Infrared Micro-Spectroscopy Observation of Plantplankton in Lake Biwa

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Recently many cyanobacteria in Lake Biwa are surrounded by mucilaginous external layers. The mucilage sheath is expected to play an important role in revealing the causative substance of the diremption phenomenon of BOD and COD in Lake Biwa [1]. The algal cell is composed mainly of protein and the mucilage sheath is composed mainly of polysaccharides plus a small amount of protein.

Infrared micro-spectroscopy is useful for studying local structures of organic materials and biomaterials. In the SR center, an infrared micro-spectroscopy beamline (SRMS, BL-15) was constructed by a JST project (from the fiscal year of 2007 to 2010) [2, 3]. The infrared micro-spectroscopy was explored as a means of discriminating between the mucilage sheath and cell of cyanobacteria.

The mucilage sheath was isolated from *Synechococcus* sp. (Ikegaya *et al*. 2012 *in preparation*). Using infrared microspectroscope is Nicolet 6700 and Continuμm XL (Thermo Fisher Scientific Inc.) equipped at the end of the beamline. The specimen suspension was dropped onto an aluminum coated glass substrate and air-dried. The measurements were using the reflection mode and 32x Cassegrain optics. The aperture size is 25 μm x 25 μm and the sampling step is 100 μm.

The peptide group, the structural repeating unit of proteins, gives up to 9 characteristics bands named amide A, B, I, II,…VII. The amide I and II are the most intense bands of the protein IR spectrum. The amide I band between 1600-1700 cm^{-1} is mainly associated with the C=O stretching vibration and is directly related to the backbone

conformation of proteins. Amide II results mainly from the N-H bending vibration. In addition, the amide A band about 3300 cm^{-1} is associated with the N-H stretching vibration. Cell was discriminated by the amide I band (1600-1700 cm^{-1}), II band (1500-1600 cm^{-1}) and A band (3300 cm^{-1}).

Figure 1 shows the reflection spectra of the cell and mucilage sheath. In spectra of the cell, three absorption peaks occur around the spectra drawn with blue lines. On the other hand, in spectra of the sheath, a definite peak is indeterminable. These spectra show that Infrared micro-spectroscopy is effective for discriminating cell from mucilage sheath. However, it is difficult to determine the constituent carbohydrate of the mucilage sheath by the spectra. The IR data analysis of the mucilage sheath needs more research for a specific absorption of various carbohydrates.

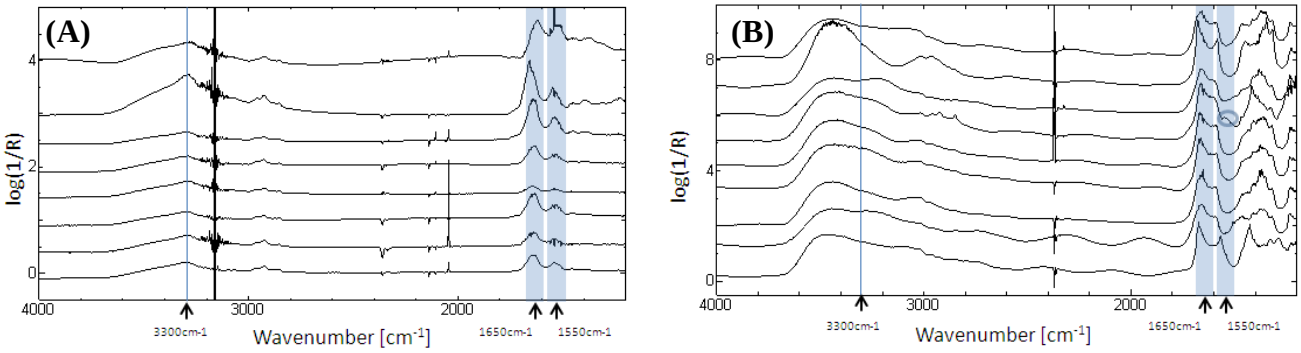

Figure 1 IR microscopic spectra from cell (A) and mucilage sheath (B). The amide I band (1600-1700cm^{-1}), II band (1500-1600cm^{-1}) and A band (3300 cm^{-1}) were used to refer to indicate the existence of cell, protein.

References

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