Experimental drought and heat can delay phenological development and reduce foliar and shoot growth in semiarid trees.

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Supporting Information.

Additional Non-Structural Carbohydrate Methods.

Non-structural carbohydrates were assayed following the protocol of Dickman et al. (2015), which was modified from that of Hoch *et al*. (2002). We added approximately 12 mg of fine ground plant material to a 2 mL deep-well plate for extraction with 1.6 mL distilled water for 60 minutes at 100°C in a water bath (Isotemp 105, Fisher Scientific, Hampton, NH). A 700 µL aliquot was removed for starch analysis and the remaining extract was centrifuged (Allegra X-15R, Beckman Coulter, Brea, CA) for 45 minutes at 4450 rpm. To determine free sugars, we used 20 µL of untreated supernatant from the centrifuged extract for conversion of fructose to glucose and glucose to gluconate-6-phosphate. The 20 µL aliquot was incubated in a microplate shaker (BioShaker M.BR-022UP, TAITEC, Koshigaya, Japan) for 45 minutes with phosphoglucose isomerase (extracted from Baker's yeast – Type III, Sigma-Aldrich, St. Louis, MO), glucose hexokinase and glucose-6-P dehydrogenase (Glucose Assay Reagent, Sigma Aldrich, St. Louis, MO). Free glucose concentrations were determined photometrically in a 96-well microplate spectrophotometer (Cary 50 UV-Vis, Agilent Technologies, Santa Clara, CA) from the optical density increase at 340 nm due to the reduction of NAD+ to NADH as glucose-6-P was oxidized, and by correcting this result relative to photometric measurement of glucose standards of a known concentration.

To determine sucrose concentrations we first hydrolysed sucrose to glucose and fructose. We incubated a 100 µL aliquot of centrifuged supernatant in a microplate shaker for 40 minutes with 50 µL of invertase (Grade VII, from Baker's yeast, Sigma-Aldrich, St. Louis, MO) buffered to pH 4.6 with 0.4 M, NaOAc (Sigma Aldrich, St. Louis, MO). Then, a 20 µL aliquot of this invertase-treated sample was used to determine total glucose as described above. Sucrose was calculated as low molecular weight sugars minus free sugars.

We determined starch concentrations from the difference between total non-structural carbohydrates (NSC) minus low molecular weight sugars (glucose, fructose, and sugars). To determine total NSC we used amyloglucosidase (from *Aspergillus niger*, Sigma Aldrich, St. Louis, MO) buffered to pH 4.5 with 0.1 M NaOAc (Sigma Aldrich, St. Louis, MO) to convert all NSC components (sugars and starches) to glucose (Pazur & Kleppe, 1962). The 700 µL of sample extract set aside following the initial extraction was transferred to a new deep-well plate and incubated overnight at 48°C in a water bath. Following incubation, the plate was centrifuged for 60 minutes at 4450 rpm, and we determined the glucose concentration photometrically of converted total NSC with a 20 µL aliquot of supernatant as described above. All NSC, sugar, and starch component values were calculated as a percent of dry sample mass.

References.

Dickman LT, McDowell NG, Sevanto S et al. (2015) Carbohydrate dynamics and mortality in a piñon-juniper woodland under three future precipitation scenarios. Plant Cell and Environment, 38, 729-739.

Hoch G, Popp M, Körner C (2002) Altitudinal increase of mobile carbon pools in *Pinus cembra* suggests sink limitation of growth at the Swiss treeline. Oikos, 98, 361-374.

Pazur JH, Kleppe K (1962) Hydrolysis of alpha-d-glucosides by amyloglucosidase from *Aspergillus niger*. Journal of Biological Chemistry, 237, 1002-1006.

Supporting Figures

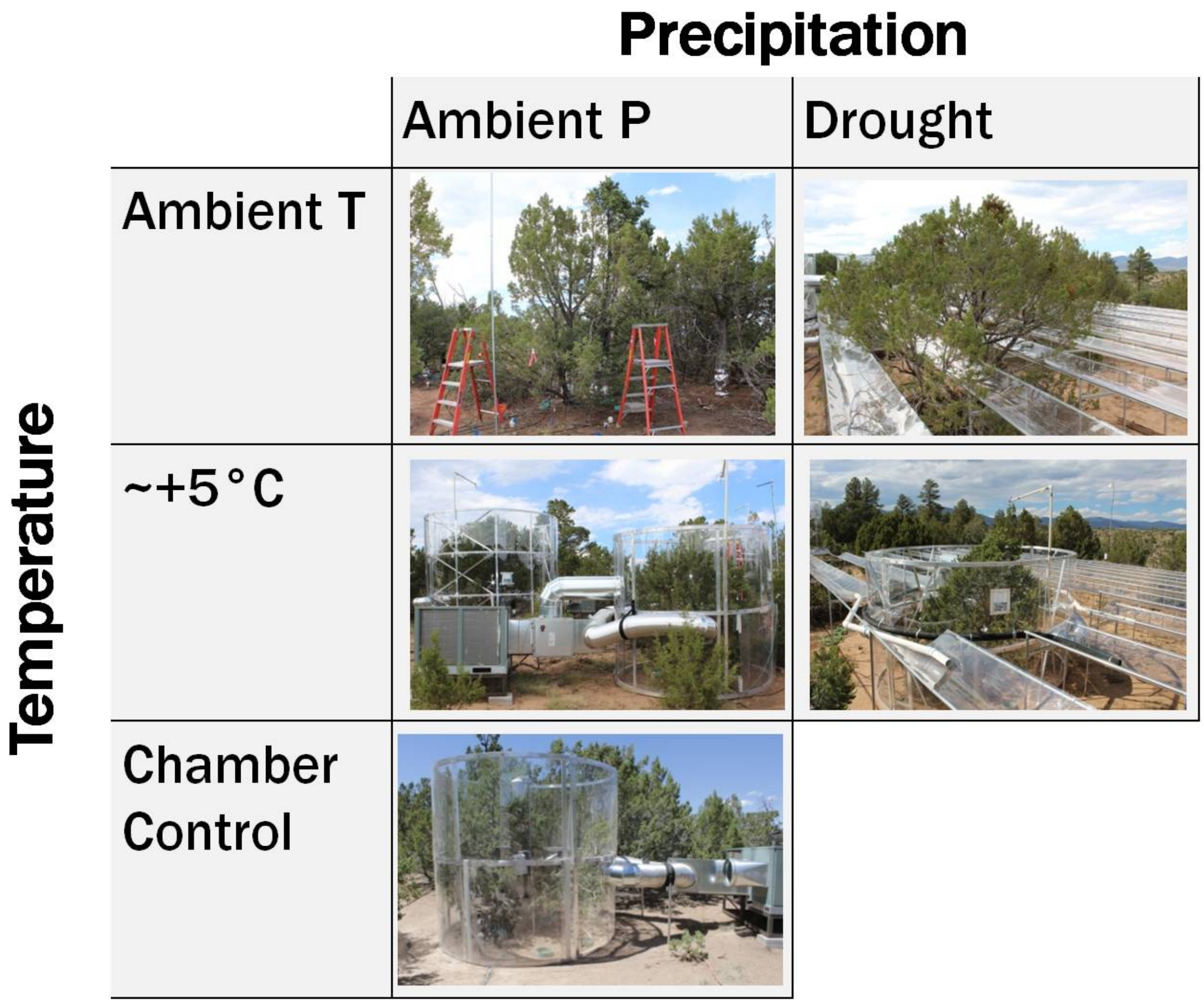

Fig S1. Design of the Los Alamos Survival-Mortality experiment (SUMO). Drought was induced with a ~45% throughfall rain-out structure and temperature was modified with transparent plastic open-top chambers regulated by heating and cooling units. Precipitation and temperature factors were combined to provide ambient, drought, heat, and drought+heat treatments. A chamber control treatment was implemented with an additional set of open-top chambers regulated to ambient field air temperature.

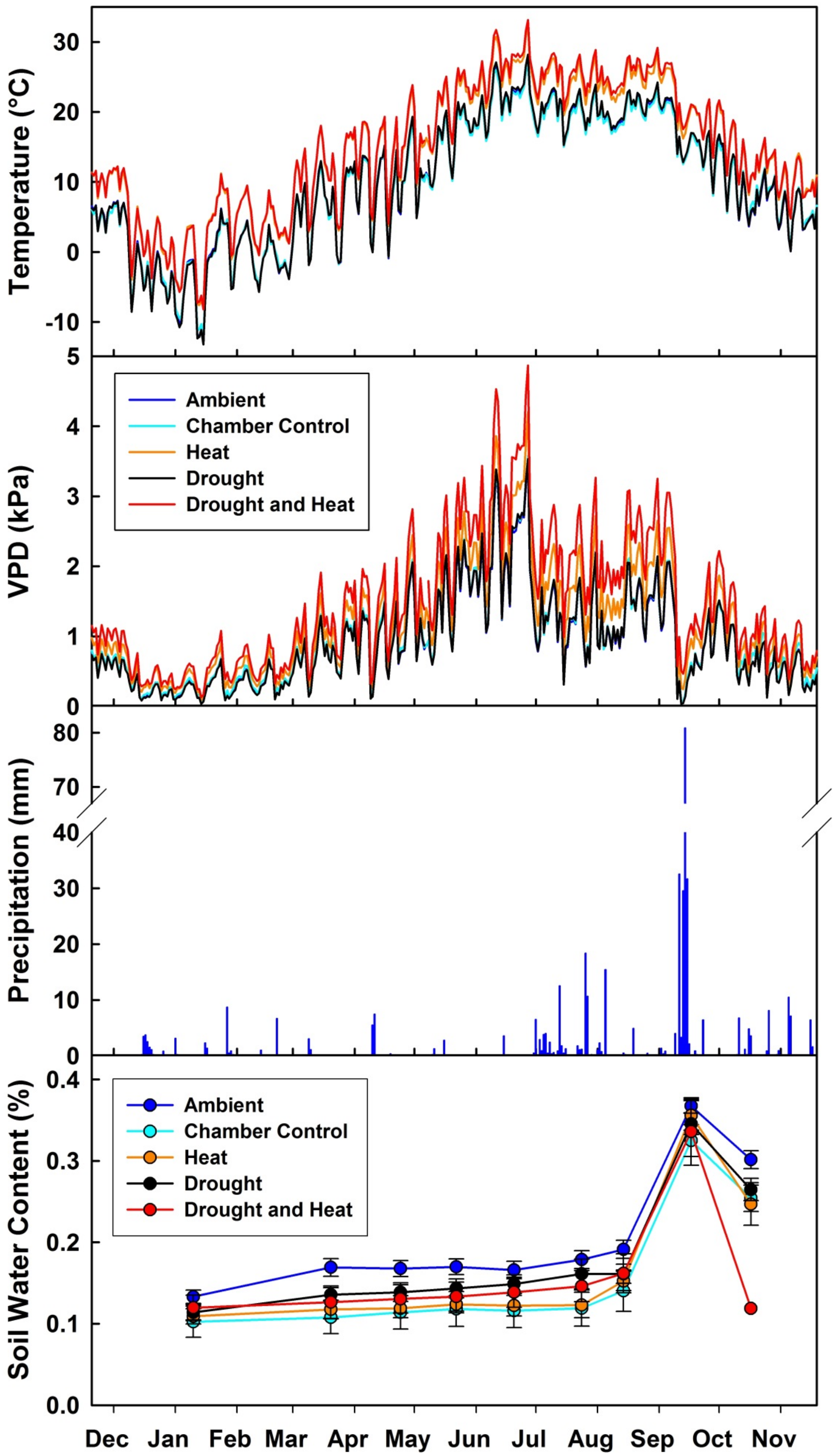
Temperature (°C)
VPD (kPa)
Ambient
Chamber Control
Heat
Drought
Drought and Heat
Precipitation (mm)
Soil Water Content (%)
Ambient
Chamber Control
Heat
Drought
Drought and Heat
Dec
Jan
Feb
Mar
Apr
May
Jun
Jul
Aug
Sep
Oct
Nov

Fig S2. Environmental data measured at the SUMO experiment from December 2012 to November 2013, including mean daily temperature and vapor pressure deficit (VPD) by treatment, total daily precipitation at the site, and period mean soil water content (10 to 40 cm) by treatment. For temperature and VPD panels, separate lines representing treatments often overlap.

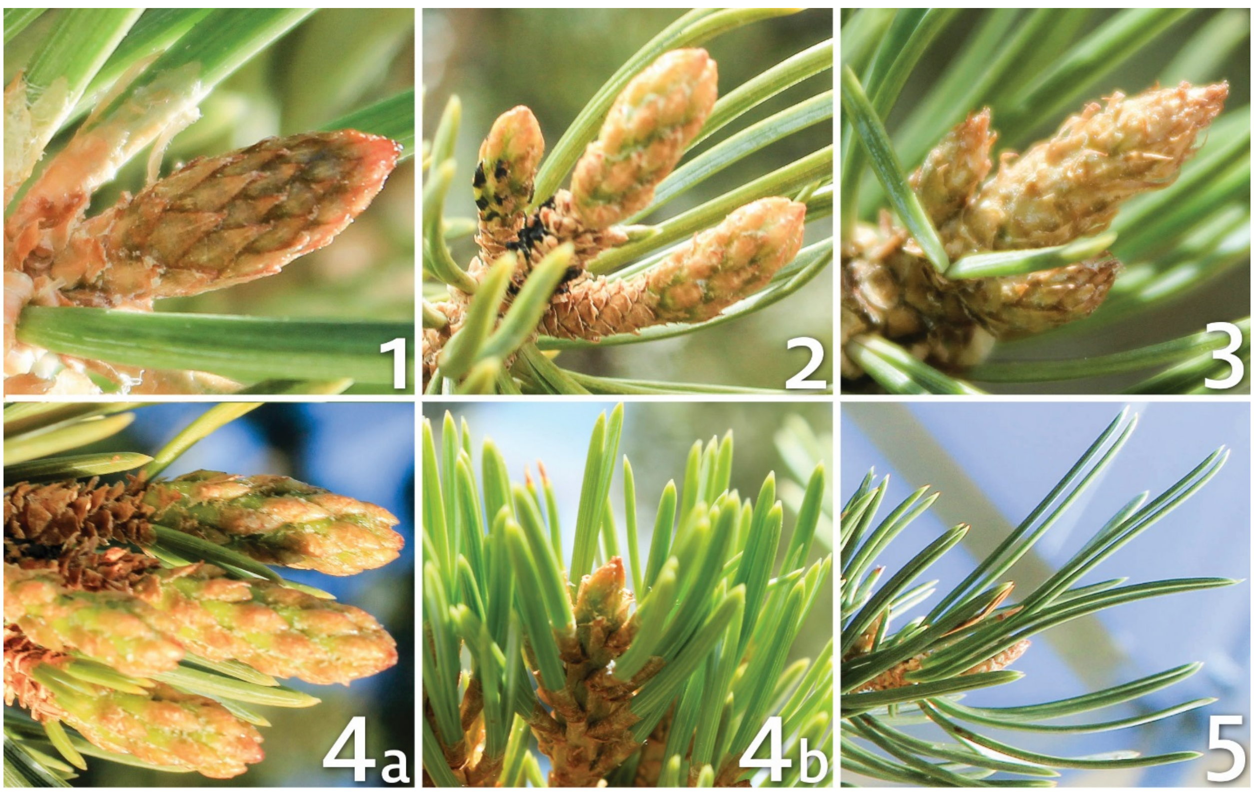

Fig S3. Phenological phases in our classification scheme for piñon pine: 1) bud dormant and unchanged in size, 2) bud swelling or growth observed, 3) needle scales open (budbreak), 4) new needle emergence and growth (both early (4a) and later (4b) examples of this stage are shown), 5) needle pairs separate.

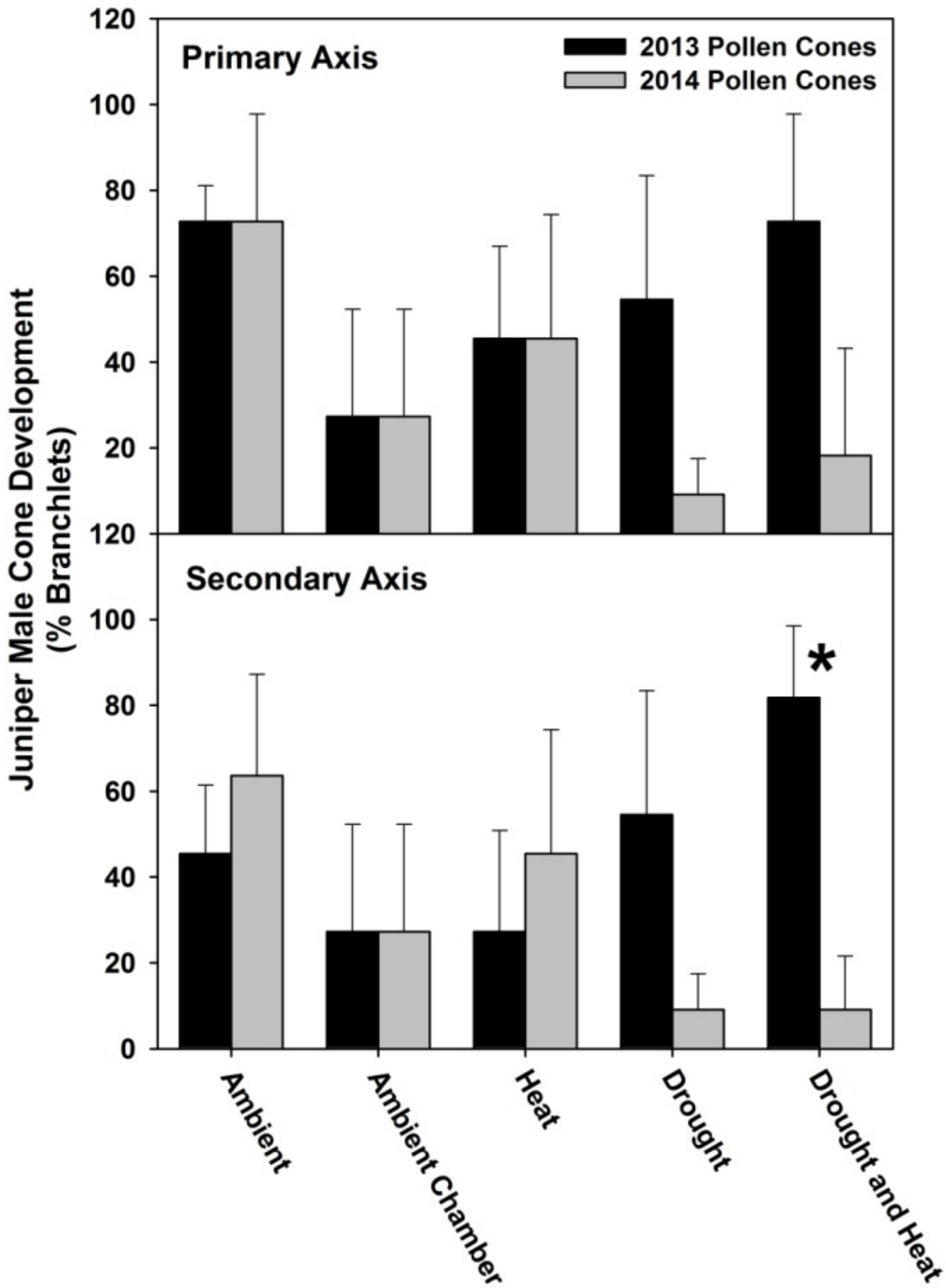

Fig S4. Male pollen cone production in one-seed juniper by treatment. Percent of juniper trees (n= 20) with male cones observed releasing pollen in early 2013 and also those developed late in the growing season for 2014 pollen release are shown. Significant differences between 2013 and 2014 cones are noted with an asterisk (Kruskal-Wallis test, $p < 0.05$). Error bars are standard errors.

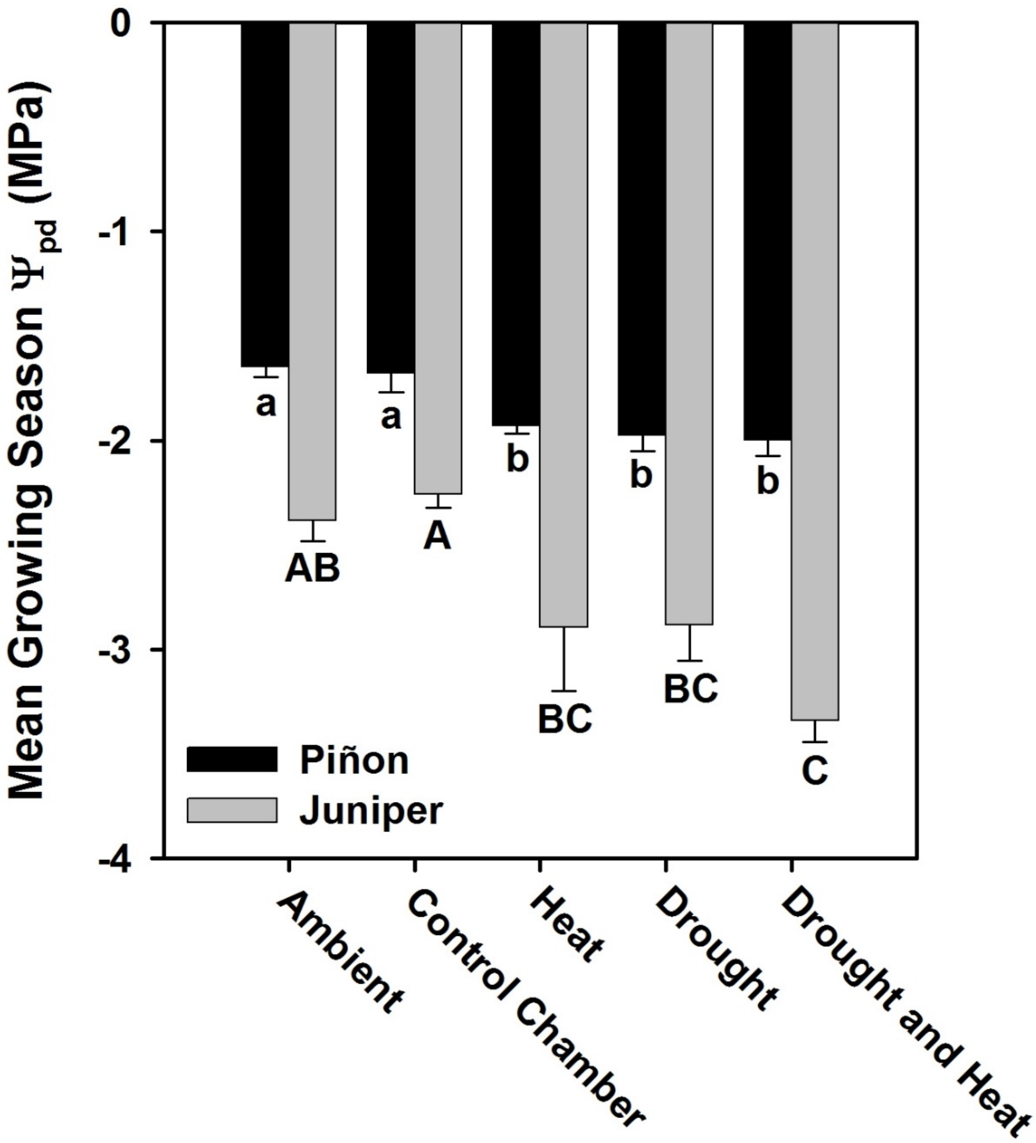

Fig S5. Mean growing season pre-dawn water potential (Ψ_{pd}) for piñon pine and juniper study trees by treatment (n = 20). Significant differences among treatments are indicated with letters for piñon pine (lower case) and juniper (upper case; ANOVA, $p < 0.05$). Error bars are standard errors.

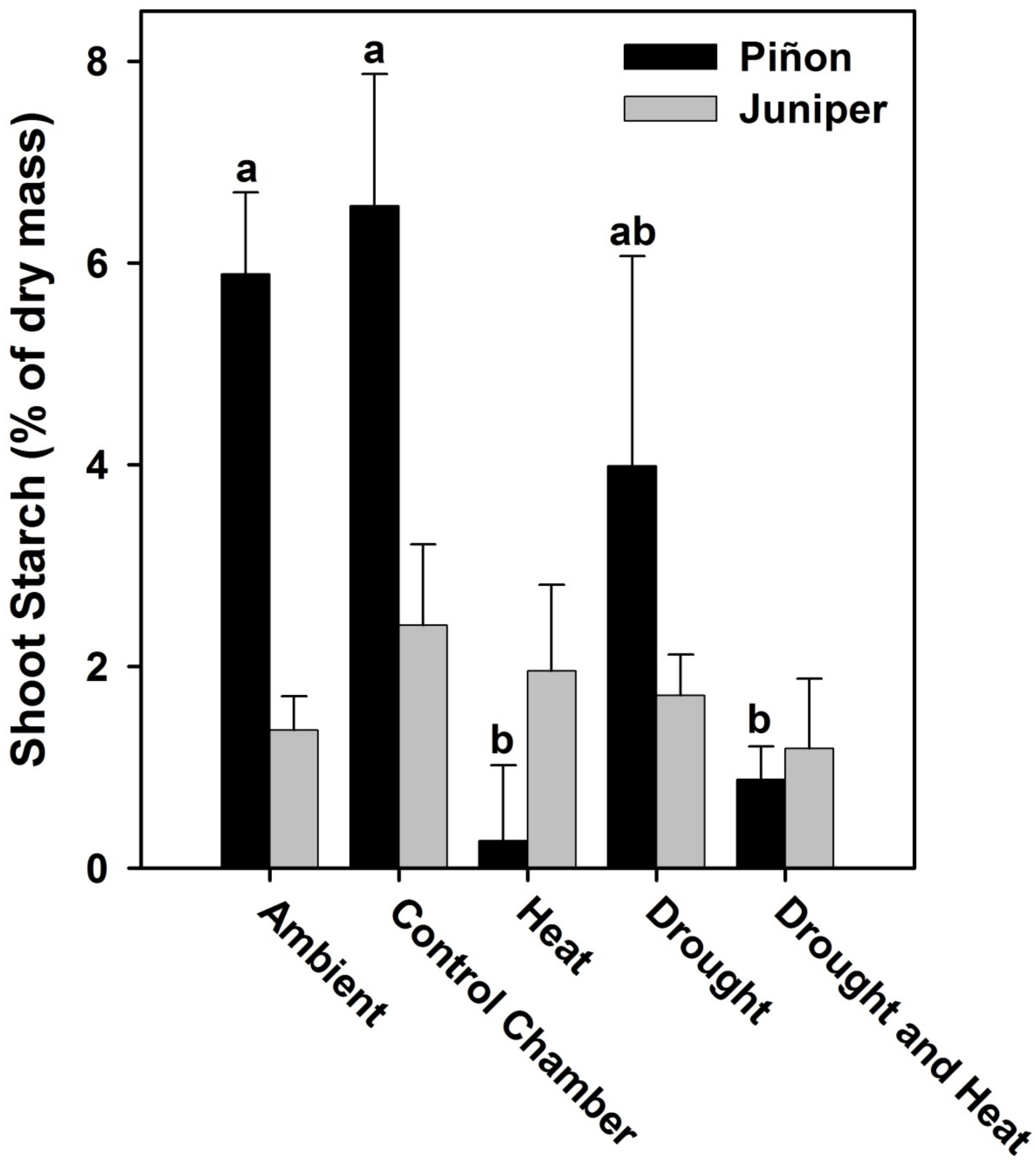

Fig S6. Mean June starch concentration in shoots of piñon pine and juniper study trees by treatment (n = 20). Significant differences among treatments are indicated with letters for piñon pine (ANOVA, $p < 0.05$), for juniper there were no significant differences among treatments (ANOVA, $p > 0.05$). Error bars are standard errors.

Supplemental Tables

Table S1. Correlations are shown between pre-dawn shoot water potential (monthly and mean growing season) and mean tree shoot growth, needle growth, and needle emergence timing in primary and secondary axis branches of piñon pine ($n = 20$ for each axis). Only significant correlation coefficients (r) are shown ($p < 0.05$). The relationships between mean growing season water potential and shoot growth, growth, and needle emergence timing are also shown in Fig 5. There were no significant correlations for juniper between shoot growth and water potential.

	Phenology	**Correlation coefficient with water potential**								
Axis	**Measurement**	**March**	**April**	**May**	**June**	**July**	**August**	**Sept.**	**Oct.**	**Mean**
Primary	Shoot growth	0.59		0.70	0.49		0.46			0.62
	Needle growth	0.66		0.66	0.48		0.59			0.66
	Needle emergence	-0.61		-0.60			-0.65			-0.70
Secondary	Shoot growth	0.73		0.64	0.48			0.50		0.71
	Needle growth	0.79		0.60	0.57		0.60	0.45		0.82
	Needle emergence									-0.55

Table S2. Correlations between June non-structural carbohydrate (NSC) concentrations and mean tree shoot growth, needle growth, and needle emergence timing in primary and secondary axis branches of piñon pine (n = 20 for each axis). Correlations are shown separately for NSC components of glucose and fructose (Gluc & Fruc), sucrose, starch, and total NSC concentrations. Only significant correlation coefficients (r) are shown ($p < 0.05$). The relationships between shoot starch content and shoot growth, growth, and needle emergence timing are also shown in Fig 5. Significant correlations were found for juniper shoot growth in secondary axis branches with shoot glucose and fructose ($r = -0.53$, $p < 0.05$) and shoot total NSC ($r = -0.46$, $p < 0.05$; data not show).

	NSC	**Primary Axis**			**Secondary Axis**		
Tissue	**Component**	**Shoot growth**	**Needle growth**	**Needle emergence**	**Shoot growth**	**Needle growth**	**Needle emergence**
Bole	Gluc & Fruc						
	Sucrose						
	Starch					0.54	
	Total NSC						
Needle	Gluc & Fruc						
	Sucrose						
	Starch			-0.45	0.51	0.55	
	Total NSC				0.47	0.46	
Root	Gluc & Fruc						
	Sucrose						
	Starch						
	Total NSC						
Shoot	Gluc & Fruc	-0.48					
	Sucrose					0.52	
	Starch	0.49	0.55	-0.65	0.62	0.77	
	Total NSC		0.51	-0.57	0.58	0.77	

Table S3. Correlations between pre-dawn shoot water potential (monthly and mean growing season) and June NSC and components in piñon pine (n = 20). Only significant correlation coefficients (r) are shown ($p < 0.05$).

	NSC	Correlation coefficient with water potential								
Tissue	**Component**	**March**	**April**	**May**	**June**	**July**	**August**	**Sept.**	**Oct.**	**Mean**
Bole	Gluc & Fruc									
	Sucrose	-0.51	-0.68							-0.52
	Starch					0.55				
	Total NSC		-0.58							
Needle	Gluc & Fruc							0.47		
	Sucrose									
	Starch	0.55						0.44		0.55
	Total NSC							0.53		
Root	Gluc & Fruc									
	Sucrose									
	Starch					0.60	0.61			0.59
	Total NSC									0.58
Shoot	Gluc & Fruc			-0.61	-0.53					
	Sucrose									
	Starch	0.50			0.67	0.58	0.49			0.70
	Total NSC	0.49			0.54	0.56	0.45			0.63

Table S4. Mean June concentrations (% of dry tissue mass) and standard error by treatment of glucose and fructose (Gluc & Fruc), sucrose, starch, and total NSC in bole, leaf, root, and shoot tissue of piñon pines (n=20).

	NSC	**Ambient**		**Control Chamber**		**Heat**		**Drought**		**Drought + Heat**	
Tissue	**Component**	**Mean**	**SE**	**Mean**	**SE**	**Mean**	**SE**	**Mean**	**SE**	**Mean**	**SE**
Bole	Gluc & Fruc	0.31	0.03	0.71	0.20	0.57	0.21	0.66	0.12	0.79	0.26
	Sucrose	0.52	0.23	0.82	0.12	0.82	0.28	0.99	0.29	0.94	0.23
	Starch	1.15	0.36	1.64	0.36	0.50	0.12	0.94	0.14	0.27	0.11
	Total NSC	1.98	0.59	3.17	0.66	1.88	0.53	2.60	0.50	2.00	0.55
Needle	Gluc & Fruc	2.45	0.28	2.17	0.31	1.64	0.38	2.33	0.13	2.24	0.28
	Sucrose	1.28	0.24	2.10	0.26	1.63	0.36	1.23	0.14	1.58	0.24
	Starch	0.82	0.47	0.78	0.20	0.15	0.07	0.21	0.06	0.00	0.00
	Total NSC	4.56	0.68	5.04	0.64	3.42	0.66	3.77	0.19	3.83	0.11
Root	Gluc & Fruc	1.53	0.50	0.96	0.00	0.97	0.06	0.73	0.18	1.03	0.00
	Sucrose	1.31	0.17	1.13	0.00	0.62	0.13	0.68	0.22	0.64	0.19
	Starch	1.69	0.76	0.81	0.00	0.77	0.22	0.87	0.23	0.36	0.31
	Total NSC	4.53	0.75	2.90	0.00	2.35	0.03	2.28	0.32	2.02	0.12
Shoot	Gluc & Fruc	2.01	0.06	2.30	0.40	2.79	0.23	2.27	0.26	2.26	0.33
	Sucrose	3.03	0.36	3.61	0.57	2.67	0.21	3.02	0.19	2.24	0.54
	Starch	5.89	0.80	6.57	1.31	0.99	0.29	3.99	2.08	0.88	0.33
	Total NSC	10.93	0.91	12.47	1.68	6.45	0.25	9.28	2.00	5.38	0.87

Table S5. Mean June concentrations (% of dry tissue mass) and standard error by treatment of glucose and fructose (Gluc & Fruc), sucrose, starch, and total NSC in bole, leaf, root, and shoot tissue of juniper trees (n=20). Root data were unavailable for juniper in the control chamber treatment.

	NSC	Ambient		Control Chamber		Heat		Drought		Drought + Heat	
Tissue	**Component**	**Mean**	**SE**	**Mean**	**SE**	**Mean**	**SE**	**Mean**	**SE**	**Mean**	**SE**
Bole	Gluc & Fruc	0.45	0.17	0.55	0.20	0.63	0.12	0.84	0.18	0.93	0.14
	Sucrose	0.26	0.12	0.28	0.09	0.32	0.11	0.60	0.10	0.32	0.07
	Starch	0.58	0.24	0.51	0.09	0.79	0.19	0.36	0.17	0.51	0.19
	Total NSC	1.29	0.45	1.33	0.37	1.74	0.25	1.80	0.35	1.77	0.14
Needle	Gluc & Fruc	2.82	0.42	3.13	0.56	3.31	0.28	4.40	0.50	4.12	0.25
	Sucrose	1.97	0.11	2.13	0.19	1.92	0.34	1.59	0.40	1.64	0.27
	Starch	5.41	1.03	7.15	0.99	3.28	1.15	3.77	0.58	3.42	0.99
	Total NSC	10.20	0.65	12.40	0.76	8.51	1.00	9.76	0.56	9.18	0.76
Root	Gluc & Fruc	1.04	0.73	NA		2.05	1.15	1.89	0.34	2.42	0.65
	Sucrose	0.39	0.26	NA		0.57	0.16	0.84	0.18	0.50	0.14
	Starch	1.79	1.14	NA		5.26	1.92	3.24	1.07	3.47	0.69
	Total NSC	3.23	1.35	NA		7.88	1.97	5.96	1.50	6.38	1.05
Shoot	Gluc & Fruc	1.65	0.18	2.19	0.41	1.68	0.30	1.43	0.28	1.97	0.25
	Sucrose	1.39	0.22	1.24	0.43	1.16	0.17	1.45	0.19	1.16	0.16
	Starch	1.37	0.34	2.41	0.80	1.96	0.86	1.71	0.40	1.19	0.69
	Total NSC	4.40	0.33	5.84	1.32	4.80	1.01	4.59	0.84	4.32	0.80