

Abstract

An object of the present invention is to provide a method for enabling rapid and accurate determination of the number of bacterial cells in a specimen using a PCR method. To
5 attain the object, the species and number of a bacterium is determined by the following steps: (1) a first PCR step of carrying out a PCR method using a nucleic acid derived from a specimen as a template and a universal primer pair for amplifying a bacterial 16S rRNA gene to obtain a first amplification product; (2) a second PCR step of carrying out a nested PCR method using a primer pair(s) for amplifying an internal sequence(s) of the sequence of the first amplification
10 product obtained by the first PCR step to obtain a second amplification product; (3) a bacterial number determination step of obtaining the number of bacterial cells in the specimen based on the amount of the second amplification product obtained in the second PCR step and using calibration data. Also, to the steps (1) to (3), the following steps (4) and (5) may be added:

(4) a bacterial species identification step of identifying the species of a bacterium in the
15 specimen; and

(5) a bacterial number correction step of correcting the provisional number of bacterial cells obtained in the bacterial number determination step, based on the control bacterium and the 16S rRNA operon copy number of the bacterial species identified in the bacterial species identification step to determine the number of bacterial cells in the specimen.

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