Application Note

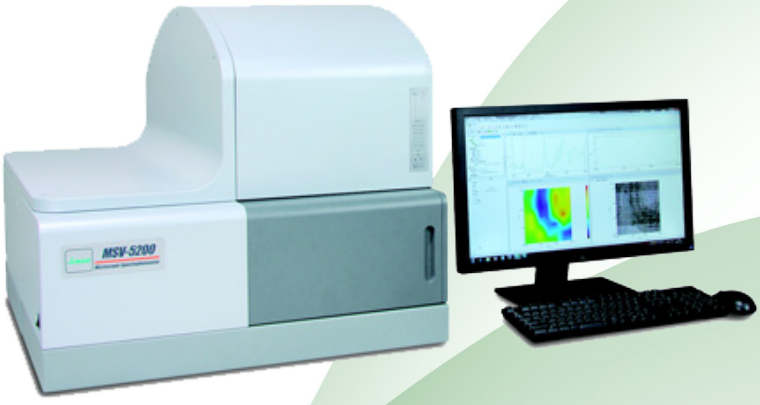
Transmission Measurement of Volvox Algae Using the MSV-5000 Series

Introduction

The MSV-5000 series micro-spectrophotometer can analyze a microscopic sample/area by both transmission/reflection measurements in the region from the UV to Near IR, which can be applied to characterization of a micro sized sample/area or impurity analysis applications.

This type of technology is also very popular in the bioscience field for the analysis of localized constituents in living cells.

Volvox, which has a localized cellular density due to its internal daughter colonies, was measured to obtain the absorption spectra and fixed-wavelength mapping data.



MSV-5000

Keyword: microscopic measurement, biochemical, spectrum imaging (mapping)

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System Configuration

MSV-5100	UV/Vis/NIR Microscopic Spectrophotometer
MAXY-501	Automatic XYZ Stage
Detector:	X-LC 3110MD

Measurement Conditions:

Spectral bandwidth (UV/Vis):	5.0 nm
Scan speed:	100 nm/min
Response:	Quick
Cassegrain objective:	16 times
Scanning speed:	1000 nm/min
Data pitch:	1 nm
Aperture:	50 mmf

Spectrum measurement

One of the daughter colonies inside of the mother colony is measured to obtain the absorption spectrum.

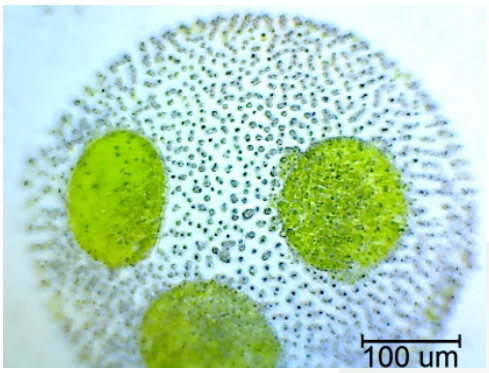

Fig. 1. Observation Image of dried Volvox

Results

Measured absorption spectrum is shown in Fig. 2. Chlorophyll a and chlorophyll b are major chlorophyll components included in inland plants and green algae 1-2) and those absorption spectra are shown in Fig. 3.2) This published data in the literature is measured under acetone solvent conditions, and the peak positions of those chlorophylls are slightly different depending on the solvent used, but the wavelength shift is only approx. 2-7 nm different. Comparing the spectra of chlorophylls with the Volvox spectrum, it is assumed that chlorophyll a and b are included in Volvox.

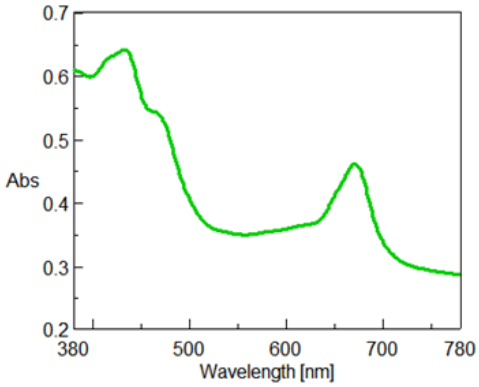

Fig. 2. Absorption spectrum of dried Volvox

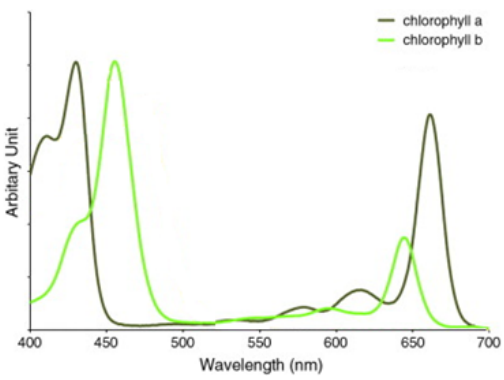

Fig. 3. Absorption spectra of chlorophylls[2)] (with Acetone solvent)

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Fixed Wavelength Mapping Measurement

A fixed wavelength mapping measurement was executed using the 672 nm peak since the major peak at 672 nm was observed in the absorption spectrum measurement. A mapping measurement at a specified fixed wavelength makes it possible to generate high speed mapping data.

Measurement Conditions:

Measurement Mode:	Lattice measurement
Measurement wavelength:	672 nm
Spectral Bandwidth:	2 nm
Aperture:	30 µmf
Response:	Fast
Cassegrain objective:	16X
Measurement interval:	30 µm

Results

The observation image and the corresponding color image obtained by the mapping measurement are shown below. The area with the higher cellular density in the observation image is in good agreement with the area of the higher absorbance values in the color mapping image.

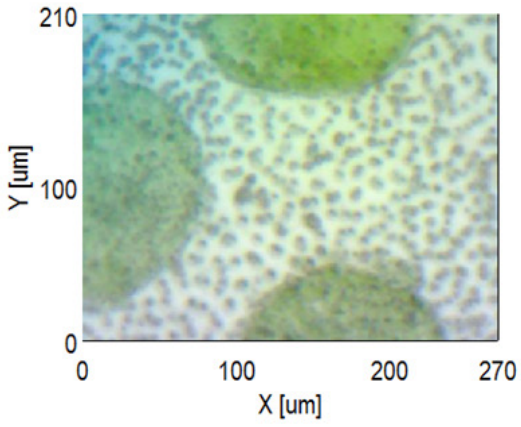

Fig. 4. Observation Image

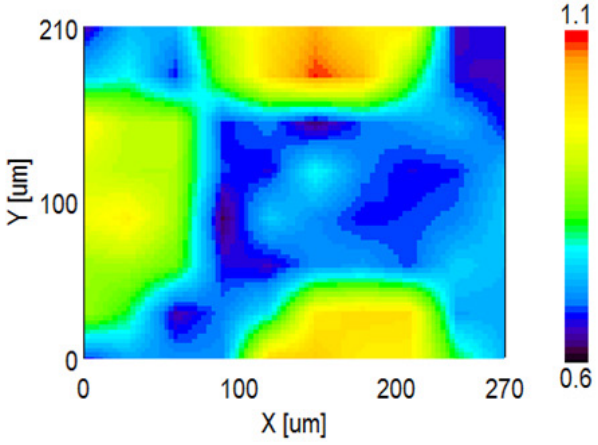

Fig. 5. False color image of the fixed wavelength mapping measurement

Reference Literatures

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