

## Juvenile growth and frost damages of poplar clone OP42 in Latvia

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**Abstract.** Short rotation plantations in the northern Europe are commonly established using poplar clone OP42 (*Populus maximowiczii* Henry × *P. trichocarpa* Torr. and Gray). We assessed its growth and suitability to the climate in central part of Latvia at juvenile age. Trees that had formed single stem were significantly higher ( $121 \pm 2.5$  cm), thicker ( $7.1 \pm 0.48$  mm) and had longer branches ( $32 \pm 1.5$  cm) than trees that had formed multiple stems. In beginning of the second growing season all trees had died stems and 19.6% of them formed new shoots from the ground level. The sprouting trees had random spatial distribution in the field. Regardless of the number of stems, the sprouting trees were significantly lower ( $110 \pm 3.9$  cm) than the dead trees ( $119 \pm 2.0$  cm). During the repeated assessment about one month later, proportion of the sprouting trees increased up to 44%, but the detected relations between measured traits of sprouting and dead trees remained. Clone OP42 had serious frost induced damages also in autumn phenology experiment (96% trees with severely damaged leaves). Our results suggest that frost prone sites are not suitable for establishment of plantations of OP42 clone.

**Key words:** *Populus maximowiczii* × *P. trichocarpa*, clone OP42, sprouting, freeze-thaw, wood dysfunction.

### INTRODUCTION

Bioeconomy concept is rapidly gaining popularity in both policy and science (Staffas et al., 2013; Pülzl et al., 2014), and it is closely linked to European Commission goal to develop a resource-efficient and low carbon economy by 2050 (EC, 2011). Bioeconomy emphasizes use of renewable raw materials (Scarlat et al., 2015), including necessity to increase biomass production in sustainably managed industrial crops (EC, 2012). Woody biomass is one of the largest sources of renewable energy in EU and its consumption is expected to increase in the following decades (Openshaw, 2011; Bentsen & Felby, 2012; Lauri et al., 2014). However native tree species in the Northern Europe (with exception of grey alder) have relatively long rotation period – from ca. 40 years for birch and aspen up to more than 100 years for pine and oak (Rytter et al., 2013); and therefore have limited flexibility to respond to changes in wood demand in a short period of time. Hence, during the last decades fast growing tree species are increasingly used (Gailis & Jansons, 2010; Uri et al., 2010; Jansons et al., 2013), and currently

ca. 50,000 ha of short rotation plantations is established in Europe (Don et al., 2012). *Populus* spp. are among the most energy-efficient perennial energy crops (Djomo et al., 2015), and are commonly used in the Northern Europe (Don et al., 2012) as well as in the Baltic States (Tullus & Vares, 2005; Bardule et al., 2016; Zeps et al., 2016). Poplars are mainly grown for bioenergy under rotation of 8–10 years (Djomo et al., 2015), but could also be grown for solid wood and veneer production in a rotation 20 years (Christersson, 2010). In Latvia, results from poplar trials indicate superior yield than the most productive native species (Jansons et al., 2014); yet results from other Northern Europe countries indicate that even higher yields could be gained (Karačić et al., 2003; Tullus et al., 2013). However, poplars in the Northern Europe are reported to suffer from frost damage at different severity (Ferm et al., 1989; Christersson, 1996; Ilstedt, 1996; Karačić et al., 2003; Christersson, 2006; Pliura et al., 2014). Pliura et al. (2014) studying poplar hybrids in two juvenile clonal trials in Lithuania found out that clones with late growth cessation were of the worse survival, possibly due to delayed shoot maturation and winter damage. In Sweden, two poplar clones had shown high biomass in one of the two experimental sites, while were completely eliminated by frost at the other (Karačić, 2005). One of the most widely used poplar clone worldwide and also in the Northern Europe is hybrid *Populus maximowiczii* (Henry) × *P. trichocarpa* (Torr. and Gray) clone OP42 (Taerøe et al., 2015). It is included in trials as far north as 60°N (Johansson & Karačić, 2011; Johansson & Hjelm, 2012b; Hjelm et al., 2015), indicating its suitability for climate at northern latitudes. Clone OP42 is also included in several trials representing different site conditions in Latvia and considered to be perspective (unpublished data). The hypothesis of the study is that poplar clone OP42 does not suffer from frost damages in Latvia. The objectives are to characterize the meteorological conditions, juvenile growth and frost damages for clone OP42, and compare the autumn phenology of OP42 to other poplar clones as a potential cause of autumn frost damages.

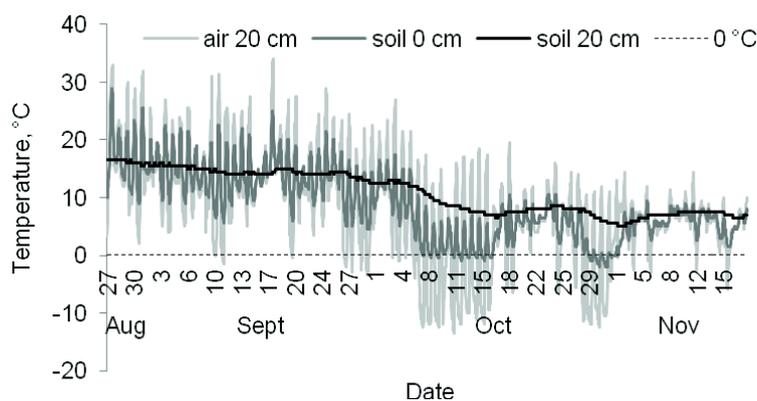
## MATERIAL AND METHODS

The study was done in poplar trial located in central part of Latvia (56°34'N, 24°31'E), near Vecumnieki.

Vecumnieki trial was established in spring 2015 on drained peat soil with pH 6,0 using 30 cm long cuttings of clone OP42 (*Populus maximowiczii* (Henry) × *P. trichocarpa* (Torr. and Gray)) obtained from trees grown in western part of Latvia. Trees were planted in 17 rows in a density of 4 × 2 m, total number of planted cuttings 3,393. Weed control (ploughing and herbicide) was carried out once in autumn 2014 (before planting) and manual weed control (ca. 25 cm around the tree) during the growing season of 2015.

In the trial, at the end of the growing season (27/08/2015–18/11/2015) temperature measurements once per hour were done at three different heights – (1) air temperature was measured 20 cm above the ground; (2) soil temperature was measured at the surface level and (3) at the 20 cm depth (Fig. 1). Air temperature first dropped below 0 °C in September 11 and until October 6 negative air temperature recurred occasionally – in nine out of 22 days. During this period, the longest frost (temperature below 0 °C) occurred in September 27 (9 hours, minimal hourly temperature -2 °C) and September 30 (7 hours, minimal hourly temperature -3 °C). In October 7 frost became more intense – air temperature was below 0 °C for 14 hours and dropped down

to  $-12.5\text{ }^{\circ}\text{C}$ ; similar temperatures remained for 10 days – mean minimal hourly temperature was from  $-9.5$  to  $-13.5\text{ }^{\circ}\text{C}$ , duration of frost was from 11 to 16 hours per day. Similar period of frost occurred in October 28–31. Likewise, on the soil surface two periods of temperature below  $0\text{ }^{\circ}\text{C}$  occurred in October, and remained for nine (October 8–16) and three (October 29–31) days. For the first time in the growing season (October 8) soil temperature at the surface level dropped below  $0\text{ }^{\circ}\text{C}$  for 2 hours; five days later – for 7 hours (minimal hourly temperature  $-1\text{ }^{\circ}\text{C}$ ). Duration of negative soil surface temperature reached 15 hours per day ( $-2\text{ }^{\circ}\text{C}$ ) in October 30 and 31. However, during the observed period no negative soil temperature at 20 cm depth was recorded.

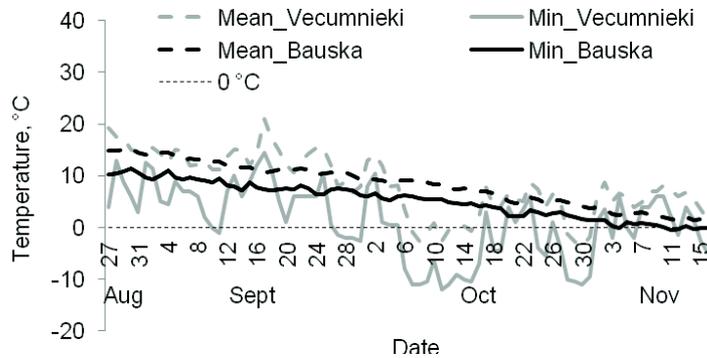


**Figure 1.** Temperature of air (20 cm above ground) and soil (at surface level – 0 cm – and in 20 cm depth) in Vecumnieki trial during 27/08/2015–18/11/2015.

Data of the long-term (30-year mean) measurements for the respective period were obtained from the nearest (distance ca. 30 km) weather station, located near Bauska ( $56^{\circ}22'\text{N}$ ,  $24^{\circ}13'\text{E}$ ; Latvian Environment, Geology and Meteorology Centre). During the analysed period, mean diurnal air temperature did not decrease below  $0\text{ }^{\circ}\text{C}$ ; but minimal temperature (30-year mean of the minimal temperature of the date) first dropped below  $0\text{ }^{\circ}\text{C}$  in November 11 (Fig. 2) – considerably later than in the study year. As can be seen the study year represent very specific meteorological conditions in the autumn thus authors did not had a chance to repeat the study with larger set of clones and sites during next growing season even so the experimental design was prepared.

In the beginning of June 2016, damage of stems (one-year-old shoots, emerged in the previous growing season) were observed. All trees had completely withered stems (no growth from the previous year shoots occurred), and part of them had new emerging shoots from the ground level. Tree height ( $\pm 1\text{ cm}$ ) and diameter ( $\pm 1\text{ mm}$ ; measured for 410 trees in 2 rows) at the stem base (root collar) of the highest one-year-old shoot (emerged in 2015) of trees were measured, number of stems (emerged in 2015) and number of branches (longer than 5 cm) per tree were counted, and length of branches ( $\pm 1\text{ cm}$ ) was measured. Trees that had more than three stems and more than three branches were pooled into groups ‘more than 3 stems’ and ‘more than 3 branches’, respectively. For each tree occurrence (0/1) of browsing damage and occurrence of new emerging shoots (further called ‘sprouting’ (1) and ‘dead’ (0) trees) were recorded. In

total, 3,025 trees were measured. In the middle of July, assessment of sprouting/dead trees was repeated in a part of the trial (5 rows, 1,018 trees).



**Figure 2.** Air temperature in the study year in Vecumnieki trial and long-term measurements of the nearest weather station (Bauska) for the period of 27/08–18/11. ‘Mean\_Vecumnieki’ – mean diurnal temperature, calculated from hourly measurements; ‘Min\_Vecumnieki’ – the lowest recorded diurnal temperature; ‘Mean\_Bauska’ – mean diurnal temperature, calculated from 30-year data of mean temperature of the respective date; ‘Min\_Bauska’ – mean of the 30-year lowest diurnal temperature of the date.

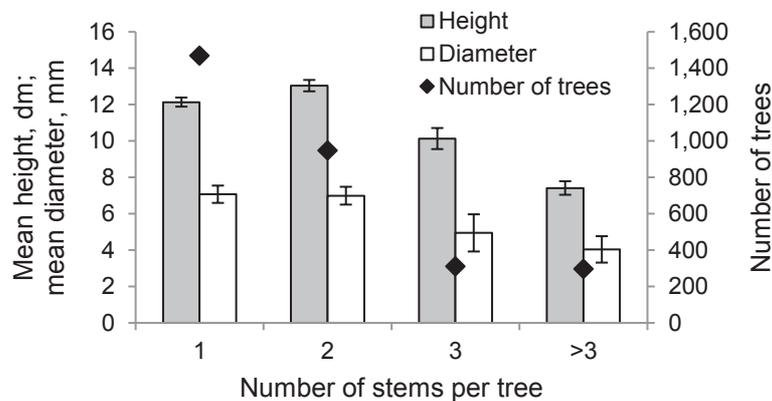
To characterize the frost hardiness of poplars the data obtained in another poplar trial located in eastern part of Latvia (56°41’N, 25°58’E), near Kalsnava, were used additionally. Besides OP42, 17 other clones were included in this trial where autumn phenology observations were made. Frequently recurring freeze-thaw cycles at the same period as in Vecumnieki were observed in Kalsnava; the frost damages for leaves and stems of two-year-old trees were assessed visually (evaluated at five grade scale) in October 15<sup>th</sup>. The detailed description of methods, as well as the frost hardiness comparison at the clone level was performed by Lazdiņa et al. (2016). In our study, clones were pooled into five groups according to their origin (Sweden, Italy, Germany, Latvia) to estimate the relative frost hardiness of OP42 in comparison to other poplar clones.

The Shapiro-Wilk test was used to assess the normality of the data. Differences of mean height, diameter, and length of branches were assessed using one-way analysis of variance (i) between trees that had formed one or multiple stems, and (ii) between trees that had formed one, two, three or more stems. The Chi-squared test was used to assess (i) distribution of the number of branches, proportion of sprouting trees and proportion of browsed trees between trees that had formed one or multiple stems and among trees that had formed one, two, three or more stems, (ii) distribution of the proportion of sprouting trees between browsed and not browsed trees, (iii) distribution of proportion of sprouting trees among plantation rows. The statistical analysis of differences between proportions of trees among the leaf and stem damage grades was hampered by insufficient number of trees in several of the grades. Pearson’s correlation was used to assess relationship between proportion of sprouting trees and mean tree height of the plantation rows. Spearman’s correlation was used to assess the relationships between (i) number of branches and number of stems per tree, (ii) length of branches and number of

branches, (iii) length of branches and number of stems per tree. Spatial autocorrelation of the sprouting trees was assessed using Moran's I. All tests were performed at  $\alpha = 0.05$ . Mean values and their confidence interval are shown both in text and figures.

## RESULTS

In Vecumnieki at the end of the first growing season 89% of the planted cuttings had formed shoots. About half (49%) of trees had formed one stem, and about half had formed multiple stems – 31% of trees had formed two, 10% of trees three and 10% of trees four and more stems. Tree height varied from 10 to 251 cm (mean  $118 \pm 1.8$  cm), and trees that had formed multiple stems were significantly ( $P < 0.001$ ) lower (mean height of the highest stem  $114 \pm 2.6$  cm) than trees that had formed one stem (mean height  $121 \pm 2.5$  cm). No overall trend between number of stems and tree height was observed (Fig. 3).



**Figure 3.** Mean tree height and diameter at root collar (both  $\pm$  confidence interval) and number of trees according to a number of stems per tree for the clone OP42 in Vecumnieki.

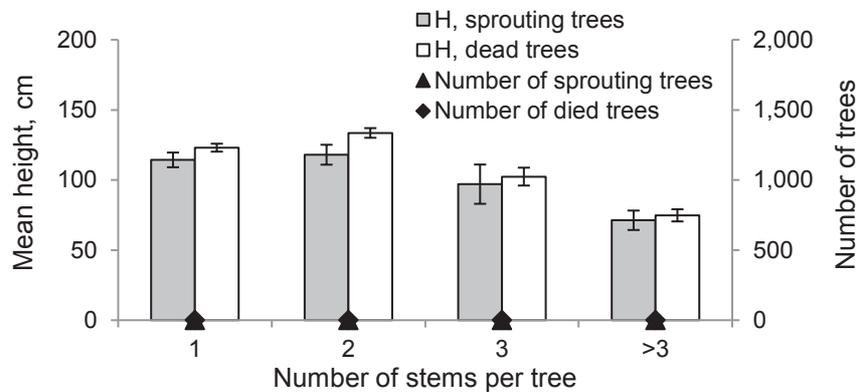
Branches were observed for 30% of trees, mean number of branches was 3.3 (from 1 to 42). One branch was observed for 11% of trees, two branches for 6%, three branches for 4% and more than three branches – for 9% of trees. Proportion of number of branches between trees that had one and trees that had multiple stems was similar ( $P = 0.86$ ). No relation between number of stems and number of branches was observed ( $r_s = -0.060$ ,  $P = 0.071$ ).

Mean length of branches was  $30 \pm 1.0$  cm, and trees that had formed one stem had slightly, but significantly ( $P < 0.001$ ) longer branches ( $32 \pm 1.5$  cm) than trees that had formed multiple stems ( $28 \pm 1.4$  cm). However, it was similar ( $P > 0.05$ ) for trees that had formed one stem ( $32 \pm 1.5$  cm) and trees that had formed two stems ( $31 \pm 2.1$  cm), but trees that had formed three ( $26 \pm 2.5$  cm) and more ( $25 \pm 2.4$  cm) stems had significantly shorter branches. Mean branch length had weak but significant correlation with number of branches ( $r_s = 0.089$ ,  $P = 0.007$ ) and number of stems ( $r_s = -0.154$ ,  $P < 0.001$ ).

Mean diameter at stem base was  $6.6 \pm 0.32$  mm. Similarly to tree height, trees that had formed smaller number of stems were thicker (Fig. 3). Mean diameter of trees that

had formed one and two stems was similar ( $P > 0.05$ ;  $7.1 \pm 0.48$  mm and  $7.0 \pm 0.49$  mm, respectively), and both significantly differed from diameter of trees that had formed three and more stems ( $4.9 \pm 1.0$  mm and  $4.0 \pm 0.73$  mm, mutually similar ( $P > 0.05$ )). Browsing damages were observed for 7.6% of trees. Proportion of browsed trees significantly differed between trees that had formed one, two, three and more stems ( $P < 0.001$ ), and it was 10.2, 6.6, 3.2 and 2.4%, respectively.

In the late June 2016, new emerging (sprouting) shoots from the ground level were found for 19.6% of trees. The sprouting trees had reached slightly, but significantly ( $P < 0.001$ ) lower height in the previous growing season ( $110 \pm 3.9$  cm) than the dead trees ( $119 \pm 2.0$  cm), regardless of the number of stems (Fig. 4). Among trees that had formed one stem, height of the sprouting and dead trees was  $114 \pm 5.2$  cm and  $123 \pm 2.8$  cm, respectively; among trees that had formed multiple stems –  $106 \pm 5.7$  cm and  $116 \pm 2.9$  cm, respectively.

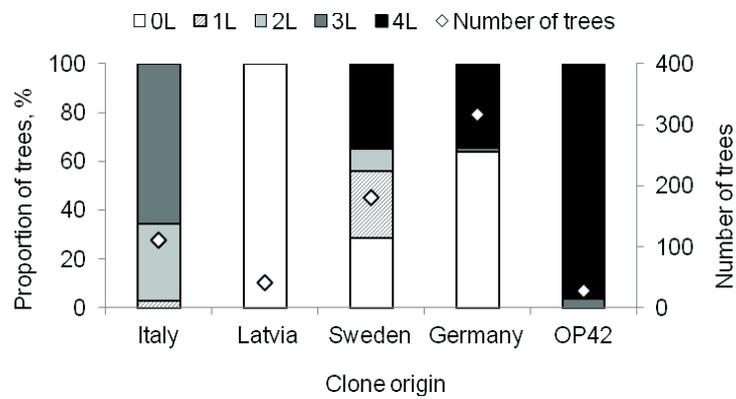


**Figure 4.** Mean height (H;  $\pm$  confidence interval) and number (N) of sprouting and dead trees according to number of stems per tree for the clone OP42 in Vecumnieki.

Proportion of the sprouting trees that had formed one, two, three and more stems was similar ( $P = 0.89$ ) – 19.4; 20.1; 20.0 and 18.2%, respectively. Significant ( $P = 0.006$ ) difference of proportion of the sprouting trees was found between browsed (12.6%) and not browsed (20.1%) trees. Proportion of the sprouting trees differed significantly ( $P < 0.001$ ) between the trial rows, but no relation ( $P > 0.90$ ) between proportion of the sprouting trees and mean tree height in the row was found. Spatial distribution of the sprouting trees in the field was random ( $P > 0.05$ ).

During the repeated assessment of sprouting/dead trees, proportion of sprouting trees was notably increased and reached 44%. However, the observed relation between measured traits remained – the sprouting trees were significantly ( $P < 0.001$ ) lower than the dead trees ( $111 \pm 4.6$  cm and  $123 \pm 4.5$  cm, respectively). Height of trees that had formed one and multiple stems was similar ( $P = 0.058$ ) –  $121 \pm 4.5$  cm and  $115 \pm 4.7$  cm, respectively. Among trees that had formed one stem, height of the sprouting trees ( $116 \pm 6.2$  cm) was significantly ( $P = 0.03$ ) lower than that of the dead trees ( $126 \pm 6.4$  cm). Similarly, among trees that had formed multiple stems, height of the sprouting trees ( $107 \pm 6.8$  cm) was significantly ( $P = 0.002$ ) lower than that of the dead trees ( $121 \pm 6.4$  cm).

Frost damage of the same frost event was assessed in another progeny trial in Kalsnava. Analysis of two-year-old trees reveals differences between clone origins. All trees of clones that were collected across Latvia were dormant (Grade 0L) and had no frost damage of leaves. Clones of Italian origin had all trees still growing, and 66% of trees had moderately damaged (Grade 3L) leaves but no severe damage (Grade 4L) was observed. Clones from Germany had relatively large proportion (64%) of dormant trees; but among still growing trees 96% had severely damaged (Grade 4L) leaves. Similarly to clones of Italian origin, all trees of clone OP42 were still growing at the time of frost event, but in contrast, trees of OP42 were more intensely damaged – 96% of trees had severely (Grade 4L) and 4% of trees had moderately (Grade 3L) damaged leaves (Fig. 5).



**Figure 5.** Proportion of two-year-old ramets with different leaf damage grades (0L, 1L, 2L, 3L and 4L) according to clone origin in Kalsnava. Damage grades: ‘0L’ – leaves were already fallen before the freezing event and buds were set; ‘1L’ – visually intact leaves; ‘2L’ – several damaged leaves; ‘3L’ – most of leaves in the current year leading shoot were damaged; ‘4L’ – most of leaves on the whole tree were damaged.

Stem damage was less frequent – in total 8% of trees had stem damage of different intensity. No stem damage was observed for clones from Latvia, which also had no leaf damage, and for clones from Italy, which had moderate leaf damage. Similarly, clones from Sweden and Germany had 1% and 15% of trees with mild stem damage, respectively. Clone OP42 had severely damaged leaves but no stem damage was observed.

## DISCUSSION

The results from rather rare combination of meteorological conditions in autumn were reported even so the study design was not initially intended for that. Considering the increasing use of the clone OP42 in Latvia and other Baltic states we did see the necessity to inform about the data so that land owners would have as objective information as possible while selecting the planting material. Although the result from the autumn phenology experiment in Kalsnava is not a direct replication of study in Vecumnieki, still it comes to similar conclusion on the risk of frost damages of the particular clone and provides a plausible reasoning for the observed problem.

Frost induced damage is not exclusively caused by extremely low temperatures; damage type and severity depends on frost event timing, duration and pattern, and the highest risk of frost damage occurs during the transition period between annual cycle of growth and dormancy (Charrier et al., 2015). We studied freeze-thaw damage in the early autumn. The freeze-thaw cycles are common to initiate embolism – bubbles are caused by dissolved air that freezes out of the sap (Tyree & Sperry 1989; Brodersen & McElrone, 2013), and sufficiently large bubbles may fill xylem conduits hampering water transport during the thawing (Pittermann & Sperry, 2006).

The first freeze-thaw cycles of the growing season occurred in the middle of September, and can be characterized by mild frost (ca. -2 to -3 °C). In the middle of October, more intense freeze-thaw occurred (Fig. 1) – temperature dropped below zero for 11–16 hours repeatedly for 10 successive nights, while the maximum temperature at day was from 6.5 to 18 °C. During the studied period the longest thaw period was 13 hours, indicating that trees experienced repeated embolism while the hydraulic conductivity had not recovered from the previous freeze-thaw cycle. Experiments of Just & Sauter (1991) showed that hydraulic conductivity of *P. × canadensis* stem segment decreased by ca. 60% after one freeze-thaw cycle, and almost no recovery was obtained during the first hour after thawing. It took 19 hours at the room temperature (21 °C) to recover 90% of the initial conductivity and about 2 days to recover completely.

Extremely high mortality in the studied site might be explained by timing of the particular freeze-thaw event. At the time of the frost all trees of clone OP42 were actively growing in the Kalsnava trial (Fig. 5), also a study of *P. tremula* × *P. tremuloides* in Latvia has revealed strong relation between height growth intensity and daily mean temperature (Zeps et al., 2015) and many other studies have showed delayed growth cessation for northward transferred species (Howe et al., 2000; Christersson, 2006; Friedman et al., 2008). Hence, most presumably clone OP42 was still growing also in Vecumnieki. During the growing season trees have higher water content than during the dormancy (Mäkinen et al., 2008; Pallardy, 2008), and more severe freeze-thaw damage is showed for trees that have high water content before the freezing occurs (Cox & Zhu, 2003). Also temperature regime before freezing significantly influences damage severity. *Betula* trees that were exposed to longer thaw period before winter freezing had increased percent length of shoot dieback and reduced length of new emerging shoots (Zhu et al., 2000). Longer thawing period was associated with increased dehardening of roots and root damage, thus, reducing root pressure, and resulted in poor recovery of embolism (Zhu et al., 2000).

Timing of the freeze-thaw cycle is also showed to affect tree recovery. *P. balsamifera* had different pattern of recovery after autumn and spring frost (Hacke & Sauter, 1996) – in the autumn when trees still had some leaves, recovery was slow and remained incomplete during the next growing season; in contrast, recovery after spring frost was complete in two months. Tree ability to recover largely depends on its vitality and damage severity. Zhu et al. (2000) have suggested that shoot damage might be directly dependent on ability of the unembolised vessels to maintain water supply for stem and branches. In turn, Cox & Zhu (2003) found that diffuse-porous trees that undergo permanent residual embolism in combination with root and shoot freezing injuries experienced higher bud mortality and shoot dieback than trees that can recover before next freeze-thaw cycle occurs.

Poplars are commonly propagated by cuttings, and trees develop shallow root system with no taproot and most lateral roots found near (within ca. 30 cm) soil surface (Puri et al., 1994; Crow & Houston, 2004; Johansson & Hjelm, 2012a). Fine roots are responsible for water uptake for trees (Block et al., 2006), and the highest biomass of fine roots was found down to 5 cm (Al Afas et al., 2008), 10 cm (Crow & Houston, 2004) depth for several poplar clones. During the studied period only slight temperature drop below zero on soil surface and no negative temperatures at the 20 cm depth was observed during the measurements (Fig. 1), therefore the particular frost event is not likely to cause substantial root damage. However, frequent recurrence of freeze-thaw cycles can cause accumulated embolism during autumn/winter (August–March) season (Sperry & Sullivan, 1992). Most deciduous trees refill frost embolized vessels in spring, and at this time nearly all conduits may be filled with air, causing as much as ca. 90% decrease of hydraulic conductivity (Sperry & Sullivan, 1992). Christensen-Dalsgaard & Tyree (2013) have observed ca. 90% loss of conductivity already after one light frost event, indicating high sensitivity of *P. deltoides* × (*P. laurifolia* × *P. nigra*) clone Walker. Later studies showed that for poplars most of the accumulated embolism occurred after the first few frost events (Christensen-Dalsgaard & Tyree, 2014). Autumn frost damage hamper winter hardening, which in turn increases vulnerability to damage (von Fircks, 1992), thus it could be assumed that root damage might occurred during the following winter when temperature dropped down to -25 °C (data from Latvian Environment, Geology and Meteorology Centre).

Root damage hamper the recovery of the hydraulic conductivity – Sperry (1993) has found that embolism of not-damaged *Betula* trees recovered from 81 to 88% during one month (April to May), while trees with root damage had 75% embolism after two months (in June). Similarly, no recovery of hydraulic conductivity was observed for *P. × canadensis* during more than three months if root pressure was absent (Hacke & Sauter, 1996). The reduced hydraulic conductivity, in turn, is showed to negatively affect timing of bud break – the lower the hydraulic conductivity at the end of the winter, the later the bud break (Wang et al., 1992). The observed emergence of new shoots of the otherwise vigorously sprouting clone OP42 (Johansson & Hjelm, 2012b) in this study was rather late and poorly performed, suggesting that trees were struggling to either rely on the remaining unembolized conduits or try to grow new conduits (Hacke & Sauter, 1996; Améglio et al., 2002).

We found that the sprouting trees were lower than the dead trees, regardless of the number of stems per tree (Fig. 4). Tree height has positive relation to vessel diameter (Martínez-Cabrera et al., 2011), which is the most important wood trait that determine vulnerability to embolism (Sperry & Sullivan, 1992). Plants most efficiently ensures conductivity by forming few, wide and long conduits (Sperry et al., 2008), but for a given tree height many small vessels are more resistant to embolism (Davis et al., 1999). Poplars are associated with high water consumption (Silim et al., 2009), and hence might have trade-off between hydraulic conductivity and resistance to embolism.

## CONCLUSIONS

Interest of growing poplars in the Northern Europe is increasing. Yet, limited number of commercial clones is available, and their growth as well as suitability to climate should be thoroughly tested due to northward transfer. We observed reasonable growth of one-year-old poplar clone OP42. However, extremely high mortality occurred due to early autumn frost (freeze-thaw cycles) at the end of the first growing season, suggesting that frost prone sites are not suitable for establishment of plantations of OP42 clone. The results emphasize necessity to include frost risk assessment, especially for the first year of establishment, in site selection and economical calculations. Our experiment was carried out in a single trial and therefore information from other sites needs to be collected before any further generalizations. Resprouting of damaged trees was rather late and poorly performed, and further assessment of vitality and growth of the sprouts is needed.

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