Jack pine becomes more vulnerable to cavitation with increasing latitudes under doubled [CO2]

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<td>19-Nov-2017</td>
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<tr>
<td>Complete List of Authors</td>
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<td>Is the invited manuscript for consideration in a Special Issue?</td>
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<tr>
<td>Keyword</td>
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Jack pine becomes more vulnerable to cavitation with increasing latitudes under doubled [CO₂]

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Abstract

Trees may migrate northward in response to climate change and become exposed to new photoperiod and soil moisture regimes. This study assessed the impacts of photoperiod and its interaction with soil moisture and carbon dioxide concentration ([CO₂]) on the hydraulic conductivity in jack pine and its vulnerability to xylem embolism. Seedlings were exposed to 400 vs. 950 µmol mol⁻¹ [CO₂], 60–70% vs. 30–40% (of field capacity) soil moisture and photoperiods of seed origin, and 5º and 10º north of seed origin in greenhouses. Cavitation vulnerability curves were measured for determining the xylem pressure at which 50% hydraulic conductivity was lost (ΨPLC50). It was found that elevated [CO₂] significantly increased hydraulic conductivity while low soil moisture decreased it. Under elevated [CO₂], the xylem became progressively more vulnerable to embolism with changes in photoperiod regime from the seed origin to 10º north of the seed origin, as indicated by the progressively less negative ΨPLC50. However, no such a trend was detected under the ambient [CO₂]. The results suggest that the species may become less resistant to drought as the atmospheric [CO₂] increases, hindering the northward migration or seed transfers. Even within its current natural distribution range, trees near its northern boundary of the range may be more vulnerable to embolism as the atmospheric [CO₂] increases even without any change in moisture conditions.

Keywords: Climate change; tree migration; jack pine; hydraulic conductivity; embolism.
1. Introduction

Global warming will increase the frequency and severity of droughts by altering the amount, intensity, frequency and type of precipitation (Christensen et al. 2007). Rainfalls are likely to increase during autumn and spring but decrease in the summer in mid-high latitudes of the Northern Hemisphere, reducing growing-season water supply to plants (IPCC 2007). Global warming will also affect the evapotranspiration and water balance of terrestrial ecosystems (Meinzer 2002; Domec et al. 2009; Way and Oren 2010) via its effects on atmospheric and soil conditions (Allison and Treseder 2008). Inevitably, the water relations in plants will be affected (Way and Oren 2010). Plant response to drought ranges from reductions in growth and photosynthesis to xylem embolism, hydraulic failure and mortality (McDowell et al. 2011). Stomatal regulation represents a short-term response to conserve water and avoid excessive embolism of the xylem (Meinzer and Grantz 1991; Baldocchi 1997; Franks 2004). When the stomatal regulation capacity is exceeded, embolism occurs, leading to shoot dieback or tree mortality (Tyree and Sperry 1989; McDowell et al. 2011). Species differ considerably in their vulnerability to embolism (Tyree and Sperry 1989). Generally, plants growing under moist conditions are more vulnerable to embolism when exposed to drought (Alder et al. 1996; Cochard et al. 1999). The ability of a species to avoid embolism and/or to recover from it can affect its ecological range (Tyree and Sperry 1989).

Atmospheric [CO$_2$] and photoperiod can influence plant water relations. Elevated [CO$_2$] increases water use efficiency by increasing photosynthetic rates and reducing transpiration via its impact on stomatal conductance (Field et al. 1995; Curtis and Wang 1998; Norby et al. 1999; Ainsworth and Long 2005; Keenan et al. 2013) and thus can partially mitigate the negative effect of drought (Keenan et al. 2013). The decrease in stomatal conductance indirectly increases
photosynthesis by reducing the level of internal water stress in the plant (Hungate et al. 2002). Furthermore, elevated atmospheric [CO₂] can enhance water uptake by increasing root growth (Rogers et al. 1994; Curtis and Wang, 1998). However, the response varies with species and other environmental conditions (Gunderson et al. 2002; Long et al. 2004; Marchi et al. 2004; Leakey et al. 2006). Longer photoperiods increase the diameter of xylem tracheid by increasing auxin production in the terminal shoot (Larson 1962). Tracheid with a larger diameter generally results in greater hydraulic conductance. However, tracheids with a larger diameter are also more vulnerable to cavitation (Lambers et al. 2008).

The global climate change, particularly changes in temperature and precipitation, will likely lead to the northward shift of the climate envelopes for many boreal tree species, leading to potential northward migrations (McKenney et al. 2007; McKenney et al. 2011). However, northward migrations (natural or assisted) will expose trees to different photoperiod regimes and other environmental conditions than those they have adapted to. The longer growing season photoperiods and faster rates of season transitions at higher latitudes may make the trees more vulnerable to frost and cavitation damages (Lambers et al. 2008). The interactive effects of photoperiod, moisture and [CO₂] are critical for predicting the potential responses of boreal trees to climate change (Curtis et al. 2000; Nowak et al. 2004) but are poorly understood.

There has been an increasing trend in tree mortality associated with climate change. The mortality is believed to have been caused by either hydraulic failure, or carbohydrate starvation, or insect attacks or deceases, or some combination of them (Adams et al. 2010; Allen et al. 2010; McDowell et al. 2011). Carbohydrate starvation is caused by a combination of reduced photosynthetic carbon fixation due to reductions in stomatal conductance and stimulated respiration under warmer, drier conditions and elevated [CO₂] (Adams et al. 2010). Hydraulic
failure is caused by the cavitation of a substantial proportion of xylem tracheid or vessels. The objective of this study is to understand how [CO₂], photoperiod and soil moisture act interactively in affecting the vulnerability of jack pine seedlings to cavitation. While seedlings are different from large trees in size, structure, anatomy and physiology, the anatomy and functioning of seedling stem xylem are similar to twigs in large trees (Pallardy 2008). Furthermore, manipulating [CO₂], photoperiod and soil moisture simultaneously is an extremely difficult, if not impossible, task to accomplish. Such a study on tree seedlings should provide valuable insight on how branches of a large tree will likely respond to similar conditions. Since elevated [CO₂] and longer photoperiods tend to increase the number and diameter of tracheid in trees and the vulnerability to cavitation is positively correlated to xylem conduit diameter (Salleo and Lo Gullo 1986; Salleo and Lo Gullo 1989; Cochard and Tyree 1990), we hypothesize that elevated [CO₂] and longer photoperiods will increase seedling hydraulic conductivity but make them more vulnerable to embolism.

2. Materials and methods

2.1 Plant materials

One-year old jack pine (Pinus banksiana Lamb.) seedlings were raised from seeds by a commercial tree seedling nursery in Thunder Bay. The seeds were collected from matured jack pine stands in the Kakabeka region of Ontario (48°57' N and 90°44' W). A total of 480 seedlings, relatively uniform in height and root collar diameter (average height/root collar diameter = 13.09/0.21 cm), were transplanted in plastic pots of 15 cm in height and 13 cm in diameter filled with a mixture of premium grade vermiculite and peat moss at a ratio of 1:1 (v/v).
2.2 Experimental design

The experiment was carried out in four greenhouses at Lakehead University’s Thunder Bay campus. The experiment design was split-split plot with \([\text{CO}_2]\) as the whole plot, soil moisture as the sub-plot and photoperiod as the sub-sub plot. There were two levels of \([\text{CO}_2]\) (400 and 950 \(\mu\text{mol mol}^{-1}\)), two levels of soil moisture (60–70% and 30–40% of field water capacity) and three photoperiod regimes (photoperiods at seed origin, 5° north of the seed origin and 10° north of the seed origin). Each of the two \([\text{CO}_2]\) treatments had two replications and was randomly assigned to the four independent greenhouses. The two levels of soil moisture were nested within each level of \([\text{CO}_2]\) and three photoperiod regimes were nested within each level of soil moisture. There were 20 seedlings per treatment combination and a total of 480 seedlings for the experiment.

2.3 Growing conditions

Desired levels of \(\text{CO}_2\) were maintained in the greenhouses using electronic ignition natural gas \(\text{CO}_2\) generators (model GEN-2E, Custom Automated Products Inc, Riverside, CA). Circulation fans were used to ensure even distribution of \(\text{CO}_2\) throughout each of the greenhouses. The \(\text{CO}_2\) in each greenhouse was monitored and controlled automatically with an Argus \(\text{CO}_2\) Control System (Argus, Vancouver, BC, Canada). The moisture content of the growing medium was measured daily with a HH2 moisture meter and a ML2x Theta probe (Delta-T Devices, Cambridge, UK) and the desired moisture levels were maintained by adding water when required. The photoperiod for each treatment was adjusted biweekly to mimic the actual biweekly average photoperiods (April 16 to October 31) at the corresponding locations. The desired photoperiod lengths were achieved by extending the natural day lengths using high-
pressure sodium lamps or shortening the natural day lengths through manual shading of the seedlings. To avoid the possible influence of light from longer photoperiod treatments, seedlings in shorter photoperiods were shaded all around using blackout tarp (silver color on one side and black on the other side) during dark periods. All the seedlings were fertilized biweekly with N:P:K fertilizer (7.14, 0.56 and 2.12 mmol per litre of water, respectively during the rapid growth phase and 1.78, 2.20 and 4.64 mmol per liter of water, respectively during the hardening phase) (Scarratt 1986). In all the greenhouses relative humidity was maintained at 55–60% during the rapid growth phase and reduced to 45–50% during the hardening phase. The day and night air temperatures were set biweekly to mimic the mean biweekly day and night air temperatures at the seed origin and the regime was the same in all the treatments. Relative humidity and air temperature were controlled automatically using an Argus Control System.

2.4 Hydraulic conductivity measurement

Three seedlings were randomly selected from each treatment combination. A branch-free stem segment was excised from each seedling and placed in a water bath at 4°C for 2 hours to reduce resin content. Hydraulic conductivity ($K_H$) was measured following the methods of Sperry et al. (1988). While submerged under water, both ends of the stem segment were re-cut using sharp clippers leaving 2 cm at each end of the pre-marked 15 cm segment. Then two ends of the segment were trimmed again with a sharp razor blade and rapidly attached to solution-filled tubing. Rubber gaskets were used in both ends of the stem segment to ensure leak-proof fitting. At this stage perfusing solution was passed through the stem from a supply tank under gravity. The sap coming out of the stem was collected in a beaker sitting on an electronic balance (Traveler Scales, Ohaus Corporation, Pine Brook, NJ, USA). The balance was attached to a computer and the weight of the beaker was recorded automatically at 1 minute intervals. The
measured flow rate was considered as the background flow rate. After that, the perfusing solution in the supply tank was pressurized to 10 kPa and the flow rate was measured again. Subsequently the background flow rate measurement was repeated. The net flow rate induced by the pressure was determined by subtracting the average background flow (prior to and after the pressurization) from the pressurized flow. Under bark cross sectional area of the larger end, attached to the supply tank by tubing, of each segment was determined and the specific hydraulic conductivity was determined by dividing the net flow rate by the induced pressure gradient.

2.5 Vulnerability to embolism

Vulnerability curves to embolism were measured as the reduction in hydraulic conductivity of the stem with increases in the pressure inside a vulnerability measuring chamber (Cochard et al. 1992). Xylem embolism was induced by successively increasing positive air pressure on stem segments inside a double-ended cavitation chamber (1505D-EXP Pressure Chamber, PMS Instrument Company, Albany, OR, USA). After measuring the hydraulic conductivity (described previously) the sample was flushed at a pressure of 175 kPa for half an hour to remove any existing emboli and the maximum hydraulic conductivity ($K_{max}$) was determined. After that, pressure in the cavitation chamber was progressively increased to 0.3, 0.5, 0.8, 1.0, 1.3, 1.5, 1.8, 2.0, 2.3 and 2.5 MPa and hydraulic conductivity ($K_h$) was measured 10 minutes after the desired pressure value was reached at each pressure step. Percentage loss in hydraulic conductivity (PLC) following each pressurization was calculated as $PLC = 100\left(\frac{K_{max} - K_h}{K_{max}}\right)$. The vulnerability curves for each treatment were constructed using Microsoft Excel (2010). The air pressures at 50% loss of the maximum hydraulic conductivity ($\Psi_{PLC50}$) were calculated from the best-fit curves generated by using Weibull functions.
2.6 Statistical analysis

Analysis of variance (ANOVA) was performed for the hydraulic conductivity and $\Psi_{PLC50}$ using R programming environment 3.1.3 (R Core Team 2015). Before the ANOVA test, the normality of distribution and homogeneity of variance were checked using Shapiro-Wilk and Bartlett tests, respectively. When ANOVA showed significant interactive effects or photoperiod effects (which had 3 treatment levels), Tukey’s HSD Post-hoc comparisons were carried out. In the analyses, [CO$_2$], soil moisture and photoperiod were treated as fixed effects and the threshold probability was 0.05.

3. Results

3.1 Hydraulic conductivity

Hydraulic conductivity in jack pine seedlings was significantly affected by carbon dioxide concentrations ([CO$_2$]) and the soil moisture (Table 1). Elevated [CO$_2$] increased the hydraulic conductivity by 34.5% (Figure 1A) while low soil moisture reduced it by 14.5% (Figure 1B). However, photoperiod or interactions between treatments had no significant effect (Table 1).

3.2 Vulnerability to embolism

The elevated [CO$_2$] at photoperiod 10º north of the seed origin resulted in significantly lower xylem pressure or less negative xylem water potential at which 50% of the hydraulic conductivity was lost ($\Psi_{PLC50} = -0.89$ MPa) compared to the same photoperiod regime under ambient [CO$_2$] ($\Psi_{PLC50} = -1.25$ MPa) and under the elevated [CO$_2$] at the photoperiod regime of the seed origin ($\Psi_{PLC50} = -1.27$ MPa) (Tables 1, Table A1, Figures 1C and 2E). However, other treatment combinations did not significantly differ from each other (Table 1, Figure 2).
4. Discussion

Elevated [CO$_2$] significantly increased hydraulic conductivity in jack pine seedlings while low soil moisture decreased it. Xylem hydraulic conductivity of trees is positively correlated with the 4$^{th}$ power of xylem vessel diameter, which is in turn positively related to growth rates (Lambers et al. 2008). Indeed the growth rates of our jack pine seedlings were greater under the elevated [CO$_2$] and high soil moisture conditions (unpublished). Other researchers have found that accelerated growth rates increase the number of tracheid and the tracheid lumen area in jack pine (Krause et al. 2011). Neufeld et al. (1992), Maherali and DeLucia (2000), Thomas et al. (2004), Cai and Tyree (2010), and Cai et al. (2014) indicate that larger-diameter tracheid/vessels are primarily responsible for the greater stem hydraulic conductivity in fast growing trees.

There is a tradeoff between stem hydraulic conductivity and stem vulnerability to embolism because larger conduits are more conductive but also more vulnerable to embolism (Cai and Tyree 2010). Indeed, the elevated [CO$_2$] in this study led to greater hydraulic conductivity but also a greater loss of hydraulic conductivity when embolism was induced experimentally. Overall the seedlings grown under elevated [CO$_2$] and the longer photoperiod corresponding to 10º north of the seed origin suffered the greatest loss of hydraulic conductivity and the loss occurred at a much less negative water potential than in other treatment combinations. Past studies find that longer photoperiods increase the number and diameter of xylem tracheid by increasing auxin production in the terminal shoot (Larson 1962), presumably leading to a greater hydraulic conductivity. The characteristics of pits on conduit walls and pit membranes have great influence on the vulnerability of xylem to embolism: wider conduits tend to have larger pit member area and larger-sized pores in their pit membrane, and are thus more prone to air-seeding and consequent cavitation (Sperry and Tyree 1990; LoGullo and Salleo
1991; Hargrave et al. 1994; LoGullo et al. 1995; Wheeler et al. 2005). However, Neufeld et al. (1992) did not observe any consistent relationship between the hydraulic conductivity or vessel diameter and xylem vulnerability to embolism.

The most interesting finding of this study is that the xylem of jack pine seedlings became more vulnerable to cavitation with changes in photoperiod regime as the latitude increased from the seed origin under the projected future atmospheric [CO₂] while it was relatively insensitive to changes in photoperiod regimes under the ambient [CO₂]. For example, the xylem pressure at which 50% of the xylem (ψₚ₅₀) was cavitated under elevated [CO₂] was -1.27 MPa under the photoperiod regime of the seed origin, but became -0.89 MPa under the photoperiod regime 10° north of the seed origin. These results suggest that the species may become less resistant to drought as the atmospheric [CO₂] increases, hindering the northward migration or seed transfers. Even with its current natural distribution range, trees near its northern boundary of the range may become more vulnerable to embolism as the atmospheric [CO₂] increases even without any change in moisture conditions. Accelerated growth under elevated CO₂ and longer growing season photoperiod at higher latitudes may provide the explanation for the increased vulnerability to cavitation.

The xylem tension at 50% loss of hydraulic conductivity (ψₚ₅₀) measured in this study was less negative than the values of the same and similar species reported by other researchers. Visual estimates from the figures of Schoonmaker et al. (2010) suggest that ψₚ₅₀ for young trees grown in shade was about -3.1 MPa for jack pine (vs. our reading of -1.3 MPa), -2.1 MPa for *Picea mariana*, -4.0 MPa for *Picea glauca* and -3.2 MPa for *Pinus contorta* while the corresponding values for trees grown under open environment were much more negative (down to -5.6 MPa). There are several explanations to the differences. Firstly the difference could be
attributable to age differences: Schoonmaker et al. (2010) measured 7-year old saplings while our measurements were conducted on 1-year old seedlings. Seedlings are generally more vulnerable to drought stress. The second factor could be the season when the measurements were taken. The measurements of Schoonmake et al. (2010) were made in the fall (late September to mid-October) while our measurements were taken on greenhouse grown seedlings at a time equivalent to mid-growing season. Drought tolerance increases towards the end of the growing season as the cold hardening progresses. The different growing conditions between the two studies could have contributed to the difference in $\psi_{\text{plc50}}$. The trees in Schoonmaker et al. (2010) were grown in the field for six years. The environmental conditions in the field are much more dynamic and stressful than the conditions in the greenhouse. The differences in $\psi_{\text{plc50}}$ between trees of the same species grown under different light conditions in Schoonmaker et al. (2010) suggest that those boreal tree species are capable of acclimating to their growing environmental conditions. Furthermore, different techniques were used in measuring the vulnerability curves between the two studies. We used a cavitation chamber which induces cavitation by pushing air into the xylem water column while Schoonmaker et al. (2010) used the centrifuging technique which pulls air into the xylem. Although the air-seeding hypothesis applies to both techniques, there could be differences in the response. However, our measurements are comparable to $\psi_{\text{plc50}}$ values reported for large trees of less drought resistant species, such as Salix alba L. and Populus deltoids Bartr. X Marsch (Cochard et al. 1992). The differences in $\psi_{\text{plc50}}$ values should not affect the response patterns that we discovered in this study.
Acknowledgements

First of all, we want to thank the Associate Editor and an anonymous reviewer for their very constructive comments on the previous version of the manuscript. Because of their comments, the presentation quality of this manuscript has been greatly enhanced. We also acknowledge Ms. Joan Lee, former Greenhouse Manager of Lakehead University for her logistic support and all out cooperation in the greenhouse operations. The study was supported by NSERC Discovery Grant to QL Dang (Project no. 203198-2013-RGPIN).

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camaldulensis due to temperature—the physiological link between water viscosity and


Table 1: P values for the effects of CO\textsubscript{2} concentration (C), soil moisture (M), photoperiod (P) and their interactions on the hydraulic conductivity (HC) and xylem pressure at 50% loss of conductivity (Ψ\textsubscript{PLC50}) in jack pine (Pinus banksiana Lamb.) seedlings. Seedlings were grown under two levels of [CO\textsubscript{2}] (400 and 950 µmol mol\textsuperscript{-1}), two levels of soil moisture (60–70% and 30–40% of field water capacity) and three levels of photoperiod (seed origin, 5º and 10º north of seed origin).

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Figure captions

**Fig. 1:** Specific hydraulic conductance of stem (HC) and xylem pressure at 50% loss of hydraulic conductivity ($\Psi_{PLC50}$) in jack pine (*Pinus banksiana* Lamb.) seedlings grown under two levels of [CO$_2$] ($C_a$-ambient, $C_e$-elevated), two levels of soil moisture ($M_h$-high, $M_l$-low) and three photoperiod regimes ($P_s$-at seed origin, $P_{nm1}$-5° north and $P_{nm2}$-10° north of seed origin).

*Note:* Bars in Figure C: Black=$P_s$, Light grey=$P_{nm1}$ and Dark grey=$P_{nm2}$.

**Fig. 2:** Vulnerability curves showing percentage loss of hydraulic conductivity (PLC) as a function of xylem pressure at different levels of [CO$_2$] (ambient ($C_a$) and elevated ($C_e$)), soil moisture (high ($M_h$) and low ($M_l$)), photoperiod regime (at seed origin ($P_s$), 5° north ($P_{nm1}$) and 10° north ($P_{nm2}$) of seed origin) and their interactions. *Note:* Lines in Figures A: Blue=$C_a$, Green=$C_e$; B: Blue=$M_h$, Pink=$M_l$; C: Cyan=$P_s$, Blue=$P_{nm1}$, Pink=$P_{nm2}$; D: Cyan=$C_a$,$M_h$, Blue=$C_a$,$M_l$, Pink=$C_a$,$M_l$, Dark green=$C_a$,$M_h$; E: Red=$C_a$,$P_s$, Green=$C_a$,$P_{nm1}$, Cyan=$C_a$,$P_{nm2}$, Blue=$C_a$,$P_{nm1}$, Pink=$C_a$,$P_{nm2}$, Dark red=$C_a$,$P_{nm2}$; F: Red=$M_h$,$P_s$, Green=$M_h$,$P_{nm1}$, Cyan=$M_h$,$P_{nm2}$, Blue=$M_l$,$P_s$, Pink=$M_l$,$P_{nm1}$, Dark red=$M_l$,$P_{nm2}$; G: Black=$C_a$,$M_h$,$P_s$, Red=$C_a$,$M_h$,$P_{nm1}$, Green=$C_a$,$M_h$,$P_{nm2}$, Yellow=$C_a$,$M_l$,$P_s$, Blue=$C_a$,$M_l$,$P_{nm1}$, Pink=$C_a$,$M_l$,$P_{nm2}$, Cyan=$C_a$,$M_l$,$P_{nm1}$, Grey=$C_a$,$M_l$,$P_{nm2}$, Dark yellow=$C_a$,$M_l$,$P_{nm2}$, Dark green=$C_a$,$M_l$,$P_{nm1}$, Dark blue=$C_a$,$M_l$,$P_{nm1}$, Dark pink=$C_a$,$M_l$,$P_{nm2}$.
Specific hydraulic conductance of stem (HC) and xylem pressure at 50% loss of hydraulic conductivity (ΨPLC50) in jack pine (Pinus banksiana Lamb.) seedlings grown under two levels of [CO2] (Ca-ambient, Ce-elevated), two levels of soil moisture (Mh-high, Ml-low) and three photoperiod regimes (Ps-at seed origin, Pnm1-5° north and Pnm2-10° north of seed origin). Note: Bars in Figure C: Black=Ps, Light grey=Pnm1 and Dark grey=Pnm2.
Vulnerability curves showing percentage loss of hydraulic conductivity (PLC) as a function of xylem pressure at different levels of [CO2] (Ca-ambient, Ce-elevated), soil moisture (Mh-high, Ml-low), photoperiod regime (Ps-seed origin, Pnm1-5° north and Pnm2-10° north of seed origin) and their interactions.

Figure 2: Vulnerability curves showing percentage loss of hydraulic conductivity (PLC) as a function of xylem pressure at different levels of [CO2] (Ca-ambient, Ce-elevated), soil moisture (Mh-high, Ml-low), photoperiod regime (Ps-seed origin, Pnm1-5° north and Pnm2-10° north of seed origin) and their interactions.

Vulnerability curves showing percentage loss of hydraulic conductivity (PLC) as a function of xylem pressure at different levels of [CO2] (ambient (Ca) and elevated (Ce)), soil moisture (high (Mh) and low (Ml)), photoperiod regime (at seed origin (Ps), 5° north (Pnm1) and 10° north (Pnm2) of seed origin) and their interactions. Note: Lines in Figures A: Blue=Ca, Green=Ce; B: Blue=Mh, Pink=Ml; C: Cyan=Ps, Blue=Pnm1, Pink=Pnm2; D: Cyan=CaxMh, Blue=CaxMl, Pink=CexMh, Dark green=CexMl; E: Red=CaxPs, Green=CaxPnm1, Cyan=CaxPnm2, Blue=CaxPs, Pink=CexPnm1, Dark red=CexPnm2; F: Red=MhxPs, Green=MhxPnm1, Cyan=MhxPnm2, Blue=MlxPs, Pink=MlxPnm1, Dark red=MlxPnm2; G: Black=CaxMhxPs, Red=CaxMhxPnm1, Green=CaxMhxPnm2, Yellow=CaxMlxPs, Blue=CaxMlxPnm1, Pink=CaxMlxPnm2, Cyan=CexMhxPs, Grey=CexMhxPnm1, Dark yellow=CexMhxPnm2, Dark green=CexMlxPs, Dark blue=CexMlxPnm1, Dark pink=CexMlxPnm2.
### App Table 1: Xylem pressures at 50% loss of hydraulic conductivity (Ψ_{PLC50}) in jack pine (Pinus banksiana Lamb.) seedlings at different levels of [CO₂], soil moisture and photoperiod and their interactions. The R²-values are from the best-fit vulnerability curves constructed by using the Weibull functions.

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<td>0.8481</td>
<td>M_l x P_{nm1}</td>
<td>1.1624</td>
<td>0.9048</td>
</tr>
<tr>
<td>P_{nm1}</td>
<td>1.2029</td>
<td>0.8962</td>
<td>M_l x P_{nm2}</td>
<td>1.1042</td>
<td>0.8245</td>
</tr>
<tr>
<td>P_{nm2}</td>
<td>1.0754</td>
<td>0.8354</td>
<td>C_a x M_h x P_s</td>
<td>1.1645</td>
<td>0.9035</td>
</tr>
<tr>
<td>C_a x M_h</td>
<td>1.2049</td>
<td>0.8925</td>
<td>C_a x M_h x P_{nm1}</td>
<td>1.2251</td>
<td>0.8982</td>
</tr>
<tr>
<td>C_a x M_l</td>
<td>1.2308</td>
<td>0.8821</td>
<td>C_a x M_h x P_{nm2}</td>
<td>1.2292</td>
<td>0.885</td>
</tr>
<tr>
<td>C_e x M_h</td>
<td>1.0989</td>
<td>0.8437</td>
<td>C_a x M_l x P_s</td>
<td>1.1866</td>
<td>0.8668</td>
</tr>
<tr>
<td>C_e x M_l</td>
<td>1.1350</td>
<td>0.809</td>
<td>C_a x M_l x P_{nm1}</td>
<td>1.1762</td>
<td>0.8981</td>
</tr>
<tr>
<td>C_a x P_s</td>
<td>1.1753</td>
<td>0.8844</td>
<td>C_a x M_l x P_{nm2}</td>
<td>1.2736</td>
<td>0.8908</td>
</tr>
<tr>
<td>C_a x P_{nm1}</td>
<td>1.2016</td>
<td>0.8972</td>
<td>C_e x M_h x P_s</td>
<td>1.1752</td>
<td>0.8538</td>
</tr>
<tr>
<td>C_a x P_{nm2}</td>
<td>1.2537</td>
<td>0.8853</td>
<td>C_e x M_h x P_{nm1}</td>
<td>1.2666</td>
<td>0.8969</td>
</tr>
<tr>
<td>C_e x P_s</td>
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<td>0.8209</td>
<td>C_e x M_h x P_{nm2}</td>
<td>0.8932</td>
<td>0.8866</td>
</tr>
<tr>
<td>C_e x P_{nm1}</td>
<td>1.2044</td>
<td>0.9005</td>
<td>C_e x M_l x P_s</td>
<td>1.3852</td>
<td>0.8207</td>
</tr>
<tr>
<td>C_e x P_{nm2}</td>
<td>0.8897</td>
<td>0.8519</td>
<td>C_e x M_l x P_{nm1}</td>
<td>1.1471</td>
<td>0.9143</td>
</tr>
</tbody>
</table>

Note: C_a and C_e are ambient and elevated [CO₂], respectively; M_h and M_l are soil moistures at 60-70% and 30-40% of field water capacity, respectively; P_s, P_{nm1} and P_{nm2} are photoperiods at seed origin, 5° and 10° north of the seed origin, respectively.