Abstract

Purpose

Nosocomial infections caused by multidrug-resistant gram-negative bacilli are a global problem. Since the conventional investigation algorithm for detection of nosocomial outbreaks seems incapable of curbing the widespread of multidrug-resistant strains, the major purposes of the present project were to establish an effective real-time detection system, which was reinforced by modern computer technology and included a genetic and epidemiologic database and a rapid strain typing method.

Material and Methods

The multilocus sequence typing (MLST) method, a recently developed bacterial typing method, was developed for genotyping Pseudomonas aeruginosa in this year. The method is based on polymorphisms of structural genes among different strains of a bacterial species. Firstly, polymerase chain reaction assays were performed to amplify 14 structural genes of P. aeruginosa and nucleotide sequencing was followed to select the most polymorphic genes. A total of 20 P. aeruginosa isolates that were found to produce OXA-type beta-lactamases were tested by MLST and the results were compared with those obtained by the gold standard typing method, pulsed-field gel electrophoresis (PFGE). The results of MLST typing can be saved easily in a digitalized manner so that they can be retrieved very quickly. A genetic and epidemiologic database that included basic data of gram-negative bacilli isolated during the study period was established. A computer-assisted surveillance system for detecting clonal outbreaks of nosocomial infections was also established. We attempted to define an alert threshold for suspected outbreaks using this system.

Results

Among the 14 structural genes tested, seven genes were found to be most polymorphic; they were arcA, aroE, gdh, prlC, tal, gor, and dadx. After phylogenetic analysis, a number was given for each variant of a structural gene for each isolate, and a MLST type was given for each isolate with seven numbers together. The 20 P. aeruginosa isolates gave 13 PFGE types and 7 MLST types, respectively. A genetic and epidemiologic database was also established. Isolates that have the same antibiotype, the same MLST type, or the same sequence of a polymorphic gene within one period can be retrieved from the database. Based on the retrieved basic data of the bacterial isolates, the link among these isolates can be investigated. An algorithm defining an alert as two standard deviations above the mean monthly rate or number of isolates with a resistance phenotype

was applied to the computer-assisted surveillance system to determine alert thresholds for suspected outbreaks. The algorithm has been shown to detect clonal outbreaks of nosocomial infections in this study. Moreover, the system was shown to detect occult nosocomial outbreaks of infections that could not be detected by the standard infection control surveillance for the detection of clonal outbreaks.

Conclusions

A genetic and epidemiological database was established in this study. MLST was developed but the typing method was found to be less discriminatory than PFGE. Since MLST is easier to perform and to save in a database, the method may become an auxiliary typing tool to PFGE or a useful screening tying method. The computer-assisted surveillance system with the two standard deviations to determine alert thresholds can be used to detect the occurrences of clonal outbreaks of nosocomial infections. However, when the rate or number of isolates with a resistance phenotype from a ward is on the increase but is below the two standard deviations, infection control and prevention teams in hospitals should be informed for early detection of clonal outbreaks of nosocomial infections. More multidrug-resistant bacterial strains should be tested to intensify the functions of the system; further retrospective and prospective studies should be performed to evaluate the usefulness of the system. Furthermore, the system may be used in a nationwide surveillance program for monitoring multidrug-resistant bacterial strains.

Keywords：nosocomial outbreak；multilocus sequence typing；bioinformatics；genetic database