BIOLOGIA PLANTARUM 45 (2): 213-220, 2002

Limitation to carbon assimilation of two perennial species in semi-arid south-east Spain

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Abstract

Diurnal and seasonal changes of net photosynthetic rate (P_n) and the efficiency of photosystem 2 (F_v/F_m) were measured on two perennial species growing on a soil catena in semi-arid south-east Spain. *Stipa tenacissima*, a tussock grass, grows on shallow soil at the top of the catena and *Retama sphaerocarpa*, a leguminous shrub, grows in the valley bottom. A linear relationship was found between light saturated photosynthetic rates (P_{max}) and diffusive leaf conductance (g_l) in both *Retama* and *Stipa* indicating that the intercellular CO_2 concentration (c_i) was maintained constant in both species diurnally. Relatively high values of calculated c_i in *Retama* cladodes suggested that was not the primary limitation to carbon assimilation. F_v/F_m for the two species when well watered was around 0.8. Although *Retama* cladodes maintained this value throughout the year, F_v/F_m decreased to a minimum of 0.43 in *Stipa* leaves, at the end of the dry season. Our data suggest that plants in the Rambla Honda can substantially reduce transpiration without reducing photosynthetic rates to the same extent by closing their stomata, because P_n is reduced primarily by high respiration, decreased mesophyll conductance and by photoinhibition or permanent damage of photosystem 2.

Additional key words: chlorophyll fluorescence, diffusive conductance, photosynthesis, *Retama sphaerocarpa*, *Stipa tenacissima*.

Introduction

Physiological and morphological adaptations of natural vegetation in Mediterranean climates have been studied extensively in North America (Mooney *et al.* 1975, Gigon 1979, Owens and Schreiber 1992) and Australia (Collatz *et al.* 1976). Substantially less information exists, however, for the Mediterranean Basin itself. Shrublands in southern Spain, in France and in Italy cover a large part of southern Europe. The low and erratic rainfall makes this type of landscape perhaps, one of the most environmentally unstable in Europe.

The vegetation protecting this ecosystem, although vital to its continued stability, is probably the component least well understood. Information on the dynamics of biomass production and ground cover within different plant communities, and their sensitivity to changes in land use and climate is required if models of water balance and erosion are to be tested over long time periods. It is therefore necessary to compare the carbon assimilation of different natural plant communities within such ecosystems to understand the differences between communities and their response to environmental changes.

Received 27 February 2001, *accepted* 7 September 2001.

Abbreviations: c_a - concentration of CO_2 in the ambient air; c_i - concentration of CO_2 at mesophyll cell walls; F_v/F_m- ratio of the variable to maximum dark-adapted fluorescence; g_l - leaf conductance to water vapour; g_m - conductance from mesophyll cell wall to carboxylation site; Γ - compensation concentration of CO_2; P_{max} - photosynthetic rate at light saturation; P_n - net photosynthetic rate; Q - photosynthetic quantum flux density.

Acknowledgements: The research was carried out as part of the MEDALUS (Mediterranean Desertification and Land Use) research project of the E.U. contract number EPOC-CT90-0014-(SMA), and was also funded by the natural environmental research council project number GR3/8169. Roberto Lázaro and Sebastian Vidal of the Estación Experimental de Zonas Áridas (CSIC) provided meteorological data, and various members of the University of Almería helped in the field data collection.

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This study has been focussed on two perennial species, *Stipa tenacissima* L., a perennial tussock grass and *Retama sphaerocarpa* (L.) Boiss., a leguminous shrub with evergreen cladodes. These species which grow on different sites along a soil catena within a catchment in south-east Spain, the Rambla Honda (Puigdefábregas *et al.* 1996, 1998), have different responses to prolonged drought. These species, growing at the top and the bottom respectively, are of the major perennials in the catchment and represent plant types typical of those found in uncultivated semi-arid areas. A combination of porometry and fluorescence measurements (Demmig and Winter 1988a,b) has been used to examine the seasonal importance of leaf conductance for carbon assimilation and fluorescence ratio in these species.

Materials and methods

Plants: The field site Rambla Honda is a dry valley on the southern slope of the Sierra de los Filabres, near Tabernas, Almería Province, SE Spain. (37°08' N, 2°22' W, altitude 600 - 700 m). Its climate is semi-arid, with a mean annual temperature of 16 °C. Mean annual rainfall is 279 mm (10-year record, 1989-1999) which falls mainly in the winter season, followed by a dry period centred on the months of June to September (Domingo *et al.* 2001, Lázaro *et al.* 2001).

Both species are adapted for dry climate, *Retama sphaerocarpa* (L.) Boiss. has small short-lived leaves and photosynthesises and transpires through its cladodes (photosynthetic stems) whereas *Stipa tenacissima* L. has stomata at the bottom of grooves in its abaxial surface and can fold its leaves. This vegetation is typical of southern Spain and representative of vegetation found over much of the Mediterranean Basin.

Measurement of gas exchange: Net photosynthetic rate (P_n) was measured with an infra-red gas analyser (*LCA-3, ADC*, Hoddesdon, UK), using a narrow leaf chamber *PLC-3(N)* on eight occasions (Table 1) to sample seasonal changes in net photosynthetic rates of *Stipa* leaves and *Retama* cladodes. Initial tests showed that it was necessary to include more than one leaf or cladode in the chamber and a specially designed clip was constructed which allowed four leaves or cladodes to be measured simultaneously. The analyser was calibrated with a previously calibrated standard mixture of CO_2 in air. For runs 4 - 5, 9 - 10 and 20 - 21, eight replicate measurements were taken on three plants, two from each cardinal direction. For runs 11, 13, four measurements, one from each cardinal direction, were made on eight plants. Gas exchange was measured on foliage in its natural orientation in space, so that variation in light within the canopy was sampled. The same sets of leaves were measured at approximately two-hour intervals from about one hour after sunrise to dusk except for runs 11, 13 which were measured in the early morning, mid-morning, midday and mid-afternoon. Values of incident

Table 1. Dates, species, environmental conditions and measurements for each run. (Runs 1 to 3 and 12 were only used for measurements of water relations, see Domingo *et al.* 2001). Max Ta - maximum air temperature; Max Da - maximum air vapour pressure deficit; ϑ - soil moisture.

Run	Date	Max Ta [°C]	Max Da [kPa]	ϑ [g(H_2O) g^{-1}]	Species	Plants	Replicates [$plant^{-1}$]	P_n	F_v/F_m	P_n/c_i
4	15 Apr 92	24.1	2.52	0.03	*Retama*	3	8	+		
5	24 Apr 92	25.2	2.49	0.03	*Stipa*	3	8	+		
6	1 May 92	22.9	2.02	0.04	*Retama*	8	4		+	
7	2 May 92	21.9	1.75	0.04	*Stipa*	8	4		+	
9	28 May 92	25.9	2.75	0.08	*Stipa*	3	8	+		
10	5 Jun 92	22.5	1.93	0.09	*Retama*	3	8	+		
11	17 Jun 92	20.0	0.90	0.17	*Retama*	8	4	+	+	
13	19 Jun 92	27.1	2.25	0.15	*Stipa*	8	4	+	+	
14	7 Aug 92	34.4	4.45	0.01	*Retama*	8	4		+	
16	9 Aug 92	29.2	3.74	0.01	*Stipa*	8	4		+	
17	29 Sep 92	24.0	2.06	0.00	*Retama*	8	4		+	
19	1 Oct 92	26.1	2.31	0.00	*Stipa*	8	4		+	
20	14 Jan 93	13.4	0.70	0.12	*Retama*	3	8	+		
21	21 Jan 93	17.0	1.72	0.10	*Stipa*	3	8	+		
22	7 Jun 94				*Retama*	2	6			+

photosynthetic quantum flux density (Q) were recorded by a PAR sensor incorporated in the *PLC-3(N)* leaf chamber during each measurement of gas exchange. Leaf area was measured by analysing images of detached leaves by image analysis software (*ACAD*, *Autodesk*, San Rafael, USA). The images had been recorded on a laptop computer with a hand-held scanner (*Scanman 256*, *Logitek*, Los Angeles, USA). A leaf area meter (*Mk2*, *Delta-T Devices*, Burwell, UK) was also used for some samples.

In June 1994, six P_n/c_i curves were determined for two *Retama* bushes in the field, using an artificial light source that clipped onto the cuvette (giving Q around 940 μmol m^{-2} s^{-1}). These response curves were measured half an hour and two and a half hours after sunrise.

Fluorescence ratio: The ratio of the variable to maximum dark-adapted fluorescence (F_v/F_m) of a leaf, is linearly correlated with maximum quantum yield of photosynthesis (Krause and Weiss 1991). The ratio indicates the intrinsic efficiency of photosystem 2 (PS2) and is useful in identifying when non-stomatal limitations to carbon assimilation become important. Fluorescence ratio was measured with a chlorophyll induction fluorometer (Plant Efficiency Analyser, *Hansatech*, King's Lynn, UK). Because of the thickness of the *Retama cladodes*, it was necessary to cut a groove into the seal of the clip to ensure that light was excluded. Measurements were replicated four times one at each cardinal point of the compass, on each of eight plants. Plants were sampled before dawn, around one hour after dawn, mid-morning, midday, mid-afternoon and at dusk. The leaf or cladode was too small to completely cover the aperture of the clip, hence the exposed leaf area was not constant, so only F_v/F_m was measured since it is independent of area. Means and standards errors were calculated with an arctan transformation of all the F_v/F_m data because of the non-normal distribution of ratio data.

Derivation and calculation of conductances to carbon dioxide transfer: Diffusive flow of CO_2 into the substomatal cavity of the leaf is controlled by boundary layer, stomatal and cuticular conductances to carbon dioxide (g_{ac}, g_{sc} and g_{cc}, respectively). These conductances are directly proportional to the diffusive conductances to water vapour (g_{aw}, g_{sw} and g_{cw}) according to the ratio of the diffusion coefficients, $g_c = g_w/1.6$. Strictly the ratio for cuticular conductances is not 1.6 but in the IRGA software a ratio of 1.6 has been used. Leaf conductance to water vapour, g_{lw}, which is a combination of the cuticular and stomatal conductances, is derived by the gas analyser. Although g_{cw} is small when the stomata when they are closed.

The CO_2 diffusion in intercellular air space encounters five other resistances to its diffusion before carboxylation, the mesophyll cell wall, plasmalemma, cytosol, chloroplast limiting membrane and interior of chloroplast. The combination of all these conductances from mesophyll cell wall to carboxylation site has been given the symbol g_m. Net photosynthetic rate (P_n) of a leaf can be expressed by equation:

$$P_n = g_m (c_i - \Gamma)$$

where Γ is the CO_2 compensation concentration and c_i is the CO_2 concentration at mesophyll cell walls. This equation has been used, acknowledging the limitation that g_m will rarely mimic a physical diffusion process. However we find this approach useful for evaluating limitations to carbon fixation.

Analysis of the conductances to CO_2 shows that the boundary layer around a *Retama* cladode does not present a major resistance to diffusion. For example, at a wind speed of 0.5 m s^{-1}, g_{ac} is around 2.2 mol m^{-2} s^{-1} (Domingo *et al.* 1996), which can be compared to the average g_{lc} of 0.125 mol m^{-2} s^{-1} (derived from data of Domingo *et al.* 2001 for g_l for water vapour) and g_m of 0.029 mol m^{-2} s^{-1} (see Results). At a maximum photosynthetic rate of 15 μmol m^{-2} s^{-1} (see Results), g_{ac} of 2.2 mol m^{-2} s^{-1} would decrease CO_2 concentration at the surface of the leaf by 7 μmol mol^{-1}, which would have little effect on photosynthetic rate. Consequently the limitation to P_n must reside in g_{lc} or g_m.

By separating the transport of carbon dioxide into two parts it is possible to see whether P_n is limited by g_{lc} or g_m (Jones 1985). There has been much interest in the sensitivity of photosynthetic rate to stomatal conductance (Farquhar and Sharkey 1982, Morison and Gifford 1983, Jones 1985, Luo 1991, Quick *et al.* 1992). The simplest statement of the limitation of the gaseous phase (l_g), *i.e.*, g_l and g_a, on P_n, can be expressed as

$$l_g = [(c_a - c_i)/c_a] \times 100,$$

where c_a is the CO_2 concentration in the ambient air. This is a simplification (Farquhar and Sharkey 1982) and can, under many circumstances, give misleading results, but is true when c_i or the 'operating point' lies on the linear part of the P_n/c_i curve (Jones 1985). Equation 2 also does not separate the effects of changes in Γ, resulting from variation in photorespiration or dark respiration, from g_m. An indication of the relative importance of stomatal versus non-stomatal control of photosynthetic rate is given by l_g.

The response of P_n to incident photosynthetic quantum flux density (Q) was assumed to be:

$$P_n = P_{max} \times [(Q/(Q + b_q)]$$

where P_{max} is the photosynthetic rate at light saturation and b_q is an empirical constant determined for each species (Jones 1992).

Results

Net photosynthetic rate: The diurnal patterns of P_n (Fig. 1*A*,*B*) were different from those of Q (Fig. 1*C*). P_n reached a maximum about 3 to 4 h before solar noon, equivalent to 14:00 local time, then declined over the day independently of Q in both *Stipa* (Fig. 1*A*) and *Retama* (Fig. 1*B*). The decline was more noticeable as the plants became more severely water stressed in June (runs 9, 10) for both *Stipa* and *Retama* (Domingo *et al.* 2001). In June, P_n were determined on both the current and previous year's cladodes of *Retama*. The previous year's cladodes had produced flowers by early June (run 10) and young fruits by mid-June (run 11). The lower P_n of old (with flowers) compared to young (*i.e.* 2 - 3 months old) cladodes may result from high respiration rates of the flowers (Fig. 1*B*). Analysis of P_n for individual sampling times indicated that rates decreased by about 30 μmol m^{-2} s^{-1} for each g of flowers or fruit (Fig. 2).

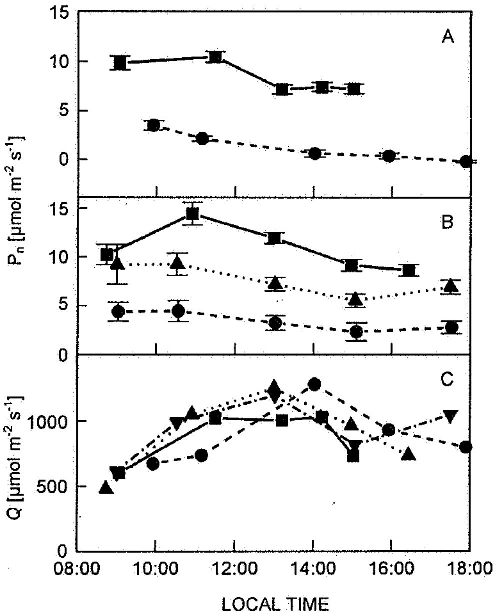

Fig. 1. Diurnal variation in net photosynthetic rate (Pn) of *A*) *Stipa* leaves in April (run 5 - *squares*), and May (run 9 - *circles*), and *B*) *Retama* cladodes in April (run 4 - *squares*), and June (run 10 - *circles*) for the old cladodes, and June (run 11 - *triangles*) for the young cladodes. *Error bars* represent SE, *n* = 24. *C* - Diurnal variation in photosynthetic quantum flux density at the place of measurement of P_n for *Retama*, runs 4 (*triagles*) and 10 (*reverse triagles*) and *Stipa*, runs 5 (*squares*) and 9 (circles).

P_{max} was compared with corresponding g_l for *Stipa* and *Retama* for runs 4, 5, 9 - 11, 13, 20 and 21 (Fig. 3). Some measurements were taken below light saturation, so variability associated with the effect of low Q on photosynthetic rate was accounted for by calculating P_{max}. Using a fixed value of b_q a linear relationship between P_{max} and g_l was found. The empirical constant b_q for *Stipa* and *Retama* was estimated to be 75 and 400 μmol m^{-2} s^{-1}, for the two species respectively. Although b_q may not remain constant throughout the year, the data suggested little variation, *i.e.* in *Stipa* by less than 25 μmol m^{-2} s^{-1} and in *Retama* by less than 50 μmol m^{-2} s^{-1}.

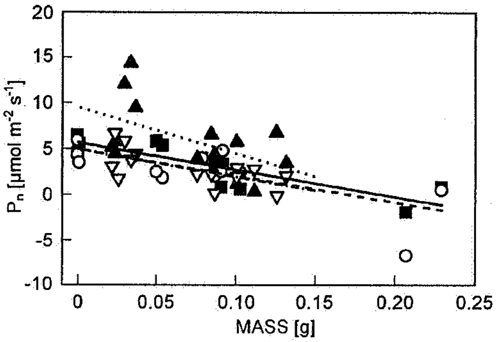

Fig. 2. The relationship between net photosynthetic rate (P_n) in *Retama* and mass of flowers or fruit per sample. Run 10, 11:00 (squares, r^2 = 0.72), run 10, 13:00 (*circles*, r^2 = 0.56), run 11, 13:00 (*closed triangles*, r^2 = 0.29) and run 11, 15:00 (*open reverse triagles*, r^2 = 0.43). The fitted lines are linear regressions of each data set; all slopes are significantly different from zero.

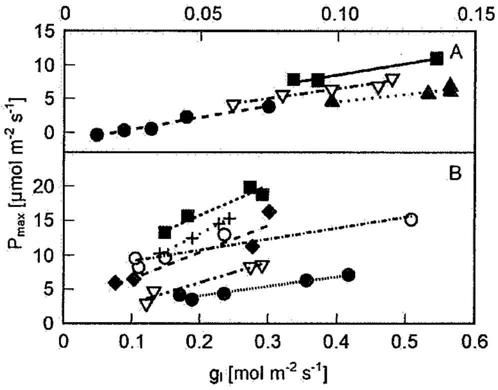

Fig. 3. The response of light-saturated net photosynthetic rate (P_{max}) to leaf conductance (g_l) in *A*) *Stipa* for run 5 (*squares*), run 9 (*circles*), run 13 (triagles), and run 21 (*reverse open triangles*), and B) *Retama* for run 4 (*squares*), run 10 old cladodes (*closed circles*) and young cladodes (*open circles*), run 11 old cladodes (*open reverse triangles*) and young cladodes (*rhombs*) and run 20 (*crosses*). The fitted lines for *Stipa* and *Retama* are linear regressions of each data set (see Table 2).

The linear relationship found between P_{max} and g_l in both *Stipa* and *Retama* (Fig. 3*A*,*B*) shows that c_i was maintained more or less constant over the day. This suggests that P_n decreased in parallel with g_l. Values of c_i for *Stipa* and *Retama* derived from data presented in Fig. 3 (Table 2) and assuming c_a = 340 μmol mol^{-1}, show that the limitation of P_n by stomatal and boundary layer conductance in *Retama* changed over the season, and was less than 25 % at all times especially in the summer. In

Table 2. Values of c_a - c_i, c_i and l_g (sensitivity of P_n to g_l) for *Stipa* and *Retama* derived from Fig. 3. Means ± SE, *n* = 24.

Species	Run	Age	c_a - c_i [µmol mol^{-1}]	c_i [µmol mol^{-1}]	l_g [%]
Retama	4	old	70 ± 10	265	21
	10	young	26 ± 6	309	7
	10	old	22 ± 3	313	8
	11	young	63 ± 17	272	19
	11	old	48 ± 7	287	14
	20	old	81 ± 2	254	24
Stipa	5		112 ± 12	228	33
	9		111 ± 11	229	33
	13		70 ± 20	270	21
	21		94 ± 13	246	28

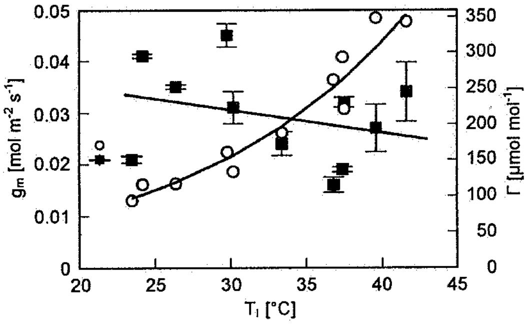

Fig. 4. The response of g_m (*squares*) and Γ (*circles*) to changes in temperature for *Retama* in June 1994. The two smaller points are thought to be in error (because of instrumentation problems) and were omitted from the regression analysis. The fitted lines are regressions of each data set.

general, there was a l_g in *Stipa* larger than in *Retama*, and l_g was least in *Retama* at the time of maximum growth (5 June, run 10) and in *Stipa* after the rain storm in June (19 June, run 13).

The relatively small dependence of P_n on CO_2 diffusion suggests that other parts of the photosynthetic system play a more important part in controlling P_n. Measured P_n/c_i curves for *Retama* exhibited a linear relationship between P_n and c_i over the range of c_i used (80 % of the relationships had r^2 > 0.98, all had r^2 > 0.94). This suggests that the above mentioned equation was valid for these data, although whether P_n/c_i relationship is linear for times of the year when the plants were less stressed, is not known.

The Γ and g_m derived from the P_n/c_i curves, were not significantly different between old and young *Retama* cladodes. Average values of g_m, obtained as the initial slope of the P_n/c_i curve, were 0.026 ± 0.009 and 0.032 ± 0.009 mol m^{-2} s^{-1} for the young and old cladodes, respectively. Calculated Γ, obtained from P_n/c_i curve when P_n = 0, was found to be very high (up to 300 µmol mol^{-1}) and variable.

It was found that much of the variability of Γ could be explained by changes in leaf temperature (Fig. 4). g_m did not change significantly as temperature increased. These data suggest that respiration may well play an important part in the constant and high c_i values in both *Stipa* and *Retama*.

Fluorescence ratio: When the plants were well watered in May, F_v/F_m for both species was around 0.8 before dawn, it declined during the day with increasing Q and recovered to 0.8 again at dusk (Fig. 5*A*). At the end of the dry season in September, F_v/F_m were substantially lower except for *Retama* which maintained F_v/F_m just below 0.8 throughout the day (Fig. 5*B*) and again showed considerable recovery with declining Q in the afternoon. At the end of the dry season *Stipa*, which was severely dehydrated (RWC *ca.* 50 - 60 %) had a low F_v/F_m at dawn, a large decline before noon and little or no recovery by the end of the day (Fig. 5*B*).

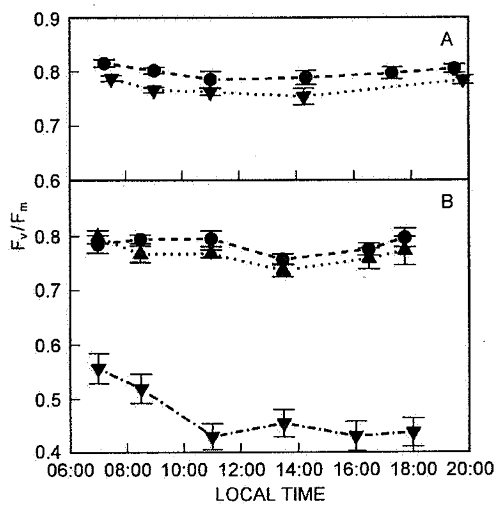

Fig. 5. Diurnal variation in F_v/F_m in May (runs 6 - 8) (*A*) and September (runs 17 - 19) (*B*) for *Retama* old cladodes (*circles*) and young cladodes (*triangles*) and *Stipa* (*reverse triangles*). *Error bars* represent SE, *n* = 24.

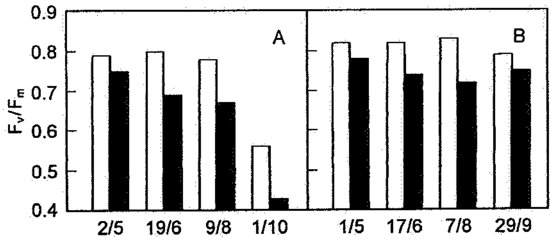

Fig. 6. Seasonal changes of dawn (*open columns*) and minimum daily (*shaded columns*) F_v/F_m for *A*) *Stipa* and *B*) *Retama*.

The daily minimum values of F_v/F_m in *Stipa* (Fig. 6*A*) showed a progressive decrease between early May and late September, but pre-dawn values only started to drop after August (run 16). *Retama* values of F_v/F_m were relatively constant throughout the year, with its minimum yearly value of F_v/F_m occurring in August and not September like *Stipa* (Fig. 6*B*).

Discussion

Under conditions where g_l is limiting P_n, c_i should decrease as stomatal conductance decreases (Farquhar and Sharkey 1982, Morison and Gifford 1983, Luo 1991). However, c_i has been reported to increase or stay constant as stomatal conductance changes (Schulze *et al.* 1975, Wong *et al.* 1979), as found here. The constant within-day values of c_i for *Stipa* varied seasonally between 230 - 270 μmol mol^{-1} (when c_a = 340 μmol mol^{-1}) and were very similar to those of eight C_3 plant species that maintained c_i at a more or less constant 220 μmol mol^{-1} (when c_a = 320 μmol mol^{-1}) (Wong *et al.* 1979). Despite the apparent stability of c_i among species, woody species have generally higher c_i than herbaceous plants. In *Retama* c_i for runs 4 and 20 (5 April 92 and 14 January 93), when the cladodes had no flowers and temperatures were low, were 264 ± 10 and 254 ± 2 μmol mol^{-1}, respectively (when c_a = 340 μmol mol^{-1}), values very similar to Wong's data. During run 11 (June 17) the young cladodes had a high c_i of 272 ± 17 μmol mol^{-1} and when they were still expanding, during run 10 (June 5), c_i was even higher (309 ± 6 μmol mol^{-1}).

The high values of c_i in *Retama* cladodes, particularly for old cladodes (5 and 17 June, runs 10 and 11) and young cladodes (run 10), may have resulted from high respiration rates (Fig. 2), which would increase Γ (Fig. 4). Γ does not vary greatly between C_3 species, values being around 40 μmol mol^{-1} (Bauer and Matha 1981). Beadle *et al.* (1981) reported that c_i of Sitka spruce increased as xylem water potential decreased, but Luo (1991) reported little variation with water potential in *Abutilon theophrasti*. The largest changes in Γ have been shown to be influenced by temperature (Bauer *et al.* 1983, Kirschbaum and Farquhar 1984, Weber *et al.* 1985) as reported here for *Retama*. One possible cause of the diurnally stable c_i is an increase in respiration rate with increasing temperature; there may also be an effect of heat stress, as this has also been shown to cause large increases in the compensation point (Bauer *et al.* 1983).

Values of l_g suggest that most of the limitation to carbon assimilation in *Retama* results not from stomatal closure but from other factors, such as high respiration rates and low g_m. *Retama* maximum l_g was 24.5 % during the winter when the plants were well hydrated, a value lower than that for another semi-arid shrub, *Larrea divaricata*, at both high and low leaf water potentials (l_g = 38 - 41 %) (Jones 1985). Net photosynthetic rates of *Stipa* were more sensitive to leaf conductance (21 - 33 %) than those of *Retama* (7 - 24 %).

The measured values of g_m for *Retama* were low (0.029 mol m^{-2} s^{-1}) when compared with maximum values compiled by Jones (1992), which range from 0.08 mol m^{-2} s^{-1} for Sitka spruce to 0.15 mol m^{-2} s^{-1} for *Larrea divaricata*. It appears that g_m decreases sharply with water stress (Luo 1991, Martin and Ruiz-Torres 1992), and decreases of 0.05 to 0.02, 0.08 to 0.02 and 0.08 to 0.03 mol m^{-2} s^{-1} were observed for three Mediterranean shrubs *Heteromeles arbutifolia*, *Salvia leucophylla* and *Salvia labitae*, respectively over a drying cycle in the field (Gigon 1979). It was also observed that g_m for these shrubs decreased with time even under irrigation, which could reflect ageing of leaves that occurs irrespective of water status.

Differences between diurnal maximum and minimum F_v/F_m (Figs. 5, 6) indicated that photoinhibition occurred during the day probably resulting from high quantum flux densities, a phenomenon which is thought to be common to most vegetation in direct sunlight (Baker *et al.* 1994). Furthermore, pre-dawn values suggest that non-reversible damage to PS2 only occurred at the end of the dry season in *Stipa* (Figs. 5*A*, 6). This contrasts with *Retama* where the pre-dawn values of F_v/F_m remained high over the year.

Reduced values of F_v/F_m seem to be related to the changes in relative water content (RWC) in both species. F_v/F_m of *Stipa* leaves appears to decline little until RWC falls below threshold values of 80 % (Aguilera, personal communication). As the plant dries below this value, F_v/F_m decreases rapidly. Taking the results of F_v/F_m in this context, lack of damage to PS2 in *Retama* may result from the plant's ability to maintain its RWC above its threshold value, which is probably below 0.8 (Domingo *et al.* 2001, Fig. 5*A*). From this data it is difficult to predict how PS2 in *Retama* cladodes would react to a decrease in RWC below its threshold, but by growing in the valley bottom it may avoid dehydration thus enabling it to maintain its intrinsic ability to assimilate CO_2 unimpaired. *Stipa* is perhaps the best adapted to protect its PS2 reaction centres since it can survive considerable drought before it dehydrates significantly (see Fig. 7 in Domingo *et al.* 2001). Consequently the intrinsic ability of *Stipa* to assimilate CO_2 is maintained, which presumably allows it to react quickly to any rain showers (Pugnaire and Haase 1996).

Over the whole season the ratio of maximum to minimum daily average P_n in *Retama* was 1.96 and in *Stipa* 6.78, which indicates the large difference in the effect of seasonally imposed stresses on the two species.

The fact that photosynthetic rates do not change much with g_l can be an useful adaptation by the plant (Aasamaa and Sõber 2001). This is because stomatal closure can restrict transpiration without substantially affecting P_n.

Thus the change in stomatal conductance with atmospheric humidity found in *Retama* (Domingo *et al.* 2001) does not limit carbon accumulation as much as water loss by the plant. Decreases in stomatal conductance in rice (*Oryza sativa*) and *Phalaris* resulting from changes in atmospheric humidity did not affect photosynthetic rates until the evaporation from the plant was so high that it affected the water status of the plant, thereby affecting g_m (Morison and Gifford 1983). Luo (1991) found that photosynthetic rates were more affected by soil water content than by atmospheric humidity. Our data suggest that plants in the Rambla Honda can substantially reduce transpiration without reducing photosynthetic rates to the same extent by closing their stomata, because P_n is reduced primarily by high respiration, decreased g_m and by photoinhibition or permanent damage of PS2.

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