Is the Molecular Epidemiology Trustworthy in Hospital Infection Control?

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Staphylococcus aureus (S. aureus) is widely known as an important pathogen in the clinical setting. The dissemination of multi-drug-resistant organisms such as methicillin-resistant S. aureus (MRSA), and the recent emergence of vancomycin-resistant S. aureus (VRSA) strains are the growing concerns in hospitals worldwide (1, 2). The success of infection control in hospitals relies on the active involvement of the clinical microbiological laboratory using molecular technology. It is essential to identify how hospital pathogens such as MRSA are disseminated between the patients. Before the era of molecular typing, phenotypic typing techniques such as antibiogram, the phage type, had been widely used. However, phenotypic typing techniques have disadvantages such as the lack of recognition of unexpressed genes and relatively poor discriminatory power. This may cause under discrimination of clinical isolates.

Since molecular technology has developed dramatically, DNA fingerprinting has developed as a useful epidemiological tool to determine the relationships amongst clinical strains. Currently, three different typing techniques such as pulsed-field gel electrophoresis (PFGE), randomly amplified polymorphic DNA (RAPD), multilocus sequence typing (MLST) have become available in research and clinical fields. Each typing technique has a unique character (3).

PFGE has been widely used as a 'gold standard' for MRSA fingerprinting in hospital settings. After the whole chromosome is digested with rare cleaving restriction enzyme, the produced fragments are separated in the agarose gel under the periodic changed electric field. Although direct probing of recognition sequences by rare cutters detects variation in less than 0.01% of chromosome, large size rearrangements, such as sequence duplication, deletion, or insertion, will be readily detected as a shift in fragment size and number. PFGE has much higher discriminatory power than other methods, it is useful enough even in places where the genetic diversity of strains is narrow. If two strains are indistinguishable, it suggests that there must be a transmission of the strain between patients during hospital stay.

RAPD is a polymerase chain reaction (PCR) based typing techniques using a single arbitrary sequenced primer at low stringent temperature. The primer randomly anneals to multiple sequences with partial homology. After running in the acrylamide gel, the fragment polymorphism is analyzed. It has less discriminatory power than PFGE. This technique however, has a much higher throughput and is less labour intensive. It allows real time infection control in hospitals based on the RAPD result. The disadvantage of RAPD is the expensive initial cost of the equipment.

MLST provides a new approach to molecular epidemiology which can identify and track the global spread of drug-resistant strains. Seven different house-keeping enzymes which are genetically stable are sequenced and typed using an automated sequencer. The data is described seven digits which are electronically transmitted through the Internet. Despite of the high accuracy and stability of the data, MLST is not suited for outbreak investigation compared to geographical and longitudinal epidemiology, because of the lower discriminatory power.

Many nosocomial outbreaks have now been investigated using these tools. The interpretation of results is more important than methods used, when it is assessed whether strains are related and thus belong to the same chain of transmission. If a set of strains shows identical DNA fingerprints, it strongly suggests a close relation between them. A problem arises when fingerprints are similar but not identical. The criteria for PFGE proposed by Tenover et al have been established since 1995; as the number of genetic differences increase, the probability of epidemiological relatedness decreases (4).

Although the fingerprinting is useful to clarify epidemiological relatedness, it should be noted that the direction of nosocomial transmission is still difficult to determine merely with molecular results. Therefore, another aspect of analysis is necessary to resolve the puzzle of epidemiological relatedness.

While there are numerous reports of outbreaks in which the inanimate hospital environment may play a role in development of nosocomial infection, evidence was scant for a decrease in patient illness when general levels of bacteria were lowered. The value of routine microbiological sampling of the inanimate environment has been controversial. Maki et al carried out extensive sampling of a newly completed hospital building before and after it was commissioned (5). Although there was a significant difference in number of organisms on the surfaces in the hospital between before and after in use, the attack rate of nosocomial infection was not different. This study suggested that organisms in the inanimate envi-
The environment did not contribute to endemic nosocomial infection. The Committee on Infections within hospitals of the American Hospital Association had immediately recommended discontinuing routine microbiological sampling in 1974. In addition, it is commonly recognized that MRSA transmission mainly occurs by direct hand contact of health care workers, and transmission from the floor to the bedridden patients is therefore unlikely. It is important to assess whether the speculation is plausible to explain the phenomenon.

Finally, the geographic information regarding to the time and place shared with the possible source is also necessary (6), otherwise, molecular results may lead to a different conclusion.

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References