510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION MEMORANDUM
ASSAY AND INSTRUMENT COMBINATION TEMPLATE

A. 510(k) Number:

K142721

B. Purpose for Submission:

To obtain a Substantial Equivalence Determination for the cobas® MRSA/SA Test.

C. Measurand:

Target DNA sequences for:
1. SCC*mec*/*orfX* junction area of methicillin-resistant *Staphylococcus aureus* (i.e., MREJ for SCC*mec* Right Extremity Junction).
2. . capsular polysaccharide enzyme CAP5N (*cpe*) gene of *Staphylococcus aureus*.

D. Type of Test:

Qualitative real-time polymerase chain reaction (PCR) assay for the amplification and detection of methicillin-resistant *Staphylococcus aureus* (MRSA) DNA and *Staphylococcus aureus* (SA) DNA.

E. Applicant:

Roche Molecular Systems, Inc.

F. Proprietary and Established Names:

cobas® MRSA/SA Test

G. Regulatory Information:

1. Regulation section:

   21 CFR 866.1640 Antimicrobial susceptibility test powder

2. Classification:

   Class II

3. Product code:

NQX - System, nucleic acid amplification test, DNA, methicillin resistant *Staphylococcus aureus*, direct specimen

OOI – Real-time nucleic acid amplification system

4. Panel:

   83- Microbiology

H. Intended Use:

1. Intended use(s):

   The cobas® MRSA/SA Test on the cobas® 4800 system, is a qualitative in vitro diagnostic real-time PCR assay, for the direct detection of methicillin-resistant *Staphylococcus aureus* (MRSA) and *S. aureus* (SA) DNA from nasal swabs to aid in the prevention and control of MRSA and SA infections in healthcare settings. The cobas® MRSA/SA Test is not intended to diagnose, guide or monitor treatment for MRSA or SA infections, or provide results of susceptibility to methicillin. A negative result does not preclude MRSA/SA nasal colonization. Concomitant cultures are necessary to recover organisms for epidemiology typing or for further susceptibility testing.

2. Indication(s) for use:

   Same as the intended use.

3. Special conditions for use statement(s):

   For prescription use only.

4. Special instrument requirements:

   cobas® 4800 System

I. Device Description:

The cobas® 4800 System uses the cobas x 480 Instrument for sample preparation, and the cobas z 480 Analyzer for amplification and detection. Both the cobas x 480 Instrument and the cobas z 480 Analyzer are controlled by a computer workstation running the cobas® 4800 System Software.

The Roche Molecular Systems (RMS) cobas® MRSA/SA Test utilizes real-time polymerase chain reaction (PCR) for the detection of Methicillin resistant *Staphylococcus aureus* (MRSA) and *Staphylococcus aureus* (SA) DNA from nasal swab specimens collected in MSwab medium to aid in the prevention and control of MRSA and SA infections in healthcare settings.

The cobas® MRSA/SA Test contains two major processes: (1) automated sample preparation to extract nucleic acids from the nasal swab specimens; (2) PCR amplification of target DNA sequences using MRSA and SA specific primers, and real-time detection of cleaved fluorescent-labeled MRSA and SA specific oligonucleotide detection probes. An Internal Control (IC), containing unrelated randomized DNA sequence, is added to all samples prior to automated sample preparation and is amplified and detected simultaneously with each sample to monitor the entire process.

The MSwab Collection, Transport and Preservation System (Copan Flock Technologies S.r.l.) is used for specimen collection, transportation and storage of specimen for the cobas® MRSA/SA Test.

The cobas® MRSA/SA Test utilizes six reagent kits:

1. cobas® 4800 MRSA/SA Amplification/Detection Kit
2. cobas® 4800 MRSA/SA Controls and Cofactor Kit
3. cobas® 4800 System Wash Buffer Kit
4. cobas® 4800 System Lysis Kit 1
5. cobas® 4800 System Internal Control Kit 1
6. cobas® 4800 System Sample Preparation Kit

J. Substantial Equivalence Information:

1. Predicate device name(s):

   Lightcycler® MRSA Advanced Test

2. Predicate 510(k) number(s):

   k091409

3. Comparison with predicate:

Similarities		
Item	**Device**	**Predicate**
	cobas® MRSA/SA Test (k142721)	**Lightcycler® MRSA Advanced Test (k091409)**
Intended Use	The cobas® MRSA/SA Test on the cobas® 4800 system, is a qualitative in vitro diagnostic real-time PCR assay, for the direct detection of methicillin-resistant *Staphylococcus aureus* (MRSA) and *S.aureus* (SA) DNA from nasal swabs to aid in the	The LightCycler® MRSA Advanced Test is a qualitative in vitro diagnostic test for the direct detection of nasal colonization with methicillin-resistant (MRSA) to aid in the prevention and control of MRSA infections in healthcare settings. The test

Similarities		
Item	**Device**	**Predicate**
	cobas® MRSA/SA Test (k142721)	**Lightcycler® MRSA Advanced Test (k091409)**
	prevention and control of MRSA and SA infections in healthcare settings. The cobas® MRSA/SA Test is not intended to diagnose, guide or monitor treatment for MRSA or SA infections, or provide results of susceptibility to methicillin. A negative result does not preclude MRSA/SA nasal colonization. Concomitant cultures are necessary to recover organisms for epidemiology typing or for further susceptibility testing.	is performed on the LightCycler® 2.0 Instrument with nasal swab specimens from patients suspected of colonization, uses swab extraction and mechanical lysis for specimen preparation followed by polymerase chain reaction (PCR) for the amplification of MRSA DNA, and fluorogenic target specific hybridization probes for the detection of the amplified DNA. The LightCycler® MRSA Advanced Test is not intended to diagnose, guide or monitor treatment for MRSA infections. Concomitant cultures are necessary to recover organisms for epidemiology typing or for further susceptibility testing.
Sample Type	Nasal swab	Same
Amplification Technology	Real-time PCR	Same
Detection Mechanism	Paired target-specific hybridization probes using Förster resonance energy transfer (FRET)	Same
MRSA Analyte Target	SCC*mec* cassette Right Extremity (RE) junction of methicillin-resistant *Staphylococcus aureus* (MRSA)	Same
Sample Preparation Procedure	Semi-automated	Same

Differences		
Item	**Device**	**Predicate**
	cobas® MRSA/SA Test (k142721)	**Lightcycler® MRSA Advanced Test (k091409)**
SA Analyte Target	Capsular polysaccharide enzyme CAPN (CPE)	No SA target detected
Result Analysis	PCR cycle threshold (C_t) analysis	Melting peak analysis

K. Standard/Guidance Document Referenced (if applicable):

Draft Guidance for Industry and Food and Drug Administration Staff. Establishing the Performance Characteristics of Nucleic Acid-Based In Vitro Diagnostic Devices for the Detection and Differentiation of Methicillin-Resistant *Staphylococcus aureus* (MRSA) and *Staphylococcus aureus* (SA). Document issued on June 15, 2011.

L. Test Principle:

Sample preparation for the cobas® MRSA/SA Test is automated with the use of the cobas x 480 instrument. Organisms in nasal swab specimens collected in MSwab medium are lysed with chaotropic agent, proteinase K, and SDS reagents. Released nucleic acids, along with added Internal Control DNA, are bound by magnetic glass particles. They are washed and then eluted into a small volume of buffer. The instrument then takes an aliquot of the eluted material and sets up the PCR reaction with an activated Master Mix.

Target Selection

The well-characterized Right Extremity (RE) junction between the Staphylococcus aureus orfX gene and SCCmec cassette carrying the mecA drug-resistant gene was chosen to be the target to specifically detect MRSA. The capsular polysaccharide enzyme CAP5N (CPE) gene was chosen as the target for Staphylococcus aureus identification. This gene is conserved in Staphylococcus aureus and present in both methicillin resistant and methicillin sensitive organisms. Internal Control (IC) DNA sequence is unique and unrelated to either MRSA or SA target sequences.

Target Amplification and Detection

The PCR cycling steps and detection of target signal occurs in the cobas z 480 Analyzer. The Master Mix reagent contains primer pairs and probes for orfX (MRSA), CPE (SA) and IC targets. If the target nucleic acid sequences are present, amplification with the corresponding primers will occur by a thermostable DNA polymerase, generating PCR products (amplicons). These products are detected by specific TaqMan probes containing a fluorescent dye and a quencher. Normally, the quencher suppresses the fluorescence of the dye.

However, if the PCR product is present, the probe hybridizes to the product and gets cleaved by the 5' to 3' nuclease activity of the polymerase. This reaction allows the fluorescence to be emitted from the dye, and the signal is recorded in real time during each PCR cycle by the cobas z 480 analyzer. The signal is interpreted by the cobas® 4800 System Software and reported as final results.

M. Performance Characteristics (if/when applicable):

1. Analytical performance:

a. Precision/Reproducibility:

Precision Study
Within-laboratory precision was evaluated for the cobas® MRSA/SA Test at one (1) site with four (4) panel members:
1) Negative,
2) High Negative (HN), < 1.0 x LoD,
3) Low Positive (LP), 1 x LoD and
4) Moderate Positive (MP), 3.0 x LoD.

The study was conducted over twelve (12) days using three (3) reagent lots of two (2) kit sizes on three (3) cobas® 4800 systems. Each run contained two (2) replicates of each of the panel members (12 days x 3 lots x 2 replicates = 72 replicates/panel member/strain).

The results for the study are summarized in Table I, below. The upper limit of the 2-sided exact binomial 95% confidence interval (CI) of the detection for panel members at or near the limit of detection was 100% for MRSA and SA culture isolates. Positive rates for all other panel members were as expected.

Table I: Precision Study

Strain	Panel	MRSA- SA-	MRSA- SA+	MRSA+ SA+	% Detected	95% CI
No Analyte	Negative	72/72	0/72	0/72	0.0%	(0.0%, 5.0%)
MRSA 10364 (NARSA 384)	HN	0/72	15/72	57/72	79.2%	(68.0%, 87.8%)
	LP	0/72	0/72	72/72	100.0%	(95.0%, 100.0%)
	MP	0/72	0/72	72/72	100.0%	(95.0%, 100.0%)
MRSA 8065 (ATCC 43300)	HN	0/72	9/72	63/72	87.5%	(77.6%, 94.1%)
	LP	0/72	0/72	72/72	100.0%	(95.0%, 100.0%)
	MP	0/72	0/72	72/72	100.0%	(95.0%, 100.0%)
SA 10851 (NARSA 164)	HN	4/71*	67/71*	0/71*	94.4%	(86.2%, 98.4%)
	LP	0/72	72/72	0/72	100.0%	(95.0%, 100.0%)
	MP	0/72	72/72	0/72	100.0%	(95.0%, 100.0%)

*One (1) of seventy-two (72) samples tested could not be processed due to sample pipetting error on the instrument.

The overall coefficient of variation (CV%) of the C_t values for cobas® MRSA/SA Test was less than or equal to 1.3% indicating good reproducibility. C_t value variability of the cobas® MRSA/SA Test is mostly attributed to random factors with contributions of reagent lot, kit size, run/day and instrument/operator being substantially (several-fold) lower or negligible across the detected targets.

The precision study results met the pre-defined acceptance criteria for LP, MP, and HN samples. The cobas® MRSA/SA Test reproducibly detected both targets at concentrations around and above the LOD of the test. Reproducibility has been demonstrated across multiple runs/days, reagent lots, kit sizes and instruments/operators.

The results of this study are acceptable and these results are described in labeling.

Reproducibility Study

The reproducibility of the cobas® MRSA/SA Test on the cobas 4800 System was established in a multi-center study. The same strains and panel members were tested as listed in the precision study above (Table I). Panels were tested at three (3) sites by two (2) operators per site with one (1) run per operator per day, for five (5) days per lot, over two (2) lots for a total of one-thousand eight hundred (1,800) tests (180 tests/panel member or 90 tests/panel member/lot). Overall, sixty (60) runs were performed per site, per condition and all were valid runs. There were no failed/invalid tests.

The MRSA/SA reproducibility test panels were prepared by seeding MRSA strains NRS384 (MRSA-384) and ATCC 43300 (MRSA-43300), or SA strain RMSCC 10851 into contrived sample matrix (simulated clinical MSwab nasal specimens with mucin and human epithelial cells) at 1 of 3 concentrations (<1 x LoD, 1 × LoD, and 3 × LoD); a MRSA/SA-negative panel member was included as a panel member control. In all, there were 10 members per test panel with 3 replicates per panel member included in each run. The results of the reproducibility study are summarized in Table II below.

Table II: Reproducibility Study

Panel Member	Ct			Percent Agreement by Site (n/N)			Total Agreement	
	Mean	SD	CV (%)	1	2	3	Percent (n/N)	(95% CI)
Negative	N/A	N/A	N/A	100.0 (120/120)	100.0 (120/120)	100.0 (120/120)	100.0% (360/360)	(98.0%, 100.0%)
HN NARSA-384	40.3	0.43	1.1	95.0 (57/60)	83.3 (50/60)	78.3 (47/60)	85.6% (154/180)	(79.6%, 90.3%)
LP NARSA-384	38.0	0.49	1.3	100.0 (60/60)	100.0 (60/60)	100.0 (60/60)	100.0% (180/180)	(98.0%, 100.0%)
MP NARSA-384	36.3	0.44	1.2	100.0 (60/60)	100.0 (60/60)	100.0 (60/60)	100.0% (180/180)	(98.0%, 100.0%)

Table II: Reproducibility Study								
Panel Member	**Ct**			**Percent Agreement by Site (n/N)**			**Total Agreement**	
	Mean	**SD**	**CV (%)**	**1**	**2**	**3**	**Percent (n/N)**	**(95% CI)**
HN ATCC-43300	40.4	0.40	1.0	91.7 (55/60)	81.7 (49/60)	88.3 (53/60)	87.2% (157/180)	(81.4%, 91.7%)
LP ATCC-43300	38.9	0.45	1.1	100.0 (60/60)	100.0 (60/60)	100.0 (60/60)	100.0% (180/180)	(98.0%, 100.0%)
MP ATCC-43300	37.4	0.51	1.4	100.0 (60/60)	100.0 (60/60)	100.0 (60/60)	100.0% (180/180)	(98.0%, 100.0%)
HN SA (NARSA 164)	38.6	0.46	1.2	23.3 (14/60)	60.0 (36/60)	66.7 (40/60)	50.0% (90/180)	(42.5%, 57.5%)
LP SA (NARSA 164)	36.8	0.49	1.3	100.0 (60/60)	98.3 (59/60)	100.0 (60/60)	99.4% (179/180)	(96.9%, 100.0%)
MP SA (NARSA 164)	35.1	0.38	1.1	100.0 (60/60)	100.0 (60/60)	100.0 (60/60)	100.0% (180/180)	(98.0%, 100.0%)

These results are acceptable and they are described in labeling.

b. Linearity/assay reportable range:

Not applicable.

c. Traceability, Stability, Expected values (controls, calibrators, or methods):

Controls:
One set of cobas® MRSA/SA Test Positive and Negative Controls are included in each run. For any run, valid results must be obtained for both the Positive and Negative Control for the cobas® 4800 Software to display the reportable cobas® MRSA/SA Test results from that run.

Internal Control
The Internal Control is a lambda phage molecule that contains randomized sequences and targets for internal control-specific primers and probe. The Internal Control is added to all specimens and the Positive and Negative Controls during sample preparation on the cobas x 480 instrument. The Internal Control monitors nucleic acid amplification and detection steps for a given specimen. The Internal Control is also required for validation of the run controls.

External Controls:
Whole-cell positive external controls are not provided with this assay to control for lysis. However, commercially available control materials, MSSA strain ATTC 29213 and MRSA strain ATTC 43300, can be used as positive controls, in addition to the

other controls provided. During the clinical trial testing was conducted using the external Positive and Negative Controls listed below.

One set of cobas® MRSA/SA Test Positive and Negative Controls are included in each run. For any run, valid results must be obtained for both the Positive and Negative Control for the cobas® 4800 Software to display the reportable cobas® MRSA/SA Test results from that run.

Positive Control
The MRSA (+) Control which is provided with this assay contains non-infectious DNA plasmids of both MRSA and *Staphylococcus aureus*. The MRSA/SA (+) Control monitors the nucleic acid amplification, and detection steps in a given run of the test. The MRSA/SA positive control result must be 'Valid'.

Negative Control
The negative control, which is provided with this assay, contains Poly rA RNA. The negative control result must be 'Valid'.

During the clinical study, the positive and negative external control isolates were tested each day of testing. All MRSA/SA positive controls were detected accurately (100%, 116/116). All negative controls were detected accurately (100%, 116/116).

Kit stability:
Many components of assay stability were previously assessed during previous submissions for the cobas HSV 1 and 2, C. diff and CT/NG tests, as components of the kit are identical.

Three production lots of new reagent components of the kits required for cobas® MRSA/SA testing were entered into the stability program to establish shelf life. The expiration date for each test kit was defined by the shortest dated component in that kit. These reagents include cobas® MRSA/SA Test specific reagent kits: the cobas® MRSA/SA Amplification/Detection Kit, the cobas® MRSA/SA Controls & Cofactor Kit; and the cobas® 4800 IC Kit 1, and Lysis Buffer 1 from the cobas® 4800 System Lysis Kit 1. Stability testing of the MRSA/SA specific reagents was carried out using the cobas® MRSA/SA Test. Stability of the cobas® 4800 IC Kit 1 was carried out using cobas® MRSA/SA Test, and stability of Lysis Buffer 1 was assessed using chemical/analytical tests.

The stability of these existing generic reagents demonstrated stability from 0-25 months in real-time experiments with the exception of Lysis Buffer-1, as was previously established. To date, the 2-8 ºC storage real time testing stability results for three (3) lots of cobas® 4800 System Lysis Buffer-1 supports a shelf life of fifteen (15) months. Testing will continue, and the initial dating will be extended when data becomes available. Accelerated stability (25 ºC storage) testing results predicts a shelf life of at least 19 months for cobas® 4800 System Lysis Buffer-1 at the storage temperature of 2-8ºC.

Specimen stability:
The study was performed to determine the stability of MRSA and SA nasal swab specimens in MSwab medium (Copan MSwab™ Collection, Preservation and Transport System) after storage under various storage conditions for use with the cobas® MRSA/SA Test.

A total of twenty-six (26) nasal swab specimens (12 MRSA, 10 SA, 2 MRSA/SA and 2 negative) were included in the stability testing. Of these twenty-six (26) samples, six (6) MRSA positive, six (6) SA positive and two (2) MRSA/SA negative samples were prepared by pooling clinical specimens to create enough volume to test at all scheduled time-points. The remaining samples were prepared by spiking MRSA culture isolates (10364 or 8065) and/or SA culture isolate (10851) at 3 x LoD into negative clinical specimen pool.

All positive and negative specimens generated positive and negative results, respectively, at all time-points of up to 40 days of cumulative storage.

The results indicated that nasal clinical specimens collected in Mswab medium were stable for:

1. up to 2 days at room temperature (15-30 °C) followed by 5 days stored at 2-8 °C;
2. up to 4 days at room temperature (15-30 °C) followed by 5 days stored at 2-8 °C

followed by 1 month frozen at -20 °C before testing with the cobas® MRSA/SA Test.

There is at least a two (2) hour interval from the completion of sample preparation and amplification/detection plate setup to the start of amplification/detection on the cobas® z480 Analyzer for the cobas® MRSA/SA Test.

d. Detection limit:

The limit of detection (LoD) of the cobas® MRSA/SA Test was determined using contrived stocks of one *Staphylococcus aureus* (SA) and two methicillin resistant *Staphylococcus aureus* (MRSA) strains per CLSI guideline EP17-A2.

The titer of each culture isolates was determined by counting colonies from plates of serial dilutions. Each of the culture isolates was diluted in collection and transport medium, then spiked into a flocked swab, which was then immersed in simulated nasal swab matrix to produce a panel consisting of nine (9) concentration levels and one (1) negative level (simulated matrix only). The swab type and medium used for these assays are components of the Copan MSwab Collection, Transport and Preservation System designated in the labeling for use with the cobas® MRSA/SA Test.
Three lots of reagents and two different batch sizes were used for this study. For each reagent lot, a minimum of three independent panel dilutions were prepared. A minimum of 19 valid replicates per testing level were generated from multiple

instruments and runs for each reagent lot, yielding a minimum of 61 valid replicates for each testing level with all three lots of reagents.

The LoD of the cobas® MRSA/SA Test is the concentration where at least 95% observed positive rate for which all higher concentrations had an observed positive rate above 95%. The LoD study results are shown in Table III below.

Table III: LoD for MRSA and SA				
Strain	**SCC*mec* Type**	***spa* Type**	**PFGE Type**	**CFU/ml**
MRSA 10364 (NARSA 384)	IVa	t008	USA300-0114	1.53×10^5
MRSA 8065 (ATCC 43300)	II	t007	Sac-15	1.23×10^6
SA (NARSA 164)	N/A	t084	N/A	1.71×10^5

These study results are acceptable and they are described in labeling.

e. Analytical reactivity:

The analytical inclusivity of the cobas® MRSA/SA Test was determined for 35 additional MRSA culture isolates and 5 additional SA culture isolates. The panel included a variety of strains, taking into account geographic origin, SCC*mec* type, RE type, SPA type, and Pulsed-Field Gel Electrophoresis (PFGE) type. The panels of at least three concentrations per isolate were used to determine the LoD for each strain in simulated nasal swab matrix. The LoD was calculated as the lowest concentration level with ≥ 95 % positive rate for which all higher concentration levels show ≥ 95 % positive rate.

For the five (5) SA isolates (Table IV) and thirty-five (35) MRSA isolates (Table V) tested the LoDs range from 1.30×10^5 and 1.23×10^6 CFU/ml. The cobas MRSA/SA Test detected the right extremity (RE) types 1, 2, 3, 4, 6, 9, 11, 14, 24 and 25. The cobas® MRSA/SA Test detected MRSA staphylococcal cassette chromosome *mec* (SCC*mec*) types I, II, III, IV, V, VI and VIII, as well as MRSA pulsed-field gel electrophoresis (PFGE) types USA 100 to 1000.

Table IV: Analytical Reactivity (SA)	
SA Isolate #	***spa* Type**
1	t238
2	t018
3	t008
4	t002
5	t088

Table V: Analytical Reactivity (MRSA)				
MRSA Isolate #	**RE Type**	**SCC*mec* Type**	***spa* Type**	**PFGE Type**
1	11	new	t002	Unknown
2	6	II	t242	Unknown
3	9/11	new	t024	Unknown
4	14	Unknown	Unknown	Unknown
5	25	Unknown	t003	Unknown
6	6	II	t216	USA100
7	2	IV	t008	USA300
8	2	II	t037	USA200
9	2	IV	t1578	USA300
10	2	II	t002	USA100
11	2	IV	t008	USA800
12	2	IV	t008	USA300
13	2	IV	t064	USA500
14	2	IV	t148	USA700
15	2	IV	t688	USA800
16	2	IV	t688	USA300
17	2	II	t042	USA100
18	2	II	t018	USA200
19	2	IV	t008	USA300
20	2	IV	t008	USA300
21	2	IV	t5576	USA800
22	2	II	t004	USA600
23	2	IV	t216	USA1000
24	2	IV	t064	Iberian
25	2	II	t266	USA600
26	2	IV	t008	USA300
27	2	IV	t008	USA300
28	2	IV	t002	USA800
29	3	V	t242	USA1000
30	24	new	t476	Unknown
31	1	I	t149	Unknown
32	3	VIII	Unknown	Unknown
33	4	IV	Unknown	Unknown
34	2	III	t030	Unknown
35	25	VI	Unknown	Unknown

In addition to thirty-seven (37) MRSA isolates and six (6) SA isolates tested in the LoD and analytical sensitivity studies shown above, two hundred and eighty-one (281) MRSA isolates from locations across Europe, United States, Japan and Australia and eighty-five (85) SA isolates from locations across the United States were tested. The MRSA collection contained MRSA isolates of different SCC*mec* types (I, II, III, IV, IVa, V, VI, VII, and new), and seventy-one (71) *spa* types. Of the eighty-five (85) SA isolates from the United States 75 *spa* types were represented. All of the SA isolates were detected by the cobas® MRSA/SA Test. Of the two hundred

eighty-one (281) MRSA isolates, two hundred seventy-seven (277, 98.6%) of the MRSA isolates were detected. Sequencing results suggest that the target regions for the four (4) MRSA isolates not detected by the cobas® MRSA/SA Test contained sequences not recognized by the primers and probes in the assay. One of the four isolates was a *mecA*$_{LGA251}$ strain (also known as *mecC*). All other results were as expected.

These study results are acceptable and they are described in labeling.

f. Analytical specificity:

Microbial cross-reactivity and interference:
The cobas® MRSA/SA Test was examined for analytical specificity by testing non-MRSA/SA microorganisms, including coagulase-negative (CoNS) and methicillin-resistant CoNS (MR-CoNS) in negative nasal matrix in the presence or absence of MRSA and SA targets.

Organisms were spiked at high concentration into the simulated nasal swab matrix, without (cross-reactivity) or with the targets (interference) at approximately 3 x LoD. All bacteria and fungi were tested at 1.00 x 10^6 CFU/ml. All viruses except for Adenovirus type 1 (1.00 x 10^4 PFU/ml) were tested at 1.00 x 10^5 PFU/ml. The same two MRSA and one SA isolates used in the LoD study were used. The list of strains used for the cross-reactivity and microbial interference studies is listed below in Tables VI and VII.

Table VI: Staphylococcal Strains Included in Analytical Cross-Reactivity and Microbial Interference

	Organisms	**Strain ID**
1	*Staphylococcus arlettae*	ATCC43957
2	*Staphylococcus auricularis* *(Methicillin-resistant)*	ATCC33753
3	*Staphylococcus caprae* *(Methicillin-resistant)*	ATCC35538
4	*Staphylococcus captis*	ATCC35661
5	*Staphylococcus carnosus*	ATCC51365
6	*Staphylococcus chromogenes*	ATCC43764
7	*Staphylococcus cohnii*	ATCC35662
8	*Staphylococcus delphini*	MayoClinicH18859
9	*Staphylococcus epidermidis* *(Methicillin-resistant)*	ATCC14990
10	*Staphylococcus epidermidis* *(Methicillin-resistant)*	ATCC35547
11	*Staphylococcus epidermidis*	ATCC35983

Table VI: Staphylococcal Strains Included in Analytical Cross-Reactivity and Microbial Interference		
	Organisms	**Strain ID**
	(Methicillin-resistant)	
12	*Staphylococcus epidermidis* *(Methicillin-resistant)*	ATCC35984
13	*Staphylococcus epidermidis* *(Methicillin-resistant)*	ATCC51624
14	*Staphylococcus epidermidis* *(Methicillin-resistant)*	ATCC51625
15	*Staphylococcus epidermidis*	ATCC700583
16	*Staphylococcus epidermidis* *(Methicillin-resistant)*	ATCC27676
17	*Staphylococcus equorum*	ATCC43958
18	*Staphylococcus felis*	ATCC49168
19	*Staphylococcus gallinarum*	ATCC35539
20	*Staphylococcus haemolyticus* *(Methicillin-resistant)*	ATCC29968
21	*Staphylococcus haemolyticus*	ATCC29970
22	*Staphylococcus haemolyticus*	ATCC43252
23	*Staphylococcus hominis*	ATCC25615
24	*Staphylococcus hominis*	ATCC35982
25	*Staphylococcus hominis subsp. Hominis*	ATCC27844
26	*Staphylococcus hominis subsp. Hominis*	ATCC27845
27	*Staphylococcus intermedius*	ATCC29663
28	*Staphylococcus kloosii*	ATCC43959
29	*Staphylococcus lentus*	ATCC29070
30	*Staphylococcus lugdunensis*	ATCC49576
31	*Staphylococcus pasteuri*	ATCC51129
32	*Staphylococcus pseudointermedius*	DSMZ21284**
33	*Staphylococcus pulvereri*	ATCC51698
34	*Staphylococcus saprophyticus*	ATCC15305
35	*Staphylococcus schleiferi* *(subspecies coagulans)*	ATCC43808
36	*Staphylococcus sciuri*	ATCC49575
37	*Staphylococcus simulans* *(Methicillin-resistant)*	ATCC27848
38	*Staphylococcus simulans*	ATCC11631

Table VI: Staphylococcal Strains Included in Analytical Cross-Reactivity and Microbial Interference		
	Organisms	**Strain ID**
39	*Staphylococcus warneri*	ATCC27836
40	*Staphylococcus warneri* *(Methicillin-resistant)*	ATCC27839
41	*Staphylococcus warneri*	RMSCC 1224
42	*Staphylococcus xylosus*	ATCC35663
43	*Staphylococcus xylosus*	ATCC29971

Table VII: Staphylococcal Strains Included in Analytical Cross-Reactivity and Microbial Interference		
	Organisms	**Strain ID**
1	*Acinetobacter baumannii*	ATCC19606
2	*Acinetobacter haemolyticus*	ATCC17906
3	*Bacillus cereus*	ATCC13472
4	*Bordetella bronchioseptica*	ATCC19395
5	*Bordetella parapertussis*	ATCC15311
6	*Bordetella pertussis*	ATCC9797
7	*Burkholderia cepacia*	ATCC25416
8	*Candida albicans*	ATCC10231
9	*Candida glabrata*	ATCC2001
10	*Candida parapsilosis*	ATCC22019
11	*Candida tropicalis*	ATCC750
12	*Chlamydia pneumoniae*	CDC-CWL-011 strain
13	*Citrobacter freundii*	ATCC8090
14	*Citrobacter koseri*	ATCC27028
15	*Corynebacterium amycolatum*	ATCC49368
16	*Corynebacterium bovis*	ATCC7715
17	*Corynebacterium flavescens*	ATCC10340
18	*Corynebacterium genitalium*	ATCC33030
19	*Corynebacterium glutamicum*	ATCC13032
20	*Corynebacterium jeikeium*	ATCC43734
21	*Cryptococcus neoformans*	ATCC32719
22	*Eikenella corrodens*	ATCC23834
23	*Enterobacter aerogenes*	ATCC13048
24	*Enterobacter cloacae*	ATCC13047
25	*Enterococcus flavescens*	ATCC49996
26	*Enterococcus gallinarum*	ATCC49573

Table VII: Staphylococcal Strains Included in Analytical Cross-Reactivity and Microbial Interference		
	Organisms	**Strain ID**
27	*Enterococcus hirae*	ATCC8043
28	*Escherichia coli*	ATCC11775
29	*Finegoldia magna*	RMSCC 974
30	*Haemophilus aphrophilus*	ATCC19415
31	*Haemophilus influenzae*	ATCC33391
32	*Haemophilus parainfluenzae*	ATCC33392
33	*Issatchenkia orientalis*	ATCC6258
34	*Klebsiella oxytoca*	ATCC33496
35	*Klebsiella pneumoniae (KPC producing)*	ATCC700603
36	*Klebsiella pneumoniae (KPC producing)*	ATCC BAA1900
37	*Lactobacillus crispatus*	ATCC33820
38	*Lactobacillus delbrueckii*	ATCC12315
39	*Legionella pneumophila*	ATCC33152
40	*Leifsonia aquatica (formerly Corynebacterium aquaticum)*	ATCC14665
41	*Listeria monocytogenes*	ATCC15313
42	*Microbacterium testaceum*	ATCC15829
43	*Micrococcus luteus*	ATCC4698
44	*Moraxella catarrhalis*	ATCC8176
45	*Mycobacterium tuberculosis avirulent*	ATCC25177
46	*Mycoplasma pneumoniae*	ATCC15531
47	*Mycoplasma salivarium*	ATCC23064
48	*Neisseria meningitidis*	ATCC13077
49	*Parvimonas micra*	ATCC33270
50	*Pasteurella aerogenes*	ATCC27883
51	*Planococcus maritimus*	RMSCC11454**
52	*Proteus mirabilis*	ATCC29906
53	*Proteus vulgaris*	RMSCC204**
54	*Providencia stuartii*	ATCC22914
55	*Pseudomonas aeruginosa*	ATCC33584
56	*Pseudomonas fluorescens*	ATCC11250
57	*Rhodococcus equi*	ATCC6939
58	*Rothia mucilaginosa*	ATCC25296
59	*Salmonella enterica subsp. Enterica (formerly Salmonella typhimurium)*	RMSCC374*
60	*Serratia marcescens*	ATCC8100

Table VII: Staphylococcal Strains Included in Analytical Cross-Reactivity and Microbial Interference		
	Organisms	**Strain ID**
61	*Shigella sonnei*	ATCC29930
62	*Streptococcus agalactiae*	RMSCC983**
63	*Streptococcus anginosus*	ATCC12395
64	*Streptococcus mitis*	ATCC33399
65	*Streptococcus mutans*	ATCC25175
66	*Streptococcus pneumoniae*	ATCC33400
67	*Streptococcus pyogenes*	ATCC12344
68	*Streptococcus salivarius*	ATCC7073
69	*Streptococcus sanguinis*	ATCC10556
70	*Streptococcus suis*	ATCC43765
71	*Yersinia enterocolitica*	ATCC9610
72	*Adenovirus Type 7*	VR-7
73	*Adenovirus Type 40*	Dugan (VR-40)
74	*Coronavirus 229E*	VR-740
75	*Coronavirus OC43*	VR-1558
76	*Cytomegalovirus*	AD-169 (VR-538)
77	*Epstein Barr Virus*	B95-8
78	*HSV 1*	MacIntyre (VR-539)
79	*Human Adenovirus type 1*	VR-1
80	*Human enterovirus 71*	VR-1775
81	*Human metapneumovirus*	Peru6-2003
82	*Influenza A/H1N1*	A/PR/8/34 (VR-95)
83	*Influenza A/H3N2 A/HongKong/8/68*	H3N2
84	*Influenza B*	N/A
85	*Measles virus*	N/A
86	*Mumps virus*	Enders (VR-106)
87	*Parainfluenza 1*	C-35 (VR-94)
88	*Parainfluenza 2*	Greer (VR-92)
89	*Parainfluenza 3*	C-243 (VR-93)
90	*Rhinovirus type 1A*	VR-1559
91	*RSV A*	VR-1540
92	*RSV B*	VR-1400
93	*HCT-15 cells (human genomic DNA)*	RMSCC3515

None of the organisms tested above cross-reacted or interfered with the assay at the concentrations tested.

These study results are acceptable and these results are described in labeling.

Co-infection Studies:
To test for competitive interference with the MRSA target, SA organisms including borderline oxacillin-resistant *S. aureus* (BORSA) strains and empty cassette variants (*mecA* drop-outs) were tested at high concentration to examine any cross-reactivity and/or interference with MRSA detection at 3 x LoD using the two (2) MRSA strains used in the LoD study. Both MRSA strains were correctly identified in every case. Ten (10) BORSA strains and sixteen (16) empty-cassette (*mecA* drop out) MRSA strains were also tested in the absence of MRSA. The cobas® MRSA/SA test correctly detected 2/2 MSSA, 10/10 BORSA and 13/16 *mecA* drop out isolates as positive for SA and negative for MRSA. Three (3) of the *mecA* drop out isolates were detected as positive for SA and MRSA. The risk of incorrect identification of MRSA from *mecA* drop-outs is mitigated in the labeling by requiring follow-up testing for results indicating the presence of MRSA. A limitation is also included in labeling indicating that false positive MRSA result may occur if an "empty cassette variant" *Staphylococcus aureus* is present.

Additionally, two (2) methicillin-sensitive (MSSA) and two (2) methicillin-resistant *Staphylococcus epidermidis* (MRSE) isolates were spiked at increasing concentrations to determine if there is any competitive inhibition effect on the detection of the MRSA and SA targets (SA strains were tested against MRSA targets only). In the presence of increasing concentration of SA or MRSE, low positive MRSA targets were consistently detected as positive. This demonstrates that there is no competitive inhibition by these organisms.

In consideration of microbial specificity overall, none of the 135 closely related *Staphylococcus* species or microorganisms commonly found in the nasal flora at tested levels yielded false positive or false negative results. The cobas® MRSA/SA Test detected all 10 BORSA isolates, 13 of 16 empty cassette variants and 2 SA isolates correctly as SA only. Increasing concentration of MSSA did not compete with the detection of low level MRSA targets and increasing concentrations of MRSE did not compete with the detection of MRSA and SA targets.

g. Interfering substances:

Twenty-seven (27) substances (Table VIII) that may potentially be present in the sampling area including human whole blood, mucin, nasal sprays, medications or moisturizers were evaluated in this study.

Table VIII: Interfering Substances
Substance
Whole blood
Mucin
Afrin Nasal Spray
Beconase Nasal Spray
Bepanthen® nasal ointment
Chloraseptic Max Sore Throat Lozenges
Fluticasone Propionate (50 mcg) Nasal Spray
FluMist© (Afluria, Influenza virus vaccine)
Flunisolide Nasal Solution USP, 0.025%
Mupirocin Ointment
Dristan™ Nasal Mist
Luffeel™
Triamcinolone Acetonide Nasal spray
NasalCrom Nasal Spray
Nasonex Nasal Spray
Neo–Synephrine
Otrivine Nasal Spray
Relenza®
Budesonide Inhalation Suspension 0.25 mg/2 ml
Azelastin HCL Nasal Solution
Equate Saline Nasal Moisturizing Spray
Rhinaris® Nasal gel
Tobramycin and Dexamethasone Ophthalmic Solution
Releev™ (for cold sores)
Zicam Nasal Gel
QVAR (40 mcg) Inhalation Aerosol
Nostrilla

For blood and mucin, three levels of each interferent were spiked into samples constructed with a simulated nasal swab matrix at 3 x LoD for reach strain used in the LoD studies. Interferents were added into the samples in the amounts corresponding to a percentage of volume/volume (v/v) or weight/volume (w/v) based on the maximum capacity of the flocked swab used with the cobas® MRSA/SA Test.

Ten (10) replicates were prepared this way for each interferent at each level, for each of the three (3) strains. Samples with interferents but no targets were also tested. At 100% v/v whole blood level and at 20% w/v mucin level, false negative results were observed for target positive samples, as listed in Table VI below.

For exogenous substance testing, substances were tested in concentrations specified per the manufacturer's labeling. Each product was spiked into simulated nasal swab matrix and tested in the absence or presence of MRSA and SA targets used in the LoD studies at approximately 3 x LoD. Each substance was introduced in the

amounts corresponding to 100% of the swab capacity, except for Relenza®, which was tested at the maximum prescription dosage. For substances that interfered with the cobas® MRSA/SA Test at initial testing concentration, lower concentrations were tested to identify the level that is tolerated by the test. Each condition was tested in triplicate.

The performance of the cobas® MRSA/SA Test was not affected by 23 out of 25 exogenous substances tested. Rhinaris® Nasal gel and Releev™ interfered with the performance of the cobas® MRSA/SA Test when present in the amounts above 15% (Rhinaris® Nasal gel) and above 25% (ReleevTM) of the swab capacity.

The minimum concentration that caused interference for each substance in the conditions tested us listed in Table IX below.

Table IX: Interfering Substances	
Strains	
Human Whole Blood	100% v/v
Type II Mucin	20% w/v
Rhinaris® Nasal gel	15% v/v
Releev™	25% v/v

These study results are acceptable. A limitation is included in labeling indicating that false negative or invalid results may occur due to interference from these various substances.

h. Carry-Over:

A cross-contamination study was conducted for the cobas® MRSA/SA Test. MRSA and negative samples were processed in a checkerboard configuration on the cobas® 4800 system. High titer samples were prepared by spiking MRSA culture to simulated nasal swab matrix to generate a cycle threshold (C_t) that exceeded 95% of signal from specimens of infected patients of the clinical specimen population.

Five (5) runs were performed on each of the three (3) cobas® 4800 systems (3 instruments x 5 runs = 15 runs total). The first run on each system contained only the negative samples to confirm the instrument was clean. The three (3) subsequent runs on each system had alternating positive and negative samples in checkerboard configurations to assess the cross contamination rate (3 instruments x 3 checkerboard runs = 9 checkerboard runs total). The last run on each system contained only the negative samples to assess the carry-over contamination rate.

There were no cross-contamination events in any of the nine checkerboard runs across three cobas® 4800 systems (a total of 423 MRSA negative samples) for an observed cross-contamination rate of 0%. All results in the last three (3) runs containing only the negative samples were negative, suggesting that there was no carry-over run-to-run contamination.

i. Assay cut-off:

The C_t cut-off values for the cobas® MRSA/SA Test was determined to achieve the balanced performance against the established reference culture method. The initial C_t cut-off values were determined using 533 nasal swab samples collected at two sites in the United States for a pre-clinical feasibility study. These samples were tested on the cobas® 4800 System using two lots of reagents. Direct and enrichment MRSA and SA culture procedures were also conducted on these specimens. Final combined direct and enrichment culture results were designated as reference culture results.

The initial cut-off values were further evaluated in an interim analysis of the United States clinical utility study data with first 1,643 subject results. The results of this analysis and final cut-off values are listed in Table X below.

Table X: Assay Cutoff

	Analyte		**Reference Culture**	
cobas® MRSA/SA Test	**SA (C_t 39.0)**		Positive	Negative
		Positive	402	67
		Negative	26	1148
				95% CI
		Sensitivity	93.9%	(91.2%-95.8%)
		Specificity	94.5%	(93.1%-95.6%)
		NPV	85.7%	(82.3%-88.6%)
		PPV	97.8%	(96.8%-98.5%)
	MRSA (C_t 41.0)		Positive	Negative
		Positive	94	39
		Negative	7	1503
				95% CI
		Sensitivity	93.1%	(86.4%-96.6%)
		Specificity	97.5%	(96.6%-98.1%)
		NPV	70.7%	(62.4%-77.7%)
		PPV	99.5%	(99.0%-99.8%)

The final C_t cut-off values were determined to be 39.0 for SA and 41.0 for MRSA in order to assure balanced performance of the cobas® MRSA/SA Test against the defined culture reference method.

The results of this study are acceptable.

2. Comparison studies:

a. Method comparison with predicate device:

Not applicable.

b. Matrix comparison:

All analytical studies were conducted in a simulated negative matrix. A comparison study was conducted between negative clinical matrix and a contrived negative matrix in order to validate the use of the contrived negative matrix in place of a clinical negative matrix for the analytical studies in section M1 above. Contrived negative matrix was constructed to mimic challenging clinical specimens, consisting of mucin, human cells and an isotonic (MSwab) media.

The performance results of the simulated matrix compared to the natural nasal swab specimen was demonstrated using two different MRSA isolates spiked into each matrix at 1 x LoD. The C_t values between the two matrices demonstrate that the simulated matrix is comparable to a natural specimen. All specimens were detected (100%). The results of this matrix comparison study are listed in Table XI below.

Table XI: Matrix Comparison Study

Panel ID	Target	Pooled Negative Clinical Matrix		Contrived Negative Matrix	
		Mean C_t	95% CI	Mean C_t	95% CI
MRSA 10364 (NARSA 384)	*orfX*	39.1	38.8 - 39.5	39.1	38.7 - 39.5
	IC	38	37.7 - 38.3	38.3	37.9 - 38.7
MRSA 10364 (NARSA 384)	*orfX*	39.2	38.9 - 39.5	38	37.6 - 38.4
	IC	38	37.6 - 38.4	38.1	37.7 - 38.5

The results of this study are acceptable.

3. Clinical studies:

This was a prospective multi-site study to evaluate the performance of the cobas® MRSA/SA Test compared with direct culture and enrichment culture in the screening of patients for nasal colonization with methicillin-resistant *Staphylococcus aureus* (MRSA) and *Staphylococcus aureus* (SA). Six (6) collection sites participated in this study and three (3) sites performed testing with the cobas® MRSA/SA Test. Reference testing using direct and enrichment culture was performed at a central reference laboratory that specializes in culture and molecular detection of MRSA and SA.

Swabs were collected from patients that met the inclusion criteria. Each subject had one (1) Standard-of-Care (SOC) swab collected, for SOC testing if applicable, and one (1) study swab collected using the Copan MSwab Collection, Transport and Preservation System. The study swab was processed using the cobas® MRSA/SA Test and used for direct and enrichment culture as the reference method.

On the day of collection, the swab was expressed into MSwab media and discarded. An aliquot of the eluted specimen was transported to a reference laboratory at 2-8^{o} C to be cultured within 48 hours of collection. The remainder of the swab-expressed media in the MSwab tube is tested with the cobas® MRSA/SA Test.

At the reference laboratory, the aliquoted swab-expressed media was divided into three (3) even fractions. Two fractions are applied to HardyCHROM™ MRSA, HhardyCHROM™ Staph aureus for direct culture. If growth was observed within twenty-four (24) hours, presumptive colonies were subcultured onto blood agar. Confirmed colonies are reported as MRSA or SA, as appropriate. The third fraction was incubated in tryptic soy broth (TSB) with 6.5% w/v sodium chloride for up to fourty-eight (48) hours, subcultured on blood agar then confirmed if direct culture results did not detect MRSA. Presumptive MRSA isolates from the MRSA plate were confirmed using the cefoxitin disk screening test for methicillin resistance.

Clinical performance was based on comparison of the cobas® MRSA/SA Test results to those obtained by a composite culture of directly plated patients' nasal swabs and culture of the transport fluid material at a central location.

a. Clinical Sensitivity:

Of the two thousand five hundred twenty-eight (2528) specimens collected (1 specimen collected/enrolled patient) in the clinical study, twelve (12) were excluded due to inclusion/exclusion criteria, withdrawal or consent errors. Twelve (12) were non-evaluable due to errors in sample handling or invalid cobas® MRSA/SA Test results. During culture enrichment, four (4) cultures were confounded due to *Proteus* overgrowth, excluding them from evaluation for enrichment. One of the four was positive for SA from direct culture evaluation.

Table XII below shows the comparison of the cobas® MRSA/SA Test with combined direct and enrichment culture results for the 2,500 evaluable specimens for MRSA and 2,501 evaluable specimens for SA. MRSA sensitivity and specificity relative to combined direct and enrichment culture were 93.1% and 97.5%, respectively. The overall PPV and NPV of the cobas® MRSA/SA Test for MRSA were 71.6% and 99.5%, respectively. Similarly, SA sensitivity and specificity relative to combined direct and enrichment culture were 93.9% and 94.2%, respectively. The overall PPV and NPV of the cobas® MRSA/SA Test for SA were 85.3% and 97.7%, respectively. Across test sites, MRSA sensitivity and specificity ranged from 91.4% to 94.5% and from 97.0% to 97.9%, respectively. The corresponding ranges for SA were from 91.6% to 96.1% and from 92.9% to 95.4%.

Table XII: Clinical Performance Data for the cobas® MRSA/SA Test vs. Direct and Enrichment Culture for SA and MRSA			
MRSA Overall			
cobas® MRSA/SA Test	**Direct and Enrichment Culture**		
	Positive	**Negative**	**Total**
Positive	149	59	208
Negative	11	2281	2292
Total	160	2340	2500
Sensitivity: 93.1% (149/160) 95% CI (88.1%-96.1%) **Specificity:** 97.5% (2281/2340) 95% CI (96.8%-98.0%)			
MRSA Site1			
cobas® MRSA/SA Test	**Direct and Enrichment Culture**		
	Positive	**Negative**	**Total**
Positive	53	21	74
Negative	5	963	968
Total	58	984	1042
Sensitivity: 91.4% (53/58) 95% CI (81.4%, 96.3%) **Specificity:** 97.9% (963/984) 95% CI (96.8%, 98.6%)			
MRSA Site 2			
cobas® MRSA/SA Test	**Direct and Enrichment Culture**		
	Positive	**Negative**	**Total**
Positive	52	21	73
Negative	3	768	771
Total	55	789	844
Sensitivity: 94.5% (52/55) 95% CI (85.1%, 98.1%) **Specificity:** 97.3% (768/789) 95% CI (96.0%, 98.3%)			
MRSA Site 3			
cobas® MRSA/SA Test	**Direct and Enrichment Culture**		
	Positive	**Negative**	**Total**
Positive	44	17	61
Negative	3	550	553
Total	47	567	614
Sensitivity: 93.6% (44/47) 95% CI (82.8%, 97.8%) **Specificity:** 97.0% (550/567) 95% CI (95.3%, 98.1%)			

SA Overall			
cobas® MRSA/SA Test	**Direct and Enrichment Culture**		
	Positive	**Negative**	**Total**
Positive	620	107	727
Negative	40	1734	1774
Total	660	1841	2501
Sensitivity: 93.9% (620/660) 95% CI (91.9%-95.5%) **Specificity:** 94.2% (1734/1841) 95% CI (93.0%-95.2%)			
SA Site 1			
cobas® MRSA/SA Test	**Direct and Enrichment Culture**		
	Positive	**Negative**	**Total**
Positive	246	55	301
Negative	17	725	742
Total	263	780	1043
Sensitivity: 93.5% (246/263) 95% CI (89.9%, 95.9%) **Specificity:** 92.9% (725/780) 95% CI (90.9%, 94.5%)			
SA Site 2			
cobas® MRSA/SA Test	**Direct and Enrichment Culture**		
	Positive	**Negative**	**Total**
Positive	221	28	249
Negative	9	586	595
Total	230	614	844
Sensitivity: 96.1% (221/230) 95% CI (92.7%, 97.9%) **Specificity:** 95.4% (586/614) 95% CI (93.5%, 96.8%)			
SA Site 3			
cobas® MRSA/SA Test	**Direct and Enrichment Culture**		
	Positive	**Negative**	**Total**
Positive	153	24	177
Negative	14	423	437
Total	167	447	614
Sensitivity: 91.6% (153/167) 95% CI (86.4%, 94.9%) **Specificity:** 94.6% (423/447) 95% CI (92.1%, 96.4%)			

The positive and negative external control isolates were tested each day during the clinical studies. All MRSA/SA positive controls were detected accurately (100%,

116/116). All negative controls were detected accurately (100%, 116/116).

These study results are acceptable.

b. Clinical specificity:

See table above.

c. Other clinical supportive data (when a. and b. are not applicable):

Not applicable.

4. Clinical cut-off:

Not applicable.

5. Expected values/Reference range:

During the multi-center clinical trials, based on combined direct and enrichment culture, the overall incidence of MRSA in nasal swab specimens during these studies was 6.4% (160/2500). The overall incidence of SA in nasal swab specimens during these studies was 26.4% (660/2501).

N. Instrument Name:

cobas® 4800 System

O. System Descriptions:

1. Modes of Operation:

Does the applicant's device contain the ability to transmit data to a computer, webserver, or mobile device?

Yes ____X___ or No _____

Does the applicant's device transmit data to a computer, webserver, or mobile device using wireless transmission?

Yes _____ or No ____X___

2. Software:

FDA has reviewed applicant's Hazard Analysis and software development processes for this line of product types:

Yes ___X___ or No _____

3. Specimen Identification:

   Specimens are identified using barcodes on specimen vials.

4. Specimen Sampling and Handling:

   Specimens are placed on the cobas x 480 instrument as open tubes and specimen processing is fully automated. After completion of specimen processing, the user transfers the plate carrier to the cobas z 480 instrument for automated amplification and detection. Specimens can be processed directly from primary collection vials or as aliquots of the specimen in secondary vials. See section I for more information on specimen handling.

5. Calibration:

   No calibration is required by the user. Roche technicians perform calibration periodically as required.

6. Quality Control:

   See section M.1.*c* for information on internal and external controls.
   See section M.3.*a* for information on external control performance during clinical trials.

P. Proposed Labeling:

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

Q. Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.