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Original Article

Performance of the BD GeneOhm Methicillin-resistant *Staphylococcus aureus* (MRSA) PCR Assay for Detecting MRSA Nasal Colonization in Taiwanese Adults

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BACKGROUND/PURPOSE: A rapid diagnostic method for methicillin-resistant *Staphylococcus aureus* (MRSA) has been implemented for surveillance of the at-risk population, but its performance in those without traditional risk factors is not clear. The objective of this study was to evaluate MRSA colonization status by comparing the performance of the BD GeneOhm MRSA polymerase chain reaction (PCR) assay with that of conventional culture during a 3-month active surveillance of Taiwanese adults in the community.

METHODS: From 1 October 2007 to 28 December 2007, adults (≥18 years old) attending a mandatory health examination arranged by their employers as a part of the workplace health promotion program at three medical centers in northern Taiwan were enrolled in the study. No healthcare workers were included. A total of 498 paired nasal swabs were prospectively obtained and used for both the BD GeneOhm MRSA PCR assay and conventional culture.

RESULTS: Of the 498 paired nasal swabs, 14 (2.8%) were positive for MRSA by conventional culture and 34 (6.8%) were positive by the BD GeneOhm MRSA PCR assay ($p < 0.005$). Thirteen specimens were both culture- and PCR-positive, and 463 samples were both culture- and PCR-negative. There were two discordant results: 21 specimens were culture-negative/PCR-positive, and one was culture-positive/PCR-negative. The simple kappa coefficient for measuring the agreement between conventional culture and the MRSA PCR assay was 0.52.

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CONCLUSION: This study demonstrates the feasibility of using both the MRSA PCR assay and conventional culture as surveillance tools. Also, the MRSA-positive rate detected by MRSA PCR assay was significantly higher than that of conventional culture.

KEYWORDS: methicillin-resistant *Staphylococcus aureus*, polymerase chain reaction, surveillance

Introduction

Methicillin-resistant *Staphylococcus aureus* (MRSA) has become one of the major nosocomial pathogens in Taiwan since the early 1980s.[1] MRSA infections not only lead to higher medical expenses and longer hospital stays, but also to higher mortality rates than infections caused by methicillin-susceptible *S. aureus*.[2,3] With the recent increase of MRSA infections, and especially the emergence of community-associated MRSA,[4–7] these infections are not limited to hospitalized patients, but are also seen in the broader community.[8,9] Considering the increasing trend for MRSA-related infections in the community, and the precept that MRSA colonization is an important risk factor for subsequent MRSA infection, it is important to evaluate the colonization status of MRSA in the community. Recent advances in and the implementation of rapid diagnosis and surveillance methods for MRSA have been timely for infection control in at-risk populations.[10–21] Previous studies have compared polymerase chain reaction (PCR) assays with selective MRSA culture media in different healthcare settings or at-risk populations in the community.[10–12] However, the performance of rapid diagnostic methods for MRSA surveillance outside the healthcare setting, or in a community population that is not traditionally at risk of MRSA colonization, is limited.[22–26] Thus, it is crucial to evaluate the performance of currently available diagnostic and screening tools for MRSA colonization in the community setting, rather than the more traditional inpatient setting. In Taiwan, previous studies of MRSA surveillance have mainly focused on pediatric populations,[27,28] and experiences of applying these rapid screening methods in adults or in a community population are limited. The objective of this study was to evaluate the MRSA colonization status of Taiwanese adults in the community by comparing the performance of the BD GeneOhm MRSA PCR assay with that of conventional culture.

Methods

Study population

From 1 October 2007 to 28 December 2007, adults (≥ 18 years old) attending a mandatory health examination arranged by their employers as a part of a workplace health promotion program at three medical centers in northern Taiwan were enrolled in the study. All participants gave written informed consent. None of these organizations or companies was healthcare institutions or long-term care facilities, so no healthcare workers were included in the study. The three medical centers were: Far Eastern Memorial Hospital (1,000 beds, Taipei County, Taiwan), Cathay General Hospital (700 beds, Taipei City, Taiwan), and Taipei Medical University-Wan Fang Hospital (700 beds, Taipei City, Taiwan). This study was approved by the Institutional Review Boards of these three hospitals (Internal Review Board approval serial number: CT9684).

Sample collection

Two nasal swabs were obtained from each volunteer. Each nasal swab was inserted into the bilateral anterior nares for collection. The first nasal swab was used for conventional MRSA culture and the second was used in the GeneOhm MRSA PCR assay [Beckton Dickinson (BD), San Diego, CA, USA]. There were no differences in terms of swab collection, specimen transportation or processing between these two methods. These swabs were transported to a central microbiology laboratory at National Taiwan University Hospital for processing.

MRSA identification and culture methods

Each nasal swab was plated directly onto Trypticase soy agar/5% sheep blood plate (TSA II, BD, Sparks, MD, USA). After overnight incubation, suspected *S. aureus* isolates were checked by catalase and Gram-staining if deemed necessary, and confirmed by coagulase latex agglutination assays.

ATCC 25923 *S. aureus* was used as a positive control and ATCC 14990 *S. epidermidis* as a negative control. Further culture and identification of MRSA was confirmed according to the methods outlined in the Clinical and Laboratory Standards Institute 2007 guidelines.[29] The identification of all nasal swabs processed by conventional culture was completed within 3 days of the initial plating.

Preparation of samples for PCR

The nasal swabs used for the PCR were placed in a buffer tube and vortexed for 1 minute. The cell lysate was transferred to a lysis tube and then centrifuged at 14,000–21,000 ×*g* for 5 minutes. The supernatant was discarded using a sterile fine-tip transfer pipette without touching the pellet. After adding fresh sample buffer the lysate was vortexed again for 5 minutes and spun down. The lysis tube was then heated to 95°C for 2 minutes, and then put on a cooling block. The PCR was performed using the GeneOhm MRSA test procedure. Analysis of all swabs processed using the BD GeneOhm MRSA assay was completed on the same day as the swab collection. BD GeneOhm MRSA PCR sample buffer was used as a negative control and sample buffer plus control DNA was used as a positive control for each run.

Statistical analysis

Differences in the MRSA nasal carriage rates between different hospitals and the two different assay methods were compared using the χ^2 test. The measure of agreement between the MRSA PCR assay and conventional culture was determined by a simple kappa coefficient test. Statistical analyses were performed using SAS version 9.1 (SAS Institute Inc., Cary, NC, USA).

Results

Between 1 October 2007 and 28 December 2007, 498 paired nasal swabs were collected from community adult volunteers. The median age was 40 years (range, 18–80 years), and 45.0% were male (224/498). Seventy-six paired nasal swabs came from Far Eastern Memorial Hospital (15.3%), 127 from Cathay General Hospital (25.5%) and 295 from Taipei Medical University-Wan Fang Hospital (59.2%).

Table 1. The methicillin-resistant *Staphylococcus aureus* nasal carriage rate at three hospitals[a]

Hospital	Culture positive	PCR positive
FEMH (n=76)	4 (5.3)	4 (5.3)
CGH (n=127)	1 (0.8)	7 (5.5)
TMU-WFH (n=295)	9 (3.1)	23 (7.8)
Total (n=498)	14 (2.8)	34 (6.8)

[a]Data presented as *n* (%). FEMH=Far Eastern Memorial Hospital; CGH=Cathay General Hospital; TMU-WFH=Taipei medical university-Wan Fang Hospital; PCR=polymerase chain reaction.

Table 2. Results from BD GeneOhm methicillin-resistant *Staphylococcus aureus* polymerase chain reaction assay and conventional culture

	PCR negative	PCR positive	Total
Culture negative	463	21	484
Culture positive	1	13	14
Total	464	34	498

PCR=polymerase chain reaction.

The overall *S. aureus* carriage rate was 19.5% (97/498) in the study population (by conventional culture). Of these *S. aureus* colonizers, 14.4% (14/97) were MRSA colonizers (by conventional culture). The overall MRSA nasal carriage rate was 2.8% (14/498) as assessed by conventional culture and 6.8% (34/498) by the BD GeneOhm MRSA PCR assay. The MRSA colonization rates in the three different hospitals assessed by culture and PCR are shown in Table 1. There was no inter-hospital difference in MRSA carriage rates by conventional culture (p=0.18) or BD GeneOhm MRSA PCR assay (p=0.62). However, the overall MRSA nasal carriage rate was significantly higher for the BD GeneOhm MRSA PCR assay than for conventional culture (p<0.005).

The results from conventional culture and the MRSA PCR assay are shown in Table 2. The two methods did produce some discordant results, including 21 PCR-positive/culture-negative specimens and one PCR-negative/culture-positive specimen. The overall agreement between the BD GeneOhm MRSA PCR assay and the culture method was 95.6% (476/498). The measure of agreement between the two methods using the simple kappa coefficient was 0.52, which is "moderate".

Table 3. Summary of studies that compare BD GeneOhm methicillin-resistant *Staphylococcus aureus* polymerase chain reaction to selective or non-selective culture for methicillin-resistant *Staphylococcus aureus* surveillance

Study	No. of case (*n*)	Population	Sensitivity (%)	Specificity (%)	PPV (%)	NPV (%)	Reference
Present study	498	Health screen volunteer	92.9	95.7	38.2	99.8	
Farely et al	602	City jail prisoner	88.5	91.0	62.6	97.9	10
Paule et al	403	University hospital patients	97.9–98.2	95.2–97.7	74.6–87.3	99.7	12
Boyce & Havill	286	Patients from MICU, SICU, Heme/Onc	100	98.6	95.8	100	11

PPV=positive predictive value; NPV=negative predictive value; MICU=medical intensive care unit; SICU=surgical intensive care unit; Heme/Onc=hematology and oncology.

In this study, the sensitivity, specificity, positive predictive value (PPV) and negative predictive value of the BD GeneOhm MRSA PCR assay were 92.9%, 95.7%, 38.2% and 99.8%, respectively (Table 3), using the results obtained by conventional culture as a reference standard.

Discussion

Of the current methods for MRSA surveillance, conventional culture is still one of the most recommended methods, along with the recently developed chromogenic agar method and PCR.[30] This study was designed to evaluate the MRSA colonization status of community adults in Taiwan using the BD GeneOhm MRSA PCR assay and a conventional culture method. Results show that the MRSA nasal carriage rate was 2.8% and 6.8% as assessed by conventional culture and MRSA PCR assay, respectively. Compared with previous studies, this prevalence rate was lower than that seen in children in Taiwan (13.2%).[28] The finding of a lower MRSA nasal carriage rate in community adults than in children may be explained by the fact that children have unique risk factors for MRSA colonization in the community that are related to their behavior (e.g. nose picking) and to their interaction with the environment (daycare centers, schools or recreational facilities).

The BD GeneOhm MRSA PCR assay gave a significantly higher positive-MRSA nasal carriage rate than conventional culture (6.8% *vs.* 2.8%) and a lower PPV (38.2%). This finding may be attributed to two major reasons: the prevalence rate of MRSA colonization in the study population and the reference method that we chose. Previous studies on the surveillance of MRSA colonization, which compared the BD GeneOhm MRSA PCR assay with selective MRSA culture, showed PPVs ranging from 62.6%[10] to 95.8%[11] (Table 3). The PPV was much lower in our study. Unlike previous studies that were targeted at traditional at-risk populations for MRSA colonization such as city jail prisoners,[10] university hospital patients,[12] and intensive care unit and hematology/oncology patients,[11] our study specifically focused on community adults, which is not a population traditionally at risk for MRSA. The different populations in the previous studies had different levels of exposure to MRSA that possibly imply various underlying prevalence rates of MRSA colonization that might further affect the PPV of these diagnostic tools. In addition, our study used a conventional culture method as the reference standard, unlike previous studies, which used MRSA chromogenic agar. The main reason why we chose the conventional culture instead of MRSA chromogenic agar as the reference method is that the conventional culture method is still commonly used in daily clinical practice for MRSA surveillance, and it is inexpensive.[30] We were also interested in evaluating the overall carriage rate of *S. aureus* (not only MRSA) in community adults, and this would be affected by selecting chromogenic agar as a reference method. Therefore, this might explain the lower PPV of the BD GeneOhm MRSA PCR assay found in this study. The MRSA PCR assay also had a high negative predictive value (99.8%), which is similar to the findings of other studies. This shows that the MRSA PCR assay served as a useful tool for surveillance and successfully identified those who were not colonized with MRSA.

In this study, there were some discordant results between the MRSA PCR assay and conventional culture. There were 21 PCR-positive/culture-negative paired nasal swabs. This might be due to the higher sensitivity of the PCR.

For one paired nasal swab the PCR result was negative for MRSA, but the culture result was positive. This might be due to a sampling problem or to errors when managing the specimen. However, for these discordant pairs, all PCR were repeated and confirmed. There was also a difference in the turn-around time between the BD GeneOhm MRSA PCR assay and conventional culture; the PCR gave results within 1 day, whereas results from conventional culture took 3 days.

There are some limitations to this study. First, only nasal swabs were used for surveillance instead of multiple sites (e.g. groin, sputum). Second, the order in which the swabs were tested (first nasal swab by conventional culture and the second nasal by the BD GeneOhm MRSA PCR assay) may have affected the PCR results. Third, the study population (adults attending a mandatory health examination arranged by their employers) might not fully represent the general population in the community. All volunteers were working adults; therefore, pediatric, non-working and unemployed populations were not included. Finally, there was no thorough secondary analysis to confirm the discordant results due to the difficulty in recalling participants and obtaining another nasal swab for analysis.

In conclusion, this study demonstrates the feasibility of using the BD GeneOhm MRSA PCR assay and conventional culture as surveillance tools for community adults. The results show that the BD GeneOhm MRSA PCR assay produces a significantly higher MRSA-positive rate than conventional culture for community adults who are not traditionally at high risk for MRSA colonization.

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