

Soil microbial drought history affects physiological response of select tree species to drought stress

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Supplementary figures and tables

Table S1 Results of linear model to investigate the effects of watering treatments, microbial history, species, and their interactions (response~Soil history*Species*Watering treatment). The sum of squares, df, F-value, and *P* are listed for each model of *A* (photosynthetic assimilation), *gsw* (stomatal conductance), Ψ_{Leaf} (leaf water potential), stem sugar NSC (% of dry mass), and stem starch NSC (% of dry mass). Bolding of *P*-values represents that $P < 0.05$

		df	F-value	<i>P</i>
<i>A</i> ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$)				
	[Intercept]	1	240	<2.2e-16
	Soil history	1	3.39	0.071
	Species	2	29.5	1.8E-09
	Watering treatment	1	47.1	5.8E-09
	Soil history*Species	2	0.241	0.79
	Soil history*Watering treatment	1	5.25	0.026
	Species*Watering treatment	2	1.25	0.30
	Soil history*Species*Watering treatment	2	7.75	0.0011
	Residuals	56		
<i>gsw</i> ($\text{mol H}_2\text{O m}^{-2} \text{ s}^{-1}$)				
	[Intercept]	1	178	<2.2e-16
	Soil history	1	2.16	0.15
	Species	2	29.7	1.6E-09
	Watering treatment	1	46.7	6.6E-09
	Soil history*Species	2	0.331	0.72
	Soil history*Watering treatment	1	3.52	0.066
	Species*Watering treatment	2	1.95	0.15
	Soil history*Species*Watering treatment	2	12.5	3.3E-05
	Residuals	56		
Ψ_{Leaf} (MPa)				
	[Intercept]	1	1980	<2.2e-16
	Soil history	1	7.40	0.0086
	Species	2	67.6	9.1E-16
	Watering treatment	1	42.0	2.3E-08
	Soil history*Species	2	5.26	0.0080
	Soil history*Watering treatment	1	2.73	0.10
	Species*Watering treatment	2	12.9	2.4E-05
	Soil history*Species*Watering treatment	2	5.29	0.0078
	Residuals	56		
Stem sugar NSC (% of dry mass)				
	[Intercept]	1	285	<2.2e-16
	Soil history	1	9.03	0.0040
	Species	2	19.9	3.6E-07
	Watering treatment	1	8.76	0.0046
	Soil history*Species	2	2.04	0.14
	Soil history*Watering treatment	1	0.0642	0.80
	Species*Watering treatment	2	10.6	0.00013
	Soil history*Species*Watering treatment	2	1.93	0.16
	Residuals	56		
Stem starch NSC (% of dry mass)				
	[Intercept]	1	258	<2.2e-16
	Soil history	1	0.480	0.49
	Species	2	14.1	1.3E-05
	Watering treatment	1	0.836	0.36
	Soil history*Species	2	2.41	0.10
	Soil history*Watering treatment	1	0.0291	0.87
	Species*Watering treatment	2	5.41	0.0073
	Soil history*Species*Watering treatment	2	1.18	0.31
	Residuals	56		

Table S2 Means $\pm$ standard error of A (photosynthetic assimilation), g_{sw} (stomatal conductance), and Ψ_{Leaf} (leaf water potential) when planted in sterilized soil for each of the treatment combinations and species. Superscript letters represent significant differences among treatments within a species for a given measurement. NA listed for SE is a result of a sample size of 1 due to mortality of trees within that treatment group

Species	Watering Treatment	Soil History	A ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$)	g_{sw} ($\text{mol H}_2\text{O m}^{-2} \text{ s}^{-1}$)	Ψ_{Leaf} (MPa)
<i>Prunus virginiana</i>					
	Well-watered	Control	15 $\pm$ 6 ^a	0.08 $\pm$ 0.10 ^a	-1.8 $\pm$ 0.2 ^a
	Well-watered	Drought-stressed	4.8 $\pm$ 3 ^a	0.08 $\pm$ 0.03 ^a	-1.5 $\pm$ 0.2 ^a
	Water-stressed	Control	6.3 $\pm$ 2 ^a	0.06 $\pm$ 0.02 ^a	-2.3 $\pm$ 0.3 ^a
	Water-stressed	Drought-stressed	6.2 $\pm$ 2 ^a	0.08 $\pm$ 0.03 ^a	-2.5 $\pm$ 0.2 ^a
<i>Quercus rubra</i>					
	Well-watered	Control	12 $\pm$ 4 ^a	0.20 $\pm$ 0.08 ^a	-1.6 $\pm$ 0.3 ^a
	Well-watered	Drought-stressed	9.3 $\pm$ 2 ^a	0.13 $\pm$ 0.02 ^a	-2.3 $\pm$ 0.08 ^a
	Water-stressed	Control	0.4 $\pm$ 1 ^a	0.01 $\pm$ 0.01 ^a	-2.6 $\pm$ 0.2 ^a
	Water-stressed	Drought-stressed	9.6 $\pm$ 4 ^a	0.15 $\pm$ 0.08 ^a	-2.3 $\pm$ 0.1 ^a
<i>Liriodendron tulipifera</i>					
	Well-watered	Control	6.1 $\pm$ 0.2 ^a	0.10 $\pm$ 0.01 ^a	-1.0 $\pm$ 0.1 ^a
	Well-watered	Drought-stressed	5.6 $\pm$ 2 ^a	0.15 $\pm$ 0.07 ^a	-1.1 $\pm$ 0.06 ^a
	Water-stressed	Control	5.5 $\pm$ NA ^a	0.09 $\pm$ NA ^a	-0.8 $\pm$ NA ^a
	Water-stressed	Drought-stressed	1.8 $\pm$ 0.8 ^a	0.01 $\pm$ 0.003 ^a	-1.5 $\pm$ 0.2 ^a

Table S3 Results of linear model to investigate the effects of watering treatments, soil history, and their interactions (physiological response~Soil history*Watering treatment*Sterilization) for *P. virginiana*. The sum of squares, df, F-value, and *P* are listed for each model of *A* (photosynthetic assimilation), *g_{sw}* (stomatal conductance), and Ψ_{Leaf} (leaf water potential). Bolding of *P*-values represents that *P* < 0.05

	df	F-value	<i>P</i>
<i>A</i> ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$)			
[Intercept]	1	101	1.2E-10
Soil history	1	0.486	0.49
Watering treatment	1	8.45	0.0072
Sterilization	1	6.13	0.020
Soil history*Watering treatment	1	9.78	0.0042
Soil history*Sterilization	1	3.09	0.090
Watering treatment*Sterilization	1	1.37	0.25
Soil history*Watering treatment*Sterilization	1	0.527	0.47
Residuals	27		
<i>g_{sw}</i> ($\text{mol H}_2\text{O m}^{-2} \text{ s}^{-1}$)			
[Intercept]	1	60.2	2.4E-08
Soil history	1	0.0849	0.77
Watering treatment	1	4.95	3.5E-02
Sterilization	1	12.3	1.6E-03
Soil history*Watering treatment	1	4.93	0.035
Soil history*Sterilization	1	0.00240	0.96
Watering treatment*Sterilization	1	3.40	0.076
Soil history*Watering treatment*Sterilization	1	4.01	0.055
Residuals	27		
Ψ_{Leaf} (MPa)			
[Intercept]	1	924	<2.2E-16
Soil history	1	4.03	0.055
Watering treatment	1	48.6	1.7E-07
Sterilization	1	0.475	0.50
Soil history*Watering treatment	1	1.39	0.25
Soil history*Sterilization	1	4.80	0.037
Watering treatment*Sterilization	1	2.69	0.11
Soil history*Watering treatment*Sterilization	1	7.35	0.012
Residuals	27		

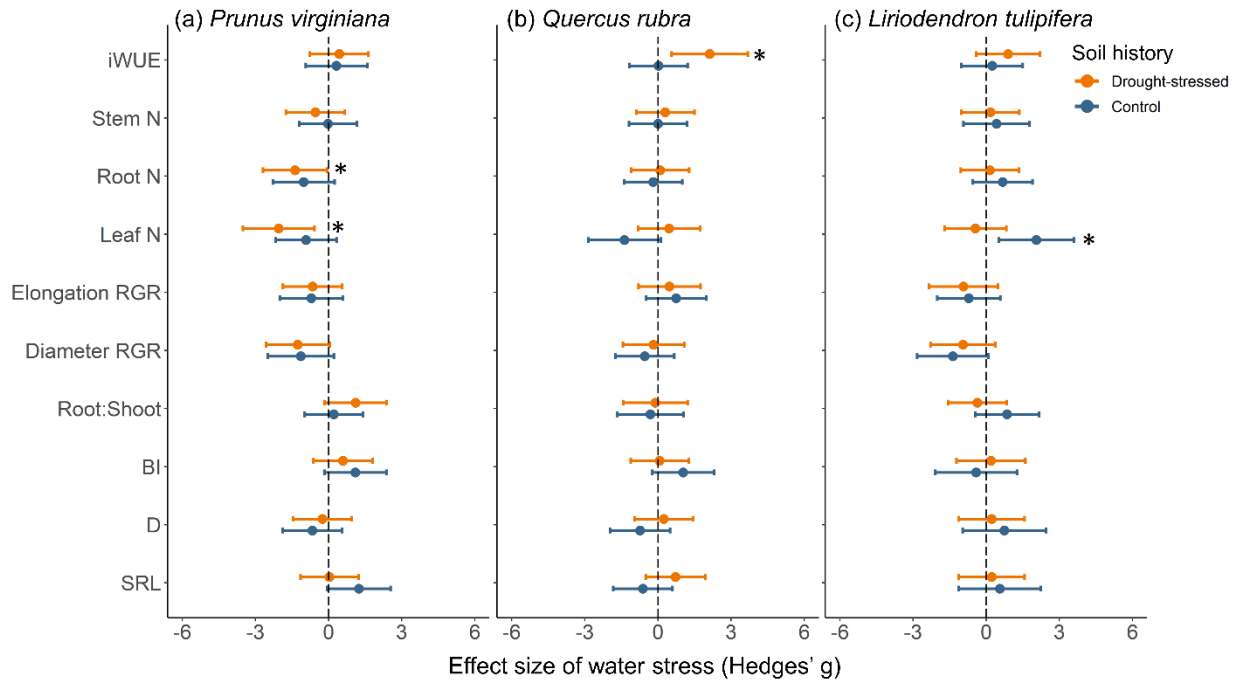


Fig. S1 Hedges' g effect sizes of water stress for iWUE (intrinsic water use efficiency), stem N, root N, leaf N, elongation RGR (relative growth rate), diameter RGR, root:shoot, BI (branching intensity), D (diameter), and SRL (specific root length) for *P. virginiana* (panel A), *Q. rubra* (panel B), and *L. tulipifera* (panel C). Effect sizes represent the difference between the well-watered and the water-stressed watering treatments within a certain soil history is designated by color of the points (control soil history – blue, drought-stressed soil history – orange). Error bars represent the 95% confidence intervals of the effect size, and the stars represent a significant effect of treatment

Species	Watering Treatment	Soil History	iWUE	Stem N (%)	Root N (%)	Leaf N (%)	Stem elongation RGR (cm)	Stem diameter RGR (cm)	Root:Shoot	BI (branch mm ⁻¹)	RD (mm)	SRL
<i>Prunus virginiana</i>	Well-watered	Control	58±3 ^a	1.1±0.11 ^{ab}	1.5±0.1 ^{ab}	2.5±0.17 ^{ab}	0.61±0.28 ^a	0.61±0.07 ^a	1.2±0.11 ^a	0.32±0.02 ^a	0.51±0.05 ^a	19±3 ^a
	Well-watered	Drought-stressed	70±5 ^a	1.0±0.10 ^a	1.9±0.16 ^a	2.7±0.09 ^a	0.89±0.40 ^a	0.62±0.11 ^a	1.1±0.13 ^a	0.32±0.01 ^a	0.44±0.05 ^a	22±5 ^a
	Water-stressed	Control	64±12 ^a	1.0±0.11 ^b	1.3±0.09 ^b	2.1±0.12 ^b	0.20±0.06 ^a	0.44±0.04 ^{ab}	1.2±0.06 ^a	0.38±0.02 ^a	0.43±0.04 ^a	24±3 ^a
	Water-stressed	Drought-stressed	79±9 ^a	0.9±0.06 ^b	1.4±0.12 ^b	2.1±0.13 ^b	0.31±0.25 ^a	0.28±0.09 ^b	1.4±0.08 ^a	0.34±0.02 ^a	0.41±0.03 ^a	22±4 ^a
<i>Quercus rubra</i>	Well-watered	Control	83±8 ^{ab}	0.6±0.03 ^a	1.2±0.13 ^a	2.4±0.08 ^a	0.02±0.01 ^a	0.63±0.08 ^a	1.5±0.15 ^a	0.60±0.07 ^a	0.57±0.11 ^a	10±2 ^a
	Well-watered	Drought-stressed	65±6 ^a	0.6±0.04 ^a	1.3±0.15 ^a	2.3±0.12 ^a	0.11±0.05 ^a	0.46±0.11 ^a	1.7±0.16 ^a	0.60±0.13 ^a	0.57±0.05 ^a	6.6±0.7 ^a
	Water-stressed	Control	84±19 ^{ab}	0.6±0.03 ^a	1.1±0.07 ^a	2.1±0.11 ^a	0.30±0.20 ^a	0.44±0.17 ^a	1.3±0.18 ^a	0.80±0.08 ^a	0.42±0.03 ^a	7.6±2 ^a
	Water-stressed	Drought-stressed	120±14 ^b	0.7±0.09 ^a	1.3±0.12 ^a	2.4±0.13 ^a	0.18±0.06 ^a	0.41±0.13 ^a	1.6±0.07 ^a	0.62±0.10 ^a	0.65±0.16 ^a	9.0±2 ^a
<i>Liriodendron tulipifera</i>	Well-watered	Control	95±11 ^a	0.7±0.12 ^a	0.95±0.06 ^a	1.6±0.08 ^a	3.0±0.82 ^a	0.45±0.07 ^a	0.73±0.07 ^a	0.65±0.28 ^a	0.39±0.02 ^a	1.8±1 ^a
	Well-watered	Drought-stressed	69±5 ^a	0.8±0.13 ^a	1.1±0.01 ^a	2.4±0.43 ^a	4.2±1.5 ^a	0.47±0.18 ^a	0.91±0.09 ^a	0.45±0.21 ^a	0.42±0.13 ^a	3.5±1 ^a
	Water-stressed	Control	112±41 ^a	0.9±0.36 ^a	1.2±0.15 ^a	2.1±0.10 ^a	1.7±0.51 ^a	0.20±0.08 ^a	0.90±0.08 ^a	0.40±0.26 ^a	0.80±0.42 ^a	7.8±4 ^a
	Water-stressed	Drought-stressed	155±47 ^a	0.1±0.45 ^a	1.1±0.10 ^a	2.0±0.14 ^a	1.9±0.67 ^a	0.19±0.03 ^a	0.82±0.09 ^a	0.53±0.04 ^a	0.48±0.07 ^a	6.0±2 ^a

Table S4 Means ± standard error of iWUE (intrinsic water use efficiency), stem N, root N, leaf N, elongation RGR (relative growth rate), diameter RGR, root:shoot, BI (branching intensity), D (diameter), and SRL (specific root length) for each of the treatment combinations and species. Letters represent significant differences in variables among treatments within species

Table S5 Number of degrees of freedom (df), denominator degrees of freedom, F-values, and *P*-values of the linear mixed-effects model comparing the effect of watering treatment and soil histories on the Ψ_{Soil} - Ψ_{Leaf} relationship of each species. Bolding of *P*-values represents a *P* < 0.05

	numDF	denDF	F-value	<i>P</i>
<i>P. virginiana</i>				
[Intercept]	1	87	1040	<0.0001
Ψ_{soil}	1	87	62.3	<0.0001
Watering treatment	1	19	0.281	0.60
Soil history	1	19	0.267	0.61
Ψ_{soil} :Watering treatment	1	87	0.00220	0.96
Ψ_{soil} :Soil history	1	87	0.239	0.51
Watering treatment:Soil history	1	19	0.886	0.36
Ψ_{soil} :Watering treatment:Soil history	1	87	0.650	0.42
<i>Q. rubra</i>				
[Intercept]	1	88	1450	<0.0001
Ψ_{soil}	1	88	19.0	<0.0001
Watering treatment	1	19	0.556	0.46
Soil history	1	19	0.700	0.41
Ψ_{soil} :Watering treatment	1	88	0.453	0.50
Ψ_{soil} :Soil history	1	88	0.466	0.50
Watering treatment:Soil history	1	19	0.0758	0.79
Ψ_{soil} :Watering treatment:Soil history	1	88	0.119	0.73
<i>L. tulipifera</i>				
[Intercept]	1	87	1950	<0.0001
Ψ_{soil}	1	87	4.11	0.046
Watering treatment	1	19	0.0373	0.85
Soil history	1	19	0.00200	0.96
Ψ_{soil} :Watering treatment	1	87	0.879	0.35
Ψ_{soil} :Soil history	1	87	0.994	0.32
Watering treatment:Soil history	1	19	0.162	0.69
Ψ_{soil} :Watering treatment:Soil history	1	87	6.18	0.015

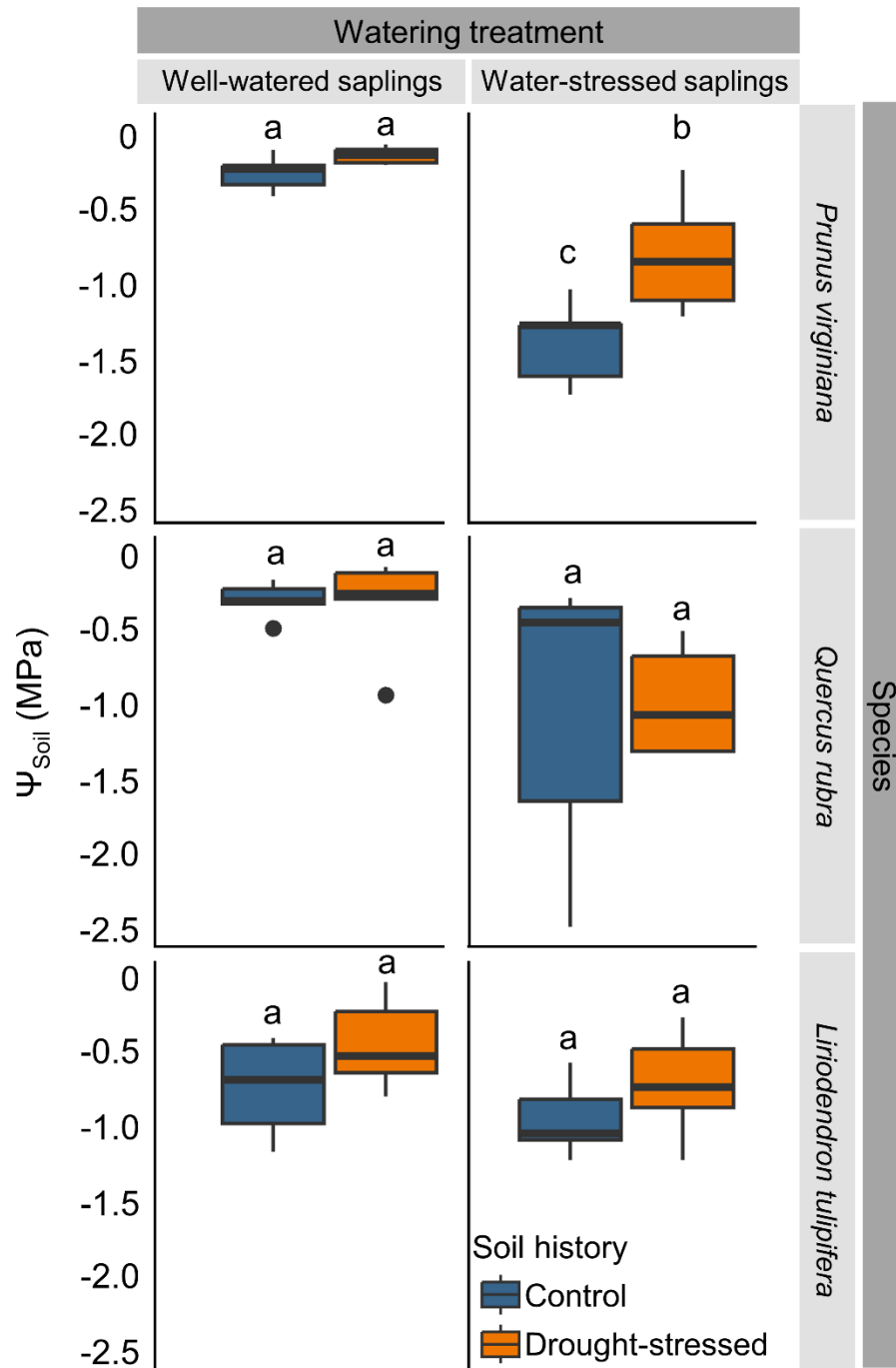


Fig S2 Soil water potential (MPa) for each of the treatment combinations and species. (control soil history – blue, drought-stressed soil history – orange). Letters above the boxplots represent a significant difference among treatment combinations for each species

Table S6 Means $\pm$ standard error of %C, %N, C:N, % sand, % silt, % clay, and bulk density (g cm⁻³) of the soil pre-treatment from the throughfall displacement experiment (source of the drought-stressed soils) and the adjacent control plot (source of the control soils). The resultant *P*-values of a t-test between the throughfall displacement experiment and control plot values are listed for each soil variable

	Throughfall displacement experiment	Control plot	<i>p</i>
Soil % C	1.9 $\pm$ 0.49	1.52 $\pm$ 0.24	0.52
Soil % N	0.12 $\pm$ 0.014	0.14 $\pm$ 0.016	0.28
Soil C:N	27 $\pm$ 16	11 $\pm$ 0.37	0.33
Soil texture			
% <i>sand</i>	2.8 $\pm$ 0.59	4.3 $\pm$ 1.5	0.31
% <i>silt</i>	74 $\pm$ 2.2	71 $\pm$ 4.7	0.48
% <i>clay</i>	23 $\pm$ 2.7	24.7 $\pm$ 4.2	0.69
Soil bulk density (g cm ⁻³)	1.20 $\pm$ 0.22	1.17 $\pm$ 0.18	0.77