



Title	Effects of snow manipulation on larch trees in the taiga forest ecosystem in northeastern Siberia
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Citation	北海道大学. 博士(環境科学) 甲第14765号
Issue Date	2022-03-24
DOI	10.14943/doctoral.k14765
Doc URL	<a href="http://hdl.handle.net/2115/85777">http://hdl.handle.net/2115/85777</a>
Type	theses (doctoral)
File Information	SHAKHMATOV_Ruslan.pdf



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令和3年度博士論文

Effects of snow manipulation on larch trees in the taiga forest  
ecosystem in northeastern Siberia

(積雪量変化が北東シベリアのカラマツタイガ林生態系に及ぼす影響)

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地球圏科学専攻 大気海洋化学・環境変遷学コース

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2022年2月

## **Abstract**

Changes in winter precipitation (snow) may greatly affect vegetation by altering hydrological and biochemical processes. To understand the effects of changing snow cover depth and melt timing on the taiga forest ecosystem, a snow manipulation experiment was conducted in December 2015 at the Spasskaya Pad experimental larch forest in Eastern Siberia, which is characterized by a continental dry climate with extreme cold winters and hot summers. Variables including soil temperature and moisture, oxygen and hydrogen isotope ratios of soil moisture and stem water, foliar nitrogen and carbon contents and their isotopes, phenology, and soil inorganic nitrogen were observed at snow removal (SNOW-), snow addition (SNOW+), and CONTROL plots. After snow manipulation, the soil temperature at the SNOW- plot decreased significantly compared to the CONTROL and SNOW+ plots. At SNOW- plot, snowmelt was earlier and soil temperature was higher than at other plots during spring because of low soil moisture caused by less snowmelt water. Despite the earlier snowmelt and higher soil temperature in the SNOW- plot in the early growing season, needle elongation was delayed. Leaf chemistry also differed between the CONTROL and SNOW- plots. The needle nitrogen content in the SNOW- plot was lower in the middle of July, whereas no difference was observed among the three plots in August. The soil inorganic nitrogen content of each plot corresponded to these results. The amount of soil ammonium was lower in the SNOW- plot than in the other plots at the end of July, however, once mineralization started at the end of August, the amount of soil ammonium in the three plots was comparable. Extremely low soil temperatures in winter and freeze-thaw

cycles in spring and dry soil condition in spring and early summer at the SNOW- plot may have influenced the phenology and mineralization of soil inorganic nitrogen.

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# Chapter 1 Snow cover parameters and its change in Northern Hemisphere

## 1-1 Introduction

Snow cover on soils is a very important factor for boreal ecosystems of the Northern Hemisphere. Snow cover insulates soil from cold air spells during winter, provides sufficient water content and regulates growing season start and duration. The ongoing global climate change is affecting snow cover on global scale. The increase in air temperature has demonstrated a negative impact on the amount of snow both by reducing the percentage of precipitation that falls as solid precipitation (snow) and increasing liquid precipitation (rain) events, and by forcing snowmelt, whereas the increase in winter solid precipitation is naturally expected to have an opposite effect, therefore the change in snow cover whether in positive or negative direction depends on both air temperature and precipitation (Raisanen, 2008). Park et al. (2013) suggests that declining sea ice also plays role in snow cover depth change, especially during snow onset phase. The increase in snow cover depth causes the increase in soil temperature and promotes permafrost degradation (Park et al., 2015), which will have an impact on larch taiga forests of Eastern Siberia, where the degradation of permafrost will have an implication for the shift from larch dominated ecosystem to pine and birch dominated ecosystem (Zhang et al., 2011). Snow cover extent, duration, depth and snow water equivalent (SWE) are snow parameters which are extensively measured and the long-term changes in these parameters globally and regionally will have major consequences for ecosystem function, anthropogenic use of snow resources, and the climate itself through feedback mechanisms (Sturm et al., 2010).



## 1-2 Snow cover parameters

### *1-2-1 Extent*

The snow cover extent significantly decreased in the northern hemisphere at estimated scale of  $3.1 \times 10^6 \text{ km}^2 (100 \text{ yr})^{-1}$  associated with increase in air temperature of  $1.26^\circ\text{C} (100 \text{ yr})^{-1}$  over  $40^\circ\text{--}60^\circ \text{ N}$  from 1915 to 1997 (Brown, 2000). In recent years, higher decrease was observed in spring and summer ( $-0.54$  and  $-0.71$  million  $\text{km}^2$  per decade, respectively) coupled with higher warming rates during these seasons from 1967-2018 (Wu et al., 2020). Future projections in snow cover extent are expected to proceed following the declining trend with linear trend corresponding to the increase in global surface air temperatures at hemispherical level, whereas regional changes are expected to vary depending on vegetation and topography (Mudryk et al., 2020).

### *1-2-2 Duration*

The duration of snow cover period is also changing in the Northern Hemisphere (Liston and Hiemstra, 2011). The decreasing trend in snow cover duration in the Northern Hemisphere is drastically different between autumn (melt) and spring (onset) periods. During autumn the decrease in duration was  $-0.49$  days per decade, whereas in spring the decrease was much higher at  $-3.43$  days per decade (Callaghan et al., 2012). The largest decrease in snow cover duration was observed in a zone, where seasonal mean air temperatures were between  $-5^\circ$  and  $+5^\circ\text{C}$  for the 1966-2007 (Brown and Mote, 2009).

Recent extensive analysis of snow cover duration trends in Northern Eurasia showed statistically significant long-term variations from 1966 to 2012: autumn onset of snow cover was delayed by approximately 1 day per decade, spring snowmelt was advanced by about 1 day per decade (Zhong et al., 2021). For Russia, as a whole, similar decreasing trends were observed with the first snowfall occurring later and spring snowmelt, on the other hand, starting earlier, and the largest decrease was observed in southern Siberia, especially regions of Altai, Sayan and Transbaikal regions ( $-4$  to  $-6$  days per decade), while Far Eastern and North Eastern coastal regions were experiencing marginally increasing trends due to proximity to the ocean (Bulygina et al., 2009). In Yakutsk region, the first day of snowfall in autumn showed a decreasing trend for the period from 1960 to 2018, with higher decrease from 1990s to late 2010s and decreasing variability (Fig. 1a), while the last day of snow in spring showed only slight increasing trend ( $R^2=0.042$ ) (Fig. 1b). However, the first day of continuous snow cover did not show any trend (Fig. 2a), whereas the last day of continuous snow cover showed slight increasing trend ( $R^2=0.106$ ), with earlier snow disappearance in the last 10 years (Fig. 2b), due to high air temperatures during spring and earlier transition from snow to rain events.

### *1-2-3 Depth and snow water equivalent*

Changes in snow depth and snow water equivalent show different trends depending on location, with strong polarizing responses between North America and northern Eurasia. Snow depth and snow water equivalent shown decreasing trend in North America, for example the mean decreasing trend for December–April is  $-0.4$  to  $-0.5$  mm/yr<sup>-1</sup> in snow

water equivalent, or about 5–8 cm in snow cover depth for the period from 1979 to 2007 (Gan et al., 2013). Regional exception in the Northern America is northern Quebec, where reconstructed data shows an increase in SWE (Begin, 2000). In Northern Eurasia, however, both annual mean and annual maximum snow depth increased during the period from 1966 to 2012 at a rate of 0.2 and 0.6 cm per decade, respectively, and the increase is higher in regions north of 50° N (Zhong et al., 2018). In Yakutsk region, maximum snow cover depth showed high variability from 1960 to 2018 with slight increasing trend, and considerable increase from 2010 to 2018 with less variability, even though mean annual air temperature showed an increasing trend, however the region is characterized by extremely low annual air temperature, therefore an increase in air temperature did not negatively impact maximum snow cover depth during extremely cold winter (Fig.3).

## **1-3 Effect of snow cover change on ecosystem**

### *1-3-1 Soil temperature*

Snow is an effective insulating material against heat transfer. Snow protects soil from freezing during seasons with negative air temperatures. Increase in snow cover depth is associated with increase in soil temperature in high latitudinal regions, while on the southern boundary of snow cover extent increase in snow cover may negatively impact soil temperatures due to overall higher air temperatures.

Snow depth increase in 5-15 cm is roughly correspondent to 1 °C increase in surface soil temperature (Zhang, 2005). Global climatic models suggest that increasing

snowfall may account for 16% of total soil warming at 1 m depth (Lawrence and Slater, 2010). In Russia both positive trend in air temperature (0.11 °C decade) and snow cover depth (0.43 cm decade) resulted in increase in soil temperature at 3.6 m depth (0.17 °C decade) and the discrepancy between higher increase in soil temperature than air temperature suggest the decoupling effect of snow cover depth, namely the higher soil temperature comparatively to air temperature due to insulation effect of snow (Park et al., 2014).

Frauenfeld et al. (2004) observed 20 cm increase in active layers depth across 242 meteorological stations in Russia from 1956 to 1990. He argues such increase might be the result of an increase in soil moisture during previous year via increase in snow cover depth, which allows deeper soil thaw, but increasing air temperature during the same period introduces uncertainty.

### *1-3-2 Soil moisture*

Soil moisture is significantly affected by snow cover. High snow cover depth is usually associated with high soil moisture availability. Whereas low snow cover depth and early snow melt may result in decrease in soil moisture availability and even result in drought condition (Iwata et al., 2010, Musselman et al., 2017). Other factors such as long period of frozen soil is associated with low soil moisture, since snow melt water cannot permeate frozen soil and results in runoff (Takata and Kimoto, 2000)

### *1-3-3 Nitrogen availability*

Nitrogen is an essential macronutrient for plants in boreal ecosystems, often limiting plant production. Soil nitrogen availability for plant uptake depends on microbial biomass and litter quality (low quality litter contains low nitrogen content and has slower decomposition rates). Usually, high snow cover depth and subsequent increase in soil temperature and moisture accelerate microbial activity and associated mineralization of inorganic soil nitrogen (Li et al., 2016; Semenchuk et al., 2016), whereas no or little snow and cold soils reduce nitrogen productivity and retention in soil via increased gaseous emissions (Brin et al., 2018), probably due to extremely cold winter soil temperatures limit soil microbial biomass in early growing season (Blankinship et al., 2011).

Microbial communities with simpler composition are especially susceptible to risk of damage by freeze-thaw cycles and hard frost, which negatively impacts decomposition of soil organic matter and nitrogen mineralization in soils, although the exact mechanism is not yet clear (Sulkava and Huhta, 2003).

### *1-3-4 Growing season and biomass production*

Snow cover both directly and indirectly via influence on above-mentioned parameters has an impact on growing season start and wood production (Vaganov et al., 1999). Snowmelt timing regulates the conditions during emergence of plants after dormant winter period. The duration of snow cover directly affects growing season length for plants

because in cold regions growth is limited by many factors associated with winter, such as soil temperature, soil moisture, nutrient availability, and solar activity (Slatyer et al., 2021).

The response is often more complex, but not simple to that the earlier snow melt equals earlier start of growing season. For example, experimentally advanced snow melt by 5-15 days resulted in early leaf opening but also was strongly affected by natural year to year variability (Darrouzet-Nadri et al., 2019).

In boreal shrubs leaf production and leaf growth increases with increase in snow free period, but the response of photosynthetic activity depends on the plant functional type and may have an opposite effect (e.g., evergreen plants) (Kudo et al., 1999). In alpine tundra, snow melt timing is important for plant growth, but indirectly due to high air temperature associated with such early snow-melt conditions (Jonas et al., 2008). Strum et al. (2001) proposed a hypothesis that shrubs by trapping snow and increasing snow cover depth by 10-20% under canopy will promote an increase in soil temperature and soil moisture, and provide positive environment for shrub growth allowing them to expand their habitat further north, but only if increased canopy size and leaf area index would not counter balance increase in soil temperature by shading the ground. Therefore, plant functional type, ecosystem and other growth limiting factors play an important role in both start of growing season and plant production, thus making difficult to address the effects of snow cover on wide scale.

## Chapter 2 Effects of snow manipulation on larch trees in the taiga forest ecosystem in northeastern Siberia

## 2-1 Introduction

Snow cover on soils plays an important role in the ecosystems of the Northern Hemisphere (Chapin et al., 2000; Vavrus, 2007). Snow cover acts as an insulator in winter and regulates the water and nutrient balances, thereby influencing material cycling in summer (Aerts et al., 2004; Kreyling et al., 2012; Freppaz et al., 2018). As stated in the IPCC Fifth and Sixth Assessment Reports, the snow-covered area is declining as the southern boundary turns into a snow-free area (Vaughan et al., 2013; Douville et al., 2021). Because of global warming, snow-covered areas are expected to decrease and precipitation events are expected to become less frequent and intense (Huntington, 2006; Deser et al., 2010; Callaghan et al., 2011).

Although decrease in the depth of snow cover is generally expected because of the decrease of snowfall, in arctic and subarctic regions snow depth may increase. According to simulation models by Park et al. (2014), snow depth is expected to increase in Siberia by 0.99 mm per year. This modeled trend is in accordance with observations from the central and western Siberia regions of Russia (Bulygina et al., 2009; Bulygina et al., 2011). Snow cover depth and duration have a major effect on ecosystems through their influence on the soil water balance and thermal regimes, nutrient availability, and duration of the growing season (Schimel et al., 2004; Grippa et al., 2005; Loranty et al., 2018). Additionally, changes in snow depth and density have a significant influence on the soil temperature (Zhang, 2005; Callaghan et al., 2011). In continuous permafrost regions, deep snow cover can lead to significant increase in mean annual ground temperatures, a reduction in the winter freezing depth, and increase in soil moisture in spring and summer (Morse et al.,



2012; Johansson et al., 2013; Park et al., 2015; Karjalainen et al., 2019). Such phenomena were observed in Eastern Siberia in 2004–2007 with heavy summer rainfall and winter snowfall (Iijima et al., 2010). Interannual variations in snow cover conditions (e.g., timing, duration, density, and thickness) also have a strong influence on the long-term ground temperature in cold regions (Aerts et al., 2004). During winter and early spring, the arctic soil temperature mainly depends on the properties of snow cover and regional climate, and even small changes in snow cover potentially have a strong impact on soil frost frequency and intensity (Walker et al., 1999). In addition to the soil temperature, an increase in snow cover may cause soil subsidence and waterlogging in permafrost regions with a high ice content (Nauta et al., 2015). In high-latitude ecosystems, snowmelt timing controls not only hydrometeorological processes, but also many biological processes such as plant phenology and productivity. There are many case studies of snow manipulation experiments exploring the effects of changing depths of snow cover (e.g., Pilon et al., 2004; Walker et al., 1999; Schimel et al., 2004; Wahren et al., 2005; Wipf and Rixen, 2010; Kreyling et al., 2012; Semenchuk et al., 2016; Watanabe et al., 2019).

Snow cover affects soil moisture in the early growing season because approximately half of the snow water equivalent (SWE) infiltrates into the soil after snow melt (Sugimoto et al., 2003). Increased water availability, especially during the early growing season, can positively affect boreal tree growth (Zhang et al., 2019), whereas large amounts of soil moisture may lead to overwetting conditions (Iwasaki et al., 2010; Tei et al., 2013), resulting in reduced gross primary production (Kotani et al., 2019). In subarctic bogs, phenology observed by early flowering dates was reported in an experiment with an open

top chamber (Aerts et al., 2004). On the other hand, early snow melt, followed by cold air temperatures in spring, reduced above ground plant productivity in Arctic Alaskan tundra (Stow et al., 2004) and boreal forests in northeastern Siberia (Kirilyanov et al., 2003). Models with coupled hydrological and biogeochemical processes (such as CHANGE) have also shown that deep snow cover causes larger net ecosystem exchange because of high soil moisture, especially during dry years (Park et al., 2011).

Figure 4 is a schematic diagram of the effects of changes in snow depth on boreal forest or tundra ecosystems. Amount of snow cover on soils influences length of growing season, soil moisture, and N availability (Figure 4). Because an increase in snow cover depth causes low thermal conductivity, leading in high soil temperatures. This can stimulate the decomposition of soil organic matter, resulting in increased nitrogen availability, as generally described (Schimel and Clein, 1996; Hardy et al., 2001; Robinson, 2002; Aerts, 2006; Wipf and Rixen, 2010). However, such effects are diverse and depend on ecosystem types, which are associated not only with climatic conditions but also with vegetation, litter quality and quantity, microbial composition, and plant N demand (Li et al., 2016). Although nitrogen mineralization in soil and uptake by plants during winter are relatively well studied in temperate forests (Andresen and Michelsen, 2005; Ueda and Tokuchi, 2013), the number of studies in boreal and tundra ecosystems is still limited (Cooper, 2014; Koyama and Kielland, 2019). Early, deep snowpacks in forest ecosystems allow microbial activity to continue during winter in Alaska, which was previously presumed to be biologically inactive due to the negative soil temperature, as well as that winter (non-growing season) nitrogen mineralization accounted for approximately 40% of the annual flux, which was

significantly higher than previously reported (Kielland et al., 2006). Moreover, deep snow cover causes high soil nitrogen availability, which leads to increased plant N uptake, foliar nitrogen content, photosynthesis rates (Leffler and Welker, 2013), leaf area index (Pattison and Welker, 2014), and plant production (Wahren et al., 2005).

Frost damage is another factor associated with a decrease in snow cover depth. Thin snow cover may cause freeze-thaw cycles in winter and have a negative effect on plant production. The experimental removal of snow cover has produced direct frost damage, followed by a 50% reduction in understory vegetation coverage (Kreyling et al., 2012). Indirectly, soil frost and frequent freeze-thaw cycles in the absence of snow cover during winter and spring potentially causes a decrease in nitrogen availability (Feng et al., 2007, Frechette et al., 2011), an increase in nitrogen and phosphorus leaching (Fitzhugh et al., 2001), and root damage, that disrupts nutrient uptake (Cleavitt et al., 2008). Forests with deeper organic layers have a lower possibility of soil frost damage during winter, even if the snow cover is less deep (Hardy et al., 2001).

Although there are many reports on snow manipulation experiments on tundra ecosystems, few reports were published on forest ecosystems. This is because snow cover manipulation in forests is a complicated process required labor intensive techniques such as snow shoveling, unlike in tundra, where snow fences can be easily used to manipulate snow cover. In temperate and boreal forests, most snow manipulation experiments had been conducted in regions with expected reductions in snow, for example, North America and Scandinavia, and therefore were mainly focused on the snow removal treatment (e.g., Groffman et al., 2001). Snow removal experiments in forested areas have shown a strong

effect of snow cover on ecosystem processes, such as heat and moisture fluxes, nutrient dynamics, and changes in phenology and diversity (Groffman et al., 2001; Frechette et al., 2011; Kreyling et al., 2012; Comerford et al., 2013; Drescher and Thomas, 2013; Martz et al., 2016; Watanabe et al., 2019). Because of direct frost damage to the root system, there have been many reports on the reduced ability of plants to uptake water and nutrients (Pilon et al., 1994; Cleavitt et al., 2008; Blume-Werry et al., 2016). A decrease in nutrient availability from shallower snow cover due to leaching or impaired microbial activity was also reported (Fitzhugh et al., 2001). Moreover, Frechette et al. (2011) observed decreases in foliar N and photosynthetic activity through spring snow removal in a Canadian boreal forest ecosystem. In a northern hardwood forest in Japan, soil ammonium increases following the enhancement of soil freeze-thaw cycles in winter because DON and DOC availabilities for soil microbes increase (Watanabe et al., 2019).

Other effects have also been observed, such as reductions in the terminal shoot length of sugar maple trees in the U.S. (Comerford et al., 2013), a decrease in the coverage of understory plants in Swedish boreal forests (Kreyling et al., 2012), and reductions in sapling survival in forests in Canada and northern Finland (Drescher and Thomas, 2013; Martz et al., 2016). These effects have negative implications for biomass production. As described above, there are many publications on Alaska and European boreal and temperate forest ecosystems; however, no snow manipulation experiments have been conducted for northeastern Siberia, which is a globally typical forest ecosystem.

Eastern Siberia is a vast territory covered by taiga, characterized by deciduous conifer larch trees (*Larix gmelinii* and *L. cajanderi*) which grow on permafrost (Abaimov et

al., 2002; Kharuk et al., 2006). This region experiences extreme cold winters, short and hot summers, and an extremely dry climate (Tei et al., 2017; Tei and Sugimoto, 2018). Because of the vast coverage area of larch forests worldwide (approximately 40% of the forested area of Russia), the response of larch forests is extremely important (Abaimov et al., 1997). Changes in snow cover depth and snow water equivalent (SWE) in this region affect soil moisture considerably. For larch trees, soil moisture derived from snowmelt water is extremely important in the early summer season when photosynthesis activity is highest (Sugimoto et al., 2002, 2003). It is also expected that larch trees will be affected by changing snow cover and the related processes described above. The objective of this research is to observe the responses of larch trees to changes in snow cover in snow manipulation experiments.

We conducted a snow manipulation experiment in December 2015 by manually removing snow from the snow removal plot (SNOW-) using shovels and adding the removed snow to the snow addition plot (SNOW+). The snow was transported with heavy duty bags. The aim of this study is to examine 1) whether or not snow cover manipulation affects soil temperature under extreme cold winter and short spring, and 2) whether or not the manipulation alters the start of the growing season. Deep snow cover causes longer period of snowmelt (Figure 4); however, soil thaw rate in spring depends on the soil temperature, which is strongly affected by soil moisture (or ice) content. We also investigate 3) when the nitrogen mineralization starts in soils, and 4) whether or not soil moisture affects the nitrogen availability. Notably, soil moisture is extremely important for larch forest in northern Siberia, because of the small amount of annual precipitation (238

mm). Therefore, not only water availability but also N availability for plants is also expected to be affected by soil moisture. Such dynamic effects will cause a change in tree biomass production. As no direct measurements of changes in the biomass production are conducted in this study, we instead employ the following approaches: 1) needle elongation measurements as a proxy for the growing season length, which can be used as a proxy for production change (Montgomery et al., 2020); 2) soil nutrient availability and foliar nitrogen content, which can be used as a proxy for the photosynthesis rate, and effects on biomass production (Field and Mooney, 1986). We also use 3) water isotopes to investigate the origin of soil moisture and carbon isotope ratios to know photosynthetic physiology and carbon allocation trends.

## 2-2 Methods

### *2-2-1 Site description*

The experiment was conducted at Spasskaya Pad Scientific Forest Station (62°25' N, 129°62' E, elevation 220 m above sea level) of the Institute for Biological Problems of Cryolithozone, Siberian Branch of the Russian Academy of Sciences (Fig. 5). The station is located 20 km northwest of Yakutsk, Sakha Republic, Russian Federation (Fig. 5). Deciduous conifers (*Larix cajanderi* and *L. gmelinii*) are the dominant species in the study area. The forest floor is covered by various plant species, including the deciduous shrub *Arctous alpina* and the evergreen shrubs *Vaccinium vitis-idaea* and *Arctostaphylos uva-ursi*, and a couple of grass species. This region is characterized by an intense continental climate with severe cold winters and hot dry summers with an extreme range of monthly mean temperatures from +19 °C in July to −40 °C in January. The average annual mean precipitation is 238 mm (from 1971 to 2000) and the average maximum snow cover depth is  $34 \pm 7$  cm (from 1959 to 2015), according to observations at Yakutsk station of the Hydrometeorological Center of Russia.

### *2-2-2 Experimental design*

During the summer of 2015, three 20 m × 20 m (400 m<sup>2</sup>) experimental plots for snow addition (SNOW+), snow removal (SNOW-), and no treatment (CONTROL) were established (Fig. 5c). Snow cover at the SNOW- plot was manually removed using shovels and transported to the SNOW+ plot using heavy duty bags on December 17–21, 2015. At the time of manipulation, the snow depth was 24 cm. After transporting the snow, the SNOW- plot had approximately 1 cm of snow cover remaining, and the snow cover depth of the SNOW + plot did not change because of compaction of existing snow cover by the added snow. Soil temperature and soil moisture observations were conducted from 2015 summer to 2016 summer. In spring, the snow depth and snowmelt timing were recorded using automatic time-lapse cameras. During the growing season, needle elongation and thaw layer thickness were measured manually.

### *2-2-3 Hydrometeorological observations*

Air temperature and precipitation were observed using an automatic weather observation system (WXT520, Vaisala, Vantaa, Finland) installed at a height of 15 m (the top of the canopy). Soil moisture was observed with dielectric permittivity probes with data loggers (HS10 with EM50 data logger, Decagon Devices, Pullman, Washington, USA) installed at five depths of 0.01, 0.1, 0.2, 0.3 and 0.5 m at the SNOW- and SNOW+ plots. Measurements began on June 4, 2015. Soil temperature measurements began on July 15,



2015, using thermistors (TMC-HD with U-12-008 data loggers (HOBO, Onset Computer Corporation, Bourne, Massachusetts, USA) at six depths of 0.1, 0.2, 0.4, 0.6, 0.8, and 1 m for SNOW- and SNOW+ plots. The date of snowmelt was determined at all treatment plots using images from a time-lapse camera according to the date at which 50% of the ground surface was visible.

#### *2-2-4 Needle elongation and samplings*

In order to reach the sunlight branches, we used young larch trees for measuring needle elongation. Three typical young larch trees (2–4 m height) were selected from each treatment plot for needle elongation measurement and sampling for foliar chemistry analysis (C and N contents, and their isotopic compositions). For each tree, three branches were marked, and the elongation of 3–9 randomly selected needles was measured at each branch using digital calipers (totally 38 to 55 replications at each plot). Each tree was located more than 4.8 m from the edge of the plot and each plot was 8 meters apart. Needle length measurements were conducted seven times from May 8 to July 8, 2016. Data was presented as percentage of the final length of needles observed on the last day of the observation (July 8). Needles were also sampled on June 17, July 16, and August 20, 2016, and current year shoots were sampled on August 20, 2016 for C and N contents and isotope analyses. Dried needles and shoots were ground and analyzed with an elemental analyzer – isotope ratio mass spectrometer online system (Flash EA 1112 and Delta V, Thermo Fisher Scientific, Massachusetts, USA). The analytical accuracy (reproducibility) was 0.3% and

0.2% for C and N content, respectively, and less than 0.2‰ for the  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values.

Delta values for carbon are defined by:

$$\delta = \left[ \frac{\left[ \frac{^{13}\text{C}}{^{12}\text{C}} \right]_{\text{sample}}}{\left[ \frac{^{13}\text{C}}{^{12}\text{C}} \right]_{\text{standard}}} - 1 \right] \times 1000$$

Delta values for nitrogen are defined by:

$$\begin{aligned} \delta^{15}\text{N} &= \left( \frac{R_{\text{sample}}}{R_{\text{standard}}} - 1 \right) \times 1000 \\ &= \left( \frac{(^{15}\text{N}/^{14}\text{N})_{\text{sample}}}{(^{15}\text{N}/^{14}\text{N})_{\text{standard}}} - 1 \right) \times 1000, \end{aligned}$$

#### *2-2-5 Isotopic analyses of stem water and precipitation*

To analyze the isotopic compositions of oxygen and hydrogen in stem water (water extracted from larch tree branches), we selected branches from mature trees. Tree branches from two mature larch trees (reaching stand height) at each treatment plot were sampled from 4–6 m height using an extendable tree pruner. Bark and cambium were removed and the remaining xylem was cut into 1-cm pieces, stored in glass vials, and stored at  $-18\text{ }^{\circ}\text{C}$  prior to extraction. Branch samples were collected on May 14, May 27, June 7, June 15, July 16, and August 12, 2015, and on May 29, June 17, July 15, and August 14, 2016. Water was extracted from the stem cryogenically on a vacuum distillation line and stored in glass vials at  $5\text{ }^{\circ}\text{C}$  prior to analysis. Rain samples were collected during rain events in situ

using a simple rain collector. Snow cover samples were collected on December 16–19, 2015, using a metal cylinder (12-cm diameter). The water samples were analyzed using an online system (GasBench equipped with MAT253, Thermo Fisher Scientific, Massachusetts, USA). Analytical accuracy (reproducibility) was 0.1 and 0.2‰ for the  $\delta^{18}\text{O}$  and  $\delta\text{D}$  values, respectively. Delta values are defined by

$$\delta^{18}\text{O} \text{ (or } \delta\text{D)} = (R_{\text{sample}}/R_{\text{SMOW}} - 1) \times 1000 \text{ (‰)},$$

where  $R_{\text{sample}}$  and  $R_{\text{SMOW}}$  are isotope ratios ( $^{18}\text{O}/^{16}\text{O}$  or D/H) of sample and standard (Vienna Standard Mean Ocean Water).

#### *2-2-6 Soil inorganic nitrogen*

For the soil inorganic N analysis, four soil core samples were collected from all treatment plots on July 16 to 18, July 22 to 25, and August 13 to 15, 2016, at four depths (organic layer, 0–10, 10–20, and 20–50 cm). Roots were removed from the soil samples, and soil N was immediately extracted from 4 g of soil sample by 40 ml of 2M KCl solution and mixed on a shaker (40 min). The solution and soil particles were then separated using a centrifuge and filtrated with a 0.45- $\mu\text{m}$  syringe filter. The extracts were stored at  $-18\text{ }^{\circ}\text{C}$  prior to analysis, and ammonium and nitrate concentrations in the extracted samples were analyzed using a continuous flow spectrophotometer autoanalyzer (Bran & Luebbe, Norderstedt, Germany).

### *2-2-7 Statistical analyses*

Statistical significance was analyzed using the Student's t-test for needle length difference among treatments and between consecutive dates. One-way Analysis of Variance with post-hoc Tukey test was used for needle chemistry analyses. Data were compared among treatments and consecutive dates based on a significance level of  $p < 0.05$ .

## 2-3 Results

### *2-3-1 Hydrometeorological conditions during the experiment*

Table 1 shows the monthly averages of air temperatures and precipitation during warm periods (April to September) as a seven-year average (2010–2016) and for 2015 and 2016. The air temperature of the years before and after snow manipulation (2015 and 2016, respectively) was close to the seven-year average, except for April 2015 ( $-6.3\text{ }^{\circ}\text{C}$ ) and September 2016 ( $7.9\text{ }^{\circ}\text{C}$ ), which were colder and warmer than the seven-year average ( $-0.6$  and  $4.7\text{ }^{\circ}\text{C}$ ), respectively. Total precipitation during the warm period in 2015 and 2016 was mostly similar to the seven-year average (119, 109, and 109 mm, respectively); however, June 2015 showed a larger amount of rainfall, and August 2015 and September 2016 received smaller amounts of rain than the seven-year average. Before snow manipulation on December 17–21, 2015, the snow density was  $106\text{ kg/m}^3$  with snow water equivalent (SWE) of 25 mm.

Before snow manipulation, soil temperatures of all three plots exhibited no significant difference, except for lower values at 0.1 m and 0.2 m depth at CONTROL from October to December (Fig. 6). After manipulation, the soil temperature in the SNOW– plot rapidly decreased and became significantly lower than those in the SNOW+ and CONTROL plots. The maximum difference between SNOW– and CONTROL was  $5.8\text{ }^{\circ}\text{C}$  at 0.1 m on December 30, 2015, and the difference decreased with an increase in depth. The difference between SNOW+ and CONTROL was  $3.7\text{ }^{\circ}\text{C}$  at 0.1 m on the same date. At the end of March, as the daytime air temperature increased, the soil temperature started to

increase simultaneously for all treatments. No significant difference in soil temperature was observed among the three plots during the period from March 24 to April 18, 2016. However, after snowmelt started on April 19 at the SNOW– plot (Fig. 7a and b), the soil temperature of all monitored soil depths (from 0.1 m to 0.4 m) became higher than the soil temperature at the SNOW+ plot at corresponding depths. This may be caused by a difference in soil moisture, as described below. The observed difference in soil temperature continued until the end of the observation period at the end of summer. The soil temperature at 0.1 m depth in the SNOW– plot exceeded 0 °C on April 27 (eight days after snowmelt), and exhibited diurnal fluctuations for 21 days (12 consecutive days) (until May 10), decreasing at or below 0 °C during nighttime (Fig. 7a), which indicates diurnal freeze-thaw cycles. However, soil temperature at 0.1 m depth in the SNOW+ plot only exceeded 0 °C on May 16 (20 days after snowmelt), and showed diurnal variations with minimum soil temperatures at or below 0 °C for only 7 days ( 5 consecutive days) (until May 23).

Before snow manipulation, there was no significant difference in the average soil thaw depth between SNOW+ ( $58 \pm 13$  cm) and SNOW– ( $61 \pm 13$  cm) plots ( $n=441$ ) on July 7, 2015 (Supplement 1 a, b), indicating similar soil properties between the two plots. However, the spatial variability of soil thaw depth showed a dependency on understory vegetation. Under patches of deciduous shrubs with broader leaves (*Arctous alpina*), the thaw depth was shallower than that under patches of evergreen shrubs (*Vaccinium vitis-idaea*, *Arctostaphylos uva-ursi*) and grasses due to shading by the higher leaf area.

After snow manipulation, in the summer of 2016, the thaw depth was measured only for designated subplots (9 m<sup>2</sup>) with a dominant type of understory plant (deciduous

shrub, evergreen shrub, grasses). On July 26, 2016, SNOW<sup>-</sup> subplots had the deepest average thaw depth, whereas SNOW<sup>+</sup> subplots were the shallowest, and CONTROL subplots exhibited intermediate thaw depths. The average thaw layer depths of the three subplots were  $84 \pm 9$  cm for SNOW<sup>-</sup>,  $72 \pm 6.7$  cm for SNOW<sup>+</sup>, and  $77 \pm 5.3$  cm for CONTROL (Table 2). As the soil thawed in the summer of 2016, soil moisture was higher at SNOW<sup>+</sup> than SNOW<sup>-</sup> in the surface soil layers (0.1 m and 0.3 m) (Fig. 7c) due to the infiltration of snowmelt water.

### *2-3-2 Snowmelt timing and needle elongation*

The snowmelt date was the earliest in the SNOW<sup>-</sup> plot (April 19), followed by the CONTROL plot (April 23), then SNOW<sup>+</sup> plot (April 25) (Fig 8). Figure 9 shows the growth of needles of young larch trees, which is a percentage of the needle length on July 8, 2016 in each plot. Needle lengths on June 16 were 103, 99.2, and 101% in the SNOW<sup>-</sup>, SNOW<sup>+</sup> and CONTROL plots, respectively, indicating that needles were already mature on June 16. Leaf opening in young larch trees in all three plots had already started on May 8, 2016, at the beginning of the observation period (Fig. 9). Despite the earlier snowmelt, SNOW<sup>-</sup> plot needles exhibited delayed elongation, with shorter length (7.4%) than those in the CONTROL (14.1%) and SNOW<sup>+</sup> (12.7%) plots on May 8 (Fig. 9). This situation continued until May 30.

In the period from May 30 to June 16, there was a different situation. Needle length in the SNOW<sup>-</sup> plot increased significantly from May 30 (64.9%) to June 16 (103%), while

only small differences were observed from May 30 (83% for both SNOW+ and CONTROL) to June 16 (99.2 and 101% for SNOW+ and CONTROL), although the differences were not statistically significant. In the early growing season, elongation of needle length in the SNOW- plot was delayed, and continued until the later stages. The needle length at SNOW- plot was smaller in length in mm compared to SNOW+ and CONTROL for the period from May 8 to May 30, but in June and July after the needle growth reached length near to final the difference became insignificant among treatments, but in absolute values SNOW+ needle were longer than SNOW- needle throughout the experiment.

### *2-3-3 Stem water isotopic compositions*

Before snow manipulation, in the summer of 2015, the  $\delta^{18}\text{O}$  values of larch stem water showed clear seasonal variations and no difference was observed among the three plots (Fig. 10a). The  $\delta^{18}\text{O}$  values were highest in May (-13.6‰), with a sharp decrease in June (-21.3‰), followed by an increase in July (-16.5‰) and August (-18.6‰). After snow manipulation, in the summer of 2016, the  $\delta^{18}\text{O}$  values of larch stem water showed clear differences (Fig. 10b). The  $\delta^{18}\text{O}$  values of the SNOW- plot exhibited the highest values throughout summer (-18.5‰), with only a slight increase by 1.3‰ from June to August. CONTROL and SNOW+ plots exhibited similar values that were lower than those of SNOW- in early summer, but almost equal to those of SNOW- (-18.1‰) in August. As shown in Table 1 and Fig. 11, summer rainfall has higher  $\delta^{18}\text{O}$  values (-18 to -10‰) than



snow cover (approximately  $-30\%$ ); thus, the decreased  $\delta^{18}\text{O}$  values in all plots in June 2015 and in SNOW+ and CONTROL plots in 2016 resulted from the uptake of infiltrated snowmelt water with extremely low  $\delta^{18}\text{O}$  values, as previously described by Sugimoto et al. (2002).

#### *2-3-4 Carbon and nitrogen contents and isotopic ratios*

From June to August 2016, the needle C content of young trees and their  $\delta^{13}\text{C}$  values exhibited decreasing trends among all three plots, although no statistically significant differences were observed (Fig. 12a and b). In addition, current year shoots and needles on them of young larch trees at SNOW+ plots showed lower  $\delta^{13}\text{C}$  values ( $-30.8 \pm 0.7\%$  and  $-30.6 \pm 0.8\%$ ) than the SNOW- plot ( $-29.9 \pm 0.5\%$  and  $-29.9 \pm 0.3\%$ ), although this difference was not statistically significant (Fig. 13). However, we suppose that the soil moisture difference between SNOW- and SNOW+ plots may have caused the higher  $\delta^{13}\text{C}$  values at SNOW-, as we will discuss later.

Needle N content at SNOW- decreased slightly from June 17 ( $1.4 \pm 0.2\%$ ) to July 16 ( $1.2 \pm 0.1\%$ ). Then, on August 20, the N content in all plots increased, especially in the SNOW- plot (Fig. 12c). The observed needle  $\delta^{15}\text{N}$  values ranged from  $-6.2\%$  to  $-2.1\%$ , revealing large variability among individual trees. Needle  $\delta^{15}\text{N}$  values of most trees exhibited an increasing trend from June 17 to August 20, excluding the two cases out of the three trees in the SNOW- plot. In the SNOW+ and CONTROL plots, the increase in  $\delta^{15}\text{N}$  values from June to August was  $0.8\%$  (Fig. 12d). Nitrogen content and  $\delta^{15}\text{N}$  of current year

shoots were not measured due to the concentration below accurate measurement threshold.

### *2-3-5 Soil inorganic nitrogen*

Soil inorganic N exhibited temporal and spatial variations at the study site (Fig. 14). Inorganic N mostly comprised ammonium at the site. A low concentration of ammonium was found in the SNOW+ plot in the middle of July (Fig. 14a). At the end of July (Fig 14b), the concentration of ammonium increased in the SNOW+ and CONTROL plots. These results agree to our previous studies (Li et al., 2016; Semenchuk et al., 2016). Soil inorganic N was small in the beginning of summer, and increased quickly after the soil temperature at 20 cm reached optimal condition, then active N mineralization began in the period from the middle of July to the beginning of August. (Popova et al., 2013). On the other hand, at the end of July, a low concentration of ammonium was observed at the SNOW- plot (Fig. 14b), and at the end of August concentration of ammonium in the SNOW- plot finally increased (Fig. 14c).

## 2-4 Discussion

### *2-4-1 Soil temperature*

In our experiment, after snow manipulation, winter soil temperature in the SNOW- plot quickly decreased when compared with that in the SNOW+ and CONTROL plots (Fig. 6). The absence of a snowpack at the SNOW- plot during the coldest period in winter reduced the effect of snowpack insulation and we expect a high thermal conductivity of the ground surface, including the snowpack, enabling winter cold waves to cool the deep soil layer. Therefore, because of the lower snowpack depth and higher thermal conductivity, the SNOW- plot had low soil temperatures until March, when air temperature rose substantially.

The SNOW+ plot showed higher soil temperatures than the CONTROL plot until mid-January (monthly average soil temperature at SNOW+ was higher than that at CONTROL by 0.2 °C in October (first month of snow) 2015 and by 2 °C in January), however, during the period from mid-January to March, the soil temperature was similar in SNOW+ and CONTROL (average monthly difference of 0.7 °C at 10 cm between the treatments)(Fig. 6). The similarity may be caused by compaction of the snowpack. After manipulation, the observed snow depth at SNOW+ was 24 cm, which was similar to that before manipulation, because the snow density was doubled by adding snow from the

SNOW– plot. Generally, cold regions such as Yakutsk have lower snow density ( $108 \text{ kg/m}^3$  in winter and  $161 \text{ kg/m}^3$  in spring, as measured in 2015 at the experimental site), which suggests a lower thermal conductivity of the ground surface, including the snowpack. Conversely, we expect larger snow density observed at the SNOW+ plot correspond to a larger thermal conductivity. Thus, compaction of the snowpack may impair the insulation effect of the snowpack (Zhang, 2005).

#### *2-4-2 Length of growing season*

The SNOW– plot experienced earlier snowmelt (April 19) than the SNOW+ and CONTROL plots (April 25 and 23), as shown in Fig. 8. However, contrary to the schematic diagram shown in Fig. 4, the SNOW– plot did not exhibit early needle opening, but delayed needle elongation (Fig. 9). Needle opening started before the first day of our observation (May 8, 2015). During the period from May 8 to May 30, needle length in the SNOW– plot was significantly shorter than that in the SNOW+ (by 5.3% of final length or by 1.6 mm on May 8 and by 19.2% of final length or by 7.2 mm on May 30) and CONTROL (by 6.7% of final length or by 1 mm on May 8 and by 19% of final length or 4.3 mm on May 30) plots, where needle elongation continued until May 30 (76.1% of final length for SNOW+ and 77.1% for CONTROL, whereas only 64.9% for SNOW–). On the other hand, needles at the SNOW– plot continued to elongate until June 16 (85.2% of final length). Since the needle elongation started before the first day of observations, we do not know the exact date needle elongation started at each plot, but by calculating daily average

elongation speed in mm between first two measurement points on May 8 and May 10 and then by converting it into percentage of needle elongation, we may potentially estimate the first day of needle elongation. Using this method, the first day of needle elongation for CONTROL was April 29 (9 days before the start of observation), for SNOW- was May 3 (5 days before the start of observation) and for SNOW+ was April 27 (11 days before the start of observation).

Clearly, needle elongation took longer at the SNOW- plot. One of the possible reasons for this is the dry conditions (Fig. 4). The limited water supply at the SNOW- plot during early summer may also be a factor influencing slower needle elongation (Fig. 7c). In the following subsection, we discuss the influence of soil moisture.

In contrast to these findings, previous studies have found that early snowmelt leads to a long growing season. According to satellite and ground observation data, Barichivich et al. (2013) showed that photosynthesis activities in northern ecosystems are enhanced by the long growing season, especially by early snowmelt and an early spring. Moreover, Sutinen et al. (2015) conducted a snow manipulation experiment and showed that soil frost in spring caused a delay in needle and shoot growth in Norway spruce. These results, which have generally been attributed to earlier snowmelt resulting in a long growing season, seem inconsistent with our data. In our study, SNOW- treatment had a high soil temperature in spring, but the dry soil condition in the SNOW- plot may cause this. The small amount of precipitation (approximately 238 mm per year) at our study site (Yakutsk) may explain the difference between our study and previous research. Another possibility not explicitly shown in Fig. 4 is that frost damage of fine roots caused by freeze-thaw cycles in spring

(Fig. 7a) might cause a delay in needle elongation (Fig. 9) through a decline in water and nutrient uptake.

### *2-4-3 Soil moisture*

In 2015 and 2016 at Spasskaya Pad, the precipitation values agreed with the seven-year average (Table 1). Therefore, our experiment was conducted in typical dry conditions, with no “overwetting” conditions in the forest after the snowmelt in 2016 spring. The origin of water in the soil can be determined using water isotope ratios (Sugimoto et al., 2002). Infiltration of larger amounts of snowmelt water in SNOW+ and CONTROL plots than in the SNOW–plot was recorded by the  $\delta^{18}\text{O}$  values of stem water (Fig. 10b). This means that trees in the SNOW– plot were unable to use snowmelt water, which has low  $\delta^{18}\text{O}$  values (Fig. 11). During summer, rainwater with high  $\delta^{18}\text{O}$  values recharged the soil, and in August, all plots showed similarly high  $\delta^{18}\text{O}$  values (Fig. 10). As seen in Fig. 7c, soil moisture in the SNOW– plot increased gradually from early summer. In our study area, which is in a continental dry climate, larch trees show high photosynthetic activity in the early growing season, because snowmelt water can be used by plants (Sugimoto et al., 2002). Snow removal treatment thus shows negative effect on tree growth.

Another proxy for soil moisture is the  $\delta^{13}\text{C}$  values of current year shoots because C used for the production of current year shoots is mainly photosynthesized during the growing season (Kagawa et al., 2006; Li and Sugimoto, 2018). In the SNOW– plot, the  $\delta^{13}\text{C}$  values of current year shoots were relatively higher than those in the SNOW+ and

CONTROL plots, which suggest dry conditions in the SNOW- plot, however, these values were not statistically significant because of the small number of samples (Fig. 13).

Delayed elongation of needles in SNOW- plot may be caused by the dry soil condition in the SNOW- plot.

#### *2-4-4 Nitrogen availability*

As shown in Fig. 12c, on June 17 and July 16, needle N content in the SNOW+ and CONTROL plots showed similar values, whereas that at SNOW- showed a large deviation on June 17, and the value on July 16 was significantly lower at SNOW- (1.15%) than those at the SNOW+ (1.42%) and CONTROL (1.4%) plots. The results show that N uptake by trees at the SNOW- plot was insufficient in the first half of summer. However, on August 20, needle N content in the SNOW- (1.54%) plot increased to a value comparable to those of the SNOW+ (1.55%) and CONTROL (1.74%) plots. Similar trends were also observed in the soil inorganic N data, as shown in Fig. 14. As most roots of larch trees are distributed in the soil layer within 50 cm depth (Sugimoto personal communication), foliar N contents are compared with the observed soil inorganic N data.

Low concentration of soil inorganic N was observed at the SNOW+ plot on July 16 to 18 ( 0.003 mgN / g dry soil) (Fig. 14a) and increased rapidly at the end of July (0.052 mgN / g dry soil) (Fig. 14b), whereas that of the SNOW- plot's soil inorganic nitrogen concentrations were below detection threshold in mid-July (0 mgN / g dry soil) and low concentration was detected in late July (0.006 mgN / g dry soil), and only increased in August (0.081 mgN / g dry soil) (Fig. 14c). Therefore, these results indicate that N

availability in the SNOW- plot was low until mid-July, which may have led to lower needle N content in the SNOW- plot during this period. Subsequently, trees in the SNOW- plot used soil inorganic N at the end of July and in August, and the needle nitrogen content in the SNOW- plot in August (0.081 mgN / g dry soil) increased to roughly the same as that in the SNOW+ (0.053 mgN / g dry soil) and CONTROL (0.045 mgN / g dry soil) plots (Fig. 12c). It should be noted that the soil temperature at the SNOW- plot (9.6 °C monthly average for June, July and August at 10 cm depth) was higher than those at the SNOW+ (6.3 °C monthly average for June, July and August at 10 cm depth) and CONTROL (7.5 °C monthly average for June, July and August at 10 cm depth) plots during the growing season (Fig. 7). The foliar  $\delta^{15}\text{N}$  values at the SNOW+ and CONTROL plots (Fig. 12d) showed a gradual increase from June to August (0.78‰ and 0.82‰, respectively), whereas only slight increase could be observed at SNOW- plot (0.25‰). Because soil organic matter in the deeper layer generally has higher  $\delta^{15}\text{N}$  values than plant tissue or atmospheric nitrogen depositions (Craine et al., 2015), observed increases in the foliar  $\delta^{15}\text{N}$  value (Fig. 12d) correspond to the use of soil N.

There are several possible reasons for the delayed increase in soil inorganic N availability at the SNOW- plot. One may be the freeze-thaw cycles in the SNOW- plot during the period from April 28 (the day when the soil temperature exceeded 0 °C) to May 8 (the day when the soil temperature stopped decreasing to 0.1 °C or lower during the night) (Fig. 7a), which may damage the microbial biomass in the soil and plant roots. Another reason may be the dry soil conditions in the first half of the summer (Fig. 7c and 10b). Under dry soil conditions, inactive microbial decomposition may lead to less



available N at the SNOW– plot, and the inactive uptake of N by plants causes low N contents in the needles. As described already, climate of study site (near Yakutsk) shows extremely low winter temperature, therefore extremely low soil temperature at the SNOW– plot might affect the soil microbes and plant roots.

At the SNOW+ plot, increase in snow depth did not significantly alter winter soil temperature. This may be because of compaction of added snow, which leads to snow density double, compared to the CONTROL plot, resulting in the decreased thermal conductivity. Besides, additional snow provided sufficient soil moisture in the early growing season, and the start of mineralization of soil inorganic N in the SNOW+ plot was earlier than in the SNOW– plot.

We could not estimate the effect of doubled snow density on the ecosystem. In the natural condition, snow density was much lower than the observed density in our SNOW+ treatment. This would affect the soil temperature and consequently N availability.

#### *2-4-5 Comparison with other regions*

Our study site is located in a continuous permafrost zone with an extreme continental subarctic climate. It has extremely cold long winters with a small amount of snowfall and dry and hot short summers. Compared to other regions, Eastern Siberia is a rather unique study area from climatological perspective, because it is as cold as high arctic tundra but as dry as alpine tundra, and contains abundant forests. Groffman et al. (2001) proposed a hypothesis whereby global warming will cause less snow cover during winter

and therefore less thermal insulation, which will lead to the “colder soils in a warmer world” phenomenon. They conducted a snow removal experiment in a northern hardwood forest in the US and reported only mild soil freezing ( $-4\text{ }^{\circ}\text{C}$ ) in winter, indicating a great potential for freeze-thaw cycles during winter. Active growth of soil microbes during winter and/or the snowmelt period was reported following snow reduction experiments in northern forests (Groffman et al., 2001; Duran et al., 2014; Sorensen et al., 2016; Watanabe et al., 2019). On the other hand, at our site, the soil in the CONTROL plot was frozen for more than three months and the soil temperature was below  $-14\text{ }^{\circ}\text{C}$ . Moreover, the soil temperature at the SNOW- plot was less than  $-19\text{ }^{\circ}\text{C}$  soon after snow removal (Fig. 6). Such low temperatures could cause mechanical or cellular root damage. In our experiment, the mineralization of soil ammonium at the SNOW- plot was delayed until the end of July (Fig. 14c). The low soil temperature at the SNOW- plot observed during winter might damage not only the roots but also the soil microbes. In addition, freeze-thaw cycles in spring (Fig. 7a) may also damage soil microbes.

Owing to these problems, snow removal in the experiments caused a delay in needle elongation during the early growing season. It has previously been reported that low spring soil temperatures affect the ecosystem. In a boreal forest in Finland, the bud burst of Norway spruce was delayed for two weeks in a spring frost experiment, in which insulation by 15 cm of hay was applied, resulting in prolonged soil frost (Sutinen et al., 2015). This indicates that spring soil frost affects the length of the growing season. In our experiment, we observed that earlier snowmelt at the SNOW- plot did not hasten needle flushing, but slowed needle elongation, although the soil temperature was higher than that at the

CONTROL plot. These results contradict the diagram illustrated in Fig. 4, where short period for snowmelt caused an early start of the growing season. Our results showed the later start of growing season, despite the short period of snowmelt and high soil temperature. Our study site is characterized by extremely low air temperature and dry conditions in winter, as well as a short snowmelt season. Therefore, it is very important for plants to obtain water, instead of warming the soil during the needle flushing period.

In the present study, we observed soil inorganic N only in summer season. Additional studies to investigate soil microbial activity, especially during winter and spring, are required in the future.

## 2-5 Conclusions

A snow cover manipulation experiment was conducted in December 2015 at the Spasskaya Pad Scientific Forest Station in the Eastern Siberia taiga forest ecosystem. The snow removal (SNOW-) plot showed a more significant effect of manipulation than the other plots. Soon after the manipulation, the SNOW- plot showed extreme cold soil temperature. Needle elongation was delayed in the SNOW- plot, despite earlier snowmelt and a higher soil temperature during the growing season. Mineralization of soil inorganic N was also delayed in the SNOW- plot, and the larch needle N content was lower than that in other plots until July. Soil ammonium at the SNOW- plot was eventually produced in August, which resulted in a comparable needle N content to the other plots. Dry soil conditions in the SNOW- plot, during the snowmelt period and early growing season, caused higher soil temperature, and also caused freeze-thaw cycles in spring, resulting in delay of soil N mineralization.

The study duration was only one year, however, we observed a clear response of larch trees to snow cover changes, including changes in phenology (needle opening and elongation). Moreover, longer experiments may cause changes to the ecosystem, such as photosynthesis and biomass production. It is also difficult to predict possible scenarios for long-term ecosystem change because other factors, such as overwetting due to increased soil moisture input by increased amount of snow or permafrost degradation due to increase in soil temperature via deeper snow cover and therefore higher insulating properties, should also be assessed.

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Table 1. Monthly data in 2015 and 2016 and the seven-year average from 2010 to 2016, and  $\delta^{18}\text{O}$  (‰) values of precipitation in 2015 and 2016 observed at Spasskaya Pad during the warm period (April to September) were shown.

Month	Air temperature (°C)			Precipitation (mm)			Precipitation $\delta^{18}\text{O}$ (‰)	
	7-year ave	2015	2016	7-year ave	2015	2016	2015	2016
Apr	-0.6	-6.3	0.3	1.7	0.1	0.1	-	-
May	8.1	6.2	7.0	10.8	12.6	10.7	-	-18.1
Jun	14.8	13.8	15.9	24.1	45.2	27.7	-	-15.1
Jul	19.9	18.5	17.6	32.7	28.8	41.8	-10.0	-14.5
Aug	14.5	15.5	13.2	25.3	13.6	27.4	-13.4	-
Sep	4.7	4.3	7.9	14.1	18.4	1.1	-	-

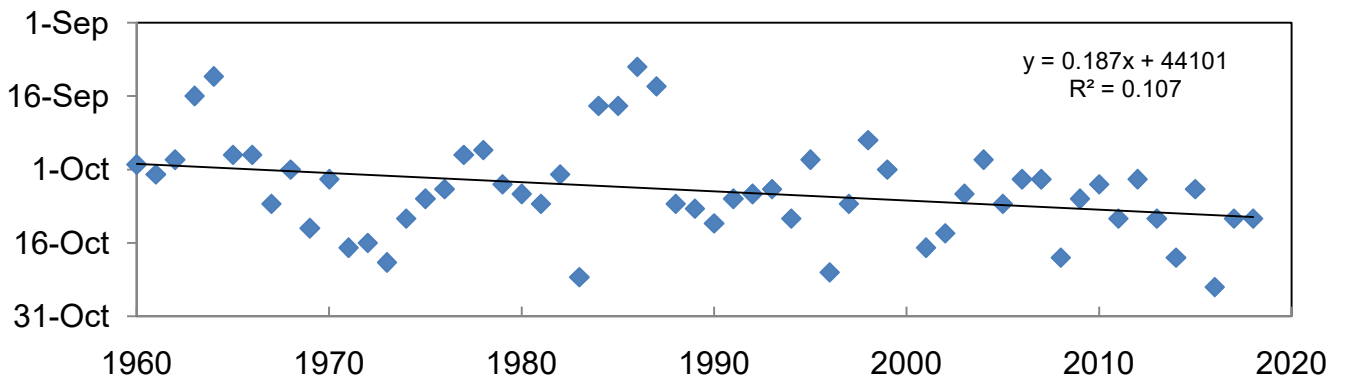
Table 2. Thaw depth in 2016. Thaw depth measured by the vegetation type subplots *Arctous alpina* (broad leaf), *Vaccinium vitis-idaea* (evergreen) and Grasses for each treatment for early and late July.

Subplot	Thaw depth (cm)		
	Snow-	Snow+	Control
	July 7-10 2015		
<i>Arctous alpina</i>	67±6	61±11	n.a*
<i>Vaccinium vitis-idaea</i>	66±8	65±10	n.a*
Grassy	70±12	81±13	n.a*
	July 26 2015		
<i>Arctous alpina</i>	85±5	67±5	74±4
<i>Vaccinium vitis-idaea</i>	73±3	69±4	76±4
Grassy	93±3	79±4	80±7

# Tables and figures

(a)

First day of Snow at Yakutsk (1960-2018)



(b)

Last day of Snow at Yakutsk (1960-2018)

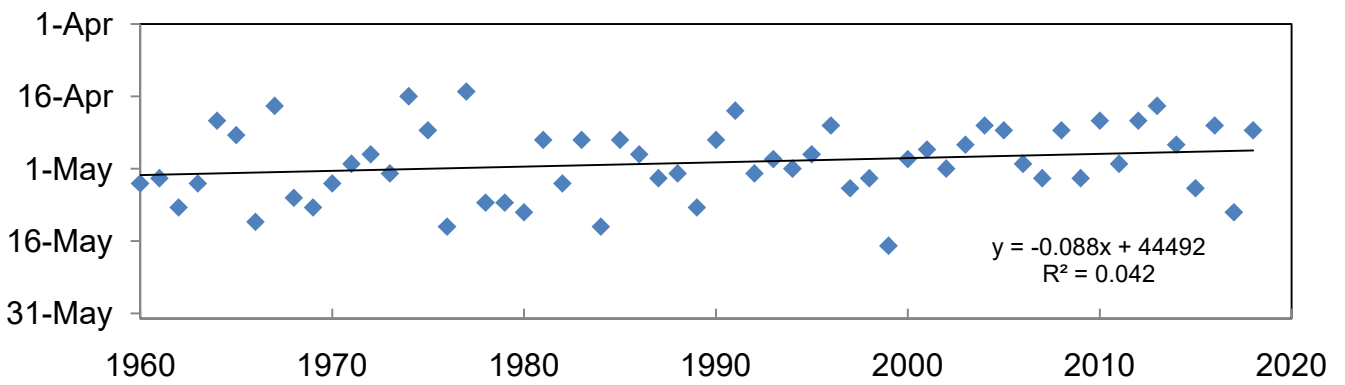


Figure 1. The first day of snow in autumn (a) and the last day of snow (spring) in Yakutsk for 1960-2018.

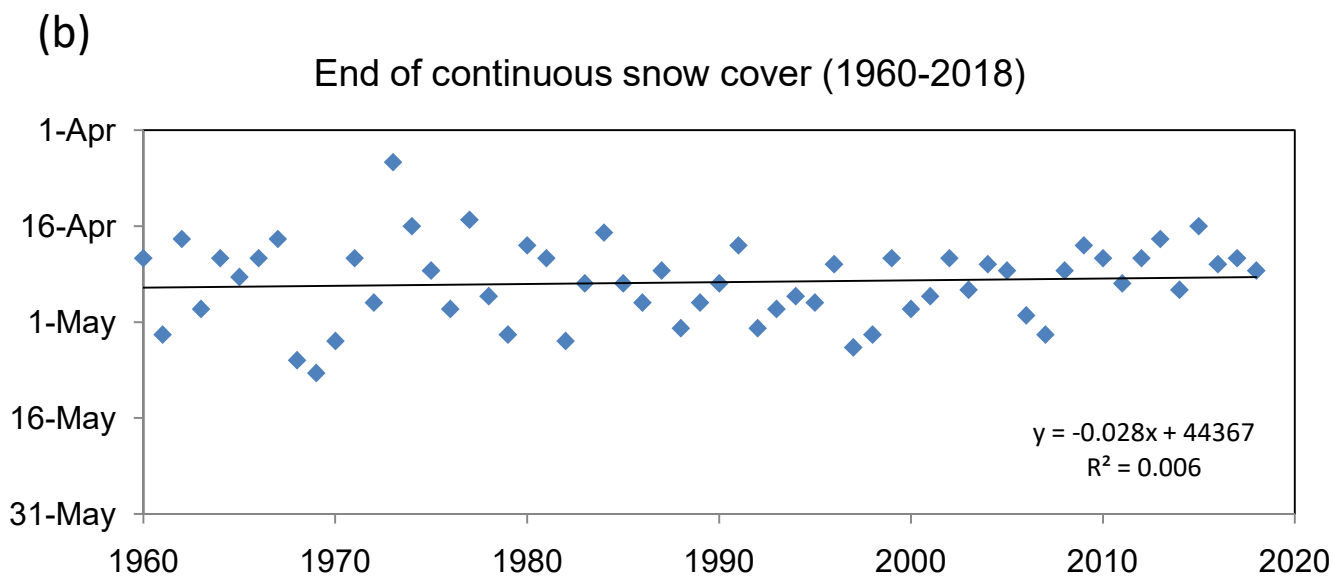
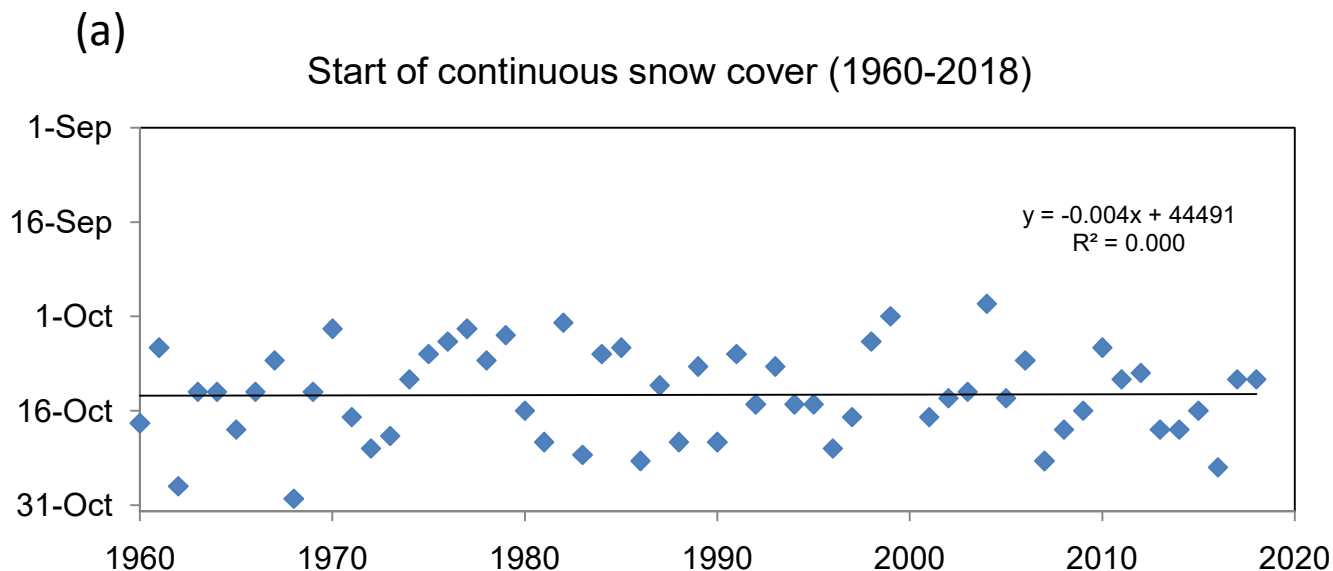


Figure 2. The duration of continuous snow cover in Yakutsk for 1960-2018. Start of the continuous snow cover in autumn (a) and end of continuous snow cover in spring (b) in Yakutsk for the period from 1960 to 2018.



### Snow depth and Air Temperature at Yakutsk (1960-2018)

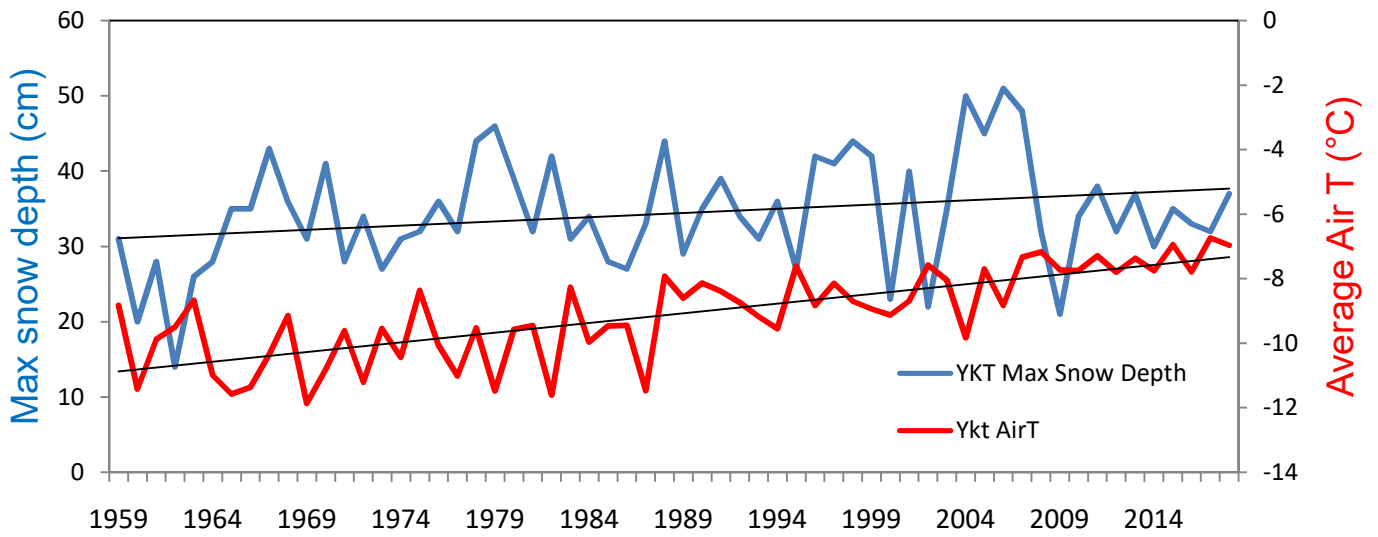


Figure 3. Maximum snow cover depth in cm (blue line) and average annual air temperature (red) in degree Celcius in Yakutsk for the period from 1960 to 2018.

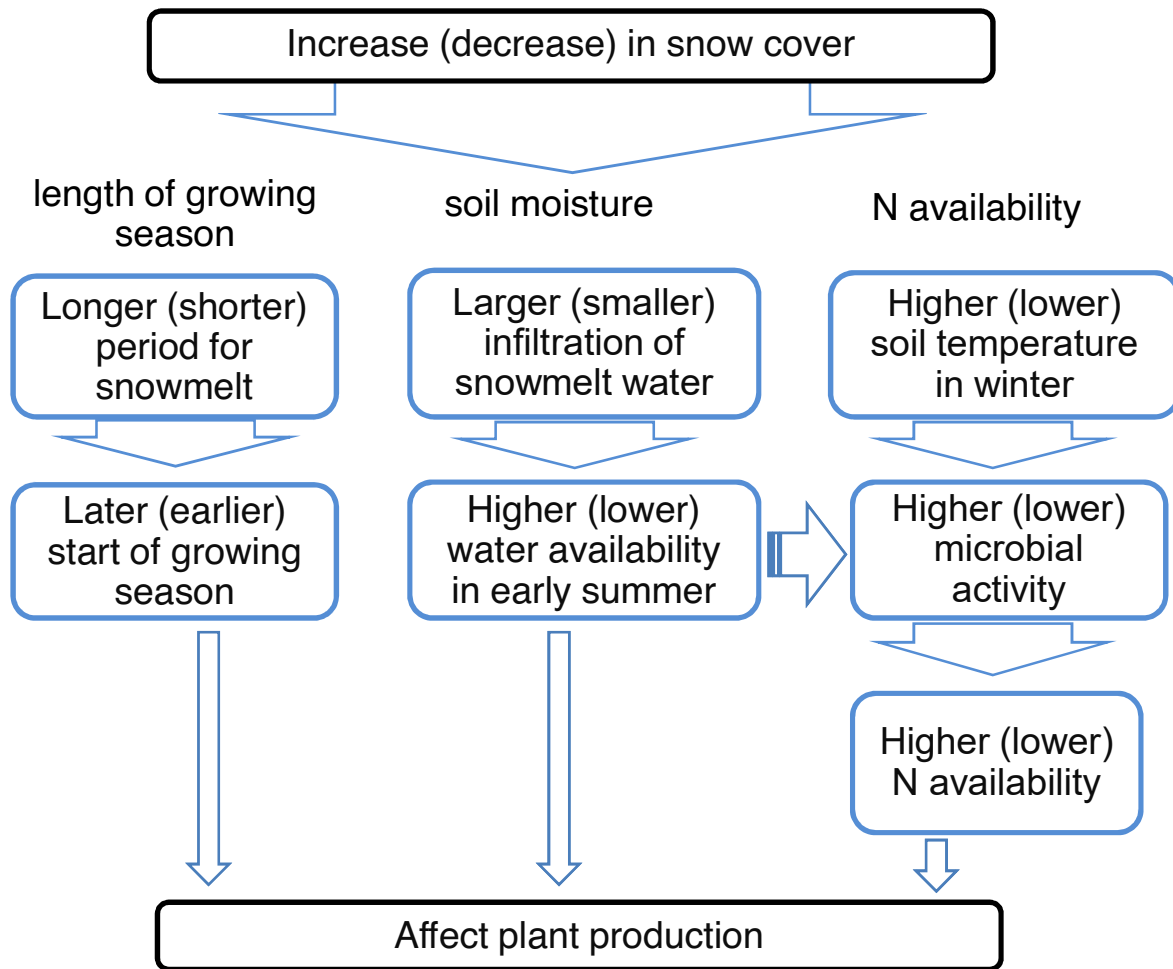


Figure 4. Schematic diagram on the effect of changing snow cover depth. Start of growing season, soil moisture and nitrogen availability may change following increase or decrease in snow cover depth, and affect plant productivity.

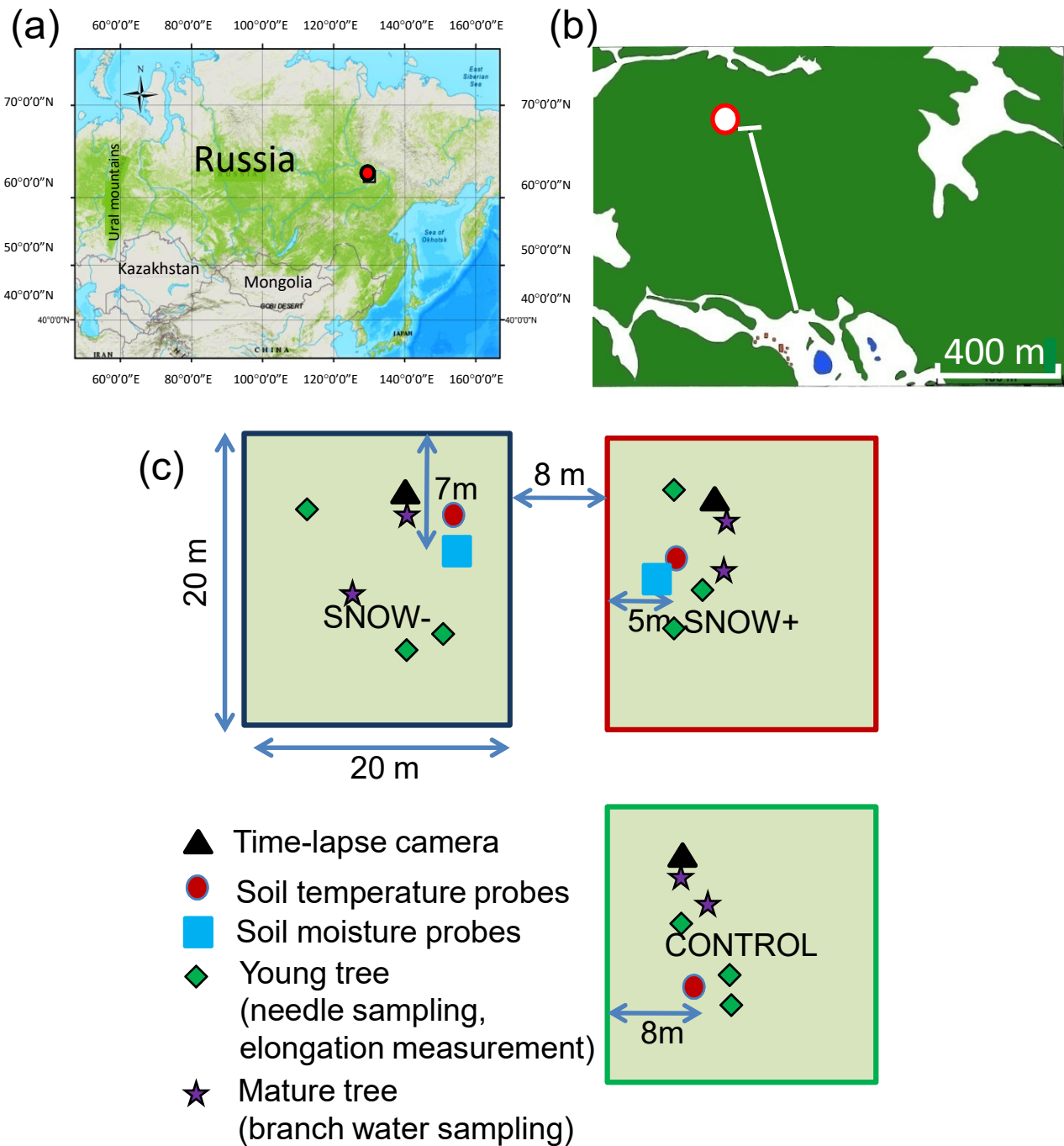


Figure 5. Maps of Eastern Eurasia with the locations of Yakutsk, Spasskaya Pad Forest Station, and experimental sites. Maps of (a) Yakutsk ( $62^{\circ} 03' N$ ,  $129^{\circ} 71' E$ ), (b) Spasskaya Pad Scientific Forest Station ( $62^{\circ} 14' N$ ,  $129^{\circ} 37' E$ ) with green and white areas indicating forest and grassland respectively, and red circle marking experimental sites, and (c) schematic maps of snow manipulation experimental sites. Purple stars show the mature sample larch trees used for stem sampling and green diamonds are young larch trees used for needle elongation observations and needle chemistry sampling.

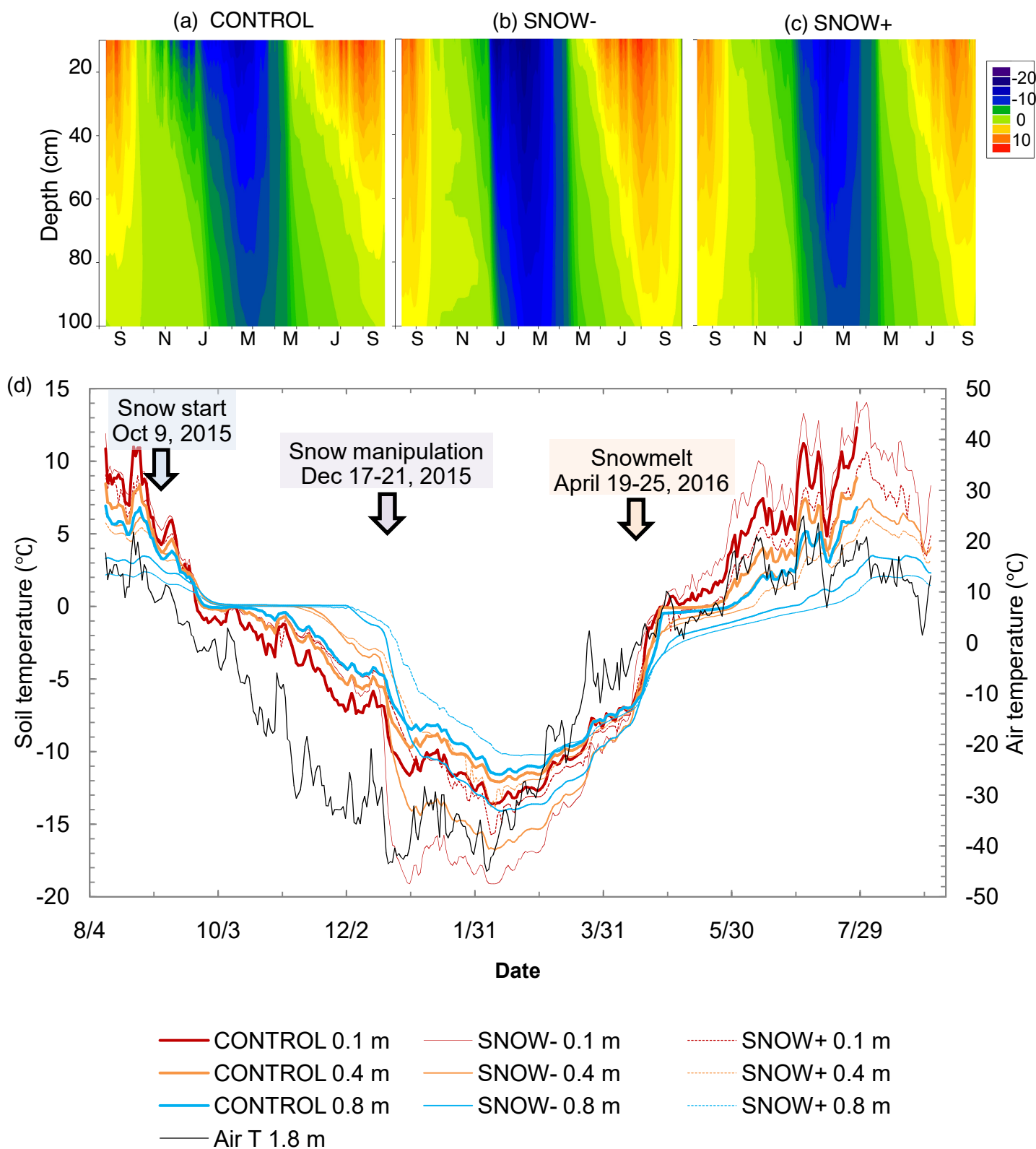


Figure 6. Depth distribution of isotherms and seasonal variation of soil temperature. Depth distribution of isotherms (daily averages of soil temperature) at the CONTROL (a), SNOW- (b), and SNOW+ (c) plots, seasonal variations in soil temperature at 0.1, 0.4, and 0.8 m, and air temperature in the CONTROL, SNOW- and SNOW+ plots (d), from August 2015 to August 2016 .

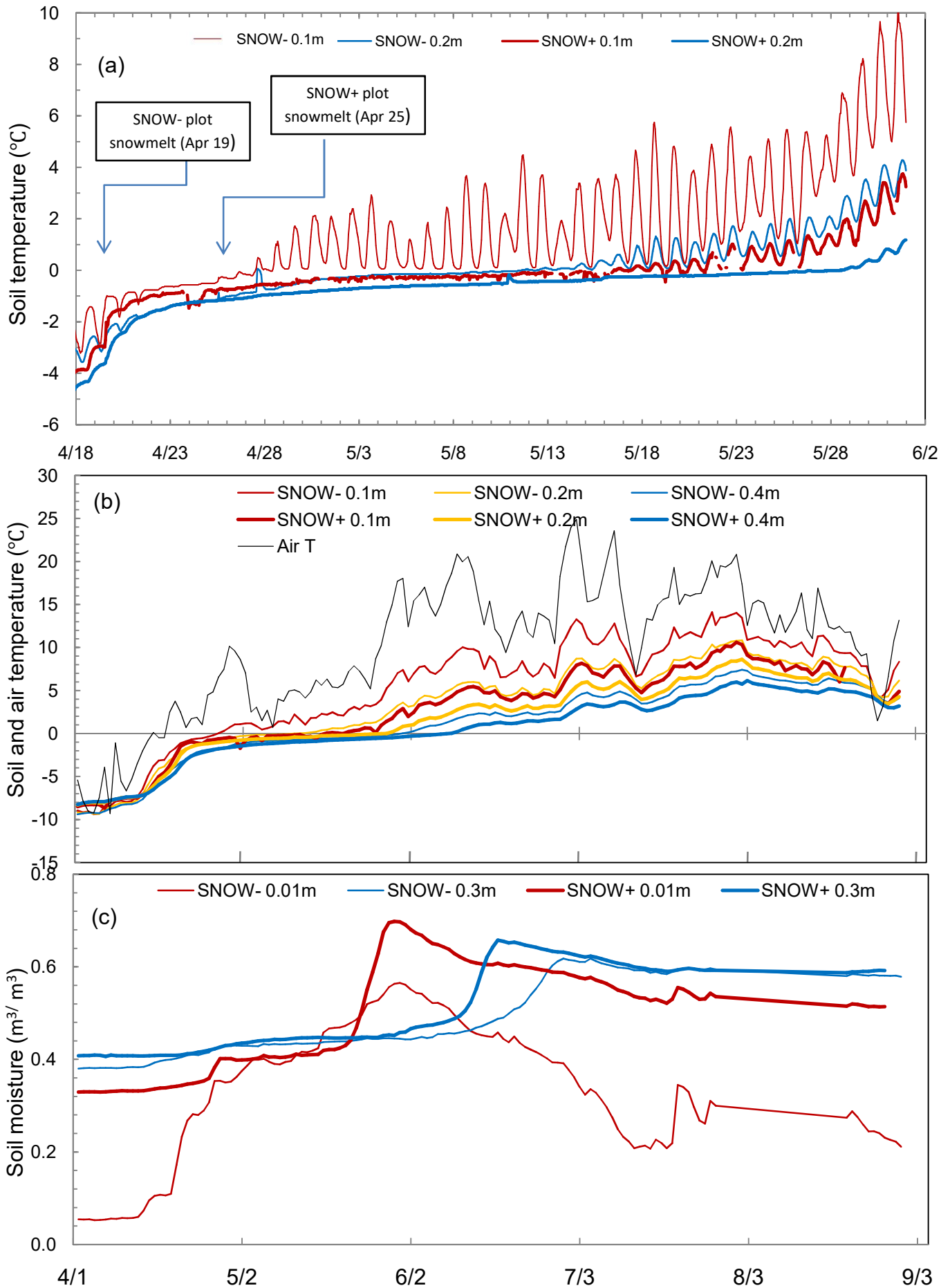


Figure 7. Soil and air temperature, and soil moisture in spring–summer 2016.

(a) Hourly soil temperature at 0.1 m and 0.2 m depth from April 18 to May 30, 2016; (b) daily average air and soil temperature at 0.1, 0.2, and 0.4 m depth, and (c) soil moisture at 0.01 m and 0.3 m depth from April to August in 2016 for SNOW- and SNOW+ plots. SNOW- and SNOW+ plots data are represented by thin and thick lines, respectively. 69

CONTROL



Apr 23

SNOW-



Apr 19

SNOW+



Apr 25

Figure 8. Snowmelt timing observed by time-lapse cameras at CONTROL, SNOW-, and SNOW+ plots.

Date of snowmelt was determined by a criteria of 50% ground surface appearance. Snowmelt dates were April 23, April 19, and April 25 for CONTROL, SNOW-, and SNOW+ plots, respectively.

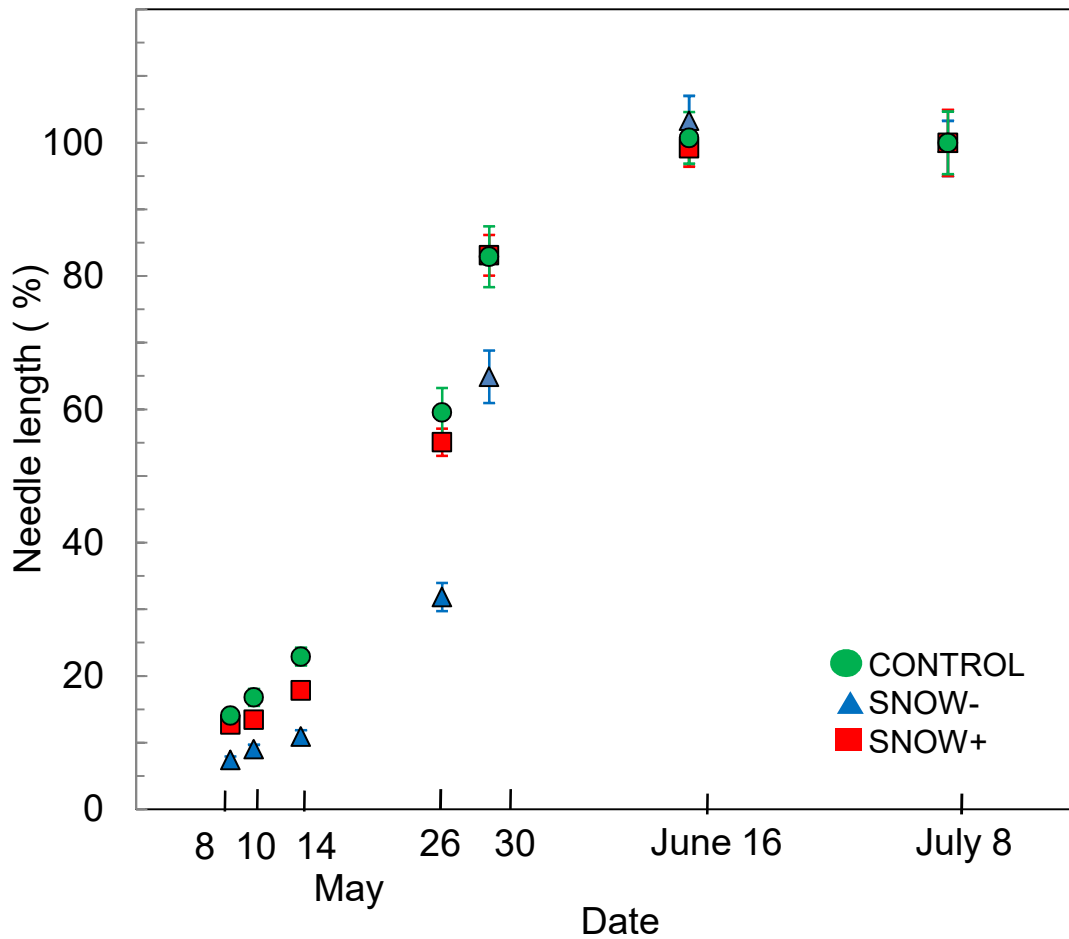


Figure 9. Growth of needle length in young larch trees in spring–summer 2016. Growth is shown as percentage of needle length at final observation date (July 8) in each plot. Average lengths at final observation date were 18.4, 21.1 and 25.1 mm for the CONTROL, SNOW- and SNOW+ plots respectively. Markers are averages and bars are standard errors in the CONTROL (green circle), SNOW- (blue triangle), and SNOW+ (red square) plots observed on May 8, 10, 14, 26, and 30, June 16, and July 8, 2016.

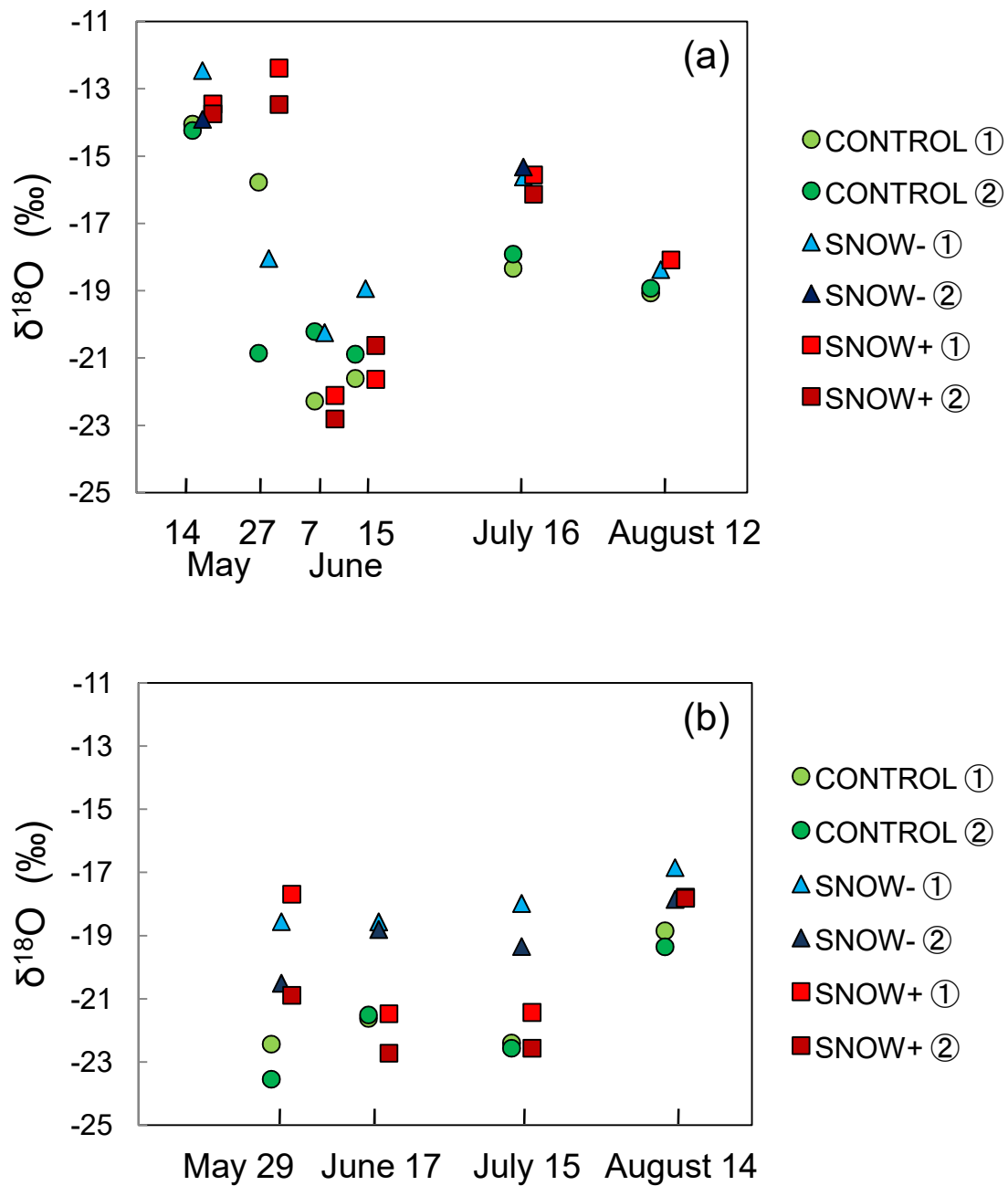


Figure 10. The  $\delta^{18}\text{O}$  values of stem water of larch trees. The  $\delta^{18}\text{O}$  values observed in (a) 2015 and (b) 2016 in the CONTROL (green circle), SNOW- (blue triangle), and SNOW+ (red square) plots are shown. “①” or “②” after plot name indicates duplicate sampling.



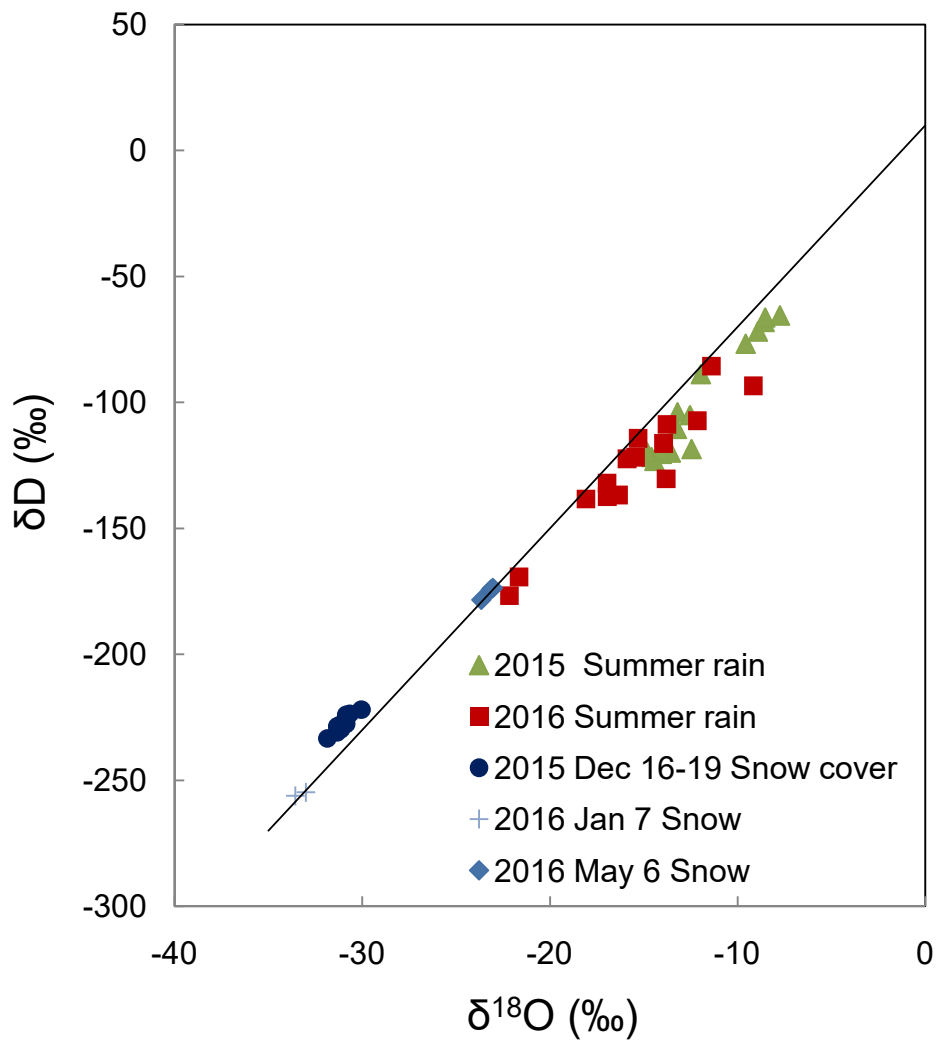


Figure 11.  $\delta D - \delta^{18}O$  plots of precipitation and snow.

Summer rainfall in 2015 (green triangle) and 2016 (red square), snow cover on December 16 to 19 in 2015 (blue circle), fresh snow on January 7 (blue cross) and May 6 (blue diamond) in 2016, and Global Meteoric Waterline are also shown. Snow cover sampled on December 16 to 19 is a snowpack sample, snow sampled on January 7 and May 6 is freshly fallen snow on the corresponding date.

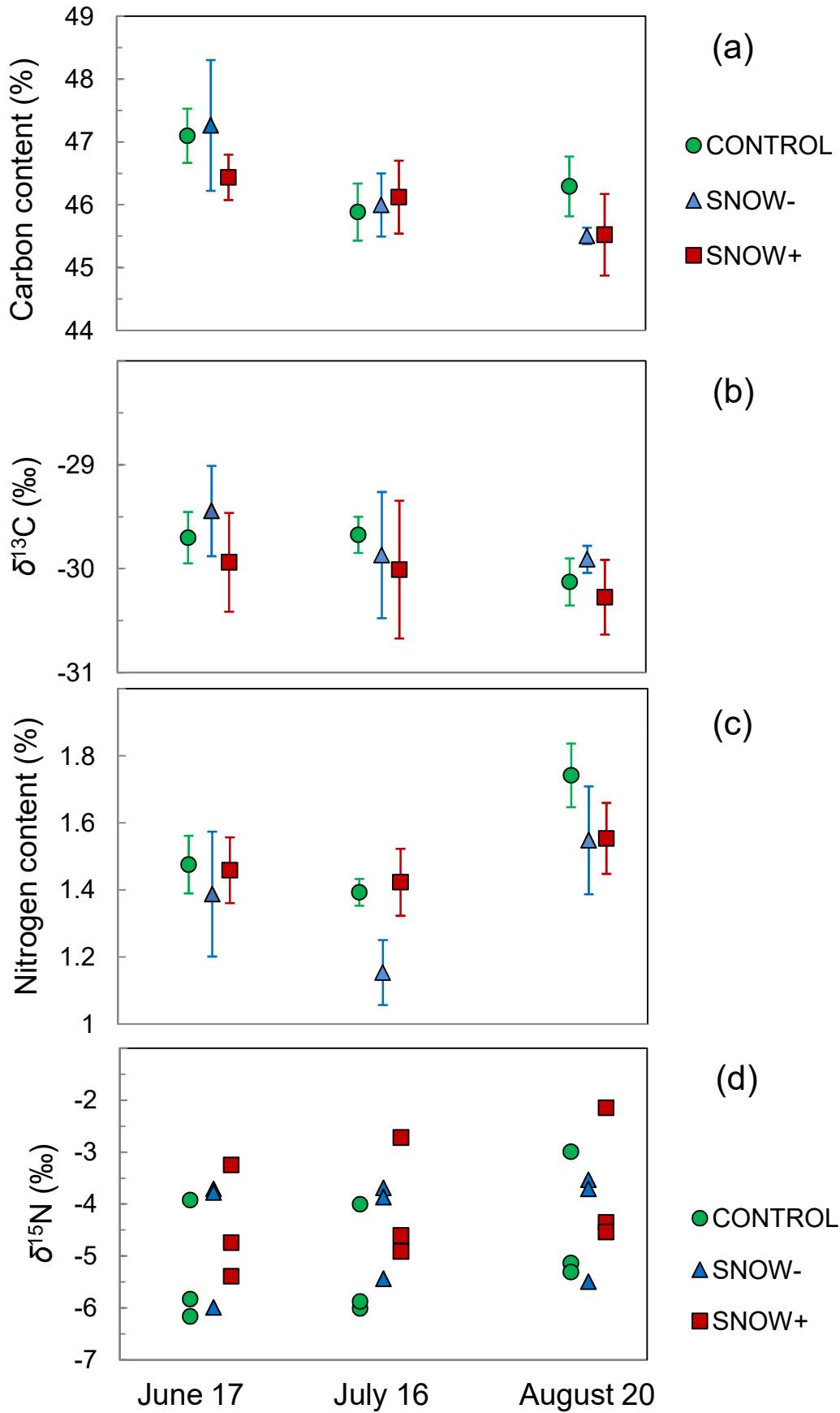


Figure 12. Carbon content,  $\delta^{13}\text{C}$ , nitrogen content, and  $\delta^{15}\text{N}$  of larch needles observed in 2016. (a) Carbon content, (b)  $\delta^{13}\text{C}$ , (c) nitrogen content, and (d)  $\delta^{15}\text{N}$  of larch needles ( $n=3$ ) in the CONTROL (green circle), SNOW- (blue triangle), and SNOW+ (red square) plots in 2016.

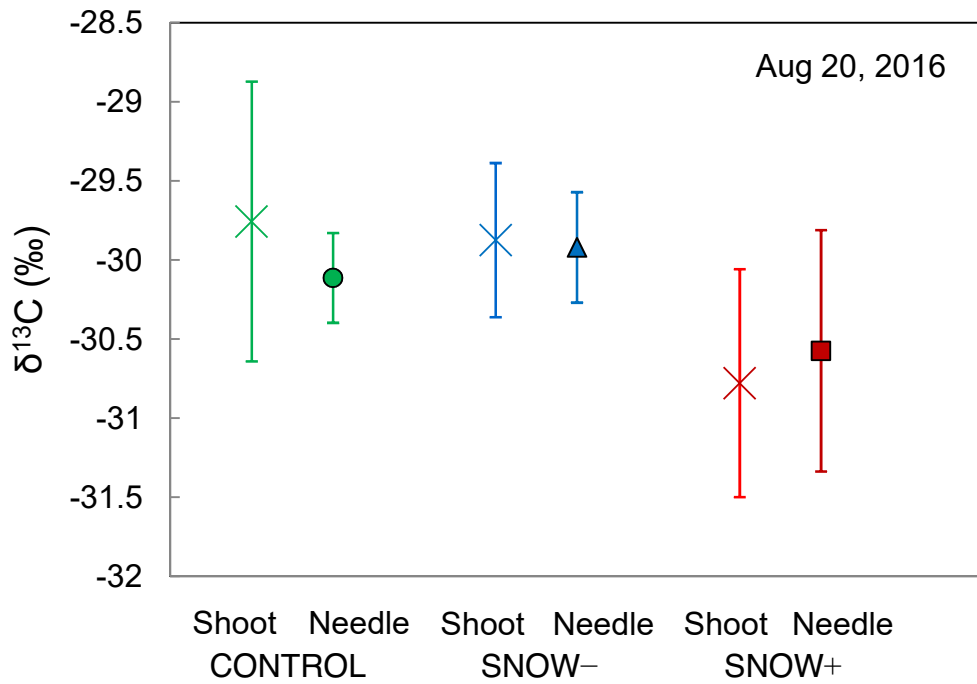


Figure 13. The  $\delta^{13}\text{C}$  values of current year shoots and shoot needles for young larch trees. Current year shoots ( $n=3$ ) and shoot needles ( $n=3$ ) of each tree sample at the CONTROL (green crossing and triangle), SNOW- (blue crossing and triangle), and SNOW+ (red crossing and square) plots sampled on August 20, 2016. Average and standard deviations are shown.

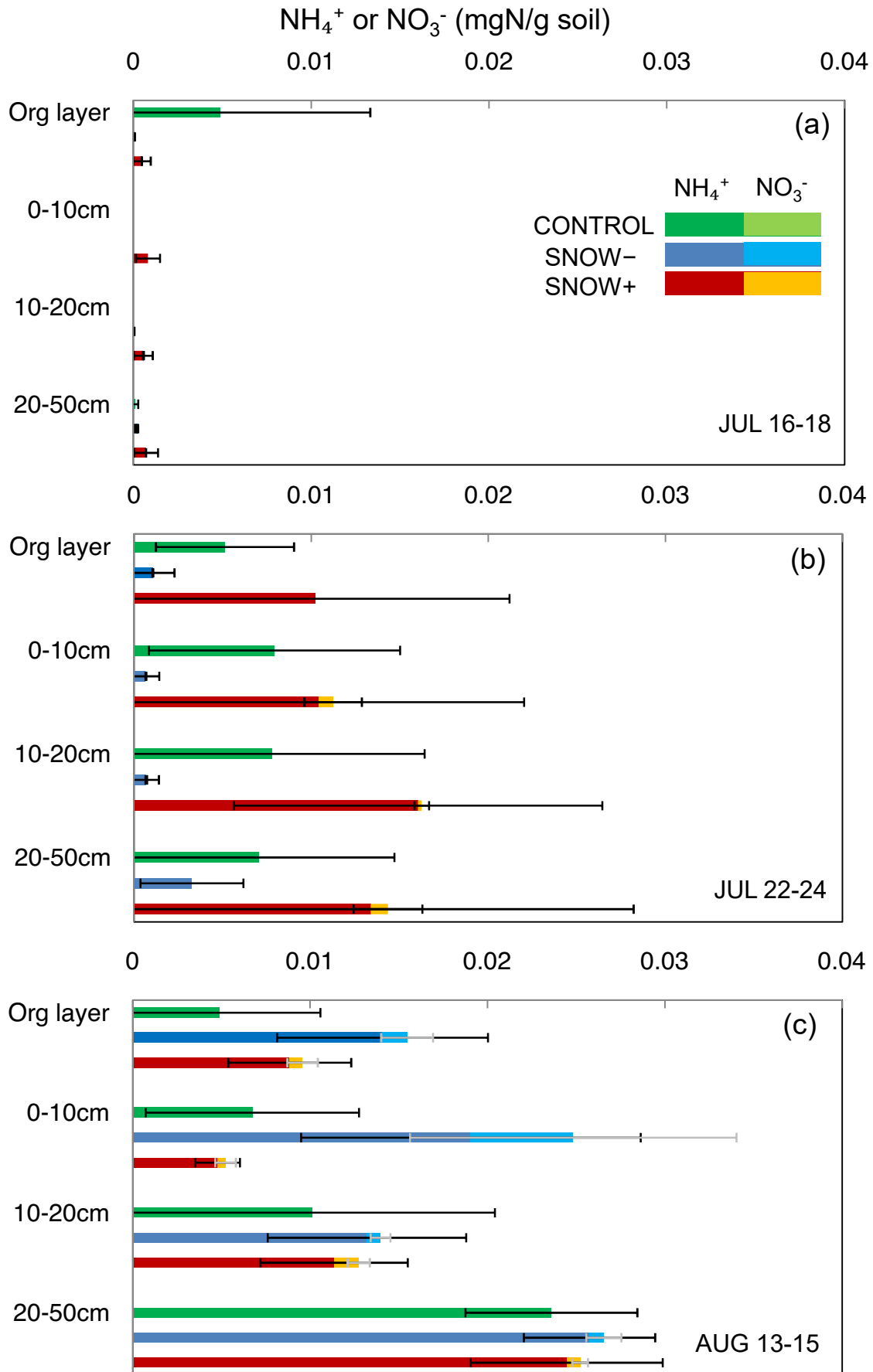
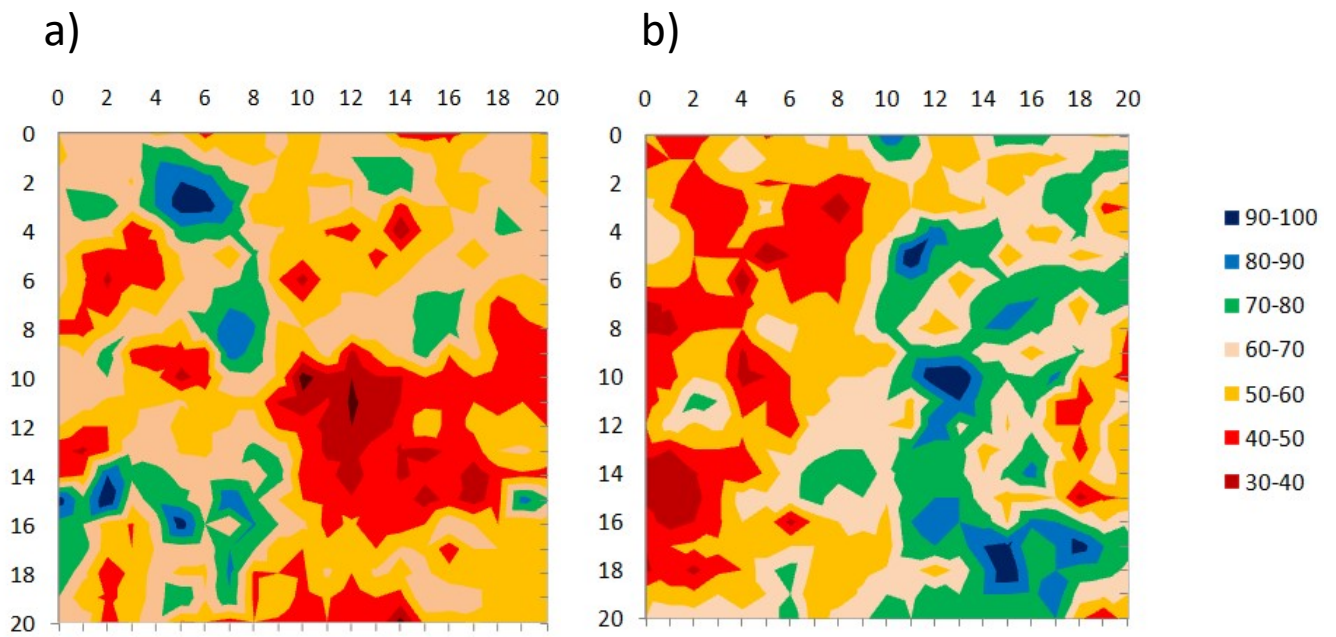


Figure 14. Soil inorganic nitrogen at each plot (n=4). Ammonium and nitrate observed in the organic layer and at 0–10 cm, 10–20 cm, and 20–50 cm in (a) mid-July, (b) end of July, and (c) mid-August at CONTROL (green), SNOW- (blue) and SNOW+ (red) plots. Ammonium and nitrate are shown by dark and lighter colors, respectively. No data means that the concentration was below the detection limit.



Supplement 1. Soil thaw depth in 2015 (before manipulation) at SNOW- (a) and SNOW+ (b) plots.