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ORIGINAL RESEARCH PAPER

Sex-specific responses of *Populus deltoides* to defoliation

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Abstract

There has been an increasing interest in understanding the differential effects of sexual dimorphism on plant stress responses. However, there is no clear pattern in the responses of the sexes to defoliation. In this study, the effects of different severity of artificial defoliation on biomass production, total nonstructural carbohydrate (NSC) concentration, and photosynthetic rate (P_N) of male and female *Populus deltoides* were examined. We used half and full defoliation to observe the differences between the sexes in three harvest dates (1 week, 4 weeks, and 8 weeks after treatments). We hypothesized that female and male *P. deltoides* compared with an undefoliated control would have compensatory growth in response to defoliation treatments. Results showed that half and full defoliation reduced the growth of both sexes. Following half defoliation, root growth was reduced, especially in males, at T2 (4 weeks after defoliation) and T3 (8 weeks after defoliation), while males showed an increase in height increment under the half defoliation compared with the nondefoliation treatments. By contrast, females were more negatively affected by defoliation than males in terms of biomass after 8 weeks. One week after defoliation, P_N increased significantly in females and males under half defoliation (+30%, +32%, respectively) and full defoliation (+58%, +56%, respectively). However, 8 weeks after defoliation, there was little difference in P_N between defoliated and undefoliated female cuttings. Increases in stomatal conductance (g_s) and leaf nitrogen were observed under fully defoliated female and male cuttings. Moreover, males had less NSC concentrations following half defoliation compared with females. Our results indicate that leaf compensatory growth in male cuttings of *P. deltoides* was maintained by obtaining greater photosynthetic capacity, higher leaf nitrogen, and lower NSC concentration following half and full defoliation. Our results highlight that females suffered from greater negative effects than did males following half defoliation, but under full defoliation, the differences between both sexes were subtle.

Keywords

artificial defoliation; *Populus deltoides*; nonstructural carbohydrates; net photosynthetic rate; sexual differences

Introduction

Defoliation caused by artificial pruning or insect herbivory can influence the growth of trees [1–3]. In Southwest China, annual mean temperature had increased 0.12°C per decade, and annual precipitation had slightly decreased during 1961–2010 [4]. This may result in increased insect damage [5,6]. Defoliation may have negative effects on tree growth by decreased leaf area and reduced carbohydrate supply [7,8]. However, the influence of defoliation on the growth of trees is not always consistent because of the differences in frequency [9], severity, pattern [10], timing [11], and growth stage [12]. Following defoliation, plants seek to maintain leaf regrowth through compensatory growth, which is defined as the restoration of morphological and physiological changes that occur in plants following defoliation [13]. Increases in photosynthetic rate commonly occur following defoliation [14], but there are also exceptions [15]. Although numerous studies have reported defoliation effects on plant growth, the physiological mechanisms driving these responses are unclear. It has been reported that defoliation causes decreases in nonstructural carbohydrates (NSC) that limits growth and survival [16,17]. Moderate and severe defoliation is known to result in reduced NSC concentrations [7,18–22], while light defoliation had no effects on NSC [23]. This implies the need for further studies on the response in trees subjected to different levels of defoliation to examine whether C limitation may explain the defoliation-induced growth reduction.

Dioecious plants are important components of terrestrial ecosystems. Approximately 6% of angiosperm species (14 620 out of 240 000) are dioecious [24]. *Populus deltoides*, a dioecious tree species, is widely distributed in midlatitude region. Defoliation of *Populus* caused by insect herbivory or branch-pruning is common [25]. Sex-related differences in growth and plant physiological responses to the stressful environment have been extensively studied in *Populus*, and females usually show a lower tolerance to abiotic stress when compared with males [26–28]. One of the main physiological mechanisms behind stress tolerance is the carbon accumulation. We expected that the sex-related differences in NSC storage will mediate male and female *P. deltoides* responses to defoliation. We hypothesized that males show a greater ability than females to mitigate the negative effects of defoliation through compensatory mechanisms, but this speculation still lacks a direct test. Although there is abundant information on the male and female *P. deltoides* responses to environmental stress, there is little understanding of defoliation effects on their photosynthesis and growth. Such information is critical when developing models to predict the effects of defoliation events on the reproduction of *Populus*, with the increases in the frequency and intensity of defoliation caused by drought and insects in the future climate.

To gain insight into different responses of dioecious species to defoliation, we chose the industrial timber species, *P. deltoides*, which plays an important role in forestation and ecological restoration. We addressed the following questions: (***i***) does the severity of artificial defoliation influence the magnitude and duration of photosynthetic upregulation and NSC; (***ii***) do responses to defoliation differ between female and male individuals?

Material and methods

Plant material and experimental design

We collected the healthy male and female cuttings of *P. deltoides* from 60 different plants sampled from 15 populations (four adult trees per population) in the Communist Youth League farm in Jingkou District of Zhenjiang, Jiangsu Province, China. After 4 weeks of rooting and stable growth, the female and male cuttings were planted into 10-L plastic pots in a greenhouse at the Chengdu Institute of Biology, the Chinese Academy of Sciences (30°67′ N, 104°06′ E). During the whole experiment, the daytime temperature range was 18–28°C, the nighttime 9–15°C, and a relative humidity was 40–85%. Plants were well watered during the whole experiment periods. The soil was collected from a depth of 10–30 cm from the plot of forest plantation. On May 1, 2015, the female (n = 45) and male (n = 45) cuttings with approximately the same crown size and equal height to eliminate the substantial initial size variation were chosen and replanted in 30-L plastic pots filled with 20 kg of homogenized soil of the same origin (one cutting per pot). The experiment lasted for 2 months from July to September.

The experiment was a completely randomized design with three factorial combinations of defoliation, sex, and time. Three defoliation treatments were applied on July 1, 2015: (***i***) D0, no leaves were removed, n = 15; (***ii***) D50, every second was removed using shears, n = 15; (***iii***) D100, the leaves were removed, excluding apical buds, n = 15. Growth conditions were consistent. We also had three different time periods of the treatment, namely 1 week after defoliation (T1), 4 weeks after defoliation (T2), and 8 weeks after defoliation (T3). In each sampling time, we collected five cuttings per treatment per sex, a total of 30 cuttings.

Biomass

The heights and diameters were measured before harvest in T1, T2, and T3. At the start of the experiment, mean height and diameter of all cuttings were 60 ±5 cm and 4.3 ±0.4 mm, respectively. Thirty cuttings (five cuttings for each sex and treatment) were measured to explore the effect of the defoliation in each duration of the treatment (T1, T2, T3). The cuttings were divided into leaves, stems, and roots and then dried at 75°C for 48 h and their dry mass was determined.

Gas exchange and leaf traits

Five cuttings from each treatment per sex in each sampling date were selected for the measurements. One week, 4 weeks, and 8 weeks after defoliation, measurement was made on the fourth, counted from the top, fully expanded and exposed leaf from each cutting at a given time. The net photosynthetic rate (P_N), stomatal conductance (g_s), transpiration rate (E), and intercellular CO_2 concentration (C_i) were measured with the LI-COR 6400 Portable Photosynthesis System (LI-COR, USA). All measurements were taken between 8 and 11 a.m. under the following conditions: a relative air humidity of 50%; the leaf temperature of 25°C; the molar flow rate of air through the chamber between 200 and 250 μmol s^{-1}; leaf-to-air vapor pressure deficit 1.5 ±0.5 KPa; the CO_2 concentration of 400 ±5 μmol mol^{-1}, and the photosynthetic photon flux density of 1500 μmol m^{-2} s^{-1}.

Total chlorophyll concentration was determined using a spectrophotometer (UV-330; Unicam, UK). After gas exchange measurements, fresh leaves were cut immediately and extracted in 80% (v/v) chilled acetone and quantified according to Lichtenthaler [29] at wavelengths of 470, 646, and 663 nm. Chlorophyll *a* and chlorophyll *b* were calculated from equations derived by Porra et al. [30].

Carbon and nitrogen contents in leaves, stems, and roots

Dry samples of leaves, stems, and roots were ground and passed through a 200-mesh screen. Concentrations of N and C were determined by the semi-micro Kjeldahl method [31] and the rapid dichromate oxidation technology [32], respectively. The total C to N ratio (C:N) was calculated as an estimate of the long-term nitrogen use efficiency [33].

Nonstructural carbohydrates

Nonstructural carbohydrate was defined as soluble sugars and starch concentrations. About 50 mg of the fine powder from the dried roots, stems, and leaves were placed in 5 mL 80% (v/v) ethanol at 80°C for 30 min and centrifuged at 5000 *g* for 10 min. According to the anthrone–sulfuric acid method [34], the total soluble sugars and starch were estimated using 1.2% anthrone in concentrated H_2SO_4 as a reagent. Then, the total soluble sugars were determined colorimetrically (Multiskan GO 1510; Thermo Fisher Scientific, Finland) at 625 nm. The starch content was determined from the pellet of plant material that remained after the removal of ethanol [35]. Fructose and sucrose were examined colorimetrically (Multiskan GO 1510) at 480 nm following the modified resorcinol method [36].

Statistical analysis

Before ANOVA, data were checked for normality and homogeneity of variance. We performed three-way ANOVA for the effects of defoliation, time, and sex to discover differences between sex response to defoliation using the R.3.3.2 (R Foundation for Statistical Computing, Vienna, Austria). Significant differences among treatment means were analyzed using Tukey's multiple comparison post hoc tests. Group regressions were used to determine the effect of sex on the relationships of P_N with g_s and leaf nitrogen.

Results

Sexual differences in growth traits

For female cuttings, both D50 and D100 treatments significantly reduced diameter increment (Fig. 1b) but only D100 treatment significantly reduced diameter in male cuttings (Fig. 1d). Compared with D0, D100 had more pronounced negative effects on female and male diameter increment at T2 (respectively −80%, −61%) and T3 (respectively −79%, −68%). Unexpectedly, compared with D0, D50 significantly increased the height increment for males at T2 and T3 (Fig. 1c).

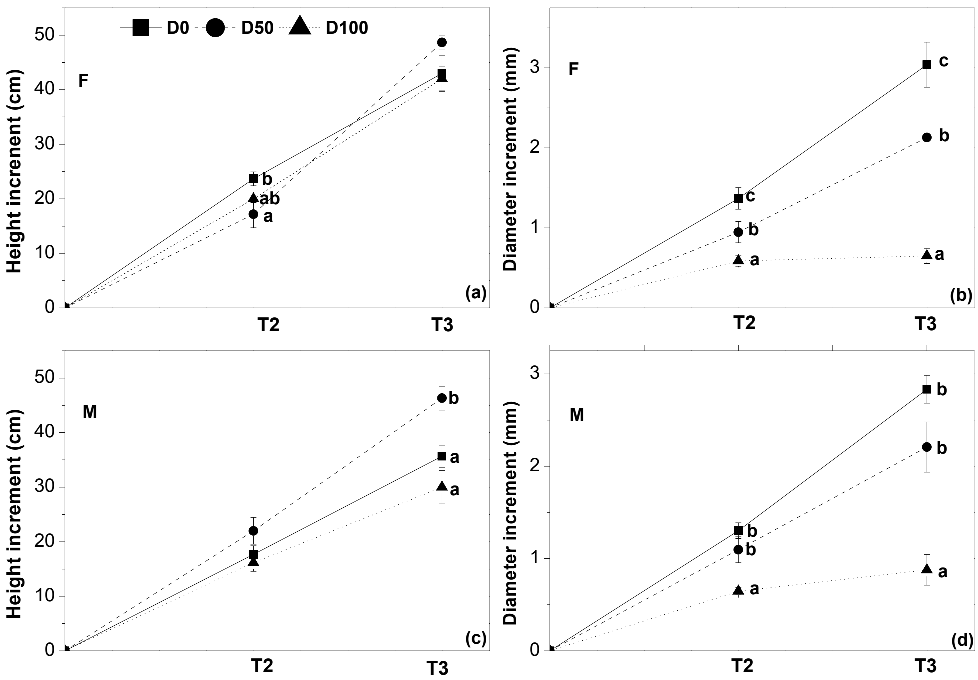

Fig. 1 Effect of defoliation on the growth of *P. deltoides* for each sampling date. Values are mean ±*SE* ($n = 5$). Different letters indicate that means are significantly different at $p < 0.05$ in a sampling date. F – female cuttings; M – male cuttings; T2 – 4 weeks after defoliation; T3 – 8 weeks after defoliation; D0 – no defoliation; D50 – half defoliation; D100 – full defoliation.

Overall, the defoliation significantly decreased stem dry mass, root dry mass, total dry mass of males and females (Fig. 2), but females exhibited higher damage under the same defoliation at T3, showing that total biomass significantly decreased in females and males under half defoliation (−27%, −22%, respectively) and full defoliation (−69%, −61% respectively). Moreover, after 4 and 8 weeks, total biomass in males showed no difference between M50 (half defoliation of males) and M0 (nondefoliation of males), whereas males under M50 had less root biomass compared with M0. Although the specific leaf area (SLA) of both sexes was decreased by D50 and D100 at T2 and T3,

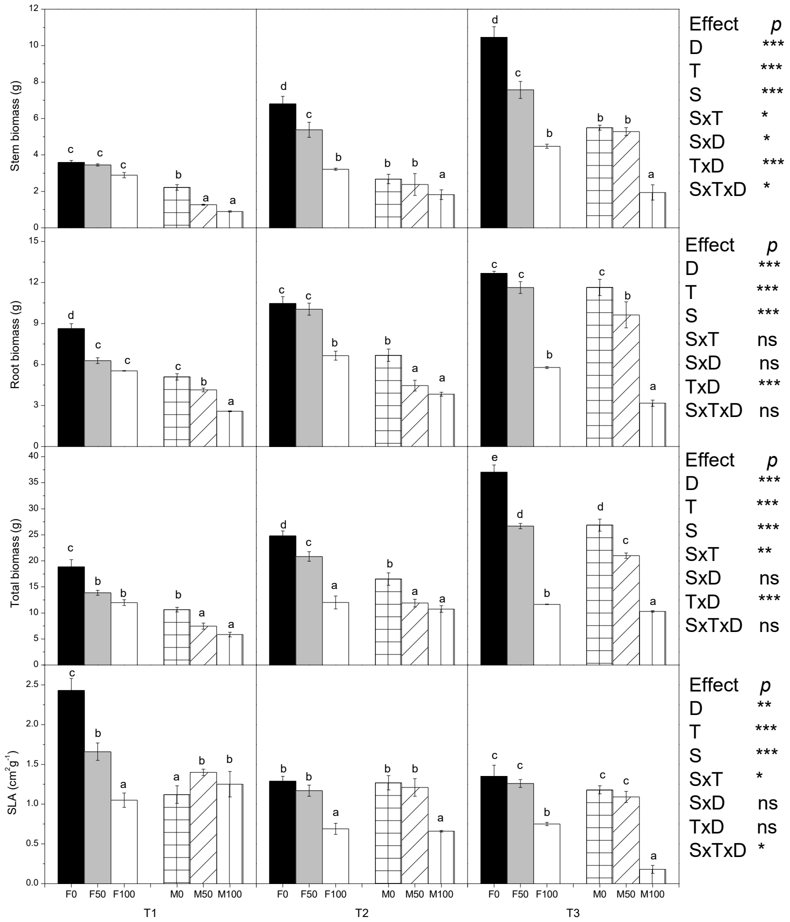

Fig. 2 Stem biomass, root biomass, total biomass, and specific leaf area (SLA) in male and female individuals of *P. deltoides* as affected by artificial defoliation in three harvest dates. Values are mean ±*SE* ($n = 5$). Treatments: F0 – nondefoliation of females; F50 – half defoliation of females; F100 – full defoliation of females; M0 – nondefoliation of males; M50 – half defoliation of males; M100 – full defoliation of males; T1 – 1 week after defoliation; T2 – 4 weeks after defoliation; T3 – 8 weeks after defoliation. Different letters indicate significant differences among treatments according to Tukey's test at a significance level of $p < 0.05$. D – defoliation effect; T – time effect; S – sex effect; S × D – Sex × Defoliation effect; S × T – Sex × Time effect; D × T – Time × Defoliation effect; D × T × S – Sex × Time × Defoliation effect. The significance values of the factorial analysis (ANOVA) are denoted as follows: ns – nonsignificant; * $0.01 < p < 0.05$; ** $0.001 < p \leq 0.01$; *** $p \leq 0.001$.

the decreases of SLA of males were not significant under D50. Compared with the same sex individuals grown under the defoliation, the females exhibited significantly greater decreases in root biomass and total biomass than males under D50 and D100. In addition, Sex × Time × Defoliation interactions were significant in the case of stem biomass and SLA ($p < 0.05$).

Sexual differences in carbon and nitrogen accumulation

Compared with nondefoliation treatment, females under D100 treatment significantly increased N contents in leaf at the first harvest date (+64%) (Tab. 1, Fig. 3), while the defoliation had a tendency to decrease the leaf N in males. For both genders, the defoliation had significant effects on the N content, whereas there was no effect on the C content in roots and stems (Tab. 1). In addition, Sex × Time × Defoliation interactions were significant in the case of the N content in stems and in leaves ($p < 0.05$). Furthermore, the C:N ratio in stems was significantly affected by the interaction of Sex × Defoliation ($p < 0.05$) (Tab. 1).

Tab. 1 The significance values of factorial analysis on the total leaf C, N, C:N ratio, and NSC (fructose, sucrose, and starch), and the allocation content shown in Fig. 2 and Fig. 3.

	$p > F_D$	$p > F_T$	$p > F_S$	$p > F_{D×T}$	$p > F_{S×D}$	$p > F_{S×T}$	$p > F_{S×D×T}$
Roots							
C	0.134 (2.12)	0.907 (0.10)	0.427 (0.65)	0.449 (0.945)	0.719 (0.33)	0.482 (0.744)	0.128 (1.919)
N	**0.044 (3.42)**	**0.032 (3.81)**	0.102 (2.81)	0.370 (1.10)	**0.024 (4.16)**	0.072 (2.83)	0.281 (1.32)
C:N	0.053 (3.19)	**0.018 (1.19)**	0.387 (0.77)	0.853 (0.33)	**0.006 (5.94)**	0.080 (2.71)	0.323 (1.21)
Soluble sugars	**<0.001 (809.48)**	**<0.001 (186.171)**	**<0.001 (19.41)**	**<0.001 (19.53)**	**0.003 (6.74)**	**<0.001 (39.76)**	**<0.001 (23.48)**
Starch	**<0.001 (445.02)**	**<0.001 (221.47)**	0.303 (1.09)	**<0.001 (122.51)**	**<0.001 (12.23)**	**0.004 (6.34)**	**<0.001 (5.95)**
NSC	**<0.001 (900.72)**	**<0.001 (396.53)**	**0.001 (11.27)**	**<0.001 (110.07)**	**<0.001 (16.62)**	**<0.001 (31.26)**	**<0.001 (12.74)**
Stems							
C	0.799 (0.23)	0.489 (0.73)	0.993 (0.00)	0.248 (1.42)	0.296 (1.26)	0.113 (2.32)	0.209 (1.55)
N	0.467 (0.78)	0.777 (0.26)	0.777 (0.08)	0.276 (1.33)	**<0.001 (8.63)**	0.597 (0.52)	**0.006 (4.37)**
C:N	0.630 (0.47)	0.685 (0.38)	0.626 (0.24)	0.370 (1.10)	**0.008 (5.48)**	0.555 (0.59)	**0.038 (2.84)**
Soluble sugars	**<0.001 (138.47)**	**<0.001 (77.73)**	**0.025 (5.46)**	**<0.001 (6.98)**	0.531 (0.65)	0.065 (2.95)	0.062 (2.47)
Starch	**<0.001 (346.76)**	**<0.001 (88.68)**	**<0.001 (150.71)**	**<0.001 (31.33)**	**<0.001 (20.99)**	**0.002 (7.71)**	**0.006 (4.34)**
NSC	**<0.001 (515.13)**	**<0.001 (144.77)**	**0.001 (138.97)**	**<0.001 (16.95)**	**<0.001 (16.55)**	**<0.001 (8.42)**	**<0.001 (6.27)**
Leaves							
C	0.430 (0.86)	**0.002 (7.18)**	**0.009 (7.70)**	0.629 (0.65)	0.125 (2.21)	0.128 (2.18)	0.907 (0.25)
N	**<0.001 (8.59)**	0.077 (2.75)	0.388 (0.76)	0.478 (0.89)	0.242 (1.48)	0.537 (0.63)	**0.002 (5.44)**
C:N	0.057 (3.11)	**0.031 (3.85)**	0.455 (0.57)	0.343 (1.17)	0.078 (2.74)	0.95 (0.05)	**0.008 (4.07)**
Soluble sugars	**<0.001 (33.09)**	**<0.001 (13.81)**	0.149 (2.11)	**<0.001 (21.84)**	**<0.001 (12.24)**	**0.009 (5.37)**	0.706 (0.54)
Starch	**<0.001 (252.25)**	**<0.001 (208.85)**	0.137 (2.31)	**<0.001 (18.90)**	**<0.001 (20.40)**	0.104 (2.41)	**<0.001 (32.40)**
NSC	**<0.001 (316.79)**	**<0.001 (134.67)**	**0.019 (6.06)**	**<0.001 (19.39)**	**<0.001 (39.16)**	0.740 (0.30)	**<0.001 (31.62)**

F_D – defoliation effect; F_T – time effect; F_S – sex effect; $F_{S×D}$ – Sex × Defoliation effect; $F_{S×T}$ – Sex × Time effect; $F_{D×T}$ – Time × Defoliation effect; $F_{D×T×S}$ – Sex × Time × Defoliation effect. Significant effects ($p < 0.05$) are shown in bold.

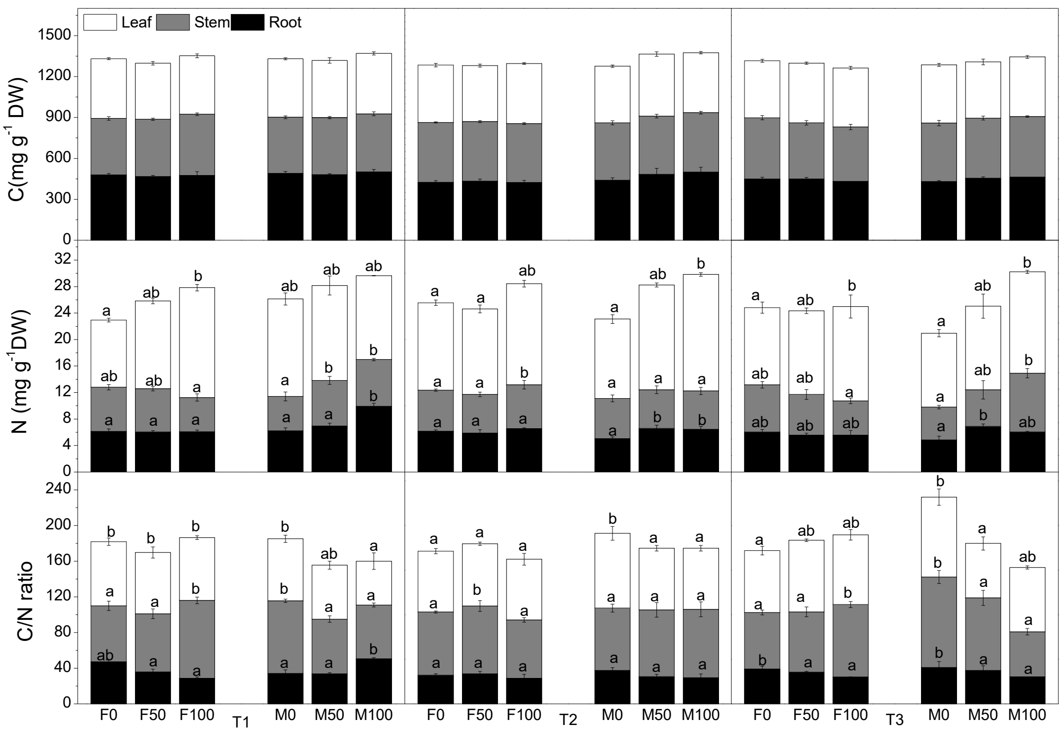

Fig. 3 Carbon, nitrogen, and C:N ratio in total leaves (white bars), stems (grey bars), and roots (black bars) of *P. deltoides* males and females under different defoliation per date (T1, T2, and T3). F0 – nondefoliation of females; F50 – half defoliation of females; F100 – full defoliation of females; M0 – nondefoliation of males; M50 – half defoliation of males; M100 – full defoliation of males. Different letters indicate significant differences among treatments according to Tukey's test at a significance level of $p < 0.05$. Values are mean ±*SE* ($n = 5$).

Sexual differences in gas exchange parameters

For both sexes, there were increases in P_N following defoliation ($p < 0.05$). After defoliation at T1, P_N in females and males increased significantly under half defoliation (+30%, +32%, respectively) and full defoliation (+58%, +56%, respectively). This photosynthetic upregulation was more pronounced in males than in females in each sampling date. By week 8 (T3), P_N of female cuttings decreased sharply in D50 and D100 (−30%, −41%, respectively). However, upregulation of P_N of the male cuttings remained across the periods of T1 and T2. Moreover, P_N, g_s, C_i, E, and chlorophyll *a*+*b* were all affected by the interaction of Sex × Time × Defoliation significantly ($p < 0.05$) (Fig. 4). Across all sampling dates, there was a positive relationship between P_N and g_s in females and males ($y = 12.60 + 0.93x$, $R^2 = 0.67$; $y = 8.43 + 18.90x$, $R^2 = 0.83$) (Fig. 5a). In addition, there was a positive relationship between P_N and the N content in leaves of females, which was not shown in males (Fig. 5b)

Sexual differences in nonstructural carbohydrates

In all sampling times, starch was the largest component of NSC in all organs (Fig. 6). Compared with D0, the soluble sugars concentrations in roots and stems of both females and males significantly decreased under D100 but no significant decreases under D50 (Tab. 1, Fig. 6). Moreover, the soluble sugar concentrations in leaves of females were higher than in roots and stems (Fig. 6). Across all the sampling dates, D100 and D50 resulted in significantly lower starch concentrations in both females and males. In addition, the starch concentration in stems of females was significantly higher than in males under the same treatment (Fig. 6). Compared with the same defoliation, the female individuals exhibited higher nonstructural carbohydrates than those in males under D0, whereas there were no significant differences in both sexes under half defoliation (Tab. 1, Fig. 6). In addition, the fructose concentration in roots, the sucrose concentration

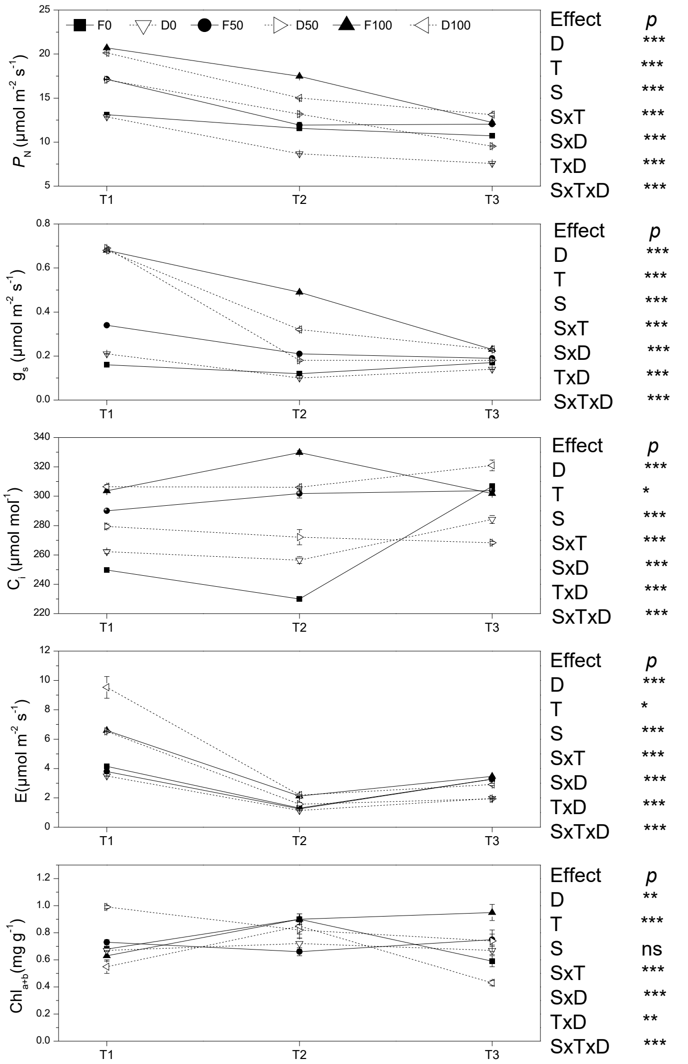

Fig. 4 Net photosynthesis rate (P_N), stomatal conductance (g_s), transpiration (E), intercellular CO_2 concentration (C_i), and total chlorophyll content (Chl_{a+b}) in the female and male individuals of *P. deltoides* as affected by artificial defoliation in different in three harvest dates. Values are mean ±*SE* ($n = 5$). Treatments: F0 – nondefoliation of females; F50 – half defoliation of females; F100 – full defoliation of females; M0 – nondefoliation of males; M50 – half defoliation of males; M100 – the full defoliation of males. D – defoliation effect; T – time effect; S – sex effect; S×D – Sex × Defoliation effect; S×T – Sex × Time effect; D×T – Time × Defoliation effect; D×T×S – Sex × Time × Defoliation effect. The significance values of the factorial analysis (ANOVA) are denoted as follows: ns – nonsignificant; * $0.01 < p < 0.05$; ** $0.001 < p \leq 0.01$; *** $p \leq 0.001$.

in roots and stems, the starch concentration in roots, stems, and leaves were significantly affected by the interaction of Sex × Time × Defoliation ($p < 0.05$) (Tab. 1).

Discussion

Defoliation interferes with plant growth, but the effects of defoliation differ among plant species and may highly depend on the type and the severity of defoliation. Douglas fir under defoliation treatments had the significant decreases in height and diameter growth [37], while artificial defoliation of *Eucalyptus globulus* only negatively affected the diameter growth [38]. Our results showed that there were significant differences in height growth of *P. deltoides* males and females when exposed to defoliation. Some studies reported that defoliation reduces the competition for light on the remaining leaves and also changes the spectral composition of light. Such a change in light quality results in morphogenetic (such as SLA) responses in plants [12,13]. Moreover, significant biomass reduction following defoliation have been observed as defoliation limits carbon uptake by reducing the total leaf area, indicating that increases in photosynthesis cannot offset carbon losses [39]. Several studies have found that defoliation reduces growth due to nitrogen loss or water stress [22] but this is not the case in our study, as individuals in our study were well watered and fertilized. Besides, we found some evidence that defoliation increases the N content in roots and leaves (Fig. 3). For males under half defoliation, stem growth is favored at expenses of root [18], while for female under half defoliation, root growth is favored at expenses of stem. Males may maintain leaf regrowth by the increase in the allocation of reserves from root to shoot [40]. Zhao et al. [41] also found that medium defoliation results in the emergence of new leaves and stimulates relative growth rate through photosynthetic upregulation, as is also shown in the present study. Furthermore, our study shows that different severity of defoliation results in different extent and duration of photosynthetic upregulation. This is similar to *Pseudotsuga menziesii* [42] and *Eucalyptus globulus* [43]. The upregulation in photosynthetic rate increases with the severity of defoliation. Similarly, more severe defoliation results in a greater increase in the net photosynthesis rate than less severe defoliation [10]. Such photosynthetic upregulation allows species to compensate, to some degree, for the loss of leaf area [12,44]. Several authors have argued that partial defoliation decreases source:sink ratio and increases demand for carbohydrates to rebuild crowns [45,46]. Many other studies have shown that partial defoliation increases stomatal conductance [47–50], as our study shows.

Various mechanisms have been proposed for compensatory growth, such as higher photosynthetic rate or stomatal conductance [51]. Although leaf loss can lead to significant reductions in stem biomass, root biomass, and total biomass [52–54], *P. deltoides* allocates more carbon to the aboveground biomass through compensatory growth. The

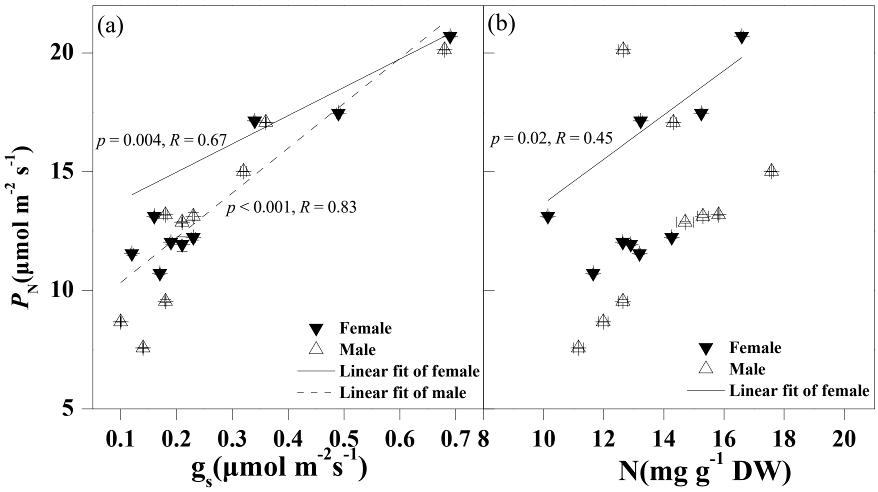

Fig. 5 Relationship between net photosynthesis rate (P_N) and stomatal conductance in female and male *P. deltoides* (**a**). Relationship between P_N and the leaf nitrogen in female and male *P. deltoides* (**b**). Values are mean ±*SE* (n = 4). Relationships are shown only if significant ($p < 0.05$).

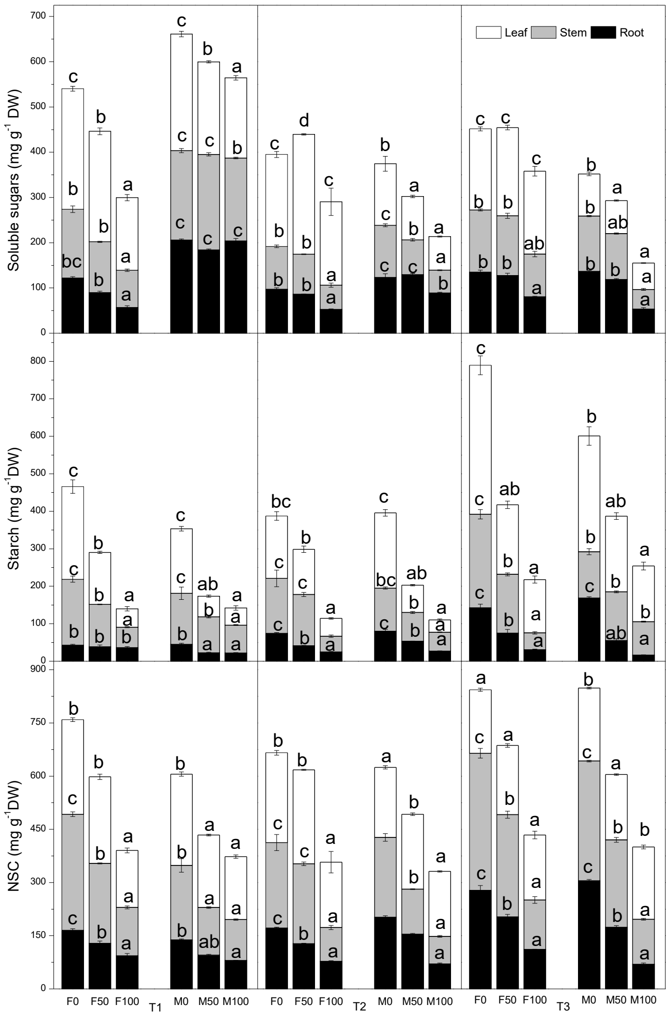

Fig. 6 Effect of defoliation on soluble sugars, starch, and NSC concentrations in different organs of females and males under T1, T2, and T3. Values are mean ±*SE* (n = 5). F0 – nondefoliation of females; F50 – half defoliation of females; F100 – full defoliation of females; M0 – non-defoliation of males; M50 – half defoliation of males; M100 – full defoliation of males. Different letters indicate significant differences among treatments according to Tukey's test at a significance level of $p < 0.05$.

present study found that defoliation was partially compensated by increased photosynthetic rate [22]. To compensate for leaf removal, plant requires large amounts of energy investment. Many studies have indicated that the rapid regrowth is highly associated with the changes in carbohydrates, C and N metabolism [55,56]. Compared with D0, we found that NSC concentration in individuals under D50 and D100 had decreased, as shown in other studies [7,55,56]. The decrease in total NSC was primarily attributable to the depletion of starch with defoliation treatment, which might in turn reduce carbon allocation to tree growth.

In conclusion, our study shows that severity of defoliation influences plant responses. The increase in photosynthetic rate increased with defoliation severity. There are sex-specific physiological differences in defoliation between males and females of *P. deltoides*. Moreover, males and females showed compensatory growth of different extent following defoliation. Males of *P. deltoides* had a greater ability than females in compensation for the negative effect of defoliation through great photosynthetic capacity, high leaf nitrogen, and low NSC concentration, showing that males maintained higher height increment and less decreased total biomass than did females. Furthermore, the full defoliation induced lesser sex-specific differences in height increment, diameter increment, and NSC concentration than did the half defoliation. Our results suggest that sex-specific difference of NSC and N allocation may explain the differences in growth between males and females of *P. deltoides* under defoliation stress. However, long-term study is required to determine in depth sex-related differences in resource acquisition and allocation under defoliation.

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