

Abstract

The purpose of the present invention is to provide an identification method capable of identifying a large number of bacteria, rather than a limited number of species, without changing the reagents or conditions for identification. Another purpose of the present invention is to provide an identification method that enables high-sensitivity detection and identification of bacteria with only one PCR, reducing the complexity of the operation and the burden on workers.

In the present invention, a method having the following steps is used to identify a bacterium. (1) A step of performing a reverse transcription reaction using a first reaction system consisting of a primer for reverse transcription to prepare a cDNA containing a base sequence for identification of a bacterium of interest, an RNA extracted from the bacterium in a sample, and an enzyme with RNA-dependent DNA polymerase activity to obtain a reaction mixture containing the synthesized cDNA and the primers for the reverse transcription, (2) A step of performing PCR using a second reaction system consisting of the reaction mixture, a primer pair for synthesis of a double-stranded DNA containing the base sequence for identification of the bacterium of interest, and an enzyme with DNA-dependent DNA polymerase activity, provided that in the second reaction system, the concentration of the primer for reverse transcription supplied from the reaction mixture is not less than 0.08nM and not more than 20nM, and (3) A step of detecting the generation of the double-stranded DNA containing the base sequence for identification of the bacterium of interest from the second reaction system after PCR.