Responses of leafing phenology and photosynthesis to soil warming in forest-floor plants

Running title: Soil warming effects on understory plants

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ABSTRACT

Phenological and physiological responses of plants to climate change are key issues to understand the global change impact on ecosystems. To evaluate the species-specific responses, a soil-warming experiment was conducted for seven understory species having various leaf habits in a deciduous forest, northern Japan; one evergreen shrub, one semi-evergreen fern, one summer-deciduous shrub, and four summer-green herbs. Soil temperature in the warming plots was electrically maintained 5ºC higher than control plots. Responses of leafing phenology highly varied among species: new leaf emergence of the evergreen shrub was delayed; senescence of overwintering leaves of the semi-evergreen fern was accelerated resulting in the shift to deciduousness; leaf shedding of the summer-deciduous shrub was accelerated. Among four summer-green species, only an earliest leaf-out species advanced growth initiation, but the period of growth season was not changed. Physiological responses to soil warming were also highly species-specific: the warming treatment increased the photosynthetic activity of the summer-deciduous shrub and one summer-green species, decreased that of the semi-evergreen fern, while other species did not show any changes in photosynthetic traits. Totally, the soil warming impacts on understory plants was apparent in spring. It was suggested that modification of snow conditions is important issue especially for plants with overwintering leaves. Responses of understory vegetation to climate change may highly vary depending on the composition of leaf habits in the cool-temperate forests.

Keywords: cool-temperate forest, leaf habit, photosynthesis, soil warming, spring phenology, understory vegetation.

1. Introduction

Initial stages of global change impact on ecosystems are changes in phenology and physiology of individual organisms (Hughes, 2000). Accordingly, with recent climate changes various phenologies are changing world-wide (e.g. Parmesan and Yohe, 2003; Menzel et al., 2006; Parmesan, 2006). Wider amplitude of phenological responses of plants is expected in high latitude ecosystems, such as boreal forests, alpine and arctic ecosystems, where temperature restricts the growing season. Specifically in the beginning of the growing season (spring), rather than in autumn, phenology is shown to be most sensitive to climate change (Parmesan, 2006; Dunne et al., 2003; Aerts et al., 2006; Delbart et al., 2008) because temperature is one of the important factors during winter and spring that influences the time of breaking dormancy for specific groups (Körner and Basler, 2010; Shen, 2011).

In deciduous forests, the phenology and physiology of understory plants demonstrate a critical period of carbon gain in early spring before canopy closure (Gill et al., 1998; Rothstein and Zak, 2001; Augsperger and Bartlett, 2003; Augsperger et al., 2005; Ida and Kudo, 2008). This is related to the relatively high light received in spring, when canopy trees have no leaves, in combination with moderate temperatures. However, understory growth is not restricted to spring and a wide array of leaf habits co-exists in a deciduous forest, such as summer-green, evergreen, semi-evergreen and summer-deciduous plants (Kikuzawa, 1989; Uemura, 1994). All these leaf habits occupy a niche in strong relation to the light availability and favorable temperature in the understory. Therefore, large changes are expected in this ecosystem with a global change in temperature. Higher temperature can directly affect the phenology and physiology of understory species but also indirectly through changes in phenology of canopy tree species or snow cover and drier conditions.

There are only a few studies on experimental warming of understory plants (Farnsworth et al., 1995; De Frenne et al., 2010; Rollinson and Kaye, 2012). For over 130 species analyzed in one experiment, the functional groups of tall forbs and large trees responded to warming by advancing leaf-out in spring, whereas the short forbs, shrubs and small trees did not alter leafing phenology (Rollinson and Kaye, 2012). In another study including short forbs and shrubs, however, summer-green herbs with dormant meristems at the soil surface advanced growth initiation in response to soil warming (Farnsworth et al., 1995; Lapointe, 2001) and shrubs with dormant buds above the soil surface did not. Also for shrubs or herbs with an evergreen, semi-evergreen and summer-deciduous leaf habit, large changes are expected in response to warming when these plants are covered by snow in winter. Because warming reduces the snow cover, leaves will be exposed to light for a longer period. This can have a positive effect on the carbon gain of leaves but also the opposite response due to freezing damage or soil desiccation when not protected by a snow cover which might even result in earlier leaf senescence (Inouye, 2000; Giménez-Benavides et al., 2007; Augspurger, 2009; Bokhorst et al., 2009; Taulavuori et al., 2011). Thus, phenological and physiological responses to warming are not necessarily limited to functional groups as defined by Rollinson and Kaye (2012).

In this study, we focus on the phenological responses to soil warming of seven dominant understory plant species in a deciduous forest in northern Japan. These include summer-green, evergreen, semi-evergreen and summer-deciduous plants for which we suggest that the response to soil warming depends on the leaf habit. Besides the recording of phenology, we also estimated the growth and measured photosynthetic rates during the growth of all species. Soil warming was initiated three years before the start of the experiment to allow for replacement of all evergreen leaves under the experimentally increased temperature conditions. Based on the previous studies mentioned before, we aim to test following predictions. First, phenological and physiological changes will be more prominent in spring than in autumn. Spring is an important carbon assimilation period for understory plants (Gill et al., 1998; Augsperger and Bartlett, 2003), while autumnal photosynthesis after canopy leaf fall may be less significant probably due to lower solar radiation (Augsperger et al., 2005; Richardson et al., 2010; but see also Fridley, 2012). Therefore, phenological and physiological changes in spring may influence the subsequent growth and performance of understory plants. Second, plants retaining leaves during winter are expected to respond more sensitively to soil warming than summer-green plants if effect of snow conditions is important. Finally, even within summer-green plants, early leaf-out species may be more sensitive to soil warming than late leaf-out species. This prediction is based on the previous study demonstrating that between-year variations in flowering phenology were larger in spring ephemerals than in summer-blooming herbs that leafed out late in spring (Kudo et al., 2008), suggesting the sensitivity of early leaf-out species to spring climate conditions.

2. Materials and Methods

2.1. Study site

The study was conducted in a deciduous forest of the Tomakomai Experimental Forest of Hokkaido University (42°40' N, 141°36' E; 70–80 m elevation) in Hokkaido, northern Japan. The mean monthly temperature ranges from –4.1°C (January) to 20.3°C (August), and annual precipitation is 1228 mm. Snow covers the ground usually from mid-December to late March, and the average snow depth is about 50 cm. The major canopy species are *Quercus crispula*, *Acer mono*, *Ostrya japonica*, and *Prunus ssiori*. Leaf emergence of canopy trees usually starts in mid-May and canopy closure is completed by mid-June (Maeno and Hiura, 2000). The forest understory is shaded from late June to early October.

2.2 Experimental warming

Experimental plots were established in the spring of 2007. Four control plots (C1 to C4) and four warming plots (T1 to T4) of 5 m × 5 m each were arranged in the forest. Electrically-heated wire (Nihon Noden, Tokyo) of 120 m length was inserted into the soil with help of a flat-bladed shovel at 20 cm intervals at the depth of 5–10 cm in each warming plot. A flat-bladed shovel was also used in the control plots, similar to the warming plots but without adding the heating cable, to have a similar disturbance between plots. The heating wire was controlled to keep the warming plots 5°C warmer than control plots throughout the year (for details, see reference in Nakamura et al., 2010). To reduce the disturbance effects of wire setting on understory vegetation and the short time effect of warming on plant responses, measurements of this study were mainly conducted in 2009 with additional measurements in 2010 for physiological traits of some plants (as mentioned later).

In this warming experiment, soil temperature at 5–10 cm depth was 5.5°C warmer (daily mean of 2008–2009) and soil surface temperature was 3.5°C warmer in the warming plots in comparison with the control plots. There were no significant differences in inorganic nitrogen (NH_4-N and NO_3-N) between control and warming plots during growing season (Ueda et al., unpublished data).

To record the daily snow condition during the wintertime, automated cameras (KADEC21-EYEII, KONA System, Sapporo) were set in one control and one warming plot. Existence of snow cover was checked by visual inspection of photographs. To evaluate the warming effects on soil moisture conditions, furthermore, volumetric soil water contents at 10 cm deep were measured at one to two-week intervals by a soil moisture sensor (HydroSense TM, Campbell Scientific Australia, Queensland) during April to September in 2009.

2.3. Target species

We selected the following seven understory species that were common in this forest: *Pachysandra terminalis* (Buxaceae), *Dryopteris crassirhizoma* (Dryopteridaceae), *Daphne kamtschatica* var. *jezoensis* (Thymelaeaceae), *Smilacina japonica* (Ruscaceae), *Trillium apetalon* (Melanthiaceae), *Parasenecio auriculata* (Asteraceae), and *Phryma leptostachya* var. *asiatica* (Phrymaceae). *Pachysandra terminalis* is an evergreen shrub of 15–20 cm high, and leaf longevity is 2–3 years. It often develops multiple branching prostrate stems forming a large clonal patch composed of many ramets. Shoot growth of individual ramets usually occurs in late spring to early summer (Yoshie and Kawano, 1986). *Dryopteris crassirhizoma* is a semi-evergreen fern whose leaves are arranged as a funnel shape, commonly 50–70 cm high. In late autumn, leaves are prostrated on the ground in a radical pattern, then overwinter on the ground. After snowmelt in next spring, overwintered leaves remain as a prostrate form on the ground, then senescence gradually from distal to basal parts in accordance with the expansion of new leaves from a frond located on the soil surface (Tani and Kudo, 2005). *Daphne kamtschatica* var. *jezoensis* is a summer-deciduous shrub whose plant height is 30–40 cm. Leaf emergence starts in mid-August and leaf expansion last until late autumn, then leaves overwinter. In next spring, several leaves are additionally produced, but all leaves are shed by late June (Kikuzawa, 1989; Lei and Koike, 1998). *Smilacina japonica* and *T. apetalon* are summer-green herbs of 20 cm high whose leaves are simultaneously produced in early spring and aboveground parts usually senesce in early summer (Ida and Kudo, 2008, 2009). *Parasenecio auriculata* and *P. leptostachya* var. *asiatica* are also summer-green herbs of 30 cm high but leaf production lasts successively from spring to early summer and aboveground parts usually senesce in autumn (Ida and Kudo, 2009).

2.4. Monitoring of leafing phenology and height growth

Measurements of leafing phenology and plant height were conducted 22 times from the beginning of April to early November in 2009 at the intervals of about seven days during spring and autumn, and about 14 days during summer. Because of the differences in growth form and plant structure among target species, we selected appropriate measurements to assess the growth pattern for individual species.

For the evergreen *P. terminalis* having a successive shoot growth pattern, we recorded height growth of individual ramets. Soon after snowmelt in early

April 2009, we randomly selected 11–12 vegetative ramets without floral buds in each plot (C1, C2, C3, C4, T1, T2, T3, T4) and marked them with numbered tags (n = 46 and 47 in the control and warming plots, respectively). Then, we recorded plant height of each ramet from the soil surface by a ruler.

For the semi-evergreen *D. crassirhizoma*, we randomly selected and marked five plants in each plot (n = 20 in each treatment) of which the senescence pattern of overwintering leaves, leafing phenology, and leaf height were recorded throughout the growth season. The senescence pattern of overwintering leaves was quantified at 10% accuracy in area per plant. Phenology of current leaves was classified into growth initiation of frond (stage 1), leaf expansion as a funnel shape arrangement (stage 2), and leaf prostration in autumn (stage 3).

For the summer-deciduous shrub, *D. kamtschatica*, the number of leaves on a single branch was recorded. In early April 2009 soon after snowmelt, 3–6 plants were randomly selected in each plot (in total, n = 20 and 17 in the control and warming plots, respectively), marked with a numbered tag on one branch per plant, and the leaf number was recorded throughout the growth season.

For four summer-green herbaceous species, plant height of randomly selected vegetative plants was recorded repeatedly from shoot emergence to senescence. The number of marked plants was 34 in the control plots and 24 in the warming plots for *S. japonica*, 38 and 39 for *T. apetalon*, 19 and 37 for *P. auriculata*, and 48 and 42 for *P. leptostachya*, respectively.

2.5. Photosynthetic measurement

To assess the warming impact on physiological activity, photosynthetic CO_2 exchange was measured in the field using a LI-6400 portable photosynthesis system (Li-Cor, Lincoln, NE, USA). Measurements were conducted for randomly selected 6–12 plants in each of the control (C1 or C2) and warming plots (T1 or T2) in early spring before canopy closure (mid-April), late spring under progressive closure (late May), late summer under complete closure (early September), and late autumn after canopy opening (early November) depending of the leaf habit of individual species. We used maximum photosynthetic rate (P_{max}) and stomatal conductance for water vapor (g_s) as an index of photosynthetic activity and water stress (Lange et al., 1971), respectively. Measurements were conducted under high irradiance (1000 μmol m^{-2} s^{-1}) of photosynthetically active radiation (PAR) using a red-blue LED light source at a controlled temperature of 15–25°C depending on ambient temperature. The ambient CO_2 concentration was maintained at 380 μL L^{-1} and the vapor pressure deficit did not exceed 1.1 hPa.

Physiological measurements for plants with overwintering leaves were conducted from 2009 to 2010 to chase same leaf cohorts. For evergreen *P. terminalis*, upper positioned leaves (i.e. current leaves) were measured in November 2009, April, May and September 2010. For semi-evergreen *D. crassirhizoma*, measurements were conducted in May, September, November 2009 (for current leaves), and April 2010 (for overwintered leaves). For summer-deciduous *D. kamtschatica*, measurements were conducted in November 2009 (for current leaves), April and May 2010 (after overwintering). Measurements for summer-green herbs with relatively short growth period, *S. japonica* and *T. apetalon*, were conducted only in May 2009. Measurements for summer-green herbs with long growth period, *P. auriculata* and *P. leptostachya*, were conducted in May and September 2009.

2.6. Data analysis

For *P. terminalis*, maximum plant height, annual height growth, and growth initiation time were compared between control and warming plots. The height growth was expressed as an increment from the first observation soon after snowmelt (6 April) to the date of maximum height record in each plant. Growth pattern was calculated by the application of logistic regression for individual ramets. Growth initiation time was defined as the day when height increment attained 50% of total height growth estimated from the logistic regression. Ramets that produced no or only a few leaves were removed from the analyses. Analysis of plant height was conducted by a linear mixed-effect model (LMM) supposing a Gaussian error distribution in which treatment was set as a fixed factor and plot as a random factor. For the LMM of height growth and growth initiation, we set both treatment and plant size (at maximum) as fixed factors to exclude the size effect on growth pattern from treatment effect, and the best-fit model was selected based on the Akaike's Information Criteria (AIC).

For *D. crassirhizoma*, the growth pattern was compared between control and warming by the leaf height at the phenology stage 2, survival rate of overwintering leaves (area percentage in early April) and the following phenological events; (1) day of complete senescence of overwintering leaves, (2) day of current-leaf expansion (first record of the phenology stage 2), and (3) day of leaf prostration in autumn (first record of the phenology stage 3). Statistical analyses were same as *P. terminalis*.

For *D. kamtschatica*, the numbers of overwintering leaves, additional leaves produced in spring, new leaves produced in autumn and the following phenological events were compared to assess the seasonal growth pattern; (1)

day of leaf senescence in early summer that was defined as a day when remaining leaf number decreased to 50% of maximum leaf number, and (2) day of leaf emergence that was defined as a day when leaf number increased to 50% of autumnal leaf production. These phenological events were estimated by the application of logistic regression for individual plants. Statistical analyses were same as *P. terminalis*.

For summer-green herbs, maximum plant height, growth initiation and senescence time, and growth period were compared between control and warming plots. The growth initiation was defined as the day when plant height attained 50% of maximum height estimated from the application of logistic regression in each plant. The senescence time was defined as the day between the first observation date of aboveground death and the previous observation date. The growth period was the duration from the growth initiation to the senescence. Statistical analyses were same as *P. terminalis*.

Analyses of P_{max} and g_s were conducted in each species by a generalized linear model (GLM) supposing a Gamma error distribution with log link function in which treatment (control vs. warming), season of the measurements (except for *S. japonica* and *T. apetalon*), and their interaction were set as fixed factors, and best-fit model was selected based on AIC. All statistical analyses were conducted using R version 2.10.0 (R Developmental Core Team, http://www.R-project.org).

3. Results

3.1. Environmental conditions

The periods with complete snow cover during wintertime were 111 days in the control plot and 59 days in the warming plot in 2008–2009, and 113 days and 73 days in 2009–2010, respectively (see Fig. A.1). Snow disappeared on 4 April in the control plot and 5 March in the warming plot in 2009, and 12 April and 15 March in 2010, respectively. Thus, understory of the warming plots was not covered by snow one month earlier than the control plots. Furthermore, understory vegetation was occasionally exposed even in mid-winter.

The soil heating system caused slight soil desiccation. Volumetric soil water contents during April to September were 24% (ranging from 13 to 33%) in the control plots, while 20% (ranging from 11 to 28%) in the warming plots in 2009.

3.2. Growth pattern

Statistical results for soil warming impacts on the performance (height growth, survival of overwintering leaves or leaf production) and phenology (growth initiation and termination, leaf expansion and senescence) of individual species are summarized in Table 1.

Pachysandra terminalis – Height growth of this evergreen shrub started in mid-April and lasted until mid-June (Fig. 1). Maximum plant height was larger in the control plots (20.7 ± 0.7 SE cm) than in the warming plots (18.2 ± 0.6 cm) only marginally ($p = 0.06$, Table A.1). Warming treatment did not influence the height growth ($p = 0.17$), while height growth was positively related to ramet size ($p < 0.001$). Interestingly, growth initiation was significantly delayed by two weeks in the warming plots (May 6 and 21 in the control and warming plots, respectively; $p = 0.002$), while size effect on growth initiation was not selected by AIC.

Dryopteris crassirhizoma – Performance of overwintering leaves of this semi-evergreen fern was strongly influenced by soil warming (Fig. 2a). Survival area of overwintering leaves in early April was significantly smaller in the warming plots (17%) than in the control plots (84%, $p < 0.001$; Table A.2). This resulted in earlier disappearance of overwintering leaves in the warming plots (April 13) than in the control plots (May 23, $p = 0.01$). Time of current leaf expansion was seven days earlier in the warming plots (May 16) than the control plots (May 23, $p = 0.005$) because height growth of frond started one week earlier in the warming plots (Fig. 2b). However, the time of leaf prostration in autumn was similar between the treatments (October 18 and 23 in the control and warming plots, respectively, $p = 0.10$). Disappearance of overwintering leaves occurred just at the same time of leaf expansion in the control plots. However, overwintering leaves of the warming plots disappeared 28 days before leaf expansion of current leaves even when leaf expansion of the warming plants occurred one week earlier. Therefore, the leaf habit changed from semi-evergreen to deciduous by soil warming (see also Fig. A.1). Plant height in summer was marginally smaller in the warming plots (53.9 ± 3.2 cm) than in the control plots (68 ± 2.4 cm, $p = 0.08$; Table A.2). Size effects on leafing phenologies of *D. crassirhizoma* were excluded by AIC from every analysis.

Daphne kamtschatica – Leaf production of this summer-deciduous shrub started in late August and continued until late October. After overwintering, several new leaves were added in early May, but all leaves were shed by mid-June (Fig. 3). Therefore, the leafless period was about two months in summer. There were no significant differences in leaf production in autumn (16.5 ± 0.7 and 16.2 ± 0.8 in the control and warming plots, respectively, $p = 0.82$) and the number of spring leaves (sum of overwintered and additional leaves) between the treatments (20.7 ± 0.8 and 19.1 ± 0.9 in the control and warming plots, respectively, $p = 0.23$; Table A.3). Time of leaf abscission in early summer was six days earlier in the warming plots (Jun 14 and 8 in the control and warming plots, respectively, $p = 0.03$), while leaf emergence in autumn occurred at the same time between the treatments (August 30, $p = 0.89$). Size effects on the leafing phenologies of *D. kamtschatica* were excluded by AIC from every analysis. Therefore, soil warming accelerated the leaf shedding in early summer independent of plant size.

Summer-green herbs – Growth patterns of four summer-green herbaceous species are shown in Figure 4. Maximum plant height, growth initiation and senescence times, and growth season length were compared between the control and warming plots. Three of four species (*T. apetalon*, *P. auriculata*, and *P. leptostachya*) did not indicate any significant warming effect for these growth variables ($p > 0.05$; Table A.4). Only *S. japonica*, that had the earliest growth initiation and termination among summer-green species, showed smaller plant height (13.7 ± 0.9 cm and 11.7 ± 0.5 cm in the control and warming plots, respectively, $p = 0.04$) and acceleration of growth initiation (April 23 and 14 in the control and warming plots, respectively, $p < 0.0001$) in the warming plots, while senescence time and growth period did not differ between the treatments ($p > 0.10$). In contrast, size effects on growth schedule were common over species in which larger plants tended to show slower growth initiation (for every species) and later growth termination (for three species except for *P. leptostachya*; Table A.4).

3.3. Photosynthetic activity and stomatal conductance

Effects of the warming treatment were excluded from the best-fit GLM model based on AIC in both P_{max} and g_s in *P. terminalis* (Table A.5). Significant seasonal trend was detected in both P_{max} and g_s, they tended to increase from spring to summer, while decrease from late autumn to early spring, i.e. before and after overwintering (Table 2).

P_{max} of *D. crassirhizoma* decreased after overwintering (Table 2) and this trend was stronger in the warming plots (significant treatment x season interaction; $p = 0.003$, Table A.5). Thus, soil warming decreased the photosynthetic activity of overwintering leaves. In contrast, the warming treatment was not selected in g_s by AIC. Stomatal conductance was the highest in late May soon after leaf emergence then decreased in late summer and remained low values after that.

P_{max} and g_s of *D. kamtschatica* were the largest in autumn and the smallest in early spring soon after overwintering (Table 2). Soil warming positively influenced the P_{max} but only marginally ($p = 0.08$) and g_s significantly ($p = 0.02$; Table A.5).

The warming treatment was not selected by AIC or not significantly ($p = 0.10$) influenced P_{max} and g_s in *S. japonica* and *T. apetalon* (Table A.5). The warming treatment was not selected in both P_{max} and g_s in *P. auriculata*. In contrast, *P. leptostachya* significantly increased P_{max} in the warming plots ($p = 0.02$). Both P_{max} and g_s tended to decrease from spring to summer in both species (Table 2).

4. Discussion

In the understory, most changes in phenology and photosynthetic activity were concentrated in spring or early summer. Furthermore, leafing phenology of plants with overwintering leaves were highly responsive to soil warming. The causes and consequences of the changes in leafing phenology and physiology of understory plants under warmer climate are discussed here. Especially, we show new responses to experimental warming of plants in the understory and confirm the advanced leaf-out reported earlier (Farnsworth et al., 1995; De Frenne et al., 2010; Rollinson and Kaye, 2012).

4.1. Earlier leaf senescence of overwintering leaves

The leaf senescence of *D. crassirhizoma* and *D. kamtschatica* under normal conditions occurred in mid-May and mid-June, respectively (Tables A.2 and A.3). In the month prior to leaf senescence, leaves of *D. crassirhizoma* had the lowest photosynthetic values of the year, whereas *D. kamtschatica* had reasonably high values (Table 2). This suggests different mechanisms might be employed that determine the earlier leaf senescence. In *D. crassirhizoma*, the earlier snowmelt might have led to exposure to freezing temperature under strong irradiation that could have damaged the leaves (e.g. Skillman et al., 1996; Taulavuori et al., 2011). Accordingly, photosynthetic capacity was negligible in early spring in the warming plots, whereas in the control plots, photosynthetic capacity was also low but still maintained at about half of the maximum values recorded during the year. Tani and Kudo (2005) reported that overwintering leaves in *D. crassirhizoma* had both photosynthetic and resource storage functions by shading and defoliation experiments during two years. Besides the photosynthetic potential loss (Karlsson, 1985), the early senescence also might have removed the storage potential of such leaves (Shaver, 1981; Jonasson, 1989) in the warming plots. Actually, the early senescence was not followed by a similar advance in leaf-out and thus the leaf habit of *D.*

crassirhizoma changed from semi-evergreen to deciduousness what might have strongly decreased the effectiveness of storage. This might have resulted in the smaller plant size in the warming plots.

Although earlier snowmelt was suggested to be unfavorable for *D. crassirhizoma*, it might have extended the active photosynthetic period for *D. kamtschatica*. Lapointe (2001) demonstrated that leaf senescence of spring ephemerals occurred when plants stored a certain level of photosynthetic carbohydrate rather than the decrease in light intensity under a canopy. Similarly, acceleration of spring photosynthetic carbon gain in *D. kamtschatica* might have resulted in the earlier leaf shedding in the warming plots (Table A.3). Thus carbon balance control could explain the leafing period for this species (Chabot and Hicks, 1982; Kikuzawa, 1991). Accordingly, there were no significant differences in leaf production between the treatments (Fig. 3) and the warming impact on the growth of *D. kamtschatica* may be neutral.

4.2. Earlier leaf-out in spring

With soil warming, *D. crassirhizoma* leafed out one week and *S. japonica* 1.5 weeks earlier in spring, respectively (Tables A.2 and A.4). Under warm conditions, leaf-out of *D. crassirhizoma* occurred at the same time (mid-May) as the initiation of canopy closure (Fig. A.1, Nakaji et al., 2011). Thus, earlier leaf-out under warm conditions might be restricted to occur after canopy cover for *D. crassirhizoma*. Because *S. japonica* leafed out well before canopy closure (mid- to late April), leaves were exposed to high light conditions longer under warm soil conditions. Irrespective of the early leaf-out in *S. japonica*, the growing season length was not different significantly between the treatments (Table A.4). Similar results were shown in an arctic herb, *Polygonum bistorta,* which showed earlier emergence and senescence by soil warming, resulting in no change in photosynthetic period (Starr et al., 2000). These results suggest that the length of growing season may be not extended under warm conditions.

4.3. Later leaf-out in spring

The new shoot growth of *P. terminalis* was delayed by two weeks in the warming plots (Table A.1). Possibly photo-inhibition in early spring (Skillman et al., 1996) delayed the growth initiation when plants in the warming plots were exposed to freezing temperature under strong irradiation without protection by snow cover. Yoshie and Kawano (1986) reported that photosynthetic activity of *P. terminalis* tended to decrease in winter season because of low temperature and water stress. Although the effects of warming treatment were excluded from the best-fit model in our analyses, probably due to large seasonal trends of both P_{max} and g_s, P_{max} values in April were much smaller in the warming plots than in the control plots. The possible negative effects, however, seemed not to be carried over to the new leaves because photosynthetic rates measured in May did not differ between the treatments (Table A.5).

4.4. No change in leafing phenology

For three of the four summer-green herbs, *T. apetalon*, *P. auriculata* and *P. leptostachya*, no changes in leafing phenologies were observed (Table A.4). This indicates that temperature is not the main determinant of phenology in these species. In the previous study conducted in the same forest, Kudo et al. (2008) reported that between-year variations in flowering phenology of summer-blooming herbs were relatively small in comparison with spring-blooming herbs that initiated growth in early spring, suggesting that growth schedule of late leaf-out herbs might be not strongly influenced by climate conditions. Our results are consistent with this prediction also for leafing phenology. Although the soil warming resulted in soil desiccation slightly (4% decrease in water content), P_{max} of *P. leptoschachya* and g_s of *D. kamtschatica* were higher in the warming plots (Table 2), indicating little drought stress in our experimental system. Similarly, phenologies of plants in subalpine (Dunne et al., 2003) and deciduous temperate forests under humid climate (Rollinson and Kaye, 2012) were reported to be not sensitive to soil water conditions.

In the summer deciduous shrub, *D. kamtschatica*, the leafing time and production of autumn leaves were independent of soil warming, whereas soil warming accelerated leaf shedding in early summer. Lei and Koike (1998) showed that the light environment influenced the leafing time and production by comparing this species growing in the shade, forest edge and under artificial shading. As canopy-forming trees (*Quercus crispula*) around the warming plots did not significantly advance the leaf flushing (Fig. A.1; Nakaji et al., 2011), the differences in light environment between the warming and control plots were small. Thus, soil warming did not influence the leafing phenology in autumn of this summer deciduous species.

Little changes in the leafing phenology of canopy trees in our experiment might be because leafing phenology of trees generally responds to air temperature rather than soil temperature (Menzel et al., 2006; Delbart et al., 2008) and/or photoperiod (Körner and Basler, 2010). Several previous studies demonstrated that temperature-dependent canopy flushing may be realized once a critical photoperiod has passed (Parmesan and Yohe, 2003; Root et al., 2003; Menzel et al., 2006; Delbart et al., 2008; Körner and Basler, 2010).

5. Conclusions

The present study revealed that phenological responses to soil warming were generally prominent in spring. However, directions of responses highly varied among species depending on the leaf habit and life history. In summer-green herbs, early leaf-out species are more sensitive to warming probably because growth is more temperature limited than late leaf-out species. Performance and phenology of plants with overwintering leaves could be influenced by the modification of snow conditions caused by soil warming. Intermittent occurrence of snow-less period during winter and too early snow release may cause physiological damage to overwintering leaves resulting in the decrease in photosynthetic activity and subsequent growth performance especially for semi-evergreen species, possible also for evergreen plants, but maybe not for summer-deciduous plants. These results indicate that careful considerations of species composition, leaf habit, and winter climate are crucial for the prediction of climate change impacts on understory vegetation. Many understory plants extensively photosynthesize during the short period from snowmelt to canopy closure (Rothstein and Zak, 2001). Therefore, not only snowmelt regime but also phenological shift of canopy trees should influence the productivity, performance, and population dynamics of understory plants (Lapointe, 2001; Ida and Kudo, 2008, 2009; Rollinson and Kaye, 2012). This indicates the importance of indirect effects of warming temperature in temperate forest ecosystems.

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References

Aerts, R., Cornelissen, J.H.C., Dorrepaal, E., 2006. Plant performance in a warmer world: general responses of plants from cold, northern biomes and the importance of winter and spring events. Plant Ecol. 182, 65–77.

Augspurger, C.K., 2009. Spring 2007 warmth and frost: phenology, damage and refoliation in a temperate deciduous forest. Funct. Ecol. 23, 1031–1039.

Augspurger, C.K., Bartlett, E.A., 2003. Differences in leaf phenology between juvenile and adult trees in a temperate deciduous forest. Tree Physiol. 23, 517–525.

Augspurger, C.K., Cheeseman, J.M., Salk, C.F., 2005. Light gains and physiological capacity of understorey woody plants during phenological avoidance of canopy shade. Funct. Ecol. 19, 537–546.

Bokhorst, S.F., Bjerke, J.W., Tømmervik, H., Callaghan, T.V., Phoenix, G.K., 2009, Winter warming events damage sub-Arctic vegetation: consistent evidence from an experimental manipulation and a natural event. J. Ecol. 97, 1408–1415.

Chabot, B.F., Hicks, D.J., 1982. The ecology of leaf life spans. Ann. Rev. Ecol. Syst.13, 229–259.

De Frenne, P., De Schrijver, A., Graae, B.J., Gruwez, R., Tack, W., Vandelook, F., Hermy, M., Verheyen, K., 2010. The use of open-top chambers in forests for evaluating warming effects on herbaceous understory plants. Ecol. Res. 25, 163–171.

Delbart, N., Picard, G., Le Toan, T., Kergoat, L., Quegan, S., Woodward, I., Dye, D., Fedotova, V., 2008. Spring phenology in boreal Eurasia over a nearly century time scale. Global Change Biol. 14, 603–614.

Dunne, J.A., Harte, J., Taylor, K.J., 2003. Subalpine meadow flowering phenology responses to climate change: integrating experimental and gradient methods. Ecol. Monogr. 73, 69–86.

Farnsworth, E.J., Núñez-Farfán, J., Careaga, S.A., Bazzaz, F.A., 1995. Phenology and growth of three temperate forest life forms in response to artificial soil warming. J. Ecol. 83, 967–977.

Fridley, J.D., 2012. Extended leaf phenology and the autumn niche in deciduous forest invasions. Nature 485, 359–364.

Gill, D.S., Amthor, J.S., Bormann, F.H., 1998. Leaf phenology, photosynthesis, and the persistence of saplings and shrubs in a mature northern hardwood forest. Tree Physiol. 18, 281–289.

Giménez-Benavides, L., Escudero, A., Iriondo, J.M., 2007. Reproductive limits of a late-flowering high-mountain Mediterranean plant along an elevational climate gradient. New Phytol. 173, 367–382.

Hughes, L., 2000. Biological consequences of global warming: is the signal already. Trends Ecol. Evol. 15, 56–61.

Ida, T.Y., Kudo, G., 2008. Timing of canopy closure influences carbon translocation and seed production of an understory herb, *Trillium apetalon* (Trilliaceae). Ann. Bot. 101, 435–446.

Ida, T.Y., Kudo, G., 2009. Comparison of light harvesting and resource allocation strategies between two rhizomatous herbaceous species inhabiting deciduous forests. J. Plant Res. 122, 171–181.

Inouye, D.W., 2000. The ecological and evolutionary significance of frost in the context of climate change. Ecol. Lett. 3, 457–463.

Jonasson, S., 1989. Implications of leaf longevity, leaf nutrient re-absorption and leaf translocation for the resource economy of five evergreen plant species. Oikos 73, 269–271.

Karlsson, P.S., 1985. Photosynthetic characteristics and leaf carbon economy of a deciduous and an evergreen dwarf shrub: *Vaccinium uliginosum* L. and *V. vitis-idaea* L. Holarc. Ecol. 8, 9–17.

Kikuzawa, K., 1989. Ecology and evolution of phenological pattern, leaf longevity and leaf habit. Evol. Trends Plants 3, 105–110.

Kikuzawa, K., 1991. A cost-benefit analysis of leaf habit and leaf longevity of trees and their geographical pattern. Am. Nat. 138, 1250–1263.

Körner, C., Basler, D., 2010. Phenology under global warming. Science 327, 1461–1462.

Kudo, G., Ida. T.Y., Tani, T., 2008. Linkages between phenology, pollination, photosynthesis, and plant reproduction in deciduous forest understory plants. Ecology 89, 321–331.

Lange, O.L., Losch, R., Schulze, E.D., Kappen, L., 1971. Responses of stomata to changes in humidity. Planta 100, 76–86.

Lapointe, L., 2001. How phenology influences physiology in deciduous forest spring ephemerals. Physiol. Plant. 113, 151–157.

Lei, T.T., Koike, T., 1998. Some observations of phenology and ecophysiology of *Daphne kamtschatica* maxim. var. *jezoensi* (Maxim.) Ohwi, a shade deciduous shrub, in the forest of northern Japan. J. Plant Res. 111, 207–212.

Maeno, H., Hiura, T., 2000. The effects of leaf phenology of overstory trees on the reproductive success of an understory shrub, *Staphylea bumalda* DC. Can. J. Bot. 78, 781–785.

Menzel, A., Sparks, T.H., Estrella, N., Koch, E., Aasa, A., Ahas, R., Alm-Kübler, K., Bissolli, P., Braslavska, O., Briede, A., Chmielewski, F.M., Crepinsek, Z., Curnel, Y., Dahl, A., Defila, C., Donnelly, A., Filella, Y., Jatczak, K., Mage, F., Mestre, A., Nordli, Ø., Peñuelas, J., Pirinen, P., Remisova, V., Scheifinger, H., Striz, M., Susnik, A., Van Vliet, A.J.H., Wielgolaski, F.E., Zach, S., Zust, A., 2006. European phenological response to climate change matches the warming pattern. Global Change Biol. 12, 1969–1976.

Nakaji, T., Oguma, H., Hiura, T., 2011. Monitoring of phenology of deciduous broad leaf forest by using compact VIS-NIR multiband camera system. J. Agri. Meteor. 67, 65–74.

Nakamura, M., Muller, O., Tayanagi, S., Nakaji, T., Hiura, T., 2010**.** A new technique of branch warming changes leaf phenology and acorn production in tall mature trees. Agri. Forest Meteor. 150, 1026–1029.

Parmesan, C., 2006. Ecological and evolutionary responses to recent climate change. Ann. Rev. Ecol. Syst. 37, 637–669.

Parmesan, C., Yohe, G., 2003. A globally coherent fingerprint of climate change impacts across natural systems. Nature 421, 37–42.

Richardson, A.D., Black, T.A., Ciais, P., Delbart, N., Friedl, M.A., Gobron, N., Hollinger, D.Y., Kutsch, W.L., Longdoz, B., Luyssaert, S., Migliavacca, M., Montagnani, L., Munger, J.W., Moors, E., Piao, S., Rebmann, C., Reichstein, M., Saigusa, N., Tomelleri, E., Vargas, R., Varlagin, A., 2010. Influence of spring and autumn phenological transitions on forest ecosystem productivity. Phil. Trans. R. Soc. B 365, 3227–3246.

Rollinson, C.R., Kaye, M.W., 2012. Experimental warming alters spring phenology of certain plant functional groups in an early-successional forest community. Global Change Biol. 18, 1108–1116.

Root, T.L., Price, J.T., Hall, K.R., Schneider, S.H., Rosenzweig, C., Pounds, J.A. 2003. Fingerprints of global warming on wild animals and plants. Nature

421, 57–60.

Rothstein, D.E., Zak, D.R., 2001. Photosynthetic adaptation and acclimation to exploit seasonal periods of direct irradiance in three temperate deciduous-forest herbs. Funct. Ecol. 15, 722–731.

Shaver, G.R., 1981. Mineral nutrition and leaf longevity in an evergreen shrub, *Ledum palustre* ssp. *decumbens*. Oecologia 49, 362–365.

Shen, M., 2011. Spring phenology was not consistently related to winter warming on the Tibetan Plateau. Proc. Natl Acad. Sci. USA 108, E91-91.

Skillman, J.B., Strain, B.R., Osmond, C.B., 1996. Contrasting patterns of photosynthetic acclimation and photoinhibition in two evergreen herbs from a winter deciduous forest. Oecologia 107, 446–455.

Starr, G., Oberbauer, S.F., Pop, E.W., 2000. Effects of lengthened growing season and soil warming on the phenology and physiology of *Polygonum bistorta*. Global Change Biol. 6, 357–369.

Tani, T., Kudo, G., 2005. Overwintering leaves of a forest-floor fern, *Dryopteris crassirhizoma* (Dryopteridaceae): a small contribution to the resource storage and photosynthetic carbon gain. Ann. Bot. 95, 263–270.

Taulavuori, K., Bauer, E., Taulavuori, E., 2011. Overwintering stress of *Vaccinium vitis-idaea* in the absence of snow cover. Environ. Exp. Bot. 72, 397–403.

Uemura, S., 1994. Patterns of leaf phenology in forest understory. Can. J. Bot. 72, 409–414.

Yoshie, F., Kawano, S., 1986. Seasonal changes in photosynthetic characteristics of *Pachysandra terminalis* (Buxaceae), an evergreen woodland chamaephyte, in the cool temperate regions of Japan. Oecologia 71, 6–11.

Table 1. Summary of growth and phenological responses to soil warming in each species. Refer Tables A.1–A.4 for statistical results.

Species	Leaf habit	Performance [a]	Spring or early summer phenology [b]	Autumnal phenology [c]
Pachysandra terminalis	Evergreen (EV)	NS	+	not available
Dryopteris crassirhizoma	Semi-evergreen (SE)	–	–	NS
Daphne kamtschatica	Summer deciduous (SD)	NS	–	NS
Smilacina japonica	Summer green (SG)	–	–	NS
Trillium apetalon	Summer green (SG)	NS	NS	NS
Parasenecio auriculata	Summer green (SG)	NS	NS	NS
Phryma leptostachya	Summer green (SG)	NS	NS	NS

–: negative performance or advanced phenological responses, +: positive performance or delayed phenological responses, NS: not significant responses.

[a] height growth (EV, SG), leaf production (SD) or survival of overwintering leaves (SE)

[b] growth initiation (EV, SG), leaf-out time (SE) or senescence time of overwintered leaves (SE, SD)

[c] growth termination (SG), leaf-out time (SD) or leaf prostration time (SE). Growth termination of *S. japonica* and *T. apetalon* occurred in summer but included in autumnal phenology here.

Table 2. Seasonal transition of maximum photosynthetic rate (P_{max}; μ mol m^{-2} s^{-1}) and stomatal conductance (g_s: m mol m^{-2} s^{-1}) of target species between control and soil-warming plots. Mean ± SE (sample size). Refer Table A.5 for statistical results.

Species*	Trait	Treatment	Early Spring	Late Spring	Late Summer	Late Autumn
Pachysandra terminalis	P_{max}	Control	3.7±0.33 (7)	5.2±0.35 (6)	5.2±0.25 (9)	4.3±0.39 (6)
		Warming	2.6±0.60 (10)	5.2±0.65 (6)	6.1±0.94 (9)	4.2±0.26 (7)
	g_s	Control	49±4.6 (7)	74±11.6 (6)	145±13.1 (9)	51±6.7 (6)
		Warming	46±8.3 (10)	66±9.7 (6)	161±26.0 (9)	53±5.4 (7)
Dryopteris crassirhizoma	P_{max}	Control	1.4±0.19 (9)	3.0±0.35 (6)	2.6±0.15 (6)	3.4±0.23 (6)
		Warming	0.8±0.19 (7)	3.8±0.29 (6)	2.4±0.23 (6)	3.4±0.23 (6)
	g_s	Control	43±3.7 (9)	75±10.8 (6)	40±4.3 (6)	52±3.7 (6)
		Warming	50±5.8 (9)	60±7.1 (6)	35±6.1 (6)	38±5.5 (6)
Daphne kamtschatica	P_{max}	Control	4.3±0.43 (10)	6.9±0.75 (12)	—	9.1±0.61 (7)
		Warming	4.9±0.38 (8)	7.9±0.76 (10)	—	10.4±0.49 (7)
	g_s	Control	36±3.4 (10)	88±9.1 (12)	—	182±39.1 (7)
		Warming	44±4.6 (8)	113±12.0 (10)	—	220±37.7 (7)
Smilacina japonica	P_{max}	Control	—	5.4±0.62 (6)	—	—
		Warming	—	6.4±0.84 (6)	—	—
	g_s	Control	—	148±21.3 (6)	—	—
		Warming	—	101±15.6 (6)	—	—
Trillium apetalon	P_{max}	Control	—	5.8±0.60 (6)	—	—
		Warming	—	6.4±0.50 (6)	—	—
	g_s	Control	—	93±13.0 (6)	—	—
		Warming	—	107±2.3 (6)	—	—
Parasenecio auriculata	P_{max}	Control	—	5.3±0.70 (6)	2.3±0.12 (6)	—
		Warming	—	5.1±0.52 (6)	2.4±0.10 (6)	—
	g_s	Control	—	92±12.7 (6)	82±14.0 (6)	—
		Warming	—	102±10.8 (6)	72±7.7 (6)	—
Phryma leptostachya	P_{max}	Control	—	4.3±0.18 (6)	2.4±0.12 (6)	—
		Warming	—	5.5±0.61 (6)	2.7±0.19 (6)	—
	g_s	Control	—	81±9.6 (6)	53±7.8 (6)	—
		Warming	—	86±10.4 (6)	47±5.0 (6)	—

Figure captions

Fig. 1 Seasonal transitions of height increment of an evergreen shrub, *Pachysandra terminalis* in the control (open) and warming plots (filled) expressed as Julian calendar date. Plant height at the first observation time (April 5 = 95) was set as a base point for individual plants. Box plots indicate 25, 50 and 75 percentile and whiskers indicate 10 and 90 percentile of data distributions. Refer Table A.1 for statistical results.

Fig. 2 Seasonal transitions of the proportion of living area of overwintering leaves (a) and seasonal transitions of plant height (b) of a semi-evergreen fern, *Dryopteris crassirhizoma* in the control (open) and warming plots (filled). Refer caption of Fig.1 for details, and Table A.2 for statistical results.

Fig. 3 Seasonal transitions of leaf number per stem of a summer-deciduous shrub, *Daphne kamtschatica* in the control (open) and warming plots (filled). Refer caption of Fig.1 for details, and Table A.3 for statistical results.

Fig. 4 Seasonal transitions of plant height of four summer-green herbaceous species (a–d) in the control (open) and warming plots (filled). Refer caption of Fig.1 for details, and Table A.4 for statistical results.

Fig.1 (Ishioka et al.)

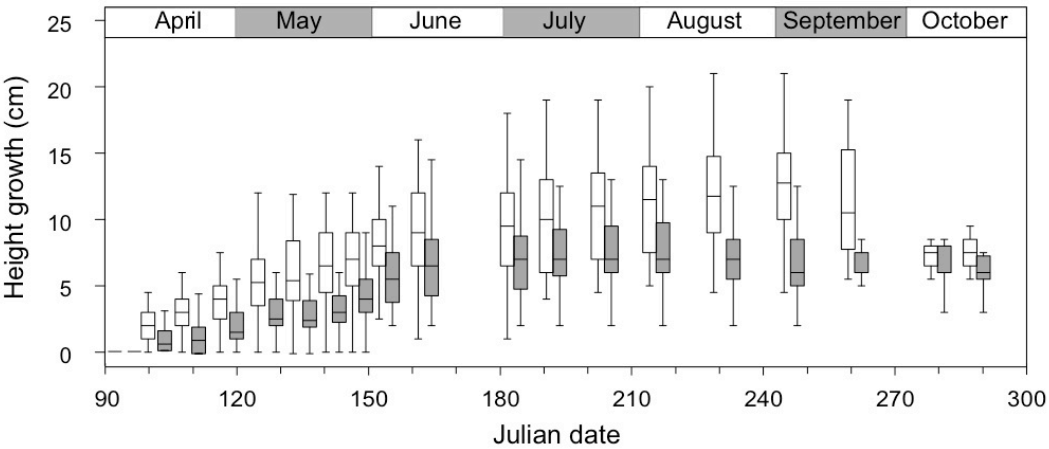


Fig. 2 (Ishioka et al.)

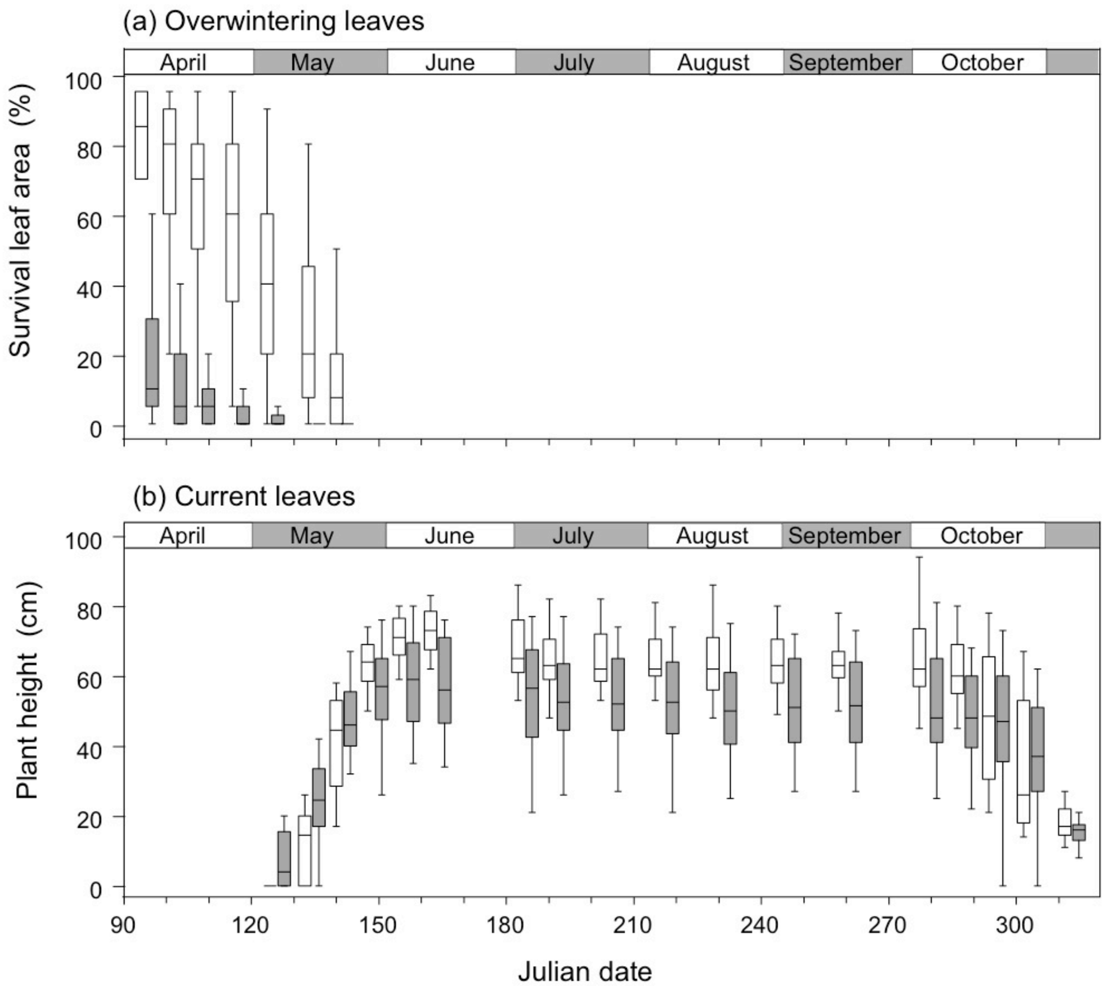


Fig. 3 (Ishioka et al.)

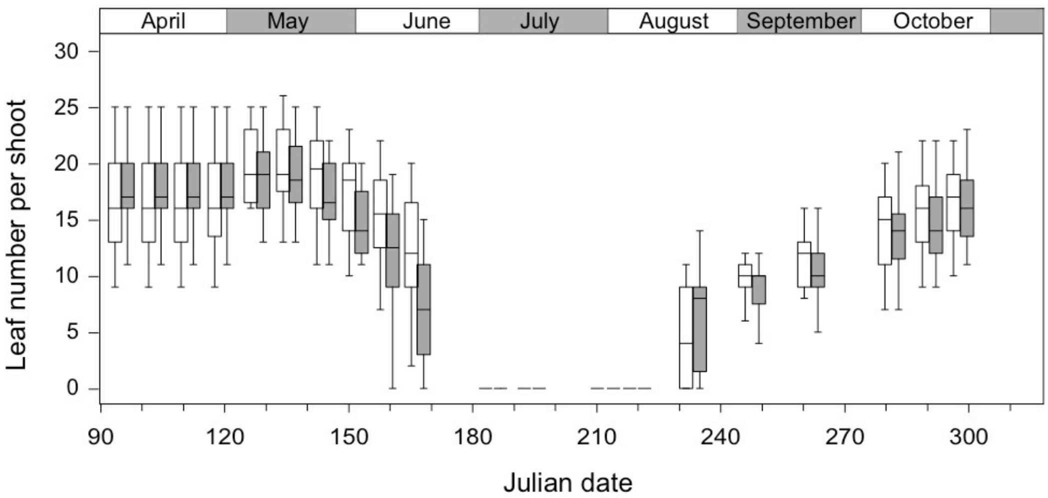
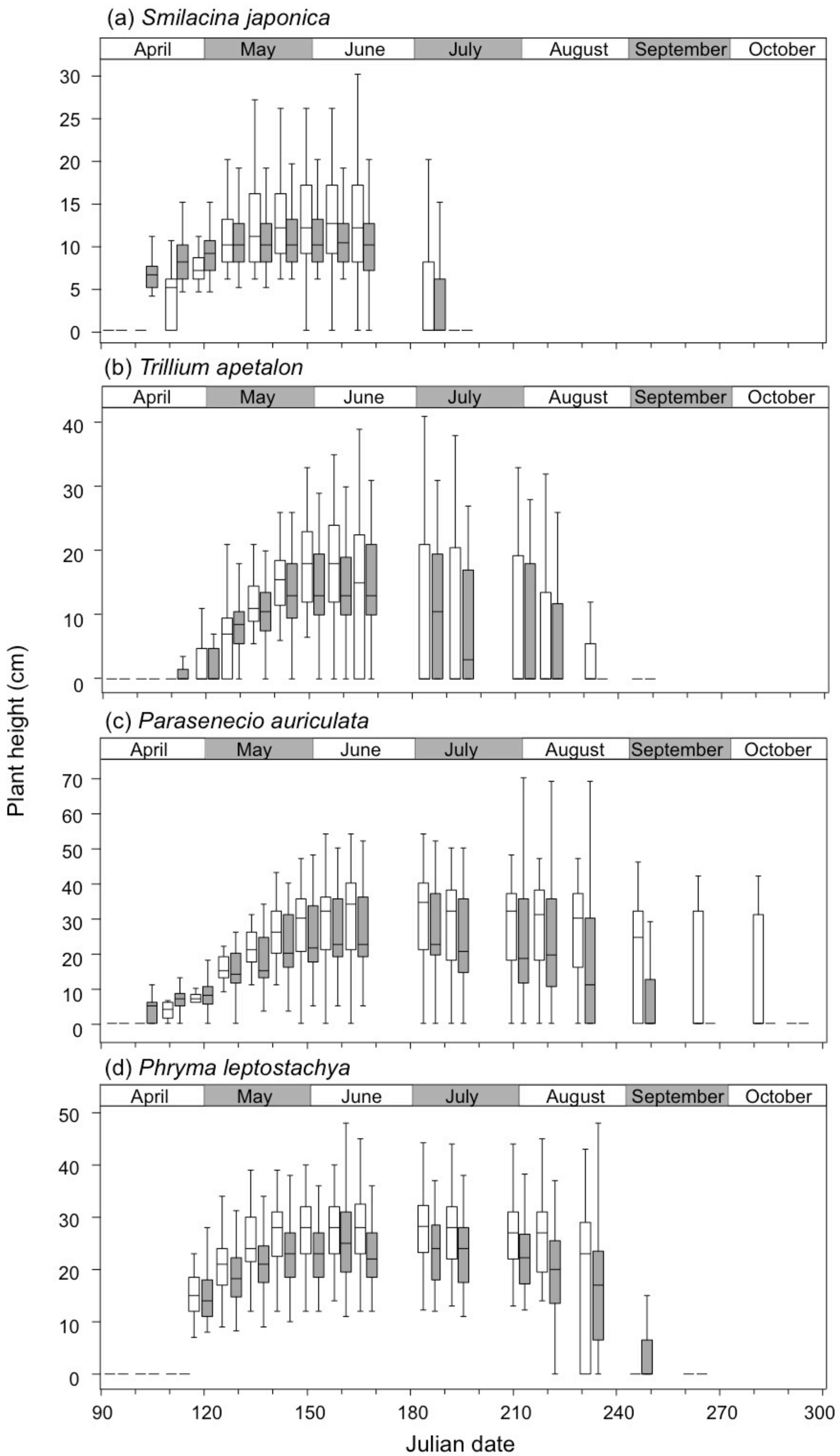


Fig. 4 (Ishioka et al.)



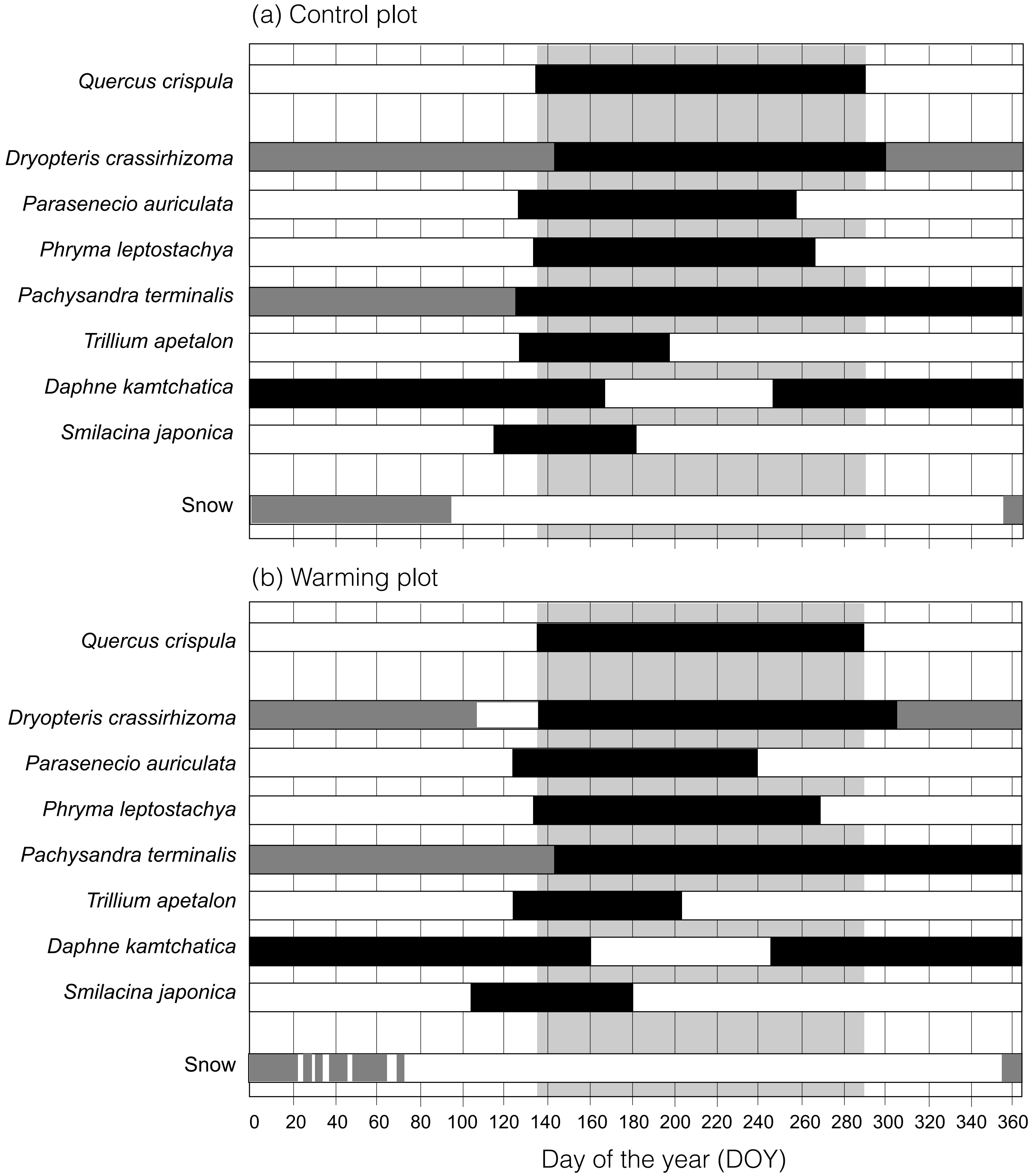


Fig. A.1. Leaf onset and senescence time of a dominant canopy tree (*Quercus crispula*) and understory species, and duration of snow cover in the control (a) and soil warming plots (b) based on the analyses of color photographs by an automatic-shooting camera. Black bars indicate when leaves are present, open bars when leaves absent, and grey bars either prostrating leaves (*Dryopteris crassirhizoma*), overwintering leaves without new leaves (*Pachysandra terminalis*) or snow cover. Data from Nakaji et al. (2011).

Table A.1. Effects of soil warming on the maximum plant height, annual height increment, and growth initiation time in an evergreen shrub, *Pachysandra terminalis*. Significant levels and *t* values by LMM are shown in which fixed factors were treatment and plant size (except for max. height). Mean ± SE, sample size is shown in parenthesis.

Treatment	Max. height (cm)	Height increment (cm)	Growth initiation date
Control	20.7 ± 0.7 (46)	9.9 ± 0.6 (46)	May 6 ± 1.7 (43)
Warming	18.2 ± 0.6 (47)	7.4 ± 0.5 (47)	May 21 ± 1.6 (36)
Warming effect	$P = 0.06$, $t_6 = -2.3$	$P = 0.17$, $t_6 = -1.5$	**$P = 0.002$**, $t_6 = 4.7$
Size effect	—	**$P < 0.001$**, $t_{84} = 9.1$	not selected

Table A.2. Effects of soil warming on the maximum plant height, survival rate of overwintering leaves (percentage in living arae in early April), time of disappearance of overwintering leaves, time of current leaf expantion, and time of leaf prostration in a semi-evergreen fern, *Dryopteris classirhizoma*. Significant levels and *t* values by LMM are shown in which fixed factor was treatment, while effects of plant size were excluded by AIC. Mean ± SE, sample size is shown in parenthesis.

Treatment	Max. height (cm)	Overwintering leaf survival rate (%)	Senescence of overwintering leaves	Leaf expansion in spring	Leaf prostration in autumn
Control	68.2 ± 2.4 (20)	83.5 ± 2.5 (20)	May 23 ± 2.1 (20)	May 23 ± 1.0 (20)	Oct. 18 ± 1.3 (20)
Warming	53.9 ± 3.2 (20)	17.3 ± 4.3 (20)	Apr. 13 ± 3.9 (20)	May 16 ± 1.2 (20)	Oct. 23 ± 1.2 (18)
Warming effect	$P = 0.06$, $t_6 = -2.3$	**$P < 0.001$**, $t_6 = -6.8$	**$P = 0.01$**, $t_6 = -3.7$	**$P = 0.005$**, $t_6 = -4.3$	$P = 0.10$, $t_6 = -1.9$

Table A.3. Effects of soil warming on the number of overwintering leaves, spring leaf production, autumnal leaf production, time of leaf shadding in early summer, and time of leaf-out in autumn in a summer-deciduous shrub, *Daphne kamtschatica*. Significant levels and *t* values by LMM are shown in which fixed factor was treatment, while effects of plant size were excluded by AIC. Mean ± SE, sample size is shown in parenthesis.

Treatment	No. of spring leaves	No. of autumnal leaves	Leaf shadding date in early summer	Leaf-out date in autumn
Control	20.7 ± 0.8 (20)	16.5 ± 0.7 (21)	June 14 ± 1.3 (20)	Aug. 30 ± 2.4 (21)
Warming	19.1 ± 0.9 (17)	16.2 ± 0.8 (23)	June 8 ± 1.6 (16)	Aug. 30 ± 2.4 (22)
Warming effect	P = 0.23, t_6 = –1.3	P = 0.82, t_6 = –0.24	***P* = 0.03**, t_6 = –2.8	P = 0.89, t_6 = –0.14

Table A.4. Effects of soil warming on the maximum plant height, time of growth initiation, time of growth termination, and the length of growth season in four summer-green herbaceous species. Significant levels and *t* values by LMM are shown in which fixed factors were treatment and plant size (except for max. height). Mean ± SE, sample size is shown in parenthesis.

Treatment	Max. height (cm)	Growth initiation	Growth termination	Growing season period (d)
Smilacina japonica				
Control	13.7 ± 0.9 (42)	Apr. 24 ± 1.0 (42)	June 27 ± 3.7 (42)	67 ± 3.6 (42)
Warming	11.7 ± 0.5 (46)	Apr. 13 ± 0.5 (46)	June 26 ± 3.1 (46)	75 ± 3.0 (46)
Warming effect	**P = 0.04**, $t_6 = -2.5$	**P = 0.002**, $t_6 = -5.5$	$P = 0.96$, $t_6 = 0.05$	$P = 0.13$, $t_6 = 1.7$
Size effect	—	**P <0.0001**, $t_{79} = 5.3$	**P = 0.01**, $t_{79} = 2.6$	$P = 0.08$, $t_{79} = 1.7$
Trillium apetalon				
Control	19.1 ± 8.3 (36)	May 8 ± 1.4 (36)	July 13 ± 7.2 (36)	66 ± 6.7 (36)
Warming	15.7 ± 1.2 (39)	May 5 ± 1.4 (39)	July 19 ± 6.6 (39)	77 ± 5.8 (39)
Warming effect	$P = 0.35$, $t_6 = -1.0$	$P = 0.59$, $t_6 = -0.5$	$P = 0.20$, $t_6 = 1.5$	$P = 0.10$, $t_6 = 1.9$
Size effect	—	**P <0.0001**, $t_{66} = 5.4$	**P <0.0001**, $t_{66} = 5.0$	**P <0.0001**, $t_{66} = 4.4$
Parasenecio auriculata				
Control	35.3 ± 2.7 (16)	May 7 ± 1.3 (16)	Sep. 23 ± 3.2 (16)	130 ± 6.4 (16)
Warming	29.8 ± 2.4 (35)	May 4 ± 0.9 (35)	Sep. 10 ± 6.9 (35)	114 ± 3.2 (35)
Warming effect	$P = 0.35$, $t_4 = -1.1$	$P = 0.73$, $t_4 = -0.4$	$P = 0.20$, $t_4 = -1.3$	$P = 0.09$, $t_4 = -2.3$
Size effect	—	**P <0.0001**, $t_{42} = 4.7$	**P = 0.01**, $t_{42} = 2.6$	not selected
Phryma leptostachya				
Control	29.9 ± 1.0 (47)	May 14 ± 0.3 (47)	Sep. 18 ± 2.5 (47)	131 ± 2.6 (47)
Warming	26.5 ± 1.3 (39)	May 13 ± 0.4 (39)	Sep. 20 ± 3.5 (39)	134 ± 3.5 (39)
Warming effect	$P = 0.36$, $t_6 = -1.0$	$P = 0.58$, $t_6 = -0.6$	$P = 0.67$, $t_6 = 0.4$	$P = 0.52$, $t_6 = 0.7$
Size effect	—	**P = 0.003**, $t_{77} = 3.1$	not selected	not selected

Table A.5. Results of GLM for maximum photosynthetic rate (P_{max}) and stomatal conductance (g_s) of target species in which treatment (control vs. soil warming), season of the measurements and their interaction are included as explanatory variables. Best-fit model based on AIC is shown in each species. Refer Table 2 for measurement values.

(1) *Pachysandra terminalis*

P_{max}	Coef.	SE	*t* value	*P* value
Intercept (late autumn)	1.44	0.10	13.9	**<0.001**
Soil warming	not selected			
Early spring	-0.33	0.14	-2.39	**0.020**
Late spring	0.21	0.15	1.42	0.16
Late summer	0.29	0.14	2.14	**0.037**

g_s	Coef.	SE	*t* value	*P* value
Intercept	-2.96	0.11	-27.8	**<0.001**
Soil warming	not selected			
Early spring	-0.10	0.14	-0.67	0.50
Late spring	0.30	0.15	1.97	0.054
Late summer	1.08	0.14	7.75	**<0.001**

(2) *Dryopteris crassirhizoma*

P_{max}	Coef.	SE	*t* value	*P* value
Intercept (late spring)	1.08	0.14	7.79	**<0.001**
Soil warming	0.24	0.20	1.24	0.22
Late summer	-0.12	0.20	-0.59	0.56
Late autumn	0.13	0.20	0.66	0.51
Early spring	-0.75	0.18	-4.21	**<0.001**
Warming x L. summer	-0.34	0.28	-1.22	0.23
Warming x L. autumn	-0.23	0.28	-0.83	0.41
Warming x E. spring	-0.81	0.26	-3.10	**0.003**

g_s	Coef.	SE	*t* value	*P* value
Intercept	-2.70	0.09	-29.9	**<0.001**
Soil warming	not selected			
Late summer	-0.59	0.13	-4.60	**<0.001**
Late autumn	-0.40	0.13	-3.14	**0.003**
Early spring	-0.38	0.12	-3.19	**0.003**

(3) *Daphne kamtschatica*

P_{max}	Coef.	SE	*t* value	*P* value
Intercept (late autumn)	2.20	0.09	25.7	**<0.001**
Soil warming	0.14	0.08	1.81	0.077
Early spring	-0.75	0.10	-7.36	**<0.001**
Late spring	-0.27	0.10	-2.77	**0.008**

g_s	Coef.	SE	*t* value	*P* value
Intercept	-1.73	0.11	-15.4	**<0.001**
Soil warming	0.26	0.10	2.53	**0.015**
Early spring	-1.63	0.13	-12.1	**<0.001**
Late spring	-0.67	0.13	-5.17	**<0.001**

(4) *Smilacine japonica*

P_{max}	Coef.	SE	*t* value	*P* value
Intercept	1.78	0.09	20.3	**<0.001**
Soil warming	not selected			

g_s	Coef.	SE	*t* value	*P* value
Intercept	-1.91	0.15	-12.8	**<0.001**
Soil warming	-0.38	0.21	-1.80	0.10

(5) *Trillium apetalon*

P_{max}	Coef.	SE	*t* value	*P* value
Intercept	1.81	0.06	29.1	**<0.001**
Soil warming	not selected			

g_s	Coef.	SE	*t* value	*P* value
Intercept	-2.30	0.07	-34.6	**<0.001**
Soil warming	not selected			

(6) *Parasenecio auriculata*

P_{max}	Coef.	SE	*t* value	*P* value
Intercept (late spring)	1.65	0.06	27.1	**<0.001**
Soil warming	not selected			
Late summer	-0.80	0.09	-9.31	**<0.001**

g_s	Coef.	SE	*t* value	*P* value
Intercept	-2.34	0.09	-25.1	**<0.001**
Soil warming	not selected			
Late summer	-0.23	0.13	-1.77	0.091

(7) *Phryma leptostachya*

P_{max}	Coef.	SE	*t* value	*P* value
Intercept (late spring)	1.49	0.06	23.2	**<0.001**
Soil warming	0.18	0.07	2.49	**0.021**
Late summer	-0.66	0.07	-8.93	**<0.001**

g_s	Coef.	SE	*t* value	*P* value
Intercept	-2.48	0.09	-29.0	**<0.001**
Soil warming	not selected			
Late summer	-0.51	0.12	-4.23	**<0.001**