



RESEARCH PAPER

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Responses to defoliation of *Robinia pseudoacacia* L. and *Sophora japonica* L. are soil water condition dependent

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Abstract

Key message: Defoliation significantly affected biomass allocation of *Robinia pseudoacacia* L. and *Sophora japonica* L., but leaf physiology readjusted to control levels at the end of the experiment. Considering carbon or sink limitation and relative height growth rate, defoliated *R. pseudoacacia* grew faster than *S. japonica* under well-watered conditions, while defoliated *S. japonica* and *R. pseudoacacia* had similar performance under drought conditions.

Context: Climate change may result in increases of both drought intensity and insect survival, thereby affecting both exotic and native trees in warm temperate forests.

Aims: In this study, we examined the interaction effects of defoliation and drought on an exotic species *Robinia pseudoacacia* and a native species *Sophora japonica* in a warm temperate area, to provide a theoretical basis for predicting the distribution and dynamics of the two species under future climate change.

Methods: In a greenhouse, both species were exposed to three soil moisture (75%, 55%, and 35% of field capacity) and three defoliation treatments (no defoliation, 50% defoliation, and 100% defoliation). Leaf physiology, biomass, and non-structural carbohydrate were determined.

Results: Leaf physiology of defoliated trees did not differ from controls trees, but defoliated seedlings allocated relatively more resources to the leaves at the end of the experiment. In well-watered conditions, defoliated *R. pseudoacacia* was not carbon or sink limited and defoliated *S. japonica* was carbon limited, while defoliated individuals of the two species were sink limited under drought. Defoliated *R. pseudoacacia* grow more rapidly than *S. japonica* in well-watered conditions. Defoliated *R. pseudoacacia* had a similar growth rate to *S. japonica* in drought.

Conclusions: Defoliation clearly affects biomass allocation of the two species, but not leaf physiology. Considering the carbon or sink limitation, the growth of *S. japonica* and *R. pseudoacacia* may be limited by future global climate change scenarios.

Keywords: Carbon limitation, Non-structural carbohydrates, Photosynthetic rate, Relative growth rate in the height, Sink limitation, Watering treatment

Handling Editor: Leonie Schönbeck

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1 Introduction

Climate change models predict that summer in East Asia may become warmer with lower frequency and intensity of precipitation (Dai, 2011; Yu et al. 2014). These changes may cause higher rates of evapotranspiration and lower soil moisture content. The annual average rainfall in northern China has shown a significant downward trend during the period 1951–2010 (Yu et al. 2014). Moreover, the increased annual average temperature is likely to increase insect damage to plants (Netherer and Schopf 2010). Increased temperature enhances the reproductive potential of insects and reduces the time of hibernation (Dale and Frank 2017). As a result, drought and insect defoliation caused by increased temperature are likely to occur simultaneously in northern China under global climate change. Simultaneous drought and insect defoliation affect exotic and native species in the warm temperate forests of northern China (Lombardero et al. 2012). A better understanding of the combined effect of drought and defoliation on exotic and native tree species will be beneficial to understanding the distribution and dynamics of alien species in warm temperate forests under future global climate change.

In trees, photosynthates can be stored as non-structural carbohydrates (NSC; soluble sugar and starch) that act as a buffer to reconcile the temporal asynchrony between carbon supply and demand (Gricar et al. 2019). NSC fuels respiratory metabolism and supports the buildup of structural biomass. NSC are often used to help distinguish between carbon and sink limitation under stresses or disturbances (Wiley et al. 2017). In general, if growth is carbon limited, NSC will be used for metabolism and maintaining growth, so that eventually NSC will decrease (Barry et al. 2011). For example, when plants are subjected to defoliation NSC will significantly decrease, indicating that the growth of the plant is experiencing carbon limitation (Wiley et al. 2017). If the stored NSC increase or are unchanged but growth is limited under stresses (e.g., drought), plants are considered sink limited (Millard et al. 2007, Piper 2020). In this case, plants are carbon sufficient, and growth would be limited by stress factors. However, long-term drought can result in decreased NSC, since plants need to use the stored carbon to survive and maintain their metabolic processes (Bréda et al. 2006, Galiano et al. 2011). Growth will be carbon-limited in such conditions. Distinguishing carbon and sink limitation is important, because some ecological factors that increase carbon availability will only enhance tree growth under carbon-limited conditions (Wiley et al. 2017).

Defoliation always affects the physiology and growth of plants (Barry et al. 2011; Quentin et al. 2012). After defoliation, plants maintain metabolism by consuming stored NSC (Jacquet et al. 2014), resulting in carbon limitation. Then, plants may increase the net photosynthetic rate of

the remaining leaves (Quentin et al. 2012) and decrease the net photosynthetic rate of the bark (Eyles et al. 2009b), thereby increasing carbon absorption to reallocate stored NSC to maintain metabolism and product new organs (Weber et al. 2018). Therefore, after a regrowth period, the growth of defoliated plants may no longer be carbon limited. By this time, defoliated plants with an increased NSC level could eventually increase total biomass, reaching or even surpassing the biomass levels of non-defoliated plants (Eyles et al. 2009a; Barry et al. 2011). Moreover, the degree of growth after defoliation depends on the defoliation intensity, species identity, and environmental context (Chen et al. 2017, Jacquet et al. 2014, Quentin et al. 2012).

There are complex interactions between drought and defoliation treatments (Itter et al. 2018, Teskey et al. 2015). The three primary scenarios that follow have been proposed. (i) Defoliation could exacerbate the adverse effects of drought. Defoliated trees may deplete NSC reserves (Maguire and Kobe 2015) and decrease the carbohydrate transport from leaves to roots, thereby limiting root growth capacity and soil water absorption under drought conditions (Teskey et al. 2015). (ii) Defoliation may mitigate the negative impact of drought on plants. Due to the reduction of foliar area, defoliation may reduce transpiration and thereby mitigate the negative impact of drought (Puri et al. 2015, Itter et al. 2018). (iii) Defoliation and drought may have no interactive effects on *Populus tremuloides* Michx., which may be relevant to the size and age of a tree (Chen et al. 2018). In addition, the interaction of drought and defoliation on a tree also depends on the intensity of drought or defoliation (Jactel et al. 2012). Hence, the effects of drought on the growth of defoliated trees remain debatable.

Robinia pseudoacacia was introduced to China from North America and has become one of the dominant trees in warm temperate forests of China (Wang and Zhou 2000). Since the 1980s in northern China, approximately 8000 ha of *R. pseudoacacia* has been planted for reforestation due to its capacity for rapid growth (Wang et al. 2020, Zhang and Xing 2009). *Sophora japonica* is a widespread native tree in northern China with significant ecological (Kwon et al. 2016) and medicinal value (Zhang et al. 2014). *S. japonica* belongs to the same subfamily (Papilionoideae) as *R. pseudoacacia*. With juicy and palatable leaves, *R. pseudoacacia* and *S. japonica* are frequently defoliated by insects (Kwon et al. 2016, Liu et al. 2015). Therefore, the exotic *R. pseudoacacia* and native *S. japonica* will be exposed to simultaneous insect defoliation damage and drought in the context of climate change. We conducted a greenhouse experiment with three defoliation and three soil moisture treatments to (1) determine whether the growth of *R. pseudoacacia* and *S. japonica* is carbon or sink limited under different

treatments; (2) reveal the responses of the two species to defoliation; and (3) reveal the responses of defoliated trees to different soil moisture conditions.

2 Materials and Methods

2.1 Study site and plant materials

This study was conducted at the Fanggan Research Station of Shandong University (36° 26' N, 117° 27' E) in the Central Mountain Region of Shandong Province, China. The site has a warm temperate monsoon climate with an average temperature of 13 ± 1 °C and annual precipitation of 700 ± 100 mm (Zhang et al. 2006). The experiment was performed in a greenhouse with a steel pipe frame covered by a plastic film to avoid natural precipitation. The mean air temperature of the greenhouse during the experiment was 29.6 °C in the daytime and 20.8 °C at night. The greenhouse was kept well ventilated by rolling up the plastic film at the sides during the experiment.

Seeds of *S. japonica* and *R. pseudoacacia* were collected in the garden of Dacheng Seed Company (Jinan, China) in autumn of 2014. The seeds were stored at 4 °C in a refrigerator. On April 29, 2015, after soaking in distilled water for 24 h, the seeds were transferred to a growth chamber for germination. After 10 days, the seeds started to germinate. Then, we chose healthy and uniformly germinated seedlings to be sown in plastic pots (25 × 21 cm, height × diameter). Each pot was filled with 4.9 kg of air-dried loam and 1.6 kg of sand that remove debris and stones beforehand. The same substrate was used by the growth chamber for germination. Weeds and insects were manually controlled.

2.2 Experimental design

Seedlings were subjected to the three soil moisture treatments: 75% (control, W1), 55% (moderate drought, W2), and 35% (severe drought, W3) of field capacity. In each soil moisture treatment, plants were subjected to either control (D0, no defoliation), 50% defoliation treatment (D1), or 100% defoliation treatment (D2).

Throughout the experiment, the pots received compensatory irrigation by weighing daily at 18:00 to maintain a constant soil moisture level. The weights of pots together with soil of W1, W2, and W3 treatments were supplemented by water to 7.6 kg, 7.3 kg, and 7 kg, respectively. The corresponding soil water contents are: W1 = 16.9%, W2 = 12.4%, W3 = 7.9% according to the formula as follows:

$$\text{Soil water content} = (W_t - W_0) / W_0$$

W_t : the weight of pots together with soil before or after watering during the experiment, W_0 : the weight of pots together with soil before the experiment began.

The weights of pots before watering were recorded at the eighth day, the twenty-second day, the thirty-sixth day, and the fiftieth day after defoliation. The soil water content before watering in different watering treatments was listed in Fig. 1 (using the above formula).

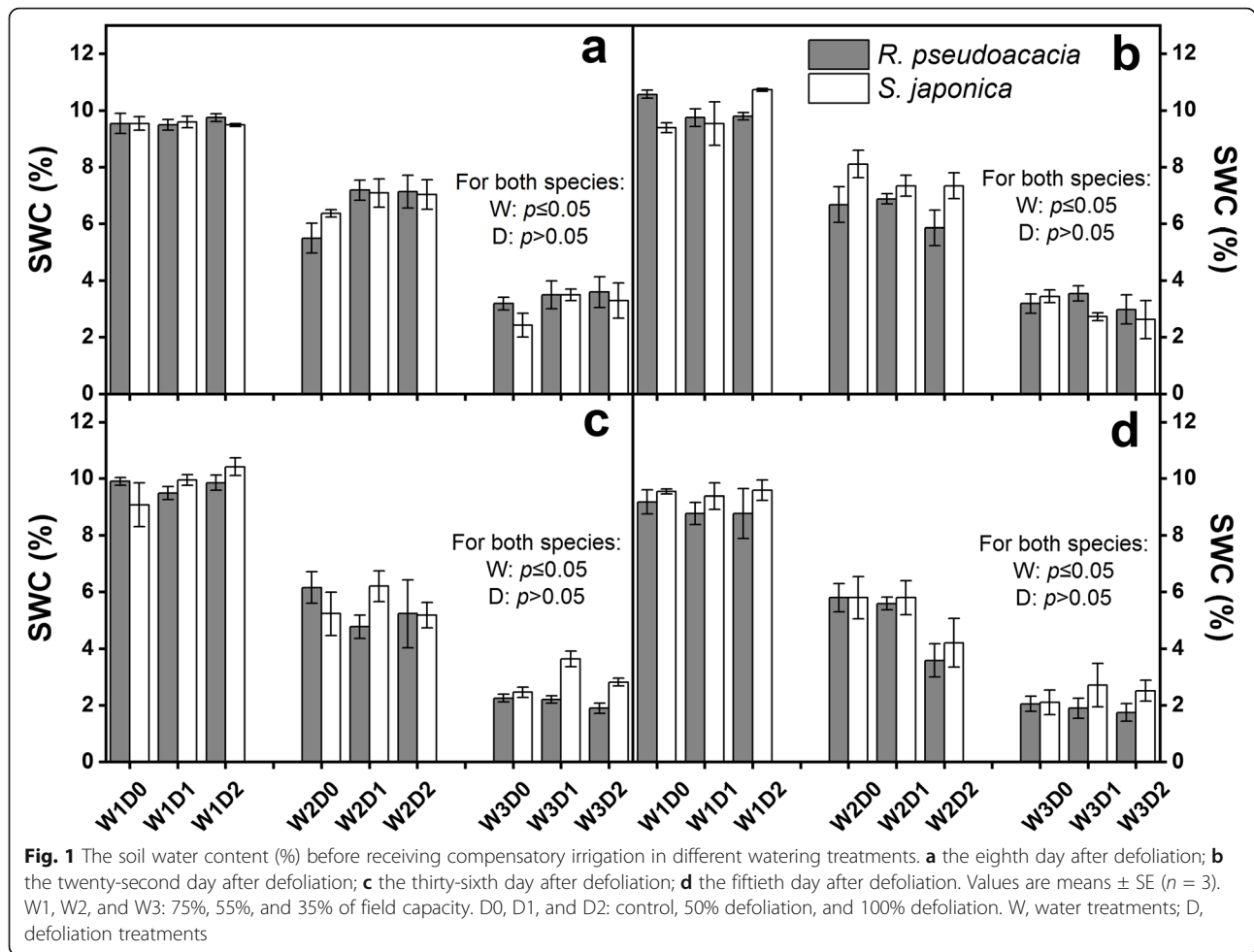
Leaves (excluding apical buds) were removed using scissors from the crown apex downwards in defoliation treatments. Total leaf area removed from individuals was determined in 100% defoliation treatment. For each seedling subjected to 50% defoliation treatment, the shoot was first separated equally into upper and lower parts. The leaves of the upper part were removed and scanned to measure the leaf area. After calculation, the removed leaf area ratio in 50% defoliation treatment was approximately 48% of the total leaf area for each seedling.

Five replicates were included for *R. pseudoacacia* and *S. japonica* at each treatment (nine combinations of treatments). As such, there were 90 pots of *R. pseudoacacia* and *S. japonica* in total. Each pot had one seedling. Approximately 20cm height of seedlings of both species were subjected to the specified experimental treatments for approximately 8 weeks, from July 12, 2015, to September 6, 2015.

2.3 Leaf traits

Gas exchange parameters were measured using a GFS-3000 Portable Photosynthesis System (Walz, Effeltrich, Germany). An LED light source mounted on the GFS-3000 Portable Photosynthesis System was set at a photosynthetic photon flux density of $1200 \mu\text{mol m}^{-2} \text{s}^{-1}$. The CO₂ concentration in the leaf chamber was set as 400 ppm using a CO₂ cylinder, and the temperature in the leaf chamber was set at 28 °C to provide a relatively stable environment. Maximum net photosynthetic rate (A_{max}), stomatal conductance (G_s), and transpiration rate (E) were logged when the system showed stable readings. Gas exchange parameters were measured between 8:30 and 12:00 on sunny days from prior to defoliation, 2, 5, and 8 weeks after defoliation (from July 8–11, July 20–26, August 16–23, and September 1–6, respectively). The three youngest fully developed leaves of three seedlings from each treatment were sampled for measurement.

Chlorophyll a and chlorophyll b concentrations were determined using a 722S visible light spectrophotometer (Leng Guang, Inc., Shanghai, China) after the leaves were extracted in 95% ethanol at end of experiment according to the method of Lichtenthaler and Wellburn (1983). Four replicates of both species in each treatment were sampled, and the concentrations were calculated on a fresh weight basis. The chlorophyll a and b concentrations (chl a+b) and the ratio of chlorophyll a and b (chl a/b) were also calculated.



2.4 Growth and biomass partitioning

At the end of the experiment, the seedlings were harvested and divided into root, stem, and leaf. Every part of five replicates for both species in each treatment was weighed after oven-drying at 80 °C for 48h. Total biomass (TB) and biomass allocation were calculated as follows:

$$\begin{aligned}
 TB &= RB + SB + LB; \\
 \text{Root mass ratio (RMR)} &= RB/TB; \\
 \text{Stem mass ratio (SMR)} &= SB/TB; \\
 \text{Leaf mass ratio (LMR)} &= LB/TB; \\
 \text{Root to shoot mass ratio (R/S)} &= RB/(SB + LB)
 \end{aligned}$$

where RB is root biomass, SB is stem biomass, and LB is leaf biomass.

2.5 Non-structural carbohydrates

Five replicates for each species in each treatment were used to measure carbohydrate concentration. Leaf, stem,

and root of the seedlings were dried at 80 °C for 48 h, weighed, and ground to powder using a grinding miller. Following the method of Palacio et al. (2007), 50 mg of dried tissue was prepared for extracting the soluble sugar (SS) in 10 ml 80% (v/v) ethanol at 100 °C in a water bath. After centrifugation at 4000 rpm for 10min, the supernatant was separated for SS measurement. The remainder of the sample was digested to glucose with 9.2 mol L⁻¹ perchloric acid to determine the starch (St) concentration. The concentrations of SS and St were determined using a phenol–sulfuric acid colorimetric assay (DuBois et al. 1956) as modified by Buysse and Merckx (1993). NSC concentration was the sum of SS and St.

2.6 The relative growth rate in the height

We measured the plant heights at both the start and end of each treatment. The relative growth rate in the height (RGR_H, cm day⁻¹) was calculated as follows (Li et al. 2019):

$$\text{RGR}_H = (\ln H_2 - \ln H_1) / t$$

where H_2 and H_1 indicate the seedling height at the end and the beginning of drought \times defoliation treatments, and t indicates the time duration (56 days).

2.7 Statistical analysis

Three-way analysis of variance (ANOVA) was applied to test the effects of species identity, soil moisture, and defoliation treatments for every performance parameter of *R. pseudoacacia* and *S. japonica*. One-way ANOVA and Duncan's multiple range tests were performed to detect significant differences between/among treatments within each species. Repeated measures ANOVA was used to examine the effects of drought, defoliation, and time on gas exchange parameters. Before applying ANOVAs, data were tested for variance normality and homogeneity and were transformed to increase normality and homogeneity of variances if necessary. To test species differences specifically, independent sample t -tests were performed between every parameter of *R. pseudoacacia* and *S. japonica* under each water regime \times defoliation treatment separately. Dataset used for analysis can be found in the figshare repository (Li et al. 2021). All of the statistical analyses were conducted using IBM SPSS Statistics 22.0 (SPSS Inc., Chicago, IL, USA). Figures were drawn using Origin 9.0 software (OriginLab Co., MA, USA).

3 Results

3.1 Leaf traits

Defoliation had a significant effect on most gas exchange parameters after 2 weeks (Table 1). Defoliation increased A_{\max} of *R. pseudoacacia* in W2 and A_{\max} of *S. japonica* in both W1 and W2 (Fig. 2b, d and e). Moreover, A_{\max} of both species began to decline 2 weeks after defoliation, and A_{\max} of both species recovered to the control levels 5 weeks after defoliation. Drought had a significant effect on A_{\max} at 5 weeks after defoliation (Table 1). Two weeks after defoliation, defoliation increased E of *S. japonica* and G_s of both species in both W1 and W2 (Appendix Fig. 8 and 9). Five weeks after defoliation, most E and G_s both species recovered to the control levels.

Eight weeks after defoliation, drought continued to have a clear effect on A_{\max} , E and G_s of the two species (Table 1). Most gas exchange parameters in W1 were clearly higher than those in W3 in any defoliation treatment (Fig. 3a, b, and c). Moreover, A_{\max} of *R. pseudoacacia* was higher than that of *S. japonica* in all soil moisture and defoliation, except W1D0 (Table 1 and Fig. 3a). Species identity, soil moisture, and defoliation had significant effects on chl a/b, and species identity had a significant effect on chl a+b (Table 1). Chl a/b of

R. pseudoacacia in W1D1 and W1D2 were higher than those of *S. japonica* (Fig. 3d and Appendix Fig. 10).

3.2 Non-structural carbohydrate concentration

Leaf NSC concentrations of both species were unaffected by defoliation or drought. Leaf NSC concentration of *R. pseudoacacia* was clearly higher than that of *S. japonica* in all drought and defoliation treatments, except W1D1 and W1D2 (Table 1 and Fig. 4a). Stem NSC concentration was affected by species identity, drought, defoliation, drought \times defoliation (the interaction between drought and defoliation), and drought \times defoliation \times species identity (the interaction among drought, defoliation, and species identity), respectively (Table 1). Under W1, stem NSC concentration of *S. japonica* remarkably decreased with defoliation intensity (Fig. 4b). However, stem NSC concentration of *R. pseudoacacia* was unaffected by defoliation. Root NSC concentration was influenced by drought, defoliation, and species identity \times drought (Table 1). Root NSC concentration of *R. pseudoacacia* was unaffected by defoliation and drought (Fig. 4c). Root NSC concentration of *S. japonica* in W2 and W3 was clearly higher than that in W1 (Fig. 4c).

3.3 Growth and biomass allocation

The species identity, soil moisture, defoliation, and species identity \times soil moisture clearly affected leaf biomass of both species (Table 1). In W1 and W3, LB of *R. pseudoacacia* showed no significant differences among the three defoliation treatments; in W2, LB of *R. pseudoacacia* in D0 was clearly higher than that in D2 (Fig. 5a). LB of *S. japonica* in D0 was higher than that in D2 under W1 and W2 treatments (Fig. 5a). And LB of *R. pseudoacacia* was higher than that of *S. japonica* in W1 and W3, except W1D0.

TB of the two species was significantly affected by species identity, soil moisture, and defoliation (Table 1). In W1, TB of *R. pseudoacacia* showed no significant differences among the three defoliation treatments; in W2 and W3, TB of *R. pseudoacacia* in D0 was clearly higher than that in D2 (Fig. 5b). TB of *S. japonica* in W1 and W2 decreased with defoliation intensity (Fig. 5b). TB of *R. pseudoacacia* was higher than that of *S. japonica* in W1 and W3 (Table 1 and Fig. 5b).

The soil moisture and species identity clearly affected most of the biomass allocation of both species (Table 1). Drought clearly restricted LMR of the two species (Fig. 6a). RMR and R/S of *S. japonica* were lower in W1 than that in W2 and W3, while those of *R. pseudoacacia* was unaffected by drought (Fig. 6 c and d).

3.4 The relative growth rate in the height

RGR_H was affected by drought, defoliation, and species identity (Table 1). In W1, RGR_H of *S. japonica* was lower

Table 1 *F* values of three-way ANOVA of different treatments on parameters of *R. pseudoacacia* and *S. japonica*

	<i>S</i>	<i>W</i>	<i>D</i>	<i>S</i> × <i>W</i>	<i>S</i> × <i>D</i>	<i>W</i> × <i>D</i>	<i>S</i> × <i>W</i> × <i>D</i>
Leaf traits							
0- <i>A</i> _{max} (μmol m ⁻² s ⁻¹)	48.193**	0.268	0.081	1.516	0.323	0.094	0.200
0- <i>E</i> (mmol m ⁻² s ⁻¹)	15.623**	0.609	0.014	0.735	0.058	0.685	0.038
0- <i>G</i> _s (mmol m ⁻² s ⁻¹)	3.833	0.032	0.016	0.009	0.12	0.029	0.017
1- <i>A</i> _{max} (μmol m ⁻² s ⁻¹)	0.261	1.812	8.223**	0.846	6.745**	1.164	2.152
1- <i>E</i> (mmol m ⁻² s ⁻¹)	4.436*	2.630	28.523**	0.733	6.518**	4.076**	0.987
1- <i>G</i> _s (mmol m ⁻² s ⁻¹)	1.284	20.538**	40.589**	0.738	0.814	4.233**	0.630
2- <i>A</i> _{max} (μmol m ⁻² s ⁻¹)	1.387	6.153**	1.353	0.237	4.778*	1.052	2.087
2- <i>E</i> (mmol m ⁻² s ⁻¹)	8.353**	0.274	6.890**	15.738**	0.451	0.917	2.637
2- <i>G</i> _s (mmol m ⁻² s ⁻¹)	0.713	6.288**	6.342**	5.364**	0.910	1.134	3.602*
3- <i>A</i> _{max} (μmol m ⁻² s ⁻¹)	230.537**	18.221**	1.632	1.285	0.550	0.263	0.566
3- <i>E</i> (mmol m ⁻² s ⁻¹)	1.761	8.751**	1.397	0.399	0.813	0.325	0.123
3- <i>G</i> _s (mmol m ⁻² s ⁻¹)	0.008	26.498**	4.661*	0.427	1.001	0.549	1.065
Chl a+b (mg g ⁻¹)	6.362*	0.315	2.052	0.509	0.105	0.583	0.595
Chl a/b	15.359**	4.169*	4.651*	0.221	2.469	1.041	1.843
Non-structural carbohydrate							
Leaf NSC concentration (%)	84.787**	0.020	2.372	2.270	1.524	0.485	2.297
Stem NSC concentration (%)	50.257**	19.514**	8.550**	2.588	0.417	2.817*	2.554*
Root NSC concentration (%)	0.869	16.157**	4.920*	10.372**	0.525	1.413	0.670
Growth and biomass partitioning							
TB (g)	58.186**	8.682**	15.759**	1.200	1.898	0.781	0.459
LB (g)	62.380**	17.618**	11.223**	3.215*	0.552	0.532	0.400
LMR	0.594	10.252**	2.043	0.070	2.071	0.956	0.581
SMR	8.477*	0.282	0.675	4.323*	0.864	0.923	0.937
RMR	4.107*	6.028**	2.300	2.154	1.133	0.973	0.186
R/S	5.533*	5.667**	2.224	2.029	1.351	1.021	0.191
RGR_H							
RGR _H (cm day ⁻¹)	24.726**	18.925**	4.779*	0.414	0.206	1.794	0.120

p* ≤ 0.05, *p* ≤ 0.01*S* species, *W* water regime, *D* defoliation

0, 1, 2, and 3: before defoliation, 2 weeks, 5 weeks, and 8 weeks after defoliation

Except for gas exchange parameters, other parameters were measured at 8 weeks after defoliation

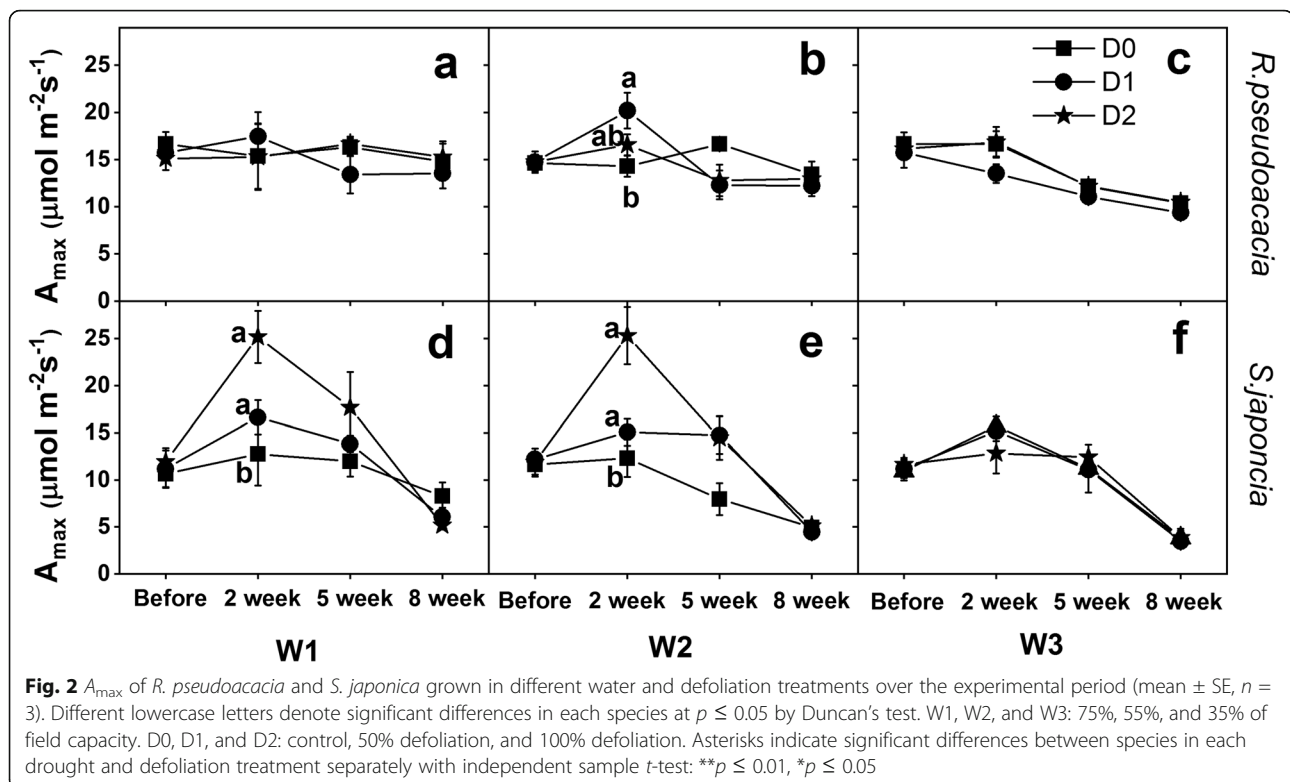
than that of *R. pseudoacacia*. In W2 and W3, RGR_H of *R. pseudoacacia* has no significant difference from that of *S. japonica* (Fig. 7).

4 Discussion

4.1 Carbon or sink limitation after defoliation and drought

Our study showed that leaf, stem, and root NSC concentrations of *R. pseudoacacia* were not affected by defoliation or drought (Fig. 4), indicating that defoliated *R. pseudoacacia* had recovered the carbon concentration of each organ to the control level, consistent with studies on *Nothofagus pumilio* and *Pinus sylvestris* (Piper et al.

2015, Susiluoto et al. 2010). Moreover, TB of defoliated *R. pseudoacacia* was not significantly different from that of undefoliated trees under high soil moisture conditions, suggesting that the growth of defoliated *R. pseudoacacia* in high moisture conditions is not carbon or sink limited. However, stem NSC concentration of defoliated *S. japonica* under high soil moisture conditions was remarkably lower than in control (Fig. 4b). Combined with the lower TB of defoliated *S. japonica* than that of undefoliated one, we inferred that defoliated *S. japonica* did not have enough carbohydrate stored to support growth, which is consistent with the definition of carbon limitation (Wiley et al. 2017). Hence,



defoliated *S. japonica* growth under high soil moisture conditions is carbon limited, while the same is not true for *R. pseudoacacia*.

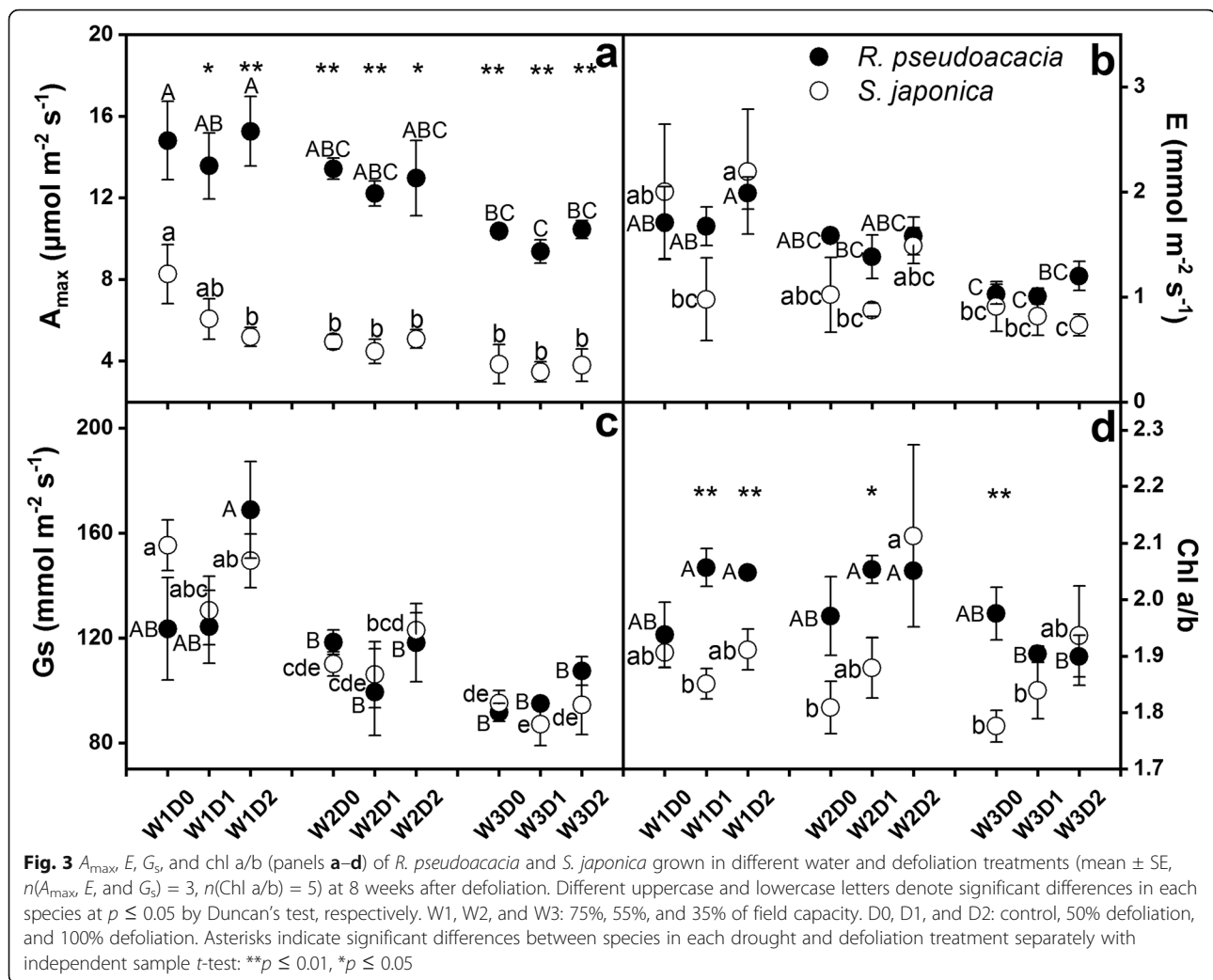
Leaf, stem, and root NSC concentrations of two species in drought had no significant difference with those of seedlings in high soil moisture conditions, indicating that seedlings with or without defoliation in drought had enough stored carbohydrate to support growth (Piper 2020). Moreover, TB of both defoliated species in drought were lower than those of undefoliated trees in the high moisture treatments, meaning that growth of defoliated trees was inhibited by drought and defoliation. In drought, defoliated *R. pseudoacacia* and *S. japonica* had enough stored carbohydrate, but could not recover their full growth, suggesting that drought inhibited the process of plant synthesis of biomass using available carbohydrates. That was in line with the characteristics of sink limitation (Wiley et al. 2017).

4.2 *R. pseudoacacia* and *S. japonica* responses after defoliation

In our study, seedlings of defoliated *R. pseudoacacia* and *S. japonica* had a transient upregulation of A_{\max} at 2 weeks after defoliation (Table 1 and Fig. 2). The results are consistent with studies in *Eucalyptus globulus* and *Pinus sylvestris* (Quentin et al. 2010, Schönbeck et al.

2020). The reason for stronger photosynthesis could be that defoliated seedlings had insufficient stores of NSC in organs to maintain growth (Wang et al. 2020). Pinkard et al. (2011a) showed that lower NSC concentration in plant organs may stimulate A_{\max} producing more carbohydrates. Therefore, seedlings increased G_s (Table 1 and Appendix Fig. 9), which was strongly correlated with the increase in A_{\max} of defoliated *Eucalyptus globulus* (Quentin et al. 2012) to increase the capacity of photosynthesis at leaf level for increased uptake of carbohydrate.

A_{\max} and E of the two species were not significantly different between defoliation treatments at 8 weeks after defoliation (Table 1, Fig. 2, and Appendix Fig. 8), indicating that these parameters of both species had grown to the control levels. Our results are consistent with the study on *Eucalyptus globulus* (Pinkard et al. 2011b). In our study, this result may be related to unchanged leaf NSC concentration under defoliation treatments (Fig. 4a). According to the positive relationship of stored NSC and A_{\max} (Pinkard et al. 2011a), we inferred that unchanged leaf NSC concentration may have no positive feedback on A_{\max} (Iqbal et al. 2012). In addition, we observed that chl a+b was unchanged by defoliation (Table 1 and Appendix Fig. 10). Pinkard et al. (2011b) showed that A_{\max} and chlorophyll content usually had a strong



positive relationship. Hence, the unchanged chlorophyll content may also have contributed to the unchanged A_{max} at 8 weeks after defoliation.

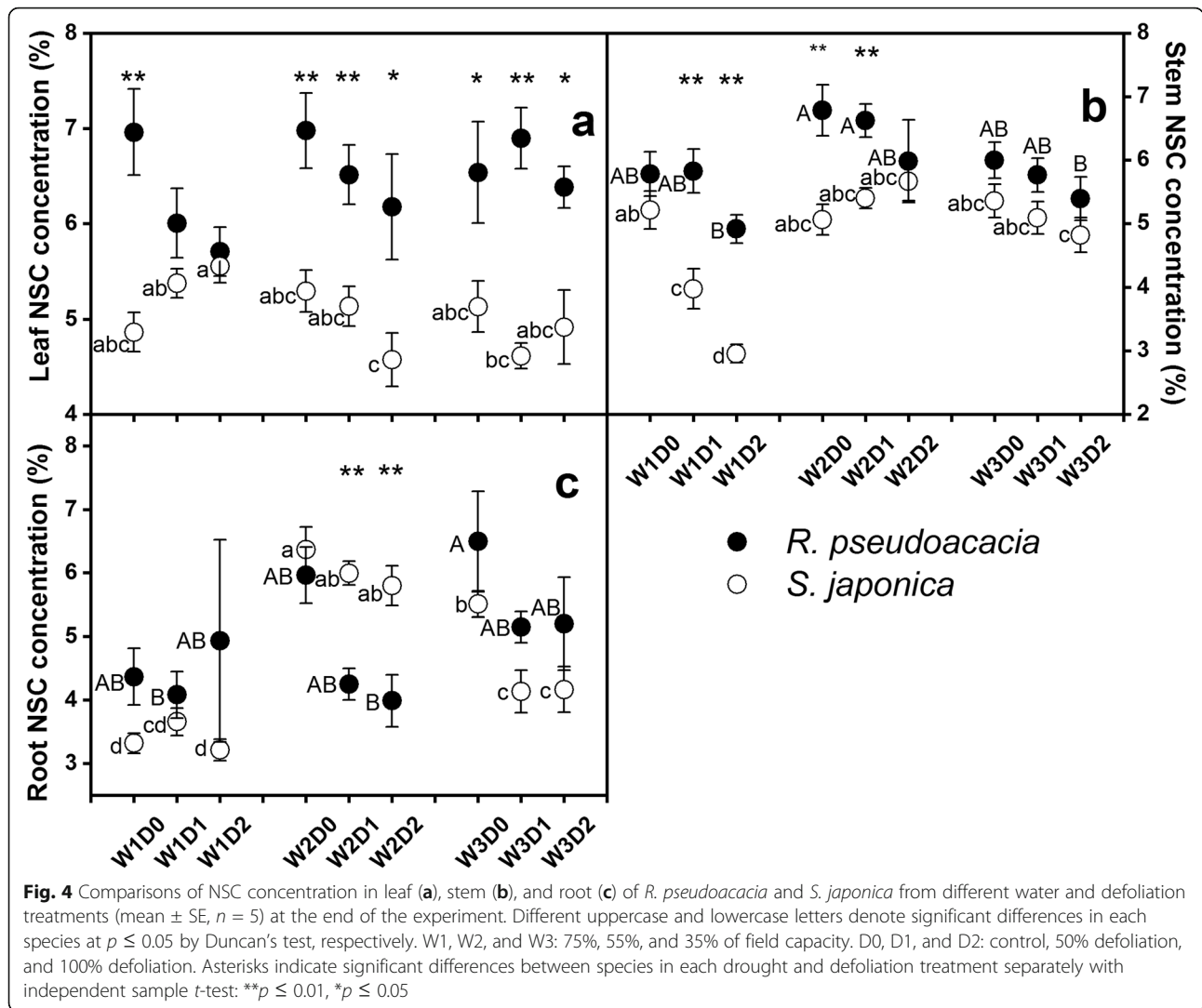
Leaf NSC concentrations of the two species were unchanged by defoliation, while stem and root NSC concentrations of the two species decreased with defoliation intensity, showing NSC concentrations of the two defoliated species did not completely recover. Importantly, this result also suggested that defoliated seedlings invested more NSC to leaf than to stem and root. Some studies have found that producing new organs requires the consumption of carbohydrates from the nearest storage pool (Dietze et al. 2014). Thus, higher leaf NSC concentration may mean more new leaves of seedlings that can improve the carbon absorption of the whole seedling and fix more carbon in order to grow.

With regard to biomass allocation, we found that both species were unaffected by defoliation. Indeed, when calculating LB in the defoliation treatments, we neglected

the removed leaf biomass at the beginning of defoliation. Therefore, LB of defoliation treatments may be lower than that of control. After a regrowth period, LMR of defoliated trees was unchanged, indicating that they may allocate more resources to the leaves. Our finding is consistent with another study showing that defoliation may increase the leaf or aboveground biomass ratio to increase carbon assimilation of the whole tree (Eyles et al. 2009a).

4.3 Defoliated *R. pseudoacacia* and *S. japonica* responses under different soil moisture conditions

There were no significant interaction effects between defoliation and drought treatments on most of the parameters (except for E and G_s at 5 weeks after defoliation and stem NSC concentration at 8 weeks after defoliation) in this study. However, some studies have identified antagonistic or synergistic effects of biotic and abiotic stresses on growth in different woody plants, such as in



Eucalyptus globulus and *Quercus rubra* (Kolb et al. 1999, Quentin et al. 2012). In our study, *R. pseudoacacia* and *S. japonica* belong to the Leguminosae, which may result in both species had similar interaction effects between defoliation and drought conditions. Therefore, we inferred that our results could be attributed to species' identities or family specific.

Stem NSC concentration of *S. japonica* decreased with defoliation under well-watered conditions, while that of *R. pseudoacacia* was unchanged by defoliation. This may have resulted from the higher A_{max} of *R. pseudoacacia* compared to *S. japonica* (Fig. 3a) causing relatively more leaf NSC of *R. pseudoacacia* to be allocated to stems. However, stem NSC concentrations of *R. pseudoacacia* and *S. japonica* were unaffected by defoliation under drought suggesting that NSC is crucial to maintaining hydraulic functioning in trees under water stress (Jacquet et al. 2014).

Defoliated *R. pseudoacacia* and *S. japonica* had different RGR_H under different water conditions: RGR_H of defoliated *R. pseudoacacia* was higher than that of *S. japonica* in high moisture conditions, while *R. pseudoacacia* and *S. japonica* did not differ significantly in RGR_H in drought. The results suggested that the exotic *R. pseudoacacia* was able to take advantage of increased water resources and enhance its growth after defoliation. Caramaschi et al. (2016) also reported that exotic species after defoliation had a greater ability to grow than native species. This is one of the characteristics of exotic species, i.e., that they often exhibit a greater response than native species when resource availability increases (Leishman et al. 2010, Luo et al. 2015). However, compared with native species, most exotic species cannot capitalize and maintain greater performance in a stressful environment (Dawson et al. 2012), which is supported in our study. In addition, A_{max} and chl a/b of the

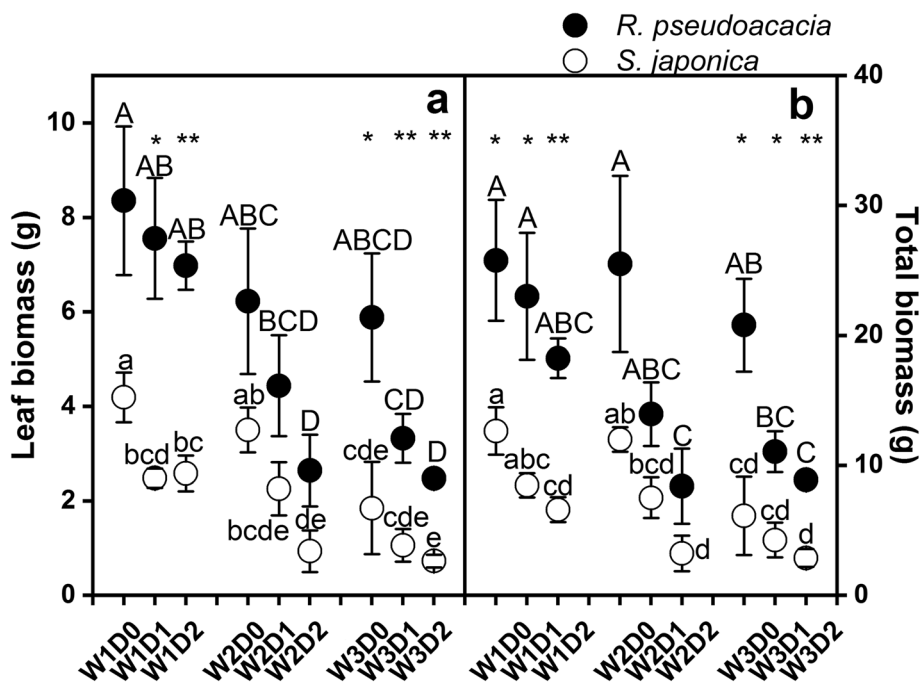


Fig. 5 Comparisons of LB (a) and TB (b) of *R. pseudoacacia* and *S. japonica* grown in different water and defoliation treatments (mean ± SE, n = 5) at 8 weeks after defoliation. Different uppercase and lowercase letters denote significant differences in each species at $p \leq 0.05$ by Duncan's test, respectively. W1, W2, and W3: 75%, 55%, and 35% of field capacity. D0, D1, and D2: control, 50% defoliation, and 100% defoliation. Asterisks indicate significant differences between species in each drought and defoliation treatment separately with independent sample t-test: ** $p \leq 0.01$, * $p \leq 0.05$.

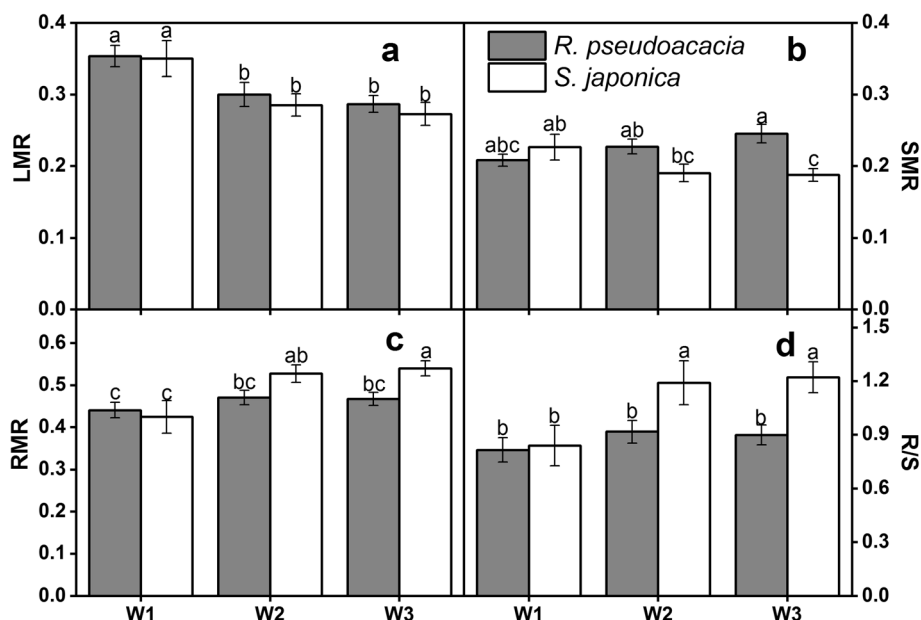


Fig. 6 Comparisons of biomass allocation of *R. pseudoacacia* and *S. japonica* grown in different water regimes (mean±SE, n = 5) at the end of experiment. **a** LMR, **b** SMR, **c** RMR, and **d** R/S. Different letters denote significant differences in each species at $p \leq 0.05$ by Duncan's test. W1, W2, and W3: 75%, 55%, and 35% of field capacity

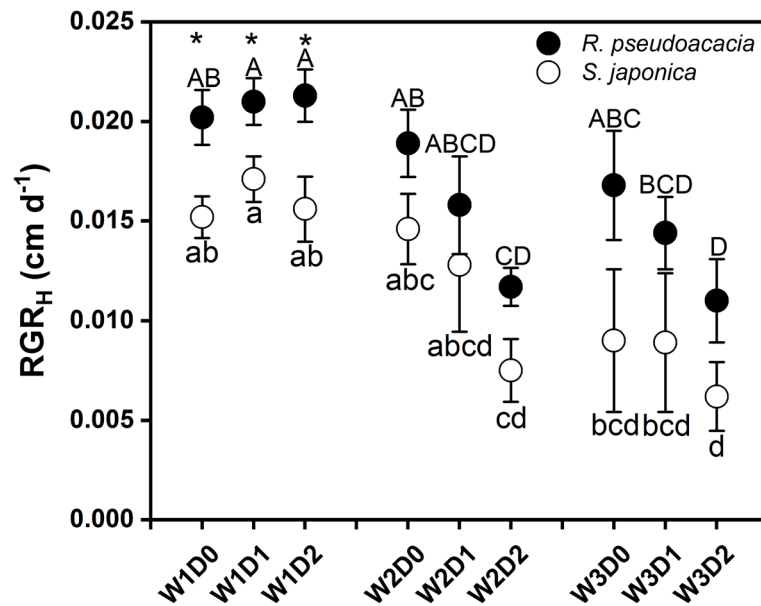


Fig. 7 RGR_H of *R. pseudoacacia* and *S. japonica* grown in different water and defoliation treatments (mean ± SE, $n = 5$). Different uppercase and lowercase letters denote significant differences in each species at $p \leq 0.05$ by Duncan's test, respectively. W1, W2, and W3: 75%, 55%, and 35% of field capacity. D0, D1, and D2: control, 50% defoliation, and 100% defoliation. Asterisks indicate significant differences between treatment within each species in each drought and defoliation treatment separately with independent sample t -test: ** $p \leq 0.01$, * $p \leq 0.05$

exotic *R. pseudoacacia* was higher than that of the native *S. japonica* at 8 weeks after defoliation under well-watered conditions (Table 1 and Fig. 3 a and d), which may be a key reason for stronger RGR_H of *R. pseudoacacia*.

In addition, we found that leaf NSC concentration of *R. pseudoacacia* had no significant difference with that of *S. japonica* in high soil moisture conditions. Combined with the high A_{\max} of *R. pseudoacacia* in well-watered conditions, these results indicated that *R. pseudoacacia* used more carbon to supply the growth of new leaves (Dietze et al. 2014). Meanwhile, more new leaf production of *R. pseudoacacia* can improve carbon fixation of whole seedlings, and this may be another reason for the stronger RGR_H of *R. pseudoacacia* under well-watered conditions.

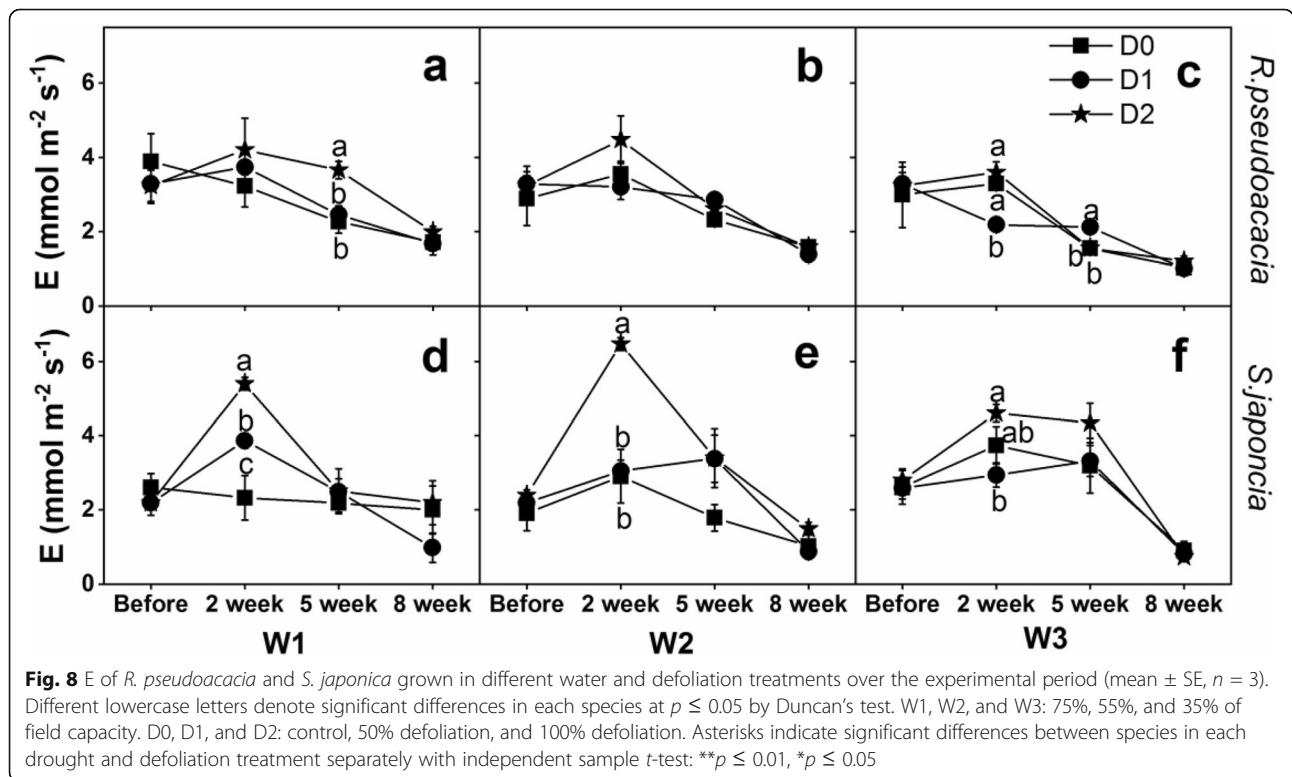
Interestingly, RGR_H of *S. japonica* and *R. pseudoacacia* did not have a significant difference in drought (Fig. 7). *S. japonica* in drought conditions had the higher R/S, RMR, and root NSC concentration than that in the well-watered conditions, suggesting that *S. japonica* allocates more resources to roots in drought conditions. This could improve root osmotic pressure (higher root NSC concentration means higher solute concentration), taking up more water and alleviating the negative effects of drought (Guo et al. 2019, Jacquet et al. 2014). Comparing the unchanged R/S, RMR, and root NSC concentration of *R. pseudoacacia* in drought, we inferred that *R.*

pseudoacacia had weaker drought tolerance than *S. japonica*. Although *R. pseudoacacia* had higher carbon absorption capacity at leaf level (A_{\max}) and leaf biomass than those of *S. japonica* under drought conditions (Fig. 3a), weak drought tolerance of *R. pseudoacacia* limited its ability to grow after defoliation. This may be the main reason why there is no significant difference in the RGR_H of the two species under drought treatments.

5 Conclusion

Our study showed that A_{\max} was upregulated at 2 weeks after defoliation, indicating that both species had a transient increase in carbon fixation at the leaf level. At the end of the experiment, the leaf physiology of both defoliated species had recovered to the levels of undefoliated trees. Defoliated seedlings allocated more resources to the leaves to increase carbohydrate production. In well-watered conditions, *R. pseudoacacia* achieved clearly greater RGR_H than *S. japonica*, while *R. pseudoacacia* had similar RGR_H to *S. japonica* in drought. In well-watered conditions, defoliated *R. pseudoacacia* did not show carbon or sink limitation. However, both species were sink limited in drought. In conclusion, the growth of both *R. pseudoacacia* and *S. japonica* may be limited under future drought and defoliation. Nevertheless, to fully reveal the different responses between sympatric *R. pseudoacacia* and *S. japonica*, long-term experiments and monitoring data are needed.

6 Appendix



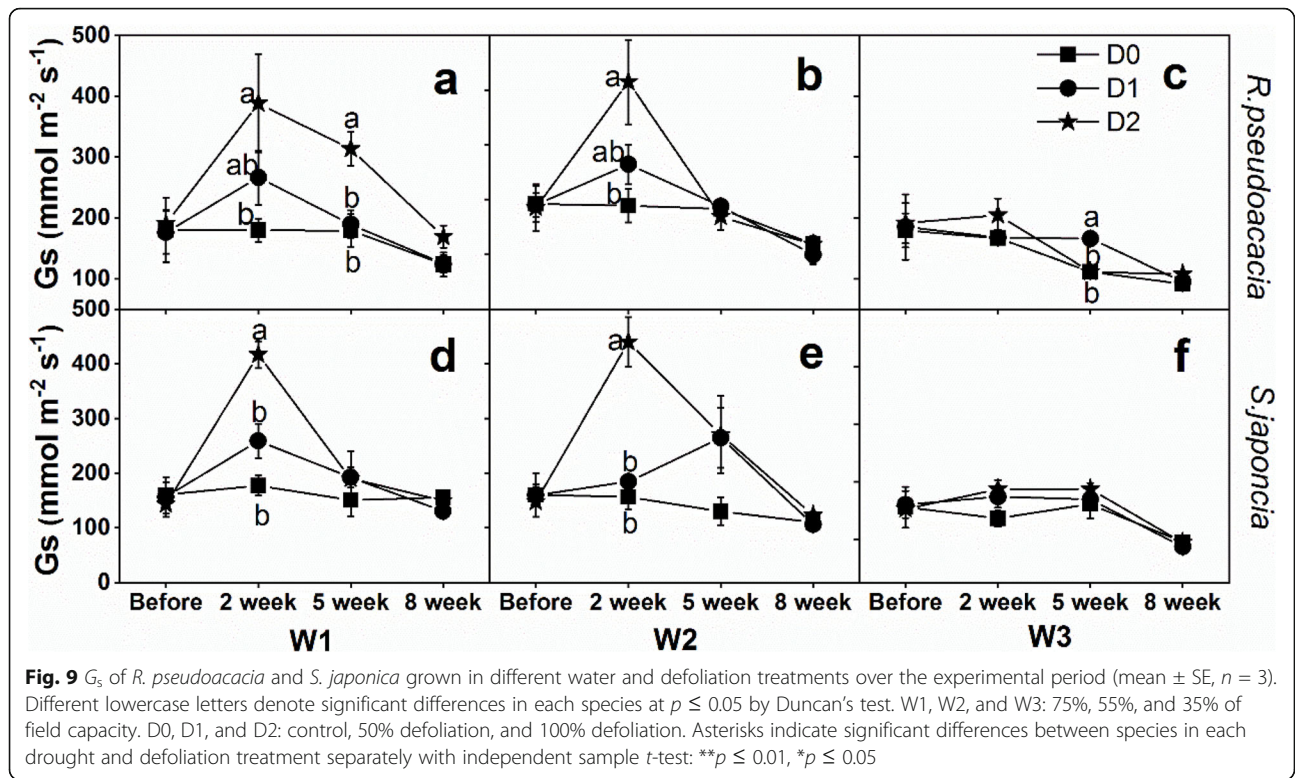


Fig. 9 G_s of *R. pseudoacacia* and *S. japonica* grown in different water and defoliation treatments over the experimental period (mean \pm SE, $n = 3$). Different lowercase letters denote significant differences in each species at $p \leq 0.05$ by Duncan's test. W1, W2, and W3: 75%, 55%, and 35% of field capacity. D0, D1, and D2: control, 50% defoliation, and 100% defoliation. Asterisks indicate significant differences between species in each drought and defoliation treatment separately with independent sample t -test: ** $p \leq 0.01$, * $p \leq 0.05$

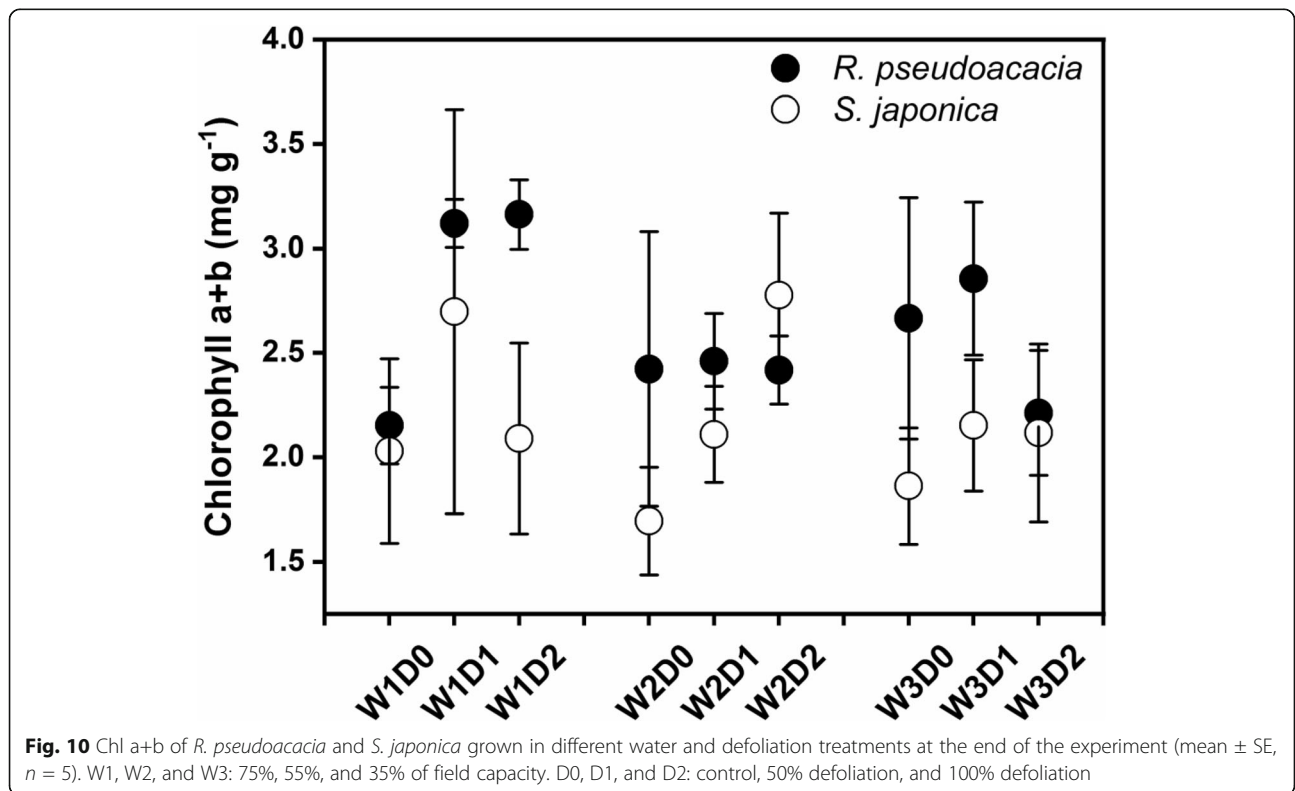


Fig. 10 Chl a+b of *R. pseudoacacia* and *S. japonica* grown in different water and defoliation treatments at the end of the experiment (mean \pm SE, $n = 5$). W1, W2, and W3: 75%, 55%, and 35% of field capacity. D0, D1, and D2: control, 50% defoliation, and 100% defoliation

Acknowledgements

We express our gratitude to Ning Wang, Zhangnan Guan, and Song Zhao for their help during the experiment and letpub Company for their valuable writing suggestions.

Code availability

Not applicable.

Authors' contributions

Conceptualization: Mingyan Li, Xiao Guo, and Ning Du; Methodology: Mingyan Li; Formal analysis and investigation: Mingyan Li and Xiao Guo; Writing—original draft preparation: Mingyan Li and Xiao Guo; Writing—review and editing: Mingyan Li, Xiao Guo, Lele Liu, Jian Liu, Ning Du, and Weihua Guo; Funding acquisition: Mingyan Li, Xiao Guo, Ning Du, and Weihua Guo. The authors read and approved the final manuscript.

Funding

This study was financially supported by the National Natural Science Foundation of China (No. 31970347), the Natural Science Foundation of Shandong Province (No. ZR2020MC035), the Forestry Science and Technology Innovation Program of Shandong Province (2019LY010), the Seed funding for International Scientific Research Cooperation of Shandong University (No. SD202015), and the Advanced Talents Foundation of Qingdao Agricultural University (No. 6631115021, 6631120094).

Availability of data and materials

The datasets generated during and/or analyzed during the current study are available in the figshare repository, <https://doi.org/10.6084/m9.figshare.17004637.v3>.

Declarations

Ethics approval and consent to participate

Not applicable.

Consent for publication

All authors gave their informed consent to this publication and its content.

Competing interests

The authors declare no conflict of interest. The funders had no role in the design of the study; in the collection, analyses, or interpretation of data; in the writing of the manuscript; or in the decision to publish the results.

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Received: 7 July 2021 Accepted: 3 March 2022

Published online: 07 April 2022

References

- Barry KM, Quentin A, Eyles A, Pinkard EA (2011) Consequences of resource limitation for recovery from repeated defoliation in *Eucalyptus globulus* Labillardière. *Tree Physiol* 32(1):24–35. <https://doi.org/10.1093/treephys/tp128>
- Bréda N, Huc R, Granier A (2006) Temperate forest trees and stands under severe drought: a review of ecophysiological responses, adaptation processes and long-term consequences. *Ann Forest Sci* 63(6):625–644. <https://doi.org/10.1051/forest:2006042>
- Buysse J, Merckx R (1993) An improved colorimetric method to quantify sugar content of plant tissue. *J Exp Bot* 44(10):1627–1629. <https://doi.org/10.1093/jxb/44.10.1627>
- Caramaschi GMCL, Barbosa ERM, da Silva DA, Braga VB, Borghetti F (2016) The superior re-sprouting performance of exotic grass species under different environmental conditions: the study case of *Paspalum atratum* (Swallen) and *Urochloa brizantha* (Hochst. ex A. Rich. - Stapf). *Theor Exp Plant Physiol* 28(3): 273–285. <https://doi.org/10.1007/s40626-016-0058-6>
- Chen L, Huang JG, Dawson A, Zhai L, Stadt KJ, Comeau PG, Whitehouse C (2018) Contributions of insects and droughts to growth decline of trembling aspen mixed boreal forest of western Canada. *Glob Change Biol* 24(2):655–667. <https://doi.org/10.1111/gcb.13855>
- Chen Z, Wang L, Dai Y, Wan X, Liu S (2017) Phenology-dependent variation in the non-structural carbohydrates of broadleaf evergreen species plays an important role in determining tolerance to defoliation (or herbivory). *Sci Rep* 7(1):10125. <https://doi.org/10.1038/s41598-017-09757-2>
- Dai AG (2011) Drought under global warming: a review. *WiRes Clim Change* 2(1): 45–65. <https://doi.org/10.1002/wcc.81>
- Dale AG, Frank SD (2017) Warming and drought combine to increase pest insect fitness on urban trees. *PLoS One* 12(3):e0173844. <https://doi.org/10.1371/journal.pone.0173844>
- Dawson W, Rohr RP, van Kleunen M, Fischer M (2012) Alien plant species with a wider global distribution are better able to capitalize on increased resource availability. *New Phytol* 194(3):859–867. <https://doi.org/10.1111/j.1469-8137.2012.04104.x>
- Dietze MC, Sala A, Carbone MS, Czimczik CI, Mantooth JA, Richardson AD, Vargas R (2014) Nonstructural carbon in woody plants. In: Merchant SS (ed) Annual review of plant biology. Annual Reviews, Palo Alto, pp 667–687, 65, 1, DOI: <https://doi.org/10.1146/annurev-arplant-050213-040054>.
- DuBois M, Gilles KA, Hamilton JK, Rebers PA, Smith F (1956) Colorimetric method for determination of sugars and related substances. *Anal Chem* 28(3):350–356. <https://doi.org/10.1021/ac60111a017>
- Eyles A, Pinkard EA, Mohammed C (2009a) Shifts in biomass and resource allocation patterns following defoliation in *Eucalyptus globulus* growing with varying water and nutrients. *Tree Physiol* 29(6):753–764. <https://doi.org/10.1093/treephys/tp1014>
- Eyles A, Pinkard EA, O'Grady AP, Worledge D, Warren CR (2009b) Role of cortical photosynthesis following defoliation in *Eucalyptus globulus*. *Plant Cell Environ*. 32(8):1004–1014. <https://doi.org/10.1111/j.1365-3040.2009.01984.x>
- Galiano L, Martínez-Vilalta J, Lloret F (2011) Carbon reserves and canopy defoliation determine the recovery of Scots pine 4 yr after a drought episode. *New Phytol* 190(3):750–759. <https://doi.org/10.1111/j.1469-8137.2010.03628.x>
- Gricar J, Zavadlav S, Jyske T, Lavric M, Laakso T, Hafner P, Eler K, Vodnik D (2019) Effect of soil water availability on intra-annual xylem and phloem formation and non-structural carbohydrate pools in stem of *Quercus pubescens*. *Tree Physiol* 39(2):222–233. <https://doi.org/10.1093/treephys/tpy101>
- Guo X, Luo YJ, Xu ZW, Li MY, Guo WH (2019) Response strategies of *Acer davidii* to varying light regimes under different water conditions. *Flora* 257:9 <https://doi.org/10.1016/j.flora.2019.151423>
- Iqbal N, Masood A, Khan NA (2012) Analyzing the significance of defoliation in growth, photosynthetic compensation and source-sink relations. *Photosynthetica* 50(2):161–170. <https://doi.org/10.1007/s11099-012-0029-3>
- Itter MS, D'Orangeville L, Dawson A, Kneeshaw D, Duchesne L, Finley AO (2018) Boreal tree growth exhibits decadal-scale ecological memory to drought and insect defoliation, but no negative response to their interaction. *J Ecol* 107(3):1288–1301. <https://doi.org/10.1111/1365-2745.13087>
- Jacquet JS, Bosc A, O'Grady A, Jactel H (2014) Combined effects of defoliation and water stress on pine growth and non-structural carbohydrates. *Tree Physiol* 34(4):367–376. <https://doi.org/10.1093/treephys/tpu018>
- Jactel H, Petit J, Desprez-Loustau ML, Delzon S, Piou D, Battisti A, Koricheva J (2012) Drought effects on damage by forest insects and pathogens: a meta-analysis. *Global Change Biol* 18(1):267–276. <https://doi.org/10.1111/j.1365-2486.2011.02512.x>
- Kolb TE, Dodds KA, Clancy KM (1999) Effect of western spruce budworm defoliation on the physiology and growth of potted Douglas-fir seedlings. *For Sci* 45:280–291
- Kwon GH, Lee MS, Eom HS, Kim YH, Kwon YD (2016) Occurrence and ecological characteristics of *Chiasmia cinerearia* (Bremer et Grey) (Lepidoptera: Geometridae) attacking *Sophora japonica*. *Ko J of Appl Entomol* 55(2):67–71. <https://doi.org/10.1023/A:1022112329990>
- Leishman MR, Thomson VP, Cooke J (2010) Native and exotic invasive plants have fundamentally similar carbon capture strategies. *J Ecol* 98(1):28–42. <https://doi.org/10.1111/j.1365-2745.2009.01608.x>
- Li MY, Guo X, Du N (2021) primary data. xlsx. [dataset]. figshare. <https://doi.org/10.6084/m9.figshare.17004637.v3>

- Li Q, Wang N, Liu X, Liu S, Wang H, Zhang W, Wang R, Du N (2019) Growth and physiological responses to successional water deficit and recovery in four warm-temperate woody species. *Physiol. Plantarum* 167(4):645–660. <https://doi.org/10.1111/ppl.12922>
- Lichtenthaler HK, Wellburn AR (1983) Determinations of total carotenoids and chlorophylls a and b of leaf extracts in different solvents. *Biochem Soc Trans* 603(5):591–593. <https://doi.org/10.1042/bst0110591>
- Liu TT, Cai YP, Wang CZ, Li HH (2015) Biology of *Chrysaster ostensackenella* (Fitch), a new invasive pest of black locust *Robinia pseudoacacia* L. plantations, and a new record of a related species, in China. *Chin J Appl Entomol* 52:942–950. <https://doi.org/10.7679/j.issn.2095-1353.2015.113>
- Lombardero MJ, Alonso-Rodríguez M, Roca-Posada EP (2012) Tree insects and pathogens display opposite tendencies to attack native vs. non-native pines. *Forest Ecol Manag* 281:121–129. <https://doi.org/10.1016/j.foreco.2012.06.036>
- Luo Y, Yuan Y, Wang R, Liu J, Du N, Guo W (2015) Functional traits contributed to the superior performance of the exotic species *Robinia pseudoacacia*: a comparison with the native tree *Sophora japonica*. *Tree Physiol* 36(3):345–355. <https://doi.org/10.1093/treephys/tpv123>
- Maguire AJ, Kobe RK (2015) Drought and shade deplete nonstructural carbohydrate reserves in seedlings of five temperate tree species. *Ecol Evol* 5(23):5711–5721. <https://doi.org/10.1002/ece3.1819>
- Millard P, Sommerkorn M, Grelet G (2007) Environmental change and carbon limitation in trees: a biochemical, ecophysiological and ecosystem appraisal. *New Phytol* 175(1):11–28. <https://doi.org/10.1111/j.1469-8137.2007.02079.x>
- Netherer S, Schopf A (2010) Potential effects of climate change on insect herbivores in European forests-general aspects and the pine processionary moth as specific example. *Forest Ecol Manag* 259(4):831–838. <https://doi.org/10.1016/j.foreco.2009.07.034>
- Palacio S, Maestro M, Montserrat-Martí G (2007) Seasonal dynamics of non-structural carbohydrates in two species of Mediterranean sub-shrubs with different leaf phenology. *Environ Exp Bot* 59(1):34–42. <https://doi.org/10.1016/j.envexpbot.2005.10.003>
- Pinkard EA, Battaglia M, Roxburgh S, O'Grady AP (2011a) Estimating forest net primary production under changing climate: adding pests into the equation. *Tree Physiol* 31(7):686–699. <https://doi.org/10.1093/treephys/tpr054>
- Pinkard EA, Eyles A, Grady APO (2011b) Are gas exchange responses to resource limitation and defoliation linked to source:sink relationships? *Plant Cell Environ* 24(10):1652–1665. <https://doi.org/10.1111/j.1365-3040.2011.02361.x>
- Piper FI (2020) Decoupling between growth rate and storage remobilization in broadleaf temperate tree species. *Funct Ecol* 34(6):1180–1192. <https://doi.org/10.1111/1365-2435.13552>
- Piper FI, Gundale MJ, Fajardo A (2015) Extreme defoliation reduces tree growth but not C and N storage in a winter-deciduous species. *Ann Bot* 115(7):1093–1103. <https://doi.org/10.1093/aob/mcv038>
- Puri E, Hoch G, Körner C (2015) Defoliation reduces growth but not carbon reserves in Mediterranean *Pinus pinaster* trees. *Trees* 29(4):1187–1196. <https://doi.org/10.1007/s00468-015-1199-y>
- Quentin AG, O'Grady AP, Beadle CL, Mohammed C, Pinkard EA (2012) Interactive effects of water supply and defoliation on photosynthesis, plant water status and growth of *Eucalyptus globulus* Labill. *Tree Physiol* 32(8):958–967. <https://doi.org/10.1093/treephys/tps066>
- Quentin AG, Pinkard EA, Beadle CL, Wardlaw TJ, O'Grady AP, Paterson S, Mohammed CL (2010) Do artificial and natural defoliation have similar effects on physiology of *Eucalyptus globulus* Labill. seedlings? *Ann Forest Sci* 67(2):203–203. <https://doi.org/10.1051/forest/2009096>
- Schönbeck L, Gessler A, Schaub M, Rigling A, Hoch G, Kahmen A, Li MH (2020) Soil nutrients and lowered source:sink ratio mitigate effects of mild but not of extreme drought in trees. *Environ Exp Bot* 169:103905. <https://doi.org/10.1016/j.envexpbot.2019.103905>
- Susiluoto S, Hiltunen E, Berninger F (2010) Testing the growth limitation hypothesis for subarctic Scots pine. *J Ecology* 98(5):1186–1195. <https://doi.org/10.1111/j.1365-2745.2010.01684.x>
- Teskey R, Werten T, Bauweraerts I, Ameye M, McGuire MA, Steppe K (2015) Responses of tree species to heat waves and extreme heat events. *Plant Cell Environ* 38(9):1699–1712. <https://doi.org/10.1111/pce.12417>
- Wang N, Zhao MM, Li Q, Liu X, Song HJ, Peng XQ, Wang H, Yang N, Fan PX, Wang RQ, Du N (2020) Effects of defoliation modalities on plant growth, leaf traits, and carbohydrate allocation in *Amorpha fruticosa* L. and *Robinia pseudoacacia* L. seedlings. *Ann For Sci* 77:15. <https://doi.org/10.1007/s13595-020-00953-1>
- Wang RQ, Zhou GY (2000) The vegetation of Shandong Province. Shandong Science and Technology Publisher, Jinan
- Weber R, Schwendener A, Schmid S, Lambert S, Wiley E, Landhäuser SM, Hartmann H, Hoch G (2018) Living on next to nothing: tree seedlings can survive weeks with very low carbohydrate concentrations. *New Phytol* 218(1):107–118. <https://doi.org/10.1111/nph.14987>
- Wiley E, Casper BB, Helliker BR (2017) Recovery following defoliation involves shifts in allocation that favour storage and reproduction over radial growth in black oak. *J Ecol* 105(2):412–424. <https://doi.org/10.1111/1365-2745.12672>
- Yu MX, Li QF, Hayes MJ, Svoboda MD, Heim RR (2014) Are droughts becoming more frequent or severe in China based on the standardized precipitation evapotranspiration index: 1951–2010? *Int J Climatol* 34(3):545–558. <https://doi.org/10.1002/joc.3701>
- Zhang B, Zhang R, Xie T, Li G (2014) Antinociceptive activity of flower buds extract of *Sophora japonica* and its main active ingredient quercetin in bee venom-induced rat model. *J Interact Ethnopharmacol* 3(1):9–14. <https://doi.org/10.5455/jice.20131031042628>
- Zhang J, Xing S (2009) Research on soil degradation of *Robinia pseudoacacia* plantation under environmental stress (in Chinese with English abstract). *Chin J Soil Sci* 40:1086–1090
- Zhang XQ, Liu J, Welham CVJ, Liu CC, Li DN, Chen L, Wang RQ (2006) The effects of clonal integration on morphological plasticity and placement of daughter ramets in black locust (*Robinia pseudoacacia*). *Flora* 201(7):547–554. <https://doi.org/10.1016/j.flora.2005.12.002>

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