ESRF Experimental Report

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OBSERVATION OF EMBOLISM FORMATION AND REFILLING IN PLANTS USING SYNCHROTRON BASED X-RAY MICRO COMPUTED TOMOGRAPHY

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Overview

Successful observations of water transport were made in a number of plant species by HRCT at the ID19 beam line. Experiments were directed towards establishing the degree to which the formation of gas emboli in xylem conduits impairs plant water transport with progressive drought. We evaluated vulnerability to embolism of Eucalyptus, Spruce and Pine species. We used two scan stages in our measurements. The larger stage allowed us to mount and scan the stem of intact plants utilizing a custom built pot holder (Fig 1). A smaller stage and a second camera were used to make higher resolution scans of detached leaves and small stems.

Measurement Quality

Initial issues with image quality were solved by adjusting the energy of the x-ray beam (18 keV). After this point, all scans taken were of excellent quality, with very good resolution (3.5 μm and 1.4 μm respectively for the first and second settings) and excellent contrast between gas, liquid and plant tissue volumes due to the great signal to noise ratio. This allowed us to easily distinguish between embolised xylem conduits from water-filled ones, as well as many other anatomical features (Fig 2). Movement of the sample was sometimes an issue during scans made on intact plants using the larger stage. This was only a minor problem in general. Scan time on the larger stage was roughly 12 mins and ca. 4 mins on the small stage (camera with small field of view). The spatial and contrast resolutions and scan time provided by the second stage were particularly impressive, allowing us to see details of tissue deformation in leaves as they dried.

Results

The 72 hours of the beam time were necessary and 100% utilized. More than 100 tomographic scans were performed, allowing us to follow the dynamics of a plant's response to water stress. The results of the experiments are very exciting and will be published in high impact journals in the field of plant sciences (e.g., *New Phytologist, Plant Physiology*). We are currently analysing the images we collected with the aim of correlating our non-invasive observations with traditional, destructive measurement techniques used across the world. The results are exciting because they represent the first in vivo measurements of vulnerability to drought-induced embolism. Scans made on Pine and Eucalyptus leaves also open up exciting possibilities for evaluating how deformation of tissue in the leaf will affect gas and liquid pathways important to photosynthesis and transpiration.

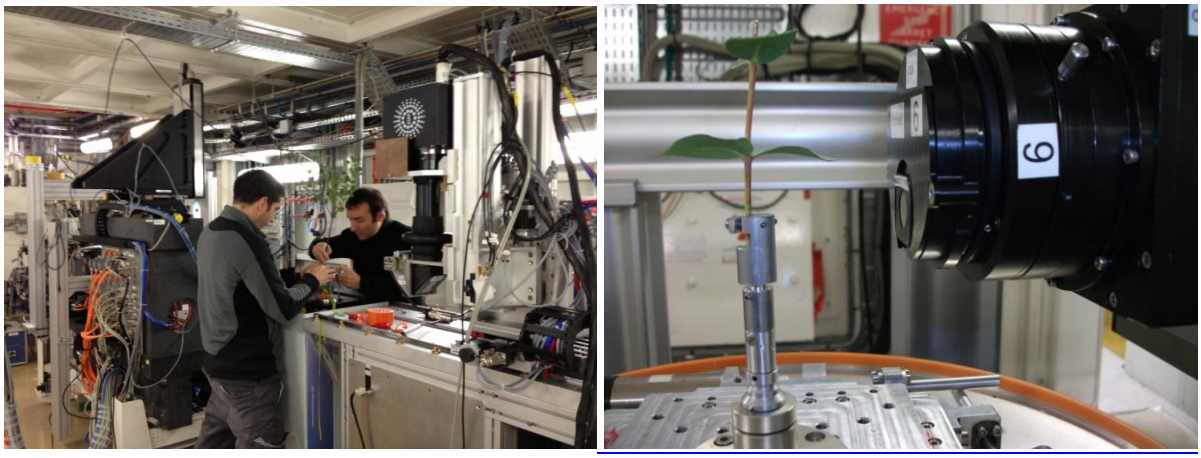

Fig 1. Setting up a potted *Eucalyptus* plans to be scanned in the ID19 hutch. Plants are held in place by a custom pot holder using the large field of view camera (A) or smaller detached samples were scanned using the high resolution settings (B).

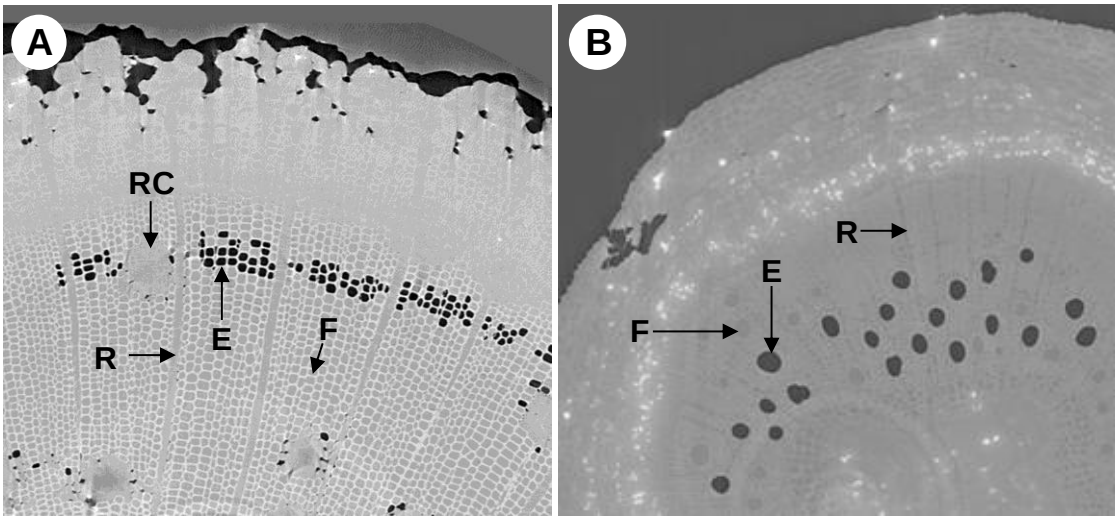

Fig 2. A. Cross sectional slice from scan of pine stem. Embolised (E) and water-filled tracheids (F) in the xylem tissue are easily distinguished. Rays (R) and resin canals (RC) can also be observed. **B.** Cross sectional slice from scan of Eucalyptus stem showing embolised (E) and water-filled (F) vessels.