

## Evaluation of *Heracleum sosnowskyi* Frost Resistance after Snow Cover Removal in Early Spring

I. F. Chadin<sup>a,\*</sup>, I. V. Dalke<sup>a</sup>, and R. V. Malyshev<sup>a</sup>

<sup>a</sup>Institute of Biology, Komi Scientific Center, Ural Branch, Russian Academy of Sciences, Syktyvkar, 167982 Komi Republic, Russia

\*e-mail: chadin@ib.komisc.ru

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**Abstract**—The development of environmentally safe and cost-effective methods for controlling invasive species Sosnowsky's hogweed (*Heracleum sosnowskyi* Manden.) is an urgent issue for the European part of Russia. The article presents findings of an experiment on the effect of snow cover removal from the areas occupied by *H. sosnowskyi* in the early spring period (the beginning of March 2018) in the vicinity of the city of Syktyvkar (Komi Republic, Russia). The snow depth reached 100 cm on the intact plots; the sum of below-zero air temperatures measured at 6 a.m. constituted  $-448^{\circ}\text{C}$ , with a minimum of  $-29.0^{\circ}\text{C}$  during the experiment. The number of *H. sosnowskyi* plants of all age groups at the experimental plots (with removed snow cover) was shown to be significantly decreased. The median seedling density (pcs. per square meter) was equal to zero. Most of the surviving plants were located along the sides and in the corners of experimental plots. This can be explained by the higher temperature of soil on the borders of plots with an intact snow cover. The results of the experiment may be used for development of invasive plant eradication technology by removal of the snow cover. This technology can be suitable for kindergartens, schools, hospitals, and water protection zones, where the use of chemical methods of plant control is limited or prohibited. The obtained data set with respect to *H. sosnowskyi* monitoring is available in the repository of Zenodo.

**Keywords:** *Heracleum sosnowskyi*, biological invasion, frost resistance, snow cover, invasion management

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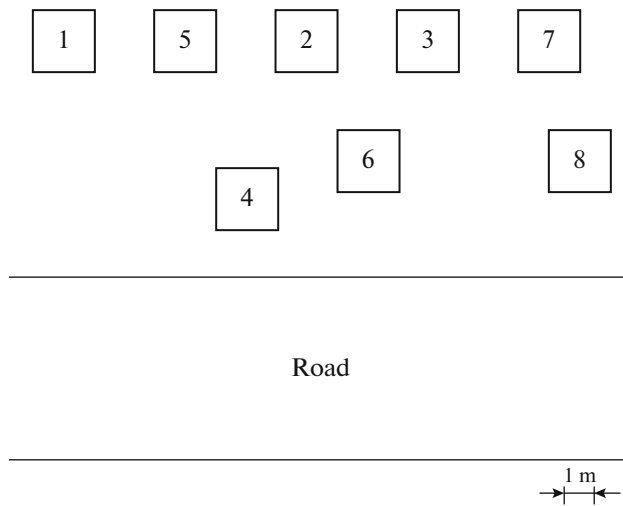
### INTRODUCTION

At present, mechanical, chemical, and thermal methods are applied to combat invasions of alien plant species (*Invasive Alien Species...*, 2001; *Invasive Species Management...*, 2009; Saunders et al., 2010). The spread and eradication of undesirable dense stands of the invasive Sosnowsky's hogweed (*Heracleum sosnowskyi* Manden.) species have attracted significant attention in Russia in the last decade. Along with other hogweed species, *H. sosnowskyi* was farmed on a large scale as forage crop in the Soviet Union and a number of East European countries in the middle of the 20th century (Satsyperova, 1984; Nielsen et al., 2005; Pyšek et al., 2012; Pergl et al., 2016; Ozerova et al., 2017). In the European part of Russia, *H. sosnowskyi* first became naturalized in the 1980s; in Siberia, the first finds of the species outside the agrocoenoses were reported in 2005 (Ebel' et al., 2018). Presently, plants of the species have been discovered in 54 subjects of the Russian Federation (Panasenko, 2017; Chadin et al., 2017; Ozerova and Krivosheina, 2018; Vinogradova et al., 2018).

Giant hogweeds are characterized by a high-level socioeconomic and environmental impact (Nielsen et al., 2005; Dergunova et al., 2012; Pergl et al., 2016;

Rajmis et al., 2016). These plants trigger dermatitis and burns in humans (Karimian-Teherani et al., 2007; Jakubowicz et al., 2012). Programs to control unwanted overgrowth of the species (*O gosudarstvennoi podderzhke...*, 2016; *O gosudarstvennoi podderzhke...*, 2017; *Ob oblastnom budzhete Leningradskoi oblasti...*, 2017) have been adopted by the subjects of the Russian Federation. The cost of mapping and eradication of *H. sosnowskyi* has amounted to nearly 314 million rubles in Russia since 2011 (Dalke et al., 2018).

Methods for the controlling and effectiveness of eradication measures with respect to *H. sosnowskyi* continue to remain a matter of urgency in rural locations (Dalke et al., 2018). As many as eight groups of methods to combat giant hogweed thickets have been suggested in the literature: soil (disk) plowing, pulling out of the soil manually (digging out), mowing, umbel cutting (removal), mulching (with a layer of covering materials), application of herbicides, exposure to microwave radiation, grazing (Nielsen et al., 2005; *Ecology and Management...*, 2007; Malyshev, 2014). The most effective proved to be repeated disk plowing and sowing substituting crops, chemical treatment, and application of covering materials (Dalke and Chadin, 2008, 2010). Mowing, soil plowing, and her-



**Fig. 1.** Location plan of the experimental (1–4 snow removed) and control (5–8 snow covered) plots in *Heracleum sosnowskyi* plant stands (lat. 61.646944° N, long. 50.757277° E).

bicide treatment are commonly preferred to control hogweed in Russia (Dal'ke et al., 2018). However, it is practically impossible to liquidate hogweed entirely by mowing (Dalke et al., 2015). Mechanical control methods require considerable resources, high-level organization of labor, and close monitoring (*Ecology and Management...*, 2007); no biological control methods have been developed to date (Reznik et al., 2008). Herbicide application is seen as the most effective for eradication of *H. sosnowskyi* plants in large areas (Sampson, 1994; Caffrey, 2001; Nielsen et al., 2005; *Ecology and Management...*, 2007; Yakimovich et al., 2013). Chemical treatment is considered to be the first step in implementation of the strategy for eradication of plants with a high fecundity, including *H. sosnowskyi* (*Ecology and Management...*, 2007). Considering the restricted use of pesticides and agrochemicals within the limits of populated localities, water protection zones, specially protected natural areas (*Gigienicheskie trebovaniya...*, 2016), there is an urgent need to develop and implement safe and effective methods for containment of unwanted vegetation.

In a seasonal climate environment, the thermal regime of the soil is among the significant factors affecting plant survival rates in wintertime. It is possible to influence this regime by altering the depth of the snow cover. Notwithstanding its high level of effectiveness in using environmental resources and resistance to unfavorable conditions during the growing season (Dalke et al., 2015; Veselkin et al., 2017), *H. sosnowskyi* was reported to be killed following dormancy in a snowless period (Aleksandrova, 1971) or as a result of severe conditions in late fall (Khantimer, 1974). Accordingly, we put forward a hypothesis that dense stands of plants can be destroyed by altering the thermal regime of the soil during the cold season.

The objective of our work is to investigate the effect of snow cover removal on the state and size of the *H. sosnowskyi* population. Provided a drop in the soil temperature causes the death of plants, this will allow the development of a safe and reasonable method for regulating the species abundance in the areas where the use of the chemical control methods is prohibited.

## MATERIALS AND METHODS

The investigations were conducted from March 5 through June 10, 2018, in the middle boreal subzone in the vicinity of Syktyvkar. Experimental plots were set up within the typical *H. sosnowskyi* habitats along roads (Fig. 1). Places for the experiment were selected on the basis of the median density of generative shoots specific to dense monospecies stands of this species, i.e., 1 plant/m<sup>2</sup> (Dalke et al., 2015). The number of generative *H. sosnowskyi* plants from the previous year was determined according to retained dry generative shoots and their traces in the soil.

The first group included the plots where the snow was removed manually with shovels down to the ground surface (experiment,  $n = 4$ ); plots of the second group were left snow covered (control,  $n = 4$ ). The plots were cleared from fallen snow within one to two days from the moment of the precipitation.

The size of approximately 2 × 2 m was defined for the plots at the experiment setup. To determine their area more accurately after the snow melted, we measured lengths of all sides and diagonals of quadrangle to an accuracy of 1 cm. The area of the plots ( $S$ ) was estimated from the formula

$$16S^2 = 4d_1^2d_2^2 - (b^2 + d^2 - a^2 - c^2)^2,$$

where  $a$ ,  $b$ ,  $c$ , and  $d$  are the sides and  $d_1$  and  $d_2$  are the diagonals of a plot.

Seedlings and plants of *H. sosnowskyi* rapidly regrow after the disappearance of the snow cover and can be easily distinguished from other species on the basis of the shape and size of its leaves. The number of the studied plants of different ontogenetic age stages was assessed three times during their growth (May 12, May 25, and June 10). The ontogenetic age stages of individual plants were determined according to the extent to which the lamina was incised (Satsyperova, 1984). We factored in the quantity of plants in the first year (sprouts and seedlings) and older than a year. The number of sprouts and seedlings was estimated in a series of 10–11 sample measurements uniformly along the plot diagonals using frames 0.028 m<sup>2</sup> in area. Data with respect to the density of hogweed plants were converted to plants/m<sup>2</sup>.

The projective cover of *H. sosnowskyi* plants was assessed from photographs using the image processing software package ImageJ (Konukhov, 2012; Schneider et al., 2012). The pictures were taken with a Canon

**Table 1.** Effects of exposure of plants to low temperatures during dormancy on population characteristics of *Heracleum sosnowskyi* plants (June 10, 2018)

Indicator	Control	Experiment	<i>P</i> *
Density of plants older than a year (median value), plants/m <sup>2</sup>	3.3	1.8	0.02
Density of first-year plants (median value), plants/m <sup>2</sup>	321	0.0	$4.79 \times 10^{-12}$
Height of plants older than a year (median value), cm	50	22	$1.68 \times 10^{-5}$
Projective cover, %	64	15	0.02

\* *P*-value calculated by the results of the Kruskal–Wallis test.

PowerShot A480 camera elevated to the height of about 4 m by means of telescopic fishing rod with the lens axis viewing the nadir with Picavet suspension (Verhoeven et al., 2009).

The functional condition of buds located on the subterranean portion of adult vegetative plant shoots was evaluated according to their respiration rate (intensity) and water content at the end of the growing period. Specimens of Sosnovsky hogweed were uprooted and transferred to the laboratory. Apical perennating buds were located and cut off from the subterranean part of the shoot. Combined samples of several buds were used for measurements. The fresh weight of one bud sample was about 1.5 g. The bud samples were placed in a thermostatically controlled chamber. The respiration was measured within the temperature range from 4 to 47°C on the basis of CO<sub>2</sub> release using an LI-7000 infrared differential gas analyzer (United States) (Sivkov and Nazarov, 1990). To determine the dry matter content, the samples were dried to constant weight in thermostat at 105°C and weighed on an analytical balance.

Meteorological data (air temperature, snow depth, and sum of precipitation) were obtained from the Raspisanie pogody (Weather Timeline) website (2018) and the website of Avtomatizirovannaya informatsionnaya sistema obrabotki rezhimnoy informatsii (AISORI) (Automated Information System of Processing Sensitive Data) (2018) for the weather station in Syktyvkar (synoptic index 23804).

Descriptive statistics were employed for the data analysis. The value of the indicator “number of plants” differed from normal distribution (the Shapiro–Wilk test). Therefore, in addition to mean values, the median, range, and minimum and maximum values were utilized to describe the sample. The control and experimental plots were compared using the nonparametric Kruskal–Wallis test. Statistical calculations were performed in the R environment (R Core Team, 2017). The standard error of the mean is shown after the ± sign. The dataset obtained as a result of monitoring *H. sosnowskyi* and the source code with computations in the R language are available in the Zenodo repository (Chadin et al., 2018).

## RESULTS

In March 2018, the depth of the snow cover varied from 60 to 100 cm and averaged 80 cm. The experimental plots were completely cleared from the snow cover during the setup of the experiment (March 5, 2018) and after the fall of new precipitation.

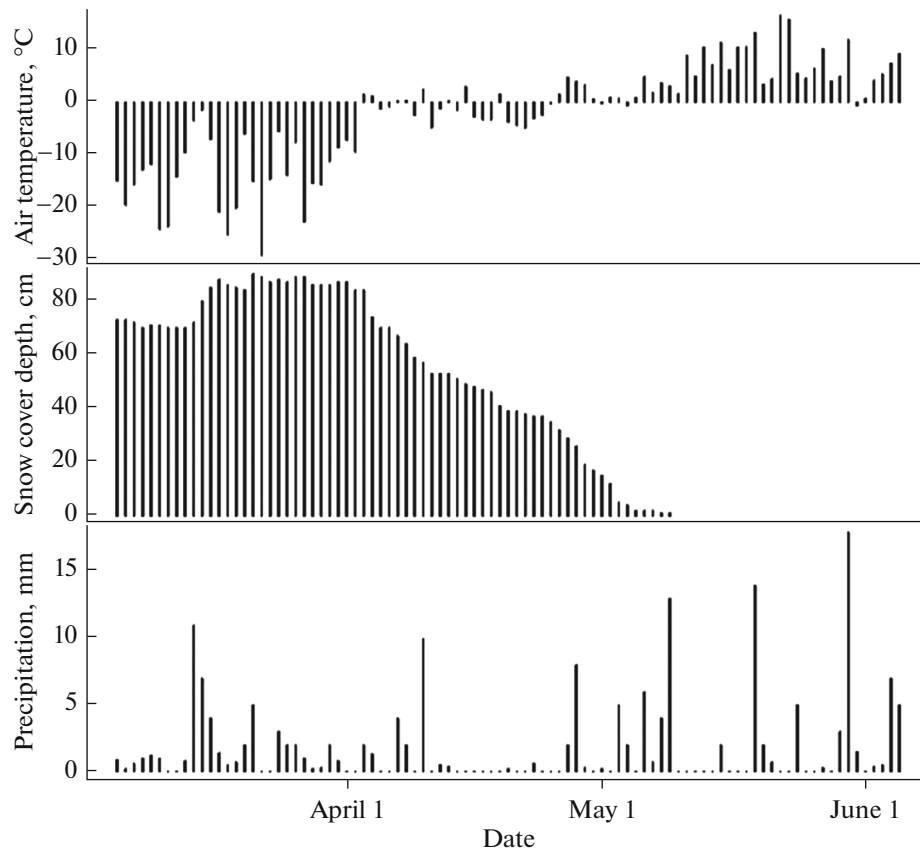
The dynamics of temperature, precipitation amount, and snow cover depth is detailed in Fig. 2. Approximately 50 mm of precipitation fell in March; the snow depth reached 70 cm. The morning temperatures were observed to vary daily from –5 to –30°C in the middle of March (March 10, 11, 18, and 22). The snow cover did not exceed 10–15 cm in April when the temperature reached values above zero. In May, the average temperature was at about 6°C; the snow entirely melted; about 80 mm of precipitation fell predominantly in the form of rain. Overnight frosts with a temperature drop to –0.7°C were observed at the beginning and end of May.

The median value of the generative plants in dense *H. sosnowskyi* stands from the previous year was 0.9 plants/m<sup>2</sup> for both the control and the experimental plots. According to the Kruskal–Wallis test, the density of the plants older than a year was significantly lower on the experimental plots (*P*-value 0.02) as compared to control plots throughout various observation periods (Table 1, Fig. 3).

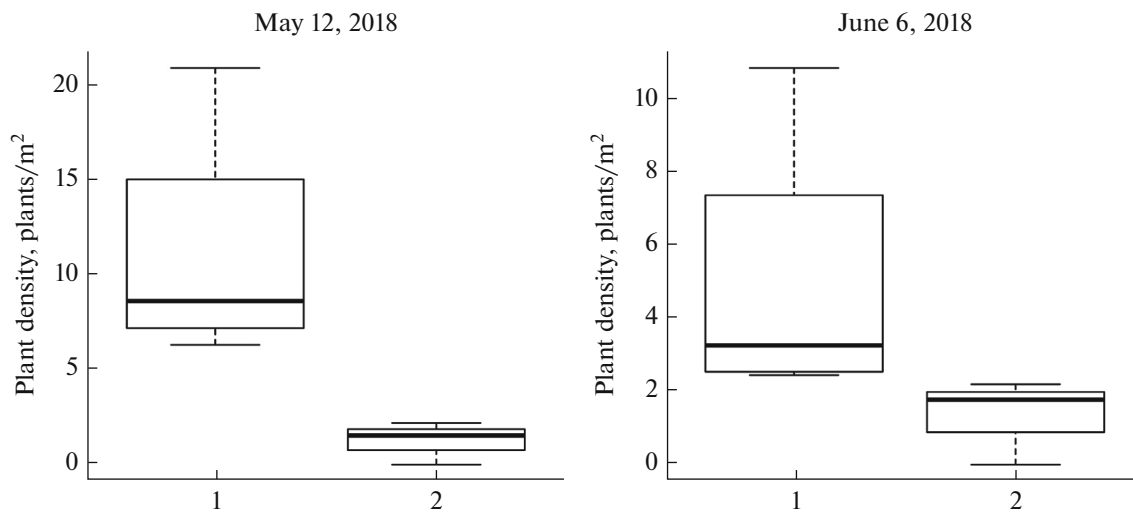
Experimental plots (Table 1, Fig. 4) exhibited a low number of first-year plants or seedlings (median 0, mean 36). The number of the seedlings was considerably higher on control plots (median 321, mean 331). The significant differences between the experiment and control are supported by the Kruskal–Wallis test (*P*-value  $\ll$  0.001).

At the beginning of June, a significant decrease was observed in projective cover (median 64% and 15% for the control and experiment, respectively, *P*-value 0.02) and height of the plants older than a year (median 50 and 22 cm, *P*-value  $\ll$  0.001) on experimental plots as compared to the control (Table 1, Fig. 5).

After *H. sosnowskyi* plants had been killed, experimental plots devoid of hogweed plants were observed to have aftergrowth of *Elytrigia repens* (L.) Nevski., fireweed (*Chamerion angustifolium* ssp. *angustifolium* (L.) Holub), and nettles (*Urtica dioica* L.).



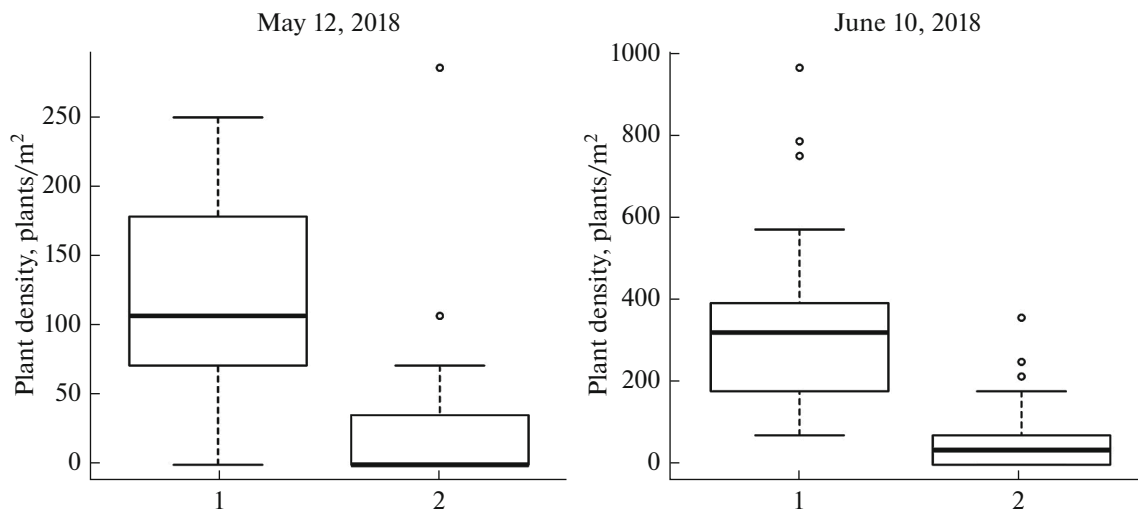
**Fig. 2.** Air temperature at 6 a.m. (upper panel), snow cover depth (middle panel), and daily total precipitation (lower panel) during the period of observation of *Heracleum sosnowskyi* plants (March 5, 2018–June 5, 2018).



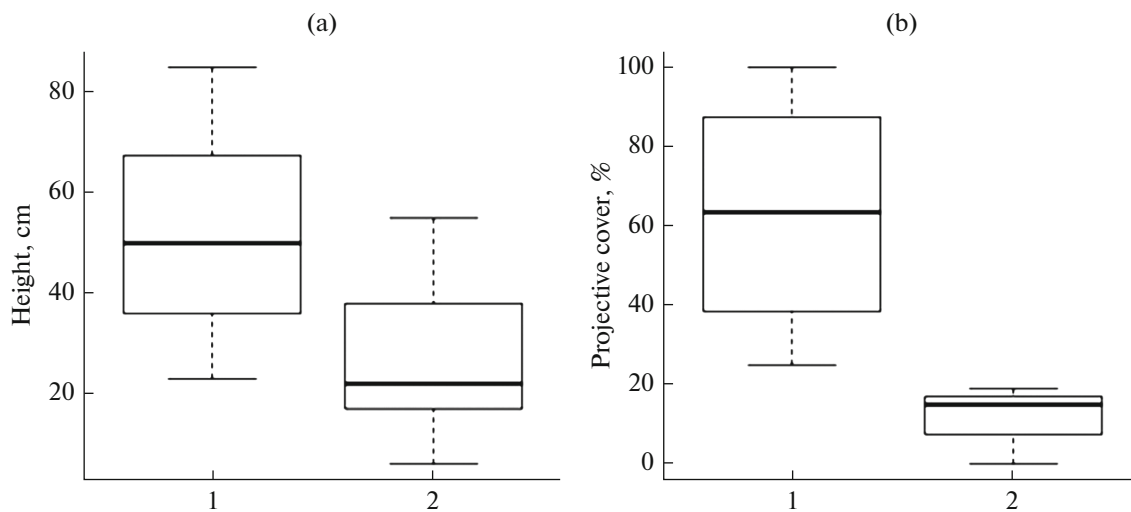
**Fig. 3.** Density of *Heracleum sosnowskyi* plants older than a year (plants/m<sup>2</sup>) on (1) control and (2) experimental plots during different observation periods. The boxplot function in the R environment was used to create the box-and-whisker plot. The box is drawn from the first to third quartiles; the line in the middle of the box is the median. The ends of whiskers are the maximum and minimum of the sample.

To assess the mechanisms that ensure frost resistance of the overwintering *H. sosnowskyi* plants, we estimated the water content and respiratory rate in api-

cal buds located on subterranean parts of the shoot. Apical buds of the adult vegetative plants contained  $85.1 \pm 0.5\%$  of water or as converted  $5.8 \pm 0.3$  mg



**Fig. 4.** Density of *Heracleum sosnowskyi* plants of the first year (plants/m<sup>2</sup>) on (1) control and (2) experimental plots. The boxplot function in the R environment was used to create the box-and-whisker plot. The box is drawn from the first to third quartiles; the line in the middle of the box is the median. The ends of whiskers are the maximum and minimum of the sample, excluding outliers. The values of outliers are indicated with circles.



**Fig. 5.** Height of *Heracleum sosnowskyi* plants older than a year (a) and projective cover (b) on (1) control and (2) experimental plots (June 10, 2018). The boxplot function in the R environment was used to create the box-and-whisker plot. The box is drawn from the first to third quartiles; the line in the middle of the box is the median. The ends of whiskers are the maximum and minimum of the sample.

H<sub>2</sub>O/mg of dry tissue matter in October. The respiration rate of the buds was 1.5 mg CO<sub>2</sub>/g of dry wt per hour at a temperature of about 5°C.

## DISCUSSION

According to the data obtained with respect to the number of the generative plants, specimens older than a year, and seedlings on the control plots, the coenopopulations in the area where experimental plots were set up were similar to the typical dense monospecies

stands of *H. sosnowskyi* of the middle boreal subzone (Dalke et al., 2015).

Snow depth is known to considerably affect the temperature of the soil, particularly in its upper layers, where perennating buds of perennial plants are stored. Overwintering plants are frequently killed by frost at the drop of the air temperature to -20°C and snow depth up to 10 cm. In this situation, difference between the soil temperature at the depth of the tillering zone and the minimum air temperature is approximately 9°C. Snow cover 40 cm in depth properly insulates the cold, while at its depth of 75 cm, soil tempera-

ture variations are minimized. Good snow cover ensures minimum mean soil temperature within the range from  $-4$  to  $-8^{\circ}\text{C}$  at the depth of 3–5 cm throughout the winter (*Agroklimaticheskie...*, 1973).

Biologically, *H. sosnowskyi* plants are known for their resistance to low temperatures throughout the growing season and good winter hardiness. Nevertheless, the presence of snow cover is a necessary condition for the winter survival and preservation of the coenopopulation. Snow prevents the plants from being killed by frost and provides conditions for the seed stratification (Skupchenko, 1989). In the taiga zone, the seedlings can be observed under the snow as early as April (Hogweed Seeds ..., 2013). Massive germination occurs immediately after melting of the snow cover. Terminal and lateral (auxiliary) perennating buds found on the caudex are buried in the soil at a depth of 10–15 cm and are protected from mechanical damage (Satsyperova, 1984; Dalke et al., 2015). Under favorable conditions, plants of *H. sosnowskyi* continue to vegetate until the late fall. The meristems of buds effectively stock energy for growth during vegetation and adapt well to a wide-range change in temperature (Maslova et al., 2018).

When measured, the respiration rate of the hogweed perennating buds revealed that they tend to transition to a state of exogenous dormancy in the late fall and winter and start growing as soon as the conditions become favorable. At the end of growing season (October), generation of the new shoots, flowering, and fruit bearing was observed once the adult plants were transferred to the optimum growth conditions (Dalke et al., 2018). There have been recorded instances of hogweed growing in winter. Exceptional conditions of warming up to  $2.5^{\circ}\text{C}$  in December 1965 triggered growth processes in these plants. The subsequent dramatic drop in temperature in the absence of the melted snow cover led to a kill of 89% of *H. sosnowskyi* and 56% of *H. lehmannianum* Bunge and *H. ponticum* (Lipsky) Schischk. ex Grossh. (Aleksandrova, 1971). Similarly, hogweed cultivation was impeded by severe late fall conditions and insufficient amount of heat, when the sum of active temperatures above  $10^{\circ}\text{C}$  during the growing season did not exceed  $800\text{--}1000^{\circ}\text{C}$ , on tundra meadows (Khantimer, 1974).

According to the Weather Timeline portal, during the observation period after the snow had been removed, the sum of below-zero temperatures taken at 6 a.m. was  $-448^{\circ}\text{C}$  with the minimum of  $-29.0^{\circ}\text{C}$  on the experimental plots. During this period, the temperature dropped to below  $-20^{\circ}\text{C}$  at least seven times. Thus, the temperature could have dropped to below  $-10^{\circ}\text{C}$  at a depth of *H. sosnowskyi* perennating bud bedding (15 cm).

In our experiment, removal of the snow cover significantly contributed to a density reduction in *H. sosnowskyi* plants of different ages, their growth characteristics, and projective cover. Additionally, the exper-

imental plots almost entirely lacked its sprouts. The likely explanation is that either the seedlings were killed after frost penetration below the critical temperature or the seeds failed to emerge from dormancy because of inability to undergo stratification.

Survival of part of the plants older than a year on the experimental plots can be explained by a higher soil temperature at the boundary with intact snow cover (edge effect). The majority of the surviving plants were found along the sides and at the corners of the plots.

The main reason for a plant cell to be killed by below-zero temperatures is ice formation, dehydration, and mechanical damage to cell structures by ice crystals (Tumanov, 1963; Samygin, 1969; Trunova, 2007). Accumulation of growth inhibitors and protective substances in buds and change in content and state of water is one of the mechanisms of adaptation of perennial plants to a period of below-zero temperatures (Klimov, 2001; Trunova, 2007). The effects of plant exposure to low temperatures significantly depend on free and bound water content in the tissues. Dormant buds lack outward signs of growth and are highly resistant to dehydration and unfavorable environmental factors (Tumanov, 1979). During the period of deep dormancy (December–January), the water content was  $0.5\text{--}0.7$  mg  $\text{H}_2\text{O}/\text{mg}$  of dry wt in woody *Betula pendula*, *Populus nigra*, and *Syringa vulgaris* and about  $1.0$  mg  $\text{H}_2\text{O}/\text{mg}$  of dry wt in *Vaccinium myrtillus* and *Vaccinium vitis-idaea* prostrate shrubs (Maslova et al., 2013; Malyshev and Atoyan, 2018). In winter, reduction of water content in the buds to  $0.5$  mg  $\text{H}_2\text{O}/\text{mg}$  of dry wt is sufficient for ensuring the resistance of *Picea obovata* and *Pinus sylvestris* to temperatures ranging from  $-35$  to  $-40^{\circ}\text{C}$  (Alaudinova, 2007).

The high respiration rate of perennating plants at  $5^{\circ}\text{C}$  indicates the absence of deep dormancy, which may translate into a low level of frost resistance in their tissues.

As opposed to “meristematically” active tissues of phanerophytes and chamephytes, meristems of the cryptophytes feature considerably higher water content, e.g.,  $9.0$  mg  $\text{H}_2\text{O}/\text{mg}$  of dry wt at the rhizome apex in *Achillea millefolium* (Maslova et al., 2013). According to our data, at the end of growing season (October), apical buds of *H. sosnowskyi* contained  $5.8$  mg  $\text{H}_2\text{O}/\text{mg}$  of dry wt, which is considerably higher than in the buds of woody plants and prostrate shrubs, but is comparable to the amount of water in subterranean meristematic tissues of other cryptophytes. Directly determined freezing temperatures of water solutions in perennating bud apex tissues revealed that more than half of the water contained in them undergoes a phase transition to ice at a temperature of  $-12^{\circ}\text{C}$  (Malyshev, 2018).

Resistance of early seedlings of *H. sosnowskyi* to frost penetration is somewhat lower compared to

perennating buds. Radicles of its seedlings which were plucked in March under the snow cover contained 7.3 mg H<sub>2</sub>O/mg of dry tissue weight. The seedling were completely frozen and killed at a temperature of –8°C (Maslova et al., 2018). When compared to seedlings, seeds in this species are more resistant to the damaging effects of below-zero temperatures. In September, the freezing temperature of seed cell solution was the same as in apical buds (–12°C). Cryoresistance of the seeds is ensured by their low (0.5 mg H<sub>2</sub>O/mg of dry wt) water content (Malyshev, 2018).

In the middle boreal subzone, the meristematic potential of *H. sosnowskyi* plants includes up to 20000 seeds and about 20 perennating buds of plants older than a year per 1 m<sup>2</sup> of the coenopopulation (Dalke et al., 2015). All these loci of perennation are sensitive to frost penetration and may be killed completely in the absence of snow cover.

The effect detected in the experiment can serve as a basis for the development of technology for controlling the numbers of *H. sosnowskyi* plants by means of snow cover removal. In regions with suitable climatic conditions, this method for eradication of undesirable dense stands can be in demand for kindergartens, schools, medical institutions, and water protection zones, where the use of chemical hogweed control methods is restricted or prohibited.

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#### COMPLIANCE WITH ETHICAL STANDARDS

*Conflict of interests.* The authors declare that they have no conflict of interest.

*Statement on the welfare of animals.* This article does not contain any studies involving animals performed by any of the authors.

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