

Water-use efficiency and carbon isotope discrimination of *Acacia ampliceps* and *Eucalyptus camaldulensis* at different soil moisture regimes under semi-arid conditions

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Abstract

Acacia ampliceps Maslin and *Eucalyptus camaldulensis* Dehnh. were grown for one year in lysimeters at three soil moisture regimes: 100 % (well-watered), 75 % (medium-watered) and 50 % (low-watered) of total plant available water. Biomass yield of both species increased with increase in soil moisture. Water-use efficiency (WUE) of *E. camaldulensis* decreased and that of *A. ampliceps* increased markedly with decrease in available soil moisture. *A. ampliceps* showed 4 - 5 times more biomass yield than *E. camaldulensis* grown at similar soil moisture. *A. ampliceps* showed almost 5, 9 and 12 times higher WUE than *E. camaldulensis* under low-, medium- and well-watered treatments, respectively. Significant negative correlation of $\delta^{13}\text{C}$ with WUE ($r = -0.99$) was observed in *A. ampliceps*. In contrast, $\delta^{13}\text{C}$ of *E. camaldulensis* showed a significant positive correlation with WUE ($r = 0.82$).

Additional key words: biomass production, plant available soil water, transpiration efficiency.

Soil salinity is a major form of land degradation in several parts of arid and semi-arid regions of the world including Pakistan (Qureshi and Barrett-Lennard 1998). *Acacia ampliceps* and *Eucalyptus camaldulensis* proved to be the most promising among the species tested under saline conditions (Davidson and Galloway 1993). *E. camaldulensis*, a moderately to highly salt and waterlogging tolerant species, is very widely planted around the world (Marcar 2002). *A. ampliceps* has proven to be one of the best performing species for survival and growth on saline land in Pakistan (Marcar *et al.* 1998, Ansari *et al.* 1998) and is potentially a valuable fuel wood and fodder species for arid subtropics. However, such plantations, particularly of *Eucalyptus*, have been criticized that they may deplete groundwater reserves by high rates of water-use (Morris 1997). The scarcity of irrigation water is a global problem that warrants efficient use of water reserves. This could be achieved by selection

of high water-use efficiency plants and/or managing irrigation according to plant water requirements.

The concept of water-use efficiency has been very well utilized in crop plants to select cultivars/species that provide maximum yields under specific climatic conditions. In contrast, most of the studies on tree species have been directed to estimate the whole-tree water-use and not the water-use efficiency. Hence, the information regarding screening of tree species for water-use efficiency is limited. In the present study, we determined the water-use efficiency of *Eucalyptus camaldulensis* and *Acacia ampliceps* grown at different soil water regimes and its relationship with carbon isotope discrimination ($\delta^{13}\text{C}$) under saline conditions in a semi-arid climate.

The study was conducted at Nuclear Institute for Agriculture and Biology (NIAB), Faisalabad (31°.2' N, 73°.05' E), Pakistan. The area is semi-arid and characterized by large seasonal variations of both

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temperature and rainfall. During winter the temperature ranges from 2 - 18 °C and during summer from 20 - 47 °C. The average annual rainfall is 250 mm, most of which occurs in monsoon (July - August). The soil collected from a saline area at Biosaline Research Station, Pacca Anna near Faisalabad, was air dried, passed through 2 mm sieve and filled into lysimeters (1 × 1 × 1 m). The soil attained average bulk density (~1.4 g cm⁻³) similar to that in the field. In the center of each lysimeter, a neutron moisture access tube was installed down to the bottom. The neutron moisture meter was calibrated in the same soil before the start of experiment by field method (Akhter *et al.* 1995). Two soil moisture tensiometers, one at 15 cm and other at 50 cm depth, were also installed in each lysimeter. The water retained by the soils at different pressures (0.03 MPa and 1.5 MPa) was determined by placing the soil samples in plastic rings in a pressure membrane apparatus. Total available water (TAW) = soil moisture at 0.03 MPa (FC) - soil moisture at 1.5 MPa (PWP), where FC and PWP refer to field capacity and permanent wilting point, respectively.

Four seedlings (*ca.* 30 cm tall) of each species, *Eucalyptus camaldulensis* and *Acacia ampliceps*, grown in plastic bags containing fertile soil were transplanted in each of 9 lysimeters containing saline sodic soil (electrical conductivity, EC 1.25 S m⁻¹, pH 8.5, sodium adsorption ratio, SAR 60) with sandy loam texture (sand 650, silt 210, clay 140 g kg⁻¹). Three plots (lysimeters) were kept as an unplanted control. The seedlings were grown for 6 months under similar conditions until they established with a uniform biomass cover. Tap water (EC 0.09 S m⁻¹; SAR_{adj} 2.6; pH 7.6) was used to irrigate the plants.

Three replicate plots of each species, selected randomly, were subjected to three water regimes. In well-watered treatment, the soil was kept at 100 % of TAW, under medium-water treatment at 75 % of TAW and in low-watered treatment at 50 % of TAW. The amount of water required for each plot to maintain the respective soil water regime was estimated on the basis of readings from the neutron moisture meter. The tensiometer readings were used as an alternate check to confirm the soil moisture status. Generally the water regimes were restored on alternate days except for few weeks of rainfall periods during the year. The volume of water required for each plot was added through a locally fabricated irrigation system including a water pump, fixed pipes, water flow meters and taps, *etc.* The rainfall during the experimental period was also recorded.

Plants were harvested year after maintaining the soil moisture treatments and fresh and dry biomass of plants was determined. Seasonal samples were taken after mixing fresh leaves collected from all trees from each lysimeter in mid of March, June and November. Plant samples (final) were also collected at the harvesting time after mixing old and fresh leaves. Plant samples ground after oven drying at 45 °C were preserved for isotopic ($\delta^{13}\text{C}$) analysis. Total water applied during the whole growing period was used to determine the water-use

efficiency (WUE) and transpiration efficiency (TE). The plant-transpired water was estimated as the difference between the water applied to control (unplanted) plots kept at a moisture regime and the total water applied to the respective treatment plots with plants. The WUE and TE were calculated using the following relations: WUE = total dry matter/total water applied and TE = total dry matter/total water transpired.

Yield response factor (K_y) for each species was derived by regressing relative yield decrease (1-Y_a/Y_m) with relative decrease in water (1-ET_a/ET_m) for each treatment. Where, Y_a is actual yield, Y_m the maximum yield, and ET_a is the actual evapotranspiration and ET_m is the maximum evapotranspiration. The isotopic ratio ($R = ^{13}\text{C}/^{12}\text{C}$) of plant samples (R_{sample}) and in standard (R_{standard}) was determined using a ratio mass spectrometer. The R values were converted to $\delta^{13}\text{C}$ using the relation: $\delta^{13}\text{C} = [R_{\text{sample}}/R_{\text{standard}} - 1] \times 1000$.

The standard was provided by International Atomic Energy Agency (IAEA), Vienna, Austria. Data were subjected to analysis of variance (ANOVA) followed by Duncan's multiple range test (DMRT) for comparison of paired means of treatments using PC package *CoStat (CoHort Software, Berkeley, USA)*.

In both species, *Eucalyptus camaldulensis* and *Acacia ampliceps*, biomass yields were significantly higher under well-watered conditions relative to low- and medium-watered plants (Table 1). *A. ampliceps* showed almost 4 - 5 times more biomass yield than *E. camaldulensis* when grown under similar conditions. Under well-watered conditions, *E. camaldulensis* consumed significantly more water and produced significantly higher biomass compared with that under medium- and low-watered treatments. Higher dry/fresh mass ratios in medium- and low-watered treatments (Table 1) indicated reduced water uptake under water deficit compared with well-watered conditions. WUE and TE of *E. camaldulensis* increased with increase in soil moisture and exhibited trends similar to biomass yields under different water treatments. The results suggest that *E. camaldulensis* has a prodigal water-use strategy and may be useful plant for areas where water availability is not a problem. Evergreen vegetation can adjust its leaf area index in response to long-term water availability (Hatton and Wu 1995) and leaf efficiency (tree water use per unit leaf area) did not vary among eucalyptus growing under similar conditions at sites with seasonal water limitations (Hatton *et al.* 1998). These observations support our findings that *E. camaldulensis* is lavish in water-use.

Under well-watered conditions, *A. ampliceps* consumed more water and produced higher biomass compared with medium-watered and low-watered plants. The WUE and TE of *A. ampliceps* increased with decrease in total plant available water (Table 1). Similar dry/fresh mass ratios of plants grown in well-watered and low-watered conditions indicated ability of the species to maintain water uptake under drought stress. Therefore, *A. ampliceps* employed a conservative water-use strategy and can be grown in water-limited and high salinity

Table 1. Biomass production, total water applied, water-use efficiency and transpiration efficiency of *Eucalyptus camaldulensis* and *Acacia ampliceps* grown under different soil moisture regimes. Values are means \pm SE of three replicates. Means followed by different letters in a row differ significantly at $P \leq 0.05$.

Species	Parameter	Well-watered	Medium-watered	Low-watered
<i>Eucalyptus . camaldulensis</i>	Total water applied [kg]	4304 \pm 130a	2805 \pm 75b	2579 \pm 247b
	Fresh mass [kg plot $^{-1}$]	13.99 \pm 0.79a	6.28 \pm 0.18b	6.39 \pm 0.61b
	Dry mass [kg plot $^{-1}$]	6.06 \pm 0.25a	2.89 \pm 0.11b	2.96 \pm 0.60b
	Dry/Fresh mass ratio	0.433	0.461	0.463
	Water-use efficiency [g kg $^{-1}$]	1.40 \pm 0.03a	1.03 \pm 0.01b	1.12 \pm 0.12b
	Transpiration efficiency [g kg $^{-1}$]	2.22 \pm 0.02a	1.71 \pm 0.03b	1.52 \pm 0.12b
<i>Acacia ampliceps</i>	Total water applied [kg]	4433 \pm 109a	1625 \pm 108b	800 \pm 3.20b
	Fresh mass [kg plot $^{-1}$]	95.78 \pm 4.76a	38.57 \pm 5.86b	32.14 \pm 3.21b
	Dry mass [kg plot $^{-1}$]	32.28 \pm 2.74a	15.02 \pm 1.15b	11.11 \pm 0.98b
	Dry/Fresh mass ratio	0.337	0.389	0.334
	Water-use efficiency [g kg $^{-1}$]	7.25 \pm 0.46b	9.19 \pm 0.12b	13.84 \pm 1.18a
	Transpiration efficiency [g kg $^{-1}$]	8.14 \pm 0.46c	11.79 \pm 0.80b	20.11 \pm 1.37a

Table 2. Carbon isotope discrimination ($\delta^{13}\text{C}$ [%]) in leaves of *Eucalyptus camaldulensis* and *Acacia ampliceps* grown under different soil moisture regimes. Values are means \pm SE of three replicates. Values for each species followed by different letters differ significantly at $P \leq 0.05$.

Species	Parameter	Well-watered	Medium-watered	Low-watered
<i>Eucalyptus . camaldulensis</i>	March	-28.51 \pm 0.032cd	-28.96 \pm 0.015ef	-29.30 \pm 0.015fg
	July	-26.04 \pm 0.339a	-29.41 \pm 0.020g	-27.36 \pm 0.045a
	November	-27.51 \pm 0.043b	-28.33 \pm 0.042a	-27.70 \pm 0.045b
	December (final harvest time)	-27.35 \pm 0.032b	-28.90 \pm 0.012de	-28.13 \pm 0.101c
<i>Acacia ampliceps</i>	March	-28.91 \pm 0.015g	-28.56 \pm 0.050f	-28.05 \pm 0.031e
	July	-28.83 \pm 0.096g	-28.50 \pm 0.010f	-28.03 \pm 0.032e
	November	-29.58 \pm 0.031i	-26.44 \pm 0.036a	-26.84 \pm 0.062b
	December (final harvest time)	-29.13 \pm 0.070h	-27.83 \pm 0.082d	-27.64 \pm 0.040c

conditions with more biomass yields relative to *E. camaldulensis*. WUE of *Changium smyrnioides* and *Anthriscus sylvestris* also increased with the decrease of soil moisture (Ge *et al.* 2003). Thomson *et al.* (1994) reported that some Australian *Acacia* species withstand long dry periods. These observations may be explained by high water-use efficiency of *A. ampliceps* determined in our study.

Variation in $\delta^{13}\text{C}$ values in both *Eucalyptus camaldulensis* and *Acacia ampliceps* at different times of year (Table 2) reflects variation in rate of photosynthesis under different soil water treatments. Higher $\delta^{13}\text{C}$ (less negative) values of *E. camaldulensis* were observed under well-watered treatment. In contrast, *A. ampliceps* showed highest $\delta^{13}\text{C}$ values at low-watered treatment followed by medium- and well-watered treatments (Table 2). Variation in $\delta^{13}\text{C}$ values in both plant species over the season (Table 2) indicated differences in photosynthesis rates at different times that may be related to variations in atmospheric conditions. The carbon isotope discrimination ($\delta^{13}\text{C}$) was significantly and negatively correlated with WUE of *A. ampliceps* (WUE

[g kg $^{-1}$] = 104.6 - 4.076 $\delta^{13}\text{C}$ [%], $r = -0.99$) subjected to different water regimes. However, a positive correlation of $\delta^{13}\text{C}$ with WUE was observed in *E. camaldulensis* (WUE = 7.29 + 0.237 $\delta^{13}\text{C}$, $r = 0.82$) under water-limited conditions and may be attributed to low rate of photosynthesis under decreasing soil moisture availability.

In conifer trees, Warren *et al.* (2001) found a strong relationship between $\delta^{13}\text{C}$ and water availability or drought stress in seasonally dry climates ($\text{P/E} < 1$) and little or no relationship under wet conditions ($\text{P/E} > 1$). Using carbon isotope discrimination, Lüttge *et al.* (2003) reported that long-term WUE in *Eucalyptus saligna* was lower than in *Podocarpus falcatus*, whereas *Eucalyptus* species also showed higher transpiration than *P. falcatus*. For *Eucalyptus microtheca* grown at three moisture levels in greenhouse, Li (1999) found significant population differences in each treatment in $\delta^{13}\text{C}$, WUE and dry matter production. Further, a negative correlation in control and drought stress I and a positive correlation under control and drought stress II was observed between WUE and dry mass production. Korol *et al.* (1999)

observed that an increase in rainfall of 900 mm in a growing season of *Pinus radiata* showed an increase in carbon isotope discrimination (Δ) of 2.0 ‰ corresponding to a decrease in transpiration efficiency of 24 %. They found that Δ was negatively correlated with mean annual vapor pressure deficit at different sites and nutrient availability showed no significant effect on Δ values. The results of present study are in agreement with the earlier findings (Lüttege *et al.* 2003, Korol *et al.* 1999, Li 1999) and suggest that availability of soil moisture exercises a controlling influence on the $\delta^{13}\text{C}$ values of both the plant species. However, the contrasting behavior in *E. camaldulensis* and *A. ampliceps* reflects genetic differences between the two tree species in processes controlling C_i/C_a ratio and, thus, ^{13}C isotopic discrimination.

Yield response factor (K_y) is an important component of evapotranspiration function that provides a useful means for defining plant water requirements (IAEA 1996, Akhter *et al.* 2003). A smaller value of K_y indicates a smaller yield reduction with decreasing soil moisture. *A. ampliceps* showed lower value of yield response factor ($K_y = 0.817$) compared to *E. camaldulensis* ($K_y = 1.373$) over the growing season indicating that relative yield reduction in the former species is lower than the latter under same water deficit conditions. In the present studies, *A. ampliceps* proved to be drought tolerant species employing a conservative water-use strategy with high biomass production and high WUE and may, therefore, be recommended for dry saline areas. *E. camaldulensis*, with a prodigal water-use strategy, is a species suitable for areas of no water shortage.

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