

## THE ROLE OF LIVING BARK IN *BETULA PENDULA* ABILITY TO REFILL EMBOLISED XYLEM

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### INTRODUCTION

Embolism formation and its potential run away spreading through the plant vascular system has received a lot of attention in the recent years, as it appears central to the observed drought-induced mortality of trees. However, the tree ability to recover from such embolism has received less attention. In particular, the tree ability to refill embolism causes by drought or freezing stress is under debate in the last recent years. This debate has been fuelled by the discovery that some earlier method used to assess tree embolism and refilling ability were prone to artefact (e.g., Wheeler et al. 2013). And if or when refilling happens, the mechanisms allowing it remains unknown despite several hypothesis (Nardini et al. 2011).

However, our recent studies have nonetheless shown that silver birch (*Betula pendula*) might be able to refill (Salmon et al. 2018), and hypothesised that it might be related to its ability to pressurised sap in the spring prior to budburst (Hölttä et al. 2018). These earlier studies led to two hypothesis about the mechanisms involved in the refilling process: 1) the phloem in the living bark plays a role in initiating the refilling; 2) the refilling process is an activate process involving energy cost for the tree, which are supplied by the stored starch and non-structural carbohydrates.

We conducted an experiment to test these two hypothesis and further explore the mechanisms underlying birch refilling ability.

### METHODS

Branches of silver birch were sampled at the greenhouse of the university of Helsinki, Viikki campus. The sampled branches were longer than 70cm and without divisions. The sample was let to bench dry until fully embolised and not conductive., with a water potential ( $\Psi$ ) lower than -3 MPa (Choat et al. 2012) as controlled by water potential measurement of a 10 cm subsample with a pressure chamber. The samples were divided in two subsamples of more than 30cm length to avoid open vessels artefacts. One subsample was peeled of its bark, phloem and cambium and wrap with PTFE tap to limit evaporation and avoid leakage during the refilling experiment.

For each subsample, hydraulic recovery was induced with a small hydraulic positive pressure provided by a water column attached to the upper end of the branches held vertically on a stand. The amount of water flowing through each subsample was recorded with balances to measure the restoration of the hydraulic conductivity.

After each measurement, a 5 to 10cm piece was cut from the subsamples and used to measure non-structural carbohydrates and starch concentrations. The cut piece was frozen in liquid nitrogen to stop enzymatic activity, dried and ground to powder and stored until analyses. The procedure continues with

shorten sub-samples until the branches stopped being able to refill or became too short for further measurements (i.e. after a maximum of three refilling attempts).

We also measured the osmolality of the first drop of sap coming out of the refilling branches to assess whether refilling might have happened by loading sugars in the xylem to osmotically draw water from surrounding cells.

### CONCLUSIONS

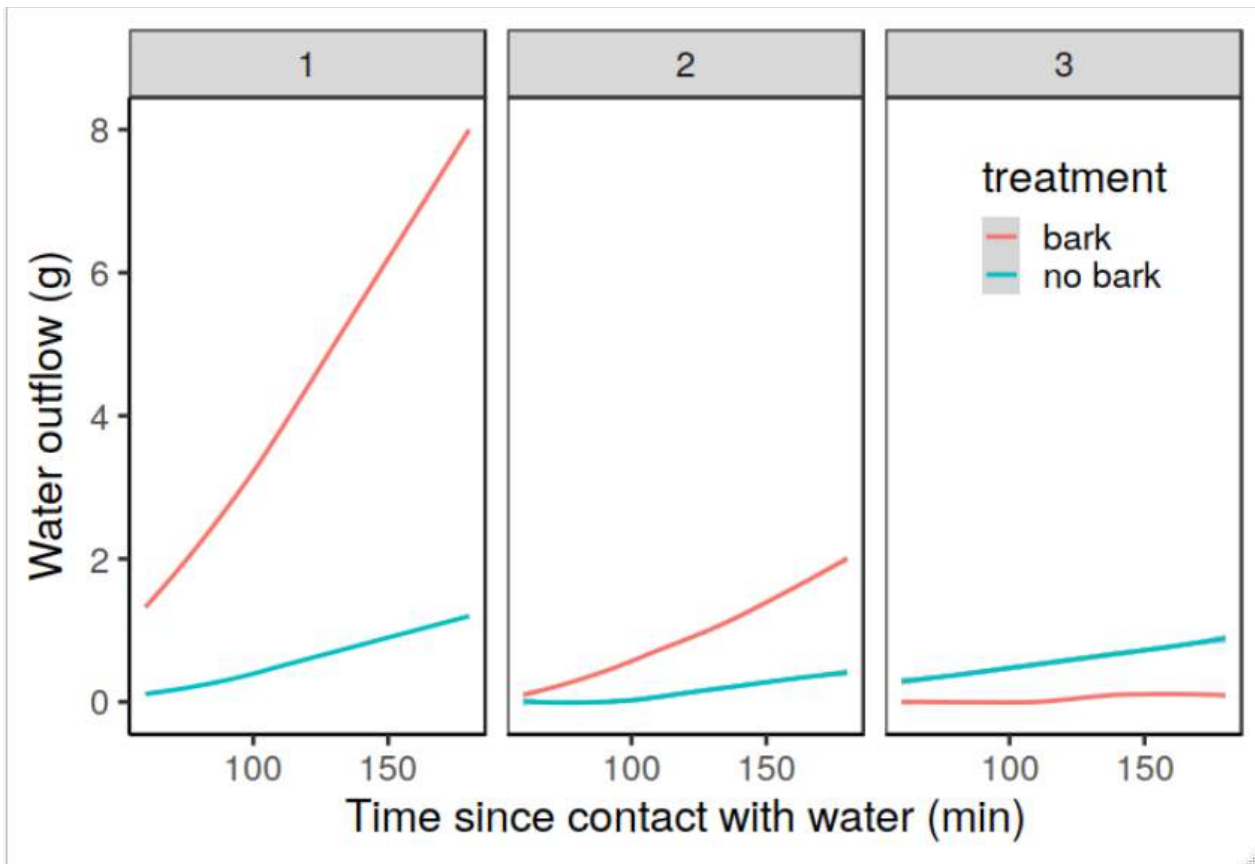


Figure 1: Amount of water conducted through a branch of silver birch as a function of time since the branches was put in contact with water per successive runs of embolism and refilling (1, 2 and 3) for sub-samples with the living bark intact (“bark”, red) or with the living bark removed (“no bark”, blue).

The preliminary results show the presence of living bark originally allows the studied branches to restore conductivity faster than the branches on which living bark has been removed (Fig. 1, panel 1). The refilling ability of branches with living bark clearly declines with the number of refilling events (compare runs 1, 2 and 3), eventually becoming smaller than that of the branches without bark.

The osmolality of the sap after refilling is not sufficient to explain the refilling through sugar loadings in the embolised xylem.

On-going measurements of non-structural carbohydrates and starch are expected to shed further light on the potential mechanisms underlying these differences between treatments as well as improve our understanding of the mechanisms underlying silver birch refilling ability.

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## REFERENCES

- Choat B., Jansen S., Brodribb T.J., Cochard H., Delzon S., Bhaskar R., Bucci S.J., Field T.S., Gleason S.M., Hacke U.G. et al. (2012). Global convergence in the vulnerability of forests to drought. *Nature* 491: 752–756
- Hölttä, T., Dominguez, M., Salmon, Y., Aalto, J., Vanhatalo, A., Bäck, J. and A. Lintunen (2018). Water relations in silver birch during springtime. How is sap pressurized? *Plant Biology*, 20: 834-847.
- Nardini, A., Lo Gullo, M.A., and Salleo, S. (2011). Refilling embolized xylem conduits: Is it a matter of phloem unloading? *Plant Sciences*. 180, 604-611.
- Salmon, Y., Lintunen, A., Lindfors, L. Suhonen, H., Sevanto, S., Vesala, T. and T. Hölttä (2018). Silver birch ability to refill fully embolised xylem conduits under tension. *Acta Horticulturae*, 1222, 67-73.
- Wheeler JK, Huggett BA, Tofte AN, Rockwell FE, Holbrook NM (2013) Cutting xylem under tension or supersaturated with gas can generate PLC and the appearance of rapid recovery from embolism. *Plant Cell and Environment*, 36, 1938-1949