A Cryogenic Sample Environment for the Analysis of Frozen Hydrated Biological Tissue at the Hard X-ray Micro/Nano-Probe Beamline P06 at PETRA III

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A cryogenic sample environment was developed at beamline P06 to minimize structural and chemical artefacts during analytical X-ray microscopy. The instrument completes a cryogenic workflow from shock freezing of small samples up to 2 mm diameter, especially hydrated biological and medical tissue. The components of the cryogenic environment are: 1) a two chamber vacuum system with ca. 10^{-8} mbar, 2) two cooling systems, one for rapid cooling at startup and one for vibration free long term temperature maintenance between -120°C and -150°C, 3) detectors, such as an ultrathin window X-ray fluorescence detector for fluorescence tomography, a PCO4000 camera for absorption and phase contrast tomography, 4) sample stages and 5) a shuttle transfer system. The top view port allows at present a macroscopic view of the sample area. This is currently extended to imaging by an additional light microscope, to allow microscopic viewing of the sample colinear with the incoming beam via a punctured mirror system.

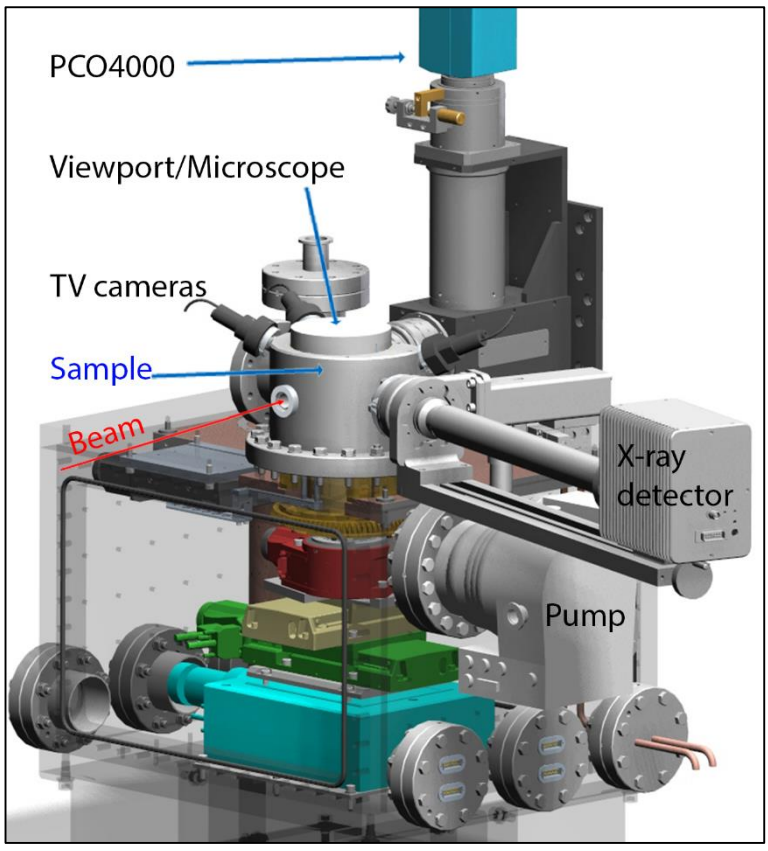

Figure 1 *The cryogenic sample environment. The lower compartment contains the stages, all electrical connections, a low vibration turbo molecular pump, an internal cooling Dewar and the rapid cooling system, using a liquid nitrogen flow system. The beam traverses the smaller upper chamber that allows to closely position the detectors and the optional beam focusing mirrors to the sample.*

Here we show examples of first applications. Samples were prepared on site and shock frozen by plunge freezing in liquid propane to -190°C, then transferred into liquid nitrogen for temporary storage. Samples were then transferred into an evacuated cryogenic shuttle for insertion into the chamber for analysis. Two TV cameras aid to align the sample for insertion into the cold stage.

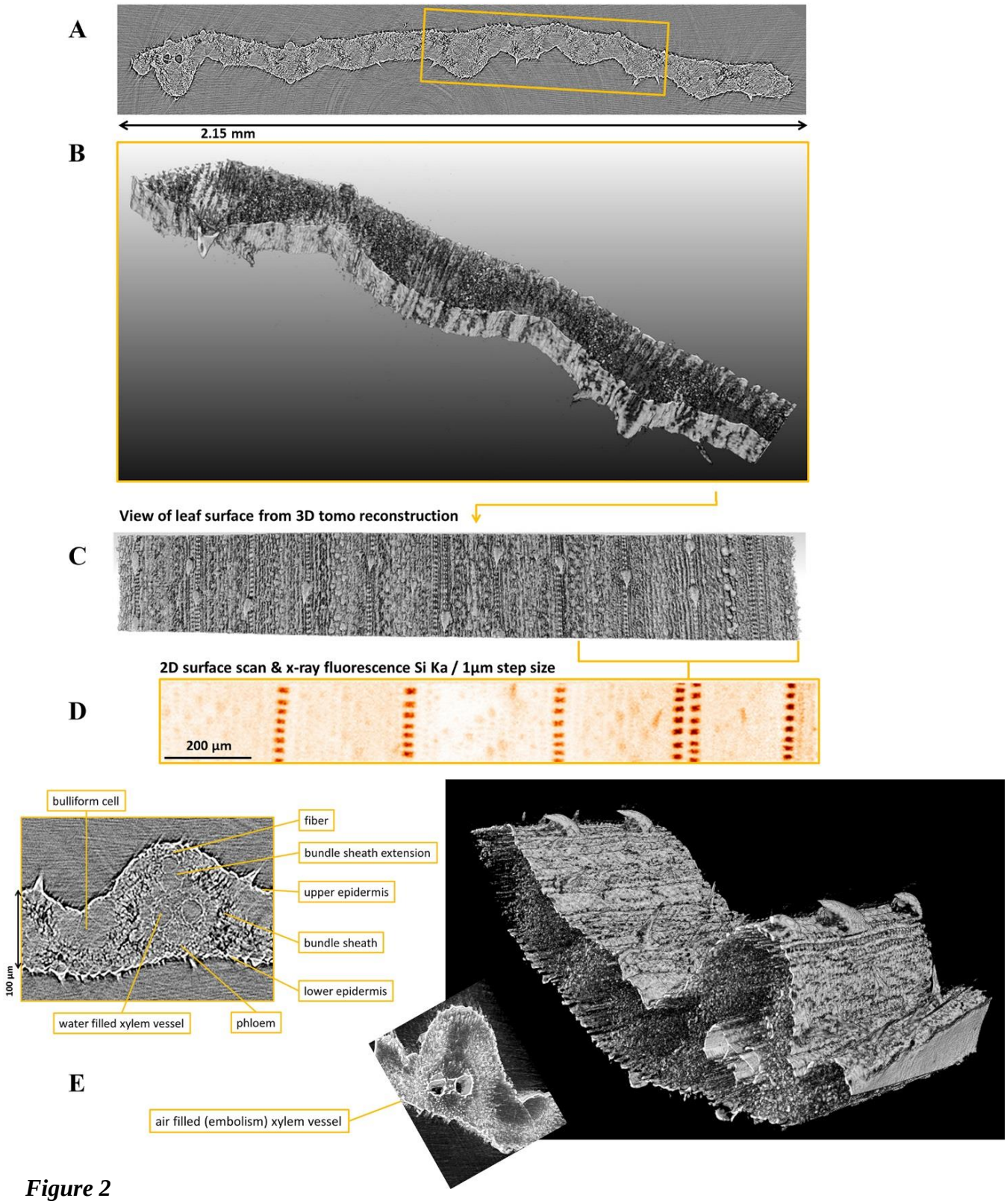

Figure 2

A frozen hydrated rice leaf serves as example of maximal size. We recorded a 3D mixed absorption/phase contrast CRYO-CT at 12 keV with 0.3° steps at 2s exposures. Image (A) shows a virtual cross section of the leaf, a single slice reconstruction. The 3D volume in (B) includes the area indicated in (A). A computed 3D volume similar to (B) is aligned for a detailed front view of leaf surface structures (C). To demonstrate the ability to detect x-ray fluorescence down to low energies the leaf surface was scanned using a focused beam of 0.8µm, $<10^{10}$ photons/s, and the X-ray fluorescence recorded to obtain 2D element maps. The element map from the surface scan shows Si (D) as an example map primarily present in lines of Si enforced structures, presumably a defence against herbivores. Some internal leaf structures are pointed out in (E).