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Fatemeh Kazemi
Edith Cowan University

Mansoure Jozay

Farzaneh Salahshoor

Eddie van Etten
Edith Cowan University

Sahar Rezaie

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Drought Stress Responses of Some Prairie Landscape C4 Grass Species for Xeric Urban Applications

Fatemeh Kazemi [1,*], Mansoure Jozay [2], Farzaneh Salahshoor [3], Eddie van Etten [1,*] and Sahar Rezaie [4]

1 School of Science, Edith Cowan University, Perth 6027, Australia
2 Faculty of Plant Production, Gorgan University of Agricultural Sciences and Natural Resources, Gorgan 4913815739, Iran
3 Faculty of Agriculture, Ferdowsi University, Mashhad 9177948974, Iran
4 School of Architecture, Planning and Landscape, University of Calgary, Calgary, AB T2N 1N4, Canada
* Correspondence: f.kazemi@ecu.edu.au (F.K.); e.van_etten@ecu.edu.au (E.v.E.)

Abstract: Creating xeric landscapes in lawns and prairies is a significant challenge and practical need in arid urban environments. This study examined the drought resistance of some C4 grass species for constructing urban lawns and prairies. A factorial experiment based on randomized complete block designs with four replications was conducted. Experimental treatments were two irrigation levels (100% and 50% Field Capacity (FC)) and five warm-season grass species (*Andropogon gerardii* Vitman, *Sorghastrum nutans* (L.) Nash, *Panicum virgatum* L., *Schizachyrium scoparium* (Michx.) Nash, and *Bouteloua curtipendula* (Michx.) Torr.). The effects of drought on physiological, morphological, and qualitative characteristics of the grass species were analyzed. Drought conditions induced a decrease in all the measured traits. However, fewer physiological, morphological, and qualitative characteristics were affected by drought stress on *Andropogon gerardii*, *Schizachyrium scoparium*, and *Bouteloua curtipendula*, compared to the other two species. Overall, warm-season grasses of *Andropogon gerardii*, *Schizachyrium scoparium*, and *Bouteloua curtipendula*, had greater adaptability to drought stress, making them promising C4 grass species for prairie or lawn landscaping in arid urban environments. Landscape professionals and decision-makers should consider using *Andropogon gerardii*, *Schizachyrium scoparium*, and *Bouteloua curtipendula*, as these were the most resilient grass species for drought-tolerant prairie landscaping schemes. *Sorghastrum nutans* and *Panicum virgatum* may be used as a second priority if a more diverse variety of grasses is required for drought-resilient prairie or lawn landscaping in arid cities.

Keywords: drought stress; morphological traits; physiological traits; prairie landscaping; warm-season grasses; water-conserving landscaping

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1. Introduction

In recent decades, landscape designers and managers have shown interest in utilizing naturalistic plantings of native and introduced herbaceous perennials, which are sometimes termed prairie landscapes [1–3]. The cultivated introduced flora of many nation-states is much larger than the equivalent native flora, providing designers with a broader spectrum of species to maximize potential ecological and cultural fit. To be robust in practice, the introduced plants should be climatically well-adapted to their new climatic and soil conditions. As key environmental features (soils, microclimate, etc.) are often much changed within urban sites from the original conditions that local plants evolved to tolerate, it is unsound to believe that natives will automatically be better suited than introduced species. In many cases, introduced species can contribute to the region's biodiversity, particularly where they can provide habitat or features for other species [4]. Maintaining high levels of plant species diversity is widely known to enhance ecosystem stability and resilience and provides a greater range of ecological functions. However, in plant communities with one or a few dominant species, intraspecific diversity is important for such ecosystem characteristics [5]. Under certain urban scenarios, exotic species may more readily provide ecosystem services and health benefits than native species [6]. Where sufficient evidence is available for their adaptability and low potential for invasiveness, genetically improved and adapted exotic species can be valuable for increasing biodiversity, aesthetics, and amenity, as well as assisting with urban heat island mitigation and hydrology [7]. Such species can positively contribute to urban green areas, which are one of the fundamental infrastructures known to improve the ecological, social, and economic sustainability of cities [8,9]. As with native species, introduced herbaceous perennials have specific ecological requirements that must be satisfied if they are to establish and survive in competition with other plants within grassy vegetation [10]. One of the critical attributes of introduced plants to be sustainable in their new region is to be manageable over the long term with relatively low inputs of resources, such as water, nutrients, and carbon, as well as demanding minimal time and labor for maintenance [11].

Bahrani et al. [12] concluded that water stress is often the most limiting factor in urban landscaping of semi-arid and arid urban environments. Despite the shortage of water in many cities worldwide, irrigation of landscapes accounts for a considerable percentage (40–70%) of urban water consumption [13]. That is why several concepts such as xeriscaping (or xeric landscaping) and water-sensitive urban design, including their essential principles and strategies, have recently been introduced or researched in some cities [14–16]. Among the essential principles and strategies for xeric landscaping is selecting drought-tolerant plant species [17]. Drought stress is one of the most critical environmental factors limiting the growth of turfgrasses, particularly in urban areas where water availability for landscape irrigation is increasingly limited [13,18]. Turfgrasses often receive substantial water and fertilizer inputs; thus, low-input grasses that maintain acceptable quality levels are desirable in limited irrigation situations [19]. This has led to shifts in attitudes on which grass species are planted and how they are managed in turf cultures. Some relatively new ideas such as naturalistic, prairie, and meadow landscaping have been developed, mainly in Europe, in which growing grass species without mowing is encouraged. All these parallel concepts promise more attractive, natural-looking, and more low-input landscapes compared to the common turfgrass cultures for the future cities [1,2,20–22].

Although some knowledge has been gained on the adaptability and growth of grass species within urban environments, most of these research works were conducted under full irrigation regimes without examining drought stress conditions (e.g., see [23]). Knowledge of variability in drought resistance and its mechanisms is essential for selecting plant species, including grasses for arid landscape prairie landscaping. Such knowledge will help improve management strategies and develop drought-resistant grass species and cultivars for urban landscaping [24]. Further, with many areas of the globe likely to experience further warming and drying over the coming decades due to human-induced climate change, adapting to this scenario through purposely selecting more drought-tolerant plant varieties and species has been recommended for urban areas [25].

Some research, also, has been conducted on the tolerance of grasses to drought and their adaptability to sustainable arid environments [26–29]. However, the effects of drought have been mainly studied in C3 plants, and much remains unknown on the drought responses of C4 plants [30]. Recently, several studies have been conducted on morpho-physiological responses of cool-season grasses to water deficits; however, less information is available on the responses of warm-season grasses to drought stress [31–33].

As there is a limited number of genetically improved grasses in some developing countries to satisfy the interior and exterior landscaping needs, attention has been given to management strategies for controlling the potential invasiveness of introduced species in arid climate regions by, for example, planting them indoors or in other segregated environments, controlling their seed production by mowing [34], or through specific strategies to limit vegetative spread by runners or rhizomes. Using such management strategies, some genetically improved species such as *Cynodon dactylon* (couch grass) and *Bouteloua dactyloides* (buffalo grass) are currently widely used successfully as introduced drought-tolerant warm-season grass cultivars in many dryland countries, including Iran. The use of these species and other introduced cultivars can be considered with care where there are currently no or few native grass species available and can broaden the plant pallets available to landscape and interior designers. One of the most effective ways to control invasive species is to prevent them from being introduced in the first place. This strategy can be applied by monitoring and regulating the import and export of plant and animal species, implementing quarantine measures, and increasing public awareness about the risks associated with invasive species. In other words, detecting invasive species as early as possible and responding quickly to prevent them from establishing and spreading is very important. Early detection can be achieved through regular monitoring and surveillance programs, and rapid response can involve a range of measures such as mechanical control, manual removal, herbicide treatment, or biological control [1,35,36]. Mechanical control for grass-dominated systems commonly involves regular mowing, which can be costly. One study that examined the relationship between mowing frequency and maintenance costs found that increasing mowing frequency from once every four weeks to once every two weeks resulted in a 45% increase in maintenance costs for a turfgrass area [37].

This study aimed to determine and compare the responses to moderate drought stress of five introduced warm-season grass species. This study was conducted on these introduced warm-season grasses to establish their relative drought tolerance for their future use in sustainable urban development and for landscaping in Iran and countries with relatively similar climatic conditions. The findings should form a basis for introducing grasses in lawn and prairie landscaping in urban environments. The selected grass species for this study were highly water-use efficient compared to many other grass species, mainly because they were warm-season C4 plant species that can reduce their water loss through transpiration by closing their stomata in some photoperiods. The overall goal of this study was to assess drought stress responses in prairie landscape grass species potentially suited to xeric urban landscaping and to identify grass species that are well-adapted to drought conditions in urban landscapes with limited water resources to promote sustainable landscaping practices and reduce water usage in urban areas. The results of this research can be applicable and significant for all the urban areas worldwide that are increasingly facing water scarcity issues due to population growth and climate change. By identifying grass species that are more drought-tolerant and water-efficient, urban planners and landscape architects can design more sustainable urban landscapes requiring less water consumption. Such planting designs can reduce the overall demand for water in urban areas and promote more efficient use of limited water resources.

2. Material and Methods

The experiment of this study was a factorial based on a randomized complete block design with four replications. Grass species type (5 levels: *Andropogon gerardii* Vitman (big bluestem grass), *Sorghastrum nutans* (L.) Nash (Indian grass), *Panicum virgatum* L. (switch grass), *Schizachyrium scoparium* (Michx.) Nash (little bluestem), and *Bouteloua curtipendula* (Michx.) Torr. (sideoats grama) and drought treatment (2 levels: 100% field capacity (FC) and 50% field capacity (FC)) were used as the factors.

2.1. Site Description

The experiment was performed in a garden adjacent to Mashhad University, Mashhad, Iran (59°38′ E and 36°16′ N; elevation 989 m). The city's climate is arid to semi-arid, with cold winters and hot, dry summers. The average annual rainfall was approximately 250 mm, with very little rainfall occurring in spring and autumn. The average minimum and maximum mean annual temperatures were 4 °C and 22 °C, respectively (National Centers for Climatology, 2020). This site was selected for the study as it was located within the city environment. It was considered an appropriate representative urban landscape site for typical prairie landscaping practices by the landscape professionals in Mashhad and still gave us the privacy and protection required for conducting research.

2.2. Plant Material and Planting Conditions

Five warm-season C4 perennial grass species were used in this experiment: *Andropogon gerardii* (big bluestem grass), *Sorghastrum nutans* (Indiangrass), *Panicum virgatum* (switchgrass), *Schizachyrium scoparium* (little bluestem), and *Bouteloua curtipendula* (sideoats grama). These species were selected as they were regularly used in prairie landscaping in North America, where they are native to great plain grasslands. None of these five species are included on the Global Invasive Species Database [38], a global compendium of known invasive species maintained by experts in this field of research. Further, published literature was perused and expert consultations were sought to ensure the imported species had low potential for invasiveness in urban landscape plantings in the city of Mashhad before conducting this main study. The seeds of these specified plant species were imported in commercial packages from United States and Australian seed companies. Therefore, the seed packages had accredited scientific plant species names, together with information on their germination, viability, and purity, on their labels. Taxonomic identifications were carried out on the seeds and the seedlings before applying the irrigation treatment to ensure the authenticity of the species. The seeds were sown in pots (20 cm in diameter and 25 cm in depth) in the spring season. The pots were filled with a loam texture soil (pH = 7.5, EC = 3.5, FC = 25%, N = 14.98 mg/kg, P = 56.6 mg/kg, K = 409 mg/kg). The pots were kept in the mentioned garden outdoors to allow the outcomes to be applicable to outdoor landscaping conditions. The plants were kept well-watered up to 100% Field Capacity before imposing the drought stress conditions. Field Capacity was measured based on the method of Salter and Haworth [39]. In this method, pots of the same weight were filled with the soil of the same weight and were irrigated until they reached saturated conditions. The tops of the pots were covered with a plastic cover to prevent water evaporation and the water was allowed to freely drain from the drainage holes at the bottom of the pots. The pots were weighed for several consecutive days until constant weights were obtained. The soil moisture after reaching the constant weight of the pots was the field capacity (%). This soil moisture percentage was kept to 100% FC for well-watered irrigation treatment in the study.

2.3. Drought Treatments and Measured Parameters

Irrigation treatments were applied to keep the soil water content to 100% field capacity (FC) or 50% field capacity (FC), and the effects of these two different levels of soil water content on the plants were evaluated.

These irrigation treatments were applied to the pots using the following formula:

$$W = G \times FC\%$$

where W is the amount of water for irrigation of the pots (liter), G is the weight of the pots (kg), and FC is the percentage of moisture in the Field Capacity condition. The soil FC was 25% and irrigation intervals were every three days based on the calculations.

The drought stress and irrigation treatments were applied over six months of the experiment, starting from planting the seeds in the spring to examining the effect of drought on the emergence of the seedlings and continuing during the establishment of the grass species in the summer. All the parameters, including the morpho-physiological and qualitative parameters and the parameters related to plant growth, were measured at the end of the experiment.

Relative Water Content (RWC), Relative Water Loss (RWL), Relative Saturation Deficit (RSD), Electrolyte Leakage (EL), chlorophyll$_a$, chlorophyll$_b$, total chlorophyll, and carotenoids were measured on the leaves only, and root and shoot length and root fresh and dry weight were the factors measured on the whole plant.

2.3.1. Physiological Parameters

Relative water content (RWC) of the leaves was determined by measuring the fresh weight of five leaf samples, their turgid weight by soaking the leaf samples in distilled water for four hours, and their dry weight following the oven-drying of turgid leaf samples at 80 °C for at least 15 h using the following formula [40]:

$$\text{RWC (\%)} = (\text{fresh weight} - \text{dry weight}/\text{turgid weight} - \text{dry weight}) \times 100 \quad (1)$$

The relative water loss (RWL) of leaves was determined by dividing the leaf samples into two parts with the same weights. For the first part, fresh weight and then the turgid weight were measured by soaking the leaves in 25 mL of distilled water for 5 h. The second part was maintained at room temperature of 25 °C for 5 h, and then its wilting weight was measured. Finally, for estimating the dry weight, the sample was placed in an oven at 70 °C temperature for 24 h. RWL was calculated using the following formula [41]:

$$\text{RWL (\%)} = (\text{fresh weight} - \text{wilting weight}/\text{turgid weight} - \text{dry weight}) \times 100 \quad (2)$$

The leaves were taken and weighed immediately to start measuring Relative Saturation Deficit (RSD). Then, they were kept for 5 h in test tubes containing 25 mL distilled water at room temperature conditions. The water was removed from the leaf surfaces, and the leaves were weighed again to measure their turgid weight (saturated weight). Relative saturation deficit was calculated using the following formula [42].

$$\text{RSD (\%)} = (\text{Saturated weight} - \text{Fresh weight}/\text{Saturated weight}) \times 100 \quad (3)$$

Electrolyte Leakage (EL) was the next physiological parameter measured. Parts of the leaves were placed in the test tubes with 10 mL of distilled water, then the test tubes were shaken (using a mechanical shaker) for about 18 h, and the initial electrical conductivity was measured. The test tubes were then placed in an autoclave with a 121 °C temperature for 15 min. The secondary electrical conductivity measures were taken after the test tubes were cooled. EL was then calculated using the following formula [43]:

$$\text{EL (\%)} = (\text{initial electrical conductivity}/\text{secondary electrical conductivity}) \times 100 \quad (4)$$

Leaf chlorophyll and carotenoid contents were measured using Dere et al.'s [44] method. First, 0.2 g of fresh young leaf tissue was completely shredded and ground in a Chinese mortar with 5 mL distilled water. The resulting mixture was poured into distilled water in a 25 mL laboratory balloon. Then, 0.5 mL of the mixture was mixed with 4.5 mL of 80% acetone and centrifuged at 3500 rpm for 15 min. The chlorophyll absorbance was read at 645 and 663 nm wavelengths using a spectrophotometer (Bio Quest UK model CE 2502). Finally, the concentration of chlorophyll contents was obtained using the following equations:

$$\text{Chl}_a\ (\mu\text{g mL}^{-1}) = (12.5\text{OD}663) - (2.55\ \text{OD}645)$$

$$\text{Chl}_b\ (\mu\text{g mL}^{-1}) = (18.29\ \text{OD}645) - (2.58\ \text{OD}663)$$

$$\text{Chl.total}\ (\mu\text{g mL}^{-1}) = \text{Chl}_a + \text{Chl}_b$$

2.3.2. Qualitative Parameters

The leaf color was ranked using a visual scoring based on a 1–9 scale, as used in the USA's National Turfgrass Evaluation Program (NTEP), which is an accredited program for measuring the qualitative factors in turf grass species [45–47]. The lowest level (1) was a very deficient turf color (light green), and the highest level (9) represented the darkest green color which is the ideal visual color in the turfgrasses.

The leaf texture refers to the texture quality of the leaves in terms of roughness and softness and is related to the width of the leaves. Visual scoring was based on a 1–9 scale, as suggested in the USA National Turfgrass Evaluation Program (NTEP) [45]. Score 1 indicated the lowest value, which refers to broader leaf blades, 9 indicated the best or highest value, and numbers six or above were considered as acceptable qualities that show the leaves with finer textures or narrower leaves.

2.3.3. Plant Growth and Morphological Parameters

Plant height, leaf width, and root length were measured using a ruler. Shoot and root fresh and dry weights were measured with a digital scale [48]. Plant biomass (dry weight) was obtained following oven-drying at 65 °C until constant weights were obtained [15]. A destructive sampling method was used to measure root length and fresh and dry weight.

2.4. Statistical Analyses

The data were subjected to analyses of variance (ANOVA) to test for effects of species and treatment on the measured plant variables using the software package of JMP v.12. (Developed by JMP, a subsidiary of SAS Institute). We checked the assumptions of the ANOVA test, including normality and homogeneity of variances, and ensured that they were met before conducting the test. Comparisons of the means were conducted using Least Significant Difference (LSD) tests. The significance of between-treatment means was tested at 0.01 and 0.05 levels of probability. The bar graphs were drawn utilizing the Microsoft Excel software package.

3. Results

3.1. Physiological Parameters

The results of the analysis of variance showed that the effects of species, irrigation levels (drought treatment), and the interaction between the species and irrigation levels on physiological parameters of warm-season grasses were all significant ($p \leq 0.01$) (Table 1).

Table 1. Analysis of variance (mean of squares) related to physiological traits of warm-season grasses.

Factors	df	RWC (%)	RSD (%)	RWL (%)	EL (%)	Chl_a ($mgg^{-1}FW$)	Chl_b ($mgg^{-1}FW$)	Chl_T ($mgg^{-1}FW$)	Carotenoid ($mgg^{-1}FW$)
Block	3	6.08 ^{ns}	0.15 ^{ns}	2.82 ^{ns}	0.46 ^{ns}	1.01 *	30.43 ^{ns}	22.68 ^{ns}	36.62 ^{ns}
Species	4	1014.54 **	1.88 **	25.99 **	5.64 **	33.28 **	1004.59 **	341.39 **	325.40 **
Irrigation	1	3768.45 **	2.65 **	415.76 **	33.00 **	214.23 **	7019.32 **	5351.55 **	3612.28 **
Species × irrigation	4	376. 62 **	1.84 **	39.67 **	11.62 **	8.98 **	2870.35 **	284.10 **	266.02 **
Error	27	15.71	0.09	1.61	0.32	0.59	20.90	15.83	10.89

**, * and ^{ns} mean significant at probability levels of 1%, 5% and non-significant, respectively. $mgg^{-1}FW$ (milligram per gram of fresh weight).

Means of interaction effects of species and irrigation levels (Table 2) showed that the effect of irrigation on relative water content (RWC) was significantly different among the different species. RWC was at the highest level in all the grass species when irrigation was applied at 100% FC levels. Moreover, irrigation with 50% FC caused a significant decrease (approximately 40%) in RWC (Table 2). Drought-treated *Andropogon gerardii*, *Sorghastrum nutans*, and *Panicum virgatum* had lower RWC compared to the control (100% FC) irrigation treatment. However, in *Schizachyrium scoparium* and *Bouteloua curtipendula*, this reduction was not significant.

Table 2. Comparison of the means of the interaction effects of the grass species types and irrigation levels (100% and 50% FC) on physiological parameters of the grass species.

Grass Species	Irrigation Level (FC) (%)	RWC (%)	RSD (%)	RWL (%)	EL (%)	Chl_a ($mgg^{-1}FW$)	Chl_b ($mgg^{-1}FW$)	Total chl. ($mgg^{-1}FW$)	Carotenoid ($mgg^{-1}FW$)
A. gerardii	100	51.62 ^{b}	16.26 ^{d}	68.43 ^{a}	38.11 ^{f}	11.35 ^{a}	2.04 ^{c}	13.4 ^{b}	1.04 ^{d}
A. gerardii	50	25.05 ^{e}	26.95 ^{c}	53 ^{cd}	82.56 ^{b}	8.86 ^{cd}	1.47 ^{cd}	10.34 ^{d}	1.11 ^{cd}
S. nutans	100	33.92 ^{d}	15.97 ^{d}	51.25 ^{cd}	24.47 ^{g}	10.77 ^{ab}	1.56 ^{cd}	12.33 ^{bc}	1.1 ^{cd}
S. nutans	50	17.09 ^{f}	48.12 ^{a}	19.82 ^{f}	63.77 ^{c}	8.55 ^{d}	1.56 ^{c}	11.53 ^{cd}	1.42 ^{bcd}
P. virgatum	100	60.17 ^{a}	25.4 ^{c}	62.13 ^{b}	94.61 ^{a}	8.06 ^{d}	3.23 ^{b}	11.29 ^{cd}	1.49 ^{bc}
P. virgatum	50	20.91 ^{ef}	43.18 ^{b}	31.87 ^{e}	55.41 ^{d}	2.5 ^{f}	1.45 ^{cd}	3.96 ^{f}	1.64 ^{b}
Sch. scoparium	100	59.02 ^{a}	17.73 ^{d}	55.62 ^{c}	47.78 ^{e}	10.77 ^{ab}	5.33 ^{a}	16.1 ^{a}	2.77 ^{a}
Sch. scoparium	50	53.06 ^{ab}	23.06 ^{c}	49.76 ^{d}	78.92 ^{b}	4.65 ^{e}	11.93 ^{cd}	6.58 ^{e}	1.39 ^{bcd}
B. curtipendula	100	51.12 ^{b}	16.37 ^{d}	61.87 ^{b}	27.93 ^{g}	9.94 ^{bc}	5.69 ^{a}	15.9 ^{a}	3.04 ^{a}
B. curtipendula	50	42.68 ^{bc}	45.41 ^{ab}	29.19 ^{e}	84.71 ^{b}	3.19 ^{f}	1.19 ^{d}	4.37 ^{f}	1.31 ^{bcd}

The same letter in each column indicates non-significant differences. RWC: Relative Water Content, RSD: Relative Saturation Deficit, RWL: Relative Water Loss, EL: Electrolyte Leakage, Chl_a: $Chlorophyll_a$, Chl_b: $Chlorophyll_b$, Total chl.: Total Chlorophyll, $mgg^{-1}FW$ (milligram per gram of fresh weight).

Means of interaction effects of species and irrigation levels on relative water loss (RWL) showed that a decrease in irrigation level caused a significant reduction in RWL ($p \leq 0.01$; Tables 1 and 2). The highest percentage of RWL was observed in *Andropogon gerardii* at 100% FC (68.43%), and the lowest rate of RWL was related to *Sorghastrum nutans* at 50% FC (19.82%). In *Sorghastrum nutans*, *Panicum virgatum*, and *Bouteloua curtipendula*, an approximately 50% reduction in RWL was observed following the irrigation level decrease.

Interaction effects between irrigation levels and grass species for the relative saturation deficit (RSD) showed that in all the grass species, RSD decreased with irrigation level. RSD was significantly different amongst species at 50% FC, whereas all species had statistically similar RSD at 100% FC, except for *P. virgatum*, which has an only slightly higher RSD than the rest (Table 2). The greatest RSD occurred in *Sorghastrum nutans* grown in 50% field capacity with a 48% deficit, which was statistically similar to that of *Bouteloua curtipendula* at 50% FC irrigation. In contrast, *Schizachyrium scoparium* possessed the lowest RSD in 50% FC (23% deficit; Table 2).

Means of interaction effects of grass species and irrigation levels on electrolyte leakage showed that a decrease in irrigation level caused a significant increase in electrolyte leakage in almost all the grass species. Moreover, in all the grass species, the reduction in irrigation level to 50% FC caused more than a 50% increase in electrolyte leakage. This increase was lower in *Schizachyrium scoparium* compared to the other grass species.

Means of interaction effects of species and irrigation levels showed that the decrease in irrigation level was associated with a reduction in $chlorophyll_a$, $chlorophyll_b$, and total chlorophyll in all the plant species (Table 2). This reduction was more considerable in *Panicum virgatum*, *Schizachyrium scoparium*, and *Bouteloua curtipendula*. In *Andropogon gerardii* and *Sorghastrum nutans*, irrigation with 50% FC did not significantly differ in total chlorophyll compared to when these plants were irrigated at 100% FC.

Means of interaction effects of grass species and irrigation levels on carotenoid showed that decreasing irrigation level in *Schizachyrium scoparium* and *Bouteloua curtipendula* caused a significant reduction in carotenoid amount compared to the carotenoid in the plants when they were treated with 100% FC irrigation water. However, in *Andropogon gerardii*, *Sorghastrum nutans*, and *Panicum virgatum*, no significant change in carotenoid amount was observed when any of the two irrigation levels were applied (Table 2).

3.2. Qualitative, Morphological and Plant Growth Parameters

The results of the analysis of variance showed that the effects of grass species, irrigation levels, and the interaction between the species and irrigation levels, on warm-season grasses, were significant for most qualitative parameters ($p \leq 0.05$; Table 3). In the case of attributes of color, texture, root length, root and shoot fresh weight, root and shoot dry weight, the interaction effect of the grass species and irrigation levels was significant ($p \leq 0.05$). Among the morphological characteristics, only simple effects of the grass species and irrigation levels were significant for plant height and leaf width (Table 3).

Table 3. Analysis of variance (mean of squares) of the morphological, qualitative, and plant growth parameters of the grass species.

Factors	df	Color (1–9 Scores)	Texture (1–9 Scores)	Height (cm)	Leaf Width (cm)	Root Length (cm)	Shoot Fresh Weight (g)	Root Fresh Weight (g)	Shoot Dry Weight (g)	Root Dry Weight (g)
Block	3	0.02 ^{ns}	0.03 ^{ns}	8.07 ^{ns}	0.00 ^{ns}	0.77 *	0.26 ^{ns}	0.02 ^{ns}	0.21 ^{ns}	0.00 ^{ns}
Species	4	26.27 **	9.90 **	341.39 **	0.18 **	79.57 **	17.36 **	12.02 **	2.81 **	0.45 **
Irrigation	1	3.02 **	8.10 **	104.65 **	0.00 ^{ns}	548.41 **	53.10 **	13.87 **	9.86 **	0.54 **
Species × irrigation	4	0.27 *	0.10 *	13.49 ^{ns}	0.00 ^{ns}	53.68 **	13.67 **	4.69 **	2.54 **	0.05 **
Error	27	0.09	0.03	7.44	0	2.19	0.39	0.14	0.13	0

**, * and ns mean significant at probability levels of 1%, 5% and non-significant, respectively.

Means of interaction effects of the grass species and irrigation levels on color (Table 4) showed that although in all the grass species, a quality reduction in color of the grasses was observed when irrigation level was reduced from 100% FC to 50% FC, this reduction was only significant in *Panicum virgatum*. In general, th highest (deepest green) color values belonged to *Bouteloua curtipendula* in both irrigation levels.

Table 4. Comparison of the means of interaction effects between irrigation levels and grass species on morphological, qualitative, and plant growth parameters of the grass species.

Grass Species	Irrigation Levels (FC) (%)	Leaf Color (1–9 Scores)	Leaf Texture (1–9 Scores)	Highest Length of Root (cm)	Shoot Fresh Weight (g)	Root Fresh Weight (g)	Shoot Dry Weight (g)	Root Dry Weight (g)
A. gerardii	100	5 ^{c}	4 ^{e}	28.3 ^{b}	12.99 ^{ef}	7.01 ^{a}	2.72 ^{d}	1.01 ^{a}
A. gerardii	50	4.5 ^{cd}	3 ^{f}	12.01^{g}	11.66 ^{f}	5.3 ^{b}	2.36 ^{d}	0.7 ^{cd}
S. nutans	100	4 ^{de}	6 ^{b}	22.28 ^{cd}	18.72 ^{a}	6.33 ^{b}	4.96 ^{a}	0.62 ^{de}
S. nutans	50	3.9 ^{de}	5 ^{c}	17.76 ^{f}	12.88 ^{e}	2.87 ^{e}	2.48 ^{d}	0.25 ^{f}
P.virgatum	100	6 ^{b}	5 ^{c}	30.75 ^{a}	17.55 ^{b}	4.52 ^{d}	4.74 ^{a}	0.88 ^{b}
P. virgatum	50	5 ^{c}	4.5 ^{d}	24.09 ^{c}	13.37 ^{e}	5.07 ^{cd}	2.8 ^{d}	0.91 ^{ab}
Sch. scoparium	100	4 ^{de}	7 ^{a}	27.56 ^{b}	15.45 ^{cd}	3.28 ^{e}	3.6 ^{c}	0.6 ^{e}
Sch. scoparium	50	3.25 ^{e}	6 ^{b}	21.09 ^{de}	14.93 ^{d}	2.24 ^{f}	3.52 ^{c}	0.23 ^{f}
B.curtipendula	100	7 ^{a}	6 ^{b}	23.14 ^{cd}	15.65 ^{cd}	5.15 ^{c}	4.16 ^{b}	0.9 ^{b}
B. curtipendula	50	6.5 ^{ab}	5 ^{c}	20.04 ^{e}	16 ^{c}	4.93 ^{cd}	4.05 ^{bc}	0.76 ^{c}

The same letter in each column indicates non-significant differences.

Regarding the texture of the grasses, reducing the level of irrigation for all the grass species was associated with significantly lower-quality (coarser-textured) grasses. The coarsest texture among all the plant species was observed in *Andropogon gerardii* under a 50% FC irrigation regime. The finest texture was observed in *Schizachyrium scoparium* in 100% FC (Table 4).

Means of interaction effects of the grass species and irrigation levels on root length showed that decreasing irritation level down to 50% FC caused reductions in root length in all the species. This reduction in *Andropogon gerardii* was more remarkable than the other species. An approximately 50% reduction in root length was observed in this species when the irrigation level of 50% FC was applied (Table 4).

Means of interaction effects of the grass species and irrigation levels on shoot fresh weight showed that a maximum amount of the shoot weight was related to *Panicum virgatum* and *Sorghastrum nutans* in 100% FC but that drought stress caused significant reduction in shoot fresh weight of both species. In the other species, no significant difference was observed in any of the two irrigation levels (Table 4).

Means of interaction effects of species and irrigation on fresh root weight showed that a maximum amount of weight was related to *Andropogon gerardii* and *Sorghastrum nutans* in 100% FC and that drought stress caused a significant reduction of fresh root weight of these species. This trend was also observed in *Schizachyrium scoparium*, *Sorghastrum nutans*, and *Schizachyrium scoparium.* In the two other species, no significant differences in this trait were measured between the two irrigation levels (Table 4).

Means of interaction effects of the grass species and irrigation levels on dry shoot weight showed that the highest dry weight was obtained from *Panicum virgatum* in irrigation treatment of 100% FC and that drought stress caused a significant reduction in dry shoot weight of this plant species. In the other species, no significant difference was measured when either of the two irrigation levels was applied (Table 4).

Means of interaction effects of the grass species and irrigation levels on root dry weight of *Panicum virgatum* under two irrigation levels showed no significant differences. However, in the other four plant species, a reduction in the irrigation level was associated with reductions in the plant species' root dry weight (Table 4).

Means of simple effects of the grass species on the height of plants showed that *Sorghastrum nutans* and *Schizachyrium scoparium* had lower heights compared to the other three plant species. Still, there were no significant differences in this trait among these three species (Figure 1).

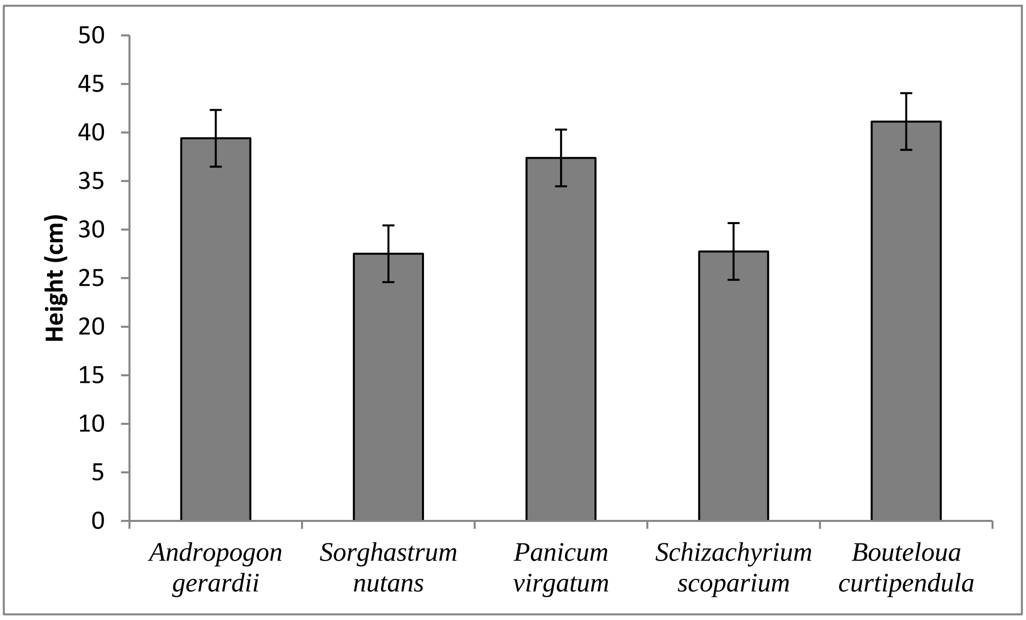

Figure 1. The simple effect of the grass species on the height of the grass species. The same letter in each column indicates non-significant differences. Error bars show +/− 1 standard error.

Simple effects of irrigation levels on height of the grasses showed that decreasing level of irrigation caused a significant reduction (of approximately 3 cm) in the height of the plants (Figure 2).

Means of simple effects of the grass species showed significant differences in the width of leaves among the species. The widest leaves belonged to *Andropogon gerardii*, and the narrowest leaves belonged to *Bouteloua curtipendula*. The three species of *Sorphastrum nutans, Panicum virgatum*, and *Schizachyrium scoparium* were the species with medium-width leaves (Figure 3). There is a public preference for narrow-leaf grass species in Iran. This study showed that the effect of plant species only (not the irrigation and drought regime) on the leaf width of the grasses was significant.

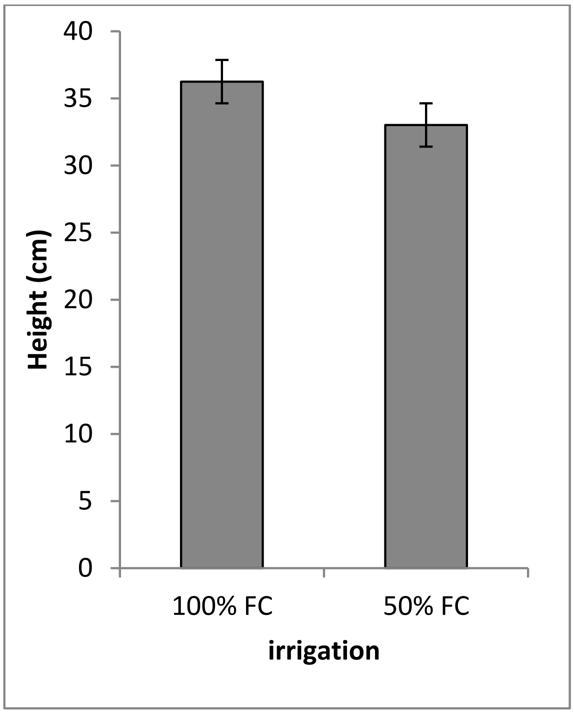

Figure 2. The simple effect of the irrigation level on the height of the grass species. The same letter in each column indicates non-significant differences. Error bars show +/− 1 standard error.

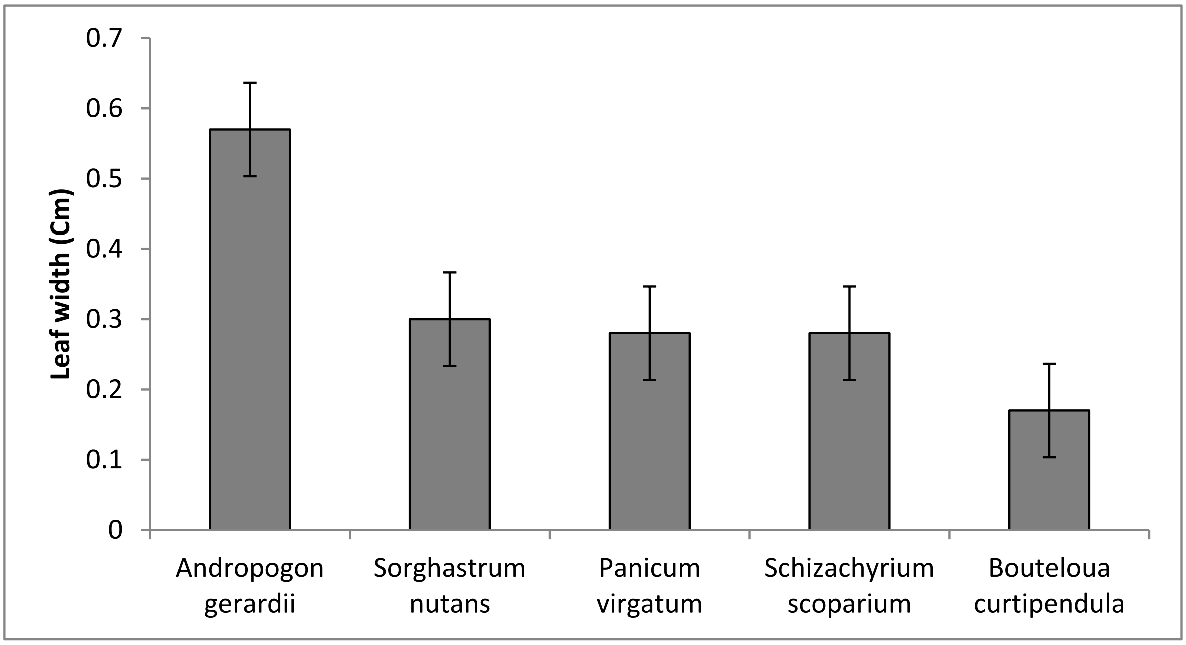

Figure 3. The simple effect of the irrigation leaves on width of the leaves of the grass species. The same letter in each column indicates non-significant differences. Error bars show +/− 1 standard error.

In this study, $chlorophyll_b$ showed a positive and significant correlation ($p \leq 0.01$, R = 0.5) with $chlorophyll_a$. However, it was negatively correlated with the number of days to germination ($p \leq 0.05$, R = −0.36) (Table 5).

Table 5. The Pearson correlation coefficient between the physiological, morphological, qualitative, and plant growth parameters measured in the grass species.

	Days to Germination	Chl_a ($mgg^{-1}FW$)	Chl_b ($mgg^{-1}FW$)	Total chl. ($mgg^{-1}FW$)	Carot. ($mgg^{-1}FW$)	Leaf Width (cm)	No. of Tillers	Root Length (cm)	Shoot Fresh Weight (g)	Root Fresh Weight (g)	Shoot Dry Weight (g)	Root Dry Weight (g)	RWC (%)	RSD (%)	RWL (%)	El. (%)
Days to germination																
Chl_a ($mgg^{-1}FW$)	0.25 ^{ns}															
Chl_b ($mgg^{-1}FW$)	−0.36 *	0.51 **														
Total chl. ($mgg^{-1}FW$)	0.047 ^{ns}	0.94 **	0.77 **													
Carot. ($mgg^{-1}FW$)	−0.47 **	0.25 ^{ns}	0.92 **	0.54 **												
Leaf width (cm)	0.88 **	0.33 *	−0.37 *	0.1 ^{ns}	−0.47 **											
No. of tillers	−0.13 ^{ns}	0.08 ^{ns}	0.26 ^{ns}	0.16 ^{ns}	0.29 *	0.03 ^{ns}										
Root length (cm)	−0.05 ^{ns}	0.17 ^{ns}	0.32 *	0.24 ^{ns}	0.23 ^{ns}	−0.25 ^{ns}	−0.08 ^{ns}									
Shoot fresh weight (g)	−0.55 **	0.81 ^{ns}	0.15 ^{ns}	0.12 ^{ns}	0.12 ^{ns}	−0.5 **	−0.11 ^{ns}	0.45 **								
Root fresh weight (g)	0.43 **	0.3 ^{ns}	−0.24 ^{ns}	0.13 ^{ns}	−0.25 ^{ns}	0.39 *	−0.49 **	0.13 ^{ns}	0.10 ^{ns}							
Shoot dry weight (g)	−0.52 **	0.07 ^{ns}	0.17 ^{ns}	0.12 ^{ns}	0.14 ^{ns}	−0.48 **	−0.11 ^{ns}	0.43 **	0.97 **	0.15 ^{ns}						
Root dry weight (g)	0.32 *	0.1 ^{ns}	0.1 ^{ns}	0.1 ^{ns}	0.1 ^{ns}	0.15 ^{ns}	−0.43 **	0.42 **	0.07 ^{ns}	0.77 **	0.18 ^{ns}					
RWC (%)	−0.15 ^{ns}	0.24 ^{ns}	0.3 **	0.35 *	0.33 *	−0.20 ^{ns}	0.43 **	0.66 **	0.44 **	−0.07 ^{ns}	0.46 **	0.2 ^{ns}				
RSD (%)	−0.18 ^{ns}	−0.66 **	−0.35 *	−0.63 **	−0.23 ^{ns}	−0.21 ^{ns}	−0.33 *	−0.35 *	−0.29 *	−0.27 ^{ns}	−0.31 *	−0.21 ^{ns}	−0.6 **			
RWL (%)	0.37 *	0.58 **	0.31 *	0.6 **	0.17 ^{ns}	0.30 ^{ns}	0.21 ^{ns}	0.45 **	0.19 ^{ns}	0.37 *	0.26 ^{ns}	0.46 **	0.68 **	−0.88 **		
El. (%)	0.12 ^{ns}	−0.52 **	−0.34 *	−0.52 **	−0.34 *	0.04 ^{ns}	0.02 ^{ns}	−0.21 ^{ns}	−0.14 ^{ns}	−0.41 **	−0.1 ^{ns}	−0.2 ^{ns}	0.005 ^{ns}	0.46 **	−0.26 ^{ns}	

**, * and ^{ns} mean significant at probability levels of 1%, 5% and non-significant, respectively. Sample: 40 number., Chl_a: $Chlorophyll_a$ ($mgg^{-1}FW$), Chl_b: $Chlorophyll_b$ ($mgg^{-1}FW$), Total chl.: Total Chlorophyll ($mgg^{-1}FW$), Carot.: Carotenoid ($mgg^{-1}FW$), RWC: Relative Water Content (%), RSD: Relative Saturation Deficit (%), RWL: Relative Water Loss (%), EL: Electrolyte Leakage (%).

Total chlorophyll had a significantly positive correlation with $chlorophyll_a$ and $chlorophyll_b$ ($p \leq 0.01$, R = 0.94, 0.76). There was a significant positive correlation between carotenoid and $chlorophyll_b$ and total chlorophyll content at the 1% probability level, respectively (R = 0.91, 0.54). However, negative correlations were observed between carotenoid level and the days to germination ($p \leq 0.01$, R = −0.46).

According to Table 5, leaf width was significantly and positively correlated, respectively ($p \leq 0.05$, R = 0.88, 0.32) with the number of days to germination and $chlorophyll_a$, but negatively correlated with $chlorophyll_b$ and carotenoid, respectively ($p \leq 0.05$, R = −0.37, −0.47). A significantly positive correlation was observed between the number of tillers and the carotenoid content ($p \leq 0.05$, R = 0.29). Root length had a positive and significant correlation with $chlorophyll_b$ ($p \leq 0.05$, 0.32). Correlation coefficients between the studied traits also showed a positive correlation between the fresh shoot weight and root length ($p \leq 0.01$, 0.45). However, shoot fresh weight was negatively correlated with the number of days to germination and leaf width, respectively ($p \leq 0.01$, R = −0.55, −0.5) (Table 5).

There was a positive and significant correlation between root fresh weight and the number of days to germination and leaf width, respectively ($p \leq 0.05$, R = 0.42, 0.39). However, a negative correlation was observed between root fresh weight and the number of tillers ($p \leq 0.05$, R = −0.49). In this study, there was a positive and significant correlation between shoot dry weight with root length and shoot fresh weight, respectively ($p \leq 0.01$, R = 0.43, 0.97), but a negative correlation was observed between shoot dry weight and the number of days to germination and leaf width respectively ($p \leq 0.01$, R = −0.52, −0.48).

Root dry weight was positively correlated with the number of days to germination, long root length, and fresh root weight, respectively ($p \leq 0.05$, R = 0.31, 0.42, 0.77). However, a negative correlation was found with the tiller number ($p \leq 0.01$, R = −0.43). RWC showed a significant and positive correlation at a 5% probability level with $chlorophyll_b$, total chlorophyll, carotenoid, number of tillers, root length, and fresh and dry weight of shoots, respectively (R = 0.3, 0.34, 0.33, 0.43, 0.66, 0.44, 0.46) (Table 5).

In this study, RSD was negatively correlated with $chlorophyll_a$, $chlorophyll_b$, total chlorophyll, number of tillers, root length, shoot fresh and dry weight, and RWC, respectively ($p \leq 0.05$, R = −0.66, −0.35, −0.63, −0.33, −0.35, −0.29, −0.31, −0.6).

There was a significant positive correlation between RWL and the number of the days to germination with $chlorophyll_a$, $chlorophyll_b$, total chlorophyll, root length, fresh and dry root weight, and RWC, respectively ($p \leq 0.05$, R = −0.36, 0.58, 0.31, 0.55, 0.45, 0.37, 0.46, 0.68). In contrast, RWL was negatively correlated with RSD ($p \leq 0.01$, −0.88). There was a positive and significant correlation between electrolyte leakage and RSD ($p \leq 0.01$, 0.46) but a negative correlation with $chlorophyll_a$, $chlorophyll_b$, total chlorophyll, carotenoid, and fresh root weight, respectively ($p \leq 0.05$, R = −0.52, −0.34, −0.52, −0.34, −0.41) (Table 5).

4. Discussion

In this study, water stress negatively affected most of the measured traits in all five C4 grass species. C4 grasses have a unique carbon-fixing pathway and keep a more steady-state stomata aperture and higher water use efficiency than C3 grass species, which helps them conserve water [49].

Bahrani et al. [12] studied ten forage types of grass. They also found that an increase in water stress decreased total water use efficiency, decreased plant height, leaf water potential, leaf area, and root dry weight in almost all the species with varying degrees of reduction. As shown in Table 6, general conclusions can be drawn to identify the resistance potential of the five studied C4 grass species to drought stress. *Andropogon gerardii*, *Schizachyrium scoparium*, and *Bouteloua curtipendula* had fewer physiological, morphological, and qualitative characteristics (nine out of 15 measured factors) affected negatively by reduced irrigation (maintained at 50% FC) compared to the control irrigation treatment of 100% FC (Table 6). However, in the other two species, *Sorghastrum nutans* and *Panicum virgatum*, 12 out of the 15 measured factors showed a significant reduction in plant performance when irrigation was reduced to the level of 50% FC. Therefore, this clearly suggests greater tolerance of *Andropogon gerardii*, *Schizachyrium scoparium*, and *Bouteloua curtipendula* (relative to the other species) to drought stress. These three species did not show sizeable reductions in the above-ground biomass (both fresh and dry weight) with deficit irrigation, which are essential positive characteristics in landscaping within arid and semi-arid cities. There are disagreements towards developing effective strategies for xeric landscaping (xeriscaping) because lowered plant biomass is commonplace in dry urban landscapes [15]. As the three mentioned species showed no decrease in their aerial plant biomass, they can contribute to creating urban landscapes with higher water use efficiency while keeping their original biomass during drought stresses of up to 50% FC. Plant biomass is an important indicator of health, aesthetic, and biodiversity in urban landscapes [13,15,16,25]. Therefore, the mentioned plant species are appropriate candidate plants for water-conserving landscaping schemes in the urban environment.

The cells with grey shading indicate where the measured factor was significantly different between irrigation treatment of 50% FC and the control irrigation treatment (100% FC). The cells with no shading showed which variables were not significantly different between treatment (50% FC) and control (100% FC) irrigation treatment for each species. While it was confirmed that *Andropogon gerardii*, *Schizachyrium scoparium*, and *Bouteloua curtipendula* did not significantly alter their aerial biomass even when their irrigation is reduced to 50% of the field capacity of their soil, *Bouteloua curtipendula* even demonstrated insignificant root dry weight changes when drought stress was applied.

Among the three drought-tolerant types of grass identified in this study, *S. scoparium* overall performed better under drought stress, as it did not show sizeable changes in its leaf water content (as measured by RWC, RWL, and RSD). *Bouteloua curtipendula* also maintained its RWC during the deficit irrigation treatment (50% FC) compared to the control (100% FC) almost as much as *S. scoparium*; however, in contrast, *A. gerardii* demonstrated a significant reduction in RWC (a near 50% decline). Maintaining consistent leaf RWC during drought conditions means maintaining cell turgidity and not wilting, which are important qualities for selective urban landscape plant species subject to drought stress conditions. In general, increasing drought stress can cause an increase in damage to cell membranes [41]. Consequently, it can cause an increase in electrolyte leakage and reduction in the swelling of the leaves, so species with high RWC and RWL and low RSD and EL are more tolerant to drought stress [50]. The better performance of *S. scoparium* compared to *A. gerardii* has also been found in previous studies [30]. Heckathorn and DeLucia [51] also confirmed that *A. gerardii* is an intermediate species in comparison to the highly xeric and drought-tolerant *S. scoparium*. Our study showed that *Panicum virgatum* was the only species to record lower electrolyte leakage during drought stress despite experiencing a very large reduction in its leaf RWC, which suggests this species may have different mechanisms to tolerate drought compared to the other species.

The results showed that leaf chlorophyll content decreased with drought stress in all the species. Chlorophyll content is representative of the ability of the plants to photosynthesize. Positive relationships have been found between chlorophyll content and the nitrogen content of the leaves. One of the reasons for decreasing photosynthesis during the drought condition is low foliar nitrogen content, because most of the nitrogen available in the young leaves is usually present in photosynthesis-related components such as photosynthetic enzymes [52]. In an experiment conducted by Heckathorn et al. [53], photosynthetic capacity decreased by 69 to 78% during drought in recently developed leaves of *A. gerardii* and *S. scoparium*. It was concluded that reductions could explain approximately one-third to one-half of this decrease in leaf N concentration and chlorophyll content. At the same time, the remainder was attributed to protective down-regulation or damage to photosynthetic metabolisms. The loss of chlorophyll content during drought stress could also be related to photo-oxidation resulting from oxidative stress [31]. Chlorophyll content is a significant indicator of photosynthetic capacity under intense stress. In other words, higher chlorophyll content and stability appears to be associated with higher drought tolerance. Therefore, selecting plant genotypes based on increased or stable chlorophyll content may prevent yield or aesthetic or physiological performance loss under drought stress [32]. This research shows drought stress caused no significant changes in $chlorophyll_b$, total chlorophyll, and carotenoid content in *Andropogon gerardii*. Such conditions appear to show that the species' photosynthetic system was not seriously damaged after drought stress [54]. Therefore, *Andropogon gerardii* appears to be a stable genotype for physiological losses during drought stress conditions."

Our finding regarding photosynthesis ability of *Andropogon gerardii* confirms the results of the previous studies. One of the fundamental mechanisms that enable C4 grasses to tolerate drought is their ability to maintain high rates of photosynthesis even under water stress conditions. This drought tolerance is achieved through several adaptations, including more efficient water use, a high stomatal density that facilitates gas exchange, and a high concentration of carbon dioxide in the leaves. Additionally, C4 grasses have a unique carbon-fixing pathway that helps them conserve water and enhance photosynthetic efficiency. Another important mechanism contributing to drought tolerance in this genus is regulation of water loss through adjusting the opening and closing of their stomata in response to changes in environmental conditions, such as temperature, humidity, soil moisture, and light intensity. Such a mechanism, common to most C4 grasses, enables water conservation during dry periods and prevents excessive water loss, which is crucial for their survival in arid regions. Furthermore, C4 grasses can maintain and even expand their root systems even under drought stress, which helps them access water from deeper soil layers. This ability is achieved through several adaptations, such as a greater root depth and a higher root/shoot ratio, which enable them to reach and absorb water from deeper soil layers where moisture is still available. Overall, the drought tolerance mechanisms of C4 grasses like *Pennisetum* spp. are complex adaptations that enable them to survive and thrive in arid environments. Understanding these mechanisms is crucial for developing drought-tolerant crops in regions affected by water scarcity. One study published investigated the physiological and biochemical mechanisms underlying drought tolerance in *Pennisetum glaucum*, a C4 warm season grass commonly known as pearl millet [55]. This study found that pearl millet exhibited several adaptive traits, such as reduced leaf water potential and stomatal conductance, increased root-to-shoot ratio, and higher activity of antioxidant enzymes. These traits allowed pearl millet to maintain its photosynthetic activity and growth even under severe drought conditions [55]. Another study explored the genetic basis of drought tolerance in switchgrass, another C4 warm season grass commonly used for forage and biofuel production. The study identified several candidate genes that may regulate water use efficiency and root growth under drought stress. The findings suggest that genetic improvement of switchgrass for drought tolerance is feasible through targeted breeding or genetic engineering [56]).

Research has proved that not changing the carotenoid content in the plants during stress conditions might also be an adaptation response in plants because of antioxidant activities. Carotenoids are the primary lipid-soluble antioxidants of plant cells [57]. The oxidative injury induced by intense drought stress is characterized mainly by the reduction in antioxidant enzymes and increased lipid peroxidation [32]. Keles and Öncel [58] also reported that carotenoid values in wheat seedlings increased under water stress and non-optimal growth temperature conditions. Carotenoids can act as a non-enzymatic antioxidant. They have multiple roles, e.g., light-harvesting and protection from oxidative damage caused by drought, in developing drought tolerance. Thus, increased contents of carotenoids are essential for stress tolerance [59].

Table 6. Significant responses of the studied grass species to drought stress conditions (change of irrigation regime from 100% FC to 50% FC) as indicated by physiological, morphological, qualitative, and plant growth parameters.

Grass Species	Color	Texture	Root Length	Shoot Fresh Weight	Root Fresh Weight	Shoot Dry Weight	Root Dry Weight	RWC	RSD	RWL	EL	Chl_a	Chl_b	Total Chl.	Carot.	No. Sig. Responses
A. gerardii		■	■		■		■	■	■	■	■	■				9
S. nutans		■	■	■	■	■	■	■	■	■	■	■	■			12
P. virgatum	■	■	■	■		■		■	■	■	■	■	■	■		12
Sch. scoparium		■	■		■		■				■	■	■	■	■	9
B. curtipendula		■	■						■	■	■	■	■	■	■	9

RWC: Relative Water Content (%), RSD: Relative Saturation Deficit (%), RWL: Relative Water Loss (%), EL: Electrolyte Leakage (%), Chl_a: $Chlorophyll_a$ ($mgg^{-1}FW$), Chl_b: $Chlorophyll_b$ ($mgg^{-1}FW$), Total chl.: Total Chlorophyll ($mgg^{-1}FW$), Carot.: Carotenoid ($mgg^{-1}FW$), No. Sig. responses: Number of significant responses.; Note: the shaded cells show significantly different responses to the measured factor in the 100% FC and 50 % FC treatments.

All the grasses studied in this experiment were dominant plants of tall grass prairies of the North American continent. While there is research evidence on the grass germplasm of West Asia, this study used the grass germplasm of North America for drought resistance research for several reasons: firstly, North America has a wealth of C4 grass species that are proven to be adapted to a range of environmental conditions, including drought. Many grasses are also essential forage crops, making them economically important. In contrast, West Asia has a limited number of C4 grass species, and those present may not be as well adapted to drought conditions. Secondly, a wealth of knowledge and resources is available for studying C4 grasses in North America. Many of the leading research institutions and researchers in the field are based in North America, and there is a long history of research on C4 grasses in the region. This opportunity makes North American germplasm an attractive choice for researchers looking to build on this existing knowledge base. Finally, selecting germplasm for drought resistance research involves balancing genetic diversity and practical considerations such as availability and ease of use. North American germplasm was chosen because it represents a diverse range of C4 grass species that are commonly used in research, making it a practical and accessible choice for researchers looking to study drought resistance mechanisms in these plant groups [60]. The species chosen for this research have also been commonly and successfully used in urban xeric landscaping settings in North America and elsewhere.

In this study, *Sorghastrum nutans* had the greatest sensitivity to the reduction in its water content. Tall grasses experience diverse environmental conditions imposed by weather variability and by biotic modification of their microclimate. Intra-seasonal and inter-seasonal variability in precipitation is high in the tall grass prairies. Moreover, grasses experience relatively frequent periods of drought characterized by high temperatures and desiccating winds, in which leaf photosynthesis and biomass production are reduced [61]. Therefore, differences in water use efficiency among these species can be related to their morphological differences [61]. In the experiment conducted by Silletti and Knapp [62] on responses of *Andropogon gerardii* and *Sorghastrum nutans* to long-term manipulations of nitrogen and water, it was found that these two species did not respond equally to climate changes. *A. gerardii*, which is currently more abundant than *S. nutans*, was relatively unresponsive to resource manipulations. Therefore, maintenance of constant physiological activities in the face of resource variability and low resource availability may favor *A. gerardii* over its potential competitors such as *S. nutans*. The findings of Silletti and Knapp [62] confirm the results of our current study.

Another species that in this experiment showed more enhanced sensitivity to drought stress was *Panicum virgatum*. In this study, *Panicum virgatum* was the only species whose root dry weight and root length did not decrease with drought conditions. In general, an increase in soil water content elicits a proportional increase in root growth and above-ground plant growth [63]. On the other hand, plants usually develop an extensive root system under water stress to avoid the stress [12]. However, in this study, *P. virgatum*, despite having a sound root system, still had less tolerance to drought stress than *A. gerardii*, *S. scoparium*, and *B. curtipendula.* This finding may be possible because *P. virgatum* is a small-seed species that initially allocates large amounts of energy to develop a robust root system. It usually reaches only 33–66% of its maximum production capacity during the first and second years and reaches its total capacity during the third year after planting [64]. Moreover, the dominant C4 grasses, such as *A. gerardii*, *S. scoparium* and *B. curtipendula*, have been shown to take up most of their water from the top 30 cm of the soil. Therefore, the performance of these shallow-rooted grasses would be expected to be closely related to rainfall conditions of their origin [30]. On the other hand, in this study, the general absence of root mass increases in grasses under water stress may be attributed to the limited space (a pot with 2.5 kg of soil) for the plants to develop their root systems. Pot-grown plants resist water stress by adjusting osmotic potential and decreasing water potential to provide the necessary potential gradient for water absorption and movement. In contrast, field-grown plants develop extensive roots for water uptake [12]. In general, comparing the growth factors among these grasses under drought and non-drought stress conditions supports their local and geographical distribution. *P. virgatum* is a mesic grass and appears to be the most sensitive grass to water stress among our studied species. Therefore, from this aspect, our results are consistent with Knapp's results [54,65]. Heckathorn and DeLucia [51] also confirmed that *A. gerardii* is an intermediate species and *S. scoparium* is a xeric and drought-tolerant species.

One critical and practical note emerging from this study was that the number of days to germination was negatively correlated with the amount of carotenoid and chlorophyll content. This finding may show that the longer the germination progressed, the greater the grass's leaf greenness and freshness would be. On the other hand, when the number of days to germination increased, leaf width was higher, but the shoot dry weight and the number of the tillers were reduced. Therefore, where grass biomass and density per unit area are the important factors in landscaping, we should select the species which germinate quicker. However, because in the grasses that germinated quicker the color of the grass type was yellower, and since the visual quality as defined by the greenness of the grass is important in urban landscaping, it is recommended to select the grass species with a greener color and later germination rate and to solve the problem of lower grass density per unit area through denser planting and increasing the amount of the seed sown per unit area for these species.

Although relative drought tolerance is an important plant characteristic, since it strongly influences survival and growth potential, there are many other factors to consider when selecting suitable species for landscaping within arid and semi-arid cities, including their potential for wider negative impacts [66,67]. Although there can be some benefits to non-native species in terms of meeting urban landscaping and greening objectives, introducing plants from other continents or regions risks invasion and spread from gardens and into native ecosystems. It is therefore recommended that risk assessments for potential invasiveness be performed before new introductions of exotic plant species [68]. Further, management techniques to reduce likelihood of plant invasions should also be implemented [34].

This study was conducted in local weather conditions of Mashhad, and the soil used in the survey was a representative soil imported in many urban landscape projects across this city. However, it still has some limitations that might be considered in future studies. Designing more time-extended experiments in larger experimental plots in urban landscapes may provide more robust recommendations for executive landscape projects in urban environments.

5. Conclusions

This study has found that drought stress influenced most of the measured qualitative and morpho-physiological traits of the introduced warm-season grasses. Drought conditions, both short and long term, are commonplace in much of the world's drylands, which includes most of Iran where this study took place; however, some species were found to be more drought-tolerant than others and hence more suitable overall for planting in such regions, especially in areas where there is limited potential for irrigation or where there is a shift to more water-conscious design and landscaping, such as in many urban areas experiencing warming and drying climates. Drought-resistance rankings of the grasses are useful for this purpose but may vary with the assessment method, duration of the imposed drought, or presence of edaphic stress that limits rooting within a soil horizon or layer. However, our results showed that the species of *Andropogon gerardii*, *Schizachyrium scoparium*, and *Bouteloua curtipendula* had more tolerance to drought stress than *Sorghastrum nutans* and *Panicum virgatum* under similar conditions of this experiment. All the studied grasses in this research are found throughout the tall grass prairies and are similar in many aspects, i.e., are C4 tall grasses of approximately similar stature. Therefore, their differing responses to water resource changes and drought stress conditions suggest possible differences in their basic eco-physiological processes. Thus, attaining knowledge in these differing responses can help landscape managers better select and manage these species for drought-resistant and xeric landscapes in urban environments.

Andropogon gerardii maintained its photosynthetic rates and stomatal conductance. *Schizachyrium scoparium* maintained its relative water content, relative water loss, and relative saturation deficit under drought stress, indicating its high drought tolerance. Keeping photosynthetic rate and chlorophyll content robust during droughts is critical for keeping the plants and the landscapes photosynthetically active. Therefore, such plants can contribute to the high environmental performance of urban landscapes in terms of photosynthesis, gas exchange capacity, and air purification.

Moreover, maintaining a robust plant water balance, relative leaf water contents, and minimal water loss during droughts indicate high drought-resilient plant species even after drought conditions. *Bouteloua curtipendula* maintained a robust relative water content during drought treatment in this study, indicating its next-ranked drought-tolerance potential among the studied plants. These three species indicated a high potential for contributing to water-use-efficient landscapes by keeping their original biomass during drought stresses of up to 50% FC.

Sorghastrum nutans and *Panicum virgatum* failed to show enough resilience during the drought treatment for most of the measured morphophysiological, qualitative, and plant growth-related parameters. However, if a wider variety of planting is required for drought-tolerant landscaping, selecting *Panicum virgatum* is preferred to selecting *Sorghastrum nutans* because it showed robust fresh and dry root weights during drought treatments of this study. Overall, we demonstrated that an ecophysiological approach is beneficial for identifying drought-tolerant species for urban landscape applications.

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