

## **STUDY ON THE APPLICATION OF MicroSEQ™ 500 16S rDNA PCR/SEQUENCING KIT TO MICROBIAL IDENTIFICATION AND THE EXPANSION OF PRIMERS AND DATABASE**

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It is a widely known marker, 16S rRNA, for microbial identification in the species level. MicroSEQ™ 500 16S rDNA PCR/Sequencing kit (abbr. MSID kit) makes it easier to do the laboratory work and the analysis process with their own developed primer mixtures targeting specific regions of 16S rRNA gene with the MicroSEQ ID (identification) 500bp library. This convenient kit, however, gives us sequences no longer than 500bp which is relatively short to specify a species. And the library has a limited amount of database as it includes about 2,100 strains of bacterial species. The company (Thermo Fisher Scientific corp.) has released the supplemental library containing about 7,100 strains of bacterial species, though. The results from MSID kit could simultaneously appear several species with 100% of concordance, or could not specify species with 100% of concordance. The reason of which could be the length of sequence is too short to specify species in same genus, or the similarity among species is too high even if the length of sequence is not too short. The purpose of this study is to establish the protocol for the microbial identification when the results using the MSID kit would not be satisfactory. We have carried out the series of processes for microbial identification using primers targeting 16S rRNA, not same with the primers in the MSID kit, and other libraries than the MicroSEQ ID 500bp library in the cases that the ambiguous results had been gotten in the species level using MSID kit. The poster shows a few results of specified bacterial strains in the case of the not specified microorganisms using MSID kit.