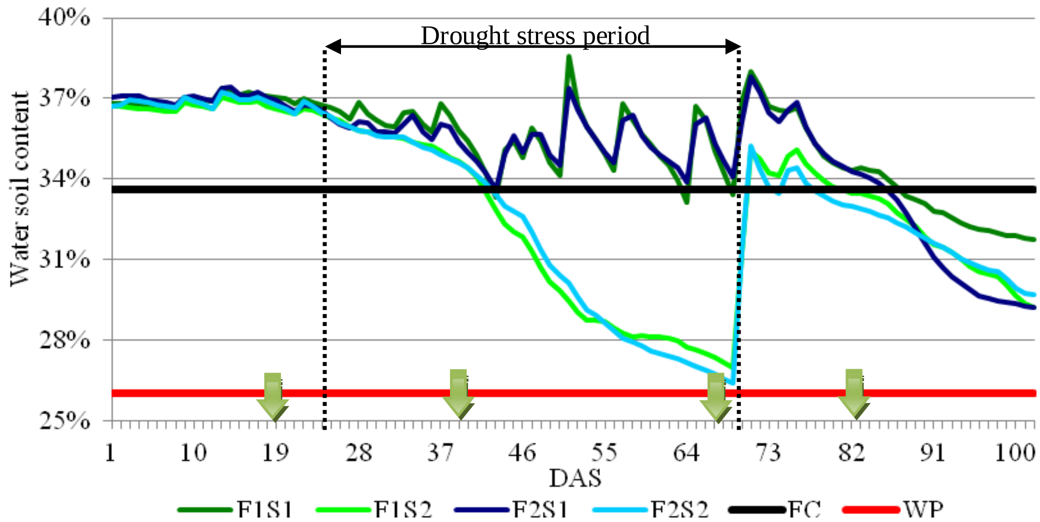

FIGURE 2. Water content in soil (%) during the experiment for each treatment, field capacity (FC), witting point (WP), drought stress induction period and the four measurements (green arrows) of photosynthetic rate, total chlorophyll index and levels of leaf nitrogen and phosphorus.

First the variables were analyzed using simple descriptive statistics (mean, standard deviation and coefficient of variation), for further analysis of variance (ANOVA). As for the comparison of different levels of the plots, subplots and treatments, the Tukey test ($p < 0.05$) has been applied. Moreover, a correlation test among all variables analyzed ($p < 0.05$). All analyzes were performed using the statistical package SAS 9.3 (SAS, 2011).

RESULTS AND DISCUSSION

Measurements of photosynthetic rates (*A*) demonstrated increments between 40 to 80% in plants under high 700 mg L^{-1} [CO_2] (Table 2). These photosynthesis values were generally higher than those found by JIFON & WOLFE (2002), BUNCE (2008) and LEE et al. (2011). These studies reported that photosynthetic rates increased between 13 to 60% for bean and other C3 plants under high [CO_2]. The largest increase in photosynthetic rate, as compared to previous results, was a result of higher [CO_2] in the atmosphere in which the plants were grown, resulting in a higher gradient of [CO_2] between the environment and the leaf, increasing the availability of CO_2 in Rubisco carboxylation site, which, probably, reduced photorespiration.

TABLE 2. Summary of descriptive analysis and variance for the four measurements of photosynthetic rate (*A*1, *A*2, *A*3 and *A*4) and four measurements of leaf concentration of nitrogen ([N]1, [N]2, [N]3 and [N]4), together with the Tukey test

Analysis of Variance (Anova)									
Source of Variation		*A*1	*A*2	*A*3	*A*4	[N]1	[N]2	[N]3	[N]4
		------------- $\mu molCO_2\ m^{-2}s^{-1}$ -------------				------------- $dagN\ kg^{-1}$ -------------			
F		**	**	**	*	*	*	**	**
S		NS	NS	*	NS	NS	NS	NS	NS
F × S		NS	NS	*	NS	NS	NS	*	NS
Mean		31.58	33.75	27.24	23.52	4.72	3.43	2.62	2.19
SD		5.13	8.69	4.50	5.34	0.33	0.44	0.35	0.19
CV (%)		16.26	25.74	16.52	22.71	7.06	12.82	13.22	8.87
Tukey Test ($p < 0.05$)[1]									
F1	S1	41.02 A	44.32 A	37.69 A	29.72 A	4.53 B	2.92 B	1.88 C	1.71 B
	S2	34.48 A	39.15 A	37.80 A	27.73 A	3.98 B	2.67 B	2.12 BC	1.73 B
F2	S1	24.27 B	24.93 B	22.46 B	18.96 B	5.31 A	4.23 A	3.68 A	2.65 A
	S2	26.55 B	26.62 B	11.02 C	17.67 B	5.09 A	3.92 A	2.79 B	2.67 A

where: * significant to $p < 0.05$; ** significant to $p < 0.01$; NS - not significant to $p < 0.05$; F - levels of [CO_2] (F1 - [CO_2] at 700 mg L^{-1}; F2 - [CO_2] at 380 mg L^{-1}); S - water availability (S1 - well watering; S2 - drought stress); SD - standard deviation; and CV - coefficient of variation. [1]mean in same column follow to the same letter are equal for the Tukey Test.

The photosynthetic rate was highly affected by the interaction between the [CO_2] and water availability only after severe drought stress (3rd measurement; Table 2). The increased availability of CO_2 was responsible for maintaining a high photosynthetic rate, even under drought stress, corroborating the observations of LEAKEY et al. (2009). They argued that plants exposed to high [CO_2] and under drought stress, like the interaction F1S2, do no experience decrease in photosynthetic rate in relation to plants under high [CO_2] and without drought stress, like the interaction F1S1. However, under current [CO_2], the effect of drought stress resulted in lower photosynthetic rate by approximately 50% (F2S2; Table 2).

The leaf concentration of nitrogen decreased throughout the crop cycle for all treatments (Table 2), which may be expected due to leaf senescence and displacement of the photoassimilates and nutrients to the grains (GARTEN JR. et al. 2008). On the other hand, MIYAGI et al., (2007) have shown increased nitrogen fixation by leguminous plants exposed to high [CO_2]. We found that common beans under high [CO_2] (F1) have presented throughout the cycle, lower [N] than plants exposed to ambient [CO_2] (F2; Table 2). Specifically, reductions have been noted in the first measurement ([N]1) between 15 to 25%, second ([N]2) between 31 and 37%, third ([N]3) between 42 and 49% and fourth ([N]4) 35%. These reductions are higher than those found by DAVEY et al. (1999), 19%, and LEE et al. (2011), 13%. Thus, the reductions of leaf [N] in plants under high [CO_2] were due to increased demand, production of photoassimilates and dry matter (Table 3), resulted of the high photosynthetic rate that plants. But this high production of photoassimilates not increases total mass of grains (*MG*), who suggest the high production of photoassimilates were drain to increase the dry matter, to produce leaves and shoots. In the third measurement ([N]3), exists an increase in the severity of drought stress in plants under ambient [CO_2] which leads to lower [N] as compared to plants without drought stress. This was due to the reduction in soil nitrogen absorption and inhibition of fixation of these plants (MIYAGI et al. 2007).

Plants under high [CO_2] and exposed to severe drought stress (F1S2) have not shown significant differences regarding plants of treatment F1S1, its might occur because, according to ROGERS et al. (2009), the high [CO_2] contributes to the maintenance of nitrogen fixation. The lower [N] throughout the growing cycle, in plants exposed to high [CO_2] (F1), might indicate that do not occur higher nodulation or nitrogen fixation by plants, but maintenance of fixation during water stress (Table 2).

TABLE 3. Summary and descriptive analysis of variance for the four measurements of variables chlorophyll total index (*CT*1, *CT*2, *CT*3 and *CT*4), total mass of grains (*MG*) and total dry matter (*DM*), together with the Tukey test

Analysis of Variance (Anova)							
Source of Variation		*CT*1	*CT*2	*CT*3	*CT*4	MG	*DM*
		chlorophyll total index				g	g
F		NS	NS	NS	NS	NS	NS
S		NS	NS	NS	*	****	***
F × S		NS	NS	NS	NS	NS	*
Mean		50.00	54.06	50.42	42.07	395.23	796.47
SD		3.23	3.47	4.01	4.80	58.91	59.25
CV (%)		6.45	6.41	7.96	11.40	19.30	7.44
Tukey Test ($p< 0.05$)[1]							
F1	S1	49.49	54.16	48.20	37.24	478.23	1030.87 A
	S2	52.23	51.04	51.05	45.98	154.96	692.99 BC
F2	S1	48.66	53.41	47.76	40.89	417.14	825.79 B
	S2	49.63	57.66	54.67	44.18	170.59	636.24 C

where: * significant to $p < 0.05$; *** significant to $p < 0.001$; NS - not significant to $p < 0.05$; F - levels of [CO_2] (F1 - [CO_2] at 700 mg L^{-1}; F2 - [CO_2] at 380 mg L^{-1}); S - water availability (S1 - well watering; S2 - drought stress); SD - standard deviation; and CV - coefficient of variation. [1]mean in same column follow to the same letter are equal for the Tukey Test.

Due to the high photosynthetic rates of plants under high [CO_2] (F1S1), the dry matter (*DM*) at the end of the crop cycle has shown a significant increase of 25% (Table 3), close to the value obtained by JIFON & WOLFE (2002), NASSAR et al. (2008) and LEAKEY et al. (2009), but lower than that obtained by MIYAGI et al. (2007), 106%. Common beans under high [CO_2] and drought stress (F1S2) experienced a distinct response, with a decrease of 16% in dry matter, while under atmosphere [CO_2] and drought stress (F2S2) experienced a decrease close to 23%, lower that found by SILVEIRA & STONE (2008). This indicates that high [CO_2] support only the photosynthetic rate, until the effects of severe drought stress are felt by plants resulting in leaves and pod fall and consequently reduced final dry matter (*DM*) and mass of grains (*MG*) (Table 3). The reduction of 63.64% in the total mass of grains (*MG*) of plants subjected to drought stress was greater than that found by SAUCEDO et al. (2006) and SILVEIRA & STONE (2008), approximately 30 and 42%, respectively, possibly due to the magnitude and period of drought stress in this experiment, because according to DAVIES (2006), when the drought stress occurs in important stages of the plant, such as flowering and pod formation, the productivity losses are considerable. It should be noted that these results are very likely associated with the fact that measurements of photosynthetic rate are performed on fully expanded leaves, under ideal conditions of light, not reflecting the conditions of all the leaves of the plant.

TABLE 4. Summary and descriptive analysis of variance for the four measurements of variables leaf concentration of P ([P]1, [P]2, [P]3 and [P]4) and efficiency of nitrogen use for photosynthesis (*AN*1, *AN*2 , *AN*3 and *AN*4), together with the Tukey test.

Analysis of Variance (Anova)									
Source of Variation		*AN*1	*AN*2	*AN*3	*AN*4	[P]1	[P]2	[P]3	[P]4
		--------- $\mu molCO_2\ m^{-2}s^{-1}\ dagN^{-1}\ kg$ ---------				----------------- $dagP\ kg^{-1}$ -----------------			
F		*	*	**	**	NS	*	*	**
S		NS	NS	NS	NS	NS	NS	NS	NS
F × S		NS	NS	NS	NS	NS	NS	**	NS
Mean		6.94	10.80	12.12	11.85	0.33	0.21	0.16	0.16
SD		1.28	3.12	3.18	3.82	$2.00\ 10^{-2}$	0.04	0.03	0.02
CV (%)		18.50	28.93	26.25	32.22	6.07	18.57	15.68	15.01
Tukey Test (p< 0.05)[1]									
F1	S1	9.11 [A]	15.18 [A]	19.99 [A]	17.64 [A]	0.33	0.18 [B]	0.10 [B]	0.14 [B]
	S2	8.79 [A]	15.19 [A]	18.36 [A]	16.07 [A]	0.29	0.14 [B]	0.16 [B]	0.16 [B]
F2	S1	4.62 [B]	5.92 [B]	6.16 [B]	7.10 [B]	0.36	0.27 [A]	0.22 [A]	0.17 [A]
	S2	5.24 [B]	6.93 [B]	3.96 [B]	6.58 [B]	0.34	0.26 [A]	0.16 [AB]	0.18 [A]

where: * significant to p < 0.05; ** significant to p < 0.01; NS - not significant to p < 0.05; F - levels of $[CO_2]$ (F1 - $[CO_2]$ at 700 mg L^{-1}; F2 - $[CO_2]$ at 380 mg L^{-1}); S - water availability (S1 - well watering; S2 - drought stress); SD - standard deviation; and CV - coefficient of variation. [1]mean in same column follow to the same letter are equal for the Tukey Test.

The leaf phosphorus concentration, [P], has shown a similar response to [N], with significant reductions in [P] over the cycle in plants exposed to high $[CO_2]$ (F1). In the second measurement ([P]2), the reduction was between 33 to 48%, in the third ([P]3), from 27 to 55% and in the fourth ([P]4), between 6 to 18% (Table 4). The reductions in [P] in C3 leguminous plants under high $[CO_2]$ have been also found by NIINEMETS et al. (1999), with reductions of 53% in [P] leaf, and JIN et al. (2012). This drop in [P] in plants under high $[CO_2]$ is associated with increased demand of nutrients due to enhanced production of photoassimilates and dry matter (Table 2), once the plant metabolism was grow, like the Rubisco Enzyme activate, Calvin Cycle and Electron Transportation (AINSWORTH & LONG, 2004).

Due to the high photosynthetic rates (*A*) and low concentrations of nitrogen ([N]) in plants under high $[CO_2]$ (Table 2), the efficiency of nitrogen use in photosynthesis (*AN*) was higher in these plants throughout the cycle (Table 4), with increments of 82% in the first measurement (*AN*1), 136% in the second measurement (*AN*2), 279% in the third measurement (*AN*3) and 146% in the fourth measurement (*AN*4). These increases in *AN* were greater than those found by LEE et al. (2011), 21% for common beans. This increase in the efficiency of nitrogen use by photosynthesis was also reported by DAVEY et al. (1999).

Despite these differences in the concentrations of nitrogen and phosphorus in plants subjected to the applied treatments discussed above, no significant differences in the rate of chlorophyll total index (*CT*) were found insofar the first three measurements have been concerned (Table 3). The only significant difference (p < 0.05) has occurred in the last measurement, but only under drought stress (S). Plants experiencing drought stress had higher levels of chlorophyll total index (*CT*), due to higher stock of nitrogen in the soil which was not consumed during drought stress. NIINEMETS et al. (1999) have no also found reduction in the concentration of chlorophyll in C3 plants under high $[CO_2]$.

The Pearson correlations (r) between the characteristics evaluated were largely non-significant (p > 0.05) and positive (Table 5). The efficiency of nitrogen use for photosynthesis (*AN*) showed a strong negative correlation with leaf phosphorus concentration, corroborating the observations of NIINEMETS et al. (1999). The *AN* has also shown strong correlation with photosynthetic rate (*A*) and high negative correlation with nitrogen concentration ([N]). This may be anticipated because *AN* is the product of the *A* times the [N]. The nitrogen concentration has shown a strong correlation with the concentration of phosphorus, because the both absorption is related to each other.

TABLE 5. Estimates of the Pearson correlation of coefficients (r) between all variables and t test for correlations

	[N]	[P]	*A*	*CT*	*DM*	*AN*	*MG*
[N]	1.0000						
[P]	0.9122 ***	1.0000					
A	-0.0744 NS	-0.0485 NS	1.0000				
CT	0.3340 ***	0.2460 *	0.1615 NS	1.0000			
DM	-0.1097 NS	-0.0817 NS	0.5133 *	-0.4053 NS	1.0000		
AN	-0.6902 ***	-0.5993 ***	0.7039 ***	-0.1646 NS	0.3895 NS	1.0000	
MG	0.1726 NS	0.0866 NS	0.2285 NS	-0.5452 *	0.8690 ***	0.1084 NS	1.0000

where: * significant to $p < 0.05$; *** significant to $p < 0.001$; NS - not significant to $p < 0.05$; [N] - leaf nitrogen concentration; [P] - leaf phosphorus concentration; *A* - photosynthetic rate; *CT* - chlorophyll total index; *DM* - total dry matter; *AN* - efficiency use of nitrogen for photosynthesis; and *MG* - total mass of grains.

The results obtained in this study corroborate the observations made by SORATTO (2004), who claimed that it is possible to use the chlorophyll total index (*CT*) as indicative of the nitrogen content in the leaf and, hence, the need for fertilization in the culture (Table 5). The chlorophyll total index was also significantly correlated with the concentration of leaf phosphorus. This result differs from that of NIINEMETS et al. (1999), because the chlorophyll total index (*CT*) show negative correlation with total mass of grains (*MG*). Different of the final dry matter of common beans (*DM*), have shown significant correlation with the photosynthetic rate (*A*) and (strong significant) total mass of grains (*MG*), which may indicate increased production of photoassimilates under high [CO_2]. But this increased production of photoassimilates were only to increase leaves and shoots.

CONCLUSIONS

Leaf concentrations of nitrogen and phosphorus are lower in bean plants exposed to high concentrations of CO_2, with similar reductions for both nutrients. These low concentrations result from an increased production of photoassimilates but only on dry matter, leaves and shoots. Plants under ambient CO_2 concentration and without drought stress present higher leaf concentrations of nitrogen and phosphorus than plans under drought stress and/or high CO_2 concentration. Due to the increase in photosynthetic rate and the reduction in leaf concentration of nitrogen, plants grown under high CO_2 concentration have shown greater efficiency in nitrogen use for photosynthesis.

The final dry matter is bigger in bean plant exposed to high concentrations of CO_2 without drought stress, but this increase production of photoassimilates are only to increase leaves and shoots, not grain. The final dry matter presents positive correlation with the photosynthetic rate and total mass of grains.

The drought stress reduces the dry matter and mass of grain, same with high [CO_2].

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