

Workshop on Sample Preparation Techniques for Life Science-Applications at Beamline L

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About ten different European user groups work on biomedical research projects at the SRXRF/XANES-microprobe beamline L. For this community a dedicated one-day workshop about life science-applications at this beamline was held at HASYLAB on 17.03.2006. The workshop placed emphasis on different aspects of sample preparation techniques, but also general topics like imaging techniques in life sciences, histological methods, and XAFS were addressed. The workshop was organized by M. Kühbacher and G. Falkenberg and had about 20 participants.

M. Kühbacher opened the workshop with a general introduction into the research on metalloproteins. Metalloproteomics requires specific bio-analytical tools for the identification of these proteins, their distribution, and biological functions. The importance of controlling the entire analytical process, starting with the right animal model and sample taking, was emphasized.

A practical exercise was given directly at the beamline on appropriate clean sample handling. A technique was demonstrated which enables plane mounting of 4 µm thin substrate Mylar films on a slide frame-sized sample holders. After sectioning with a cryotome, the thin tissue section is placed on the substrate film and is fixed by electrostatic force. Different other approaches for sample mounting were discussed by participants.

M. Lankosz from the Univ. Science and Technology, Cracow, talked about “Preparation of central nervous system tissue for micro-SRXRF and micro-XANES”. Different experiences with freezing, fixation, mounting procedures, conditions of sample storing, the effect of radiation damage, and photo-reduction during X-ray measurements have been addressed.

As the first step after sample-taking, the freezing procedure is important for the preservation of the morphology of the tissue samples. Different experiences with freezing with liquid nitrogen and problems with damaged tissue as a consequence of time delays especially with human post-mortem samples were discussed.

The role of the so-called fixation of the tissue was discussed in detail. The histological fixation is a chemical process in which biological tissue is preserved from degradation. Fixation terminates enzymatic reactions, and increases the mechanical stability of the tissue. Formaldehyde is one of the most commonly used fixatives. Better known under the name formalin it cross-links the proteins. Although this process is usually used in many histological staining procedures, it has the disadvantage of washing out elements like chlorine, potassium, and others. Beyond that, it is also a potential source of contamination. As a result of the open discussion, there was agreement about the need of further investigations on the effect of fixation on the microscopic distribution of metals.

Closely linked with the fixation procedure is the subject of contamination. Because most of the elements under investigation are present in trace amounts even in clean laboratories, it is quite important to avoid contaminations. Possible sources of contamination are the operation cutlery, the steel microtome knife, and the blood in the tissue itself, or the used perfusion solution. To limit contamination it is mandatory to work with a well-trained staff and even better to control the whole process with the eyes of an analytical chemist.

In the final talk, G. Falkenberg reported about recent technical developments at beamline L, namely capillary optics, detector development, and fast scanning mode. Sensitivity and reliability of the instrument enable high sample throughput and statistics, which is a basic requirement for biomedical research.