COMPARATIVE WATER RELATIONS AND PHOTOSYNTHESIS OF MYCORRHIZAL AND NON-MYCORRHIZAL BOUTELOUA GRACILIS H.B.K. LAG EX STEUD.

Michael F. Allen

William K. Smith

Thomas S. Moore Jr.

Martha Christensen

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COMPARATIVE WATER RELATIONS AND PHOTOSYNTHESIS OF MYCORRHIZAL AND NON-MYCORRHIZAL BOUTELOUA GRACILIS H.B.K. LAG EX STEUDEL.

BY MICHAEL F. ALLEN*, WILLIAM K. SMITH, THOMAS S. MOORE, JR AND MARTHA CHRISTENSEN

Department of Botany, University of Wyoming, Laramie, Wyoming 82071, U.S.A.

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SUMMARY

The rangeland grass, Bouteloua gracilis, was inoculated with its mycorrhizal symbiont, Glomus fasciculatus, to determine the influence of vesicular-arbuscular mycorrhizae on water status, stomatal behaviour and photosynthesis as well as gross plant morphology, biomass and phosphorus content. Mycorrhizal infection increased transpiration rates by over 100% with 50 to 70% lower leaf resistances to water vapour diffusion. Leaf xylem pressure was not different between mycorrhizal and non-mycorrhizal plants indicating that whole-plant resistance to water transport was reduced by more than 50%. Photosynthetic rates under saturating light conditions increased 68% with infection as a consequence of a 33% reduction in stomatal resistance and a 67% reduction in mesophyll resistance to CO₂ uptake. Mycorrhizal infection did not affect biomass or gross plant morphology after 30 weeks of growth, but increased chlorophyll and phosphate concentrations by 28% and 70% respectively. These physiological changes indicate that mycorrhizae may substantially alter survival ability of Bouteloua gracilis.

INTRODUCTION

Vesicular-arbuscular (VA) mycorrhizae improve survival and growth of many plants, especially those living in marginal habitats (Aldon, 1975; Kahn, 1975; Ruehle and Marx, 1979). Direct transport of several nutrients from soil to plant by mycorrhizal fungi has been demonstrated (Hattingh, Gray and Gerdemann, 1973; Rhodes and Gerdemann, 1978, 1979) suggesting that the increased absorption provided by hyphae increased plant growth (Sanders and Tinker, 1971). However, the effects of mycorrhizal infection on other physiological processes that influence growth, such as water relations, stomatal behaviour, and CO₂ uptake, are relatively unknown.

In many arid and semiarid regions, the major factor limiting plant production and nutrient uptake may be water availability (Harner and Harper, 1973; Fischer and Turner, 1978; Lauenroth, Dodd and Sims, 1978). Safir, Boyer and Gerdemann (1971, 1972) demonstrated that, under mesic soil water conditions, VA mycorrhizal infection lowered resistance to water uptake by 40% in soybean and Mosse and Hayman (1971) reported that non-mycorrhizal onions wilted when transplanted, whereas mycorrhizal plants did not. Aldon (1975) noted that survival of Altriplex canescens on coal mine spoils in New Mexico increased with VA mycorrhizal infection and he hypothesized that increased water uptake was a contributing factor to improved survival.
factor. More recently, Levy and Krikun (1980) found that VA mycorrhizal infection increased transpiration and $^{14}$CO$_2$ uptake in citrus upon recovery from drought.


**MATERIALS AND METHODS**

*B. gracilis* was grown from seed (Sharp Brothers Seed Company, Healy, Kansas) in 10 cm diameter pots (five seedlings per pot, 15 pots per treatment) containing steam-sterilized grassland sandy loam (Olney soil series) collected from the Pawnee grassland IBP site near Ft. Collins, Colorado, U.S.A. Each pot was inoculated with approximately 150 sterile (non-mycorrhizal treatment) and viable (mycorrhizal treatment) *G. fasciculatus* spores collected from clones of *B. gracilis* in the Pawnee grasslands by a sucrose flotation method (Allen *et al.*, 1979). The greenhouse was adjusted to approximate a typical growing season for a shortgrass steppe region of the north-central United States (16 h day, 30 to 35 °C, 8 h night, 15 to 20 °C, 13 to 23 % relative humidity) (Lauenroth and Sims, 1976). Plants were watered regularly until experiments began. No fertilizers were added throughout the study. Lighting was supplemented with high-intensity incandescent lamps. Following inoculation, axenic conditions were not maintained.

**Plant growth and nutrient status**

After 5 months growth, infection frequency was determined as described by Allen and Allen (1980) and roots and shoots were washed and dried at 55 °C for 48 h for dry wt determination. It was impossible to separate all soil particles from the roots; therefore, following dry wt determinations, the roots were ashed at 550 °C for 6 h and the ash weight was subtracted from the dry wt. Leaf size was estimated on one plant randomly selected from each pot; leaf length and width were measured just above the sheath and triangular leaf area computed. From the total number of leaves per plant the leaf area per plant was determined. Stomatal characteristics were determined at midday by spreading a thin layer of Duco cement (Du Pont Co.) over the surface of the flag leaf, near the sheath. After drying, the cement was carefully stripped off and stomatal length, width and density measured microscopically. Other leaves were frozen and hand-sectioned just above the sheath to determine mesophyll cell size, bundle sheath cell size and number and leaf thickness.

Root and shoot phosphate concentrations were analyzed as described by Dittmer and Wells (1969). Leaf concentrations of total chlorophyll, chlorophyll $a$ and $b$ were measured spectrophotometrically and chlorophyll $a/b$ ratios calculated (Ross, 1974).

**Water relations**

Pots of mycorrhizal and non-mycorrhizal plants were allowed to dry out and soil water potential ($\psi_s$), air temperature and absolute air humidity monitored until $\psi_s$ reached the lowest measurable limit of the soil thermocouple psychrometers.
Water relations of mycorrhizae

Measurements of leaf temperature, stomatal resistance to water vapour diffusion \( R_{svw} \) and leaf xylem pressure \( \psi_x \), were made on one plant from each pot at midday daily throughout the drying period. Aerodynamic resistance was estimated to be approximately 10 s m\(^{-1}\) for both treatments (Nobel, 1974; Taylor, 1975). Total diffusion resistance \( R_{svw} \) was then computed as the parallel sum of the aerodynamic and stomatal resistance for both sides of the leaf (Nobel, 1974).

Transpiration rates \( J_{svw} \) for mycorrhizal and non-mycorrhizal plants were computed using the Ohm's Law analogy for water vapour diffusion (Nobel, 1974) and resistance to water flux from soil to leaves calculated as described by Nye and Tinker (1977). Leaf temperatures were measured with a Barnes Instatherm (Model 14-220-4) and air temperature and humidity using a ventilated thermocouple psychrometer in order to calculate the difference in water vapour concentration between leaf interior and surrounding air (Nobel, 1974). Stomatal resistance to water vapour diffusion was measured with a diffusion resistance porometer (Lambda Inst. Corp., Model HAW-103), leaf xylem pressure with a Scholander-type pressure bomb (PMS model 1000) and soil water potential with thermocouple psychrometers (Wescor Inc. model PT51-10) and a Dewpoint microvoltmeter (Wescor Inc. model HR-33T).

Stomatal behaviour and light

Mycorrhizal and non-mycorrhizal plants in saturated soils (\( \psi_s \sim 0.01 \) MPa, \( n = 15 \)) were taken from the greenhouse at midday when the stomata were fully open and placed in a ventilated dark growth chamber at 30 °C (20% relative humidity). Changes in \( R_{svw} \) for leaves of three individual plants (alternating within a pot) were measured each minute for the first 10 min and every 2 min thereafter for 30 min using the diffusion resistance porometer. Two additional sets of plants \( (n = 15) \) were kept in the dark for 24 h at 30 °C and were then placed in a growth chamber at 30 °C using supplemental incandescent and fluorescent lighting at an illumination of 400 \( \mu \)E m\(^{-2}\) s\(^{-1}\). Stomatal resistance was monitored and leaf resistance calculated approximately every minute for the first 6 min and every 2 min thereafter for 30 min as above.

Photosynthesis

\( \text{CO}_2 \) uptake by mycorrhizal and non-mycorrhizal plants was monitored using a flow-through infrared gas exchange system incorporating a Beckman model 865 \( \text{CO}_2 \) analyzer under near saturating light conditions (1600 \( \mu \)E m\(^{-2}\) s\(^{-1}\)). Individual plants (8 to 10 blades) were sealed inside a Plexiglass cuvette and blade temperatures were monitored with fine-wire thermocouples (36 gauge). Temperature was controlled (±1.6 °C) by circulating cold water through a false-bottom in the cuvette and by filtering the light source through 10 cm of water. The \( \text{CO}_2 \) concentration entering the cuvette was maintained at approximately 320 \( \mu \)l l\(^{-1}\) in \( \text{N}_2 \). The influence of mycorrhizae on the \( \text{CO}_2 \) diffusion pathway from the ambient air to the carboxylation sites in the chloroplasts was quantified using electrical circuit analogies (Nobel, 1974). The linearity of the equations when the internal concentration of \( \text{CO}_2 \) was assumed negligible was confirmed by plotting \( \text{CO}_2 \) flux \( J_{\text{CO}_2} \) against ambient \( \text{CO}_2 \) concentration over the range 90 to 400 \( \mu \)l l\(^{-1}\) of \( \text{CO}_2 \) in \( \text{N}_2 \).
Statistics

Standard data transformation and appropriate parametric statistical techniques were used as described by Zar (1974) and Neter and Wasserman (1974). Student's t-test was applied to biomass, nutrient content, morphological features, \( R_p \) and \( \text{CO}_2 \) uptake, also to the linear regressions for water status of mycorrhizal and non-mycorrhizal plants. Values of \( R_{uv} \) for leaf resistance changes with light were transformed logarithmically and comparisons based on linear regressions.

Table 1. Plant biomass, morphology and anatomy of VA mycorrhizal and non-mycorrhizal Bouteloua gracilis expressed as mean ± s.e. mean (n)

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Treatment</th>
<th>Non-mycorrhizal</th>
<th>Mycorrhizal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Infection frequency (%o)</td>
<td></td>
<td>0±0 (5)</td>
<td>48±5 (15)</td>
</tr>
<tr>
<td>Root dry wt (mg per plant)</td>
<td></td>
<td>189±21 (15)</td>
<td>229±19 (15)</td>
</tr>
<tr>
<td>Shoot dry wt (mg per plant)</td>
<td></td>
<td>160±19 (15)</td>
<td>197±14 (15)</td>
</tr>
<tr>
<td>Total dry wt (mg per plant)</td>
<td></td>
<td>350±28 (15)</td>
<td>425±31 (15)</td>
</tr>
<tr>
<td>Average leaf width at base (mm)</td>
<td></td>
<td>0.79±0.02 (10)</td>
<td>0.79±0.02 (10)</td>
</tr>
<tr>
<td>Average leaf length (mm)</td>
<td></td>
<td>56.1±2.5 (10)</td>
<td>59.6±2.3 (10)</td>
</tr>
<tr>
<td>Average leaf thickness (( \mu m ))</td>
<td></td>
<td>124±13 (10)</td>
<td>136±4 (10)</td>
</tr>
<tr>
<td>Leaf area (( \text{mm}^2 ) per plant)</td>
<td></td>
<td>1059±137 (10)</td>
<td>1223±150 (10)</td>
</tr>
<tr>
<td>Average no. of leaves per plant</td>
<td></td>
<td>20.6±1.8 (10)</td>
<td>24.4±1.9 (10)</td>
</tr>
<tr>
<td>Stomatal density (number ( \text{mm}^{-2} ))</td>
<td></td>
<td>205±7 (10)</td>
<td>227±14 (10)</td>
</tr>
<tr>
<td>Stomatal length (( \mu m ))</td>
<td></td>
<td>20.2±1.9 (10)</td>
<td>22.4±1.7 (10)</td>
</tr>
<tr>
<td>Stomatal width (( \mu m ))</td>
<td></td>
<td>13.4±1.5 (10)</td>
<td>11.8±2.2 (10)</td>
</tr>
<tr>
<td>No. cells per bundle sheath</td>
<td></td>
<td>9±1 (10)</td>
<td>9±0 (10)</td>
</tr>
<tr>
<td>Cell diameter (( \mu m ))</td>
<td></td>
<td>20±3 (10)</td>
<td>20±1 (10)</td>
</tr>
<tr>
<td>Mesophyll cell width (( \mu m ))</td>
<td></td>
<td>2±0 (10)</td>
<td>2±0 (10)</td>
</tr>
<tr>
<td>Mesophyll cell length (( \mu m ))</td>
<td></td>
<td>13±1 (10)</td>
<td>13±3 (10)</td>
</tr>
</tbody>
</table>

Table 2. Chlorophyll and phosphate concentrations of VA mycorrhizal and non-mycorrhizal Bouteloua gracilis

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Leaf chlorophyll (mg kg(^{-1}) fresh wt)</th>
<th>Phosphate (mmol kg(^{-1}) fresh wt)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Total</td>
<td>( a/b )</td>
</tr>
<tr>
<td>Non-mycorrhizal</td>
<td>619±50</td>
<td>624±78</td>
</tr>
<tr>
<td>Mycorrhizal</td>
<td>867±98*</td>
<td>961±53</td>
</tr>
</tbody>
</table>

* Denotes significant differences at confidence level \( \geq 0.95 \).
† Significant differences at confidence level \( \geq 0.95 \) by Mann-Whitney U-test.

Leaf chlorophyll was measured on seven composite samples of four plants [mean ± s.e. (mean)], phosphate on three composite samples of four plants (mean values only).

RESULTS

Plant growth, anatomy and nutrient status

After 4 to 5 months growth, no significant differences in biomass or gross plant morphology were apparent between mycorrhizal and non-mycorrhizal plants (Table 1); the same applied to mesophyll cell size, bundle sheath cell size and number and stomatal size and density (Table 1). Mycorrhizal infection increased
leaf chlorophyll concentrations by 28%, did not affect chlorophyll a/b ratios and increased P concentration by 89 and 47% in leaves and roots respectively (Table 2).

**Water relations**

Although no significant differences in leaf xylem pressure potentials were noted between mycorrhizal and non-mycorrhizal plants [Fig. 1(a)], $R_{wv}$ was significantly lower in mycorrhizal plants throughout the soil drying period, ranging from 50% less than control plants in saturated soil to 70% less in extremely dry soils [Fig. 1(b)]. Transpiration fluxes for mycorrhizal plants were more than twice those for non-mycorrhizal plants throughout the range of $\psi_s$ [Fig. 1(c)]. Stomatal resistances of mycorrhizal plants remained low down to the lowest measurable $\psi_s$ (approximately minus 6 MPa), whereas those of many non-mycorrhizal plants increased until no measurable transpiration occurred at $\psi_s$ values near minus 5.5 MPa. However, no significant differences in $\psi_s$ with drying could be detected between pots containing mycorrhizal and non-mycorrhizal plants (Table 3).
Mycorrhizae also appeared to affect water transport from soil to leaves. At high $\psi_s$ (0 to minus 0.5 MPa) there was a 50\% reduction in $R_p$ (Table 4). Similar differences between mycorrhizal and non-mycorrhizal plants occurred in moderately dry soils (minus 0.6 to minus 1.5 MPa), although the variability among individual plants was much greater. In dry soils (less than minus 1.5 MPa), the variability in $R_p$ within treatments was greater than differences between mycorrhizal and non-mycorrhizal treatments.

**Table 3.** Water potentials ($\psi_s$) of drying soil containing mycorrhizal and non-mycorrhizal Bouteloua gracilis expressed as mean ± s.e. (mean) (n = five per day per treatment)

<table>
<thead>
<tr>
<th>Days of drying</th>
<th>Non-mycorrhizal</th>
<th>Mycorrhizal</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.01 ± 0.0</td>
<td>-0.01 ± 0.0</td>
</tr>
<tr>
<td>2</td>
<td>-0.01 ± 0.0</td>
<td>-0.01 ± 0.0</td>
</tr>
<tr>
<td>4</td>
<td>-0.1 ± 0.0</td>
<td>-0.1 ± 0.1</td>
</tr>
<tr>
<td>6</td>
<td>-1.1 ± 1.0</td>
<td>-1.6 ± 1.4</td>
</tr>
<tr>
<td>7</td>
<td>-1.6 ± 0.4</td>
<td>-2.6 ± 0.8</td>
</tr>
</tbody>
</table>

**Table 4.** Whole plant resistance to water flux ($R_p$) of VA mycorrhizal and non-mycorrhizal Bouteloua gracilis expressed as mean ± s.e. (mean) (n)

<table>
<thead>
<tr>
<th>$\psi_s$ range (MPa)</th>
<th>$R_p$ (MPa m$^2$ s mg$^{-1}$ x $10^{-3}$)</th>
<th>Percentage reduction with infection</th>
</tr>
</thead>
<tbody>
<tr>
<td>0 to -0.5</td>
<td>20 ± 2 (15)</td>
<td>50</td>
</tr>
<tr>
<td>-0.6 to -1.5</td>
<td>15 ± 3 (10)</td>
<td>53</td>
</tr>
<tr>
<td>&lt; -1.5</td>
<td>9 ± 10 (9)</td>
<td>89</td>
</tr>
</tbody>
</table>

* Significant difference between treatments at confidence level $\geq 0.99$.
† Significant difference between treatments at confidence level $\geq 0.90$.

**Stomatal behaviour and light**

Although there was little $R_{wv}$ response to decreases in leaf xylem pressure potentials during the day, there were significant differences in response to light–dark transitions between mycorrhizal and non-mycorrhizal plants (Fig. 2). Leaf diffusion resistance of mycorrhizal plants remained approximately 50\% lower than in non-mycorrhizal plants during a 30 min exposure to light [Fig. 2(a)], but became much greater during darkness [Fig. 2(b)].

**Photosynthesis**

VA mycorrhizal infection significantly increased CO$_2$ uptake by 68\%, as compared with non-mycorrhizal plants (Table 5). One-third of the increase was due to a 51\% reduction in the combined gas-phase resistance ($R_{CO_2}^{gas}$) and two-thirds to a 77\% reduction in the liquid-phase resistance of the mesophyll cells ($R_{CO_2}^{un}$).

**Discussion**

*Bouteloua gracilis* is a dominant grass in western North America and has VA mycorrhizal infection frequencies of 88 to 100\% in the field (Davidson and
Water relations of mycorrhizae

Fig. 2. Diffusion resistance of opening (a) and closing (b) stomata of VA mycorrhizal (●) and non-mycorrhizal (○) Bouteloua gracilis in light and dark. Mean values for $R_w \pm$ s.e. (mean).

Table 5. Gas exchange of VA mycorrhizal and non-mycorrhizal Bouteloua gracilis expressed as mean ± s.e. (mean)

<table>
<thead>
<tr>
<th>Treatment</th>
<th>No. of plants</th>
<th>$R_{wF}$ (s m$^{-1}$)</th>
<th>$R_{CO_2}$ (s m$^{-1}$)</th>
<th>$R_{CO_2}$ (s m$^{-1}$)</th>
<th>$J_{CO_2}$ (mmol m$^{-1}$ s$^{-1}$)</th>
<th>Percentage increase due to $R_{CO_2}$</th>
<th>Percentage increase due to $R_{wF}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Non-mycorrhizal</td>
<td>11</td>
<td>840 ± 311*</td>
<td>1310 ± 430*</td>
<td>2050 ± 780*</td>
<td>3.8*</td>
<td>—</td>
<td>—</td>
</tr>
<tr>
<td>Mycorrhizal</td>
<td>13</td>
<td>560 ± 200</td>
<td>870 ± 310</td>
<td>1160 ± 560</td>
<td>6.4†</td>
<td>33</td>
<td>67</td>
</tr>
</tbody>
</table>

* All measured and calculated values are significantly different between treatments at confidence level ≥ 0.95.
† Indicates a 68% increase in photosynthesis in mycorrhizal as compared with non-mycorrhizal plants.

Christensen, 1977; Allen, personal observations). Infection with VA mycorrhizae has been shown to enhance water status in soybean (Safir, Boyer and Gerdemann, 1972), onion (Mosse and Hayman, 1971), citrus (Levy and Krikun, 1980) and to improve survival of Atriplex canescens in arid mine spoils (Aldon, 1975). However, only a few investigators have considered possible physiological mechanisms which could contribute to these effects. Daft and Okusamya (1973) reported an increased number of vascular bundles in maize following mycorrhizal infection which could reduce resistance to water transport. Safir et al. (1972) estimated that root resistance to water uptake was reduced 40% by mycorrhizal infection in soybean. They also noted that phosphate fertilizers reduced root resistance and attributed the improved water transport to improved nutritional status, especially phosphorus.
However, Levy and Krikun (1980) suggested that in similarly sized plants, mycorrhizae only increase stomatal conductance and do not alter root resistance. Transpiration rates were significantly higher in mycorrhizal than in non-mycorrhizal plants with no changes in xylem pressure potentials (Fig. 1). These results were similar to those reported for citrus (Levy and Krikun, 1980). Under high transpirational demands such as in western grasslands (Lauenroth and Sims, 1976) and in our greenhouse (less than 25% humidity and higher than 30 °C air temperature), xylem pressure potentials closely approached leaf water potentials (Weatherly, 1975). Thus, whole-plant resistance ($R_p$) was reduced by mycorrhizal infection. Assuming that $R_p$ approaches the inverse of the hydraulic conductivity at high transpiration rates (Fiscus and Markhardt, 1979), B. gracilis had fairly high whole-plant resistances when compared with crop plants (Boyer, 1971; Safir et al., 1972; Fiscus and Markhardt, 1979). However, as reported previously by Safir et al. (1972), whole-plant resistances dropped sharply with mycorrhizal infection.

Whole-plant resistance is a composite term which incorporates many possible factors including root surface area and permeability (Nye and Tinker, 1977). Although root biomass for B. gracilis was not altered significantly here, increased root branching (Allen et al., 1981) could lead to substantial increases in root surface area without changes in total biomass. The absorbing area for water uptake also could be increased in mycorrhizal plants by the fungal hyphae, especially by reducing or eliminating ‘dry zones’ which might surround the slower growing roots during low soil moisture periods (Nye and Tinker, 1977). Finally, changes in hydraulic conductivity have been observed following changes in phytohormone levels (Markhardt et al., 1979; Tal et al., 1979) such as have been attributed to mycorrhizal infection (Allen, Moore and Christensen, 1980; Allen, 1980).

Leaf resistance to water vapour diffusion in mycorrhizal plants was lower than in non-mycorrhizal plants throughout the drying period [Fig. 1(b)] and in response to illumination (Fig. 2), but became considerably higher in response to darkness (Fig. 2). The reduced resistance could have resulted from enhanced water uptake and thus, higher leaf water potentials or possibly increased photosynthesis (Meidner and Mansfield, 1968). Additionally, leaves from VA mycorrhizal B. gracilis grown in axenic culture can have elevated cytokinin levels (Allen et al., 1980) which may stimulate stomatal opening (Incoll and Whitelam, 1977) and transpiration (Cooper, Dibgy and Cooper, 1972). The increased $R_{wv}$ for mycorrhizal plants during darkness (Fig. 2) suggests either incomplete stomatal closure in non-mycorrhizal plants (stomatal sizes and densities were not different between mycorrhizal and non-mycorrhizal plants, Table 1) or a higher cuticular resistance to water vapour diffusion in mycorrhizal plants. To our knowledge, the possibility of a better developed cuticle or other more impermeable epidermal covering following mycorrhizal infection has not been investigated.

The increase in photosynthesis by B. gracilis infected with VA mycorrhizae was due to a 51% reduction in the gas-phase and a 77% reduction in the liquid-phase resistance (Table 5). The reason for the reduction in gas-phase resistance is probably the same as for the observed reduction in stomatal resistance to water vapour transport (see also Levy and Krikun, 1980). We found higher phosphate levels in mycorrhizal than in non-mycorrhizal plants which might reduce the liquid-phase resistance to CO$_2$ uptake, but would not be expected to alter the gas-phase resistance (Longstreth and Nobel, 1980). The liquid-phase resistance also incorporates resistance to CO$_2$ uptake associated with changes in photophosphorylation (Nobel, 1974). The increased amounts of leaf chlorophyll without
significant differences in leaf thickness or chlorophyll $a/b$ ratios, may indicate an increase in the number of photosynthetic units (Alberte, Fiscus and Naylor, 1975) contributing to the lower resistance.

Despite increased photosynthesis, we found no significant increase in biomass of mycorrhizal plants. We are not sure why this was so. Possibly there was no major phosphate response as has been reported elsewhere (Safir et al., 1972) since phosphate is not as limiting, generally, as water and nitrogen in the Pawnee grassland soils (Lauenroth et al., 1978). Furthermore, $B. gracilis$ is a slow growing perennial and may require much longer establishment periods for significant growth enhancements to be observed. According to Tranquillini (1964), as much as 38% of the CO$_2$ fixed by $Pinus cembra$ may be incorporated in its ectomycorrhizae. Thus, the possibility exists that a substantial portion of the increased photosynthate in our mycorrhizal plants may have been incorporated into the fungus (see Sanders, Mosse and Tinker, 1975). Further research is needed to clarify the role of carbon allocation in the symbiotic relationships of VA mycorrhizal fungi and their hosts.

$B. gracilis$ is one of the few $C_4$ perennial plants predominating in western North American high plains. In the field, CO$_2$ fixation was still substantial even at soil water potentials below minus 3 MPa (Brown and Trlica, 1977). Leaf potentials as low as minus 6 to minus 7 MPa have been measured in the field in areas where soil water potentials commonly fall to minus 6 or minus 8 MPa during the growing season (Hutcheson, 1972; Allen, 1979). Therefore, $B. gracilis$ must be able to conduct water vapour and CO$_2$ at extremely low soil water potentials. We noted that conductance of water vapour, and therefore CO$_2$, was greater in mycorrhizal than in non-mycorrhizal plants down to the lowest measurable soil and leaf water potentials. Furthermore, even though midday transpiration rates were higher in mycorrhizal than in non-mycorrhizal plants, the rate of soil water depletion was not significantly different (Table 3). This suggests that mycorrhizal plants are more efficient in obtaining the available moisture and that a mycorrhizal relationship might be an essential adaptation in these habitats.

The enhanced water and nutrient uptake and photosynthetic rates of $B. gracilis$ with mycorrhizal infection indicates an important role for this association in plant survival. As marginal habitats are utilized increasingly for agriculture and energy development, the successful re-establishment of desirable native plant species may require the restoration of VA mycorrhizal associations.

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**References**


Water relations of mycorrhizae


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