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# Evaluation of *Alnus subcordata* for urban environments through assessment of drought and flooding tolerance

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**Abstract:** The urban environment is stressful and trees experience multiple stresses, including drought, flooding, and extreme heat, all of which are likely to increase under future climate warming and increasing urbanisation. In the selection of tree species to maximise ecosystem services, tolerance to site characteristics such as flooding and severe drought is of critical importance. This study evaluated the suitability of a rare species, *Alnus subcordata* C.A. Mey (Caucasian alder) from the Hyrcanian forests of southern Azerbaijan, for its functionality as an urban tree.

A total of 48 pot-grown, two-year-old saplings of *A. subcordata* were tested in a greenhouse experiment using a complete randomised block design. Each block contained four replicates of three treatments (waterlogging, drought, control), with 16 plants per treatment. Height differences between treatments were measured, and water status was estimated by determination of midday leaf water potential ($\Psi_L$) and stomatal conductance ($g_s$). To estimate drought tolerance reaction in the treatments, leaf water potential at turgor loss ($\Psi_{P0}$) was used together with broken-stick modelling of water status over time.

There was a significant difference in tree height between the different treatments. In the drought treatment, *A. subcordata* plants showed no height increase, while plants in both the waterlogged and control treatments increased in height during the nine-week experiment. Over 63 days of flooding, plant water status was slightly more negative in the waterlogging treatment, but did not deviate essentially from the control. In the drought treatment, plant water status rapidly deviated from the control. There was a significant difference in $\Psi_{P0}$ between treatments, with drought-treated plants showing the lowest value (−2.31 MPa).

This study demonstrated that *A. subcordata* has limited tolerance to drought and seems to rely more on water loss-avoiding strategies. However, the species may be usable at periodically waterlogged sites, due to its high tolerance to flooding. It could therefore be recommended for wet urban environments and stormwater management facilities, for which reliable guidance on suitable trees is currently lacking.

**Keywords:** climate change, ecosystem services, urban forestry, stress tolerance, adventitious roots

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## Introduction

With growing awareness of the critical importance of ecosystem services and green infrastructure, trees are gaining recognition in research and policies as a suitable means to achieve resilience and sustainability in cities worldwide (Morgenroth et al., 2016; Pauleit et al., 2017). Recent research has demonstrated the value of urban trees in e.g. mitigating air pollution, managing stormwater, and mitigating heat (Connop et al., 2016; Livesley et al., 2016; Fitzky et al., 2019). However, most of these ecosystem services are strongly connected with tree size and health (Xiao & McPherson, 2002; Gratani & Varone, 2006; Gómez-Muñoz et al., 2010; Vos et al., 2013). Thus tree species tolerance to local climate and site conditions forms the basis for sustainable urban forestry. The urban environment is stressful and trees often experience multiple stresses, including drought, flooding, and extreme heat, all of which are likely to increase under future climate scenarios and increased urbanisation (Allen et al., 2010; Teskey et al., 2015). Thus, in the selection of tree species to maximise ecosystem services provision, tolerance to site conditions such as flooding and severe drought events is of critical importance. During recent years, there has also been growing interest in using trees in open stormwater solutions or purposely directing stormwater runoff to urban tree pits, to improve the tree growth while simultaneously managing stormwater (Szota et al., 2019). Reuse of water in tree pits can improve their growth and water status and promote ecosystem services provision, but may also cause temporary flooding that can have negative effects on tree performance. Thus, trees in modern installations need to tolerate both temporary inundation and prolonged drought spells.

Future climate change will increase the importance of having systems with high species diversity, to enhance the resilience of urban forests to both abiotic and biotic challenges (Hooper et al., 2005; Alvey, 2006). The strategic value of diversifying urban forest is acknowledged, but progress is compromised by a lack of familiarity among urban tree planners and nurseries regarding utilisation and production of species that are not currently well-known (Sjöman & Nielsen, 2010). Therefore, research-based guidance is needed on the use of species that are not widely observed in urban landscapes. One way to provide the necessary guidance and instil confidence among tree planners is to provide quantitative data on the reaction of candidate tree species to specific stressors arising in urban environments today and in the future. However, much of the existing research on stress reactions in trees focuses on common tree species already used in great numbers in urban environments (Sjöman et al., 2018). For less common species and ecotypes, research efforts have been much more limited. Evaluating and measuring the reactions of these species and ecotypes to drought and flooding in controlled environments, such as greenhouse facilities, is an essential step in screening for their suitability for future urban environments.

This study evaluated the suitability of a rare and promising tree species, *Alnus subcordata* C.A. Mey (Caucasian alder) from the Hyrcanian forests of southern Azerbaijan, for its functionality as an urban tree at sites that experience periodic flooding. This species is not currently used as an urban tree and is very rare even in tree collections at botanical gardens and university arboretums, so it seldom features in the horticultural and urban forestry literature. In dendrological publications, the species is mainly described by information on its origin and taxonomic explanations, and not site preferences in cultivation (Bean, 1980; Krüssmann, 1986), while the horticultural literature does not include the species due to its rarity. However, on reviewing literature about the ecological habitat in the Hyrcanian forests of the Caspian mountains, it is clear that *A. subcordata* can tolerate flooded conditions, due to its widespread occurrence in wetlands and river ecosystems (Sabeti, 2006). One study also mentions that *A. subcordata* is a fast-growing tree in the wild and is therefore a popular timber tree, and is also useful in areas where soil erosion is a problem along streams and rivers (Akbarian et al., 2011). *Alnus subcordata* can also be important for reforestation with *Fagus orientalis*, since it creates adequate growing conditions for the beech saplings (Taleshi et al., 2009). A controlled evaluation of the capacity of *A. subcordata* to cope with flooding and drought, and its reaction to these stressors, is needed in order to evaluate its future potential for use in urban environments.

## Material and methods

### Plant material

*Alnus subcordata* is native to the Hyrcanian forests of the Caspian Mountains in southeast Azerbaijan and Northern Iran, and is found from sea level to 2000 m a.s.l. It is a fast-growing tree in the wild and is mainly found in river valleys, but can also thrive in other types of growing conditions, especially at higher altitudes, where it is adapted to a large variety of ecological conditions (Akbarian et al., 2011; Safarov & Farzaliyev, 2019). The plant material evaluated in this study consisted of seeds collected at 690 m a.s.l. in the Talysh Mountains in southern Azerbaijan, during a joint expedition in 2017 by the Plant Collecting Collaborative (PCC) and the Institute of Botany in Baku. Collection was carried out at high altitudes and the northern boundary of the species, in order to obtain winter-hardy plant material.

The seeds were germinated at Gothenburg Botanical Garden, Sweden, and transferred to 2-L pots containing a mixture of 1/3 peat, 1/3 green waste compost, and 1/3 sand. In total, 48 two-year-old plants in pots were used in a controlled experiment in the greenhouse at the Swedish University of Agricultural Science in Alnarp between July and October 2019. All plants were well-rooted in the potting substrate and had no abiotic or biotic injuries at the start of the experiment.

## Drought and flooding evaluation

The experiment was laid out in a complete randomised block design, with the blocks containing four replicates subjected to three treatments: waterlogging, drying, and control (16 plants per treatment). All plants in the experiment were kept well irrigated in the greenhouse from early spring until the treatments started in June. On the start date, all plants were watered to field capacity and their initial height (mean = 1.488 m, SD = 0.185) and pot total weight were measured. The seedlings in the control group were kept well irrigated in well-drained pots throughout the experiment. The waterlogged seedlings were submerged in water-filled boxes for 63 days of the experimental period. To ensure that the water always reached the surface of the topsoil, the boxes were refilled periodically to compensate for any evaporative water loss. The plants subjected to the drought treatment received no irrigation throughout the 9-week experiment. The pots in the drought treatment were weighed during the experiment. At the end of the experiment, plant height, root collar diameter, and the presence of adventitious roots were recorded. Stem diameter growth is often increased in flood-tolerant species subjected to waterlogging, since they may produce intercellular spaces and lower density cells to achieve more effective oxygen transport (Glenz et al., 2006).

Water status was estimated daily during the first week of the experiment and then every second day for the following two weeks, by which time the drought-stressed seedlings were completely wilted. Thereafter, measurement continued only for the control and waterlogged groups, once a week until the end of the experiment. Plant water status was estimated by determination of midday leaf water potential ($\Psi_L$) and stomatal conductance ($g_s$). For $\Psi_L$, one light-exposed mature leaf from each seedling was collected and immediately installed in a pressure chamber (Model 1000, PMS Instrument Company, OR, USA). In parallel, $g_s$ was determined on one light-exposed leaf using a porometer (METER Group AG, Münich, Germany). Measurement of $\Psi_L$ and $g_s$ followed the treatment block order on all occasions, to account for any differences arising from the time of measurement.

## Leaf water potential at turgor loss

Traits that confer tolerance to water deficiency are of inherent interest to growers selecting trees for urban use, as they allow the tree to maintain physiological processes for a longer period during the drying cycle. Leaf water potential at turgor loss ($\Psi_{P0}$) provides a robust measure of plant drought tolerance (Sack et al., 2003; Lenz et al., 2006). Species with low (more negative) $\Psi_{P0}$ tend to maintain leaf gas exchange, hydraulic conductance, and growth at lower soil water potential ($\Psi_{soil}$) (Mitchell et al., 2008; Blackman et al., 2010). $\Psi_{P0}$ is also related to leaf and stem conductivity, hydraulic traits reflecting drought impacts on the water supply for transpiration and photosynthesis (Bartlett et al., 2016). Therefore, $\Psi_{P0}$ indicates the capacity of a species to grow in dry environments such as paved urban sites and to continue delivering ecosystem services by remaining biologically active under harsh climate conditions (Sjöman et al., 2015). Comparison of $\Psi_{P0}$ values is also useful for detecting differences in drought tolerance between treatments and between different ecotypes within a species (Hannus et al., 2020; Hirons et al., 2020).

On the sampling occasion, one sun-exposed twig with no symptoms of abiotic or biotic damage was collected from 14 individual trees per treatment (waterlogged, drought, control) in the early evening on day 10 of the experiment, when transpiration was relatively low. After overnight rehydration, one disc per leaf was taken from fully expanded leaves using an 8 mm core borer. The sample was taken from the mid-lamina region between the mid-rib and leaf margin, avoiding sampling lamina regions with first- and second-order veins. All discs were tightly wrapped in foil to limit condensation or frost after freezing. The foil-wrapped leaf discs were then submerged in liquid nitrogen for 2 min to fracture the cell membranes and walls and punctured 10-15 times with sharp-tipped forceps to facilitate evaporation through the cuticle and decrease equilibration time (Kikuta & Richter, 1992). The leaf discs were then placed in a vapor pressure osmometer (Vapro 5600, Westcor, Logan, UT, USA) using a standard 10 $\mu$L chamber. Solute concentration ($c_s$, mmol kg$^{-1}$) was determined after 10 min equilibration time (initial value) and then at ~2 min intervals until the concentration was <5 mmol kg$^{-1}$. Solute concentration was converted to osmotic potential ($\Psi\pi$) using the Van't Hoff's equation:

$$\Psi_{P0} = -RTc_s \qquad \text{(Eq. 1)}$$

where R is a gas constant, T is the temperature in Kelvin and $c_s$ is the solute concentration.

Bartlett et al. (2012b) developed an equation allowing prediction of $\Psi_{P0}$ from the osmotic potential at full turgor ($\Psi_{\pi 100}$), based on a global dataset that included data from tropical biomes. For trees in the temperate biome, Sjöman et al. (2015a) used a subset (woody temperate, Mediterranean/temperate-dry and temperate conifer species) of the supplementary data published by Bartlett et al. (2012a) to generate a new equation for deriving $\Psi_{P0}$ from $\Psi_{\pi 100}$ in temperate tree species:

$$\Psi_{P0} = -0.2554 + 1.1243 \times \Psi_{\pi 100} \quad \text{(Eq. 2)}$$

That equation was used in the present study, as it provided a higher coefficient of determination ($R^2$; 0.91 vs. 0.86) and therefore a more reliable means of predicting $\Psi_{P0}$.

## Statistical analyses

All statistical testing was performed in R, with a significance level $p<.05$. To test for differences in height growth between treatments, a generalised least squares (GLS) model was used to account for the difference in variance between treatments, using the gls function in the nmle package in R (Pinheiro et al., 2019) with a varIndent variance structure and REML estimation. The response used was the difference in tree height before and after treatment, while the explanatory variable was treatment (fixed) with three levels. Model validation was based on plotting of residuals according to the approach in Zuur et al. (2009). The emmeans package (Lenth, 2019) was used to calculate least-squares means based on the Satterthwaite d.f. method and pairwise comparisons with Bonferroni correction for multiple testing. The same approach was used to test for differences in slenderness index, i.e. the ratio of height to root collar diameter, after treatment. Testing for differences in $\Psi_{P0}$ was performed according to the same GLS approach. Since $\Psi_L$ and $g_s$ at midday are relative measures dependent on e.g. solar radiation, temperature, and humidity, midday $\Psi_L$ and $g_s$ values were plotted as time-series including their 95% confidence intervals, together with the average temperature derived from the porometer readings in the greenhouse. For individual plants that had wilted, zero stomatal conductance and the maximum pressure that could be achieved in the pressure bomb were imputed and the trend was plotted until the confidence intervals covered these imputed minimum values. Based on this, it was evident that the behaviour of plants in the waterlogged and drought treatment differed markedly. Therefore, to evaluate the full trend of treatments, the effect of flooding compared with the control for the whole experimental period was modeled separately as a repeated measurement model. To evaluate the effects of the drought treatment, and especially biologically important change points segmented modelling (broken stick models was used to define breakpoints in the linear relationship between the response variable and the number of days of treatment (Muggeo, 2008).

To evaluate when the plant had used up all available water in the drought treatment, a mixed broken stick model (Muggeo et al., 2014) of the change in total weight was created, accounting for any individual differences, i.e. the individual plant was included as a random effect. All mixed broken stick modelling was done using the segmented package (Muggeo, 2016) and additional scripts presented by Muggeo (2016) and a diagonal covariance structure. A mixed broken stick model was also used to explore the change point in midday $\Psi_L$ and $g_s$ in the drought treatment, i.e. an approximation was made of the number of days before loss of biological activity and wilting of tree leaves. Based on this, the breakpoint within this period before wilting was modelled concerning the difference between drought and control in each of the four blocks, to account for any differences related to the time of measurement. Therefore, the block was used as a random factor instead of the individual plant for this analysis.

To model the effect of flooding on midday $\Psi_L$ and $g_s$ as responses, the gls function with different variance-covariance structures was used to incorporate the repeated structure of the data. The best-fitting co-variance structure was based on Akaike Information Criterion (AIC) together with log-likelihood test and inspection of residual plots. Explanatory variable included was the flooding treatment and the number of days since it started and their interaction. Block was also included in the model.

# Results

There was a significant difference ($F_{2.45}=254.6$; $p<0.0001$) in tree height increase between the different treatments. The drought-treated plants showed no height change, while both waterlogged and control plants showed an increase in height during the treatments. However, the height increase was greatest in the control and all treatments were significantly different from each other (Table 1).

There was also a significant difference in slenderness index ($F_{2,.45}=46.1$; $p<0.0001$), where the waterlogged species had the lowest value, followed by the control and drought treatments (Table 2). All treatments were significantly different from each other. All waterlogged seedlings developed adventitious roots, while those in the control and drought treatments failed to produce adventitious roots (Fig. 1).

Table 1. Least square means (LSM), standard error (SE), degrees of freedom (df), and 95% confidence interval of tree height growth (m) in the drought, waterlogged, and control treatments. Treatments with different letters (a,b,c) are significantly different

| Treatment | LSM (m) | SE | df | 95% Conf. int. | | |
|---|---|---|---|---|---|---|
| Drought | −0.009 | 0.005 | 15 | −0.020 | 0.003 | a |
| Waterlogged | 0.226 | 0.030 | 15 | 0.162 | 0.289 | b |
| Control | 0.465 | 0.021 | 15 | 0.419 | 0.511 | c |

Table 2. Least square means (LSM), standard error (SE), degrees of freedom (df), and 95% confidence interval for slenderness index in the drought, waterlogged, and control treatments. Treatments with different letters (a,b,c) are significantly different

| Treatment | LSM | SE | df | 95 % Conf. Int. | | |
|---|---|---|---|---|---|---|
| Drought | 18.4 | 0.768 | 15.2 | 16.3 | 20.4 | c |
| Waterlogged | 10.9 | 0.357 | 15.0 | 9.9 | 11.8 | a |
| Control | 14.0 | 0.726 | 15.2 | 12.1 | 16.0 | b |

Table 3. Least square means (LSM), standard error (SE), degrees of freedom (df), and 95% confidence interval of leaf water potential at turgor loss ΨP0(MPa) in the drought, waterlogged and control treatments. Treatments with different letters (a,b,c) are significantly different

| Treatment | LSM (MPa) | SE | df | 95 % Conf. Int. | | |
|---|---|---|---|---|---|---|
| Drought | −2.23 | 0.026 | 13 | −2.29 | −2.18 | a |
| Waterlogged | −1.93 | 0.021 | 13 | −1.98 | −1.89 | b |
| Control | −1.83 | 0.022 | 13 | −1.88 | −1.78 | c |

There was a significant difference in $\Psi_{P0}$ between treatments ($F_{2,.39}$ = 72.8; p<0.0001), with the control having the lowest $\Psi_{P0}$ values, followed by the waterlogged and drought treatments (Table 3). All treatments were significantly different from each other.

The $\Psi_L$ value in the waterlogging treatment was similar to that in the control during the first week and then dropped to slightly more negative values, leading to a small but overall significant ($F_{1,569}$=13.6; p = 0.0224) mean difference of −0.0739 (SE = 0.0329) between these treatments

Fig. 1. (Left) Waterlogged *Alnus subcordata* plant with a profusion of adventitious roots at the base. The picture was taken after 63 days of waterlogging; (right) control plants

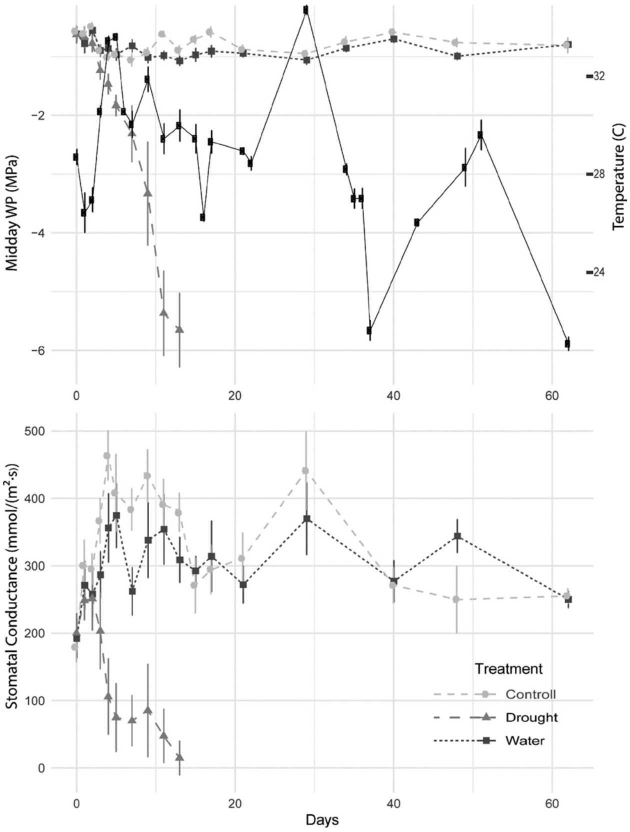


Fig. 2. Mean values (with 95% confidence intervals) of (upper panel) midday leaf water potential ($\Psi_L$) and (lower panel) stomatal conductance ($g_s$). The temperature on non-measurement days, when available, are included to provide a better trend line over time and indicated in the upper panel with the black solid line

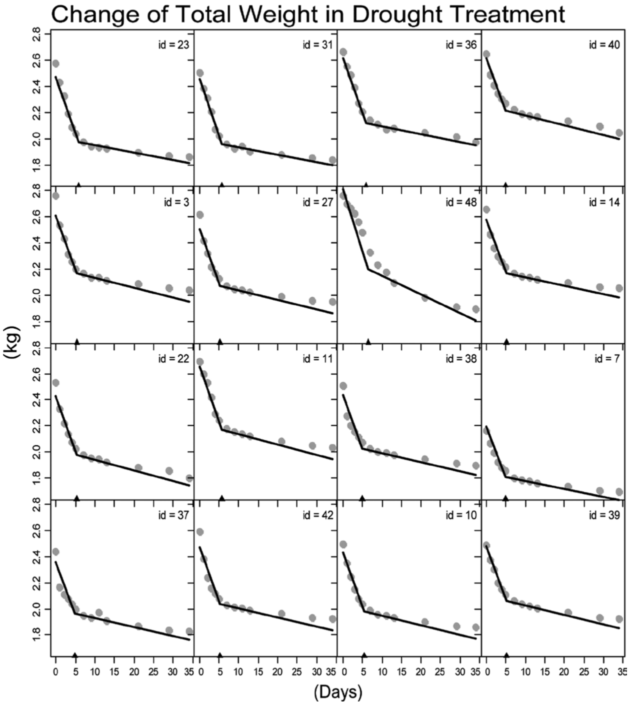


Fig. 3. Panels from random broken stick modelling of the relationship between pot (plus tree) weight and days of drought treatment. Each panel illustrates an individual seedling (n=16) in the drought treatment and the breakpoint at which it showed a marked shift in weight loss

(Fig. 2). Stomatal conductance varied more over time and showed a significant interaction ($F_{1,569} = 22.7$; $p<0.0001$) with the flooding treatment, which could be related to the variations in average temperature (Fig. 2). On average over the experimental period, flooding was estimated to give a significant ($F_{1,569} = 57.5$; $p<0.0001$) decrease in stomatal conductance of 64.96 (SE = 7.59). Changes in the drought treatment followed a more general trend not related to overall temperature, and after only a few days a clear difference between the control and the drought treatment was evident (Fig. 2).

Between day 5 and day 6 (95% CI: 5.1 to 5.8 days) of the drought treatment, pot weight (and thus water availability) changed markedly in that treatment, indicating that the available water had been consumed by the plants at this point (Fig. 3).

From day 9 to day 13 (95% CI: 9.6 to 12.7 days), stomatal conductance changed markedly in the drought treatment (Fig. 4). A shift in $\Psi_L$ occurred slighter later in that treatment (Fig. 4), from around day 13 to day 15 (95% CI: 13.0 to14.6 days). Looking at the change point in relation to the control treatment over the four blocks (Fig. 5, the change in $\Psi_L$ occurred after 5 to 8 days (95% CI: 4.8 to 7.9 days), and the change in $g_s$ after 3 to 5 days (95% CI: 3.4 to 5.2 days).

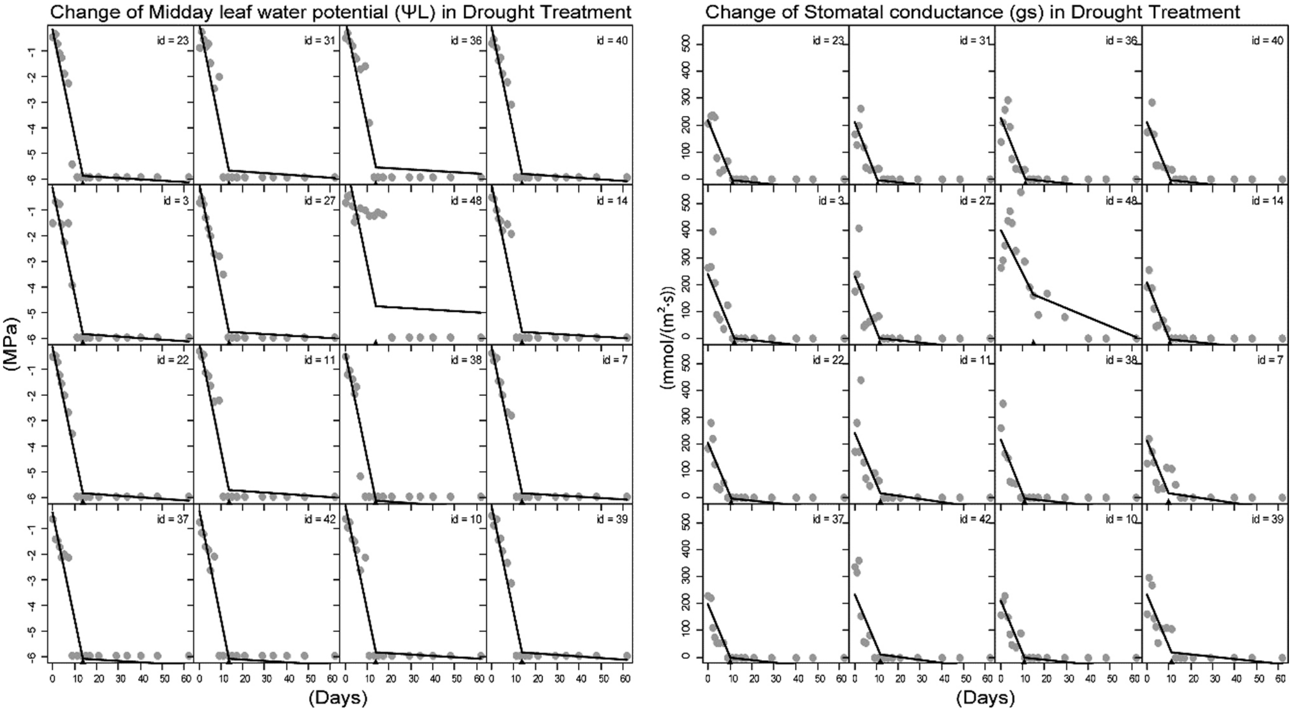
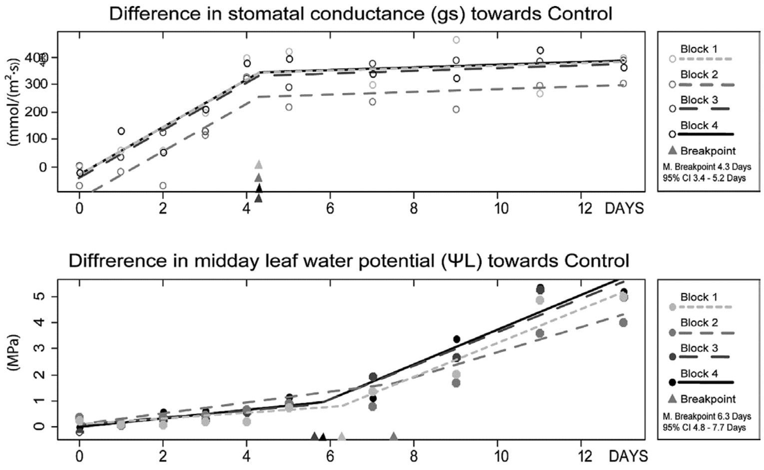


Fig. 4. Panels from random broken stick modelling of the relationship between days of drought treatment and (left) midday leaf water potential ($\Psi_L$) and (right) stomatal conductance ($g_s$). Each panel illustrates an individual seedling (n=16) in the drought treatment and the breakpoint at which it showed a marked shift in $\Psi_L$ or $g_s$


Difference in stomatal conductance (gs) towards Control

Diffrerence in midday leaf water potential (ΨL) towards Control

Fig. 5. Panels for the random broken stick model of differences in stomatal conductance (gs) as well as midday leaf water potential (ΨL) against the control and drought treatment for each block in relation to a number of days of treatment. Each panel illustrates the different blocks and their breakpoint where there is a marked shift in differences in water status between drought treatment and control

# Discussion

High species diversity is vital to enhance the resilience of urban forests to abiotic and biotic challenges in the future (Hooper et al., 2005; Alvey, 2006). Despite the recognition that many trees in urban environments are vulnerable to a changing climate, urban tree planners face uncertainties in robust species selection for future environments (Khan & Conway, 2020), especially as regards rare and/or unconventional tree species. Therefore, a better understanding of the capacity of different candidate trees to tolerate relevant abiotic stresses (drought and flooding) should be the focus of those seeking to provide species selection guidance to urban forest planners and the landscape community.

Evaluation of *A. subcordata* plants in the drought treatment showed that the plants had consumed all water in the pots after 5-6 days. Soon after that they showed a marked decrease in water status and were totally wilted after 15 days, following a trend with consistently negative water potential compared with the control. The drought-treated plants showed no increase (investment) in height during the experimental period, which is a common response to drought stress to save resources (Kozlowski & Pallardy, 1997). Evaluation of leaf water potential at turgor loss ($\Psi_{P0}$) revealed a significant difference between the treatments showing plasticity within the species. The drought-treated plants had the lowest $\Psi_{P0}$ value (-2.31 MPa). Thus, based on the Hirons and Sjöman (2018) categorisation of drought tolerance by $\Psi_{P0}$, *A. subcordata* can be considered a sensitive species with a limited capacity to tolerate drought. The vapour pressure osmometry technique for prediction of $\Psi_{P0}$ (via $\Psi_{\pi100}$) is sensitive enough to resolve differences in drought tolerance between different treatments. From a practical perspective, the technique has been proven to be more time-efficient (Bartlett et al., 2012b) and more sensitive than pressure-volume curves (Banks & Hirons, 2019), a more traditional technique used to determine $\Psi_{P0}$ in leaves. Therefore vapour pressure osmometry can be used to identify plant lineages likely to contribute to drought tolerance.

The results obtained in the waterlogging treatment are well in line with findings in previous studies of *A. subcordata* showing high survival over long periods of waterlogging (Ghanbary et al., 2012a, 2012b). In this study, the root system of plants in the waterlogging treatment was kept submerged for 63 days. Water potential in the waterlogging treatment was similar to that in the control during the first week and then dropped to slightly more negative values, but with no major deviations compared with the control. A lack of effect of waterlogging on $\Psi_L$ has been found in earlier studies (Blake & Reid, 1981; Bradford & Hsiao, 1982; Dreyer et al., 1991). According to Bradford and Hsiao (1982), this is achieved by stomatal closure. However, in the present study, the *A. subcordata* plants maintained their water potential while still achieving relatively high stomatal conductance. This shows high functional performance despite the waterlogged conditions to which the plants were exposed. Typical responses of plants to flooding include losses of plant biomass, changes in plant morphology and anatomy, suppression of vegetative and reproductive growth, and often death (Kozlowski, 1997; Shiono et al., 2008). Most plants indigenous to flooded areas have adapted to flooding in various ecological and physiological ways. Mechanisms related to flooding tolerance include avoidance strategies (formation of lenticels, aerenchyma, and adventitious roots) (Kozlowski, 1997; Dat et al., 2004; Glenz et al., 2006), escape strategies (enhanced shoot growth) (Parolin, 2002), physiological resistance strategies (reductions in stomatal conductance, photosynthesis, and root hydraulic conductivity) (Parent et al., 2008) and decreases in chlorophyll and protein levels (Kozlowski, 2002; Yordanova & Popova, 2007). Several of these traits were observed in the waterlogged *A. subcordata* plants in this study, such as restricted height growth compared with control plants and formation of adventitious roots. Producing more intercellular spaces and lower density cells to achieve more effective oxygen transport is another suggested adaptation strategy (Glenz et al., 2006). Many flood-tolerant species show increased stem diameter growth and thus decreases in slenderness index, when subjected to waterlogging, as seen in this study for *A. subcordata*. Furthermore, all waterlogged *A. subcordata* plants developed large quantities of adventitious roots (Figure 1), which are reported to have higher hydraulic conductivity under flooding conditions compared with non-adventitious roots (Kozlowski, 1997). Adventitious roots, which have a high proportion of intercellular spaces, facilitating longitudinal oxygen transport, are produced when the primary root system of the tree is impaired because of soil oxygen deficiency. The flooding tolerance of tree species seems to be closely linked to their ability to form adventitious roots (Zhang et al., 2017). For example, Whiteman et al. (1984) investigated the flooding tolerance of tropical pasture legume trees and found that species with the capacity to produce medium to large adventitious root systems had superior flooding tolerance. A study on Central Amazonian trees, such as *Salix martiana* and *Tabernaemontana juliana*, demonstrated that the main entry point of atmospheric oxygen was gas-permeable pores in the stem, near the origin of adventitious roots (Haase & Rätsch, 2010). Evidence of the role of adventitious roots in the survival of waterlogged trees is also available from temperate regions of the world, with species such as *Larix laricina, Platanus occidentalis,* and *Picea mariana* showing strong tolerance to flooding by investing in adventitious roots (Tsukahara & Kozlowski, 1985; Calvo-Polanco et al., 2012; Pernot et al., 2019). For *Alnus rubra*, Batzli and Dawson (1997) observed a drop in stomatal conductance during the process of investing in adventitious roots when flooded. It seems reasonable to assume that a similar correlation between stomatal conductance and adventitious root development exists for *A. subcordata*, although this was not directly tested in the present study. Moreover, in the study by Batzli and Dawson (1997), *Alnus viridis* ssp. *sinuata* formed few adventitious roots and did not recover its stomatal conductance. Uptake of oxygen seems also to be possible along adventitious roots growing at the water surface and in contact with the atmosphere (Haase et al., 2003). Such oxygen uptake is required for the maintenance of mitochondrial respiration (Kludze et al., 1994; Li et al., 2013). Other important functions of adventitious roots are absorption of water and nutrients to compensate for the damaged primary root system (Barlow, 1986; Calvo-Polanco et al., 2012). However, the large investment in adventitious roots is an energy-intensive process that is carried out at the expense of other investments, such as shoot growth, as observed in waterlogged trees in several studies (Herrera, 2013; Kreuzwieser & Rennenberg, 2014). In the present study, height growth was restricted in the waterlogged trees compared with the control, indicating that resources might have been allocated to root development.

Based on the results from this study, it is clear that *A. subcordata* has limited capacity to endure drought while maintaining its functionality, which makes the species unsuitable for warm and periodically dry inner-city environments or other paved environments. Therefore *A. subcordata* as an urban tree should be restricted to park and garden habitats where almost unrestricted root growth is possible. However, based on the results, *A. subcordata* may be useful for stormwater management facilities (raingarden plantings, swales, etc.) due to its high tolerance to flooding. It can therefore be recommended for wet urban environments and stormwater management areas, for which reliable guidance on suitable species is currently lacking. For example, *Acer rubrum* and *Acer ×freemanii* are frequently proposed for stormwater management plantings, but when tested for flooding tolerance by Zwack et al. (1999) showed stomatal conductance functionality that was below the capacity of *A. subcordata*. Current knowledge on physiological and molecular aspects of flooding/waterlogging tolerance in trees is far behind that in herbaceous species (Kreuzwieser et al., 2009), which makes this study important in the work of creating confidence in selecting trees for urban environments and ecosystem services.

This study focused explicitly on one species, *Alnus subcordata* (Caucasian alder), for which there is very limited knowledge outside its natural distribution, and can thus be seen as a case study. It is difficult to compare the findings with literature values, since previous studies reporting controlled evaluations and screening of tree species for flooding are rare. Further research is essential to gain a broader understanding of candidate urban tree species for the future. Results from previous controlled evaluations and screening of tree species for drought tolerance are available (Saini, 2017), but not with *A. subcordata* included. With a $\Psi_{P0}$ of -2.31 in drought conditions, *A. subcordata* can be considered sensitive to drought and comparable with other drought-sensitive species such as *Magnolia salicifolia* and *Halesia monticola* (Sjöman et al., 2018; Hirons et al., 2020).

Based on the natural habitats of *A. subcordata* close to streams and rivers, its apparent tolerance for flooding is not surprising, and controlled evaluations can be conducted to quantify and analyse the extent of its tolerance. An important aspect to consider is that an association to flood-prone areas and rivers does not necessarily indicate high tolerance to waterlogging (Sakio & Tamura, 2008) or lack of drought tolerance (Schrader et al., 2005). After 63 days of flooding, a reduction in growth was the main response, with continuously high functional performance otherwise. This indicates that the species can be a highly valuable future urban tree for open stormwater facilities or solutions purposely directing stormwater runoff to urban tree pits (Szota et al., 2019). The reuse of water in tree pits can improve their growth and water status and promote ecosystem services provision, but might also cause temporary flooding, so trees in these modern installations need to tolerate temporary inundation. Kreuzwieser and Rennenberg (2014) concluded that adult trees tolerate waterlogging and flooding better than seedlings of the same species. The present study was based on two-year-old saplings, and the capacity for flood tolerance could be even stronger in adult trees, but this remains to be investigated. Moreover, given the high complexity of invasiveness (Lustig et al., 2017) and evidence of negative invasion impacts of introduced tree species (Richardson & Rejmánek, 2011), the introduction of new species such as *A. subcordata* should be evaluated for each specific urban geographical context of the introduction to enable local recommendations for where and how it can be planted to limit associated invasion risks while delivering wanted ecosystem services (Sjöman et al., 2016).

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