



Sensibilidade de genótipos de feijão ao estresse hídrico

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Resumo - Este trabalho teve como objetivo caracterizar agronomicamente genótipos de feijão crioulo e comercial para tolerância ao estresse hídrico, bem como verificar o efeito do estresse hídrico nos caracteres agrônômicos de interesse. No Rio Grande do Sul o feijão é cultivado entre agosto a abril, período em que, frequentemente ocorre estresse hídrico. Primeiramente, foram caracterizados a campo 25 genótipos de feijão (19 crioulos e 6 variedades comerciais). Posteriormente, os genótipos que se destacaram na avaliação a campo e mais duas testemunhas (BAT477 e IPR Juriti) foram submetidos a duas condições hídricas: i) irrigados conforme a necessidade hídrica da cultura durante todo o ciclo e; ii) irrigados conforme a necessidade hídrica da cultura até o aparecimento do primeiro botão floral (estágio R6), quando a irrigação foi suspensa por um período de 10 dias. O estresse hídrico teve influência negativa sob todos os caracteres, resultando em: i) aumento da temperatura foliar; ii) redução na capacidade fotossintética; iii) redução no número de legumes por planta e; iv) redução no número de grãos por legume. Os genótipos BAG40, BAG100 e BAG102 podem ser promissores para a tolerância ao estresse hídrico, uma vez que foram insensíveis na maior parte dos caracteres avaliados, principalmente aqueles relacionados à produtividade.

Palavras-chave: *Phaseolus vulgaris* L. Déficit hídrico. Banco de germoplasma.

Sensitivity of common bean accesses to water stress

Abstract - This study aimed to characterize agronomically landraces and commercial genotypes for tolerance to drought stress and investigate the effect of water stress on the agronomic traits of interest. In Rio Grande do Sul, beans are grown from August to April, when water stress is usually observed. First, 25 bean genotypes (19 landraces and 6 commercial varieties) were characterized in the field. Then, the genotypes that excelled in the field and two controls (BAT477 and IPR Juriti) were submitted to two water conditions: i) irrigation according to the water requirement of the crop throughout the cycle and; ii) irrigated according to the water requirement of the crop until the appearance of the first floral bud (stage R6), when irrigation was suspended for a period of 10 days. Water stress had a negative effect on all traits, and caused: i) increased temperature; ii) reduced photosynthetic capacity; iii) reduced number of legumes per plant and; iv) reduced number of grains per legume. Genotypes BAG40, BAG100 and BAG102 may be promising for tolerance to water stress, since they were insensitive in most traits assessed, especially those related to productivity.

Key words: *Phaseolus vulgaris* L. Water deficit. Genebank.

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Introduction

Plants are often exposed to drought and heat stresses, which reduce crop yields worldwide (LIPIEC et al., 2013). Beans are among the plant species considered sensitive to water stress, mainly due to their low capacity to recover after water stress and their poorly developed root system (FRANCISCO et al., 2016). Water stress, caused by water deficit, is the second cause that limits bean production in Cuba (POLÓN et al., 2014) and in Latin America (SUÁREZ et al., 2016). In Brazil, beans (*Phaseolus vulgaris* L.) are traditionally grown in three seasons, in the water season (September to November), in the dry season (January to March) and in Autumn-Winter (May-July). However, in Rio Grande do Sul, beans are grown only between the months of August and December (harvest) and January and April (second season), which coincides with periods of increased sensitivity to drought in the summer months. According to Ávila (1994), the probability of rainfall overcoming the potential evapotranspiration in the months from December to February is less than 60% in almost the entire state. This indicates high frequency of water deficit and, consequently, reduced grain yield, which is one of the main limitations to bean cultivation in Rio Grande do Sul (MALUF et al., 2001). Furthermore, there is evidence of increased temperature for the coming years (IPCC, 2007), and water stress periods will probably worsen. Cerri et al. (2007) reported that climate change and variability, such as drought and other extreme phenomena related to climate changes, directly affect the quality and quantity of agricultural production and, in many cases, even damaging it.

In this scenario, it is evident that beans will be one of the crops most affected by rising temperatures and droughts in the coming years, especially if they occur simultaneously. The combined effect of high temperature and water stress on the yield of many crops is stronger than the effect of each of these stresses individually (DREESEN et al., 2012; ROLLINS et al., 2013). Acosta-Gallegos and Shibata (1989) found reduction in all production components when beans were subjected to water stress, and this reduction in output was greater when the stress was applied during the reproductive phase, compared to the vegetative phase, which was ascribed to the reduced leaf area and reduced number of pods per plant. Still when the water deficit occurs during the reproductive stage, it has adverse effects on the yield of beans (BOICET, 2010).

Given the importance of water for the better development of crops becomes essential studies that understand the physiological responses of depending on the variation in availability water (CHAVARRIA et al., 2015). Therefore, it is clear the need for development in breeding aiming at improving drought tolerance. Thus, it is important to develop new cultivars tolerant to drought (water stress). Therefore, knowledge of the physiological and morphological traits of some genotypes related to tolerance to water deficits has practical effects on the selection, breeding and recommendation of superior individuals.



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This study aimed to characterize agronomically genotypes of landraces and commercial beans for tolerance to drought stress and assess the effect of water stress on the agronomic traits of interest.

Material and methods

In 2012/13 (harvest and second harvest), 25 genotypes at Bean Genebank from the Fundação Estadual de Pesquisa Agropecuária (FEPAGRO) were characterized in the field of the experimental area of Centro de Pesquisa do Litoral Norte (Research Center of the North Coast) - FEPAGRO Litoral Norte. Nineteen of them were landraces, followed by the numbers related to the sequence of their introduction into the seed bank. They were described as follows: BAG01, BAG17, BAG18, BAG19, BAG22, BAG24, BAG36, BAG37, BAG40, BAG47, BAG51, BAG53, BAG55, BAG61, BAG77, BAG97, BAG100, BAG102, BAG104 and six commercial cultivars, namely, FEPAGRO26, Guapo Brilhante, IPR Jurití, IPR Tuiuiú, Iraí and Ouro Branco. The FEPAGRO Litoral Norte experimental area is located in the municipality of Maquiné/RS, latitude 29° 54' South, longitude 50° 19' W, altitude 38 m, climate Cfa, annual rainfall of 1679.3 mm (Matzenauer et al. 2011) and typical Orthic Haplic Chernosol. The experiment was arranged in a randomized block design with three replicates per treatment. Each plot consisted of four 4-m long rows, spacing of 0.45 m, and total area of 7.2 m² for each genotype. The useful area was composed of two central lines, and 0.5 m of the ends were discarded. The experiment was conducted under natural rainfall conditions. Weed control was carried out through hand weeding. Insect control was conducted through the use of chemicals, when severe damage to the experiment was caused by insects. Fertilizers were applied according to soil analysis.

At the end of the morphological characterization in the field, ten accessions agronomically superior for traits of agronomic interest were identified and sown in greenhouse in 2013/14, in order to assess tolerance to water stress (Table 1).

Two commercial varieties were also used as controls; one tolerant (BAT477) and the other, susceptible (IPR Jurití) to water stress. The seeds of the genotypes were planted in plastic pots containing 10 kg of soil. Five seeds were sown in each pot to ensure the achievement of the desired number. After two weeks, thinning was conducted and two plants were maintained in each pot. At the time of sowing, the soil received treatment, according to the results of the chemical analysis performed. The topdressing fertilization was performed with urea (N) when the plants were at the V3-4 stage. When necessary, the weeds were removed manually. The experiment was arranged in a randomized block design with three replicates per treatment.



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Table 1. Characteristics of 10 genotypes of beans from the genebank collection of FEPAGRO/Centro de Pesquisa do Litoral Norte (BAGFE).

Passport Data					
Accessions	Name	Origin	Category ⁽¹⁾	Group	Color
BAG01	Paulista	Iraí/RS	TC	Black	Black
BAG19	Chumbinho	Salto do Jacuí/RS	TC	Black	Black
BAG36	Unknown	Vista Alegre/RS	TC	Carioca	Carioca
BAG40	Morrinho	Erebango/RS	TC	Manteigão	Carioca/purple
BAG47	Taquariano	Erebango/RS	TC	Black	Black
BAG91	IPR Jurití	IAPAR/PR	BC	Carioca	Carioca
BAG97	SM 9809	Maquiné/RS	BC	Black	Black
BAG100	SM 0014	Maquiné/RS	BC	Manteigão	Carioca/purple
BAG102	Muçum	Muçum/RS	TC	Black	Black
BAG171	BAT 477	EMBRAPA	BC	Manteigão	Cream

⁽¹⁾TC – Traditional Cultivar (*Landrace*); BC – Breeding Cultivar and lines. (IPGRI, 2001)

The genotypes were subjected to two moisture conditions, following the methodology proposed by Aguiar et al. (2008), with modifications: i) the accessions were irrigated according to the water requirement of the crop throughout the cycle and; ii) the accessions were irrigated according to the water requirement of the crop until the emergence of the first floral bud (stage R6). Then, irrigation was interrupted for 10 days. After this period, it was resumed according to the water requirement of the crop until the completion of the cycle.

Leaf temperature and total chlorophyll were verified daily, between 7 and 9 am, from the start of water restriction, with the aid of a digital infrared thermometer Model ST-600, produced by Incoterm® and a chlorophyll meter ClorofiLOG® CFL 1030, produced by Falker, which gives the results in a specific index called FCI: Flaker Chlorophyll index. A total of 2052 observations were recorded for leaf temperature, and 2052, for chlorophyll content. The following traits were assessed: Leaf temperature in °C (LET), total chlorophyll (CLO), plant cycle in days (PC), plant height in cm (PH), first pod insertion in cm (FPI), stem diameter in mm (STD), root length in cm (RL), root dry weight in g (RDW), shoot dry weight in g (SDW), number of pods per plant (NPP), number of grains per plant (NGP) and number of grains per pod (NGP).



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The plants were plucked for investigation of the root system. First, the soil was slightly revolved in the pots with pointed rods and then the plants were removed from the soil. Next, the roots were washed in running water and dried with paper towels water for the performance of measurements of the length of major axis with the aid of a calibrated millimeter measuring tape. Then, the roots were individually placed in paper bags and taken to a drying oven at 48 °C, for 24 hours, for the determination of the dry mass. After this period, the roots were weighed on a digital scale with a precision of 0.001 g.

The results of the variables were subjected to analysis of variance by the F test at 5% error probability. Comparison of means of treatments with main effect was conducted using the Tukey test at 5% significance. The degrees of freedom of the interaction were unfolded through simple effect (slice). The statistical package SAS University Edition® (SAS INSTITUTE Inc., 2014) was used in this procedure. For the preparation of the graphics SciDAVis free program available for download on <http://scidavis.sourceforge.net/index.html> was used.

Results and Discussion

A significant effect of water condition was also detected on most characters, as well as significant interaction between genotype and water condition for most characters, except plant height, insertion of first legume, stem diameter, root length and root dry weight. Thus, water condition affects genotypes, but with different intensities, depending on the characters. Once significant interaction is detected, it is necessary to decompose the degrees of freedom of the factors involved in the interaction (in this case, genotypes and water condition). For the characters presenting no interaction, work should be conducted only with the main effects.

The analysis of the comparison of means revealed that genotypes BAG36, BAG40 and BAG47 presented higher value for plant height (Table 2).

Genotypes BAG47 and BAT477 showed the highest and lowest values, respectively, for the character insertion of first legume. Genotype Jurití presented the highest value for stem diameter, while genotype BAG47 stood out with the highest average (Table 3) for the character root dry weight. In general, genotype BAG47 stood out with the highest averages for the characters studied. In relation to water conditions *per se*, water stress reduced the insertion of the first legume (Table 2). The characters plant height, insertion of first legume and stem diameter vary according to the breeding program, in which priority is given to either higher or lower values. However, limited soil moisture reduces plant size, leaf area and the amount of potential storage locations of the dry matter produced (ABEBE et al., 2002). Thus, under water stress, the traits of interest may be more reduced than desired by the breeding program, which may spoil the new variety.



Table 2. Means comparison of ten genotypes and two water conditions for the characters plant height (PH), first pod insertion (FPI), stem diameter (STD), and root dry weight (RDW).

Genotypes	Traits			
	PH (cm)	FPI (cm)	STD (mm)	RDW (g)
BAG01	40.63 bc	8.00 bc	0.58 b	1.49 ab
BAG100	31.20 c	7.54 bc	0.50 b	1.07 b
BAG102	40.76 bc	9.38 ab	0.53 b	1.17 b
BAG19	34.96 c	7.78 bc	0.53 b	1.37 ab
BAG36	46.42 ab	9.59 ab	0.50 b	1.01 b
BAG40	49.29 ab	8.88 abc	0.49 b	1.36 ab
BAG47	54.96 a	11.00 a	0.56 b	1.94 a
BAG97	31.87 c	7.73 bc	0.53 b	1.15 b
BAT477	32.83 c	6.70 c	0.55 b	1.42 ab
JURITI	32.96 c	8.80 abc	0.65 a	1.46 ab
Water condition				
Without stress	40.58 a	9.01 a	0.54 a	1.29 a
With stress	38.84 a	8.07 b	0.54 a	1.39 a

Same letters do not differ statistically by the Tukey test ($P < 0.05$)

Significant differences and varied behaviors were observed among the genotypes compared in the two water conditions (Table 3).



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Table 3. Unfolding of the degrees of freedom through the simple effect (slice), by fixing the level of a factor and varying the levels of the other (genotype - fixed - and water conditions - variant) and means of the traits leaf temperature in °C (LET), total chlorophyll (CLO), plant cycle in days (PC), shoot dry weight in g (SDW), number of pod per plant (NPP), number of grains per plant (NGP) and number of grains per pod (NGP) of ten bean genotypes grown under two water conditions.

Genotype	Simple Effect – F value (GL = 1)						
	LET	CLO	PC	SDW	NPP	NGP	NGP
BAG01	2.67	0.02	19.27*	4.63*	14.21*	19.14*	2.04
BAG100	68.19*	0.02	0.01	0.04	0.24	1.24	1.30
BAG102	11.64*	1.26	0.22	0.10	0.13	0.86	3.05
BAG19	2.67	6.54*	8.20*	2.75	5.85*	11.76*	2.99
BAG36	120.59*	6.49*	0.30	0.69	12.86*	9.20*	0.44
BAG40	116.22*	12.41*	19.27*	1.03	0.45	2.34	2.22
BAG47	0.43	0.01	1.20	5.17*	1.79	22.06*	12.83*
BAG97	66.31*	9.16*	8.94*	19.98*	15.12*	20.98*	1.68
BAT477	6.71*	0.11	0.69	0.33	4.09*	1.51	1.18
JURITI	6.66*	10.79*	9.01*	0.63	9.24*	1.69	2.50

Genotype	Water condition	Traits Means						
		LET	CLO	PC	SDW	NPP	NGP	NGP
BAG01	With stress	22.64	47.53	80.33	6.54	8.50	27.17	3.38
	Without stress	22.17	47.43	77.67	3.48	13.67	54.33	3.95
BAG100	With stress	22.58	47.13	77.33	3.59	8.92	37.25	4.25
	Without stress	20.18	47.26	77.33	3.31	9.58	44.17	4.71
BAG102	With stress	22.63	45.05	77.33	5.38	9.33	37.42	4.04
	Without stress	21.63	45.99	77.00	5.89	8.78	43.11	4.76
BAG19	With stress	22.92	44.55	78.82	5.60	9.55	34.09	3.56
	Without stress	22.44	46.69	77.00	3.12	12.92	55.92	4.28
BAG36	With stress	23.68	46.83	77.67	4.27	8.25	37.58	4.60
	Without stress	20.48	48.97	77.33	3.08	13.17	56.42	4.33
BAG40	With stress	23.46	43.83	81.00	5.83	8.83	16.83	2.05
	Without stress	20.33	40.88	78.33	7.28	9.75	26.33	2.65
BAG47	With stress	22.77	47.35	80.00	7.45	10.33	25.83	2.75
	Without stress	22.58	47.27	79.33	4.21	12.17	55.00	4.19
BAG97	With stress	22.69	45.78	75.18	9.73	9.82	32.36	3.51
	Without stress	20.33	48.32	77.00	3.14	15.25	61.50	4.05
BAT477	With stress	22.44	46.96	79.00	4.98	10.00	44.67	4.53
	Without stress	21.56	47.30	78.45	4.11	12.82	52.27	4.06
JURITI	With stress	22.46	48.71	79.00	5.91	9.00	36.50	4.11
	Without stress	21.71	51.46	77.00	4.58	13.33	45.08	3.46

*Significant at 5% error probability by the F test.



It is appropriate to point out that the ideal genotype would be insensitive or less sensitive to changes in water condition. For the trait leaf temperature, genotypes BAG01, BAG19 and BAG47 showed no significant differences by the simple effect, ie, they were considered insensitive to the water conditions of the experiment. Guimarães et al. (2011) observed a relationship between canopy temperature and water condition of plants. They also observed that the number of grains per pod and the number of pods per plant were reduced with increased leaf temperature. It suggests that infrared thermometry is efficient to infer plant water condition, and is therefore useful for discriminating genotypes in programs targeting tolerance to drought. Genotypes BAG19, BAG36, BAG40, BAG97 and Juriti were sensitive for the character total chlorophyll. In many cases, including beans, water deficiency leads to reduced chlorophyll (GRZESIAK et al., 2007). Drought stress is usually characterized by loss of chlorophyll and progressive decline in the photosynthetic capacity of plants (SILVA et al., 2014). BAG102, BAG36, BAG47 and BAT477 were insensitive for plant cycle. Three genotypes can be considered sensitive for air dry weight: BAG01, BAG47 and BAG97. Rosales-Serna et al. (2004) found that, under water stress, tolerant cultivars showed reduced number of days to maturity. However, Teran and Singh (2002) observed similar physiological maturity between genotypes with different levels of tolerance to abiotic stress, which leads to the conclusion that response of the cycle to water stress depends on the material studied. Regarding the three traits related to grain yield (number of legumes per plant, number of grains per plant and number of grains per legume), genotypes BAG100, BAG102 and BAG40 showed no differences for water conditions. Genotype BAG47 was not sensitive for the trait number of legumes per plant, and genotypes BAT477 and Juriti were not sensitive for number of grains per plant. BAG47 was the only genotype that showed a significant difference for the number of grains per legume (Table 3). Weaver et al. (1984) observed that decreased water potential in the soil reduces the number of pods and seeds per plant and the total yield per plant in 20% to 40%; but the number of seeds per pod and 100 seed weight were not reduced. Miorini (2012) also considers that the variable number of pods was severely affected by water suppression at flowering (it is the most critical treatment in relation to the number of pods) it leads to abortion and fall of flowers and reduced number of pods per plant.

The mean values of all genotypes in relation to both water conditions (Table 3) is also provided. By matching this information with the results of the interaction, it is possible to observe differences between the values of the genotypes subjected to stress and those which were not, for the seven traits assessed. The negative effect of water stress condition on every trait is clear, since it results in: i) increased leaf temperature; ii) reduced photosynthetic capacity; iii) reduced number of legumes per plant; and iv) reduced number of grains per legume (Table 4). It is also evident that genotypes BAG40, BAG100 and BAG102 can be considered promising for



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tolerance to water stress, since they were insensitive in most traits studied, especially those related to productivity.

As already said, morphological changes were observed in the genotypes assessed, due to water stress. According to Taiz and Zeiger (2009), plants use three main lines of defense against drought, namely, i) reduction in leaf area; ii) elongation of roots and; iii) stomatal closure. Pimentel and Perez (2000) found that leaf water potential is a good indicator of the effect of water deficit on bean, together with the leaf area and dry matter mass of shoots, it can discriminate genotypes more tolerant to water stress. Thus, the genotype BAG47 presented the highest root dry weight and one of the highest aerial dry weights (although its traits related to yield decreased), whereas for genotypes BAG100 and BAG102, no significant differences were found between the aerial dry weight when subjected to the two water conditions. No reduction was found for the three genotypes for chlorophyll levels. Although genotype BAG40 showed increased leaf temperature and reduced chlorophyll content during the period of water restriction, no changes were observed in traits related to yield. It indicates that this material may have some recovery mechanism, which justifies maintaining it as promising.

It is interesting to point out that two of the most promising genotypes have different genetic constitution, one is manteigão (BAG40) and the other, rajado (BAG100). The other genotype (BAG 102) belongs to the black group, but is a rustic variety. This may indicate that genotypes from groups different from black and carioca or more rustic groups may be more tolerant to water stress. Muñoz-Perea et al. (2006) evaluated 13 genotypes for drought tolerance, and found that the Mexican red bean was tolerant to water stress. There is evidence that the genotype BAT477, with cream grains, is tolerant to drought (GUIMARÃES et al., 1996.), and that genotypes of Durango beans, with beige grains, are more tolerant to drought (SINGH, 2007). One hypothesis is that, due to their hardness, the mechanism of tolerance to water stress is maintained in these materials, probably present in wild species. However, it is reduced in present varieties because of domestication and breeding. A fact that corroborates this hypothesis is that most of the modern varieties is either carioca or black type. Further studies, nevertheless, are needed to confirm and identify the traits of these genotypes that provide tolerance to water stress.

The graphs of leaf temperature and total chlorophyll show the behavior of genotypes over the days when they were subjected to both moisture conditions (Figures 1 and 2, respectively).

In general, the genotypes that maintained daily values of leaf temperature below the overall average, with approximate values in both water conditions, were those called insensitive: BAG01, BAG19 and BAG47 (Figure 1).

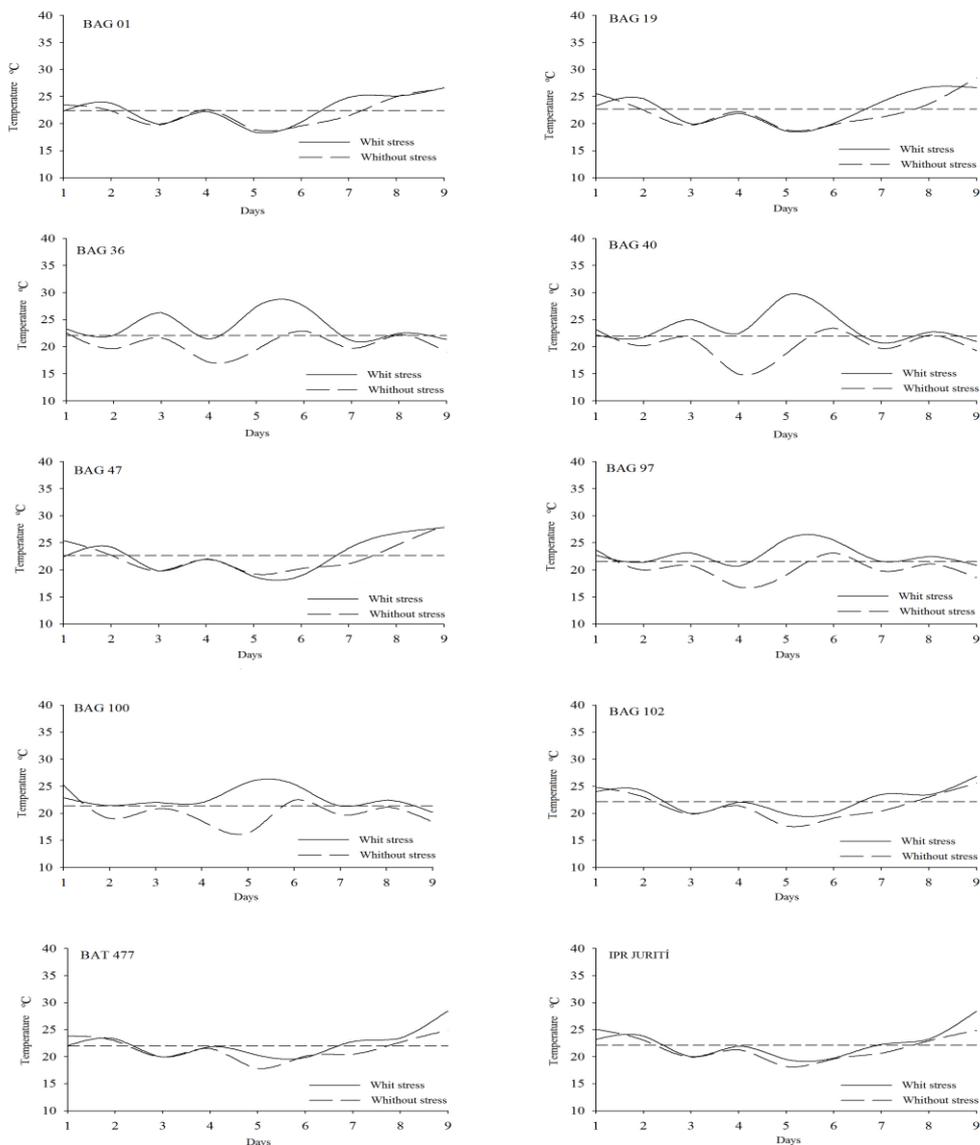


Figure 1. Values of leaf temperature obtained daily in 10 bean genotypes grown under two water conditions (with and without water stress).

On the other hand, genotypes BAG36, BAG40, BAG97 and BAG100 obtained the highest daily fluctuations, including significant difference between the water regimes, which agrees with the values found in the unfolding of the interaction, where the highest values of F were observed (Table 3). The other genotypes could be classified in an intermediate group, with sensitivity for leaf temperature during water stress, but not as



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much. In relation to the total chlorophyll content (Figure 2), minor fluctuations were verified for genotypes BAG01, BAG47, BAG100, BAG102 and BAT477.

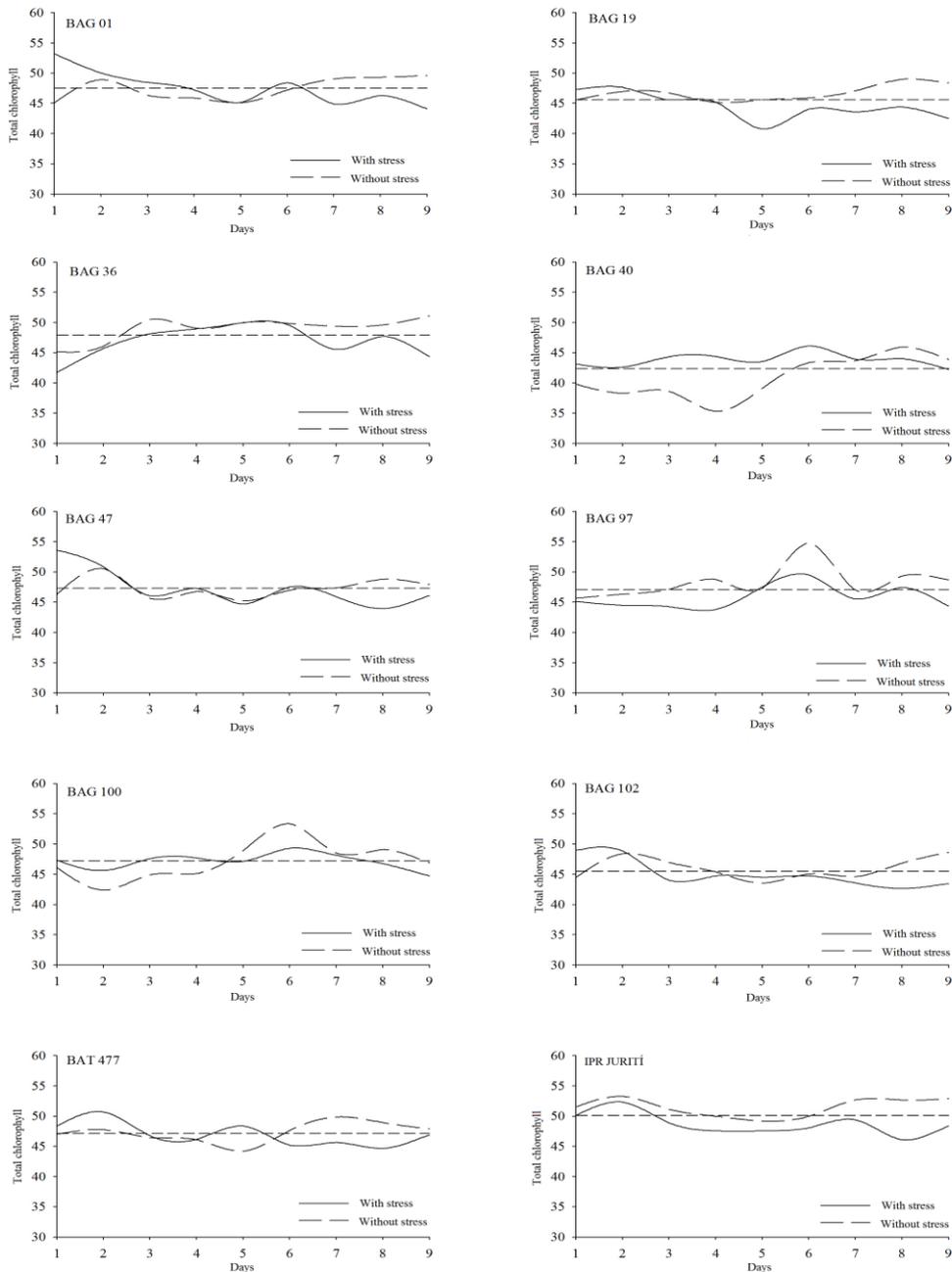


Figure 2. Values of total chlorophyll obtained daily in 10 bean genotypes grown under two water conditions (with and without water stress).



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Besides, the values between the chlorophyll content under both conditions were close to and below the overall average over the days. It is interesting to point out that genotype Jurití achieved the highest chlorophyll content when it was not submitted to stress over time. However, throughout the period of water stress, its chlorophyll content was below the overall average. The sensitivity of this genotype with leaf temperature and chlorophyll content and other agronomic traits agrees with the results found by Vale et al. (2012), which indicate its susceptibility to water stress.

Cultivars that show the least possible reduction in their yield when submitted to adverse conditions are regarded as drought tolerant. Thus, the results presented are promising, since they indicate genotypes that can be used in breeding programs with genetic potential for drought tolerance. Results also reveal that breeding programs should focus on genotypes with more rustic genetic constitution, usually those belonging to color groups (such as brindle, cream, sulfur, red, etc.), since they may be more tolerant to water stress. Further studies should be carried out to corroborate this hypothesis and assess tolerance to water deficit in different groups, at different levels of breeding or in different crop cycles, etc.

Conclusions

Water stress negatively affected various materials for most traits assessed and resulted in: i) increased leaf temperature; ii) reduced photosynthetic capacity; iii) reduced number of legumes per plant and; iv) reduced number of grains per legume. Genotypes BAG40, BAG100 and BAG102 can be considered promising for tolerance to water stress, since they were insensitive for most traits investigated, especially those related to productivity.

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