## 〈一般論文〉

## 香料を対象とした抗菌活性の簡便測定法

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## Improved Methods for Estimation of Antimicrobial Activities of Volatile and Hydrophobic Fragrance Ingredients

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## Abstract

Antimicrobial activities of chemical and natural substances are generally estimated based on the standardized methods established by Clinical and Laboratory Standards Institute (CLSI). These methods are, however, not successfully applicable to fragrance ingredients because of their highly volatile and hydrophobic characteristics. In the present study, we developed methods for precise and convenient estimation of antimicrobial activities of the fragrance ingredients by modifying the standard CLSI methods. In the modified methods, surfactants were introduced to allow homologous dispersion of the fragrance ingredients in the test medium and wells of the testing plates were individually sealed by a plate-seal to avoid effects brought by evaporation of the test compounds. Antimicrobial analyses against 4 bacterial and 2 fungal strains of thyme red oil and cinnamic aldehyde as reference natural and chemical fragrance ingredients carried out by the modified methods revealed that the deviations of minimum inhibitory concentration (MIC) values from those obtained by the standard CLSI methods ranged from 2 to more than 8 fold depending on the fragrance ingredients or microbes tested. These differences might be caused by fine or less dispersion of the fragrance ingredients in the test medium and also by antimicrobial effects brought by vapor compounds from another wells. In contrast, the results obtained by the modified broth micro-dilution and agar-dilution methods are well coincided and reproducible. The high reproducibility of the modified broth micro-dilution method was further confirmed using additional 29 fragrance ingredients. Consequently, we propose that the modified methods established in the present study can be applicable as convenient standard methods for precise estimation of antimicrobial activities of fragrance ingredients. In addition, for a purpose of survey of antimicrobial activities on many kinds of samples, the modified micro broth-dilution method would be recommended because of its less time-consuming and paintaking workability.

Key words: fragrance ingredients, antimicrobial activities, improved standard method, CLSI protocol.