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Lim KT, Hanifah YA, Mohd Yusof MY, Ito T, Thong KL.

Microbiology Division, Institute of Biological Science, Faculty of Science, University of Malaya, 50603 Kuala Lumpur, Malaysia.

**Abstract**

**BACKGROUND:** Methicillin-resistant *Staphylococcus aureus* (MRSA) is one of the main bacterial pathogens responsible for a variety of nosocomial infections worldwide. The purpose of this study was to determine changes in antimicrobial resistance profiles and genotypes of MRSA strains isolated in 2003 and 2008 in a Malaysian tertiary hospital.

**METHODS:** Antibigrams of 163 MRSA strains were determined by disk diffusion test. The genotypes of MRSA strains were determined by pulsed-field gel electrophoresis, MLST, SCCmec and spa typing.

**RESULTS:** All the strains were resistant to oxacillin only but sensitive to vancomycin. Majority of the MRSA strains in 2003 and 2008 remained sensitive to linezolid (99%), teicoplanin (99%), mupirocin (97%) and rifampicin (96%). In contrast, most of them were resistant to gentamicin (85%), ciprofloxacin (91%), clindamycin (95%) and erythromycin (96%). Although three SCCmec types were observed in this study, both SCCmec type IV and SCCmec type V were only detected in 2008 strains. PFGE, MLST and spa typing subtyped the strains into 63 pulsotypes, 9 MLST types and 15 spa types, respectively.

**CONCLUSION:** Besides vancomycin, four other antimicrobial agents, including linezolid, teicoplanin, mupirocin and rifampicin are active agents 'in vitro' against MRSA infections in this tertiary hospital. All three different DNA fingerprinting methods indicate that MRSA strains from this local hospital were genetically related and correlation between DNA profiles (PFGE, spa and MLST types) and resistotypes was observed. The presence of more spa and MLST types in 2008 strains indicates that MRSA strains isolated in later years were slightly more diverse when compared to MRSA strains from earlier year.

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**Supplement:**

Methicillin-resistant *Staphylococcus aureus* (MRSA) is an important nosocomial pathogen and is a big concern in hospital settings because of the limited choice of antimicrobial agents available for treatment of life-threatening cases. In this study, we would like to determine potential changes in antimicrobial resistance profiles and the genetic association between MRSA strains from a teaching hospital in 2003 and 2008.

Disc diffusion tests were performed on all the MRSA strains. Genotypic characterization by using PFGE and sequence typing were performed following published work (1-2). Statistical software (STATISTICA, Version 8.0, StatSoft, Inc, Tulsa, OK, USA) was used for data analysis. Fisher exact test was used for the comparison of certain variables whereas Spearman's rank correlation coefficient test was used for the determination of associations among antimicrobial agents.

This study shows the antimicrobial resistance trends of MRSA strains from the teaching hospital in 2003 and 2008 (Fig. 1). We found that MRSA strains remained susceptible to vancomycin. This information is relevant and important as vancomycin is one of the few remaining antibiotics left for the treatment of MRSA infections (3). However, the use of vancomycin for treatment of MRSA should be treated with caution as cases of vancomycin treatment failure in vancomycin-susceptible MRSA strains have been reported in Malaysia (4).

High sensitivity of MRSA strains towards linezolid (99%), teicoplanin (99%), mupirocin (97%) and rifampicin (96%) could be due to limited usage of these few drugs in this tertiary hospital. In vitro testing showed that linezolid and teicoplanin are highly effective against Malaysian MRSA infections (5). The resistance rates of MRSA strains in the year 2003 and year 2008 are summarized in Figure 1.

Based on Spearman's rank correlation coefficient test, the correlation between erythromycin and ciprofloxacin resistance (R = 0.607, P < 0.05) was observed. Similarly, correlation between erythromycin and tetracycline (R = 0.1922, P < 0.05) and ciprofloxacin and tetracycline (R = 0.0795, P < 0.05) were also observed. These values indicate that tetracycline-resistant strains were most likely to show co-resistance towards ciprofloxacin and erythromycin. This is a cause of concern because choices of antimicrobial agents for treatment of life-threatening cases will be limited as usage of tetracycline, ciprofloxacin and erythromycin is still common in Malaysian hospitals for treatment of respiratory tract and other nosocomial infections (6).

Three SCCmec types were observed in this study with SCCmec type III being predominant (90%), followed by SCCmec type IV (9%) and SCCmec type V (1%). Both SCCmec type IV and SCCmec type V were only present in MRSA strains isolated from 2008. All SCCmec type IV strains are hospital acquired MRSA, while the only SCCmec type V strain is community-acquired MRSA (4).
PFGE, MLST and spa typing subtyped the strains into 63 PFGE types and 9 MLST types and 15 spa types, respectively. Among the 15 spa types, three spa types (i.e. 1037, 11544 and 1421) were detected in 2003 whereas the remaining 12 spa types were found in 2008 strains. On the other hand, two MLST types, ST920 and ST229 were detected in year 2003 while the remaining seven MLST types were found in 2008 strains. This suggests that the MRSA strains have diversified.

Most of the strains that were clonally related by PFGE (more than 80% similarity) shared very similar resistotypes. For example, strains with Pulsotype-9 to Pulsotype-14 and spa-MLST types, t037-ST239 and t1593-ST241 were resistant to erythromycin, gentamicin and ciprofloxacin (1).

SCCmec type IV strains in this study belonged to spa-MLST types, t304-ST6, t032-ST22, t14184-ST22, t1037-ST22, t1107-ST1178, t1378-ST22 whereas the only SCCmec type V strain belonged to t1657-ST772. In addition, we also found that eleven SCCmec IV strains which were grouped together by PFGE were closely related by spa typing and MLST, these were from spa-MLST types t032-ST22, t1378-ST22 and t14184-ST22. This again shows the close association between the MRSA strains isolated from different time periods from this tertiary hospital. Furthermore, this also indicates that PFGE is a useful subtyping tool for differentiating SCCmec type IV strains.

A total of 35 MRSA strains which were indistinguishable by PFGE were differentiated into five different combined spa-SCCmec-MLST types (t037-III-ST239, t1421-III-ST239, t1452-III-ST239, t1450-III-ST239, t032-III-ST22). This indicates that combined spa-SCCmec-MLST typing was more discriminatory than PFGE alone in subtyping MRSA strains.

In conclusion, five antimicrobial agents, including vancomycin, linezolid, teicoplanin, mupirocin and rifampicin are active agents ‘in-vitro’ against MRSA infections in this tertiary hospital. All three different DNA fingerprinting methods indicate that MRSA strains from this local hospital were genetically related and correlation between DNA profiles (pulsotypes, spa and MLST types) and resistotypes was observed. The presence of more spa and MLST types in 2008 strains indicates that MRSA strains isolated in later years were slightly more diverse when compared to MRSA strains from earlier year.

Reference


Resistance rate of Malaysian MRSA strains in year 2003 and 2008

![Resistance rate of Malaysian MRSA strains in year 2003 and 2008](image)

Figure 1. Resistance rates of Malaysian MRSA strains in year 2003 and 2008 from a tertiary hospital

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Contact:
Prof Dr Kwai-Lin Thong

Microbiology Unit, Institute of Biological Sciences, Faculty of Science, University of Malaya, 50603, Kuala Lumpur, Malaysia.

Email: thongkl@um.edu.my.