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Evaluation of six agglutination tests for *Staphylococcus aureus* identification depending upon local prevalence of meticillin-resistant *S. aureus* (MRSA)

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Most routine laboratory detection of *Staphylococcus aureus* isolates is based on rapid agglutination test systems. Failure of agglutination assays to identify meticillin-resistant *S. aureus* strains (MRSA) has been demonstrated. The aim of this study was to evaluate six commercially available agglutination tests for the detection of meticillin-sensitive *S. aureus* (MSSA) and *mecA*-positive MRSA strains. The Dry Spot Staphytect Plus® test (Oxoid), the Pastorex Staph Plus® test (Bio-Rad), the Slidex Staph-Kit® and Slidex Staph Plus® test (bioMérieux), the Staphaurex Plus® test (Remel) and the Staphylase Test® (Oxoid) were used. As determined by pulsed field gel electrophoresis, 52 distinct MRSA strains from five countries, 83 MSSA strains and 150 coagulase-negative staphylococci were included. Species identification and determination of susceptibility patterns were performed using colony morphology, Gram stain, catalase testing, tube coagulase testing, DNase testing, mannitol fermentation, susceptibility testing towards oxacillin by Etest®, coagulase gene PCR, fibrinogen receptor gene PCR and PCR of the *mecA* gene. Sensitivity of the agglutination tests ranged from 82·7 to 100·0 % for MRSA strains and 92·8 to 100·0 % for MSSA strains, respectively. Specificity of the test systems ranged from 91·3 to 99·1 %. None of the six agglutination assays produced correct reactions for all staphylococci tested. Only the Dry Spot Staphytect Plus® test correctly identified all 52 MRSA strains. For the other tests kits, sensitivity of MRSA detection was lower than for MSSA isolates. Depending upon the local MRSA prevalence and the parameter of interest (sensitivity or specificity), these test systems may be useful for routine diagnostic purposes.

INTRODUCTION

Unlike coagulase-negative staphylococci (CNS), *Staphylococcus aureus* strains are able to secrete free plasma coagulase, which is an important virulence factor for these bacteria. *S. aureus* is a common pathogen in nosocomial infections, so exact identification of *S. aureus* isolates is essential for microbiology laboratories (Emori & Gaynes, 1993). The proportion of clinical *S. aureus* isolates that are resistant to meticillin (MRSA) has increased over recent years (Tiemersma *et al.*, 2004). Compared with meticillin-sensitive *S. aureus* (MSSA), infections by MRSA strains are associated with increased morbidity and mortality in affected patients (Cosgrove *et al.*, 2003).

Today, the gold standard for *S. aureus* identification is proof of free plasma coagulase in the tube coagulase test (Bannerman, 2003). However, confirmation of *S. aureus* by this method may take as long as 24 h. Rapid *S. aureus* agglutination tests have been developed as an alternative for routine diagnosis (Essers & Radebold, 1980). In these test systems, particles precipitate with one or multiple *S. aureus* surface antigens, and allow *S. aureus* and CNS isolates to be distinguished within a few seconds. Bacteria identified as *S. aureus* by these tests are then further checked for meticillin resistance. Unfortunately, the accuracy of these test systems is limited. In particular, MRSA may be easily misclassified

Abbreviations: CNS, coagulase-negative staphylococci; MRSA, meticillin-resistant *S. aureus*; MSSA, meticillin-sensitive *S. aureus*; NPV, negative predictive value; PFGE, pulsed-field gel electrophoresis; PPV, positive predictive value.

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in agglutination tests because of false-negative results (Aldridge *et al.*, 1984; Brakstad *et al.*, 1993; Croize *et al.*, 1993; Fournier *et al.*, 1989; Lairscey & Buck, 1987; Piper *et al.*, 1988; Ruane *et al.*, 1986; Winblad & Ericson, 1973), which might be due to changes in various surface components, such as capsular polysaccharides, the clumping factor or protein A (Fournier *et al.*, 1987, 1989; Kuusela *et al.*, 1994). However, correct identification of MRSA is essential for appropriate treatment strategies and sufficient infection control measures for the prevention of nosocomial infections (Muto *et al.*, 2003).

The aim of this study was to assess the sensitivity, specificity and positive and negative predictive values (PPV and NPV) of six commercially available *S. aureus* agglutination assays for clinical MSSA and MRSA isolates.

METHODS

Source of bacterial strains. The MSSA strains tested were clinical isolates collected from patients, staff and environmental swabs from different wards in six hospitals in Berlin. MRSA strains came from five different countries (Germany, Switzerland, Argentina, Belgium and Canada). In addition, strains ATCC 43300 (MRSA) and ATCC 6538 (MSSA) were used as controls. CNS strains were obtained from two microbiology laboratories serving seven different hospitals in Berlin. If duplicate strains had been provided, only one isolate per person was included in the MSSA, the MRSA or the CNS group.

Genus determination for *S. aureus*. MSSA and MRSA strains were included in this study if all of the following four criteria had been fulfilled: (i) typical morphology of colonies (Bannerman, 2003) on 5 % Columbia sheep blood agar, (ii) Gram stain showing Gram-positive cocci in clumps, (iii) at least one positive reaction in the tube coagulase test evaluated after 2, 4 and 24 h (bioMérieux) or detection of the coagulase gene encoding free coagulase by genotyping using PCR and (iv) detection of the fibrinogen receptor gene encoding the clumping factor by PCR. As it is known that there are DNase-negative (Menzies, 1977) or catalase-negative (Tu & Palutke, 1976) *S. aureus* strains, and some *S. aureus* strains fail to grow on mannitol salt agar (Kampf *et al.*, 1997), we did not exclude strains with these characteristics.

Genus and species determination of CNS. Strains were designated as CNS if all of the following five criteria were fulfilled: (i) typical morphology of colonies, (ii) positive Gram stain, (iii) negative tube coagulase test, (iv) proof of the absence of the coagulase gene by PCR and (v) proof of the absence of the fibrinogen receptor gene by PCR. Further species determination was performed using API 32 STAPH. If no exact species could be assigned by this test system, catalase-positive strains were included as 'non-*S. aureus* strains' in the CNS group. As DNase-positive CNS strains are documented (Menzies, 1977), as well as growth of CNS on mannitol salt agar (Bannerman, 2003), we did not exclude strains with these characteristics.

Determination of meticillin resistance. After species determination, all *S. aureus* isolates were differentiated by susceptibility towards oxacillin via Etest®. For confirmation of the antibiotic susceptibility phenotype, all *S. aureus* isolates were then genotyped by PCR. A PCR for detection of the *mecA* gene that encodes the penicillin-binding protein-2 was performed, as it is considered to be the 'gold standard' for identification of meticillin resistance in staphylococci (Wallet *et al.*, 1996).

PCR detection of coagulase, fibrinogen receptor and *mecA* genes. Assays for the *mecA* and fibrinogen receptor genes were performed simultaneously using a multiplex PCR. DNA extraction for all PCR systems was performed using Easy DNA® (Invitrogen) according to the manufacturer's instructions. Template DNA was stored at 4 °C if PCR was started within 24 h or at −20 °C for longer time periods until use. Coagulase gene identification was performed by a nested PCR system published previously (Goh *et al.*, 1992) and modified by Schwarzkopf (1995). In brief, four to eight repetitive sequences (81 bp each) of the 3′-region of the coagulase gene were amplified in a thermocycler, using primers COAG2 and COAG3 as described (Goh *et al.*, 1992). This PCR leads to corresponding DNA fragments with a length of 654 to 978 bp. Multiplex PCR for the fibrinogen receptor gene and the *mecA* gene was performed using primers Sa-fibrec1 (5′-AGAAGTAAAGAAGCAGATG-CAAGTG-3′), Sa-fibrec2 (5′-CTAATTCCTCCGCATTTGTATTGC-TTG-3′), mecA1 and mecA2 as described by Murakami *et al.* (1991). The fibrinogen receptor gene consists of 320 bp and the *mecA* gene of 533 bp. Amplification parameters in this multiplex PCR were chosen as described for *mecA* PCR (Murakami *et al.*, 1991). Until further processing in 2 % agarose gel electrophoresis, all PCR products were stored at 4 °C. PCR products were detected by ethidium bromide stain.

DNA macrorestriction analysis for determination of MRSA clonal variability. MSSA and CNS are known to show great genetic diversity (Struelens *et al.*, 1992; Valentine *et al.*, 1988). In contrast, diversity of MRSA strains is limited, which might be due to a clonal MRSA origin (Kreiswirth *et al.*, 1993). Pulsed-field gel electrophoresis (PFGE) has proven to be the most discriminative method for MRSA (Saulnier *et al.*, 1993; Struelens *et al.*, 1992). Contour-clamped homogeneous electric field electrophoresis was performed as described (Witte & Grimm, 1992) based on a previously published method (Chu *et al.*, 1986). Bacteria were grown on sheep blood agar and then prepared according to Maslow *et al.* (1993); *Sma*I was used as the cutting enzyme (Tenover *et al.*, 1995). PFGE parameters were chosen according to Bannerman *et al.* (1995). A lambda DNA ladder was used in all PFGE gels for standardization. Analysis of PFGE results was carried out visually as well as by a computer-assisted method (GelCompar; Applied Maths). MRSA isolates that had been obtained from different continents (North America, South America and Europe) were considered epidemiologically unrelated if they showed at least four differences in electrophoresis band patterns. For strains from the same continent (Belgium, Germany and Switzerland), criteria for distinct strains were applied as proposed by Bannerman *et al.* (1995) and Tenover *et al.* (1995).

Panel of latex agglutination assays. Six commercially available *S. aureus* agglutination tests kits were evaluated: Dry Spot Staphytect Plus® (Oxoid), Pastorex Staph Plus® (Bio-Rad), Slidex Staph-Kit® and Slidex Staph Plus® (bioMérieux), Staphaurex Plus (Remel) and Staphylase Test® (Oxoid). Further details of these test systems are described in Table 1. Test kits were used according to manufacturers' instructions. Autoagglutination reactions in the negative control were excluded from calculations.

Statistical analysis. Assuming a P value $\leqslant 0{\cdot}05$ for statistical significance, it was calculated that 115 isolates in the *S. aureus* and the CNS group each were enough to assess a 4 % precision as required. Statistical analysis of results was performed using 2×2 tables (Epi Info 6). The McNemar χ^2 test was used to assess significant differences between each agglutination test kit and the results of free plasma coagulase testing in the tube coagulase test.

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Table 1. *S. aureus* agglutination test systems

Test kit	Agglutinating particles	Detection of *S. aureus* surface antigens			Species in which false-positive results may occur according to manufacturer
		Clumping factor	Protein A	Other	
Dry Spot Staphytect Plus®	Latex particles	Yes	Yes	Staphylococcal capsular polysaccharides type 5 and 8	*S. intermedius*, *S. lugdunensis*, *S. schleiferi*, *S. hyicus*, *S. saprophyticus*
Pastorex Staph Plus®	Latex particles	Yes	Yes	Staphylococcal capsular polysaccharides type 5 and 8	*S. intermedius*, *S. lugdunensis*, *S. schleiferi*, *S. hyicus*
Slidex Staph-Kit®	Latex particles and erythrocytes	Yes	Yes	Glycopolysaccharide antigen 18	*S. intermedius*, *S. lugdunensis*, *S. schleiferi*
Slidex Staph Plus®	Latex particles	Yes	Yes	Glycopolysaccharide antigen 18	*S. intermedius*, *S. lugdunensis*, *S. schleiferi*, *S. hyicus*, *S. delphini*
Staphaurex Plus®	Latex particles	Yes	Yes	Staphylococcal capsular polysaccharides type 5 and 8	*S. intermedius*, *S. lugdunensis*, *S. schleiferi*, *S. hyicus*, *S. saprophyticus*
Staphylase Test®	Erythrocytes	Yes	No	None	*S. sciuri*, clumping-factor-negative *S. aureus*

RESULTS AND DISCUSSION

Characteristics of panel of strains

A total of 295 strains were investigated in this study using phenotype and genotype screening. Eight staphylococcal strains were excluded according to our criteria. The phenotypic and genotypic characteristics of the remaining 287 strains (137 *S. aureus* and 150 CNS) are shown in Table 2. PCR of the *mecA* gene confirmed the identification of all 83 MSSA and 54 MRSA strains as previously indicated by Etest® (data not shown). In addition, 86 of the 150 CNS also tested *mecA*-positive (data not shown). Visual PFGE analysis of all MRSA isolates was more discriminative than computer-assisted analysis. In two of the 54 MRSA strains, different PFGE patterns were observed in repeat PFGE and these strains were therefore excluded from further use (data not shown). The remaining 52 MRSA strains had been obtained from Canada ($n=19$), Germany ($n=18$), Belgium ($n=5$), Switzerland ($n=5$) and Argentina ($n=5$). All of these MRSA strains were distinct by PFGE. As determined by API 32 STAPH®, strains in the CNS group consisted of *Staphylococcus epidermidis* ($n=90$), *S. hominis* ($n=17$), *S. haemolyticus* ($n=13$), *S. capitis* ($n=12$), *S. warneri* ($n=5$), *S. lugdunensis* ($n=4$), *S. saprophyticus* ($n=1$), *S. intermedius* ($n=1$), *S. simulans* ($n=1$) and *S. cohnii* ($n=1$). Five strains in the CNS group could not be identified reliably to species level; these strains were then designated as 'non-*S. aureus*'.

Sensitivity and specificity of test kits

Finally, 83 MSSA, 52 MRSA and 150 CNS were used for the evaluation of the different agglutination tests in this study. All strains were tested with every test kit except the Slidex Staph Plus® test kit. Thirty-four CNS isolates could not be tested with the Slidex Staph Plus® test because the manufacturer had ended production of this kit during the study period for unknown reasons. Table 3 shows the results of agglutination reactions of the test kits used. Only the Dry Spot Staphytect Plus® test identified all MSSA and MRSA isolates correctly. For three test kits (Slidex Staph-Kit®, Staphaurex Plus® and Staphylase Test®), false-positive reactions were observed in the negative control due to autoagglutination. In 23 of 35 false-positive reactions, the species involved had not been mentioned in information from the corresponding manufacturer (Table 1). CNS species that led to false-positive reactions were *S. epidermidis* ($n=13$), *S. lugdunensis* ($n=12$), *S. hominis* ($n=6$), *S. haemolyticus* ($n=2$), *S. capitis* ($n=1$) and *S. warneri* ($n=1$). Autoagglutination reactions were excluded for calculation of sensitivity and specificity of agglutination test kits for MSSA and MRSA detection as shown in Table 3. As mentioned above the Dry Spot Staphytect Plus® test showed 100 % sensitivity for MSSA and MRSA strains. All other test kits showed a lower sensitivity for MRSA than MSSA. Significant differences for the agglutination tests compared with the tube coagulase test were observed for the Dry Spot Staphytect Plus® test, the Slidex Staph-Kit® test and the Staphylase Test® kit.

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Table 2. Genotype and phenotype patterns of strains investigated ($n=287$)

Group	Strains (n)	Genotyping		Phenotyping			
		Fibrinogen receptor gene	Coagulase gene	Tube coagulase test	DNase	Catalase	Growth on mannitol salt agar
S. aureus ($n=137$)	120	+	+	+	+	+	+
	3	+	−	+	+	+	+
	8	+	+	−	+	+	+
	1	+	+	−	+	+	−
	1	+	+	+	+	−	+
	4	+	+	+	+	+	−
CNS ($n=150$)	106	−	−	−	−	−	−
	9	−	−	−	+	+	−
	16	−	−	−	−	+	+
	17	−	−	−	+	+	+
	2	−	−	−	−	−	−

PPV and NPV of test kits

The PPV and NPV of test systems depend upon the prevalence of the factor tested. There are great differences in MRSA prevalence in different countries. According to the National Nosocomial Infections Surveillance system in the USA, the proportion of nosocomial *S. aureus* among all staphylococci varies depending upon the site of infection (e.g. surgical site infections, 58 %; pneumonia, 91 %) (Emori & Gaynes, 1993). Therefore, given a mean *S. aureus* prevalence of 80 %, we calculated PPV and NPV for laboratories with a large (50·0 %), medium (25·0 %) or small (2·0 %) proportion of MRSA among *S. aureus* isolates (Table 4).

Routine microbiology laboratories need to perform rapid and accurate pathogen identification. For this purpose, several *S. aureus* agglutination test systems have been developed and are widely used. In most routine laboratories, MRSA detection is based on prior *S. aureus* species determination. Further rapid differentiation between MSSA and MRSA may then be performed by MRSA-specific agglutination test systems (Cavassini *et al.*, 1999). Reliable *S. aureus* species determination is therefore essential as a first step. Evaluation of *S. aureus* agglutination test kits has to take into account the phenomenon that species misclassification is more likely to occur for meticillin-resistant strains (Aldridge *et al.*, 1984; Brakstad *et al.*, 1993; Croize *et al.*, 1993; Fournier *et al.*, 1989; Lairscey & Buck, 1987; Piper *et al.*, 1988; Ruane *et al.*, 1986; Winblad & Ericson, 1973). MRSA prevalence has increased worldwide and great variation in MRSA prevalence is seen in different countries (Tiemersma *et al.*, 2004), so the rate of non-detectable MRSA isolates in laboratories influences the PPV and NPV of *S. aureus* agglutination tests. This is very important, because MRSA-positive patients, in particular, require special infection-control measures according to official guidelines in order to prevent inter-patient spread (Muto *et al.*, 2003). The aims of our study included evaluation of the sensitivity of MRSA detection by a variety of agglutination test systems. The test kits used in this study have been tested for this purpose before.

Wichelhaus *et al.* (1999) calculated sensitivity for MRSA agglutination of 99·4 % using PFGE-typed MRSA strains in the Dry Spot Staphytect Plus® test kit. Macrorestriction profiles revealed 90 different genotypes among the 181 MRSA strains that they used. Their results are confirmed by our findings. The data presented in this study showed an MRSA sensitivity of 100·0 % in 52 non-identical MRSA isolates.

The Pastorex Staph Plus® test identified 96·2 % of MRSA correctly in our panel of strains. Several other studies have evaluated MRSA sensitivity of this test kit, and all these studies found a sensitivity of 95·1 % or above (Fournier *et al.*, 1993; Tveten, 1995; van Griethuysen *et al.*, 2001; Wichelhaus *et al.*, 1999). Three other studies (Davies, 1997; Luijendijk *et al.*, 1996; Personne *et al.*, 1997) documented 100·0 % MRSA sensitivity in 40, 78 and 144 MRSA strains, respectively. However, the latter studies did not perform prior MRSA genotyping so the use of duplicate strains cannot be excluded.

MRSA sensitivity of the Slidex Staph® kit from our data was 97·9 %. This result is concordant with strains PFGE-typed by others (Wichelhaus *et al.*, 1999) giving a sensitivity of 97·2 %. Results of other MRSA evaluations of this test system ranged from 95·4 to 100·0 % (Croize *et al.*, 1993; Davies, 1997; Wilkerson *et al.*, 1997). Personne *et al.* (1997) reported an MRSA sensitivity of no more than 93·0 % for this test kit. Although only epidemiologically unrelated MRSA strains had been used, their panel of strains were not typed for verification of actual genetic diversity. All published reports have in common that MSSA sensitivity was shown to be greater than sensitivity for MRSA detection.

The second generation of this test series, the Slidex Staph Plus®, was also evaluated. In contrast to others, who observed an increased MRSA sensitivity compared to the first-generation test kit (100·0 % vs 93·0 %) (Personne *et al.*, 1997), in our strain panel, sensitivity actually decreased slightly for MRSA (92·3 vs 97·9 %) and MSSA (97·6 vs

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Table 3. Results of agglutination test kits

Criterion	Dry Spot Staphytect Plus®	Pastorex Staph Plus®	Slidex Staph-Kit®	Slidex Staph Plus®*	Staphaurex Plus®	Staphylase Test®
MSSA (*n*=83)						
Positive	83	82	80	81	83	77
False-negative	0	1	1	2	0	6
Autoagglutination	0	0	2	0	0	0
Sensitivity (%) (95 % CI)	100·0 (94·5–100·0)	98·8 (92·5–99·9)	98·8 (92·4–99·9)	97·6 (90·8–99·6)	100·0 (94·5–100·0)	92·8 (84·4–97·0)
MRSA (*n*=52)						
Positive	52	50	46	48	46	43
False-negative	0	2	1	4	6	9
Autoagglutination	0	0	5	0	0	0
Sensitivity (%) (95 % CI)	100·0 (91·4–100·0)	96·2 (85·7–99·3)	97·9 (87·3–99·9)	92·3 (80·6–97·5)	88·5 (75·9–95·2)	82·7 (69·2–91·3)
All *S. aureus* (*n*=135)						
Positive	135	132	126	129	129	120
False-negative	0	3	2	6	6	15
Autoagglutination	0	0	7	0	0	0
Sensitivity (%) (95 % CI)	100·0 (96·6–100)	97·8 (93·1–99·4)	98·4 (93·9–99·7)	95·6 (90·2–98·2)	95·6 (90·2–98·2)	88·9 (82·0–93·4)
CNS (*n*=150)						
False-positive	13	3	13	1	3	2
Negative	137	147	136	115	146	147
Autoagglutination	0	0	1	0	1	1
Specificity (%) (95 % CI)	91·3 (85·3–95·1)	98·0 (93·8–99·5)	91·3 (85·2–95·1)	99·1 (94·6–100·0)	98·0 (93·8–99·5)	98·7 (94·7–99·8)
Comparison with gold standard (*P* value)	<0·01	0·71	0·01	0·16	0·53	<0·01

*Only 116 CNS isolates were tested in the Slidex Staph Plus® kit.

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Table 4. PPV and NPV for *S. aureus* identification depending upon MRSA prevalence

Total *S. aureus* prevalence was 80 %.

Test kit	Specificity (%)	2 % MRSA/98 % MSSA			25 % MRSA/75 % MSSA			50 % MRSA/50 % MSSA		
		Total *S. aureus* sensitivity (%)	PPV (%)	NPV (%)	Total *S. aureus* sensitivity (%)	PPV (%)	NPV (%)	Total *S. aureus* sensitivity (%)	PPV (%)	NPV (%)
Dry Spot Staphytect Plus®	91·3	100·0	97·9	100·0	100·0	97·9	100·0	100·0	97·9	100·0
Pastorex Staph Plus®	98·0	98·7	99·5	95·0	98·2	99·5	93·2	97·5	99·5	90·7
Slidex Staph-Kit®	91·3	98·8	97·8	95·0	98·6	97·8	94·2	98·4	97·8	93·4
Slidex Staph Plus®	99·1	99·4	99·8	97·6	96·3	99·8	87·0	95·0	99·8	83·2
Staphaurex Plus®	98·0	99·8	99·5	99·2	97·1	99·5	89·4	94·3	99·5	81·1
Staphylase Test®	98·7	92·6	99·7	76·8	90·3	99·6	71·8	87·8	99·6	66·9

98·8 %). The different results may in part be explained by the origin of MRSA strains that were tested. In our study, an MRSA panel from five different countries was used, while all strains tested by Personne *et al.* (1997) were obtained from the French Reference Centre for Staphylococci. Van Griethuysen *et al.* (2001) determined geographically stratified MRSA sensitivities of this test kit in strains from Switzerland, France and the Netherlands; they found sensitivities of 95·9, 97·3 and 98·0 %, respectively. Gupta *et al.* (1998) also evaluated the accuracy of the Slidex Staph Plus® test kit. They used 158 'unique clinical MRSA isolates' and calculated an MRSA sensitivity of 98·1 % by this approach. However, a clonal relationship of some isolates cannot be excluded, since no further typing was performed. To our knowledge, no other comparable MRSA sensitivity evaluation of this test system has been published in which only non-identical MRSA strains, as determined by genotyping, have been used.

The Staphaurex Plus® test kit revealed an 88·5 % MRSA sensitivity in our work, but all MSSA strains had been identified correctly as *S. aureus*. The range of MRSA as determined by others is 97·0 to 100·0 % (Gupta *et al.*, 1998; Luijendijk *et al.*, 1996; Personne *et al.*, 1997; Summers *et al.*, 1998; van Griethuysen *et al.*, 2001; Wichelhaus *et al.*, 1999; Wilkerson *et al.*, 1997). There was also a wide range in terms of specificity in the Staphaurex Plus® test. Compared with our data (98·0 %) and that of Summers *et al.* (1998) (97·9 %), lower specificities were calculated in all other studies. The lowest specificity was 72·7 % (Personne *et al.*, 1997). Again, one may speculate that the selection of the bacterial panel has had a strong impact on the variation of results.

Besides the present study, only one other publication has reported on the sensitivity of the Staphylase Test® kit for MRSA identification as *S. aureus* species (Davies, 1997). In contrast to our results (43 of 52 strains), all MRSA isolates ($n=40$) were detected in their work. They had used recent clinical isolates and did not document any prior typing of strains to exclude possible duplicates. Of course, considering the small number of strains that were tested, this difference may alternatively be explained by chance.

To assess the quality of agglutination assays, characterization of test strains is very important and genetically diverse isolates should be tested. We used a panel of 52 non-identical MRSA strains as determined by PFGE. All test systems used in this study for genus and species determination of panel strains are well established. Depending upon the parameter that is within the scope of a survey (sensitivity or specificity), all evaluated agglutination test systems may be valuable instruments for routine use in diagnostic microbiology laboratories. However, as shown in Table 4, the NPV of test kits that miss a great proportion of MRSA strains, such as the Staphylase Test®, decreases considerably as the local MRSA prevalence increases (NPV = 66·9 % at 50 % MRSA prevalence).

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