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Growth and physiology of yarrow species *Achillea millefolium* cv. Cerise Queen and *Achillea filipendulina* cv. Parker Gold at optimum and limited moisture

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Abstract

Appropriate management of a limited water supply would be an effective tool to reduce production costs, minimize nutrient leaching, and conserve water. Yarrow is considered a high value herbaceous perennial for landscape in United States, and have solid, mat-forming rhizome/root system with fine feathery leaves which make it drought resistant. However, little information quantifies the physiological performance of yarrow species under limited moisture and recovery from drought. Two yarrow species (*Achillea millefolium* cv Cerise Queen and *Achillea filipendulina cv* Parker Gold) plants were subjected to four irrigation intervals: irrigated daily (control), 3, 6 and 9 days under greenhouse conditions in 2004 and 2005. Irrespective of the species, the plants exposed to drought had lower predawn leaf water potential (ψ_{pd}), stomatal conductance (S_c), transpiration rate (T_s), cell osmotic potential ($^{\Psi}_{s}$), and relative water content (RWC) than controls. Leaf area (LA), leaf weight (LW) and root weight (RW) of the controls were over twice that of plants irrigated every 9 days in 2004 and over four times as high in 2005. Leaf area ratio (LAR) was lowest in the control and increased with each level of drought stress. Root-to-shoot dry weight ratio (RSR) of the control was highest and decreased with each stress level and lowest RSR was observed for plants irrigated every 9 days. Drought did not affect net assimilation rate (NAR). In both years, relative growth rate (RGR) of the control was twice as high as plants irrigated every 9 days. Both species performed better in irrigated conditions compared with drought stress. However, the production of larger leaf area (3209 cm^2) heavier leaves (35.99 g) and better osmotic adjustment by the Cerise Queen specie in response to drought enabled it to deposit more solute and lowered leaf water potential without dehydration than Parker Gold, and thus may be grown successfully on limited moisture.

Keywords: leaf area indices, growth, osmotic potential, drought, yarrow.
Abbreviations: leaf water potential (ψ_{pd}), stomatal conductance (S_c), transpiration rate (T_s), cell osmotic potential ($^{\Psi}_{s}$), relative water content (RWC), Leaf area (LA), leaf weight (LW), root weight (RW), Leaf area ratio (LAR), Root-to-shoot dry weight ratio (RSR), net assimilation rate (NAR), relative growth rate (RGR), Drought treatment (D), species (S) and drought cycles (C), turgid weight (TW), fresh weight (FW), dry weight (DW)

Introduction

Water is the main limiting factor affecting crop production throughout the world. Establishing limited water supply as a management tool could be effective in reducing production costs, conserve water and minimize leaching nutrients and pesticides into ground water (Nuruddin *et al*., 2003). However, before adopting limited water supply as a management tool, its effect on growth, physiology and yield must be evaluated (Kirda, 2002). There is a need to identify the drought adaptation mechanisms on physiological basis of those plants that can be grown on limited water supply.

Reduced carbon assimilation (Stoneman *et al*., 1994), stomatal conductance as well as transpiration (Balok and St. Hilaire, 2002), osmotic adjustment (Chartzoulakis *et al*., 1993) and leaf water potential (Feser and St. Hilaire, 2005) and higher antioxidant activities are reported in drought conditions. Drought also reduced the plant growth (Fernandez *et al*., 2002), leaf area (Feser and St. Hilaire, 2005), plant biomass partitioning (Graves and Wilkins, 1991), and ratios of leaf surface area to root dry weight as well as leaf dry weight to root dry weight (Balok and St. Hilaire, 2002; Feser and St. Hilaire, 2005). Considerable differences in response to drought are known to exist among plant genotypes (Bauerle et al., 2003). Drought tolerant species have higher values of carbon assimilation, stomatal conductance (Padgett-Johnson *et al*., 2003), water potential and relative water content (Lo Gullo *et al*., 2003) compared to non drought tolerant species. Genotypic variability in cultivar responses to drought could be exploited to improve cultivar management in a nursery and landscape setting. Yarrow is an herbaceous perennial, and is best suited to

cottage rather than a formal garden (Halevy, 1999). It has medicinal and cosmetic uses (Rohloff *et al*., 2000), and extensively grown in drought-prone environments due to its numerous leaf and several stems developed from the horizontal rootstock (Bartram, 1995). However, drought adaptation mechanisms of yarrow species to limited irrigation are unknown. Information on the effects of moisture deficits on yarrow species *Achillea millefolium* cv Cerise Queen and *Achillea filipendulina cv* Parker Gold is non available. The objectives of this study were to quantify short term effect of drought on Ψ_{pd} , T_s, S_c, Ψ_s RWC and biomass production and partitioning in yarrow species Achilla "Cerise Queen" and "Parker Gold".

Results

Plant water relations and gas exchange

Drought treatment (D), species (S) and drought cycles (C) significantly affected predawn water potential (ψ_{Pd}) in 2004 , while the effects of D and C was only significant in 2005 (Fig. 1A-D). Interactions between D × S and D × C showed non significant effects in both years. Maximum ψ_{Pd} (-0.5 MPa) was recorded for *A. mellifolium* in 2005 irrigated daily (Fig 1B). ψ_{Pd} decreased with increase in irrigation interval and minimum ψ_{Pd} (-3.8MPa) was recorded for *A. filipendulina* watered every 9 days (Fig. 1 D). Drought treatments, species and drought cycles significantly affected stomatal conductance (S_c) in 2004, while only D and C showed significant effects on S_c in 2005 (Fig. 2A-D). None of the interactive effect was significant in both years. S_c decreased with increase in drought level and minimum S_c (9 m.m mol.$m^{-2}.s^{-1}$) was recorded for *A. filipendulina* in 2004 irrigated every 9 days (Fig. 2C). Maximum S_c (600 m.mol.$m^{-2}.s^{-1}$) was recorded for *A. filipendulina* in 2005 irrigated daily (Fig. 2 D). Number of drought cycles significantly impacted S_c and the highest S_c values (365mmol $m^{-2}.s^{-1}$) were recorded at 2nd drought cycle in 2005 (Fig. 2 D).

Transpiration rate (T_s) was significantly affected by D, S, C and S × C interaction in both years while no statistical difference in T_s was observed for S in 2005 (Fig. 3A-D). D × S interaction was non significant in both years (Fig. 3A, B). Maximum T_s (10 m. mol. m^{-2} s^1) was recorded for *A. millefolium* in 2004 at drought cycle 7 (Fig 3A). T_s decreased with increase in drought level and minimum T_s (0.2mmol m^{-2}) was recorded in 2005 for *A. millefolium* and also for *A. filipendulina* in drought cycle 2 watered every 9 days (Fig. 3 A, C).

Cell osmotic potential and relative water content

Drought treatments, S and C significantly affected cell osmotic potential (ψ_s) in both years, while D × C interaction was significant only in 2005 (Fig. 4A-D). None of the other interaction showed a significant effect on $\psi_{s..}$ Control had highest ψ_s (-0.8 MPa) for both species in 2004 (Fig 4A-C) which decreased with increase in drought level and minimum ψ_s (-4.7 MPa) was recorded for *A. millefolium* in drought cycle 8 in 2005 that received water every 9 days (Fig. 4 B).

Drought levels significantly affected relative water content (RWC) in both years (Fig. 5A-D), while S and D × S interaction was significant only in 2004 (Fig. 5A-C) and C was significant in 2005 (Fig. 5B-D). None of the other interaction showed significant effect on RWC. Highest RWC (88%) was recorded in *A. millefolium* irrigated daily in drought cycle 8 in 2005 (Fig. 5 B) which decreased with increase in drought level and minimum RWC (40%) was recorded for A. *filipendulina* in 2005 that received water every 9 days (Fig. 5 D).

Growth parameters

Drought significantly impacted leaf area (LA) in both years (Table 1, 2) while S affected LA only in 2004 (Table 1). Leaf area decreased with increase in drought stress. Control plots had two times higher LA of (3690 cm^2) in 2004 and four times (3435 cm^2) higher in 2005 than plants irrigated every 9 days. *A. millefolium* produced more leaf area (3209, 1656) than *A. filipendulina* (2270, 1648 cm^2) in both years (Table 1,2). Leaf weight (LW) and root weight (RW) were significantly affected by drought stress in both years while S affected RW in 2004 only (Table 1,2). Drought decreased LW and RW. Control plots had two times higher LW (38.97 g) and RW (63.25 g) and four times higher LW and RW (10.11g) than plants irrigated every 9 days in 2004 and 2005, respectively (Table 1, 2). Leaf area ratio (LAR) was significantly affected by D in 2004 only (Table 1). LAR of the control (22.84) was lowest and increased with each level of drought stress and was highest (39.96) for plants irrigated every 9 days (Table 1). Drought and S significantly affected root-to-shoot dry weight ratio (RSR) in both years (Table 1, 2). Control had highest RSR (3.19) which decreased with increasing stress level and lowest RSR (1.58) was observed for plants irrigated every 9 days (Table 1, 2). *A. filipendulina* gave high RSR (2.80, 1.79) than *A. millefolium* (1.79, 1.12) in both years (Table 1, 2). Species significantly affected NAR in 2004 only, while D showed non significant effect on NAR in both years (Table 1,2). *A. filipendulina* resulted in maximum NAR (0.120 mg. cm^2. d^{-1}) than *A. millefolium* (0.115 mg. $cm^{-2}.d^{-1}$). Drought showed significant effect on relative growth rate (RGR) in both years (Table 1, 2). RGR of the control (0.151, 0.059) was twice as high as plants irrigated every 9 days in both years (Table 1,2).

Discussion

Plants watered daily (control) showed maximum ψ_{pd} which decreased with increase in irrigation interval and lowest ψ_{pd} was recorded for plants watered every 9 days. The low ψ_{pd} of drought stressed plants suggests that the leaves were not fully rehydrated at the end of drought cycle (Feser and St. Hilaire 2005; Balok and St. Hilaire, 2002). Droughted plants tended to exhibit the lowest ψ_{pd} indicating that the quantity of water in the root zone or the capacity of conducting system to transport water was insufficient to allow rehydration of leaves during night (Close *et al*. 1996). Stomatal conductance ($S_{c)}$ decreased with increase in drought level and minimum S_c was noted for highly stressed plants. This indicates that the severity of our drought treatments were sufficient to cause severe reduction in S_c. The high S_c in control treatment may be due to optimum availability of water which offered low resistance to S_c compared to the stressed treatments which offered high resistance to S_c due to shortage of water, small changes in the growing substrate moisture level can trigger

Table 1. Means values for various growth and development parameters of yarrow species subjected to water stress during 2004.

Parameter	Irrigation (days)				*Achillea*	
	control	3	6	9	*millefolium*	*filipendulina*
Leaf area (cm^2)	3690a*	2963b	2449bc	1853c	3209a	2270b
Leaf weight (g)	38.97a	34.67a	23.48b	19.46b	35.99a	22.30b
Root weight (g)	63.25a	53.23b	37.78c	27.47d	48.74a	42.13b
LAR	22.84c	25.61c	38.31b	39.96a	34.12a	29.23a
Root –shoot ratio	3.19a	2.50a	1.91b	1.58b	1.79b	2.80a
NAR ($mg.cm^{-2}.d^{-1}$)	0.151a	0.140a	0.099a	0.079a	0.115a	0.120b
RGR($mg.g^{-1}.d^{-1}$)	0.023a	0.019b	0.014c	0.011d	0.016a	0.017a

* = Means within a row followed by similar letters are non significant at P≤ 0.05 using Fisher's LSD within each category

Table 2. Mean values for various growth and development parameters of yarrow species subjected to water stress during 2005.

Parameter	Irrigation (days)				*Achillea*	
	Control	3	6	9	*millefolium*	*filipendulina*
Leaf area (cm^2)	3435a*	1378b	1074c	720d	1656a	1648a
Leaf weight (g)	33.46a	15.52b	11.61c	8.54d	19.02a	15.55b
Root weight (g)	70.10a	20.81b	14.03bc	10.11c	26.82a	30.69a
LAR	33.84a	38.78a	44.08a	38.70a	40.56a	37.15a
Root –shoot ratio	2.19a	1.37b	1.24b	1.21b	1.21b	1.79a
NAR ($mg.cm^{-2}.d^{-1}$)	0.520a	0.319a	0.260b	0.226b	0.303b	0.359a
RGR ($mg.g^{-1}.d^{-1}$)	0.059a	0.043b	0.038c	0.033c	0.038a	0.048a

* = Means within a row followed by similar letters are non significant at P≤ 0.05 using Fisher's LSD within each category

stomatal closure (Croker *et al*., 1998). It is likely possible that the closer of stomata in plants exposed to moisture stress might be one strategy that yarrow plants use to tolerate low moisture environment. Species responded differently to moisture levels in 2004, showing that stomatal sensitivity of plants varies with species (Balok and St.Hilaire, 2002; Croker *et al*., 1998). Variation in S_c by years suggests that species varies in response to planting season of the year as the experiment was conducted during fall to winter season in 2004; while in 2005 the same experiment was conducted in spring. Transpiration rate (T_s) was higher in control treatments compared with stressed treatments. High T_s in control treatment may be due to more availability of water. Transpiration may decrease in response to decrease in plant conductivity to water (Brodribb and Hill, 2000) and drought (Balok and St.Hilaire, 2002; Fernandez *et al*., 2002). T_s were lower for the drought stressed plants in the drought cycle 2 compared with later cycles in 2004, due to more foliage production. However, this trend was not observed in 2005, and the results were unknown. T_s of water stress imposed plants were consistently lower at all sampling date, indicating that drought may have reduced root hydraulic conductivity (Ramos and Kaufmann, 1979). Control treatments had high cell osmotic potential (ψ_s) than drought treatments. ψ_s decreased with increased drought stress showing that drought have reduced ψ_s. Since water was not a limiting factor for control treatment, therefore control plants showed maximum ψ_s compared with stressed treatments (Feser and St. Hilaire, 2005). The increase in solute concentration in drought exposed plants lowered ψ_s. The increase in cell solutes that is triggered by exposure to water deficits lowers the water potential at which stomatal closure occurs (Turner and Jones, 1980). Additionally cell solutes may play a significant role in maintenance of turgor and survival by protecting the plant from dehydration. The influx of water into the tissue will maintain turgor and enable the plant to continue growth despite the fact that plant is faced with moisture deficits (Chaves, 1991). *A. millefolium* deposited more solute than *Achillea filipendulina*; suggesting that it may perform better under drought condition. Relative water content (RWC) was highest for the control treatment which decreased with increase in water stress level and lowest RWC was recorded for plants watered every 9 days. High RWC in control may be due to the availability of more water (Feser and St. Hilaire, 2005). Photosynthetic activity may decreases when RWC drop down to 40-70% due to increased cell solutes which may inhibit enzymatic activity in chloroplast (Chaves, 1991). High RWC (74.50%) was recorded in *A. filipendulina* compared with *A. millefolium* (69.8%). RWC seems to have direct relation with carbon assimilation, which drop down to zero when RWC falls down below 70%, consequently the possibility of the growth ceases (Chaves, 1991). High RWC in *A. filipendulina* suggests that it may perform better under drought condition. Leaf area of control plants was twice high in 2004 and four times higher in 2005 than that of plants irrigated every 9 days. The greater leaf area in the following year would be associated with the higher photosynthetic radiation, which accelerate the plant growth and hence the leaf area. The increase in LA of unstressed plants was due to production of more and larger leaves. We observed that drought stressed plants resulted in drying of the leaves and thus LA decreased. Reduction in LA may be considered among the first morphological traits affected by water stress because cell expansion depends on the availability of water to maintain turgor (Hale and Orcutt, 1987). Drought reduced leaf weight (LW) and root weight (RW). LW and RW of the controls were over twice as high in 2004 and over four times as high in 2005 (Table 1, 2).

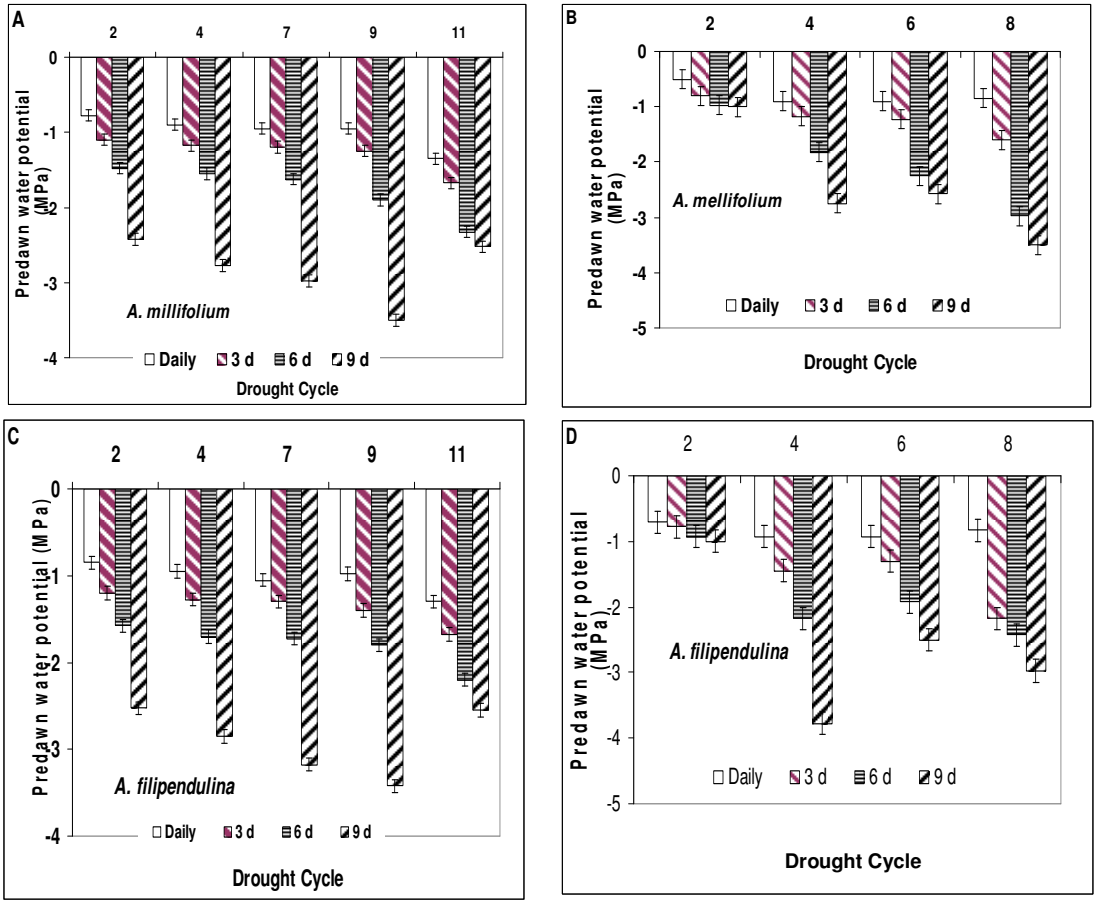

Fig 1. Predawn water potential of two yarrow species as affected by drought levels A,C= 2004, B,D=2005. Plants completed 11 drought cycles in 2004 and 8 drought cycles in 2005. Bar on column represent standard error.

The reduction in leaf and root weight was due to production of fewer and smaller leaves and roots in drought stressed plants compared with control. LW was significantly affected by S in both years (Table 1, 2) showing that LW is species dependent and *A. millefolium* produced heavy leaves than *A. filipendulina* even if planted in different growing seasons. Drought stress significantly affected leaf area ratio (LAR) in 2004 only. Control produced lowest LAR. LAR increased with each level of drought stress and highest LAR (39.96) was observed for plants in 2004 irrigated every 9 days (Table 1). High LAR values of drought stressed plants was due to reduction in total number of leaves, smaller LA of individual leaf and thus lighter leaves. Root-to-shoot dry weight ratio (RSR) of the control was highest which decreased with each stress level and lowest RSR was observed for plants irrigated every 9 days. It was observed that drought stress severely reduced shoot growth compared with root growth. Reduction in shoot growth under drought may be considered an avoidance mechanism which minimizes water losses (Ruiz-Sanchez *et al*., 2000). Species significantly affected NAR in 2004 only, while D showed non significant effect on NAR (Table 1,2). No statistical differences in RGR and NAR between *A. millefolium* and A *filipendulina* were observed except in 2004 where NAR was higher for A *filipendulina* *than A. millefolium*. These results showed that *A. filipendulina* is more sensitive to variation in environment compared with *A. millefolium*. Although the experiment was conducted in the greenhouse, yearly differences in mean maximum, minimum air temperature and relative humidity were noted that might have affected NAR. Drought showed significant effect on RGR in both years. RGR of the control was twice higher than plants irrigated every 9 days. Our results showed that RGR is sensitive to drought and may drastically reduce if plants are exposed to drought stress. Drought might reduce gas exchange, the growth of expanding tissues by reducing cellular expansion and productivity (Sanchez-Blanco *et al*., 2002, Nilsen and Orcutt, 1996).

Materials and methods

Experimental site

The experiments was conducted in a greenhouse at New Mexico State University, Las Cruces, N.M USA in 2004 and repeated in 2005. The experimental site is located at 1183 m elevation, 32° 16'4"N latitude, 106°46'18"W, longitude.

Experiment 1: Seeds of *Achillea millefolium* cv. Cerise Queen and *Achillea filipendulina* cv. Parkers Gold were sown in flat plastic trays on June 2, 2004. Plants were removed

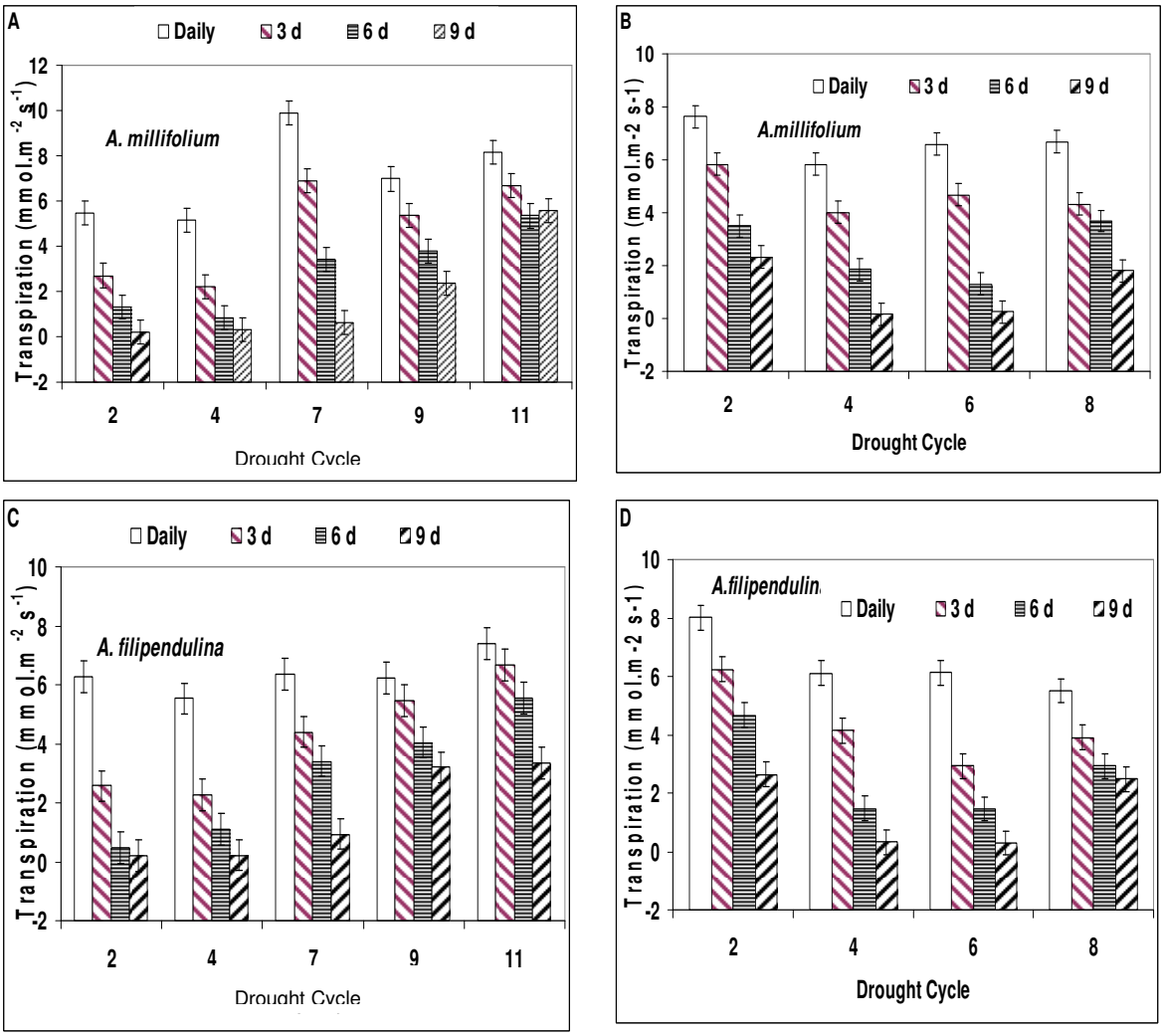

Fig 2. Stomatal conductance of two yarrow species as affected by drought levels A,C= 2004, B,D=2005. Plants completed 11 drought cycles in 2004 and 8 drought cycles in 2005. Bar on column represent standard error.

from the original flat plastic tray on July 19 and repotted into the plastic pots (3.8 liter) of 18 cm height and diameter. Pots were filled with growing substrate consisted of peat, composted bark, sphagnum peat, perlite and a wetting agent containing 0.01% available phosphorus derived from ammonium phosphate (The Scotts Advantage, Scott Company, Marysville, OH). The seedlings were watered every two days with automatic drip irrigation system and fertilized weekly with 2840 mg L^{-1} Peters solution containing 20:20:20 N:P_2O_5:K_2O, 0.05% each magnesium and iron (Scott Company, Marysville, OH). During the experimental period, daily maximum and minimum air temperature and humidity were recorded with portable thermo-hygrometer sensor (Spectrum Technologies, Plainfield, IL, USA) and is reported in Fig. 1A, B.

Experimental design and irrigation treatment

The experimental design was a randomized complete block design, with four irrigation levels (1 = control; irrigated daily, 2= irrigated every 3 days, 3 = irrigated every 6 days, and 4 = irrigated every 9 days) and two species (*Achillea millefolium* cv. Cerise Queen and *Achillea filipendulina* cv. Parkers Gold with four replications. In each pot, a single plant was left after harvesting four plants during initial harvest. These irrigation intervals were selected because they were associated with a similar degree of wilting assessed visually and plants were unable to tolerate stress longer than 9 days.

Control plants were irrigated daily with 1 liter of tap water and plants in the moisture deficit treatments were irrigated in cycles. Control plants were fertilized weekly with Peters at 2840 mg L^{-1} H_2O containing 20:20:20 N:P_2O_5:K_2O, 0.05% each magnesium and iron (Scott Company, Marysville, OH) . Moisture stressed plants were fertilized at the end of each drought cycle using the same fertilizer and quantity. During measurements, leaf temperature averaged 22±4°C. Maximum /minimum temperature in the greenhouse averaged 35± 2°C/12±1. Maximum relative humidity averaged 73± 8 /19±1%. Photosynthetically active radiation at canopy level averaged 964± 167 $\mu mols^{-1}m^{-2}.s^{-1}$. No artificial radiation was provided. Leaf temperature was measured with steady state porometer (LI-1600; LI-COR, Lincoln, NE, USA).

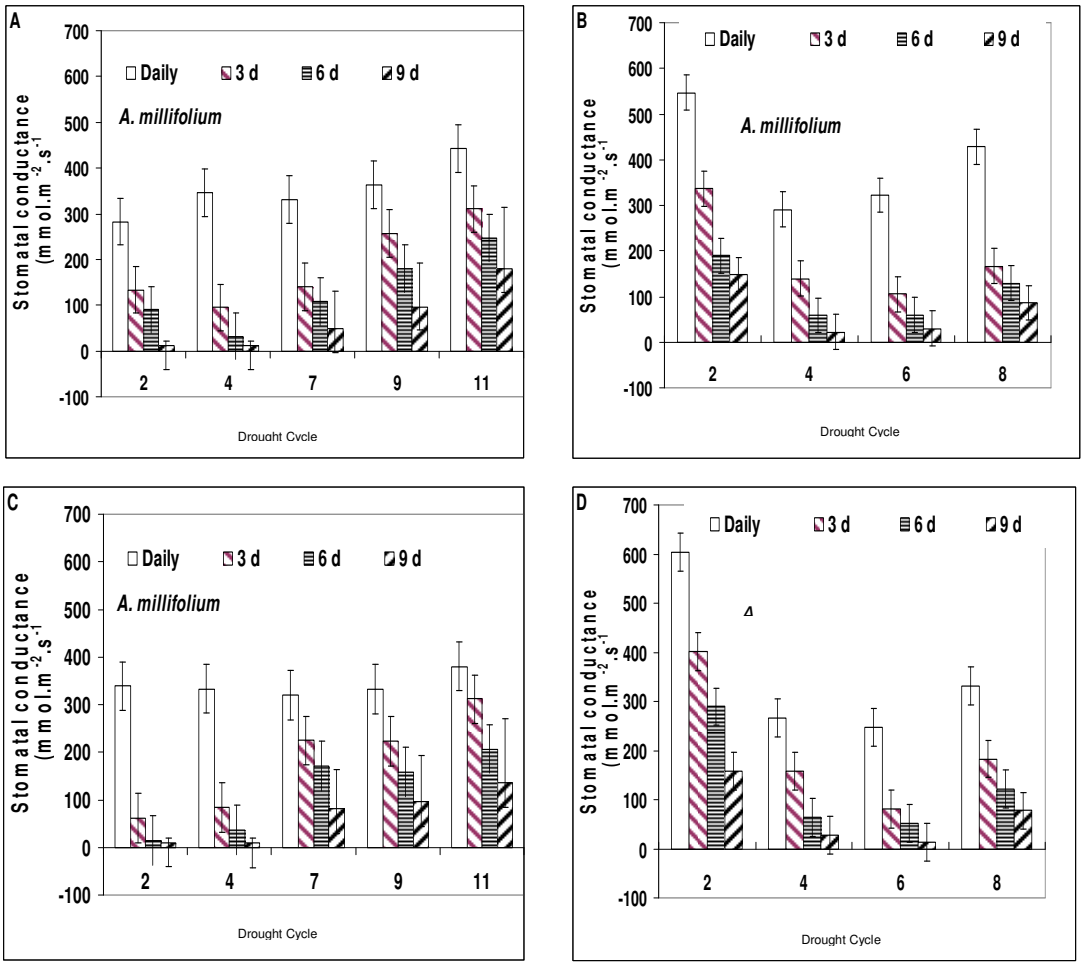

Fig 3. Transpiration of two yarrow species as affected by drought levels A,C= 2004, B,D=2005. Plants completed 11 drought cycles in 2004 and 8 drought cycles in 2005. Bar on column represent standard error.

Observations and measurements

Initial harvest

Four plants were randomly uprooted from each pot after three months of sowing. The fresh weight of seedlings was about 10 g pot^{-1}. The seedlings were separated into roots, leaves and stem. Roots were washed free of growing substrate using water. All components were oven dried for 14 d at 65° C.

Plant water relations and gas exchange

Predawn leaf water potential (ψ_{Pd}) was measured on young fully expanded leaves with a pressure chamber (Model 3005, Soil Moisture Equipment Corp, Santa Barbara, CA, USA) on the same day in all the treatments at the end of drought cycle, when coincided. Similarly, the transpiration rate (T_s) and stomatal conductance (S_c) were measured between 11:00 to 14:00 hr at same day, when drought cycle coincided with the help of steady-state porometer (LI-1600; LI-COR, USA) on youngest fully expanded leaf. All these measurements were made at the end of cycles 2, 4, 7, 9 and 11, at September 27, October 9, November 10, November 24 and December 15. On those dates the end of a drought cycle coincided for the 1, 3, 6 and 9 days treatments.

Cell osmotic potential (ψ_s)

Cell osmotic potential was measured at the end of drought cycles 4, 7, 9 and 11. A young fully expanded leaf was selected, excised and sealed in a zip lock plastic bag, placed on ice, immediately transported to laboratory and stored in freezer at -20° C in dark for 3-5 days. Leaves were removed from the freezer, rolled, placed into a Markhart leaf press (Model LP-27; Wescor, Logan, UT, USA) and pressed to squeeze out cell contents. A 10µL aliquot of the cell contents was transferred onto paper discs (SS-033 sample disc, Wescor, USA). Discs were then placed in a self calibrating vapor pressure osmometer (Vapro model 5520; Wescor, Logan, Utah) to determine cell osmolality. Values for cell osmolality (mmol kg^{-1}) were converted to cell osmotic potential (-MPa) using van't Hoff's equation.

Relative water content (RWC)

Relative water content was measured at the end of drought cycles 4, 7, 9 and 11. A young fully expanded leaf was selected, excised, sealed in a zip lock plastic bag, placed on ice and immediately transported to the laboratory. Each leaf was weighed to determine fresh weight (FW) and rehydrated in deionized water overnight. Each leaf was blotted with lintless paper to remove excess moisture and re-weighed to obtain turgid weight (TW). Each leaf was then dried in

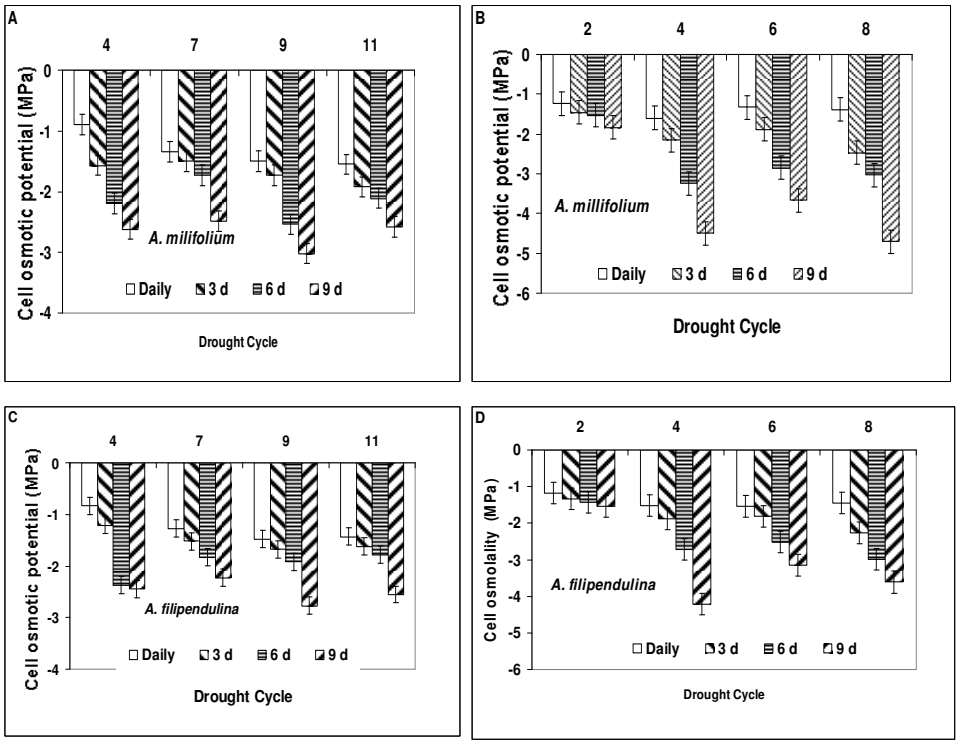

Fig 4. Cell osmotic potential of two yarrow species as affected by drought levels A,C= 2004, B,D=2005. Plants completed 11 drought cycles in 2004 and 8 drought cycles in 2005. Bar on column represent standard error.

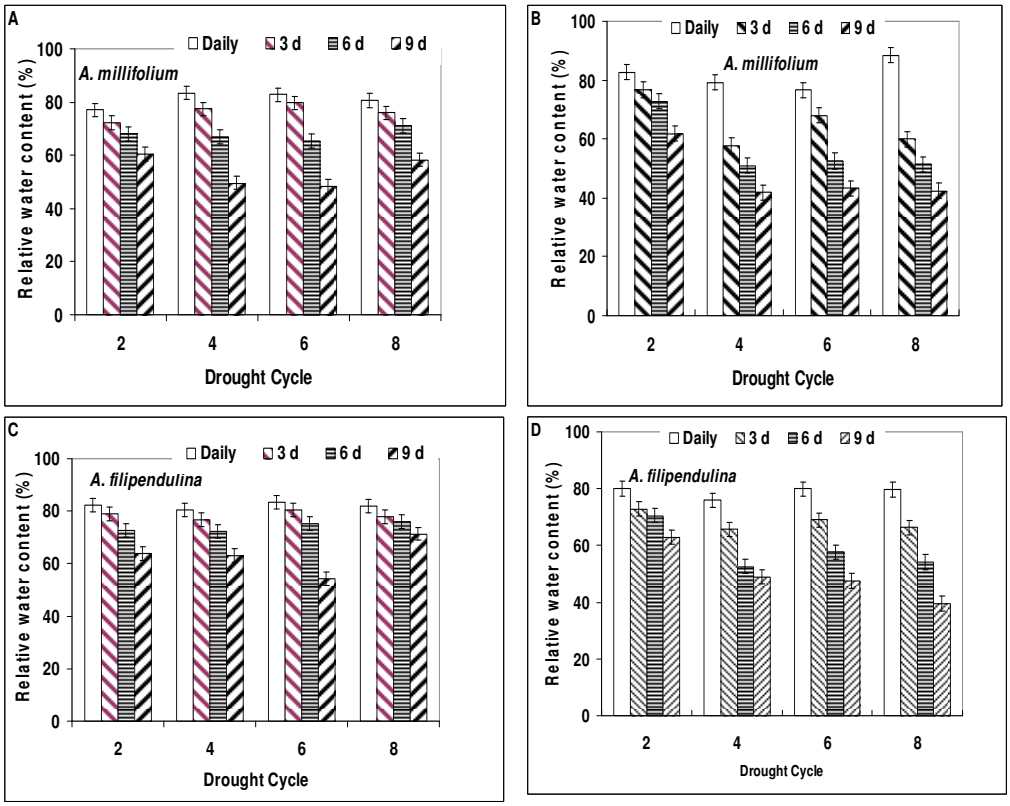

Fig 5. Relative water content of two yarrow species as affected by drought levels A,C= 2004, B,D=2005. Plants completed 11 drought cycles in 2004 and 8 drought cycles in 2005. Bar on column represent standard error.

an oven for 10 h at 85° C. Dry weight (DW) was recorded, and RWC was determined using the formula; RWC (%) = (FW-DW)/(TW-DW) ×100.

Final harvest and growth measurements

The experiment was terminated after 99 d of irrigation treatments. The experiment took 197 days as a whole starting from sowing the seed till final harvest. Leaves were cut 2 cm above the growing substrate surface. Leaf area of all plants was measured with the leaf area meter (LI 3000 A; LI-COR, Lincoln, Nebr.). Roots were water-washed free of growing substrate. Leaves and roots were oven dried at 65° C for 14 days at 65 ° C. Leaf surface area (LA) was measured with leaf area meter (LI 3000 A; LI-COR, Lincoln, Nebr, USA). The leaf area ratio (LAR), and root shoot ratio were calculated from the dry weights of roots, shoot and leaf area. Net assimilation rate (NAR) was calculated by using the equation of Harper (1977): Relative growth rate (RGR) was calculated using an equation modified from Gutschick and Kay (1995).

Experiment 2: Same treatments and experimental design were used for the 2nd experiment as was used for the 1st experiment. Seeds were planted on November 22, 2004, and plants were repotted into the plastic pots using the same procedures outlined in 1st experiment after three months. Irrigation treatments were initiated after two months of the repotting of the plants. In this experiment plants in the moisture stress treatments were irrigated in 8 cycles as against 12 cycles in experiment 1st. After completion of 4 cycles, irrigation interval was reduced to 5 and 7 days in treatments 3 and 4 respectively due to high evaporation losses after mid April. During drought treatments plants were fertilized as in experiment 1. During physiological measurements, leaf temperature averaged 22± 1°C. Maximum/minimum temperature in the greenhouse averaged 36±2°C/14±4°C, and relative humidity averaged 91± 8 /28±4%. Photosynthetically active radiation at canopy level averaged 1689± 200µmol $sm^{-2}s^{-1}$ and received no supplemental radiation. Data on ψ_{Pd}, S_c , T_s, ψ_s , RWC, NAR and RGR, and other growth parameters were recorded using procedures outlined in experiment 1. Data for ψ_{Pd}, S_c , T_s, ψ_s and RWC were recorded on March 23, April 11, April 25 and May 7, 2005. On May 11, 2005, plants were uprooted 67 days after 8th drought cycles. The experiment took 171 d from sowing to final harvest.

Statistical analysis

Data were analyzed using SAS/STAT software for windows Version 9.1 (SAS Inst. Cary, NC, USA 2004). Means of leaf area, leaf weight, root weight, LAR, root shoot ratio, NAR and RGR were separated using Fisher's least significant difference (LSD) at $P \leq 0.05$ after analysis of variance. The relationship of leaf water potentials, transpiration, stomatal conductance, relative water content and cell osmotic potential with drought cycle was analyzed using repeated measures of the Proc Mixed procedure of SAS (SAS Inst., 2004) to assess species (S) effect, drought treatment (D) effects, drought cycle (C) effect, and all interactions (S × D, S × C, S × D × C).

Conclusion

Drought stress reduced ψ_{pd} , S_c , T_S , ψ_s , RWC, LA, leaf and root weights, RSR and RGR compared to control plots where no water stress was imposed. Both species performed better in irrigated condition compared with drought stress. Cerise Queen might be more suited to the arid regions of Southwest as it produced larger and heavier leaves and showed better osmotic adjustment in response to drought which enabled it to deposit more solutes and lowered leaf water potential without dehydration than Parker Gold.

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