Quality Improvement of Soft X-Ray Image in FCSXRM ETL Mark III

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The table-top Flash Contact Soft X-ray Microscope (FCSXRM) ETL Mark III is the soft x-ray microscope system for convenient use in laboratory [1-2]. Laser-produced plasma is suitable for a flash x-ray source. An x-ray image of a specimen is recorded in terms of contact mode on a photo resist PMMA (polymethyl methacrylate) membrane supported on a silicon chip of 5-mm square. After chemical development of the PMMA chip, the x-ray image is enlarged with an atomic force microscope (AFM). The principle of soft x-ray imaging for a living cell is carbon imaging, which provides a carbon density map of the specimen in water. In the soft x-ray wavelength range, carbon and oxygen have absorption edges at 4.4nm and 2.3nm respectively. The mass absorption coefficient of carbon is about 10 times greater than that of oxygen within the wavelength range between these absorption edges. Using this wavelength range (so called “water window”), one can obtain the soft x-ray image of the living cell in water.

Quality of the x-ray image obtained by FCSXRM ETL Mark III depends on the uniformity of the thickness of water surrounding the specimen. In this system, an x-ray window (0.5-mm square) made of silicon nitride membrane separates a vacuum chamber for plasma production from a sample holder kept under atmospheric conditions. Flexure of the x-ray window due to pressure difference between the vacuum chamber and the sample holder causes the non-uniformity of water thickness under the x-ray window, and then it produces crucial unevenness of contrast in the x-ray image. In case of a large specimen with several hundreds micrometers in size, the unevenness of contrast is critical at the central part of the x-ray window. To reduce the flexure of the x-ray window during x-ray exposure, the vacuum chamber was filled with helium gas and/or a spacer was located between the PMMA membrane and the x-ray window. These improvements gave successful x-ray images of large cells cultured on the PMMA chip. Using this technique, extra cellular matrix of 3Y1 cells were examined. A possible role of soft x-ray imaging in biological research will be discussed.

References

[1] Shimizu, H., Tomie, T., Majima, T., Stead, A.D., Ford, T. W., Miura, E., Yamada, M. and Kanayama, T. “A Table-top Flash X-Ray Microscopy.” In: X-ray Microscopy and Spectroscopy. (Thieme, J. et al., eds.), Springer-Verlag, Berlin, pp.**I**-157 (1998).

[2] Majima, T. “Soft X-ray imaging of living cells in water: flash contact soft X-ray microscope.” Trends in Analytical Chemistry, **23**, 520-525 (2004).