

MECHANISMS OF DROUGHT TOLERANCE IN THE C3 DECIDUOUS SHRUB, *Ipomoea carnea*

MECANISMOS DE TOLERANCIA A LA SEQUIA EN EL ARBUSTO C3 CADUCIFOLIO *Ipomoea carnea*

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ABSTRACT

Ipomoea carnea Jacq. is a C3 perennial deciduous shrub commonly found in the semi-arid zones of northwestern Venezuela. In order to gain knowledge about the possible mechanisms of drought resistance in this species, water relations and gas exchange were studied in plants growing in the field and under greenhouse conditions. Morning water potential was relatively high in the field throughout the year (minimal value, -2.27 MPa). Leaf conductance in the field reached a maximum of 48 mmol m⁻² s⁻¹ and was undetectable by a porometer during months with little or no rainfall. Under greenhouse conditions, leaf conductance in well-watered plants was higher (370 mmol m⁻² s⁻¹), diminishing 98% after 10 days of water deficit, when no photosynthesis, but respiration, was measured. During the first 20 days of drought there was no significant decrease in leaf turgor and leaf water content decreased only 19%. Carboxylation efficiency (dA/dCi) remained unchanged within 12 days of drought. In greenhouse plants subjected to water deficit xylem water potential, stomatal conductance and photosynthetic rate reached control values within three days after re-watering. In the field as well as in greenhouse samples, soluble protein content on a leaf area basis diminished with drought, this decrease being significant in the greenhouse experiments only after 31 days of drought. The initial decrease in assimilation rate was apparently not due to either an increase in relative stomatal limitation or a decrease in carboxylation efficiency. No osmotic adjustment was observed during drought even though an increase by dry matter was found in sugars. The relative tolerance of leaves of *I. carnea* to drought may reside mainly in a strict stomatal control that prevents dehydration and allows leaves to resume a high metabolic activity very shortly after rewatering, all of which is important in the habitat of this species, characterized by unpredictable rains.

KEY WORDS: drought tolerance, *Ipomoea carnea*, osmotic adjustment, photosynthesis

RESUMEN

Ipomoea carnea Jacq. es un arbusto C3 perenne caducifolio que se encuentra comunmente en las zonas semiáridas del noroeste de Venezuela. Para conocer los posibles mecanismos de resistencia a la sequía en esta especie, se estudiaron, tanto en campo como en invernadero, las relaciones hídricas y el intercambio gaseoso. El potencial hídrico matutino medido en el campo fue relativamente alto durante todo el año (con un mínimo de -2.27 MPa). La conductancia estomática en el campo alcanzó un máximo de 48 mmol m⁻² s⁻¹ y se hizo tan baja en los meses con poca o ninguna lluvia que no pudo ser medida con un porómetro. En el invernadero, la conductancia en plantas regadas fue mayor (370 mmol m⁻² s⁻¹), disminuyendo en un 98% cuando se sometió a las plantas a déficit hídrico por 10 días; en este momento no se detectó fotosíntesis sino respiración. Después de 20 días de sequía no hubo una disminución significativa en la turgencia y el contenido de agua foliar disminuyó sólo en 19%. La eficiencia de carboxilación (dA/dCi) permaneció constante durante 12 días de sequía. En el invernadero, el potencial hídrico del xilema en plantas en sequía, la conductancia estomática y la tasa fotosintética alcanzaron los valores del control dentro de los tres primeros días de re-irrigación. En muestras tanto de campo como de invernadero, el contenido de proteína soluble disminuyó con la sequía, siendo esta disminución significativa en el invernadero sólo después de 31 días de sequía. El descenso inicial en la tasa de asimilación no pareció deberse ni a un aumento en la limitación estomática relativa ni a una disminución en la eficiencia de carboxilación. No se observó ajuste osmótico durante la sequía, aun cuando se encontró un incremento en azúcares en base a peso seco. La tolerancia relativa de las hojas de *I. carnea* a la sequía debe residir principalmente en un control estomático estricto que impide la deshidratación y le permite a las hojas alcanzar de nuevo una alta actividad metabólica muy poco después de la reirrigación, todo lo cual es importante en el habitat de esta especie, caracterizado por lluvias impredecibles.

PALABRAS CLAVE: tolerancia a la sequía, *Ipomoea carnea*, ajuste osmótico, fotosíntesis

INTRODUCTION

Ipomoea carnea Jacq. (Convolvulaceae) is a perennial deciduous shrub growing abundantly under full sun exposure in the thorn scrubs and open fields of northwestern Venezuela. It presents the C3 pathway of carbon fixation (Fonseca and Herrera 1983, Herrera and Cuberos 1990). Its leaves are large (c. 1 dm²) and remain green for as long as two months after the end of rainy periods.

Setting aside the typical avoiders, which lose their leaves early during drought, other responses to drought imply some form of tolerance to varying lengths of treatment. A diversity of mechanisms related to water relations and leaf gas exchange have been detected in plants in response to drought. In *Amaranthus palmeri* an osmotic adjustment of 0.35 MPa apparently allowed photosynthetic rate to become zero at a water potential 1.35 MPa lower than that for turgor loss in irrigated plants (Ehleringer 1983). On the other hand, plants of *Macroptilium atropurpureum* (Ludlow and Ibaraki 1979) retained an elevated water potential after prolonged periods of water deficit, while stomata closed early during the treatment.

Turner and Burch (1983) proposed that drought resistance may be divided into escape, tolerance at a high water status and tolerance at a low water status. Tolerance can be defined, within this context, as the capability to maintain carbon gain under water deficit. This classification, which is the product of extensive research on agricultural plants, may be too rigid for wild plants growing in their natural environments, for which responses may lie on an interphase difficult to define between escape and tolerance (Mooney 1981, Hinckley *et al* 1983).

The question of what causes a decline in assimilation rate during drought, whether stomatal closure or reduced mesophyll activity, is still a matter of much debate (Chaves and Pereira 1992) which has been partly clarified in experiments under controlled conditions but becomes obscured by the interaction of external factors under field conditions (Chaves 1991). The present study was done on the water relations and gas exchange of *I. carnea* in order to gain knowledge on the mechanisms of

drought tolerance in this species. We were particularly interested in looking for osmotic adjustment as a possible mechanism of water conservation. Another possible explanation for the observed leaf maintainance during drought could be the operation of a set of mechanisms that reduce water loss: increased stomatal resistance, water absorption and hydraulic conductivity (Turner and Burch 1983).

MATERIALS AND METHODS

Field studies were conducted in a thorn scrub near the city of Coro, Estado Falcón (Venezuela), located at 11°25'N, 69°36'W, at c. 20 m. This scrub belongs to the Subregion C.1., Lara-Falcón Hills System (Huber and Alarcón 1988). It is composed of a sparse evergreen and deciduous tree stratum, with *Prosopis juliflora*, *Cercidium praecox* and *Capparis* spp. as dominant species; an arbustive stratum dominated by *I. carnea* with *Jatropha gossypifolia* as a frequent constituent, and a herb stratum dominated by *Alternanthera crucis*. Columnar and herbaceous cacti are abundant.

Measurements were done during daily courses on March, June, August, September, October, November and December 1977, and February and March 1978. Microclimatic and physiological parameters were measured each time as detailed below. Further studies conducted during 1985-1991 determined that the aspect of the vegetation and leaf responses continued to be essentially similar to the 1977-78 study. Between 6 and 10 leaves were tagged every field trip in order to follow their duration.

One adult individual of average height (c. 1.5 m) was dug up and the length of its tap-root measured.

For experiments under greenhouse conditions, cuttings from the field were rooted in water and grown in 8-L pots filled with commercial garden soil and watered daily for at least 2 months before the beginning of the experiments.

Plants were grown in a greenhouse in Caracas (c. 1000 m). Temperature ranged from 22°C (0300h) to 35°C (1400h). Photosynthetic photon flux

density (PPFD, 400-700 nm) was $2000 \mu\text{mol m}^{-2} \text{s}^{-1}$ (instant highest) and $17.4 \pm 0.9 \text{ mol m}^{-2} \text{ d}^{-1}$ (total daily). Several independent experiments were carried out.

Ten plants were used in each experiment. The experiments consisted of withholding irrigation for a length of time determined by the survival of fully expanded leaves tagged at the beginning of each experiment. Leaves lasted for a minimum of 30 days under drought. All measurements were done on three of these leaves.

PPFD was measured with a LI-COR quantum sensor mod. 190S connected to a LI-COR meter mod. LI-185 (Nebraska, USA). Air and leaf temperature (T) were measured with YSI thermistors and telethermometers (Yellow Spring Instruments Co., Ohio, USA). Soil matric potential (Ψ_m) was determined by dew-point psychrometry using a C-52 chamber connected to an HR33T microvoltmeter (Wescor Inc., Utah, USA) in samples taken at 1600h with a cork borer from a depth of 10 cm. Soil water content in the field was determined in 4 samples taken at 30-50 cm depth, placed in airtight metallic containers, weighed, dried at 100°C for 72 hours and weighed.

Leaf diffusive resistance to water vapour was measured on the abaxial face of the same leaves throughout the day in field studies with a transient porometer mod. LI-60 connected to an LI-205 cuvette calibrated before each field trip, being capable of reading resistances between 0.59 and 44.4 s cm^{-1} (conductances between 678 and $9.0 \text{ mmol m}^{-2} \text{ s}^{-1}$). Values of resistance were converted to conductance (g) following Percy *et al.* (1989).

Water vapour and CO_2 exchange (A) were measured at 1100h in greenhouse experiments using a portable system consisting of an LCA2 infrared gas analyzer connected to a PLC(B) assimilation chamber and an ASU(MF) air supply unit, all from the Analytical Development Co. Ltd. (Hoddesdon, U.K.). Previous determinations showed that maximal rates of A were obtained at the time of day indicated above. Data were collected manually and processed with a custom-made programme according to the equations given by Long and Hällgren (1985).

Response curves of assimilation to intercellular CO_2 concentration (C_i) were done passing part of a compressed air stream at supra-atmospheric CO_2 concentration through a soda-lime column, mixing it with the same stream unfiltered and feeding the mixture to the PLC chamber at the desired concentrations. Measurements were done under a PPFD of $600 \mu\text{mol m}^{-2} \text{ s}^{-1}$ provided by mercury vapour lamps. Leaf chamber temperature was maintained at $27 \pm 1^\circ\text{C}$ by means of a fan directed toward it. Relative stomatal limitation was calculated after Farquhar and Sharkey (1982).

Xylem water potential (Ψ) was measured with a pressure bomb (PMS, Oregon, U.S.A.) at 0600h-1800h (field) or 0800h (greenhouse) in petioles with leaves attached. Also, in greenhouse experiments leaf water potential was measured by psychrometry as above on leaf disks of 0.2 cm^2 taken at 0900h and maintained for 30 min in the chamber. Each value is the mean of measurements done on one disk from three different leaves.

Osmotic potential (Ψ_s) was measured in the same disks as for leaf water potential frozen in liquid nitrogen and defrosted; measurements were done by psychrometry as above, leaving samples for 10 min in the chamber. Turgor potential (P) was calculated as the difference between osmotic and water potential with no correction for apoplastic water contamination.

Pressure-volume (P-V) curves were done after Wilson *et al.* (1979, 1980) on leaves resaturated for 24h at room temperature in the dark and under a water-vapour saturated atmosphere. Relative water content was calculated as $\text{RWC} = (\text{FW} - \text{DW}) / (\text{TW} - \text{DW}) \times 100$, where FW=fresh weight after rehydration, DW=oven-dried weight (60°C for 3 days) and TW=turgid weight (obtained from the interpolation of FW v. pressure at pressure=0).

Leaf water content (LWC) was determined on leaf disks taken at 1100h from greenhouse plants, weighed, dried at 60°C for 3 days and weighed. Stem water content (SWC) was determined likewise on stem segments of known length and diameter in an independent experiment in which the entire foliage of 3 watered plants was collected to determine area and LWC.

Chlorophyll content was determined after Bruinsma (1963) in acetone extracts of the pellet obtained from centrifuged leaf aqueous extracts.

Soluble protein was determined after Bradford (1976) in the trichloroacetic acid (10%) precipitate of unbuffered aqueous extracts of leaf disks taken at 1100h. No protein precipitation was evident during the extraction procedure.

Free amino acid content was measured by the method of Moore and Stein (1948) using glycine as a standard in the supernatant of the aqueous extracts centrifuged for protein determination.

Soluble sugars and starch contents were determined as glucose equivalents after Dubois *et al.* (1956) in samples collected at 0800-0900h, dried at 60°C and processed by the method of McCready *et al.* (1950).

The relative contribution of soluble metabolites to Ψ_s was calculated using the van't Hoff relation assuming that these solutes could be acting as osmotica inside the vacuole.

Values presented are means of triplicates and their standard errors. Significance was assessed by either Student's t-test or ANOVA at $p < 0.05$.

RESULTS

Field studies

The pattern of precipitation for the year of this study followed approximately the mean pattern for 30 years, although rain regime is erratic and unpredictable, as shown by the fact that on the same month either the maximum or no rainfall occurred (Figure 1). During the field studies rainfall was 52% less than for the period 1941-1970.

Morning Ψ followed very roughly the trend in precipitation but a much closer relationship was found between morning Ψ and soil water content (Figure 2a and b). This was not the case of minimum Ψ , since the lowest values corresponded to both plants with high g on a wetter soil (November 1977) and plants with low g on a drier soil (September 1977; Figure 2c). Therefore, morning Ψ was a

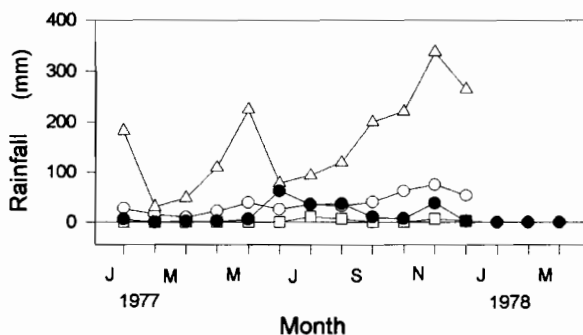


Figure 1. Precipitation in the field site for 1941-70 (triangles, maximum; open circles, mean; squares, minimum), and 1977 and the first months of 1978 (closed circles). Total rainfall was 430 mm during 1941-70 and 205 mm in 1977.

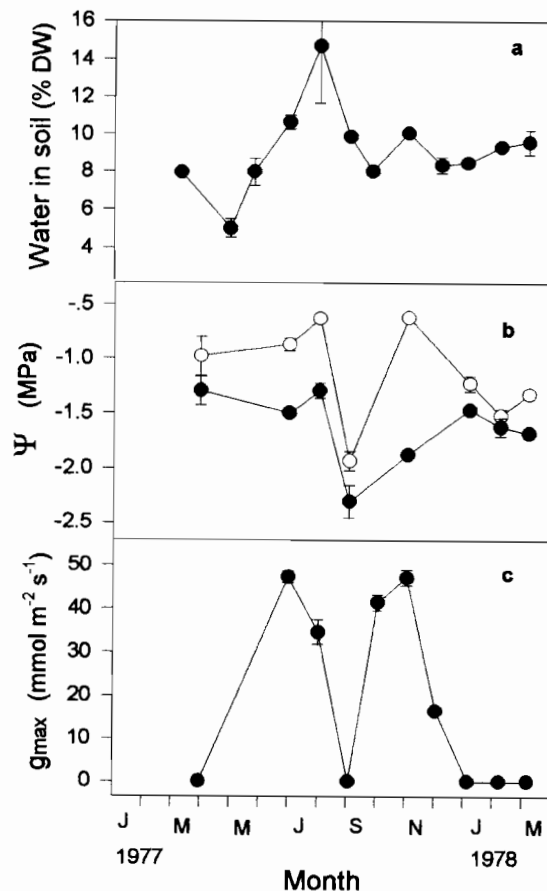


Figure 2. Changes in: a, water content in the soil; b, Ψ in the field during 1977-78 (open circles, morning; closed circles, minima), and c, maximal g . Vertical bars are one standard error.

better indicator of water status. Evidently, a higher conductance and, presumably, transpiration rate may account for the lower minimum Ψ of plants growing on wetter soil. During April and May 1977 plants did not have any foliage.

Radiation and leaf temperature were high during the entire period of study, typical daily courses resembling those in July and September (Figure 3).

In plants measured in the field morning Ψ diminished 1.3 MPa between July and September (Figure 4a). Conductance showed a maximum of 34.6 mmol m⁻² s⁻¹ during July but could not be detected with the porometer (i.e. denoted as zero on the graph) in September during the whole day (Figure 4b). The highest value of *g* detected during the year of study was 48.3 mmol m⁻² s⁻¹.

The tap-root in the individual excavated was longer than 1.5 m.

Chlorophyll content showed no significant variations throughout the year but soluble protein content in December was 41% of the value in both June and November (Table 1).

Laboratory studies

Leaves subjected to drought in the greenhouse remained green and attached to stems for approximately 30 days. Shortly after, leaf shedding began. The response of Ψ and *g* to drought was very similar between independent experiments.

A decrease in Ψ was found from the start of the dry period; five days into the dry cycle and until its end, Ψ was higher than Ψ_m , the latter decreasing to a minimum of -3.46 MPa whereas minimum Ψ was

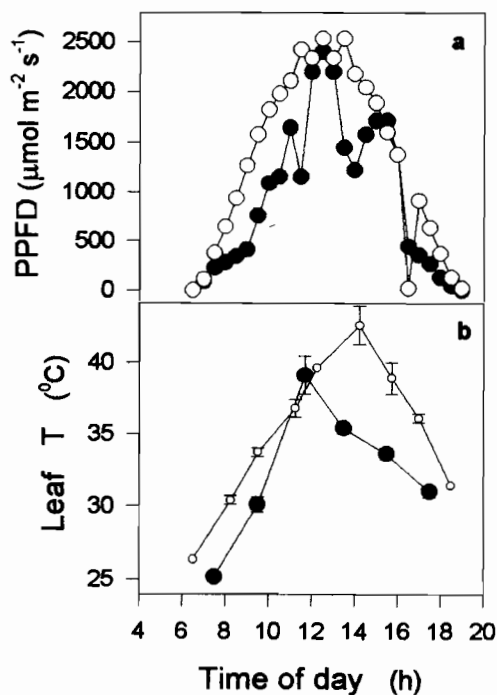


Figure 3. Daily courses in the field site of: a, PPFD, and b, leaf temperature in July (open circles) and September (closed circles). Vertical bars are one standard error.

Table 1. Leaf chlorophyll and soluble protein contents in field plants measured during 1977.

MONTH	Chlorophyll (μg cm ⁻²)	Soluble protein (μg cm ⁻²)
Jun.	31.1±0.5	176±13
Jul.	27.5±0.5	90±3
Sep.	21.3±0.7	-
Oct.	29.4±0.5	110±8
Nov.	25.2±0.7	176±11
Dec.	27.6±0.3	72±3

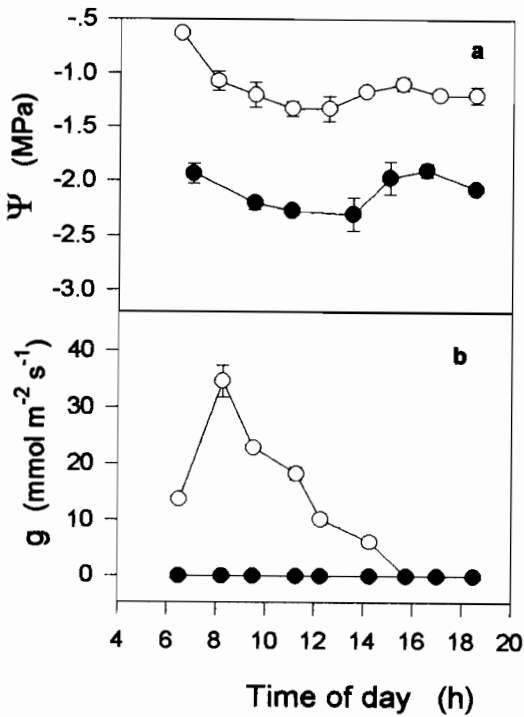


Figure 4. Daily courses in plants growing in the field during July (open circles) and September (closed circles). a, Ψ ; b,g. Vertical bars are one standard error.

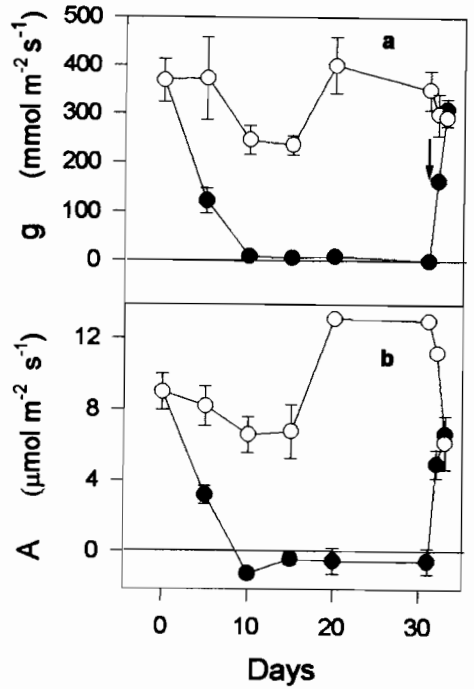


Figure 6. Changes with drought for plants growing in the greenhouse in: a, g and b, A. Closed circles, droughted plants; open circles, control plants. Vertical bars are one standard error. The arrow indicates the moment of rewatering.

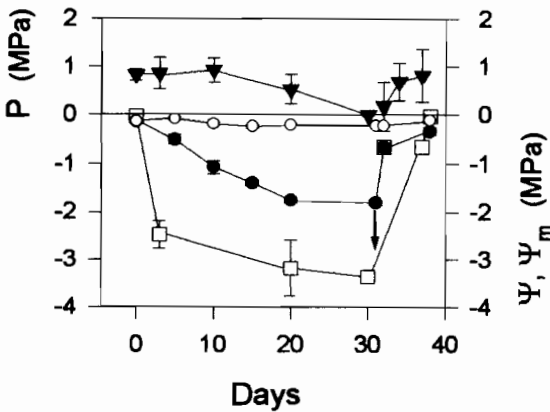


Figure 5. Changes with drought in Ψ (closed circles, droughted plants; open circles, control plants), P (triangles) and Ψ_m (squares) for plants grown in the greenhouse. Vertical bars are one standard error. The arrow indicates the time of rewatering.

-1.80 MPa (Figure 5). A significant decrease in P was found only after 20 days of drought. Recovery of both Ψ and P after rewatering was rapid, taking place within 2 days.

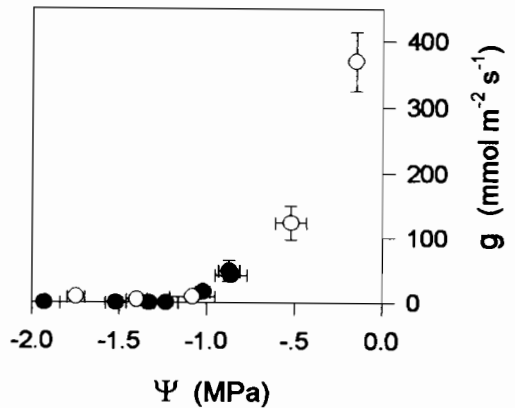


Figure 7. Changes in g with Ψ for plants growing in the greenhouse (open circles) and in the field (closed circles). Vertical and horizontal bars are one standard error.

Stomatal closure started at the beginning of a drought experiment from an initial value of g of $370 \text{ mmol m}^{-2} \text{ s}^{-1}$ and was practically completed by day 10 at $9 \text{ mmol m}^{-2} \text{ s}^{-1}$ and a morning Ψ of -1.08

MPa (Figure 6a). Conductance increased to control values 24 h after re-irrigation. The response of A to drought showed a similar trend to g: no assimilation, but respiration, was measured 10 days after the beginning of drought and A rose to nearly control values 24h after rewatering (Figure 6b). No evidence was found of a threshold value of Ψ for a decrease in g and the same was true for measurements in the field (Figure 7).

Leaf water content on an area basis remained unchanged during the first 21 days of drought, showing a decrease of 19% after 31 days of drought (Figure 8a), whereas on a DW basis a larger decrease (48%) was found (Figure 8b). This could be explained by an increase of 34% in DW/area after 31 days of drought (Table 2). A significant decrease of 22% in SWC on a DW basis was noticed. A SWC of 756 ± 37 mg water (cm³ stem volume)⁻¹ was measured on watered plants. The ratio stem volume/leaf area in well-watered plants was 0.040 ± 0.000 cm³ cm⁻². Rewatering brought about a recovery of LWC to control values.

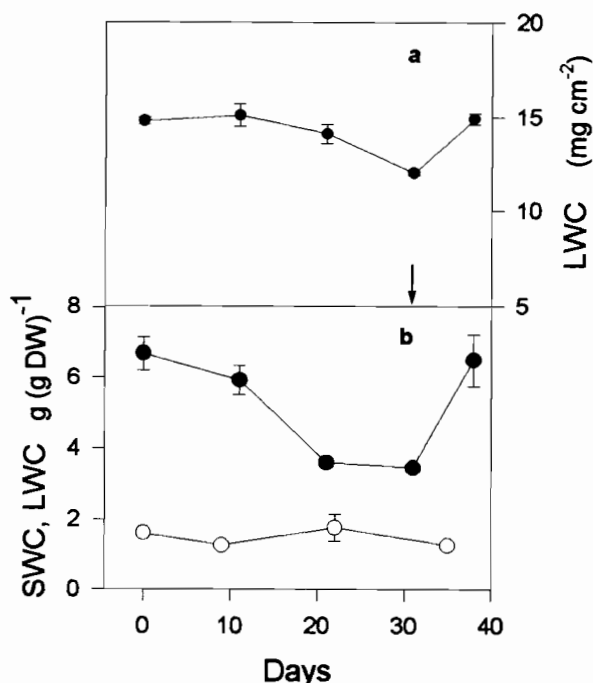


Figure 8. Changes with drought in greenhouse plants in a, LWC on an area basis and b, LWC (closed circles) and SWC (open circles) on a DW basis. Vertical bars are one standard error. The arrow indicates the time of rewatering. Rewatered values of SWC were not measured.

Table 2. Parameters measured after on month of drought in plants grown in the greenhouse. Values within brackets indicate the percent increase or decrease relative to day 0 of drought.

PARAMETER	
Ψ (MPa)	-1.80±0.1 (-88)
Ψ_s (MPa)	-1.79±0.2 (-46)
LWC (mg cm ⁻²)	14.9±0.5 (-19)
DW/area (mg cm ⁻²)	3.5±0.1 (34)
Total amino acids (µmol g ⁻¹ DW)	73±5 (-21)
Sugars (µmol glucose g ⁻¹ DW)	459±67 (53)
Starch (µmol glucose g ⁻¹ DW)	169±22 (-47)

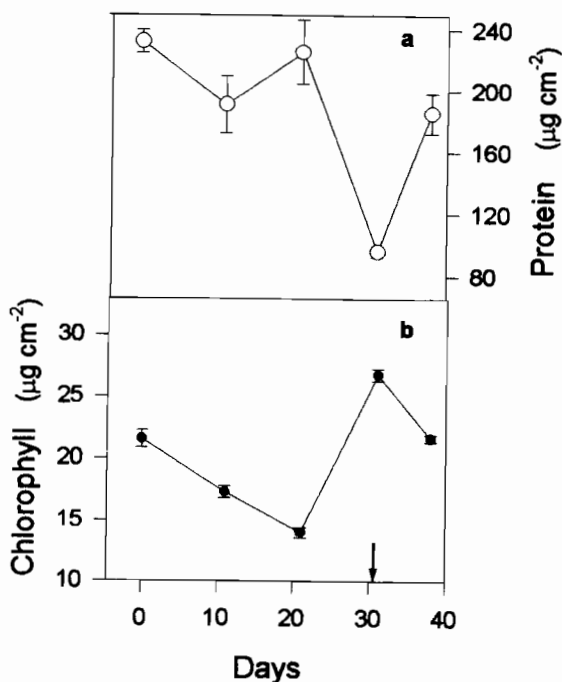


Figure 9. Changes with drought in: a, soluble protein and b, chlorophyll content. Vertical bars are one standard error. The arrow indicates the time of rewatering.

Leaf soluble protein content (on an area basis) decreased 59% after 31 days of drought but the changes between 0 and 21 days were not statistically significant (Figure 9a). The same consideration for LWC on a DW basis may hold here, in that the marked decrease in protein content may be an artifact due to tissue compaction. The corresponding decrease on a DW basis was 68%, therefore, 25% of the decrease in protein content could be attributed to tissue compaction. Chlorophyll content decreased 35% after 21 days of drought but an increase was measured between the beginning and the end of the dry treatment (Figure 9b); the latter could again be explained by tissue compaction. Rewatering provoked an increase in protein content to control values within 8 days, whereas chlorophyll content returned to control values.

In another independent experiment undertaken to examine the photosynthetic response to intercellular CO_2 concentration (A-Ci curves; Figure 10), no change in carboxylation efficiency

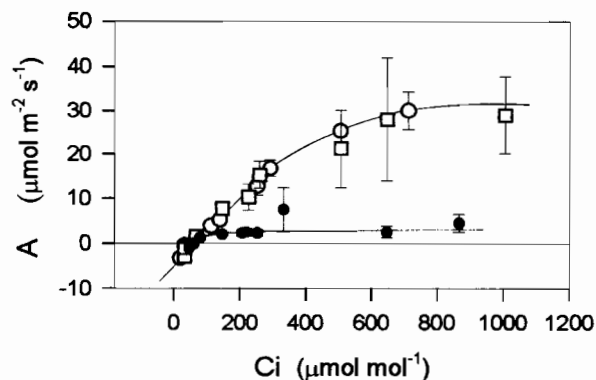


Figure 10. A-Ci curves at 0 (open circles), 9 (squares) and 12 (closed circles) days of drought in plants grown in the greenhouse. Vertical bars are one standard error. The values of r^2 for the linear portion of each curve are: 0.99 (day 0); 0.97 (day 9) and 0.99 (day 12).

(dA/dC_i) was detected during 12 days of drought. The larger variance in values of A for 9-days-stressed plants could be due to the occurrence of patchy stomatal closure, which may produce non-uniform changes in A and C_i (Downton *et al.* 1988). Relative stomatal limitation was 28% in watered plants and 27% in 9-days-droughted plants but could not be calculated in 12-days water-stressed plants since A at infinite conductance was on the plateau.

Soluble sugars accumulated during drought, whereas free amino acid and starch contents decreased (Table 2). The contents of soluble sugars, total amino acids and starch increased after rewatering (not shown).

The contribution of the different metabolic solutes to Ψ_s at the end of the dry treatment was calculated to be: amino acids, 2.3% and sugars, 2.1%.

Determinations of osmotic potential by P-V curves done on plants subjected to drought for 29 days showed that leaves of *I. carnea* were not capable of osmotic adjustment (Table 3). The comparison was made with irrigated plants measured after the drought cycle to avoid a developmental effect obscuring interpretation. Both osmotic potential and RWC at turgor loss decreased with drought. An increase in the volumetric elasticity modulus (ϵ) with drought relative to control was

Table 3. Water relations parameters from P-V curves done on droughted and control greenhouse plants after 29 days of drought. Ψ_0 , osmotic potential at turgor loss; RWC_0 , relative water content at turgor loss. The asterisk denotes significant differences (at $p < 0.05$) from a Student's t-test on independent samples.

PARAMETER	TREATMENT	
	control	droughted
Ψ_{100} (MPa)	-0.63±0.03	-0.63±0.05
Ψ_0 (MPa)	-0.83±0.04	-1.18±0.06*
RWC_0	0.96±0.06	0.84±0.04*
ϵ (MPa)	8.89±0.46	10.61±0.55*
C (MPa ⁻¹)	0.092±0.001	0.041±0.003*

found. Capacitance calculated as pressure/RWC on the initial portion of a P-V curve decreased with drought.

DISCUSSION

Leaves of plants of *I. carnea* growing in the field showed throughout the year of study changes in g and Ψ that followed the pattern of soil water content. This, together with the general aspect of the vegetation, allows us to affirm that plants measured during July 1977 were at a high water status, whereas those measured in September were at a low water status. The same leaves suffered drought for at least 40 days. Plants subjected to drought in the greenhouse responded to water deficit with an early stomatal closure which was completed within 10 days.

Minimum Ψ of plants with leaves in the greenhouse after 31 days of drought was similar to the minimal value in the field for plants with leaves after 40 days of drought, i.e. -1.80 versus -2.27 MPa. These values were higher than in other xerophytes. Minima of -5.0 MPa have been found in North American drought-deciduous shrubs (Smith and Nobel 1986) and of -8.0 MPa in *Alternanthera crucis* in the same ecosystem as *I. carnea* (Herrera *et al.* 1993).

Values of g measured in the field shortly after rainfall (maximum, 48.3 mmol m⁻² s⁻¹) were lower

than in greenhouse irrigated plants. Values higher than 400 mmol m⁻² s⁻¹ were measured in 1990 in the same ecosystem in plants with Ψ of -0.50 MPa (Tezara 1991). This could be due to higher soil water contents, but the author did not provide them. The steady-state porometer used in the present work had a low sensitivity at the lower range of resistance, as pointed out by Jones (1986).

The root system of *I. carnea*, albeit long (> 1.5 m), may not reach the water table. The occurrence of high values of Ψ under severe drought may imply that roots become isolated from the soil, as has been demonstrated in cacti (Nobel and Sanderson 1984). In the greenhouse Ψ was higher than Ψ_m after only a few days of drought. The results clearly suggest that droughted plants of *I. carnea* do not absorb water from the soil.

In *I. carnea* g was nil, and respiration instead of assimilation was measured, in greenhouse experiments at a Ψ of -1.08 MPa. In *Macroptilium atropurpureum*, a drought avoider, the corresponding value was -1.90 MPa (Ludlow *et al.* 1983). In *Cercidium microphyllum*, a drought-deciduous shrub, zero photosynthesis occurred only when Ψ had decreased to -3.00 MPa (Smith and Nobel 1986). On the contrary, in drought-tolerant grasses (Smith and Nobel 1986) and in *Alternanthera crucis* (Ríos 1993) g was zero at around -8.00 MPa. In *Macroptilium atropurpureum*, an early stomatal closure during

drought seems to contribute to the maintenance of a high Ψ (Ludlow and Ibaraki 1979).

The decrease in g followed a negative exponential from the beginning of drought, whereas Ψ diminished in a manner approximately linear for the first 20 days of drought. The decrease in g followed fairly closely that in Ψ_m , which suggests the possible existence of a root signal eliciting stomatal closure well before Ψ decreases strongly (Davies *et al.* 1990).

Under greenhouse conditions, A diminished with drought following a trend close to that of g . There was no evidence for a threshold value of Ψ eliciting stomatal closure or a decrease in A . Such a threshold Ψ has been found in other species subjected to water deficit, such as *Manihot esculenta* (Ike and Thurtell 1981). Recovery of both g and Ψ was fast, in contrast with examples of a lag after rewatering (Ike and Thurtell 1981).

The reduction in A followed a similar trend to the decrease in g . Nevertheless, relative stomatal limitation remained unchanged during 9 days of drought, although g had diminished 98% after 10 days of drought. Soluble protein content, as an indirect measure of Rubisco content, did not decrease significantly during 21 days of drought. Also, no decrease in dA/dCi was found during the first 12 days of drought. No parallel decrease in dA/dCi and Rubisco content was found in sunflower plants subjected to drought; rather, a diminution in A could be correlated with a decrease in RubP (Tezara and Lawlor 1995). Carbon assimilation may diminish to values close to zero without any significant decline in mesophyll photosynthetic capacity (Chaves 1991).

Soluble protein content decreased in droughted plants in the laboratory to a similar degree and in the same range of Ψ as in plants growing in the field, but results suggest that a significant part of the drastic loss of protein can be adjudicated to tissue compactation. Chlorophyll was maintained in field as well as greenhouse plants.

Our results suggest that photosynthetic limitation may be divided in two stages: an initial one, with a decrease in A not explained by changes

in either relative stomatal limitation, dA/dCi or soluble protein content and a later one, in which stomatal closure is completed and soluble protein content may be markedly reduced. At the first stage, parameters not measured in this work, such as RubP content, could help explain the decrease in A .

The rapid stomatal closure observed in greenhouse experiments may have prevented a drastic reduction in LWC (on an area basis). No evidence was found of osmotic adjustment during drought. The observed reduction in Ψ_0 can be interpreted as the occurrence of passive osmotic adjustment, a phenomenon to which a role in the maintenance of water status has been adjudicated (Meinzer *et al.* 1983). The fact that RWC_0 and Ψ_0 were lower than in irrigated controls implies that a reduction in LWC was delayed possibly through mechanisms not related to active osmotic adjustment.

A near constancy in LWC (on an area basis) and P for 20 days of drought bore a relationship to the presence of relatively elastic cell walls. Values of ϵ in water-stressed plants were comparable to those reported in the literature, as in water-stressed *Cenchrus ciliaris* (7 MPa; Wilson and Ludlow 1983) and droughted *Larrea tridentata* (10 MPa, Meinzer *et al.* 1986) but lower than in salt-stressed *Ellymus mollis* (20 MPa; Pavlik 1984). There was a significant increase in ϵ with drought but values were still within the lower range.

Stem water content ($756 \pm 37 \text{ mg cm}^{-3}$) was similar to that of the sympatrid, *Jatropha gossypifolia*, also with succulent stems ($794 \pm 37 \text{ mg cm}^{-3}$) and higher than that of another sympatrid, *A. crucis* ($563 \pm 45 \text{ mg cm}^{-3}$), which does not have succulent stems. Capacitance was ten times higher than in leaves of *A. crucis* (calculated from data of Ríos 1993). The ratio stem volume/leaf area, as a measure of relative capacitance, in well-watered plants was similar to the ratio pith volume/leaf area of seven species of *Espeletia* ($0.048 \pm 0.012 \text{ cm}^3 \text{ cm}^{-2}$) in which a leaf water conservation mechanism was proposed consisting of water translocation from the pith to the leaves (Meinzer and Goldstein 1986). A high capacitance in *I. carnea* may have contributed, along with the features abovementioned, to the

maintainance of leaf water status, assuming that water was readily translocated from the stem to the leaves in droughted plants.

Soluble sugars were accumulated during drought, as is frequently found (Munns *et al.* 1979). Although they could potentially act as osmotica, their small relative contribution to Ψ_s at the end of the dry cycle discards that possibility. Also, their possible role, together with free amino acids, as compatible solutes in the cytoplasm is discarded, given the absence of osmotic adjustment. Their accumulation on a DW basis may well be a mere consequence of a lowering in anabolic rate and a rising in catabolic rate during drought. Contrary to common findings (Ackerson and Hebert 1981), starch not only did not accumulate, but its content decreased.

We propose that the drought-resistance response of the deciduous *I. carnea* consists of tolerance at a high water status, rather than avoidance. Mechanisms would include an early and strong stomatal closure and the maintenance of leaf water content during drought through both a reduction in transpiration and a possible translocation of water from the stem. After rainfall, a fast recovery of soluble protein and of photosynthesis could be possible due to the former. Such set of mechanisms would allow leaves which have been under drought for more than one month to resume high photosynthetic rates within a few days of the first rains. This set of mechanisms would be more adaptive in the short term, i.e. a few days without rains, which seems to be a common occurrence in the ecosystem. In the long term avoidance by leaf shedding is the strategy employed by plants of *I. carnea* when drought extends for months.

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