IN SITU SECONDARY METABOLITES DETECTION FROM ENVIRONMENTAL MICROBIAL COLONIES BY RAMAN SPECTROSCOPY

Shunnosuke Suwa*[1,2], Masahiro Ando[2,3], Takuji Nakashima[3], and Haruko Takeyama†[1,2,3,4]

[1]School of Advanced Science and Engineering, Waseda University – Japan
[2]CBBD-OIL, AIST-Waseda University – Japan
[3]Research organization for Nano and Life Innovation, Waseda University – Japan
[4]Institute for Advanced Research of Biosystem Dynamics, Waseda Research Institute for Science and Engineering, Waseda University – Japan

Abstract

Environmental microbe produces varieties of secondary metabolites, the potential drug candidates for diseases. The screening has been performed all over the world, however, it takes massive culture and sample processing, only to find known compounds in most cases. We present a new analytical method for microbial secondary metabolites by using Raman spectroscopy, detecting secondary metabolites directly from colonies, with the aim of realizing a rapid and non-destructive screening(1). Previously, direct Raman measurement of microbial colonies was difficult owing to optical limitations. First, we improved the culture apparatus and spectrometer to maximize the Raman signal. Secondary, to evaluate metabolites production, we applied semi-supervised MCR-ALS to the recorded data using reference spectra of biomolecules. It aimed at detecting both known and unknown compounds simultaneously during cycle-fitting calculation to discover potential drug candidates. As a result, Raman spectra with high signal-to-noise ratio were recorded from *Escherichia coli* and actinomycetes colonies, exhibiting several biomolecules including secondary metabolites, with the measurement period of 1 minutes per colony at best. Spectral analysis successfully distinguished actinorhodin and undecylprodigiosin production by *Streptomyces coelicolor* A3(2) and amphotericin B from *S. nodosus* colonies, regardless of prior spectral information. Interestingly, constructed Raman images indicated heterogeneity in secondary metabolites production along with microbial differentiation. We have developed a new analytical method for microbe by means of improvement in culture method and spectral analysis. Our technique will significantly contribute to advance a rapid and effortless screening in the future.

(1) Suwa, Takeyama et al., *Analytical Chemistry* 96, 14909-17 (2024)

Keywords: secondary metabolites, microbial colony, spectroscopy, machine learning

*Speaker
†Corresponding author: haruko-takeyama@waseda.jp