

Photosynthetic performance in jack bean [*Canavalia ensiformis* (L.) D.C.] under drought and after rehydration

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The effects of drought and rehydration on *Canavalia ensiformis* (L.) D.C. (jack bean) plants were evaluated using the following gas exchange parameters: net carbon assimilation rate (A), stomatal conductance (g_s), C_i/C_a ratio and transpiration rate (E); chlorophyll a fluorescence: F_v/F_m and F_v/F_0 ratio. The plants were cultivated under greenhouse conditions and after 30 days from the emergence, irrigation was suspended in the plants submitted to drought, to obtain the following predawn leaf water potential (Ψ_{pd}): -0.40 MPa (control), -1.00 MPa (moderate drought) and -2.30 MPa (severe drought). Afterwards, the gas exchange and fluorescence analysis were initiated, and 24 h after rehydration the same analyses were repeated. The A , E , g_s and C_i/C_a values decreased significantly under both drought treatments, without however changing the F_v/F_m and F_v/F_0 values. The gas exchange parameters recovered after rehydration. It seems that drought affected photosynthesis by stomatal inhibition, as shown by the decreased g_s and C_i/C_a values, besides the maintenance of PSII photochemical efficiency. The recovery of gas exchange after rehydration could be due to plant protection mechanisms.

Key words: fluorescence, moderate drought, photosynthesis, severe drought, stomatal conductance.

Desempenho fotossintético de feijão-de-porco [*Canavalia ensiformis* (L.) D.C.] sob déficit hídrico e após reidratação: Os efeitos do déficit hídrico e posterior reidratação foram avaliados em *Canavalia ensiformis* (L.) D.C., mediante parâmetros de trocas gasosas: fotossíntese (A), condutância estomática (g_s), razão C_i/C_a e transpiração (E); fluorescência da clorofila a : razão F_v/F_m e F_v/F_0 . As plantas cresceram em casa de vegetação e após 30 dias da emergência, suspendeu-se a irrigação naquelas submetidas ao déficit hídrico, obtendo-se os seguintes potenciais hídricos na antemãhã (Ψ_{am}): -0,40 MPa (controle), -1,00 MPa (déficit hídrico moderado) e -2,30 MPa (déficit hídrico severo). Após, foram realizadas as análises de trocas gasosas e de fluorescência. As plantas foram reidratadas e, após um período de 24 h, as mesmas análises foram repetidas. Os regimes de déficit hídrico provocaram decréscimos significativos em A , E , g_s e na razão C_i/C_a ; contudo, não alteraram as razões F_v/F_m e F_v/F_0 . Após a reidratação houve o restabelecimento das trocas gasosas. Conclui-se que o déficit hídrico afetou negativamente a fotossíntese, mediante uma limitação estomática, o que se confirma pelos decréscimos em g_s , na razão C_i/C_a e na manutenção da eficiência fotoquímica do FS II. Provavelmente, mecanismos de proteção tenham sido responsáveis pelo restabelecimento das trocas gasosas após a reidratação.

Palavras-chave: condutância estomática, déficit hídrico moderado, déficit hídrico severo, fluorescência, fotossíntese.

During centuries, the jack bean [*Canavalia ensiformis* (L.) D.C.] legume has been used by local inhabitants of the southwest United States, Central America, Mexico, Brazil, Peru, Equator and of the west of India. The great adaptability of *C. ensiformis* to adverse conditions, mainly soil related,

has been of great relevance for the high protein production in regions inept for agriculture. Besides the grains being a good source of protein and its use as livestock feed, this plant is also used in soil recovery in several countries (Lynd and Anzman, 1989).

For most plants, drought is one of the factors that most limits photosynthesis (García-Plazaola and Becerril, 2000). The intensity of water deficit is commonly evaluated by the leaf water potential (Ψ_w). Values of -0.9; -1.5 and -1.3 MPa represented moderate drought for bean (Cornic et al., 1992), coffee (Da Matta et al., 1997) and tomato (Haupt-Herting et al., 2001), respectively. The values of -2.7 MPa for coffee (Da Matta et al., 1997) and -1.8 MPa for tomato (Haupt-Herting et al., 2001), were assumed to be severe drought.

The stomatal closure is among the first responses to the water stress, and is assumed to be the main cause of impaired photosynthesis induced by drought, since the stomatal closure limits CO_2 availability to the mesophyll (Chaves, 1991). In view of this, a decrease in net photosynthesis under drought depends more on the availability of CO_2 in the chloroplast than of leaf water potential (Sharkey, 1990). This fact can be interpreted as a direct adjustment of photosynthesis to CO_2 availability, which acts by regulating the activity of Rubisco (Perchorowicz and Jensen, 1983).

Under natural conditions drought usually occurs in association with high temperatures and high irradiance (Pereira and Chaves, 1993), resulting in photoinhibition of

photosynthesis, characterized by a decrease in the PSII photochemistry efficiency. Certainly, PSII is the main target of photoinhibitory damage (Barber and Anderson, 1992), although Genty et al. (1987) and Havaux (1992) have inferred that drought has little effect on PSII functioning. Though an evaluation of photosynthetic performance, the aim of the present work was to investigate the effect of drought and rehydration in jack bean plants.

The experiment was carried out at the Plant Physiology Department of the State University of Campinas (UNICAMP), SP, Brazil (22°54'S and 47°05'W), from September to October 2002, in the greenhouse under natural light and temperature conditions. Seeds of jack bean [*Canavalia ensiformis* (L.) D.C.] were sown in trays filled with vermiculite. After emergence the seedlings were inoculated by immersing roots in a suspension of previously selected *Rizobium*, and then transferred to 5 L polyethylene pots with vermiculite as substrate. The plants were supplied with Hoagland and Arnon (1950) N-deficient nutrient solution twice weekly, and water as required. Thirty days after emergence, watering was suspended for the first lot of 15 plants, in order to induce severe drought (SD). Seven days later, the same procedure

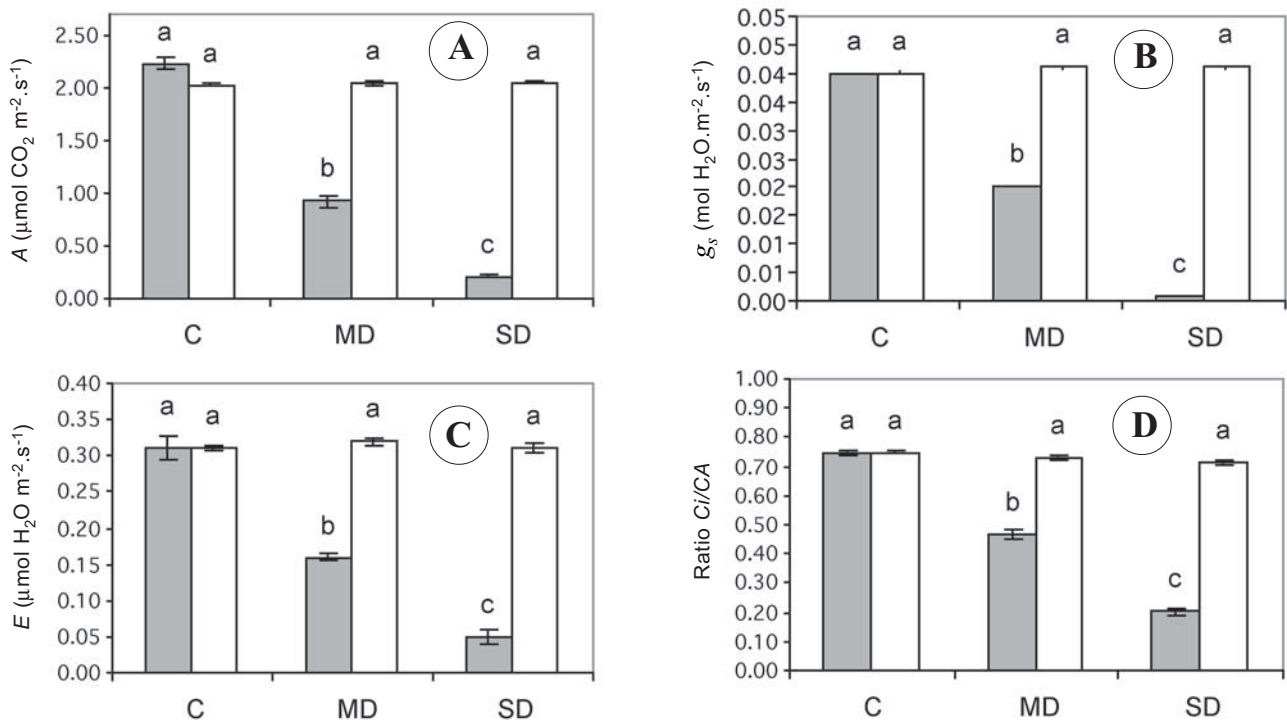


Figure 1. Net carbon assimilation rate - A (A), stomatal conductance - g_s (B), transpiration rate - E (C) and C_i/C_a ratio (D) in *C. ensiformis* plants under drought and rehydration. Control = C: $\Psi_{pd} = -0.40$ MPa; moderate drought = MD: $\Psi_{pd} = -1.0$ MPa; severe drought = SD: $\Psi_{pd} = -2.30$ MPa. Different small letters represent statistical significance between means for each treatment ($p \leq 0.05$, Duncan's test). The bar represents standard error of five replicates.

Table 1. F_v/F_m and F_v/F_0 ratios in *C. ensiformis* plants under drought and rehydration. Control: $\Psi_{pd} = -0.40$ MPa; moderate drought: $\Psi_{pd} = -1.0$ MPa; severe drought: $\Psi_{pd} = -2.30$ MPa.

Treatment	F_v/F_m^a	F_v/F_0
Control	0.78 ± 0.013 a	3.8 ± 0.31 a
Moderate drought	0.81 ± 0.006 a	4.3 ± 0.18 a
Severe drought	0.80 ± 0.006 a	4.2 ± 0.16 a

^a Each value represents the mean ± Standard Error of five replicates. Different small letters represent statistical significance between means for each treatment ($p \leq 0.05$, Duncan's test).

was carried out for another lot of 15 plants, to induce moderate drought (MD). By this means, 26 and 19 days after suspending water, the following predawn leaf water potentials (Ψ_{pd}) were obtained: -1.0 MPa (MD) and -2.30 MPa (SD). In the control plants (C) which remained watered, the value of Ψ_{pd} was -0.4 MPa.

The predawn leaf water potential was evaluated by means of pressure chamber (PMS). The gas exchange parameters, net carbon assimilation rate (A), stomatal conductance (g_s) and transpiration rate (E), were measured under natural light at about 8:30 am (temperature: 29°C; PAR: 801 $\mu\text{mol}\cdot\text{m}^{-2}\cdot\text{s}^{-1}$), using a portable open-system infrared gas analyzer (LCA4, Analytical Development Company). The internal CO_2 concentration (C_i) and the ambient CO_2 concentration (C_a) values were used in order to calculate the C_i/C_a ratio. The chlorophyll a fluorescence, in the form of F_v/F_m and F_v/F_0 ratios, was measured at room temperature using a portable mini-pulse amplitude modulation fluorometer (MiniPAM, Walz), in dark-adapted leaves for 30 min. All the analyses were carried using the middle foliole of the third expanded trifoliate leaf from the apex. The plants were distributed in a completely randomized layout, with five replicates. Each experimental plot was composed of one plant per tray. Statistical significance of mean differences were analyzed by Duncan's test, at $P \leq 0.05$.

All the gas exchange parameters decreased under both drought treatments, but they recovered within 24 h after rehydration (figure 1). The observed decline in g_s and E (figure 1), indicates that drought caused stomatal inhibition with negative reflexes on the photosynthetic CO_2 uptake and transpiration rate. The decrease in the C_i/C_a ratio in the present of drought is consistent with this interpretation (figure 1), since increases in this ratio demonstrate decreased uptake of CO_2 due to non-stomatal inhibition of photosynthesis. According to Baker (1993), there is a direct relationship between the reduction of intercellular CO_2 concentration, due to stomatal closure, and decreases in CO_2 assimilation.

Although drought impaired photosynthesis it did not damage the photosynthetic apparatus, as demonstrated by the F_v/F_m and F_v/F_0 ratios (table 1). F_v/F_m values were 0.78, 0.81 and 0.80 in control, MD and SD treatments, respectively, while the F_v/F_0 ratio ranged from 3.8 to 4.3. According to Flexas et al. (2002) the F_v/F_m ratio in plants of grapevine under drought remained around 0.8, as in the present work in spite of significant decreases in the stomatal conductance.

The reduction in photosynthesis in plants of *Myracrodruon urundeuva* under drought occurred mainly because of stomatal closure rather than damage to PSII (Queiroz et al., 2002). In coffee plants under drought conditions partial maintenance of the quantum yield of PSII was observed in spite of photosynthesis suppression. In this case, some processes could contribute to the maintenance of electron flow, such as the Mehler reaction and the photorespiratory process (Lima et al., 2002). In this context, the fall of net carbon assimilation rate caused by drought was not accompanied by decreases in F_v/F_m , showing that PSII is resistant to drought, as has been shown for the cotton plant (Genty et al., 1987), coffee tree (Da Matta et al., 1997) and *Casuarina equisetifolia* (Sanchez-Rodriguez, 1997). However, photoinhibitory damages to PSII were found by He et al. (1995), who verified degradation of the D1 and D2 proteins (mainly D2) in plants under drought. In olive trees, the photosynthetic apparatus was resistant to both weak and moderate drought, stomatal closure being the main factor limiting photosynthesis. However, under severe drought, decreases in the photosynthesis were attributed to photoinhibitory phenomena associated with an over-excitation of PSII (Angelopoulos et al., 1996).

In view of this decline in photosynthesis, less photochemical energy would be spent on CO_2 assimilation. Consequently, the photochemical energy would need to be consumed by alternative pathways. One possibility is the loss of energy by heat (Krause and Weis, 1991). Data on non-photochemical quenching (NPQ) gives information on the fraction of luminous energy lost as heat (Lima et al., 2002). In this regard, Casper et al. (1993) suggest that under drought, protection mechanisms such as the zeaxanthine cycle could be active and thereby prevent damage to the photosynthetic apparatus.

Considering the results of this study, we conclude that the two drought treatments negatively affected photosynthesis and all gas exchange parameters, and these could be re-established within 24 h after rehydration. The inhibitory effect of drought on photosynthesis could be attributed to some

stomatal limitation. Neither of the two drought-imposed treatments affected the maintenance of PSII photochemistry efficiency, which could be attributed to protection mechanisms.

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