Cepheid®

Instructions for Use

Xpert® MRSA

For use with GeneXpert® System with Touchscreen

Catalog Numbers

REF GXMRSA-100N-10

REF GXMRSA-120

303-0142 | Rev. B | 2024-05

℞only IVD *In Vitro* Diagnostic Medical Device

Trademarks, Patents, and Copyright Statements

See Revision History for a description of changes.

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Getting Started

Product Information

Proprietary Name

Xpert® MRSA

Common or Usual Name

Xpert MRSA test

Intended Use, Summary, and Principle of Procedure

Intended Use

The Xpert MRSA test performed in the GeneXpert® Instrument Systems is a qualitative *in vitro* diagnostic test designed for rapid detection of methicillin-resistant *Staphylococcus aureus* (MRSA) from nasal swabs in patients at risk for nasal colonization. The test utilizes automated real-time polymerase chain reaction (PCR) to detect MRSA DNA. The Xpert MRSA test is intended to aid in the prevention and control of MRSA infections in healthcare settings. The Xpert MRSA test is not intended to diagnose MRSA nor to guide or monitor treatment for MRSA infections. Concomitant cultures are necessary only to recover organisms for epidemiological typing or for further susceptibility testing.

Summary and Explanation

Staphylococcus aureus (SA) is a major healthcare-associated pathogen that causes a range of diseases including endocarditis, osteomyelitis, toxic shock syndrome, food poisoning, carbuncles and boils. In the early 1950s, acquisition and spread of beta-lactamase-producing plasmids thwarted the effectiveness of penicillin for treating *S. aureus* infections. In 1959, methicillin, a semi-synthetic penicillin, was introduced. By 1960, methicillin-resistant *S. aureus* strains were identified. This was determined to be the result of *S. aureus* acquiring the *mecA* gene. In the US today, MRSA is responsible for approximately 25% of healthcare-associated infections and reports of community-acquired MRSA are increasing, resulting in significant morbidity and mortality. In an attempt to limit the spread of these infections, control strategies and policies are being developed and implemented in healthcare settings. Controlling MRSA is a primary focus of most

hospital infection prevention programs. Currently detecting MRSA can be accomplished using culture or molecular methods, the latter of which is very laborious and time intensive rapid and more sensitive method for surveillance of MRSA.[1,2,3,4,5]

Principle of the Procedure

The GeneXpert Instrument Systems automate and integrate sample purification, nucleic acid amplification, and detection of target sequence in simple or complex samples using real-time PCR and RT-PCR tests. The system consists of an instrument, personal computer, and preloaded software for running tests on collected samples and viewing the results. The system requires the use of single-use disposable GeneXpert cartridges that hold the PCR reagents and host the PCR process. Because the cartridges are self-contained, cross-contamination between samples is significantly reduced. For a full description of the system, see the system operator manual.

The Xpert MRSA test includes reagents for the detection of MRSA as well as a sample processing control (SPC) to control for adequate processing of the sample and to monitor the presence of inhibitor(s) in the PCR reaction. The Probe Check Control (PCC) verifies reagent rehydration, PCR tube filling in the cartridge, probe integrity, and fluorophore stability. The primers and probes in the Xpert MRSA test detect a proprietary sequence for the presence of a cassette inserted into the *S. aureus* chromosome.

Reagents, Instruments, and Materials

Reagents

Material Provided

The Xpert MRSA (GXMRSA-100N-10) kit contains sufficient reagents to process 10 specimens or quality control samples. The Xpert MRSA (GXMRSA-120) kit contains sufficient reagents to process 120 specimens or quality control samples. The kit contains the following:

Xpert MRSA Cartridges with Integrated Reaction Tubes	**10**	**120**
Bead 1, Bead 2, and Bead 3 (freeze-dried)	1 per cartridge	1 per cartridge
Reagent 1 (Sodium Hydroxide)	3.0 mL per cartridge	3.0 mL per cartridge
Reagent 2	3.0 mL per cartridge	3.0 mL per cartridge
Xpert MRSA Reagent Pouch	**1**	**1**
Elution Reagent (Guanidinium thiocyanate)	10 x 1.5 mL per vial	120 x 1.5 mL per vial
CD	1	1

- Assay Definition File (ADF)
- Instructions to import ADF into GX software
- Instructions for Use (Package Insert)

Note Safety Data Sheets (SDS) are available at www.cepheid.com or www.cepheidinternational.com under the **SUPPORT** tab.

Note The bovine serum albumin (BSA) in the beads within this product was produced and manufactured exclusively from bovine plasma sourced in the United States. No ruminant protein or other animal protein was fed to the animals; the animals passed ante- and post-mortem testing. During processing, there was no mixing of the material with other animal materials.

Materials Required but Not Provided

- GeneXpert system with touchscreen running Cepheid OS software version 2.0 or higher, GeneXpert instrument, touchscreen unit, and operator manual.
- Printer: If a printer is required, contact Cepheid Technical Support to arrange for the purchase of a recommended printer.
- Cepheid Sample Collection Device (900-0370) or Copan equivalent

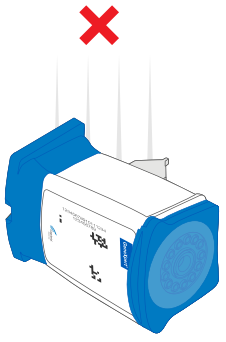
- Vortex mixer
- Disposable transfer pipettes
- Sterile Gauze

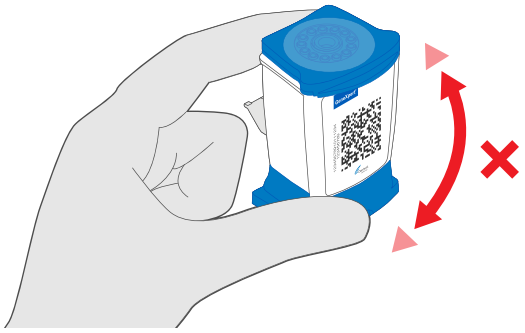
Materials Available but Not Provided

KWIK-STIKs™ from Microbiologics catalog #0158MRSA as external positive controls and #0371MSSE (methicillin-susceptible *Staphylococcus epidermidis*) as external negative control.

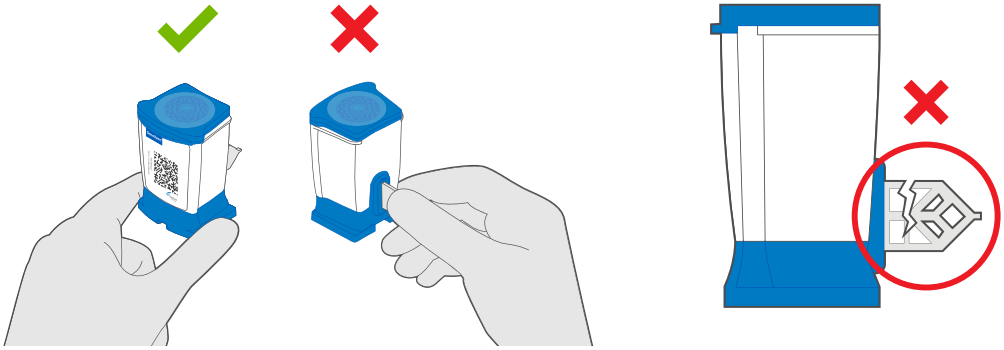
Warnings and Precautions

- For *in vitro* Diagnostic Use. Rx only
- Treat all biological specimens, including used cartridges, as if capable of transmitting infectious agents. Because it is often impossible to know which might be infectious, all biological specimens should be treated with standard precautions. Guidelines for specimen handling are available from the U.S. Centers for Disease Control and Prevention,[7] and the Clinical and Laboratory Standards Institute.
- Follow your institution's safety procedures for working with chemicals and handling biological samples.
- Biological specimens, transfer devices, and used cartridges should be considered capable of transmitting infectious agents thus requiring the use of standard precautions. Follow your institution's environmental waste procedures for proper disposal of used cartridges and unused reagents. These materials may exhibit characteristics of chemical hazardous waste requiring specific national or regional disposal procedures. If national or regional regulations do not provide clear direction on proper disposal,biological specimens and used cartridges should be disposed per WHO [World Health Organization] medical waste handling and disposal guidelines. Consult your institution's environmental waste personnel on proper disposal of used cartridges and unused reagents.
- The Xpert MRSA test does not provide susceptibility testing results. Additional time is required to culture and perform susceptibility testing.
- Do not substitute Xpert MRSA test reagents with other reagents.
- Do not open the Xpert MRSA cartridge lid except when adding sample.
- Do not use a cartridge that has been dropped or shaken after you have added the sample and reagents.





- Hold the cartridge by the base. Do not touch the reaction tube at the rear of the cartridge, as this could cause damage that would interfere with light passing through it during the test. Do not use a cartridge that has a damaged reaction tube.



- Do not place a label on the cartridge lid or barcode label.

- Each single-use Xpert MRSA cartridge is used to process one test. Do not reuse spent cartridges.
- Store the Xpert MRSA test kit at 2 – 28 °C.

Chemical Hazards, Storage and Handling

Chemical Hazards[8, 9]

- UN GHS Hazard Pictogram:

- Signal Word: WARNING
- **UN GHS Hazard Statements**
  - Harmful if swallowed
  - Causes skin irritation
  - Causes serious eye irritation
- **UN GHS Precautionary Statements**
  - **Prevention**
    - Wash hands thoroughly after handling.
    - Do not eat, drink, or smoke when using this product.
    - Avoid release to the environment.
    - Wear protective gloves/protective clothing/eye protection/face protection
  - **Response**
    - IF ON SKIN: Wash with plenty of soap and water.

- Take off contaminated clothing and wash before reuse.
- Specific treatment, see the supplemental first aid information.
- If skin irritation occurs: Get medical advice/attention.
- IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
- If eye irritation persist: Get medical advice/attention
- IF SWALLOWED: Immediately call a POISON CENTER or doctor/physician if you feel unwell.
- Rinse mouth.

- **Storage Disposal**
  - Dispose of content and/or container in accordance with local, regional, national, and/or international regulations.

Storage and Handling

- Store the Xpert MRSA cartridges and reagents at 2 – 28°C.
- Do not use reagents or cartridges that have passed the expiration date.
- Do not open a cartridge lid until you are ready to perform testing.
- Do not use any reagents that have become cloudy or discolored.

Specimen Collection, Testing, and Results

Specimen Collection

Specimen Collection, Transport and Storage

To obtain an adequate specimen, follow the instructions in this section closely.

1. Open the Cepheid Collection Device by peeling back the outer packaging.
2. Ask the patient to tilt his/her head back. Insert dry swabs approximately 1–2 cm into each nostril.
3. Rotate the swabs against the inside of the nostril for 3 seconds. Apply slight pressure with a finger on the outside of the nose to help assure good contact between the swab and the inside of the nose.
4. Using the same swabs, repeat for the second nostril, trying not to touch anything but the inside of the nose.
5. Remove the plastic transport tube. Twist off the tube cap and discard it. Place the swabs into the plastic transport tube. The swabs should go all the way into the tube until they rest on top of the sponge at the bottom of the tube. Make sure the red cap is on tightly. The swabs should stay attached to the red cap at all times.
6. Label the plastic transport tube with patient ID and send to the laboratory.
7. Store swab specimen at room temperature (15–30 °C) if it will be processed within 24 hours, otherwise store swab at 2–8 °C. The swab specimen is stable up to 5 days when stored at 2–8 °C.

Procedure

Preparing the Cartridge

Important **Start the test within 15 minutes of adding the sample to the cartridge.**

Note Use only one of the swabs. The second swab is required for repeat testing.

To add the sample into the cartridge (Xpert MRSA):

1. Remove the cartridge and reagent from the kit.
2. Remove the swabs from the transport container and then remove one swab from the red cap.

3. Insert the swab into the tube containing the elution reagent.
4. Use sterile gauze to minimize risks of contamination.
5. Hold the swab by the stem near the rim of the tube, lift the swab a few millimeters from the bottom of the tube and push the stem against the edge of the tube to break it. Make sure the swab is short enough to allow the cap to close tightly.
6. Close the cap and vortex at high speed for 10 seconds.
7. Open the cartridge lid. Using a sterile transfer pipette, transfer the entire contents of the elution reagent to the sample chamber of the Xpert MRSA cartridge.
8. Close the cartridge lid.

Figure 1 Xpert MRSA Cartridge (Top View)

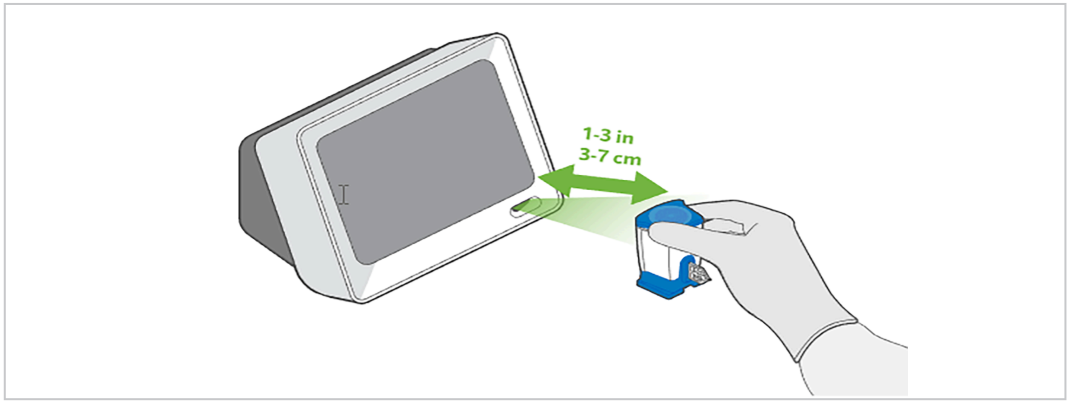
Starting the Test: GeneXpert System with Touchscreen

Important **Before you start the test, make sure that:**

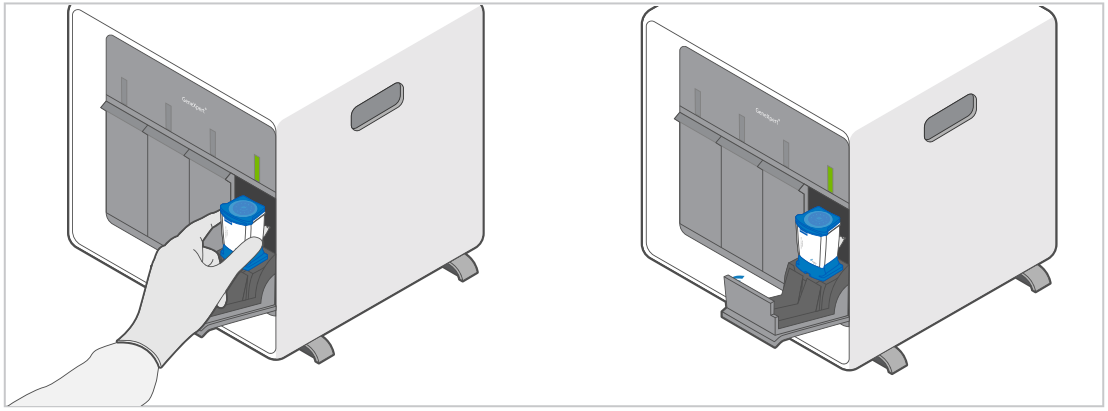
- **The system is running the correct Cepheid OS software version shown in section - Materials Required but Not Provided.**
- **The correct assay definition file is imported into the software.**

Note The default workflow is shown. Your system administrator may alter the workflow.

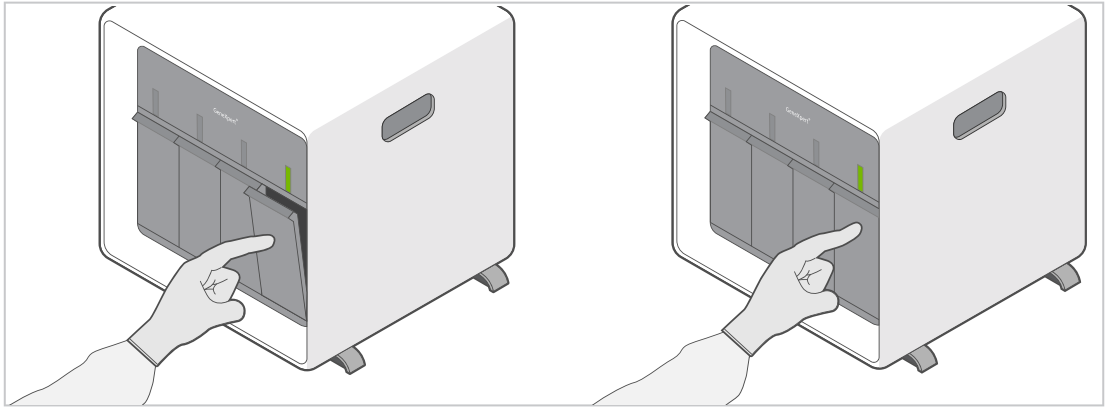
1. Turn on GeneXpert system with touchscreen.
2. Log on to system software using your username and password.
3. On the Modules tab, touch **Start Test**.
4. Follow onscreen prompts to create new test and enter patient and sample information.
5. Scan or manually input the cartridge serial number. If scanning, hold the cartridge about 1-3 inches (3-7 cm) away from the scanner. The scanner projects a green crosshair, which you center on the barcode. Scanning is complete when you hear an audible beep. Touch **Continue**.



6. Select the desired test and touch **Continue**.
7. Watch the cartridge preparation video, if needed.
8. On the Confirm screen, review all data and touch **Confirm**.
9. Open the module door under flashing green light and insert the cartridge.



10. Close cartridge module door completely by pressing until it latches. The test starts.



11. When the test completes, the **Results Summary** screen appears. Open the module door and remove cartridge.
12. Dispose of used cartridge in appropriate waste container according to your institution's standard practices.

Viewing Results: GeneXpert System with Touchscreen

The GeneXpert system with touchscreen results screen will automatically interpret test results for you and clearly show them in the **View Results** window.

1. Tap **Results**.
2. Tap the test to be viewed in the Results screen.
3. Click **OK**.
4. To generate a PDF report file, touch **View Report**. More detailed instructions for viewing and uploading results are available in your system operator manual.

Quality Control

Each test includes a Sample Processing Control (SPC) and Probe Check Control (PCC).

Sample Processing Control (SPC)—Ensures the sample was correctly processed. The SPC verifies that lysis of MRSA has occurred if the organisms are present and verifies that specimen processing is adequate. Additionally, this control detects specimen-associated inhibition of the real-time PCR test. The SPC should be positive in a negative sample and can be negative or positive in a positive sample. The SPC passes if it meets the validated acceptance criteria.

Probe Check Control (PCC)—Before the start of the PCR reaction, the GeneXpert Instrument Systems measure the fluorescence signal from the probes to monitor bead rehydration, reaction-tube filling, probe integrity and fluorophore stability. Probe Check passes if it meets the assigned acceptance criteria.

External Controls—KWIK-STIK™ (Microbiologics, catalog # 0158 MRSA as positive control and # 0371 MSSE as negative control) may be used for training, proficiency testing and external QC of the GeneXpert Instrument System. External controls may be used in accordance with local, state, and federal accrediting organizations, as applicable. Follow the Microbiologics external control procedure described below:

1. Tear open the pouch at the notch and remove the KWIK-STIK.
2. Pinch the bottom of the ampoule in the cap to release the hydrating fluid.
3. Hold vertically and tap to facilitate flow of fluid through shaft into bottom of unit containing pellet.
4. To facilitate dissolution of the lyophilized cell pellet, crush the pellet and gently pinch the bottom chamber.
5. Pull apart the KWIK-STIK to release the swab, and insert the swab into the tube containing the Elution Reagent.
6. The KWIK-STIK swab is now ready for Xpert MRSA testing.

Results

The results are interpreted by the GeneXpert Instrument Systems from measured fluorescent signals and embedded calculation algorithms and will be shown in the View Results window. Possible results are:

Result	Interpretation
MRSA POSITIVE	MRSA target DNA is detected (presumptive positive for MRSA colonization). • MRSA—POSITIVE: MRSA target has a Ct within the valid range and endpoint above the minimum setting. • SPC — NA (not applicable); SPC is ignored because MRSA amplification may compete with this control. • Probe Check — PASS; all probe check results pass.
MRSA NEGATIVE	MRSA target DNA is not detected (presumed not colonized with MRSA), SPC meets acceptance criteria. • MRSA—NEGATIVE: MRSA target DNA is not detected. • SPC — The SPC must be positive if the MRSA target is negative or the test is invalid. • Probe Check — PASS; all probe check results pass.
INVALID	Presence or absence of MRSA cannot be determined, repeat test with the extra swab. SPC does not meet acceptance criteria, the sample was not properly processed, or PCR is inhibited. • MRSA—INVALID: Presence or absence of MRSA DNA cannot be determined. • SPC—FAIL: MRSA target result is negative and the SPC Ct is not within valid range and endpoint below minimum setting. • Probe Check—PASS: All probe check results pass.
ERROR	Presence or absence of MRSA cannot be determined, repeat test according to instructions in the section below. The Probe Check Control failed, which is probably due to an improperly filled reaction tube, a probe integrity problem, or because the maximum pressure limits were exceeded. • MRSA — NO RESULT • SPC — NO RESULT • Probe Check — FAIL*; one or more of the probe check results fail. * If the probe check passed, the error is caused by a system component failure.
NO RESULT	Presence or absence of MRSA cannot be determined, repeat test according to instructions in the section below. Insufficient data were collected to produce a test result. For example, this can occur if the operator stopped a test that was in progress. • MRSA — NO RESULT • SPC — NO RESULT • Probe Check — NA (not applicable)

Reasons to Repeat the Test

Repeat the test using a new cartridge and new Elution Reagent (do not re-use the cartridge) or initiate alternate procedures if oneof the following test results occurs:

- An **INVALID** result indicates that the control SPC failed. The sample was not properly processed or PCR was inhibited.
- An **ERROR** result indicates that the Probe Check Control failed and the test was aborted possibly due to the reaction tube being filled improperly, a reagent probe integrity problem was detected, or because the maximum pressure limits were exceeded.
- A **NO RESULT** indicates that insufficient data were collected. For example, the operator stopped a test that was in progress.

Limitations

Limitations of the Procedure

- The performance of the Xpert MRSA test was validated using the procedures provided in this instructions for use only. Modifications to these procedures may alter the performance of the test. Results from the Xpert MRSA test should be interpreted in conjunction with other laboratory and clinical data available to the clinician.
- Xpert MRSA test performance has not been evaluated in patients less than two years of age.
- Nasal swab specimens from neonatal patients having high levels of *mecA* gene-containing coagulase negative staphylococci may yield false positive results due to the presence of an *SCCmec* sequence.
- Erroneous test results might occur from improper specimen collection, not following the recommended sample collection procedure, handling or storage, technical error, sample mix-up, or because the low number of organisms in the specimen is not detected by the test. Careful compliance to the instructions in this insert is necessary to avoid erroneous results.
- Because the detection of MRSA is dependent on the number of organisms present in the sample, reliable results are dependent on proper specimen collection, handling, and storage.
- Rerunning the Xpert MRSA when results are **INVALID**, **ERROR**, and **NO RESULT** should depend on practices and policies within each facility. Alternate procedures (e.g., culture using selective agar plates with or without overnight incubation in a selective enrichment broth) should be available. For culturing, the remaining swab specimen should be placed in an appropriate transport system and cultured within 4 days.
- A positive test result does not necessarily indicate the presence of viable organism. It is, however, presumptive for the presence of MRSA.
- Testing with Xpert MRSA test should be used as an adjunct to other methods available.
- Test results might also be affected by concurrent antimicrobial therapy. Therefore, therapeutic success or failure cannot be assessed using this test because DNA might persist following antimicrobial therapy.
- Mutations or polymorphisms in primer or probe binding regions may affect detection of new or unknown MRSA variants resulting in a false negative result.

Expected Values

In the Xpert MRSA clinical study, a total of 1077 nasal specimens were collected from 1077 subjects at 7 enrolling sites across the United States. The study population was grouped into subjects in nursing homes or extended-stay facilities, hospitalized over 3 days, hospitalized for 3 days or less, out patient clinic and staff or others. The number and percentage of positive and negative cases relative to the reference culture method are calculated and presented in the table below.

Table 1. Expected Values for MRSA in Different Populations

Group	Positive n (%)	Negative n (%)	Total (%)a[a]
Nursing homes, long-term and extended-stay facilities	62 (25.5)	181 (74.5)	243 (22.6)
Hospitalized >3 days	61 (23.0)	204 (77.0)	265 (24.7)
Hospitalized ≤3 days	29 (13.1)	193 (86.9)	222 (20.7)
Out-patient clinic	46 (17.7)	214 (82.3)	260 (24.2)
Staff and others	11 (12.9)	74 (87.1)	85 (7.9)
Total	**209 (19.4)**	**866 (80.6)**	**1075**

a. Two culture positive hospitalized subjects had unknown admission dates.

Specific Performance Characteristics

Clinical Performance

Performance characteristics of the Xpert MRSA test were determined in a multi-site prospective investigation study at seven institutions by comparing the MRSA test on the GeneXpert System (Xpert MRSA test) with a second FDA-cleared nucleic acid amplification test (NAAT), and enriched culture, the most sensitive culture method. Subjects included individuals and medical staff at risk for nasal colonization. Each subject was enrolled in the study only one time. Subjects who had received systemic or topical-nasal antimicrobial agents in the period 48 hours to one week prior to study enrollment, those under 2 years of age, and those who had contraindication to nasal swab collection were excluded from the study. Only those subjects meeting the inclusion criteria were enrolled.

Four Nasal swabs were collected from each subject. One swab was tested by the Xpert MRSA test and another swab by the second FDA-cleared NAAT test. The two types of NAAT tests were performed at each participating institution and an additional swab was sent to a centralized laboratory for culture testing.

At the centralized laboratory, the swab was directly streaked on to a selective chromogenic agar plate containing cefoxitin and the plate was incubated for 24–48 hours at 35 ±2 °C. The swab was transferred to trypticase soy broth (TSB) with 6.5% sodium chloride and incubated for 18–24 hours at 35 ±2 °C. If the direct culture was negative at 24 hours, the enriched TSB was streaked onto another chromogenic agar plate with cefoxitin and incubated for 24–48 hours at 35 ±2 °C. Confirmation of presumptive positives colonies from either culture method was performed with a tube coagulase test and Gram stain.

Assay performance of the Xpert MRSA test and the second FDA-cleared NAAT test were calculated relative to the central laboratory culture results (reference culture).

A total of 1077 eligible subjects (one specimen per patient) were tested for MRSA by Xpert MRSA, and a second FDA-cleared NAAT test and culture. The Xpert MRSA identified 86.3% of the specimens positive for MRSA and 94.9% of the specimens negative for MRSA relative to the reference culture method. For the subjects tested, the positive predictive value was 80.5% and the negative predictive value was 96.6%.

Table 2. Xpert MRSA Compared to Reference Culture Method

		Culture				
		+	-			
Xpert MRSA	+	182	44	226	Positive Agreement:	86.3%
	-	29	819	848	Negative Agreement:	94.9%

		Culture				
		+	-			
	Total	211	863	1074[a]	PPV[b]:	80.5%
					NPV:[c]	96.6%

a. Three specimens did not give Xpert results on two attempts.

b. Positive predictive value

c. Negative predictive value

When compared to the direct culture method (swabs directly streaked on selective chromogenic agar plates with cefoxitin without TSB enrichment and incubated for 24–48 hours at 35 ±2 °C), Xpert MRSA identified 94.3% of the specimens positive for MRSA and 93.2% of the specimens negative for MRSA; the positive predictive value was 73.0% and the negative predictive value was 98.8%.

Table 3. Xpert MRSA Compared to Direct Culture Method

		Culture				
		+	-			
Xpert MRSA	+	165	61	226	Positive Agreement:	94.3%
	-	10	838	848	Negative Agreement:	93.2%
	Total	175	899	1074	PPV[a]:	73.0%
					NPV:[b]	98.8%

a. Positive predictive value

b. Negative predictive value

The following tables show the performance of Xpert MRSA and MRSA prevalence at each clinical site compared to the reference culture and direct culture methods.

Table 4. Performance of Xpert MRSA by Site Compared to Reference Culture Method

Site	MRSA prevalence[a]	Positive Agreement (n)(95% CI)[b]	Negative Agreement (n)(95% CI)[c]	No. of indeterminate results
1	20.2% (78/387)	87.2% (n=78) (77.7-93.7%)	93.9% (n=309) (90.6-96.3%)	10
2	5.2% (3/58)	100.0% (n=3) (29.2-100.0%)	98.2% (n=55) (90.3-100.0%)	3
3	44.4% (12/27)	91.7% (n=12) (61.5-99.8%)	100.0% (n=15) (78.2-100.0%)	3
4	12.3% (20/162)	80.0% (n=20) (56.3-94.3%)	97.2% (n=142) (92.9-99.2%)	9
5	20.5% (46/224)	89.1% (n=46) (76.4-96.4%)	94.9% (n=178) (90.6-97.7%)	1
6	22.3% (42/188)	81.0% (n=42) (65.9-91.4%)	93.2% (n=146) (87.8-96.7%)	6
7	35.7% (10/28)	90.0% (n=10) (55.5-99.8%)	94.4% (n=18) (72.7-99.9%)	2
Total	**19.6% (211/1074)**	**86.3% (n=211) (80.9-90.6%)**	**94.9% (n=863) (93.2-96.3%)**	**34**

a. Determined from results by reference culture method

b. Number of positive determined by reference culture method

c. Number of negative determined by reference culture method

Table 5. Performance of Xpert MRSA by Site—Comparison to Direct Culture Method

Site	Positive Agreement	Negative Agreement
1	95.4% (87.1-99.0%)	92.2% (88.8-94.9%)
2	100.0% (29.2-100.0%)	98.2% (90.3-100.0%)
3	91.7% (61.5-99.8%)	100.0% (78.2-100.0%)
4	81.3% (54.4-96.0%)	95.2% (90.4-98.1%)
5	94.9% (82.7-99.4%)	93.0% (88.3-96.2%)
6	97.1% (84.7-99.9%)	92.9% (87.6-96.4%)
7	100.0% (54.1-100.0%)	81.8% (59.7-94.8%)
Total	**94.3% (89.7-97.2%)**	**93.2% (91.4-94.8%)**

Performances of Xpert MRSA, the 2nd FDA-cleared NAAT and direct culture method from individual sites relative to the reference culture method are presented in the tables below.

Table 6. Results from Xpert MRSA, Direct Culture Method and Second FDA-cleared NAAT Test with Specimens Positive for MRSA by Reference Culture Method

Positive Agreement (95% CI)			
Site	**Xpert MRSA**	**2nd NAAT**	**Direct Culture[a]**
1	87.2% (77.7-93.7%)	80.8% (70.3-88.8%)	83.3% (73.2-90.8%)
2	100.0% (29.2-100.0%)	100.0% (29.2-100.0%)	100.0% (29.2-100.0%)
3	91.7% (61.5.-99.8%)	83.3% (51.6-97.9%)	100.0% (73.5-100.0%)
4	80.0% (56.3-94.3%)	78.9% (54.4-93.9%)	80.0% (56.3-94.3%)
5	89.1% (76.4-96.4%)	89.1% (76.4-96.4%)	84.8% (71.1-93.7%)
6	81.0% (65.9-91.4%)	78.6% (63.2-89.7%)	81.0% (65.9-91.4%)
7	90.0% (55.5-99.7%)	100.0% (69.2-100.0%)	60.0% (26.2-87.8%)
Total	**86.3% (80.9-90.6%)**	**83.3% (77.6-88.1%)**	**82.9% (77.2-87.8%)**

a. Swabs directly streaked on selective chromogenic agar plates with cefoxitin and incubated for 24-48hours at 35 ±2 °C.

Table 7. Results from Xpert MRSA, Direct Culture Method and Second FDA-cleared NAAT Test with Specimens Negative for MRSA by Reference Culture Method

Negative Agreement (95% CI)			
Site	Xpert MRSA	2nd NAAT	Direct Culture[a]
1	93.9% (90.6-96.3%)	92.2% (88.7-95.0%)	100.0% (98.8-100.0%)
2	98.2% (90.3-100.0%)	98.2% (90.3-100.0%)	100.0% (93.6-100.0%)
3	100.0% (78.2-100.0%)	100.0% (79.4-100.0%)	100.0% (79.4-100.0%)
4	97.2% (92.9-99.2%)	97.9% (93.9-99.6%)	100.0% (97.5-100.0%)
5	94.9% (90.6-97.7%)	93.8% (89.2-96.9%)	100.0% (97.9-100.0%)
6	93.2% (87.8-96.7%)	94.5% (89.5-97.6%)	100.0% (97.5-100.0%)
7	94.4% (72.7-99.9%)	94.4% (72.7-99.9%)	100.0% (81.5-100.0%)
Total	**94.9% (93.2-96.3%)**	**94.4% (92.7-95.9%)**	**100.0% (99.6-100.0%)**

a. Swabs directly streaked on selective chromogenic agar plates with cefoxitin and incubated for 24-48hours at 35 ±2 °C.

Analytical Performance

Analytical Sensitivity

The analytical sensitivity of the Xpert MRSA was determined using 6 strains of MRSA representing the six SCCmec types and subtypes (I, II, III, IV, IVa and V). Cultures of these strains were quantified then diluted to values spanning the range of 10 to 1000 colony forming units (CFU) per swab. All dilutions were tested in replicates of 4. Limit of detection obtained for each type or subtype tested shows the lowest number of CFU/swab at which all 4 replicates were reported positive. All of the strains representing the SCCmec cassette types I - V were detected by the Xpert MRSA test.

Table 8. Detection of SCCmec Types

SCCmec	(CFU/swab)
type I	10
type II	10
type III	10
type V	10
type IV	50
type IVa	100

Additional studies using type II cells were performed to determine the 95% confidence interval for the analytical limit of detection (LOD) of this test. The limit of detection is defined as the lowest number of MRSA colony forming units (CFU) per swab that can be reproducibly distinguished from negative samples with 95% confidence. Results indicate that the Xpert MRSA test will produce a positive result with 95% confidence for a swab containing 80 CFU.

Analytical Specificity

Cultures from 51 American Type Culture Collection (ATCC) and Network on Antimicrobial Resistance in

Staphylococcus aureus (NARSA) strains representing species phylogenetically related to *S. aureus* and members of the nasal commensal flora, 32 strains of methicillin-sensitive coagulase negative staphylococci, and 12 strains of methicillin-resistant coagulase-negative staphylococci were tested. Three replicates of each isolate were tested at =1 × 10^6 CFU/swab. None of the isolates were detected by the test. The specificity was 100%.

Interfering Substances

Potentially interfering substances evaluated include blood, mucus, and nasal sprays used to relieve decongestion, nasal dryness, or irritation. The presence of these substances did not significantly inhibit PCR and did not give invalid or erroneous results.

In the investigational study for Xpert MRSA test, potential interfering substances (blood, mucus or both) were reported on 45 of 1077 (4.2%) nasal swab specimens. Of the 31 specimens that gave an equivocal result on initial testing, three specimens had mucus and one specimen had blood on the swab. Three of the four specimens gave a result on retesting while one that contained mucus remained indeterminate.

Reproducibility

A panel of specimens with varying concentrations of MRSA and methicillin-susceptible *Staphylococcus epidermidis* (*mecA*-negative)were tested in triplicate on 10 different days at each of the three sites (4 specimens × 3 times /day × 10 days × 3 sites). One lot of Xpert MRSA kit was used at each of the 3 testing sites. Xpert MRSA tests were performed according to the Xpert MRSA procedure.

Table 9. Summary of Reproducibility Results

Specimen ID	MRSA in CFU/ swab	MSSE CFU/ swab	Site 1	Site 2	Site 3	Total Agreement	% Total Agreement
Negative	0	2.6 × 10^6	30/30	30/30	30/31[a]	90/91	98.9%
Weak positive	117	2.6 × 10^6	30/30	30/30	27/29 [a]	87/89	97.8%
Positive	800	2.6 × 10^6	30/30	30/30	30/30	90/90	100.0%
Strong positive	2.6 × 10^4	2.6 × 10^6	30/30	30/30	30/30	90/90	100.0%
Total Agreement			90% 120/ 120	120/ 120	117/ 120	357/360	99.2%
% Agreement			100.0%	100.0%	97.5%		

a. Xpert MRSA assay was inadvertently performed on one additional negative specimen and one less weak positive specimen

Appendix

Bibliography

1. Mainous AG, Hueston WJ, Everett, et al. 2006. Nasal Carriage of Staphylococcus aureus and Methicillin-Resistant S aureus in the United States, 2001-2002. An Family Medicine. 4 (2):132-137.
2. National Nosocomial Infections Surveillance (NNIS) System Report, data summary from January 1992 through June 2004, issued October 2004. Am J Infect Control 2004;32:470-85.
3. Chaix C, Durand-Zileski I, Alberti C, Buisson B. 1999. Control of Endemic Methicillin Resistant Staphylococcus aureus. JAMA 282 (19):1745-51.
4. Shopsin B, Kreiswirth BN. 2001. Molecular Epidemiology of Methicillin-Resistant Staphylococcus aureus. Emerging Infectious Diseases 7(2) 323-6.
5. Salgado CD et al. 2003. Community-Acquired Methicillin-Resistant Staphylococcus aureus: A Meta-analysis of Prevalence and Risk Factors. CID 36:131.
6. Centers for Disease Control and Prevention. Biosafety in microbiological and biomedical laboratories. Richmond JY and McKinney RW (eds) (1993). HHS Publication number (CDC) 93-8395.
7. Clinical and Laboratory Standards Institute. Protection of laboratory workers from occupationally acquired infections; Approved Guideline. Document M29 (refer to latest edition).
8. REGULATION (EO) No 1272/2008 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 16 December 2008 on the classification labeling and packaging of substances and mixtures amending and repealing. List of Precautionary Statements, Directives 67/548/EEC and 1999/EC (amending Regulations (EO) No 1907/2007)
9. Occupational Safety and Health Standards, Hazard Communication, Toxic and Hazard Substances (March 26, 2012) (29 C.F.R, pt. 1910, subpt. Z).

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Table of Symbols

Symbol	Meaning
REF	Catalog number
IVD	*In vitro* diagnostic medical device
	Do not reuse
LOT	Batch code

Symbol	Meaning
	Consult instructions for use
	Caution
	Manufacturer
	Country of manufacture
	Contains sufficient for *n* tests
CONTROL	Control
	Expiration date
	Temperature limitation
	Biological risks
	Warning
Rx only	For prescription use only

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IVD

Revision History

Description of Changes: 303-0142 Rev. A. to B

Purpose: Correction

Section	Description of Change
Materials Available but Not Provided	Removed KWIK-STIKS catalog #0360MSSA.