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Comparison of the Next-Generation Xpert MRSA/SA BC Assay and the GeneOhm StaphSR Assay to Routine Culture for Identification of *Staphylococcus aureus* and Methicillin-Resistant *S. aureus* in Positive-Blood-Culture Broths

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A bloodstream infection with *Staphylococcus aureus*, including methicillin-resistant *S. aureus* (MRSA), is a serious condition that carries a high mortality rate and is also associated with significant hospital costs. The rapid and accurate identification and differentiation of methicillin-susceptible *S. aureus* (MSSA) and MRSA directly from positive blood cultures has demonstrated benefits in both patient outcome and cost-of-care metrics. We compare the next-generation Xpert MRSA/SA BC (Xpert) assay to the GeneOhm StaphSR (GeneOhm) assay for the identification and detection of *S. aureus* and methicillin resistance in prospectively collected blood culture broths containing Gram-positive cocci. All results were compared to routine bacterial culture as the gold standard. Across 8 collection and test sites, the Xpert assay demonstrated a sensitivity of 99.6% (range, 96.4% to 100%) and a specificity of 99.5% (range, 98.0% to 100%) for identifying *S. aureus*, as well as a sensitivity of 98.1% (range, 87.5% to 100%) and a specificity of 99.6% (range, 98.3% to 100%) for identifying MRSA. In comparison, the GeneOhm assay demonstrated a sensitivity of 99.2% (range, 95.2% to 100%) and a specificity of 96.5% (range, 89.2% to 100%) for identifying *S. aureus*, as well as a sensitivity of 94.3% (range, 87.5% to 100%) and a specificity of 97.8% (range, 96.1% to 100%) for identifying MRSA. Five of six cultures falsely reported as negative for MRSA by the GeneOhm assay were correctly identified as positive by the Xpert assay, while one culture falsely reported as negative for MRSA by the Xpert assay was correctly reported as positive by the GeneOhm assay.

Bloodstream infection (BSI) is a serious condition, resulting in >500,000 hospitalizations per year in the United States, accounting for up to 11% of intensive care unit admissions (1, 2). Mortality associated with BSI can range from 25% to 80%, depending on underlying illnesses, and it is higher in nosocomial versus community-acquired infections (3–5). Bloodstream infections also carry a high monetary burden, ranging from $36,000 to $40,000 in additional expenses per patient as a result of prolonged hospitalization (4–6). The leading causes of community- and hospital-acquired BSI are *Staphylococcus aureus*, *Staphylococcus epidermidis*, and various other coagulase-negative *Staphylococcus* (CoNS) species (5, 7). Within this group of organisms, infection with methicillin-resistant *S. aureus* (MRSA) is most critical and has been associated with a mortality rate 1.70 to 1.93 times higher than that of methicillin-susceptible *S. aureus* (MSSA) strains (8, 9). The outcome of these infections can be positively impacted by early diagnosis and effective antimicrobial therapy (10–12). Rapid diagnostic methods, such as peptide nucleic acid fluorescence *in situ* hybridization (PNA FISH), matrix-assisted laser desorption ionization–time of flight mass spectrometry (MALDI-TOF MS), and nucleic acid amplification and detection have been successfully applied to directly analyze positive blood culture broths (13–17). These techniques reduce the time to the identification of pathogens, including *Staphylococcus* spp., by 18 to 48 h compared to that with conventional culture and biochemical identification methods. The differentiation of *S. aureus* from CoNS (commonly associated with culture contamination or inadequate skin preparation prior to specimen collection) in blood culture broths can result in a reduced length of hospital stay, vancomycin usage, and overall cost of care (13). Further benefits in patient outcome and cost of care can be achieved using nucleic acid amplification-based tests to detect and differentiate MRSA from MSSA and CoNS (10). This enables the optimization of antimicrobial therapy for MRSA versus MSSA infections (12).

The molecular tests for identification of *Staphylococcus* spp., including MRSA, directly from positive blood culture broths include the GeneOhm StaphSR assay (BD, Sparks, MD), Verigene BC-GP (Nanosphere, Northbrook, IL), FilmArray blood culture identification (BCID) (BioFire, Salt Lake City, UT), and the Xpert MRSA/SA BC assay (Cepheid, Sunnyvale, CA). Multiplexed assays, such as the Verigene BC-GP and FilmArray BCID, employ

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separate probes for detecting *S. aureus* and *mecA*, the gene that confers resistance to methicillin. This approach has proven to be sensitive for detecting MRSA; however, the independent detection of these markers can result in false-positive results when a culture contains both MSSA and a methicillin-resistant CoNS (MR-CoNS) (14, 18). In contrast, more traditional real-time PCR (RT-PCR)-based assays, including the GeneOhm StaphSR and Xpert MRSA/SA BC assays, target the junction of the staphylococcal cassette chromosome *mec* element (SCC*mec*) (a chromosomal cassette harboring *mecA*) and *orfX* in order to specifically identify MRSA. A benefit to this approach is the ability to discriminate MRSA from *S. aureus* and MR-CoNS in mixed cultures. The Xpert MRSA/SA BC assay contains additional primers and probes to ensure that *mecA* is present, reducing the chance of a false-positive result. Initial evaluations of the performance of the GeneOhm and Xpert MRSA/SA BC assays reported 98.3% to 100% sensitivity and 98.4% to 99.4% specificity for the identification of MRSA (17, 19). A drawback to the use of a surrogate marker (SCC*mec-orfX*) for identifying MRSA is the potential for genetic rearrangements or point mutations that affect either the SCC*mec-orfX* primer binding sites or the *mecA* gene itself, which can result in false-negative results. Recent studies using pure cultures of previously characterized isolates have reported sensitivities as low as 50% to 92% for identifying MRSA in simulated blood cultures, many of which contained the *mecC* determinant, or as a result of variant SCC*mec* types or genetic rearrangements/deletions within the SCC*mec* cassette (20–23).

We compared the performance of the next-generation Xpert MRSA/SA BC assay (Xpert) to that of the GeneOhm StaphSR assay for identifying and detecting *S. aureus* and methicillin resistance in prospectively collected blood culture broths containing Gram-positive cocci. The new Xpert MRSA/SA BC assay cartridge contains all the reagents required to run the assay on-board, and the time to results is approximately 10 min longer due to changes in the automated sample processing. The two molecular tests were compared to routine culture and identification methods using latex agglutination and cefoxitin disk diffusion tests as the reference method.

MATERIALS AND METHODS

Study enrollment. Eight clinical centers representing different geographic locations within the United States participated in a prospective study to assess the clinical performance of the next-generation Xpert MRSA/SA BC assay. Each center enrolled blood cultures using BD Bactec Plus Aerobic/F (BD, Sparks, MD), VersaTREK REDOX 1 (Thermo Fisher), or bioMérieux BacT/Alert SA Standard Aerobic (bioMérieux, Hazelwood, MO) medium. The cultures that were identified as positive by an automated blood culture system were Gram stained to confirm the presence of bacteria. Cultures containing Gram-positive cocci as individual cells or in clusters were considered for enrollment in the study. The specimens were tested using the next-generation Xpert MRSA/SA BC assay and the GeneOhm assay, according to the product insert criteria for each test. Only one blood culture per patient was enrolled to avoid duplicate analysis of a single bacterial isolate. The study included blood cultures obtained from both adult and pediatric patients; however, pediatric patients were not the focus of the study and comprised <3% of the cultures enrolled in the study. To avoid bias, the results from the molecular assays were not known to the personnel conducting the reference culture method testing. This study protocol was independently approved by the institutional review board (IRB) at each clinical center.

Xpert MRSA/SA BC assay. For the Xpert MRSA/SA BC assay, blood cultures containing Gram-positive cocci were tested within 24 h of culture positivity if held at room temperature or within 72 h if held at 2 to 8°C. A 50-μl aliquot of the specimen was transferred to an elution reagent vial (provided) and vortexed for 10 s. The entire contents of the elution reagent vial were then transferred to an Xpert MRSA/SA BC test cartridge, which was sealed and inserted into the GeneXpert for analysis. The Xpert MRSA/SA BC assay targets *spa*, *mecA*, and the SCC*mec-orfX* junction using proprietary primer and probe sequences. The detection of all 3 targets was interpreted as positive for MRSA. If *spa* was detected alone or in conjunction with SCC*mec* but *mecA* was not detected, the result was interpreted as *S. aureus* (i.e., MSSA). If *spa* and *mecA* were detected in the absence of the SCC*mec-orfX* junction, the result was also interpreted as *S. aureus* (i.e., MSSA). If *spa* was not detected, the result was interpreted as negative for *S. aureus* regardless of SCC*mec-orfX* and/or *mecA* being detected.

GeneOhm StaphSR assay. The blood cultures were tested within 48 h of positivity using the GeneOhm assay. A 2-μl aliquot of culture medium was transferred to a sample buffer tube (provided) and vortexed for 10 s. Fifty microliters of the homogenized mixture was transferred to a second tube containing glass beads (provided), vortexed for 5 min to ensure lysis, and heated to 95°C for 2 min. Following reconstitution, 25 μl of reaction master mix and 3 μl of each lysed heat-inactivated sample were added to individual SmartCycler PCR tubes. Real-time PCR was conducted in the Cepheid SmartCycler II. The results were interpreted as positive for MRSA, positive for *S. aureus*, or negative based upon the detection of amplicon corresponding to SCC*mec-orfX* and/or proprietary *S. aureus*-specific genetic targets. The specimens generating invalid results (i.e., a reading of "invalid," "error," "no result," or "unresolved") on either assay were repeated once. The specimens generating a second invalid result were excluded from statistical analysis.

Reference culture method. The blood culture broths were plated to agar medium containing 5% sheep blood and incubated for 18 to 48 h. Beta-hemolytic colonies were identified as *S. aureus* using Gram stain morphology, a positive catalase test, and positive *S. aureus* latex agglutination (BactiStaph; Remel, Lenexa, KS). All isolates identified as *S. aureus* were tested for oxacillin/methicillin resistance using the cefoxitin disk diffusion method in accordance with Clinical and Laboratory Standards Institute (CLSI) standards M02-A11 (24) and M100-S22 (25). In brief, a 0.5 McFarland suspension of the test strain was plated on Mueller-Hinton agar (MHA). A 30-μg cefoxitin disk was placed onto the plate, and the culture was incubated aerobically for 16 to 18 h at 35°C, at which point the zone of inhibition was measured and interpreted (≤21 mm, resistant; ≥22 mm, susceptible).

Data analysis. The results for each of the molecular assays were compared to those of the reference culture and antimicrobial susceptibility testing methods. The sensitivities and specificities were calculated using standard methods. The 95% confidence interval was determined using the binomial expansion method. The poolability of the data between sites was examined using Fisher's exact test. If *P* values were <0.05, the difference between the groups was considered to be statistically significant. The statistical significance between the performance (sensitivity and specificity) of each assay was established using McNemar's test (26).

RESULTS

Study population. A total of 795 blood culture broths (468 BD Bactec Plus Aerobic/F sites A, B, C, F, and H; 197 VersaTREK REDOX 1 sites D and G; and 130 BacT/Alert SA Standard Aerobic site E) meeting the study criteria were collected and tested at 8 clinical centers using the Xpert MRSA/SA BC assay. The prevalences based on culture-confirmed results were 29.7% (range, 20.8% to 38.6%) for *S. aureus* and 13.3% (range, 7.0% to 17.6%) for MRSA. The Xpert MRSA/SA BC assay successfully returned results for 764/795 broths (96.1%) following the initial test. This increased to 792/795 (99.6%) broths following a single retest of the specimens initially reported to be invalid, error, or no result. A

TABLE 1 Performances of Xpert MRSA/SA BC and GeneOhm StaphSR assays for detection of *S. aureus* compared to that of routine culture method

Test	Site	Total no. of specimens tested	No. with result[a]: TP	FP	TN	FN	Sensitivity (% [95% CI])[b]	Specificity (% [95% CI])
Xpert MRSA/SA BC	A	63	19	0	44	0	100 (82.4–100)	100 (92.0–100)
	B	91	32	0	59	0	100 (89.1–100)	100 (93.9–100)
	C	44	17	0	27	0	100 (80.5–100)	100 (87.2–100)
	D	70	26	0	44	0	100 (86.8–100)	100 (92.0–100)
	E	130	27	2[c]	100	1[d]	96.4 (79.7–99.8)	98.0 (92.4–99.6)
	F	211	65	1[e]	145	0	100 (94.5–100)	99.3 (96.2–100)
	G	126	31	0	95	0	100 (88.8–100)	100 (96.2–100)
	H	57	18	0	39	0	100 (81.5–100)	100 (91.0–100)
	Total	792	235	3	553	1	99.6 (97.7–99.9)	99.5 (98.4–99.9)
GeneOhm StaphSR	A	60	18	0	42	0	100 (81.5–100)	100 (91.6–100)
	B	91	32	2	57	0	100 (89.1–100)	96.6 (88.3–99.6)
	C	52	20	1	30	1	95.2 (76.2–99.9)	96.8 (73.2–95.8)
	D	63	25	0	38	0	100 (86.3–100)	100 (90.7–100)
	E	131	28	4	99	0	100 (87.7–100)	96.1 (90.4–98.9)
	F	202	64	8	130	0	100 (94.4–100)	94.2 (88.9–97.5)
	G	127	30	0	96	1[d]	96.8 (83.3–99.9)	100 (96.2–100)
	H	56	17	4	30	0	100 (80.5–100)	89.7 (75.8–97.1)
	Total	782	234	19	527	2	99.2 (97.0–99.9)	96.5 (94.6–97.9)

[a] TP, true positive; FP, false positive; TN, true negative; FN, false negative.
[b] CI, confidence interval.
[c] 1 identified as MRSA and 1 identified as *S. aureus* (MSSA) by the Xpert MRSA/SA BC assay.
[d] Identified as MSSA by culture.
[e] Identified as *S. aureus* (MSSA) by the Xpert MRSA/SA BC assay.

total of 806 specimens (474 BD Bactec Plus Aerobic/F, 197 VersaTREK REDOX 1, and 135 BacT/Alert SA Standard Aerobic) were tested using the GeneOhm assay and reference culture, according to the study criteria. The GeneOhm assay generated a valid result for 776/806 (96.3%) specimens on the initial test. A single repeat test was conducted with 21/30 samples that failed the initial analysis, of which 6 generated a valid result. The remaining 9 samples with an initial invalid result were not retested within the time frame specified by the product insert. This resulted in a final valid result rate of 782/797 (98.1%).

Identification of *S. aureus*. The two molecular tests demonstrated similar sensitivities (Xpert, 99.6%; GeneOhm, 99.2%; $P =$ 0.99 to 1.00, Fisher's exact test) for identifying *S. aureus* (MRSA and MSSA) in blood culture broths (Table 1). The sensitivities of the molecular tests ranged from 95.2% to 100% among the 8 clinical centers; however, the intersite variability for each test was not statistically significant ($P > 0.05$, Fisher's exact test). Additionally, there was not a statistically significant difference in performance between the three blood culture systems used by the various study sites. One sample tested falsely negative for *S. aureus* by the Xpert assay but it tested positive by the GeneOhm assay (Table 1, site E). Two samples tested falsely negative by the GeneOhm assay but were positive for *S. aureus* by the Xpert assay (Table 1, sites C and G). The specificity of the Xpert test was 99.5% (553/556), with three false-positive *S. aureus* results reported. All three specimens were also reported as either positive (i.e., MRSA) or reactive (i.e., *S. aureus*) by the GeneOhm assay, all of which matched the corresponding Xpert assay results. The GeneOhm assay was comparatively less specific (96.5% [527/546]), reporting 19 false-positive *S. aureus* results ($P < 0.001$).

Identification of MRSA. The identification of MRSA in blood culture broths showed greater variability between the 2 molecular tests (Table 2). The sensitivity of the Xpert assay was a combined 98.1% (103/105), while the sensitivity of the GeneOhm assay was 94.3% (99/105). The sensitivity of the Xpert assay ranged from 87.5% to 100% across the eight clinical centers but was a combined 100% (77/77) at six of the eight clinical centers. This intersite variability was not statistically significant ($P > 0.05$, Fisher's exact test), nor was there a statistically significant difference in performance between the culture media. Across all test sites, the Xpert assay reported two blood cultures to be *S. aureus* that were confirmed to be MRSA by the cefoxitin disk diffusion test (i.e., false-negative Xpert assay results). Also, one of two blood cultures was negative for MRSA by the GeneOhm assay (Table 3, specimen C039), while the other tested positive (Table 3, specimen G013). Of note, specimen G013 was reported to contain MRSA by reference culture, while specimen C039 was reported to contain both MRSA and MSSA. Across 5 test sites, GeneOhm assay reported 6 cultures to be negative for MRSA, for a combined sensitivity of 94.3% (range, 87.5% to 100%). All 6 false-negative reports were had a positive result for *S. aureus* by the GeneOhm assay only, but 5/6 tested positive for MRSA by the Xpert assay (Table 3).

The specificity of the Xpert assay for identifying MRSA was >99.6% (684/687), with 3 false-positive results across all 8 test sites (Table 2). Two of these results were obtained from blood cultures containing methicillin-susceptible *S. aureus*; however, one sample was negative for *S. aureus* by reference culture, indicating possible contamination or the presence of a nonviable organism. The specificity of the GeneOhm assay was 97.8% (662/677). Of the 15 false-positive results for MRSA, 7 blood cultures contained methicillin-susceptible *S. aureus*, and 9 were negative for *S. aureus* by reference culture.

TABLE 2 Performances of Xpert MRSA/SA BC and GeneOhm StaphSR assays for detection of MRSA compared to that of routine culture method

Test	Site	Total no. of specimens tested	No. with result[a]: TP	FP	TN	FN	Sensitivity (% [95% CI])[b]	Specificity (% [95% CI])
Xpert MRSA/SA BC	A	63	11	0	52	0	100 (71.5–100)	100 (93.2–100)
	B	91	16	0	75	0	100 (79.4–100)	100 (95.2–100)
	C	44	7	0	36	1	87.5 (47.3–99.7)	100 (90.3–100)
	D	70	11	1[c]	58	0	100 (71.5–100)	98.3 (90.9–100)
	E	130	10	1[d]	119	0	100 (69.2–100)	99.2 (95.4–100)
	F	211	25	1[c]	185	0	100 (86.3–100)	99.5 (97.0–100)
	G	126	19	0	106	1	95.0 (73.0–99.7)	100 (96.2–100)
	H	57	4	0	53	0	100 (81.5–100)	100 (91.6–100)
	Total	792	103	3	684	2[e]	98.1 (93.3–99.8)	99.6 (98.7–99.9)
GeneOhm StaphSR	A	60	10	0	49	1	90.9 (58.7–99.8)	100 (96.4–98.8)
	B	91	14	1[d]	74	2	87.5 (61.7–98.4)	98.7 (92.8–100)
	C	52	8	1[c]	42	1	88.9 (51.8–99.7)	97.7 (87.7–99.9)
	D	63	10	0	53	0	100 (69.2–100)	100 (93.3–100)
	E	131	10	3[f]	118	0	100 (69.2–100)	97.5 (92.9–99.5)
	F	202	24	7[g]	170	1	96.0 (79.6–99.9)	96.1 (92.0–98.4)
	G	127	19	1[c]	106	1	95.0 (75.1–99.9)	99.1 (94.9–100)
	H	56	4	2[h]	50	0	100 (39.8–100)	96.2 (86.8–99.5)
	Total	782	99	15	662	6[i]	94.3 (88.0–97.9)	97.8 (96.4–98.8)

[a] TP, true positive; FP, false positive; TN, true negative; FN, false negative.
[b] CI, confidence interval.
[c] Identified as MSSA by culture.
[d] Negative for *S. aureus* by culture.
[e] Identified as *S. aureus* (MSSA) by the Xpert MRSA/SA BC assay.
[f] One identified as *S. aureus* (MSSA), and two negative for *S. aureus* by culture.
[g] Two identified as *S. aureus* (MSSA), and five negative for *S. aureus* by culture.
[h] One identified as *S. aureus* (MSSA), and one negative for *S. aureus* by culture.
[i] Identified as *S. aureus* (MSSA) by GeneOhm assay.

DISCUSSION

The next-generation Xpert MRSA/SA BC assay is an on-demand sample-to-result molecular test for the identification of *S. aureus* and MRSA directly from positive blood culture broths. The detection and differentiation of *S. aureus* and MRSA are achieved by incorporating three nucleic acid targets, including the SCC*mec*-*orfX* junction, *mecA*, and *spa*. The BD GeneOhm StaphSR test uses a two-target approach, relying on the detection of the SCC*mec*-*orfX* junction site and an *S. aureus* species-specific target for the identification of MRSA (17). The initial clinical evaluations of the original Xpert MRSA/SA BC assay and the GeneOhm StaphSR test demonstrated sensitivities of 93.7% to 100% for *S. aureus* and 98.3% to 100% for MRSA in positive blood culture broths (17, 19, 27). Subsequently, several published reports indicated sensitivities as low as 50% to 92% for detecting MRSA using the GeneOhm and Xpert MRSA/SA assays (20–23). These false-negative results were primarily attributable to mutations in the junction region of SCC*mec*-*orfX*, the target of the GeneOhm assay, or to variant SCC*mec* types, including type IVa, which in some institutions comprise up to 33% of the SCC*mec* types (20, 22, 23, 28). Additionally, the specificities of these tests for identifying MRSA suffer for strains containing truncated SCC*mec* cassettes or "empty cassettes" lacking a functional *mecA* gene (29, 30). Such strains may account for up to 4.6% of the *S. aureus* strains carrying the cassette (28).

TABLE 3 Specimens resulting in false-negative MRSA calls by the Xpert MRSA/SA BC and/or GeneOhm StaphSR assay

Specimen ID[a]	Zone diam (mm)[b]	Xpert MRSA/SA BC assay result[c]	GeneOhm StaphSR assay result
A004	None	TP	FN
B042	8	TP	FN
B080	7	TP	FN
C039	12, 26[d]	FN	FN
F094	10	TP	FN
G013	6	FN	TP
G022	6	TP	FN

[a] ID, identification.
[b] Zone of inhibition measured by cefoxitin disk diffusion test.
[c] TP, true positive; FN, false negative.
[d] Specimen contained two isolates of *S. aureus*. One MRSA and one MSSA (showing 12-mm and 26-mm zones of inhibition for cefoxitin, respectively).

The Next-generation Xpert MRSA/SA BC assay incorporates a modified sample preparation protocol to improve assay accuracy, although the specific changes to the assay are not publicly available. Though not statistically significant, the Xpert MRSA/SA BC assay demonstrated greater sensitivity than that of the GeneOhm StaphSR test for identifying MRSA. One of the two false-negative results reported by the Xpert assay contained both MSSA and MRSA upon reference culture, and the GeneOhm assay also failed to identify MRSA in this specimen. Taken together, this might suggest contamination of the reference culture with MRSA subsequent to molecular testing. The specificity of the Xpert MRSA/SA BC assay was >99.5% for both *S. aureus* and MRSA, which is equal to or greater than that of the GeneOhm assay. The three specimens with false-positive results for *S. aureus* by the Xpert assay were also detected by the GeneOhm assay, suggesting that the reference culture results may have been falsely negative for these specimens.

Seven specimens generated false-negative results for MRSA on at least one of the molecular assays (Xpert MRSA/SA BC, $n = 2$; GeneOhm, $n = 6$). The possible causes for this include variant SCC*mec* cassettes, as previously discussed, the presence of the *mecC* resistance determinant, or high-level expression of penicillinases leading to borderline oxacillin resistance (31–34). All strains were confirmed to be phenotypically methicillin resistant, demonstrating zones of inhibition ranging from undetectable to 14 mm with the cefoxitin disk diffusion test. It is likely that the resistances in these strains were mediated by *mecA* as opposed the overexpression of a penicillinase, which typically results in cefoxitin zones of inhibition of ≥28 mm (35). MRSA harboring *mecC* will display high-level phenotypic resistance to cefoxitin, which is consistent with the MRSA strains not detected by the GeneOhm assay in this study. However, all but two of these strains were correctly identified as MRSA by the next-generation Xpert MRSA/SA BC assay, effectively ruling this out as an explanation for the false-negative results. Combined, this suggests that the false-negative MRSA results observed may have been due to variant SCC*mec* types not recognized by the GeneOhm assay.

A strength of this study is the participation of 8 clinical centers located in different geographic locations within the United States and the enrollment of a large number of clinical specimens ($n = 795$), which should account for regional and institutional strain variability. The clinical performance measures (sensitivity and specificity) of the next-generation Xpert MRSA/SA BC assay among all sites were statistically equivalent. This indicates the ability to accurately identify *S. aureus* and MRSA strains across different geographic locations, accounting for regional and institutional strain diversity. Additionally, these results demonstrate the ability of the next-generation Xpert MRSA/SA BC assay to generate equivalent results independent of laboratory variables, including different technologists, laboratory workflow practices, and blood culture media. Another strength of this study is the head-to-head comparison of the next-generation Xpert MRSA/SA BC assay with the GeneOhm StaphSR assay, a second commercially available FDA-cleared molecular test for the identification of *S. aureus* and MRSA in positive blood culture broths. Our data demonstrate statistically equivalent performances for these two tests; however, the workflow was simpler with the Xpert MRSA/SA BC assay, which required fewer preanalytic processing steps than the GeneOhm StaphSR and could be conducted on-demand using the random-access GeneXpert system.

A potential weakness of this study was the inclusion of specimens obtained from clinical centers located only within the United States. Several of the studies indicating poorer performances of the GeneOhm and Xpert assays were conducted in the European Union or Australia (20, 21, 23). Clinical evaluations of the redesigned Xpert MRSA/SA BC test in these locations will be necessary to confirm the improved performance demonstrated in the current study. Additionally, specimens that tested as false negative for MRSA were not fully characterized to establish the root cause of the false-negative result; however, only a single specimen tested as false negative on both molecular assays, suggesting that differences in the target and primer design between the tests accounted for the additional false-negative results observed with the GeneOhm assay. Finally, the specimens were not tested simultaneously on both molecular assays. Because the GeneOhm test lends to batch processing, these tests may have been initiated after the initiation of the Xpert test. It is possible that delayed testing negatively impacts results if the specimen contained nucleases that degrade target sequences or accumulated other inhibitory substances resulting from specimen degradation. Alternatively, delayed testing may increase sensitivity due to additional bacterial growth. These factors were not evaluated; however, both molecular tests were performed according to the specimen acceptability criteria set forth in the respective product insert.

The benefits of the rapid detection of *S. aureus* and MRSA directly from positive blood culture broths are well documented. Specifically, molecular testing for *S. aureus* and MRSA resulted in a 21% decrease in the number of patients receiving anti-MRSA therapy and a mean reduction of 12.2 h in the duration of therapy for patients with blood cultures containing Gram-positive cocci that tested negative for *S. aureus* (36). Likewise, the time to optimal antimicrobial therapy for the patients with cultures positive for MSSA was reduced by 38.4 to 44.6 h following the implementation of a molecular test (10, 36). These rapid results contributed to a mean reduction in the length of hospital stay of 6.2 days and a reduction of $21,387 in the total hospital cost per septic episode compared to those of patients diagnosed using routine culture and susceptibility testing methods (10). Importantly, these advantages are realized only when molecular testing can be performed on-demand and the results are actively reported to the clinician. The use of batched testing formats and passive reporting of results does not significantly reduce the time to optimal antimicrobial therapy despite definitive identification of *S. aureus* and MRSA ~13 h sooner than with culture methods (37). The Xpert and GeneOhm assays demonstrated statistically equivalent sensitivities and specificities for identifying *S. aureus* and MRSA in positive blood cultures compared to those with the culture method. A potential advantage of the Xpert MRSA/SA BC assay is the simplified sample-to-result workflow and on-demand capability, which gives it the potential to reduce the turnaround time for blood cultures containing *S. aureus* or MRSA.

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