

X-ray imaging of mucilaginous sheath of phytoplankton in Lake Biwa by soft X-ray microscope

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Lake Biwa is the largest lake in Japan and occupies approximately 1/6 of Shiga Prefecture. Recently, its chemical oxygen demand (COD) index is increasing in spite of a decrease in the values of biochemical oxygen demand (BOD) index. In order to elucidate the mechanism, the mucilaginous sheath of phytoplankton lived in Lake Biwa has been observed with a soft X-ray microscope.

Mucilage of blue-green algae or green algae is considered as an important source of dissolved organic matter from natural sources. *Phormidium tenue* is a single trichome within the mucilaginous sheath. Nevertheless, the mucilaginous sheath of *P. tenue* has been not experimentally-identified. *Microcystis wesenbergii* is a spherical cell and the cells are embedded in thick colorless homogeneous mucilage with clearly defined contours. To observe the natural ambient images of mucilaginous sheath, we examined *M. wesenbergii* and *P. tenue* under the soft X-ray microscope.

Laboratory-cultured *M. wesenbergii* and *P. tenue* cells have been observed by soft X-ray microscope (BL-12). There is no requirement for fixation, staining, or coating. Air-dry and wet samples were observed under atmospheric pressure at 1.9 nm and 2.3 nm wavelength.

Figures 1 show X-ray microscopic images of *M. wesenbergii*. Moist cells shrank during drying, as seen in (A). On the other hand, cells under the moisture condition remain spherical shape, as seen in (B). There are two kinds of cells that show a

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different X-ray absorption characteristic. One shows high X-ray absorption and the other shows low X-ray absorption. The former is a normal and healthy cell, and the latter is a skeleton cell due to loss of internal content. The contrast ratio of the healthy cell, the skeleton cell, and a latex sphere is about 5:1:3. It was impossible to define contours by X-ray image since the cells produced mucilage in huge quantities.

Figures 2 show X-ray microscopic images of *P. tenue*. Each cell has a granule with a diameter of about 1 μm . With 1.9 nm observation the mucilaginous sheath is not experimentally-identified. In order to elucidate the existence of the mucilaginous sheath, a new observational method will have to be introduced.

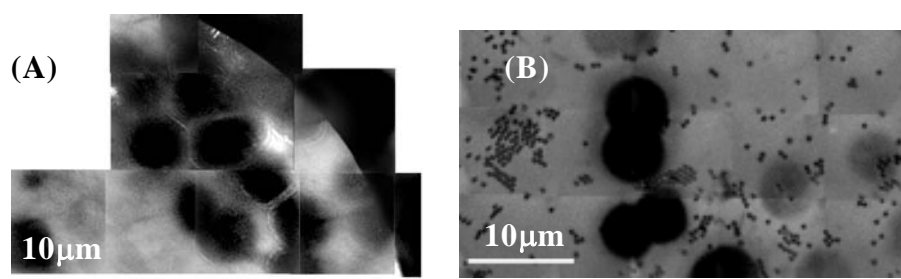


Fig.1 X-ray microscopic images of *M. wesenbergii*. (A) is dry sample and (B) is wet sample with blue latex spheres (500 nm in diameter). Observation wavelength is 2.3 nm and exposure time is 2 min/field.

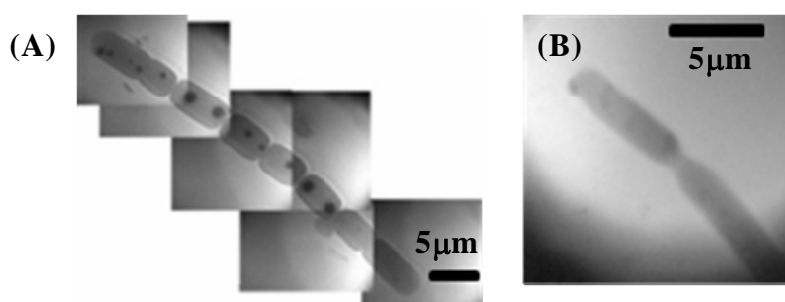


Fig. 2 X-ray microscopic images of *P. tenue*. (A) is dry sample and (B) is wet sample. Observation wavelength is 1.9 nm and exposure time is 1 min/field.