Analysis of Microbial Culture Medium by LC-MS

During LC analysis, connecting a Diode Array Detector (DAD) and a MS Detector in series allows simultaneous collection of UV spectra and mass spectra. As a result, a large amount of information about a sample can be obtained. This approach is effective for the monitoring of reactions or the confirmation of by-products during a synthesis. In this report, the dual-detector approach is applied to the screening of active ingredients in microbial culture medium.
The analysis result of the scan mode obtained by the MS Detector is presented in the form of a contour plot, which is a familiar representation of Photo Diode Array data. This style of plot allows confirmation of the overall spectra as a function of time.

5610 MS Detector

LC-MS Analysis

■ Analytical Conditions

Table 1 MS Detector Settings

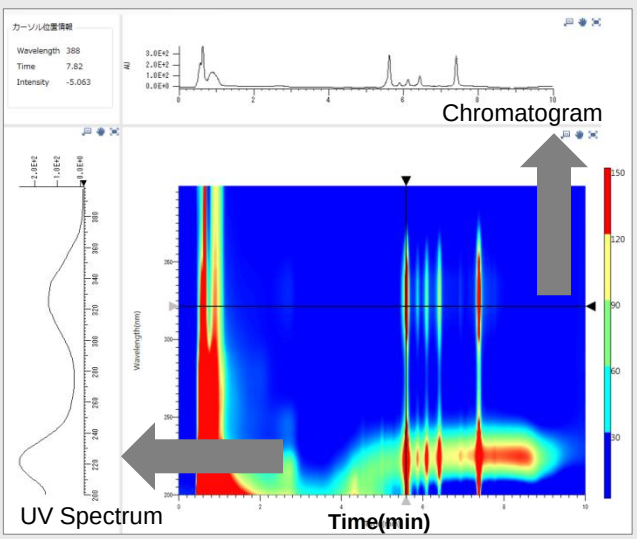
Ionization method	ESI
Ionization mode	Positive
Ionization voltage	2700 V
Measurement mode	Scan: (*m/z* 200-400)

Table 2 Analytical Conditions for HPLC

Column	LaChromUltra (1.9 µm) 2.0 mm I.D. x 50 mm
Mobile phases	A: 0.1% HCOOH in H_2O (v/v) B: 0.1% HCOOH in CH_3CN (v/v) %B = 20(0-0.5min) - 100(3-5min) - 20(5.1-10min)
Flow rate	0.2 mL/min (Split ratio = 1:50)
Injection vol.	20 µL

■ LC-DAD-MS Analysis

DAD



MS Detector

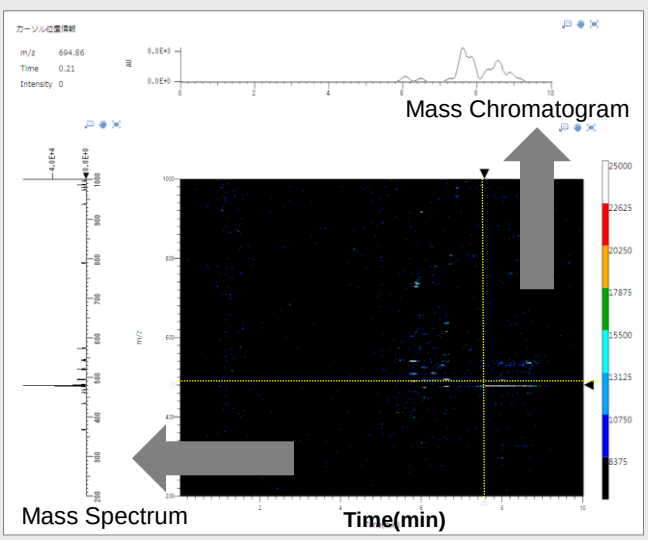


Figure 1 Contour Plot Based on DAD and MS Scan Analysis Results

20 µL of the culture medium supernatant extract was injected. The purpose of this analysis was to screen the active ingredients in the microbial culture medium. Therefore, a short column with a length of 50 mm and 10-minute cycle with gradient elution were selected for the separation conditions on the LC side. The spectral information of an arbitrary component can be obtained from the DAD analysis result, and at the same time, the mass information is provided by the MS detector.
In the short, 10 minutes analysis, useful information for screening the active ingredients can be obtained.

Confirmation of UV Spectrum and Mass Spectrum

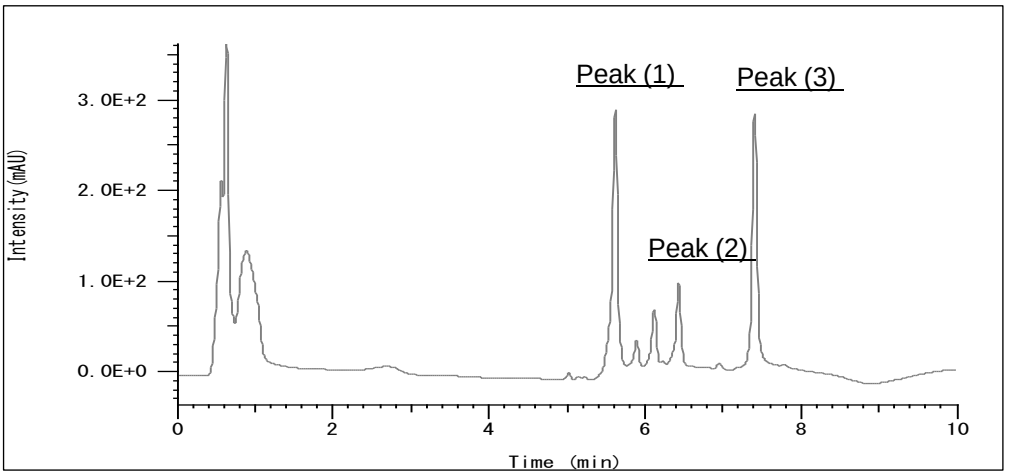


Figure 2 Chromatogram at UV 324 nm

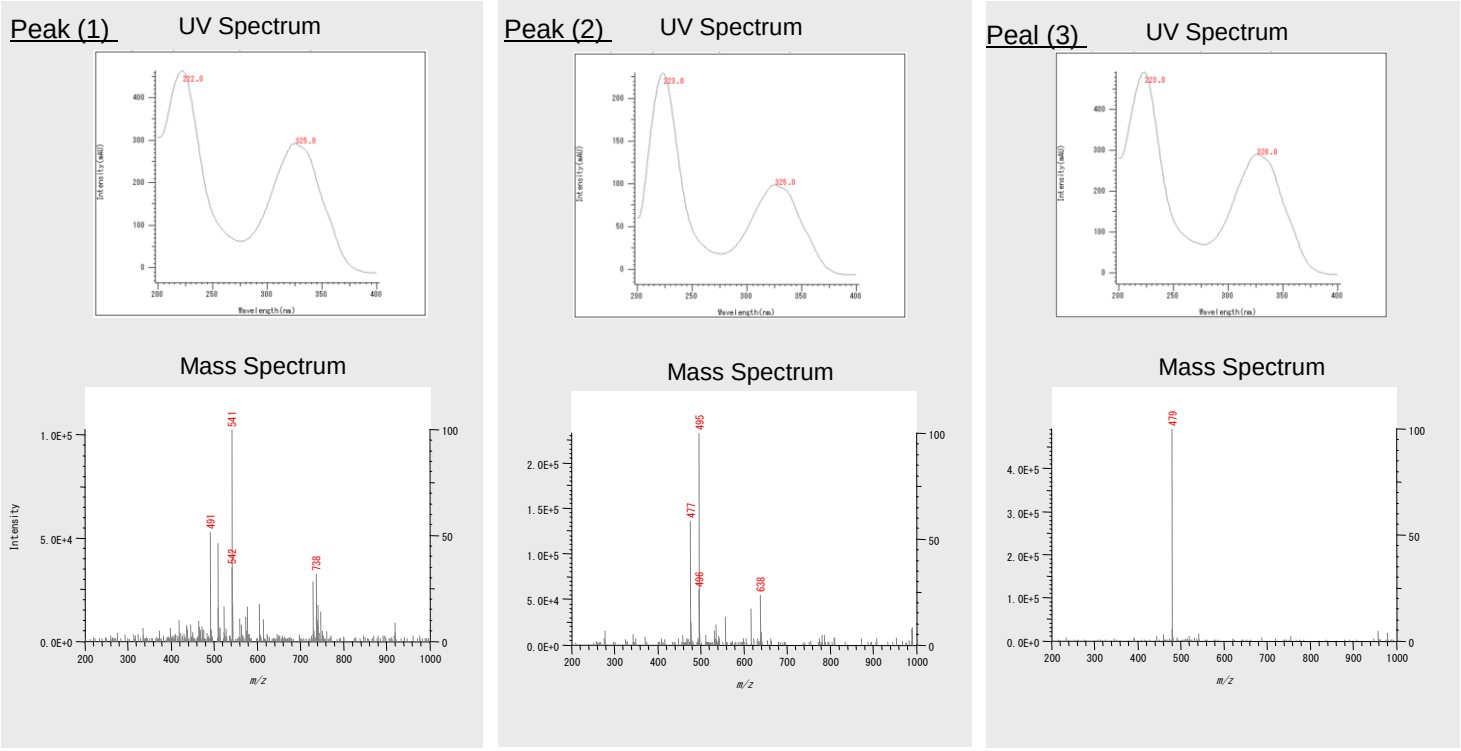


Figure 3 UV Spectra and Mass Spectra of Separated Components

From the data obtained by the DAD and MS detector, the UV spectra and mass spectra of the peaks (1), (2) and (3) were extracted. While the UV spectra are similar, the mass information was confirmed to be different for each of them. Both pieces of information are useful for the screening of active ingredients.

*The data introduced here were provided by Microrobial Chemistry Lab., Kitasato University School of Pharmacy.

<Main system configuration> Chromaster 5110 Pump, 5210 Autosampler, 5310 Column Oven, 5430DAD, 5610 MS Detector

NOTE: These data are an example of measurement; the individual values cannot be guaranteed.

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