

# ENHANCED CRYOGENIC COOLING FOR IMAGING OF LARGE SAMPLES BY X-RAY MICROSCOPY

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Developments in instrumentation including cryogenic sample environments provide platforms for multimodal tomography application. To increase cryogenic cooling capabilities for large samples, we developed Cryostream 1000 Wide Nozzle. It provides a much larger gas volume when compared to the Standard model and facilitates vitrification of samples up to 3 mm in diameter. The Wide Nozzle has been successfully used to cool large crystals and biological specimens for X-ray and neutron diffraction and scattering studies, and hard X-ray microscopy tomography.

We present the development of a lab-based cryogenic hard X-ray imaging system (microCT), designed for sub-micron resolution of biological specimens with reduced need for contrast agents which might affect biological systems.

Utilising the Oxford Cryosystems Cryostream 1000 Wide Nozzle, in conjunction with a Zeiss Versa 610 X-ray Microscope, we demonstrate that delicate biological samples, otherwise degraded under lab X-ray imaging conditions, can be successfully imaged under stable cryogenic conditions over a period of several hours.

Whilst further tests are required, we observed enhancement of absorption contrast and signal-to-noise ratio by reduction of thermal scattering. We visualized fine structural details without heavy metal staining and other associated artefacts that often arise in conventional X-ray imaging.

To map thermal gradients within the cryogenic stream, we used a thermal diode providing precise temperature data across the gas column, helping to guide future optimisation of cooling dynamics. These insights are important for ongoing work aimed at expanding the Cryostream's application to a range of materials, with potential implications for fields like biomedicine, tissue preservation, tomography, aerospace, energy materials, and heritage artefacts.

This cryogenic X-ray system, therefore, represents a significant step towards lab-based cryogenic X-ray imaging, and further developments will focus on quantifying the contributions of X-ray absorption and scatter components at cryogenic temperatures, broadening the system's capabilities for multi-modal microscopy and in situ analyses.

**Keywords:** X-ray microscopy, hard X-ray, microCT, multimodal microscopy, sample environments, cryogenic cooling, sample preservation, vitrification, tomography, open flow cooling, Cryostream 1000, Cobra, Wide Nozzle