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Evaluation of different methods for the rapid diagnosis of methicillin-resistance in *Staphylococcus aureus*

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Since its first identification in the early 1960s, Methicillin-Resistant *Staphylococcus aureus* (MRSA) has been recognized as a major human pathogen. The aim of this study was to identify the prevalence of MRSA in Alexandria Main University Hospital and to settle on a simple, rapid, accurate and cost-effective phenotypic test for the detection of MRSA from clinical specimens. One hundred *S. aureus* isolates, including 71 MRSA isolates, as confirmed by PCR for the presence or absence of the mecA gene as the gold standard, were isolated from patients from different departments at Alexandria Main University Hospital over a six month-period. They were tested for methicillin resistance by comparing five phenotypic tests (Mannitol salt agar-cefoxitin [MSA-FOX] , oxacillin disc diffusion, cefoxitin disc diffusion, oxacillin MIC by broth microdilution and latex agglutination for PBP2a) to the gold standard genotypic test (detection of mecA gene by PCR). It was found that both oxacillin disc diffusion and latex agglutination showed 100% sensitivity, negative and positive predictive values of 100 and 97.3%, respectively. Both were found to be highly sensitive phenotypic tests for the detection of MRSA. However, the oxacillin disc diffusion test is much more cost-effective. The MSA-FOX, whose sensitivity was 95.8%, was found to be a highly sensitive, cost-effective screening medium for the detection of MRSA.

Key words: *Staphylococcus aureus*, MRSA, methicillin resistance, oxacillin resistance.

INTRODUCTION

Staphylococcus aureus has long been recognized as a major human pathogen responsible for a wide range of infections, from mild skin infections to wound infections and bacteraemia. Although the introduction of antibiotics has lowered the mortality rate from *S. aureus* infections, the bacteria have developed resistance mechanisms to all antimicrobial agents that have been produced (Hardy et al., 2004).

In 1960, the year methicillin was developed, the first isolation of methicillin-resistant *S. aureus* (MRSA) was reported (Jevons, 1961). MRSA is a specific strain of the *S. aureus* bacterium, which is intrinsically insensitive to methicillin and all -lactams. Later use of oxacillin as an alternative to methicillin in susceptibility tests resulted in the term 'oxacillin-resistant *S. aureus'* (ORSA). These designations are used interchangeably in the literature and are synonymous (Brown et al., 2005). Since 1961, successive waves of epidemic MRSA have spread throughout hospitals and other chronic healthcare facilities worldwide, to the extent that it is now the most commonly isolated antimicrobial-resistant pathogen in many countries (Diekema et al., 2004; Goosens 2004). Over the last few years, reports have documented an increase in community- acquired MRSA (CA-MRSA) in patients with or without risk factors for MRSA infection, which may suggest a changing epidemiology (Chambers, 2001).

The mechanism of methicillin resistance is an altered penicillin-binding protein (PBP2a) in MRSA that markedly reduces affinity for all available -lactam antibiotics, while maintaining effective cell wall-building activity (Hartman and Tomasz, 1981). PBP2a is encoded by the mecA gene that is carried on a mobile DNA element, the staphylococcal cassette chromosome mec (SCCmec) (Katayama et al., 2000).

Timely detection of MRSA is still problematic with the majority of techniques taking longer than 48 h to produce

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a result (Hardy et al., 2004). The accurate rapid diagnosis of MRSA in microbiology laboratories is vital for patients' management. It is also essential for meaningful interpretation of surveillance data. Currently, surveillance data for MRSA is difficult to interpret because there is no uniform testing method for the detection of MRSA, and laboratories vary in their standard operating procedures and interpretation of breakpoint values (Krishanan et al., 2002).

Accurate routine phenotypic detection of MRSA is difficult using standard microbiological methods such as disc diffusion, minimal inhibitory concentration (MIC) determination, agar screening and methods that detect PBP2a. This has been ascribed to the heterogeneous methicillin resistance in most strains of MRSA where only a few cells within the total population of cells express resistance, while the majority of cells do not (Hartman and Tomasz, 1984; Hackbarth and Chambers, 1989; Chambers, 1997). No phenotypic test is completely reliable for detection of MRSA (Skulnick et al., 1992). Therefore, detection of the mecA gene is considered the gold standard (Chambers, 1997). Perhaps, the best phenotypic approach would be to have several methods available, and use an alternate test when resistance is suspected but not detected by the routine method (Swenson et al., 2001).

Although the vast majority of infections caused by *S. aureus* result in asymptomatic carriage, this species nevertheless represents a serious public health burden, particularly in the hospital setting, where clones resistant to methicillin and other classes of antibiotics are endemic, and insensitivity to vancomycin is on the increase (Feil et al., 2003).

The objectives of the present study were to detect the prevalence of MRSA in clinical specimens delivered to the laboratory of the Microbiology Department at Alexandria Main University Hospital, to evaluate the different phenotypic methods for the detection of MRSA with the aim of settling on a rapid, sensitive, specific, cost-effective and easily applicable method for MRSA detection in the routine Microbiology laboratory, and to compare the phenotypic methods with a molecular diagnostic assay (PCR for mecA gene) for confirmation.

MATERIALS AND METHODS

Specimen collection and processing

The materials of this study included 2,900 clinical specimens delivered to the routine laboratory of the Microbiology Department at Alexandria Main University Hospital (AMUH), over a six month-period, starting from the first of June 2006 till the end of November 2006. The specimens included swabs from surgical wounds, bed sores, pus, pleural and ascitic fluid, endotracheal and bronchial aspirates, sputum, urine, cerebrospinal fluid, as well as blood samples for blood culture. Only one clinical specimen was obtained from each patient. Each clinical specimen was inoculated onto both Columbia blood agar (Oxoid) and MacConkey's agar plates (Oxoid) and incubated at 37°C for 24 - 48 h aerobically. For the blood culture, 10 ml of the blood sample was inoculated into the blood culture bottle (Egyptian Diagnostic), which was subcultured on the same above-mentioned media after 24 and 48 h before discarding (usually seven days). After plating the specimens on the aforementioned media, they were also suspended in 5 ml phosphate buffered saline (Oxoid), then 1 l of the resulting suspension was inoculated onto MSA-FOX mannitol salt agar (Biolife) supplemented with 4 mg/l cefoxitin (Sigma-Aldrich, Germany). One l of the urine samples was directly inoculated on MSA- FOX; also 1 l from the blood culture bottles was subcultured on MSA-FOX after 24, 48 h and seven days. Incubation of MSA-FOX was done for 48 h at 35°C (Perry et al., 2004; Smyth and Khlmeter, 2005; Stoakes et al., 2006).

All suspected *S. aureus* colonies on Columbia blood agar, and suspected mannitol-fermenting yellow colonies grown on MSA-FOX (MRSA) and *S. aureus* colonies that grew on blood agar and did not show growth on MSA-FOX, were identified on the following bases: Gram-stained film, positive catalase test, positive tube coagulase test and mannitol fermentation (Baird, 1996). Also, yellow colonies other than *S. aureus* grown on MSA-FOX were identified and included in the study. All other isolated strains (non-staphylococcal) were presumptively identified based on colonial morphology and the results of standard routine laboratory tests (Baird, 1996). Fully identified *S. aureus* colonies on Columbia blood agar and yellow colonies on MSA-FOX, which were confirmed to be *S. aureus*, were subjected to the following tests:

Oxacillin disc susceptibility testing; was performed according to CLSI recommendations using a 1 g oxacillin disc.

Cefoxitin disc susceptibility testing: This was performed according to the CLSI recommendations using a 30 µg cefoxitin disc (Oxoid). Two control strains were used for quality control of the disc diffusion test, that is, a standard Methicillin sensitive *S. aureus* (MSSA) strain (ATCC 25923), and a MRSA strain (ATCC 43300).

Oxacillin MICs. This was determined by microdilution with Muëller–Hinton broth supplemented with 2% NaCl, following the CLSI criteria. A standard MSSA strain (ATCC 29213) was used for quality control.

Latex agglutination. The mecA product (PBP2a) was detected using the Slidex MRSA kit (Biomerieux, Marcy L'Etoile, France). The method was used according to the manufacturer's instructions. A standard mecA-negative *S. aureus* strain (ATCC 25923), as well as a mecA-positive strain (ATCC 43300) were used for quality control.

Detection of the mecA gene by PCR; DNA extraction from bacteria was performed by the method described by Schmitz et al. (1998). The supernatant was used as a template in the PCR reaction. PCR reaction was performed in a 25 l reaction volume. The oligonucleotide primers (Geno-Mechanix) were designed to amplify the 310-base pairs (bp) fragment of the mecA gene (Vannuffel et al., 1995). The primer sequences were as follows: forward primer (5'- TGG CTA TCG TGT CAC AAT CG -3') and reverse primer (5'- CTG GAA CTT GTT GAG CAG AG-3'). The conditions of the amplification reaction were: an initial denaturation step at 95°C for 5 min, 35 amplification cycles of denaturation at 95°C for 30 s, annealing at 56°C for 30 s, extension at 72°C for 30 s, followed by a final cycle of extension at 72°C for 3 min.

Antibiotic susceptibility testing

Antibiotic susceptibility testing was evaluated by using penicillin (10 units), amoxicillin-clavulanic acid (20/10 g), cephradine (30 g), ceftriaxone (30 g), cefepime (30 g), imipinem (10 g), gentamycin (10 g), erythromycin (15 g), tetracycline (30 g), ciprofloxacin (5 g), clindamycin (2 g), trimethoprim- sulfamethoxazole (1.25/23.75 g), chloramphenicol (30 g), rifampin (5 g), fusidic acid (10 g), and vancomycin (30 g) (Oxoid). Zone diameters were measured fol-

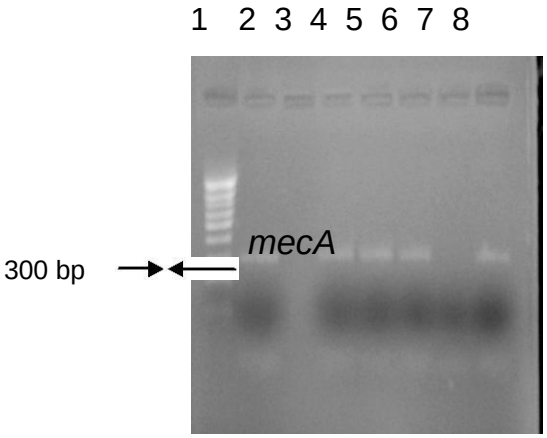

Figure 1. Agarose gel stained with ethidium bromide of few MRSA isolates. A single band of amplified PCR product of mecA gene of 310 bp at lanes 2, 4, 5 and 6. Lane 1; 100 bp DNA ladder; lane 7, negative control; lane 8, positive control.

lowing the National Committee for Clinical Laboratory Standards (NCCLS) criteria (Katayama et al., 2000). Multiresistance was defined as an isolate with resistance to three or more drug classes other than -lactam antibiotics.

Statistics

Data were fed to SPSS/Win. Analysis of data was done using count, percentage, sensitivity, specificity, positive predictive value and negative predictive value.

RESULTS

Culture-positive specimens (confirmed nosocomial pathogens) constituted 63.2% (1,833 specimens) out of the total 2,900 clinical specimens. Staphylococcus species were isolated from 203 clinical specimens, representing 11.1% (103 coagulase-negative staphylococci [CONS] representing 5.6% and 100 *S. aureus* representing 5.5%), and non-staphylococcal organisms from 1,630 clinical specimens, representing 88.9%. Among the 2,900 clinical specimens, 827 specimens showed no growth, and 240 grew mixed normal flora. *S. aureus* was mostly isolated from pus specimens (34%) and rarely from pleu-ral fluid (2%). The highest percentage of isolates was from Intensive Care Units (ICUs) (34%).

The 2,900 clinical specimens were tested on MSA-FOX. A total of 125 specimens allowed the growth of yel-low colonies on the medium when incubated at 35°C for 48 h. Among the 125 yellow colonies, 75 (60%) were found to be *S. aureus*, 4 (3.2%) were CONS and 46 (36.8%) were non-staphylococcal colonies (27 Klebsiella spp.,10 Enterococcus spp. and 9 Diphtheroids).

Out of 100 *S. aureus* isolates included in the study, 71 were found to be mecA-positive (MRSA) and 29 were found to be mecA-negative (MSSA) by PCR. Presence or absence of the mecA gene was considered the reference method for detection of MRSA (Figure 1). Accordingly, among the confirmed 1 833 nosocomial pathogens, 3.9% were found to be MRSA.

The results of all phenotypic tests in comparison to mecA gene detection by PCR (the gold standard) revealed that the oxacillin disc diffusion test, as well as the latex agglutination test identified all mecA-positive isolates. No phenotypic test identified all mecA-negative isolates (Table 1).

According to the confirmed results of PCR, 71 isolates were mecA-positive and 29 isolates were mecA-negative. MSA-FOX detected 66 MRSA isolates after a 24 h incubation (93% sensitivity) . It detected two additional MRSA isolates after extending the incubation to 48 h, raising the sensitivity to 95.8%. Three MRSA isolates failed to grow on MSA-FOX and were confirmed to be MRSA when isolated from conventional blood agar. As regards MSSA, MSA-FOX inhibited the growth of all but seven MSSA isolates. The specificity of the test medium was 75.9% at 24 and 48 h. After a 48 h incubation, the positive predicttive value (PPV) and the negative predictive value (NPV) were 90.7% and 88%, respectively.

As regards the oxacillin disc diffusion test, the diameter of zone of inhibition was measured as follows: Susceptible (13 mm), Intermediate (11 - 12 mm) and Resistant (13 mm). Any growth within the zone of inhibition was an indication of oxacillin resistance.

All 71 mecA-positive isolates were correctly identified as oxacillin resistant by oxacillin disc diffusion, resulting in 100% sensitivity. The oxacillin disc diffusion test identified 27 mecA-negative isolates as oxacillin susceptible, resulting in 93.1% specificity. Therefore, the test had a PPV of 97.3% and a NPV of 100%.

In the cefoxitin disc susceptibility tests, the interpretation of inhibition zone diameters was as follows: Resistant (19 mm), Sensitive (20 mm). It detected oxacillin resistance correctly in all mecA-positive isolates, except one isolate, which gave a zone diameter of 35 mm, resulting in a sensitivity of 98.6%. As regards the mecA-negative isolates, the cefoxitin disc diffusion test identified eight of the isolates as oxacillin resistant. This resulted in 72.4% specificity. The test had a PPV of 89.7% and a NPV of 95.5%.

As regards the broth microdilution, equivalent MIC breakpoints for *S. aureus* were Resistant (4 g/ml), Sensitive (2 g/ml) . Out of 29 mecA negative isolates, 28 isolates had an oxacillin MIC of 2 µg/ml. This provides the test with 96.6% specificity. Out of the 71 mecA- positive isolates, 66 isolates had an oxacillin MIC of 4 µg/ml. The remaining five isolates had an oxacillin MIC of 2 µg/ml. The sensitivity of the test in relation to PCR was 93%. The test had a PPV of 98.5% and a NPV of 84.8%.

Considering PCR as a gold standard test for MRSA detection, the latex agglutination test demonstrated 100% sensitivity, 93.1% specificity, 97.3% PPV and 100% NPV. Out of the 71 mecA-positive isolates, 63 isolates were

Table 1. Results of all phenotypic tests in comparison to PCR for the 100 *S. aureus* isolates.

Phenotypic Tests	mecA +ve (n = 71)		mecA -ve (n = 29)		Sensitivity (%)	Specificity (%)	PPV[d]	NPV[e]
	True +ve	False -ve	True -ve	False +ve				
MSA-FOX	68	3	22	7	95.8	75.9	90.7	88
OX DD[a]	71	0	27	2	100	93.1	97.3	100
FOX DD[b]	70	1	21	8	98.6	72.4	89.7	95.5
OX MIC[c]	66	5	28	1	93	96.6	98.5	84.8
Latex	71	0	27	2	100	93.1	97.3	100

a, OX DD: oxacillin disc diffusion; b, FOX DD: cefoxitin disc diffusion; c, OX MIC: oxacillin MIC by broth microdilution method; d, PPV: positive predictive value; e, NPV: negative predictive value.

Table 2. Antimicrobial susceptibility testing results of the 71 MRSA isolates.

Antibiotic	MRSA (n = 71) Resistance	
	No.	%
Penicillin	71	100
Amoxicillin- Clavulanic acid	55	77.5
Cephradine	64	90.2
Ceftriaxone	66	93
Cefepime	69	97.2
Imipinem	43	60.6
Erythromycin	38	53.5
Gentamycin	55	77.5
Tetracycline	56	78.8
Clindamycin	17	24
Chloramphenicol	17	24
Rifampin	13	18.3
Ciprofloxacin	53	74.6
Trimethoprim-Sulfamethoxazole	25	35.2
Fusidic acid	17	24
Vancomycin	0	0

oxacillin resistant in all phenotypic tests used in the study, while eight mecA-positive isolates showed discre-pant results in one or more phenotypic tests. Three iso-lates did not grow on MSA-FOX (after 48 h), four were sensitive to oxacillin in the broth microdilution test (their MIC values < 2 g/ml), and one had an oxacillin MIC of 2 g/ml and 35 mm zone diameter with the cefoxitin disc. On the other hand, out of the 29 mecA-negative isolates, 17 isolates were oxacillin sensitive in all tests, while 12 isolates showed discrepant results. Four grew on MSA-FOX after 24 h of incubation, five isolates were resistant to the cefoxitin disc (zone diameter < 15 mm), one grew on MSA-FOX and was also resistant to the cefoxitin disc (with a zone diameter of 10 mm), one isolate was resistant to oxacillin in all phenotypic tests and one isolate was oxacillin resistant in all tests except for a sensitive MIC (2 g/ml).

Concerning the distribution of MRSA among the differ-rent clinical specimens, sputum was the most frequent specimen from which MRSA was isolated (28 isolates representing 39.4%), 19 isolates (26.8%) were isolated from pus, 12 isolates (16.9%) from blood, eight isolates (11.3%) from wound swabs, three isolates (4.2%) from bed sore swabs and only one isolate (1.4%) from pleural fluid.

Regarding the hospital sites from which MRSA was detected, the ICU was the most common site from which MRSA was isolated (28 isolates representing 39.4%) followed by the Emergency Unit where nine isolates (12.7%) were isolated. Six isolates (8.5%) were isolated from each of the following departments, Surgery, Internal Medicine, and Dermatology, four isolates (5.6%) from Chest, three isolates (4.2%) from each of Cardiothoracic Surgery, and Oncology, two isolates (2.8%) from the Gangrene Unit and one isolate (1.4%) from each of the following departments, Neurology, Cardiology, Urosur-gery and the Burn Unit.

As regards the antibiotic susceptibility to 16 antibiotics (other than oxacillin and cefoxitin), all MRSA isolates were sensitive to vancomycin and resistant to penicillin. The resistance to -lactam drugs varied; cefepime (97.2%), ceftriaxone (93%), cephradine (90.2%), amoxi-cillin-clavulanic acid (77.5%) and imipinem (60.6%). The resistance to non- lactam drugs ranged from 78.8% for tetracycline to 18.3% for rifampin. Multi-drug resistant MRSA accounted for 83% of MRSA isolates (Table 2).

DISCUSSION

MRSA is probably the most challenging bacterial patho-gen that currently affects patients in hospital and in the community (Rayner, 2003). Methicillin resistance renders *S. aureus* resistant to all - lactam antibiotics, the most important group of antibiotics in the treatment of staphylo-coccal infections. Accurate and rapid detection of methi-cillin resistance in staphylococci is therefore important, not only for choosing appropriate antibiotic therapy for the

individual patient, but also for control of the endemicity of MRSA (Skov et al., 2003). Although multiple methods of detection of methicillin resistance have been developed, identification of the mecA gene is the most reliable reference method of detecting MRSA isolates (Velasco et al., 2005). This study was carried out aiming to settle on a rapid, reliable, cost-effective and easily applicable method for MRSA detection in the routine Microbiology laboratory.

S. aureus isolates in our study constituted 5.5% (100 out of 2 900 isolates) of nosocomial pathogens. This low percentage of *S. aureus* as compared to the study of Savas et al. (2005), where *S. aureus* was isolated from 39.6% of 871 various clinical specimens, and the study of El-Farrash et al. (2003), where 370 *S. aureus* isolates (17.2%) were identified, could be attributed to the large number of randomly collected specimens from different departments of the hospital with no selection of high risk patients.

Among the 100 *S. aureus* isolates identified in the present study, 71 (71%) were found to be MRSA constituting 3.9% of nosocomial pathogens, in accordance with the study conducted by APIC (Association for Professionals in Infection and Epidemiology) (2007) on the prevalence of MRSA in U.S. healthcare facilities that showed that 3.4% of patients were infected with MRSA and that it accounts for as many as 50 - 70% of the *S. aureus* infections acquired in healthcare facilities.

On the other hand, the prevalence of MRSA among *S. aureus* in the present study was higher than that reported by Akpaka et al. (2006) 12.8%, Ontengco et al. (2004) 18% and El-Farrash et al. (2003) 24.6%. The high prevalence of MRSA in the present study may reflect the inappropriate use of antibiotics, as well as a compromise in infection control policies.

It is also evident that the prevalence of MRSA among *S. aureus* isolates in our hospital has increased from 32% in 1989, as stated by Zaki et al. (1989), to 71% as report-ed in the present study. This probably signifies the emergence of MRSA as a common nosocomial pathogen in AMUH. This finding was consistent with data from the National Nosocomial Infections Surveilance NNIS System (USA), according to which the prevalence
of MRSA among hospitalized patients increased from 31.9% in 1996 to 60.7% in 2004.

In the present study, MSA supplemented with 4 g/ml cefoxitin (MSA-FOX) was evaluated as a screening medium for the detection of MRSA directly from clinical speci-mens. Failure of the three MRSA isolates to grow on MSA-FOX may be due to sensitivity to the selective agents in the medium base and/or the low number of organisms present in the inoculum. On the other hand, the growth of the seven MSSA isolates on MSA- FOX may be contributed to degradation of cefoxitin in the medium.

Smyth and Kahlmeter (2005) also stated that mannitol salt cefoxitin medium was superior to screening media containing oxacillin because the majority of the strains tested produced visible colonies after 18 h of incubation and the shelf-life of the medium is 30 days at a temperature of 2 - 8°C. In contrast, selective media containing oxacillin deteriorate after one week.

In this study, the 1 g oxacillin disc diffusion test correctly identified all the 71 mecA-positive (MRSA) and 27 mecA-negative (MSSA) strains in accordance with the study conducted by Fawzi et al. (2007), where the oxacillin disc diffusion test showed 100% sensitivity and 95.6% specificity.

Regarding cefoxitin disc diffusion, several groups of investigators have reported that the results of cefoxitin disc diffusion tests correlate better with the presence of mecA than do the results of disc diffusion tests using oxacillin (Felten et al., 2002; Boutiba et al., 2004; Cauwelier et al., 2004 and Swenson and and Tenover , 2005).

In the present study, the 30 µg cefoxitin disc diffusion identified 70 out of 71 MRSA isolates. The same finding was reported by Kircher et al. (2004), who found that one MRSA isolate out of 203 tested MRSA strains produced a zone diameter of 24 mm. This isolate also had an MIC to oxacillin of 2 g/ml.

In the present study, the broth microdilution test detected 66 out of 71 MRSA isolates and 28 out of 29 MSSA isolates. Failure of the broth microdilution method to detect five MRSA isolates may be attributed to the heterogeneous resistance of MRSA

The latex agglutination test in the present study was easy to perform, gave results rapidly and was amenable to the processing of a large number of samples. The use of a larger inoculum or induction by a -lactam was not needed. This is in agreement with other studies that have evaluated the latex agglutination test, which showed sensitivity without -lactam induction ranging from 93.5 to 100% and specificity ranging from 96.9 to 100% (Nakatomi and Sugiyama, 1998; Van Griethaysen et al., 1999; Cavassini et al., 1999; Sakoulas et al., 2001).

In the present study, in a PCR reaction, 71 mecA-positive strains and 29 mecA-negative strains were identified. However, two mecA-negative strains were confirmed to be MRSA by most phenotypic tests. This finding may be due to point mutation or deletion in the mecA gene, or other mechanisms of oxacillin resistance such as hyperproduction of -lactamase, which was not assessed in our study, or modified PBPs. Our results were in accor-dance with that of Warren et al. (2004), who found three mecA-negative isolates phenotypically resistant when grown on oxacillin screen agar, suggesting that methicillin resistance in these isolates was mediated by methods other than PBP2a, such as hyperproduction of -lactamase or modified PBPs.

The antibiogram of MRSA isolates in the present study was in agreement with the studies of Tai et al. (2006) and Anbumani et al. (2006). However, the resistance to gentamycin in this study was higher than that reported in the studies of Zaki et al. (1989) and El-Hefnawy (1993). Inte-

restingly, among the tested -lactam drugs in our study, cefepime and ceftriaxone gave the best indication of oxacillin resistance when both intermediate and resistant zone diameters were considered as clues for oxacillin resistance, as recommended by CLSI.

In conclusion, no single phenotypic test is completely reliable for the detection of oxacillin resistance in *S. aureus*. A cost- effective option in most laboratories would be to adopt a well standardized phenotypic technique with stringent quality control measures for day-to-day testing, and to retest ambiguous results with a second conventional phenotypic method. It was found that MSA-FOX is a good, cost-effective screening medium for the detection of MRSA from clinical specimens, as well as MRSA carriage in healthcare personnel after 24 h in a single step. Also, the oxacillin disc diffusion test and the latex agglutination test are the best tests for identification of MRSA. Isolates that give inconsistent results with two different conventional tests could then be tested with the latex agglutination test and sent to a reference laboratory for mecA detection. Vancomycin is still the drug of choice for treatment of MRSA; however, regular monitoring of vancomycin sensitivity should be carried out.

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