

Comparative Gas Exchange of *Juniperus ashei* (Ashe Juniper, Cupressaceae) at Ambient and Elevated Levels of Light, CO₂ Concentration and Temperature with Potential Influences on Community Structure

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Abstract

Juniperus ashei (Ashe juniper, Cupressaceae) is a shrub or small tree found in woodlands and savannas of central and western Texas, northern Mexico, Missouri, Arkansas and Oklahoma. Environmental conditions where this species is found are stressful due to mostly shallow soils, high summer temperatures, low and inconsistent rainfall. *Juniperus ashei* has increased in density from the distant past not because of elevated CO₂ or increased temperature, but lower fire frequency and reduced competition with prairie grasses because of heavy grazing. This study demonstrates that *J. ashei* functions as a sun species and juveniles beginning growth in canopy shade most likely will not be recruited into the canopy. *Juniperus ashei* is a sun species because the A_{max} (maximum photosynthetic rate) is high and increased significantly (45%) with increased CO₂ concentrations to 17.20 $\mu\text{mol CO}_2\cdot\text{m}^{-2}\cdot\text{s}^{-1}$ at the highest CO₂ concentration tested (780 $\mu\text{L/L}$) but did not change with increased temperature. Dark respiration (R_d) doubled when temperature increased but did not change with CO₂ concentrations. Intercellular CO₂ increased as the CO₂ concentration increased but not with temperature. Light saturation (I_{sp}) and water use efficiency (WUE) increased significantly when light levels and CO₂ concentrations were elevated but not when temperatures were elevated. Stomatal conductance decreased when the CO₂ concentration increased, but not by elevated temperature. There was a significant interaction for WUE that increased with increased light level and CO₂ concentration, but decreased overall with temperature. This suggests *J. ashei* juveniles in canopy shade will be more stressed at high CO₂ and elevated temperatures in low light reducing recruit-

ment into the canopy.

Keywords

Light Response, Gas Exchange, Photosynthesis, Respiration, Water Use Efficiency, WEU, Potential Recruitment

1. Introduction

As worldwide climates have changed, the distribution and composition of plant communities have shifted, and are expected to continue adjusting in the future [1]-[3]. The atmospheric CO₂ level was approximately 1500 ppm at the end of the Cretaceous (55 × 10⁶ years ago) which did not decline for 1000 years [4] [5]. Approximately 125,000 years ago the atmospheric CO₂ level was about 230 ppm with a corresponding temperature 6°C below the current level [6]. Current atmospheric CO₂ levels are increasing at approximately 3 parts per million per year (2024 mean level = 427 ppm) with temperatures increasing at 0.20°C/10 years (2024 mean level = 16.24°C) [7].

Plant communities and animal populations changed in the past and existing communities and populations will change in density and distributions in the future as atmospheric carbon levels and temperatures increase, but how populations will change is uncertain [1] [4] [8].

The land surface of central North America was grasslands and covered 20% of the surface in the past several thousand years [9]. These grasslands changed due to cultivation and the introduction of large herds of domestic ungulate [10]-[12]. Many of these overgrazed grasslands have been encroached by various *Juniperus* species from the Atlantic to the Pacific coasts through the Great Plains to the low and mid-elevations of the mountains of North America [13]-[15] and have been treated as stable communities [16] [17]. Nevertheless, studies suggested the *Juniperus* are pioneer species and will develop into various woodlands [18]-[20].

Central Texas currently has many communities with various densities of *J. ashei* Buchh. and *Quercus virginiana* Mill. (Ashe juniper and hill country live oak, [15] [21] [22]). The Texas *Juniperus* communities are generally similar in overall structure and environmental characteristics to *Juniperus* woodlands from all over the world [23]. Although, associated species are very different and sometimes have very limited distributions [21] [24]. Canopy density is highly variable in Texas with *Juniperus* communities having cover between 40% and 90% [25] [26] and open areas that have juvenile woody species, grasses and herbaceous plants [12] [27]. There are few studies that have investigated understory woody seedlings and herbaceous plants responses to predicted future climate changes of higher CO₂ concentration and temperature [1] [4] [8]. Due to the shifting climate, some of the current low-density woody species may be lost or recruited into the canopy. Therefore, the future density of species and structure for these areas in central

Texas is unknown.

The shift in an ecosystem from a grassland to a woodland would entail major shifts in biomass from mostly belowground for grasslands to mostly aboveground for woodlands [28]. These ecosystem shifts could alter regional terrestrial and atmospheric biogeochemistry especially if the woodlands act as a carbon and nitrogen sink [29]-[31]. Processes could include nutrient cycling and availability which would influence primary production, competition for resources, species richness, community composition as well as species interactions [32]. But how these factors would change in an atmosphere high in CO₂ and temperature are not known. In addition, it is not known how seedlings of the adult *Juniperus* and other species would respond.

CO₂ concentration has been shown to be limiting in some communities and increasing levels promoted higher photosynthetic uptake and growth for some species [33]-[35]. However, work showing effects of elevated CO₂, temperature, and light levels on *J. ashei* or on any *Juniperus* woodland communities has not been carried out. There have been a few studies that examined the gas exchange responses of associated species but none dealing with *J. ashei* [2] [34] [36].

Currently, *J. ashei* dominates the woodlands across central Texas similarly to other worldwide *Juniperus* communities dominated by their specific local *Juniperus* species, but the associated species are quite different with some in Texas being endemic to the central Texas region [24] [36] [37]. We hypothesize that due to the changing environmental conditions, specifically increased CO₂ concentration and temperature, the future community composition and structure will also change. In this study, we investigated the photosynthetic response of mature *J. ashei* to short-term increases in temperature and CO₂ concentrations through various light levels and compared them to their current ambient responses. These results were then used to evaluate their potential effects on *J. ashei*'s role in the future community dynamics of central Texas.

2. Materials and Methods

2.1. Community Structure

All gas exchange measurements were made on mature *Juniperus ashei* leaves on the south side of trees on the western part of the University of Texas at San Antonio campus in central Texas (29.58030 N and -98.62403 W). Area topography is rolling with low slopes between 4.5° and 13.5° and soils that are clayey-skeletal, smectitic, thermic lithic calciustolls [38] in the Tarrant association with surface horizons between 0 and 25 cm thick [39]. The area geology is composed of heavily fractured limestone over limestone bedrock. Climate is subtropical sub-humid [40] having a mean annual temperature of 20°C with means of 9.6°C in January and 29.4°C in July [41]. Precipitation is also highly variable usually 78.7 cm/yr and bimodal, very little in June and July and peaks in May 10 with 7 cm and September with 8.7 cm [41]. There was no domestic grazing or domestic livestock present in the study area for the past 75 years. There are large areas of *Juniperus ashei* *Quercus*

virginiana woodlands or savannas on former grassland sites in central Texas which are considered representative of similar communities found in this area [42].

Mature *J. ashei* plants in relatively undisturbed *J. ashei*/*Q. virginiana* woodland communities were selected randomly for measurements. Mean density and basal area of the communities examined was determined but limited information about community structure is presented here [2] [33]. Trees found were all identified, counted and measured. Concise but succinct information about community structure is presented below for the major species found.

2.2. Gas Exchange

Gas exchange responses at both ambient and elevated levels of CO₂ and temperature were made. Three plants were randomly selected in the summer of 2007. On each replicate plant one main stem tip at breast height, approximately 137 cm above the soil surface was selected and only mature, non-damaged stems were used. Steady state photosynthetic light response curves (A_{net} vs. PPFD) were measured on full grown stem tips with completely expanded leaves at mid-day (1000 - 1400 hrs) when relative humidity had stabilized [43]. Leaves one cm from the growing stem tip were 1.07 ± 0.27 mm (mean \pm 1.0 SD) in length and those five cm from the growing tip were 2.17 ± 0.34 mm in length and leaves surrounded the entire stem tip.

To measure leaf gas exchange, a branch tip or cluster with several secondary branches all with many small leaves was placed into the cuvette chamber. The stems were all parallel in the chamber in a single plane configuration and all at 90° to the light source. At least 66% of the cuvette chamber was covered. After the gas exchange measurements were made Leaf area within the chamber was measured with a LI-COR LI 3000A portable area meter. Each sample was measured in the scanner three times and the mean was determined and used as a correction and manually entered into the Li-6400® to adjust each curve. Measurements made and recorded were: A_{net} (net photosynthesis = $\mu\text{molCO}_2 \cdot \text{m}^{-2} \cdot \text{s}^{-1}$), C_i (intercellular [CO₂] = $\mu\text{molCO}_2 \cdot \text{molair}^{-1}$), T_{leaf} (chamber leaf temperature = °C), T_{air} (air temperature outside the chamber = °C), PPFD (photosynthetic active radiation = $\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$), g (stomatal conductance = $\text{molH}_2\text{O} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$) and E (transpiration = $\text{mmolH}_2\text{O} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$).

2.3. Chamber Set-Up

The gas exchange chamber was used to mimic varying degrees of environmental modifications with the stem cluster in the chamber attached to the plant. Conditions adjusted or manipulated were the light level, CO₂ concentration, and temperature. Relative humidity was kept at 30% - 40% and the gas flow rate was set at $400 \mu\text{mol} \cdot \text{s}^{-1}$. Coefficient of variation stabilized (<1%) before recording and moving to the next measurement. Light levels started at $1800 \mu\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$ and decreased to 1600, 1400, 1200, 1000, 800, 600, 400, 200, 100, 75, 50, 25, 10, 5 and finally $0 \mu\text{mol} \cdot \text{m}^{-2} \cdot \text{s}^{-1}$. Light curves and CO₂ response curves were measured for

different combinations of the leaf chamber CO₂ and temperature environments.

The leaf chamber was first set at the mean atmospheric CO₂ level (390 µL/L) for 2007 and a temperature of 35°C. This temperature was chosen based on the mean high temperatures for San Antonio during the summer months of June, July and August. Light curves were repeated holding the ambient CO₂ constant while raising the chamber temperature to 40°C and then to 45°C. Next, the leaf chamber CO₂ was raised to 1.5 times the 2007 CO₂ levels to 585 µL/L. Light curves were completed at a temperature of 35°C, 40°C and 45°C. This process was then repeated with the leaf chamber CO₂ level set at twice the 2007 ambient level at 780 µL/L. Lastly, CO₂ response curves were measured at a canopy shade light level (700 µmol·m⁻²·s⁻¹). Measurements were made at 35°C, 40°C and 45°C.

2.4. Analysis

The data analysis was completed using Microsoft Excel® and JMP® IN 5.1. Significant differences were measured using the JMP® IN 5.1 software with a repeated measures MANOVA on the photosynthetic rate curves including intercellular CO₂ concentrations, stomatal conductance and transpiration. The light level, PPF, was the repeat variable [44]. Water use efficiency (WUE) was calculated by dividing the photosynthetic rate by the transpiration rate and also analyzed using a repeated measures MANOVA. Significance levels used for all tests were $P \leq 0.05$. Normality was checked with the Shapiro-Wilk W test and homogeneity of variance with Bartlett's test and log transformed as necessary. A standard least squared ANOVA was used to detect significant differences in each curve at each CO₂ concentration and temperature combination. However, this is a curve-to-curve comparison and individual CO₂ uptake was not compared at individual light levels on each plant and each replication.

For other measurements, they were derived from Excel® plots of the LICOR® Li-6400 measurements. Included were maximum photosynthetic rate (A_{max}) which was the highest A_{net} measured for each replicate or a mean of the highest A_{net} values that were not significantly different. The dark respiration rate (R_d) was the gas exchange rate at PPF = 0 µmol·m⁻²·s⁻¹. The quantum yield (Φ) was the linear initial slope relationship calculated using the dark values and A_{net} at increasing PPF until the regression coefficient of the slope decreased. The light compensation point (L_{cp}) was calculated as the PPF when $A_{net} = 0$ µmolCO₂·m⁻²·s⁻¹ using the linear regression of the initial response. The light saturation point (L_{sp}) was the light level when the initial slope reached A_{max} . A standard least squared ANOVA was used to determine significant differences for the CO₂ concentration and temperature effects. Tukey-Kramer HSD multiple comparison tests were used to determine differences between pair wise comparisons [44].

3. Results

3.1. Community Structure

The community overstory had a mean canopy density of 1840 plants/ha and was

found to be dominated by *Juniperus ashei* with a relative density in the canopy of $61\% \pm 12\%$ (mean \pm one standard deviation). The other major canopy species was *Quercus virginiana* with a relative canopy density of $36\% \pm 6\%$. Additional community species with relative densities of 0.06% - 1.80% were *Celtis laevigata* (sugarberry or hackberry), *Diospyros texana* (Texas persimmon), *Prosopis glandulosa* (mesquite), *Calia secundiflora* (Texas mountain laurel), *Ulmus crassifolia* (cedar elm), and *Ungnadia speciosa* (Mexican buckeye).

3.2. Photosynthetic Curves

The mean curves of the photosynthetic rates for *Juniperus ashei* are shown by temperature and CO₂ concentration with light level being fixed (Figure 1(A) and Figure 1(B)). The photosynthetic rates by temperature show a statistical difference (MANOVA, $P = 0.0452$). When the temperature was increased there was a non-significant decrease in photosynthetic rate by approximately 9% between the 35°C and 40°C curves and an additional non-significant decrease of 13% between the 40°C to 45°C ($P = 0.4104$ and $P = 0.2876$ respectively). However, there was a significant decrease in photosynthesis between the 35°C and the 45°C curves ($P = 0.0446$, Figure 1(A)).

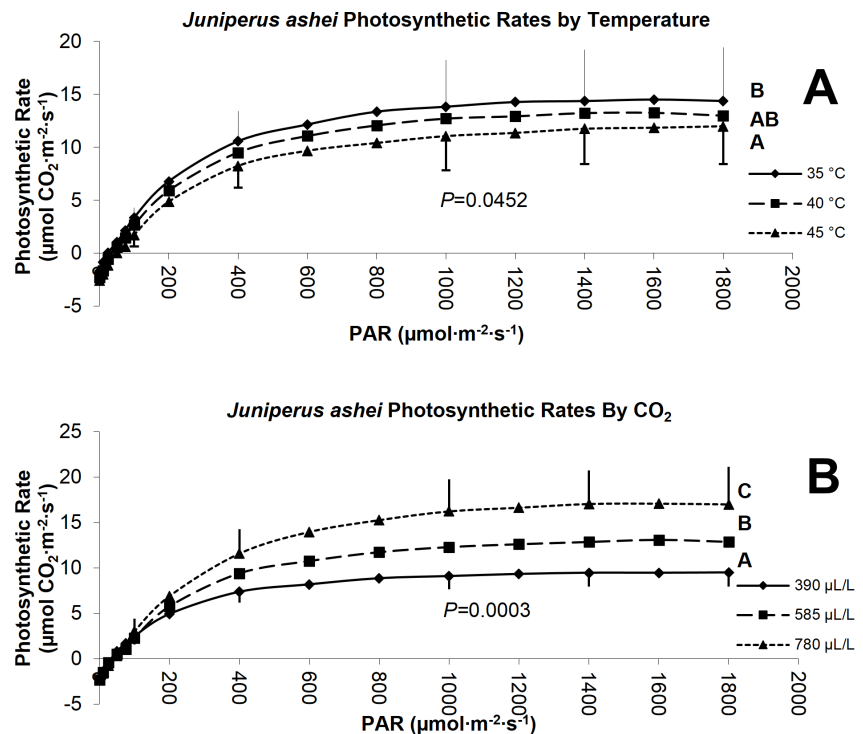


Figure 1. Mean repeated measures MANOVA curves of the photosynthetic rates for *Juniperus ashei* displayed by temperature (A) and CO₂ (B) treatment is for main effects only. P -values are shown from the repeated measures MANOVA for the main effects (Temperature and CO₂). Like letters at the end of the curves indicate no significant difference between the curves. Data is from three replicates at three concentrations of CO₂ (390, 585 and 780 $\mu\text{L/L}$) and three temperatures (35°C, 40°C and 45°C). Representative error bars are shown indicating \pm one standard deviation.

The comparison of the mean photosynthesis rates by CO₂ levels shows a significant difference between the three curves at different CO₂ levels (MANOVA, $P = 0.0003$, **Figure 1(B)**). The total increase was 51% for the curves as the CO₂ concentration increased. The CO₂ curves showed a similar significant difference between the ambient (390 μL/L) or low CO₂ concentration and the middle (585 μL/L) concentration ($\approx 25\%$ increase, $P = 0.0306$) and the middle and high (780 μL/L) CO₂ concentration ($\approx 26\%$ increase, $P = 0.0318$).

Repeated measures MANOVAs were used to investigate the main effects of light levels, CO₂ concentration and temperature on *Juniperus ashei*. Because the interactions were not significant except for water use efficiency they were removed from the MANOVA (**Table 1**). For *J. ashei* as light levels were increased all factors measured changed as the temperature and the CO₂ concentrations were increased. However, there were no significant temperature effects on; conduction, intercellular [CO₂] concentrations, or transpiration (**Table 1**). Temperature only had a significant effect on the photosynthetic rates and water use efficiency (WUE). For CO₂ concentration there were significant effects on photosynthetic rates, stomatal conduction, intercellular [CO₂] concentration, and WUE. Transpiration rate was not significantly affected by increased temperature or CO₂ (**Table 1**).

Table 1. Table of P -values for repeated measures MANOVAs of gas exchange curves for *Juniperus ashei* comparing main effects of temperature and CO₂ at 16 light levels that were held constant (interactions were not significantly different and removed from the models). Data is from three replicates at three CO₂ concentrations (390, 585 and 780 μL/L) and three temperatures (35°C, 40°C and 45°C). Significant entries are bold.

Repeated measures MANOVA P -values for <i>Juniperus ashei</i> -without interaction					
Main Effects	Photosynthetic Rate	Stomatal conductance	Intercellular CO ₂ concentration	Transpiration	Water Use Efficiency
Temperature	0.0452	0.1732	0.5070	0.2026	<0.0001
CO ₂	0.0003	0.0044	<0.0001	0.1047	<0.0001

The maximum photosynthetic rate (A_{\max}) did not change with temperature (ANOVA, $P = 0.1468$, **Table 2**). Temperature had little effect on the mean A_{\max} although it did drop by 18% with a standard error of 0.92 μmolCO₂·m⁻²·s⁻¹. The A_{\max} did change significantly with CO₂ concentration (ANOVA, $P < 0.0001$, **Table 2**). In addition, Tukey comparisons of the CO₂ effect showed significant differences between each concentration. Elevating CO₂ increased A_{\max} from 9.42 μmolCO₂·m⁻²·s⁻¹. for the ambient CO₂ concentration to 12.97 μmolCO₂·m⁻²·s⁻¹. for the middle CO₂ concentration and to 17.20 μmolCO₂·m⁻²·s⁻¹. for the high CO₂ concentration. This was a total A_{\max} increase of approximately 45% from the low to high CO₂ concentration. The interaction term was not significant ($P = 0.5961$, not shown).

The light saturation point (L_{sp}) was not significantly different by temperature (ANOVA, $P = 0.8776$, **Table 2**). The mean L_{sp} for the three temperatures were

different by 5% with a standard deviation of $23.8 \mu\text{molCO}_2 \cdot \text{m}^{-2} \cdot \text{s}^{-1}$. Elevating CO_2 had a significant effect on L_{sp} (ANOVA, $P = 0.0001$, **Table 2**). A Tukey comparison showed a significant difference between values for the ambient CO_2 L_{sp} and both the medium CO_2 and high CO_2 L_{sp} . There was no significant difference for L_{sp} between the middle and the high CO_2 concentrations. The interaction term was not significant ($P = 0.8929$, not shown).

Table 2. Factors measured and P -values for Standard Least Squared ANOVAs for *Juniperus ashei*. Data is from three replicates at three CO_2 concentrations (390, 585 and 780 $\mu\text{L/L}$) and three temperatures (35°C, 40°C and 45°C). Interactions were not significantly different and not shown. Bold entries are significant at 0.05 or less. Capital letters next to a value in a column are significantly different.

Factors And Significance Levels					
Treatment	A_{max}	L_{sp}	L_{cp}	R_{d}	\emptyset
CO_2 390	9.42A	258A	21.8A	1.82A	0.047A
CO_2 585	12.97B	402B	35.6A	1.72A	0.037A
CO_2 780	17.20C	423B	33.1A	1.98A	0.047A
Significance	$P = 0.0001$	$P = 0.0001$	$P = 0.3729$	$P = 0.4446$	$P = 0.2075$
35°C	14.46a	353a	23.5a	1.33a	0.046a
40°C	13.32a	359a	26.0a	0.88ab	0.044a
45°C	11.82a	370a	41.0b	2.32b	0.041a
Significance	$P = 0.1468$	$P = 0.8776$	$P = 0.0003$	$P = 0.0007$	$P = 0.6881$

The light compensation point (L_{cp}) showed a significant difference by temperature (ANOVA, $P = 0.0003$, **Table 2**) but not by CO_2 concentration (ANOVA, $P = 0.3729$, **Table 2**). A Tukey comparison showed the 35°C L_{cp} was not significantly different than the 40°C L_{cp} . The 45°C L_{cp} was significantly different from both the 35°C L_{cp} and the 40°C L_{cp} . There were no significant differences in the L_{cp} for CO_2 concentration (ANOVA, $P = 0.3729$). The interaction term was not significant ($P = 0.5166$, not shown).

The ANOVA for the dark respiration rates (R_{d}) showed a significant difference by temperature but not by CO_2 concentration ($P = 0.0007$ and $P = 0.4446$, **Table 2**). Tukey comparison showed the 35°C R_{d} was significantly different from the 45°C R_{d} while the 40°C R_{d} was not significantly different from either the 35°C or the 45°C R_{d} . The CO_2 concentration showed no trend with R_{d} values with increasing CO_2 concentration (ANOVA, $P = 0.4446$). The interaction term was not significant ($P = 0.4226$, not shown).

The quantum yield (\emptyset) showed no statistical trend based on temperature (ANOVA, $P = 0.6881$, **Table 2**). As with temperature, the CO_2 ANOVA showed no significant effect on the \emptyset values ($P = 0.2075$, **Table 2**). The interaction term was not significant ($P = 0.9163$, not shown).

The mean curves of the water use efficiency (WUE) for *J. ashei* are shown by temperature, CO₂ and light effects (Figure 2(A) and Figure 2(B)). Water use efficiency was significantly different when compared by temperature (MANOVA, $P < 0.0001$, Figure 2(A)). Water use efficiency values decreased from a plateau of approximately 3.5 mmol·mol⁻¹ to approximately 2.5 mmol·mol⁻¹ as temperature increased to 45 °C or by a total of 21%. The comparisons by CO₂ concentration were also statistically significant between the curves (repeated measures MANOVA, $P < 0.0001$, Figure 2(B)). The curves generally increased as the light levels increased and as the CO₂ concentration increased. At the ambient (390 μL/L) or low CO₂ concentration the WUE value was lowest at approximately 2.1 mmol·mol⁻¹ (Figure 2(B)). At the highest CO₂ concentration, the WUE value increased approximately 50% to a value of approximately 4.2 mmol·mol⁻¹.

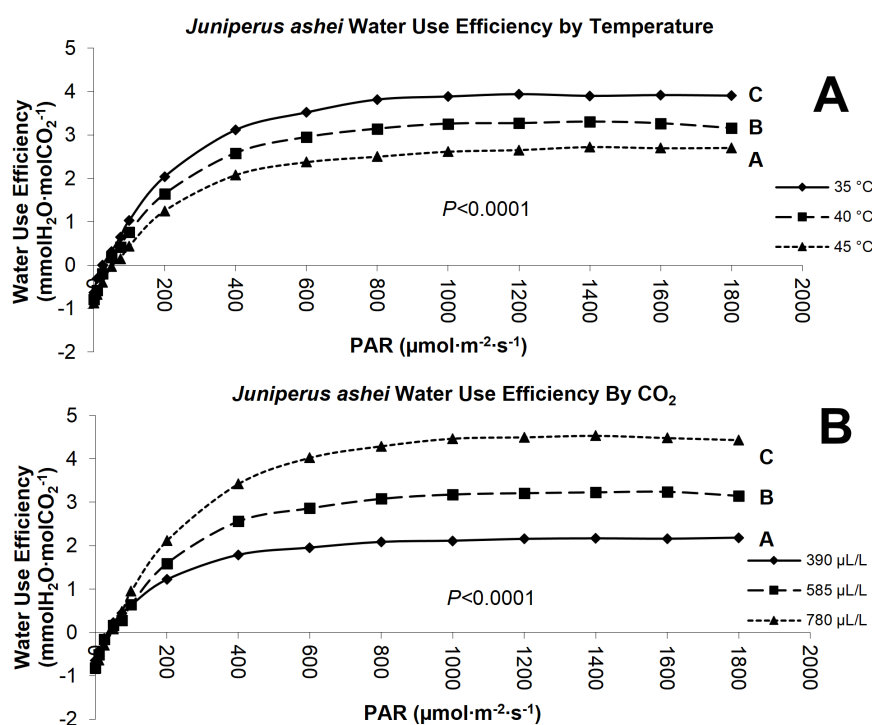


Figure 2. Presented are repeated measures MANOVA curves of main effects on water use efficiency for *Juniperus ashei* displayed by temperature (A) and CO₂ concentration (B) with light levels held constant. P -values are shown from the repeated measures MANOVAs. No like letters at the end of the curves indicate significant difference between curves. There were three concentrations of CO₂ (390, 585 and 780 μL/L) and three temperatures (35 °C, 40 °C and 45 °C).

Because there was a significant interaction between light level, temperature, and CO₂ concentration for the WUE response an additional figure is presented to demonstrate the effect (Figure 3). The WUE increased with light level (PFD, right axis front to back) and with CO₂ concentration (light, dotted to gray shade within each temperature level), but decreased overall with each temperature (X-axis-left to right).

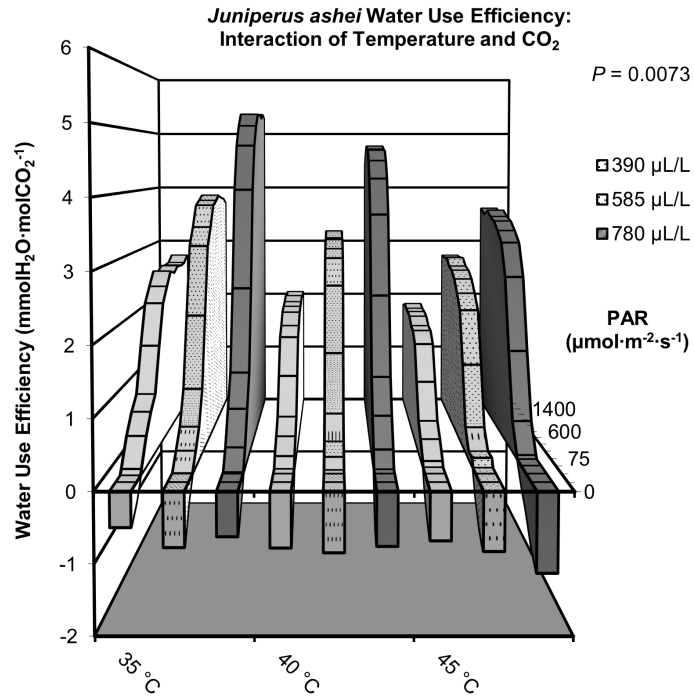


Figure 3. Mean repeated measures MANOVA curves of the water use efficiency for *J. ashei* interaction of temperature and CO₂ concentration. The CO₂ concentrations are shown below the plot and represent the three plots within each temperature array. Data is from three replicates at three concentrations of CO₂ (390, 585 and 780 μL/L) and three temperatures (35°C, 40°C and 45°C) and 16 light levels.

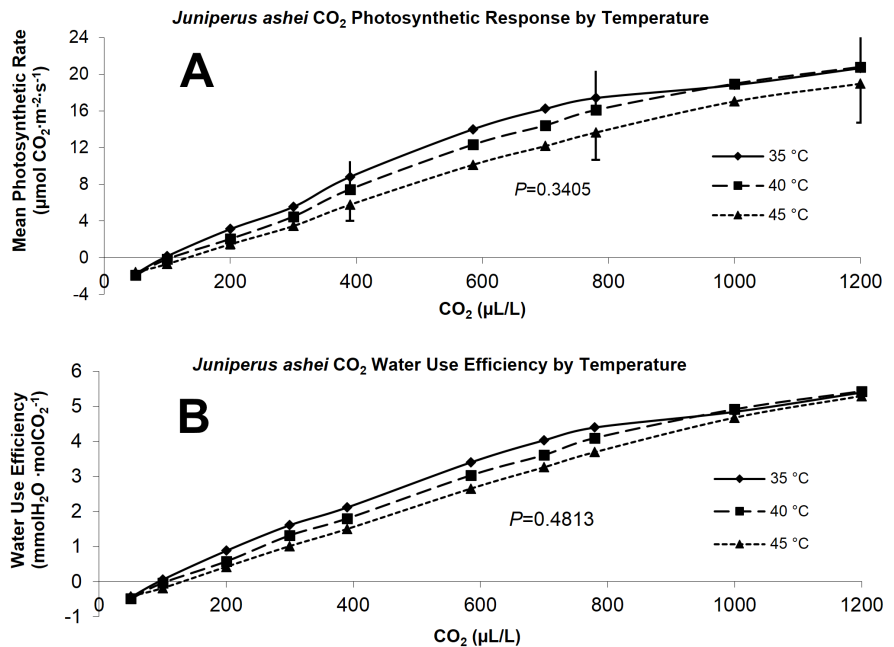


Figure 4. Photosynthetic response curves for increasing CO₂ levels (A) and three temperature (35°C, 40°C and 45°C) and (B) water use efficiency curves for *J. ashei* at increasing CO₂ levels, a light level of 700 μmol·m⁻²·s⁻¹ and three temperatures. Each curve was plotted from a mean of three replicates. *P*-values are shown from the repeated measures MANOVAs. Error bars are shown indicating ± one standard deviation.

Figures that represent the photosynthetic rate and water use efficiency CO₂ response to temperature are shown below (**Figure 4(A)** and **Figure 4(B)**). Both the CO₂ photosynthetic response and water use efficiency increase with increasing CO₂ concentration. Neither curve seems to reach a plateau. The curves follow the same basic trend, with no significant difference to increasing temperature. None of the repeated measures MANOVAs performed on the photosynthetic response, intercellular [CO₂], stomatal conductance, and transpiration were significantly different by temperature (Data not shown; $P = 0.3405$, $P = 0.4603$, $P = 0.1345$, $P = 0.9629$, respectively).

4. Discussion

There are approximately 60 *Juniperus* species that are present in various places across the Northern Hemisphere occurring from near the equator in Africa and Central America north to the arctic Circle [23]. There are 13 species of *Juniperus* in North America stretching from Canada in the north through the United States and south into Mexico and Central America [45] [46] *Juniperus ashei* is a major species in many central Texas woodland communities [37] [47]. In this study, we examined effects of ambient and elevated light levels, atmospheric CO₂ and associated higher temperatures on *J. ashei* gas exchange responses. Comparisons of our results with other species [2] [33] [36] [48] strengthen the position that *J. ashei* is not a shade-adapted species but more of a sun species [49]-[51].

Sun species tend to have a high maximum photosynthetic rate (A_{\max}), light saturation point (L_{sp}), light compensation point (L_{cp}) and dark respiration rates (R_{d}). *Juniperus ashei* was shown in this study to have a relatively high A_{\max} value under ambient and at elevated CO₂ levels (9.42 - 17.20 $\mu\text{molCO}_2\cdot\text{m}^{-2}\cdot\text{s}^{-1}$), as well as also high L_{sp} , L_{cp} , and R_{d} , which indicates it is a low sun or shade intolerant species and will remain so in an elevated CO₂ atmosphere. *Juniperus ashei* A_{\max} values did not change significantly with temperature (11.82 - 14.46 $\mu\text{molCO}_2\cdot\text{m}^{-2}\cdot\text{s}^{-1}$) but did increase significantly when CO₂ levels were increased (9.42 - 17.20 $\mu\text{molCO}_2\cdot\text{m}^{-2}\cdot\text{s}^{-1}$). The light saturation point (L_{sp}) did not change significantly with temperature but did increase significantly with elevated levels of CO₂ which tracks the values of the A_{\max} . The light compensation was not significantly affected by elevated levels of CO₂ but almost doubled at the highest temperature *J. ashei* leaves were exposed to. This was due to increased metabolism at the higher exposure temperature [1] [49].

While the MANOVA analysis showed an overall slight significant decrease in photosynthetic response as the temperature increased for *J. ashei*, the A_{\max} Least Standard Squares ANOVA comparison did not. A strong reaction would most likely be considered a hindrance to their competitive ability in a higher temperature environment, so this mixed result suggests a minimal impact. *Juniperus ashei* may further have an ameliorating effect to any temperature influenced competitive disadvantage since it showed a significant increase in photosynthetic rate to elevated CO₂. In the semi-arid environment found in central Texas, water use efficiency may play a significant role in species survival and competitive advantage,

and changes in plant response due to elevated temperature and CO₂ directly affect the species water use efficiency.

The modification to the overall WUE is of note and it closely mimicked the light curves. This is due to the photosynthetic responses decreasing in the low light environment while the transpiration rate only decreased slightly over the lower light levels. As light and CO₂ levels increase the water use efficiency increases but there was a significant decrease with increasing temperature. This significant interaction shows that at lower light levels, this species can not properly regulate water loss which is further exacerbated by a rise in temperature. *Juniperus ashei* juveniles have high mortalities at low light levels and a decrease in water use efficiency might play a role in future survival and recruitment below a canopy [52]. Species that are shade-tolerant usually regulate water use efficiency at reduced light levels allowing them to persist below the canopy. Some of the other species in these woodland understory communities are shrubs with sun species growth characteristics found at canopy edges or canopy breaks [37]. In these communities, there was an absence of many shade species found in the understory which is not fully explained by physiological characteristics of *J. ashei* the target species in this study.

Over the past century, there has been a large increase in the overall number and density of the large native herbivores in central Texas [53] [54]. Increases in density and number of large herbivores have been found to cause alterations to the local plant community compositions and dynamics in habitats all over the world [19] [55]-[59]. Almost all woody species in the study area are susceptible to juvenile herbivory and populations have a minimal cohort for recruiting except *J. ashei* [37]. The herbivory of *J. ashei* was not examined directly in the current study, however it is not eaten by *Odocoileus virginianus*, white-tailed deer [20]. This herbivore has been shown to cause establishment difficulties for many plant species in central Texas woodlands [54] [55] [57] [60].

Juniperus woodlands appear to be successional communities [1] [17] [37]. In the eastern North American deciduous forests, *Juniperus* plants are often found in gaps, blow downs or on shallow soil in glades [61]. In western North America, *Juniperus* tends to occur above the desert communities and above the arid or semiarid grasslands, but usually below the higher-elevation pine, spruce, or fir forests [12] [16] [17]. In central Texas, *J. ashei* establishes on hillsides and in former grasslands on shallow soil [22] [27].

Juniperus woodlands in many parts of the world are probably caused by a number of factors, with constant high levels of grass herbivory and a reduction of grassland fire frequency being dominant [10] [22] [37]. *Juniperus* and other early successional woody species, are favored with the reduction of grassland fire frequency producing various savannas and woodlands [59]. Overgrazing by domestic animals reduces the biomass and growth of the grasses while allowing the woody species to take advantage of the reduced competition for resources [1] [59] [61]. This difficulty in maintenance of the C₄ southern grasses is certainly domes-

tic animal herbivory but for various woody species it seems possibly to be competition for water by more shade tolerant species [19] [58] [62]-[65]. The study species is found in arid and semi-arid zones where droughts are common. In related species like *J. excelsa*, a decrease in photosynthesis during drought has been observed [66]. Also, in drought studies, *Juniperus* A_{max} was greatest near zero water potential and lowest when stressed at low water potential [65] [66]. Furthermore, *J. ashei* seedlings are strongly influenced by light levels, with higher light levels leading to interactions between water and nutrients that support continued growth [28]. In future studies the importance of drought on *J. ashei* seedlings should be examined along with interactions with other important environmental factors.

5. Conclusion

Over the past century, plant communities have been changed through the increased browsing pressure from large herbivores as well as the suppression of grassland fires. In the more recent decades and going into the future this has been and will be further complicated by the increasing air temperatures and CO₂ concentrations. Some species will be able to take advantage of the new conditions and expand their numbers, while other species lose competitive advantage and decline in number potentially resulting in different and new dominant species and community structure. We expect these central Texas *Juniperus/Quercus* woodlands will be part of these shifts and dynamic changes. Based off our study results, we believe *J. ashei* will gain some competitive advantages in the higher light levels of the open woodland canopy gaps with higher future CO₂ concentrations which should allow it to encroach and establish better in those areas. But below the canopy in the lower light levels, it does not seem to maintain the same advantages which means it may be replaced or at least lose some of its dominance within the existing woodland areas. Studying community dynamics and predicting future community composition has always been a challenging mission but with the added complications of forecasted environmental changes this has become an even more enigmatic puzzle to tease apart.

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Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

References

- [1] Begon, M. and Townsend, C.R. (2021) Ecology: From Individuals to Ecosystems. Wiley and Company. <https://www.amazon.com>

- [2] Grunstra, M.B. and Van Auken, O.W. (2024) Gas Exchange Rates of Texas Persimmon (*Diospyros texana*) in Central Texas Woodlands. *American Journal of Plant Sciences*, **15**, 329-348. <https://doi.org/10.4236/ajps.2024.155024>
- [3] World Meteorological Organization (WMO) (2024) State of the Global Climate 2023. WMO-No 1347.
- [4] Ehleringer, J.R., Cerling, T.E. and Dearing, M.D. (2005) A History of Atmospheric CO₂ and Its Effects on Plants, Animal, and Ecosystems. Springer Science + Business Media. <https://doi.org/10.1007/b138533>
- [5] Kunzig, R. (2011) World without Ice. *National Geographic* 220, 90-109. <https://www.nationalgeographic.com>
- [6] Petit, J.R., Jouzel, J., Raynaud, D., Barkov, N.I., Barnola, J., Basile, I., *et al.* (1999) Climate and Atmospheric History of the Past 420,000 Years from the Vostok Ice Core, Antarctica. *Nature*, **399**, 429-436. <https://doi.org/10.1038/20859>
- [7] NOAA Climate (2024) National Oceanic and Atmospheric Administration.
- [8] Keddy, P.A. (2017) *Plant Ecology: Origins, Processes, Consequences*. Cambridge University Press. <https://doi.org/10.1017/9781316321270>
- [9] Sims, P.L. (1988) Grasslands. In: Barbour, M.G. and Billings, W.D., Eds., *North American Terrestrial Vegetation*, Cambridge University Press. <https://assets.cambridge.org/97805215/50277/sample/9780521550277wsn01.pdf>
- [10] Collins, S.L. and Wallace, L.L. (1990) Fire in North American Tallgrass Prairies. University of Oklahoma Press. <https://www.oupres.com>
- [11] Archer, S., Schimel, D.S. and Holland, E.A. (1995) Mechanisms of Shrubland Expansion: Land Use, Climate or CO₂? *Climatic Change*, **29**, 91-99. <https://doi.org/10.1007/bf01091640>
- [12] Van Auken, O.W. (2000) Shrub Invasions of North American Semiarid Grasslands. *Annual Review of Ecology and Systematics*, **31**, 197-215. <https://doi.org/10.1146/annurev.ecolsys.31.1.197>
- [13] Dick-Peddie, W.A. (1993) *New Mexico, Vegetation: Past, Present and Future*. University of New Mexico Press. <https://www.amazon.com>
- [14] Breshears, D.D., Nyhan, J.W., Heil, C.E. and Wilcox, B.P. (1998) Effects of Woody Plants on Microclimate in a Semiarid Woodland: Soil Temperature and Evaporation in Canopy and Intercanopy Patches. *International Journal of Plant Sciences*, **159**, 1010-1017. <https://doi.org/10.1086/314083>
- [15] Van Auken, O.W. (2018) *Ecology of Plant Communities of South-Central Texas*. Scientific Research Publishing. <https://www.amazon.com>
- [16] Barbour, M.G. and Billings, W.D. (1988) *North American Terrestrial Vegetation*. Cambridge University Press. <https://www.amazon.com>
- [17] Barbour, M.G., *et al.* (1999) *Terrestrial Plant Ecology*. Addison Wesley Longman. <https://www.amazon.com>
- [18] McKinley, D.C. and Van Auken, O.W. (2005) Influence of Interacting Factors on the Growth and Mortality of Juniperus Seedlings. *The American Midland Naturalist*, **154**, 320-330. [https://doi.org/10.1674/0003-0031\(2005\)154\[0320:ioifot\]2.0.co;2](https://doi.org/10.1674/0003-0031(2005)154[0320:ioifot]2.0.co;2)
- [19] Kain, M., Battaglia, L., Royo, A. and Carson, W.P. (2011) Over-Browsing in Pennsylvania Creates a Depauperate Forest Dominated by an Understory Tree: Results from a 60-Year-Old Deer Exclosure. *The Journal of the Torrey Botanical Society*, **138**, 322-326. <https://doi.org/10.3159/torrey-d-11-00018.1>
- [20] Adkins, J.S. and Van Auken, O.W. (2023) Survival and Mortality of Protected and Non-Protected Juniperus Ashei Seedlings in Central Texas Juniper/Live Oak Wood-

- land. *Phytologia*, **1053**, 84-90.
- [21] Amos, B.B. and Gehlbach, F.R. (1988) Edwards Plateau Vegetation: Plant Ecological Studies in Central Texas. Baylor University Press. <https://www.baylorpress.com/9780918954503/edwards-plateau-vegetation/>
- [22] Elliott, L.F. *et al.* (2014) Ecological Mapping Systems of Texas: Summary Report. Texas Parks and Wildlife Department. <https://tpwd.texas.gov/gis/programs/landscape-ecology/supporting-documents/final-summary-report/>
- [23] Hora, B. (1981) The Oxford Encyclopedia of Trees of the World. Oxford University Press. <https://www.amazon.com>
- [24] Poole, J.M., *et al.* (2007) Rare Plants of Texas. Texas A&M Nature Guides. Texas Parks and Wildlife. Ever Best Printing. <https://www.tamupress.com>
- [25] Smeins, F.E. and Merrill, L.B. (1988) Long-Term Change in a Semiarid Grassland. In: Amos, B.B. and Gehlbach, F.R., Eds., *Edwards Plateau Vegetation*, Baylor University Press.
- [26] Van Auken, O.W., *et al.* (2022) Isolated Deciduous Woodlands in Central Texas. *Phytologia*, **104**, 13-23. https://www.phytologia.org/uploads/2/3/4/2/23422706/104_3_13-23vanaukendeciduous_woodlands8-15-22.pdf
- [27] Terletsky, P.A. and Van Auken, O.W. (1996) Comparison of Cedar Glades and Associated Woodlands of the Southern 8 Edwards Plateau. *Texas Journal of Science*, **48**, 55-67. <https://www.thefreelibrary.com/Comparison+of+cedar+glades+and+associated+woodlands+of+the+southern...-a0128678772>
- [28] Auken, O.W.V. (2000) Characteristics of Intercanopy Bare Patches in *Juniperus* Woodlands of the Southern Edwards Plateau, Texas. *The Southwestern Naturalist*, **45**, 95-110. <https://doi.org/10.2307/3672449>
- [29] McKinley, D.C., *et al.* (2008) Altered Ecosystem Processes as a Consequence of *Juniperus virginiana* L. Encroachment into North American Tallgrass Prairie. In: Van Auken, O.W., Ed., *Western North American Juniperus Communities. A Dynamic Vegetation Type*, Springer.
- [30] Mosier, A.R. (2001) Exchange of Gaseous Nitrogen Compounds between Agricultural Systems and the Atmosphere. *Plant and Soil*, **228**, 17-27. <https://doi.org/10.1023/a:1004821205442>
- [31] Jandl, R., Lindner, M., Vesterdal, L., Bauwens, B., Baritz, R., Hagedorn, F., *et al.* (2007) How Strongly Can Forest Management Influence Soil Carbon Sequestration? *Geoderma*, **137**, 253-268. <https://doi.org/10.1016/j.geoderma.2006.09.003>
- [32] Melillo, J.M., Butler, S., Johnson, J., Mohan, J., Steudler, P., Lux, H., *et al.* (2011) Soil Warming, Carbon-Nitrogen Interactions, and Forest Carbon Budgets. *Proceedings of the National Academy of Sciences of the United States of America*, **108**, 9508-9512. <https://doi.org/10.1073/pnas.1018189108>
- [33] Vitousek, P. (1982) Nutrient Cycling and Nutrient Use Efficiency. *The American Naturalist*, **119**, 553-572. <https://doi.org/10.1086/283931>
- [34] Grunstra, M.B. (2008) Investigation of Juniperus Woodland Replacement Dynamics. Ph.D. Thesis, The University of Texas. <https://www.scirp.org>
- [35] Eddy, K. (2019) How Does Increased Atmospheric Carbon Dioxide Concentration Affect Plant Competition? Ph.D. Thesis, The University of Texas.
- [36] Wang, S. *et al.* (2020) Recent Global Decline of CO₂ Fertilization Effects on Vegeta-

- tion Photosynthesis. *Science*, **370**, 1295-1300.
- [37] Wayne, E.R. and Van Auken, O.W. (2012) Comparisons of Gas Exchange of Several Sedges and C4 Grasses in Associated Savanna Communities. *Phytologia*, **94**, 71-90. https://www.phytologia.org/uploads/2/3/4/2/23422706/94171-90wayne_and_vasedges_and_c4_grasses.pdf
- [38] Van Auken, O.W. (1988) Woody Vegetation of the Southeastern Escarpment and Plateau. In: Amos, B.B. and Gehlbach, F.R., Eds., *Edwards Plateau Vegetation*, Baylor University Press, 43-55. <https://www.baylorpress.com>
- [39] Van Auken, O.W., Bush, J.K., Taylor, D.L. and Singhurst, J.R. (2023) Lack of Woody Species Recruitment in Isolated Deep Canyon Deciduous Woodlands in Central Texas, Usa1. *The Journal of the Torrey Botanical Society*, **150**, 525-537. <https://doi.org/10.3159/torrey-d-23-00009.1>
- [40] United States Department of Agriculture (USDA) (2000) Natural Resources Conservation Service. Soil Survey Division. Official Series Descriptions. <https://soilseries.sc.egov.usda.gov/>
- [41] Taylor, F.B., Hailey, R.B. and Richmond, D.L. (1966) Soil Survey of Bexar County, Texas. USDA, Soil Conservation Service. <https://www.scirp.org>
- [42] Arbingast, S.A. *et al.* (1976) Atlas of Texas. 5th Edition, Bureau of Business Research, University of Texas Press. <https://tamug-ir.tdl.org>
- [43] National Oceanic and Atmospheric Administration (NOAA) (2024) National Climatic Data Center. <https://www.ncdc.noaa.gov/cdo-web/>
- [44] Van Auken, O.W., Ford, A.L. and Allen, J.L. (1981) An Ecological Comparison of Upland Deciduous and Evergreen Forests of Central Texas. *American Journal of Botany*, **68**, 1249-1256. <https://doi.org/10.1002/j.1537-2197.1981.tb07832.x>
- [45] Turner, C.L. and Knapp, A.K. (1996) Responses of a C₄ Grass and Three C₃ Forbs to Variation in Nitrogen and Light in Tallgrass Prairie. *Ecology*, **77**, 1738-1749. <https://doi.org/10.2307/2265779>
- [46] Sall, J., Creighton, L. and Lehman, A. (2011) JMP Pro. SAS Institute, Inc. <https://www.scirp.org>
- [47] Little, E.L. (1971) Atlas of United States Trees. Volume 1, Conifers and Important Hardwoods. U.S. Dept. of Agriculture, Forest Service. <https://doi.org/10.5962/bhl.title.130546>
- [48] Elias, T.S. (1980) The Complete Trees of North America. Van Nostrand Reinhold. <https://www.amazon.com>
- [49] Van Auken, O.W. and Smeins, F. (2008) Western North American Juniperus Communities: Patterns and Causes of Distribution and Abundance. In: Van Auken, O.W., Ed., *Western North American Juniperus Communities*, Springer, 3-18. https://doi.org/10.1007/978-0-387-34003-6_1
- [50] Hull, J.C. (2002) Photosynthetic Induction Dynamics to Sunflecks of Four Deciduous Forest Understory Herbs with Different Phenologies. *International Journal of Plant Sciences*, **163**, 913-924. <https://doi.org/10.1086/342633>
- [51] Valladares, F. and Niinemets, Ü. (2008) Shade Tolerance, a Key Plant Feature of Complex Nature and Consequences. *Annual Review of Ecology, Evolution, and Systematics*, **39**, 237-257. <https://doi.org/10.1146/annurev.ecolsys.39.110707.173506>
- [52] Van Auken, O.W. and Bush, J.K. (2009) The Role of Photosynthesis in the Recruitment of Juvenile *Quercus Gambelii* into Mature *Q. Gambelii* Communities. *The Journal of the Torrey Botanical Society*, **136**, 465-478. <https://doi.org/10.3159/09-ra-035.1>
- [53] Van Auken, O.W., Jackson, J.T. and Jurena, P.N. (2005) Survival and Growth of Ju-

- niperus Seedlings in Juniperus Woodlands. *Plant Ecology*, **175**, 245-257. <https://doi.org/10.1007/s11258-005-0022-z>
- [54] Doughty, R.W. (1983) Wildlife and Man in Texas: Environmental Change and Conservation. Texas A & M University Press.
- [55] Krueger, L.M., Peterson, C.J., Royo, A. and Carson, W.P. (2009) Evaluating Relationships among Tree Growth Rate, Shade Tolerance, and Browse Tolerance Following Disturbance in an Eastern Deciduous Forest. *Canadian Journal of Forest Research*, **39**, 2460-2469. <https://doi.org/10.1139/x09-155>
- [56] Anderson, R.C. and Loucks, O.L. (1979) White-Tail Deer (*Odocoileus virginianus*) Influence on Structure and Composition of *Tsuga Canadensis* Forests. *The Journal of Applied Ecology*, **16**, 855-861. <https://doi.org/10.2307/2402859>
- [57] Augustine, D.J. and Frelich, L.E. (1998) Effects of White-Tailed Deer on Populations of an Understory Forb in Fragmented Deciduous Forests. *Conservation Biology*, **12**, 995-1004. <https://doi.org/10.1046/j.1523-1739.1998.97248.x>
- [58] Russell, F.L., Zippin, D.B. and Fowler, N.L. (2001) Effects of White-Tailed Deer (*Odocoileus virginianus*) on Plants, Plant Populations and Communities: A Review. *The American Midland Naturalist*, **146**, 1-26. [https://doi.org/10.1674/0003-0031\(2001\)146\[0001:eowtdo\]2.0.co;2](https://doi.org/10.1674/0003-0031(2001)146[0001:eowtdo]2.0.co;2)
- [59] Abrams, M.D. and Johnson, S.E. (2012) Long-Term Impacts of Deer Exlosures on Mixed-Oak Forest Composition at the Valley Forge National Historical Park, Pennsylvania, USA. *The Journal of the Torrey Botanical Society*, **139**, 167-180. <https://doi.org/10.3159/torrey-d-11-00075.1>
- [60] Bond, W.J. (2008) What Limits Trees in C₄ Grasslands and Savannas? *Annual Review of Ecology, Evolution, and Systematics*, **39**, 641-659. <https://doi.org/10.1146/annurev.ecolsys.39.110707.173411>
- [61] Leonard, W.J. and Auken, O.W.V. (2013) Light Levels and Herbivory Partially Explain the Survival, Growth, and Niche Requirements of *Streptanthus bracteatus* A. Gray (*Bracted Twistflower*, Brassicaceae), a Rare Central Texas Endemic. *Natural Areas Journal*, **33**, 276-285. <https://doi.org/10.3375/043.033.0306>
- [62] Van Auken, O.W. and McKinley, D.C. (2008) Structure and Composition of *Juniperus* Communities and Factors That Control Them. In: Van Auken, O.W., Ed., *Western North American Juniperus Communities*, Springer, 19-47. https://doi.org/10.1007/978-0-387-34003-6_2
- [63] Van Auken, O.W. and Bush, J.K. (2013) Invasion of Woody Legumes. Springer. <https://doi.org/10.1007/978-1-4614-7199-8>
- [64] Cadenasso, M.L., Pickett, S.T.A. and Morin, P.J. (2002) Experimental Test of the Role of Mammalian Herbivores on Old Field Succession: Community Structure and Seedling Survival. *Journal of the Torrey Botanical Society*, **129**, 228-237. <https://doi.org/10.2307/3088773>
- [65] Wayne, E.R. and Van Auken, O.W. (2024) Drought Tolerance and Recovery of the Sedge *Carex planostachys* (Cyperaceae) from Central Texas Woodlands. *American Journal of Plant Sciences*, **15**, 746-765. <https://doi.org/10.4236/ajps.2024.159048>
- [66] Korsakova, S., Plugatar, Y., Pashtetsky, A. and Ilnitsky, O. (2020) Influence of Abiotic Factors on CO₂-Gas Exchange of *Pinus pallasiana*, *Juniperus excelsa* and *Arbutus andrachne*. *E3S Web of Conferences*, **224**, Article ID: 04017. <https://doi.org/10.1051/e3sconf/202022404017>