September 25, 2019

Luminex Corporation
Kate Linak
Manager, Regulatory Affairs
12212 Technology Blvd
Austin, Texas 78727

Re: K191742
Trade/Device Name: ARIES MRSA Assay
Regulation Number: 21 CFR 866.1640
Regulation Name: Antimicrobial Susceptibility Test Powder
Regulatory Class: Class II
Product Code: NQX
Dated: June 28, 2019
Received: July 1, 2019

Dear Kate Linak:

We have reviewed your Section 510(k) premarket notification of intent to market the device referenced above and have determined the device is substantially equivalent (for the indications for use stated in the enclosure) to legally marketed predicate devices marketed in interstate commerce prior to May 28, 1976, the enactment date of the Medical Device Amendments, or to devices that have been reclassified in accordance with the provisions of the Federal Food, Drug, and Cosmetic Act (Act) that do not require approval of a premarket approval application (PMA). You may, therefore, market the device, subject to the general controls provisions of the Act. Although this letter refers to your product as a device, please be aware that some cleared products may instead be combination products. The 510(k) Premarket Notification Database located at https://www.accessdata.fda.gov/scripts/cdrh/cfdocs/cfpmn/pmn.cfm identifies combination product submissions. The general controls provisions of the Act include requirements for annual registration, listing of devices, good manufacturing practice, labeling, and prohibitions against misbranding and adulteration. Please note: CDRH does not evaluate information related to contract liability warranties. We remind you, however, that device labeling must be truthful and not misleading.

If your device is classified (see above) into either class II (Special Controls) or class III (PMA), it may be subject to additional controls. Existing major regulations affecting your device can be found in the Code of Federal Regulations, Title 21, Parts 800 to 898. In addition, FDA may publish further announcements concerning your device in the Federal Register.

Please be advised that FDA's issuance of a substantial equivalence determination does not mean that FDA has made a determination that your device complies with other requirements of the Act or any Federal statutes and regulations administered by other Federal agencies. You must comply with all the Act's

requirements, including, but not limited to: registration and listing (21 CFR Part 807); labeling (21 CFR Part 801 and Part 809); medical device reporting (reporting of medical device-related adverse events) (21 CFR 803) for devices or postmarketing safety reporting (21 CFR 4, Subpart B) for combination products (see https://www.fda.gov/combination-products/guidance-regulatory-information/postmarketing-safety-reporting-combination-products); good manufacturing practice requirements as set forth in the quality systems (QS) regulation (21 CFR Part 820) for devices or current good manufacturing practices (21 CFR 4, Subpart A) for combination products; and, if applicable, the electronic product radiation control provisions (Sections 531-542 of the Act); 21 CFR 1000-1050.

Also, please note the regulation entitled, "Misbranding by reference to premarket notification" (21 CFR Part 807.97). For questions regarding the reporting of adverse events under the MDR regulation (21 CFR Part 803), please go to https://www.fda.gov/medical-devices/medical-device-safety/medical-device-reporting-mdr-how-report-medical-device-problems.

For comprehensive regulatory information about medical devices and radiation-emitting products, including information about labeling regulations, please see Device Advice (https://www.fda.gov/medical-devices/device-advice-comprehensive-regulatory-assistance) and CDRH Learn (https://www.fda.gov/training-and-continuing-education/cdrh-learn). Additionally, you may contact the Division of Industry and Consumer Education (DICE) to ask a question about a specific regulatory topic. See the DICE website (https://www.fda.gov/medical-devices/device-advice-comprehensive-regulatory-assistance/contact-us-division-industry-and-consumer-education-dice) for more information or contact DICE by email (DICE@fda.hhs.gov) or phone (1-800-638-2041 or 301-796-7100).

Sincerely,

Steven Gitterman, M.D., Ph.D.
Deputy Director
Division of Microbiology Devices
OHT7: Office of In Vitro Diagnostics
and Radiological Health
Office of Product Evaluation and Quality
Center for Devices and Radiological Health

Enclosure

510(k) Summary

This Summary of 510(k) safety and effectiveness information is being submitted in accordance with the requirements of 21 CFR 807.92.

Date of Preparation: 09/13/2019

A. 510(k) Number:

K191742

B. Purpose for Submission:

Traditional 510(k), New Device

C. Measurand:

Target DNA sequences for
(1) *mecA* and *mecC* genes for methicillin resistance
(2) *orfX* complex gene of *Staphylococcus aureus*
(3) SCC*mec* junction region

D. Type of Test:

Qualitative Real Time Polymerase Chain Reaction (PCR)

E. Applicant:

Kate Linak
Luminex Corporation
12212 Technology Blvd
Austin, TX 78727

Tel: 512-381-4397

F. Proprietary and Established Names:

ARIES® MRSA Assay

G. Regulatory Information:

Product Code	Classification	Regulation Section	Panel
NQX	II	21 CFR 866.1640—Antimicrobial susceptibility test powder	Microbiology

H. Intended Use:

1. Intended use(s):

   The ARIES® MRSA Assay is an integrated, real-time, polymerase chain reaction (PCR) based qualitative in vitro diagnostic test for the direct detection of methicillin–resistant *Staphylococcus aureus* (MRSA) DNA from nasal swabs in patients at risk for nasal colonization.

   The ARIES® MRSA Assay is intended to aid in the prevention and control of MRSA infections in healthcare settings.

   The assay is not intended to guide, diagnose, or monitor treatment for MRSA infections. It is not intended to provide results of susceptibility to oxacillin/methicillin. A negative result does not preclude MRSA nasal colonization. Concomitant cultures are necessary to recover organisms for epidemiological typing or for further susceptibility testing.

   The ARIES® MRSA Assay is indicated for use with ARIES® Systems.

2. Indication(s) for use:

   Same as intended use.

3. Special conditions for use statement(s):

   For prescription use only.

4. Special instrument requirements:

   For use with ARIES® Systems.

I. Device Description:

The ARIES® MRSA Assay is a polymerase chain reaction (PCR)-based qualitative *in vitro* diagnostic test system which consists of the ARIES® System or the ARIES® M1 System with their included ARIES® Software, a sample processing tube, an assay-specific cassette, and an assay-specific protocol file. The ARIES® MRSA Assay cassette is a disposable, single-use cassette containing nucleic acid purification reagents, internal sample process control

(SPC), and an assay-specific master mix capable of performing the designated assay on one sample. The ARIES® MRSA Assay cassette directly detects methicillin-resistant *Staphylococcus aureus* (MRSA) from nasal swabs in patients at risk of nasal colonization. Specifically, the ARIES® MRSA Assay cassette detects the methicillin resistance genes (*mecA* and *mecC*), *Staphylococcus aureus orfX* gene, the SCC*mec* junction region, and a DNA Sample Processing Control.

Nasal swab specimens are collected using the Liquid Amies Elution Swab (ESwab™) Collection and Transport System, or equivalent. A portion of the sample is transferred to the provided 2 mL ARIES MRSA Sample Processing Tube and vortexed. The processed sample is then transferred to the ARIES® MRSA Assay cassette.

The specimen is lysed and nucleic acid is extracted using an ARIES® instrument. An extractable sample processing control (SPC) target present in the ARIES® MRSA Assay cassette is processed with the specimen. The SPC controls for recovery of extracted nucleic acid, for inhibitory substances and for PCR reagent and instrument integrity. The Ct value of the SPC is designed to verify nucleic acid extraction, to identify PCR inhibition, if any, and verify proper function of the extraction system and real-time instrument.

The extracted nucleic acid and SPC are transferred via magnetic beads through the cassette to the ARIES® MRSA Assay lyophilized PCR reagents in the PCR tube that contain primer pairs and probes specific to *mecA*/*mecC*, *orfX*, SCC*mec* and the SPC sequence. Each probe is labeled with a distinct fluorophore and detected in a distinct channel of an ARIES® System. PCR amplification is performed and assay fluorescence is monitored. Hybridization of a fluorescently labeled probe to the amplified target results in the release of quenching and generation of fluorescence signal that is indicative of PCR generated amplicon. Following amplification, the reaction is heated to separate the fluorescent-labeled probe from the amplified target, a process that results in a decrease in the fluorescence signal. The reaction fluorescence is measured during this process and the temperature at which the change in fluorescence is the maximum is the T_m of the amplicon. The instrument fluorescence output is analyzed and test results are determined using the ARIES® System software and the ARIES® MRSA Assay protocol and run files. ARIES® MRSA Assay results may be reported from the ARIES® Software or from the optional SYNCT® Software.

J. Substantial Equivalence Information:

1. Predicate device name(s):

   BD MAX™ MRSA *XT*

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

   K133605

3. Comparison with predicate:

   The following tables compare Luminex's ARIES® MRSA Assay to GeneOhm Sciences Canada, Inc. (BD Diagnostics) BD MAX™ MRSA *XT* assay (K133605).

Table 1: Similarities between New Device and Predicate

Item	New Device	Predicate: K133605
Intended Use	The ARIES® MRSA Assay is an integrated, real-time, polymerase chain reaction (PCR) based qualitative in vitro diagnostic test for the direct detection of methicillin–resistant *Staphylococcus aureus* (MRSA) DNA from nasal swabs in patients at risk for nasal colonization. The ARIES® MRSA Assay is intended to aid in the prevention and control of MRSA infections in healthcare settings. The assay is not intended to guide, diagnose, or monitor treatment for MRSA infections. It is not intended to provide results of susceptibility to oxacillin/methicillin. A negative result does not preclude MRSA nasal colonization. Concomitant cultures are necessary to recover organisms for epidemiological typing or for further susceptibility testing. The ARIES® MRSA Assay is indicated for use with ARIES® Systems.	The BD MAX™ MRSA *XT* assay performed on the BD MAX™ System is an automated qualitative in vitro diagnostic test for the direct detection of methicillin-resistant *Staphylococcus aureus* (MRSA) DNA from nasal swabs in patients at risk for nasal colonization. The test utilizes real-time polymerase chain reaction (PCR) for the amplification of MRSA DNA and fluorogenic target-specific hybridization probes for the detection of the amplified DNA. The BD MAX™ MRSA XT assay is intended to aid in the prevention and control of MRSA infections in healthcare settings. It is not intended to diagnose MRSA infections nor guide or monitor treatment for MRSA infections. A negative result does not preclude nasal colonization. Concomitant cultures are necessary to recover organisms for epidemiological typing or for further susceptibility testing.
Assay Targets	(1) *orfX* , *S. aureus* complex gene (2) SCC*mec* junction region	(1) SCC*mec*/*orfX* junction area of methicillin-resistant *Staphylococcus aureus* (i.e., MREJ for SCC*mec* Right

	(3) *mecA* and *mecC* genes, for methicillin resistance	Extremity Junction). The BD MAX™ MRSA *XT* assay is designed to detect MREJ types i, ii, iii, iv, v, vi, vii, ix, xiii, xiv, and xxi; and (2) *mecA* and *mecC* genes for methicillin resistance.
Sample Types	Nasal swabs	Nasal swabs
Assay Type	Real-time PCR	Amplification: PCR
Assay Results	Qualitative	Qualitative
Assay Controls	Sample Processing Control (SPC)	Specimen Processing Control (SPC)

Table 2: Differences between New Device and Predicate

Item	New Device	Predicate: K133605
Extraction Method	Automated by the ARIES® Systems	Automated by the BD MAX™ System
Assay Instrument	ARIES® Systems	BD MAX™ System
Detection	Fluorescent reporter probes for each target, and melting curve analysis.	Fluorogenic target-specific hybridization.

K. Test Principle:

The ARIES®MRSA Assay PCR amplification and detection reagents contain primers and probes specific to the methicillin resistance genes (*mec*A and *mec*C), a *Staphylococcus aureus* complex gene (*orf*X), the SCC*mec* region, and the Sample Processing Control (SPC) sequence. Each of the probes are labeled with a distinct fluorophore and detected in distinct channels of the ARIES® System. The probes contain a fluorophore on the 5' end of the oligonucleotide sequence and a quencher at the 3' end of the oligonucleotide sequence such that in random coil state when the probe is not hybridized to the target sequence, the fluorescent signal is quenched. PCR amplification is performed and assay fluorescence is monitored. Hybridization of a fluorescently labeled probe to the amplified target results in the release of quenching and generation of fluorescence signal that is indicative of PCR generated amplicon.

L. Performance Characteristics:

1. Analytical performance:

   a. *Precision/Reproducibility:*

   Precision

Within-laboratory precision/repeatability of the ARIES® MRSA Assay was conducted by testing a blinded panel of samples by two operators from a single site using a single lot of reagents and assessing the variabilities of the results. The panel consisted of five samples: two methicillin-resistant *Staphylococcus aureus* (MRSA) strains each prepared at 1x and 5x Limit of Detection (LoD) in simulated nasal matrix in Modified Liquid Amies (SNM+LA) and a negative sample (SNM+LA only). Each sample was tested in triplicate by each of the two operators each day for five non-consecutive days. The results showed that each operator generated 100% expected MRSA results, inclusive of MRSA positive and MRSA negative calls demonstrating that the ARIES® MRSA Assay is reproducible between independent operators. The results of the precision/repeatability study are presented in Table 3.

Table 3: Precision Study

Strain/Level	Positive/Tested (%)
MRSA *mecA*+ Moderate Positive 5X LoD[1]	30/30 (100)
MRSA *mecA*+ Low Positive 1X LoD	30/30 (100)
MRSA *mecC*+ Moderate Positive 5X LoD	30/30[2] (100)
MRSA *mecC*+ Low Positive 1X LoD	30/30 (100)
Negative	0/30 (0.0)

[1] LoD; Limit of detection for MRSA Strains BEI NR-46232 and ATCC BAA-2313 5X LoD MRSA *mecA*+ strain BEI NR-46232: 9.75E+04 CFU/mL; 1X LoD MRSA *mecA*+ BEI NR-46232: 1.95E+04 CFU/mL; 5X LoD MRSA strain *mecC*+ ATCC BAA-2313: 3.88E+05 CFU/mL; 1X LoD MRSA *mecC*+ ATCC BAA-2313: 7.75E+04 CFU/mL

[2] A single sample was reported as Invalid on initial testing; reported as Positive upon repeat testing.

Reproducibility

Site-to-site reproducibility of the ARIES® MRSA Assay was conducted by testing a blinded panel of samples at 3 independent sites. The panel consisted of 5 samples: 2 methicillin-resistant *Staphylococcus aureus* (MRSA) strains each prepared at 1x and 5x Limit of Detection (LoD) in simulated nasal matrix in

modified Liquid Amies (SNM+LA) and a negative sample (SNM+LA only). Each sample was tested in triplicate by 2 operators for 5 non-consecutive days using a single lot of assay reagents at each site. The ARIES® MRSA Assay generated 100% expected MRSA positive results for MRSA positive samples and 100% expected MRSA negative results (0% MRSA positivity) for MRSA negative samples demonstrating reproducibility of the assay. The results of the reproducibility study are presented in Table 4.

Table 4: Site-to-Site Reproducibility

Strain/Level	Positive/Number (%)			
	Site 1	Site 2	Site 3	Overall
MRSA *mecA*+ Moderate Positive 5X LoD[1]	30/30 (100)	30/30 (100)	30/30 (100)	90/90 (100)
MRSA *mecA*+ Low Positive 1X LoD	30/30 (100)	30/30 (100)	30/30 (100)	90/90 (100)
MRSA *mecC*+ Moderate Positive 5X LoD	30/30 (100)	30/30 (100)	30/30[2] (100)	90/90 (100)
MRSA *mecC*+ Low Positive 1X LoD	30/30[2] (100)	30/30 (100)	30/30 (100)	90/90 (100)
Negative	0/30 (0.0)	0/30[3] (0.0)	0/30 (0.0)	0/90 (0.0)

[1] LoD; Limit of detection for MRSA Strains BEI NR-46232 and ATCC BAA-2313 5X LoD MRSA *mecA*+ strain BEI NR-46232: 9.75E+04 CFU/mL; 1X LoD MRSA *mecA*+ BEI NR-46232: 1.95E+04 CFU/mL; 5X LoD MRSA strain *mecC*+ ATCC BAA-2313: 3.88E+05 CFU/mL; 1X LoD MRSA *mecC*+ ATCC BAA-2313: 7.75E+04 CFU/mL

[2] A single sample was reported as Invalid on initial testing; reported as Positive upon repeat testing.

[3] A single sample was reported as Invalid on initial testing; reported as Negative upon repeat testing.

b. Linearity/assay reportable range:

Not applicable. The ARIES® MRSA Assay is a qualitative assay.

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

Controls:

Process Control

Each ARIES® MRSA Assay cassette contains a Sample Processing Control (SPC), which is processed with the sample and analyzed during the amplification reaction. The SPC verifies nucleic acid extraction, and proper reagent, cassette, ARIES® System, and assay protocol performance. The SPC has a known melting

temperature (T_m) range and Ct range. Each time an assay is run, the system measures the temperature and fluorescence intensity of the SPC control to ensure the thermal and optical subsystems have remained in calibration.

External Controls

External Positive and Negative Controls were tested on a daily basis during the prospective Clinical Study using a total of 5 ARIES systems and 3 ARIES MRSA Assay cassette lots. The Positive External Control comprised a standardized suspension of a strain of MRSA at 1.95E+05 CFU/mL (10X LoD). The Negative External Control comprised a standardized suspension of a strain of *S. epidermidis* at 4.23E+04 CFU/mL (10X LoD). On initial testing, 182/183 (99.5%) Positive and 178/183 (97.3%) Negative External Controls produced the expected results. Upon repeat testing, all controls produced the expected results.

External controls should be tested according to guidelines or requirements of local, provincial and/or federal regulations or accreditation organizations. A reference methicillin-resistant *Staphylococcus aureus* strain or well characterized methicillin-resistant *Staphylococcus aureus* clinical isolates may be used as positive controls. The ARIES® MRSA Assay Kit does not include external positive or negative controls.

Stability:

Specimen Stability

Specimen stability was evaluated using contrived specimens, stored at 2°C and 30°C and tested with the ARIES® MRSA Assay. This was assessed by testing six replicates of each of two methicillin-resistant *Staphylococcus aureus* (MRSA) strains prepared at 3x and 10x Limit of Detection (LoD) in Natural Nasal Matrix (NNM), as well as a negative sample (NNM only). Following testing on the day of sample preparation, specimens stored at 2±2°C were tested at 5 time points over 10 days and specimens stored at 30±1°C were tested at 7 time points over 10 days. The results demonstrated that MRSA specimens stored at both temperatures (2°C and 30°C) generated 100% expected MRSA positive results across all time points tested. An overall 0.53% (4/757) cassette invalid rate was observed for the testing of MRSA strains and NNM samples in this study. MRSA specimens are stable for up to 10 days when stored at 2-30°C.

Shelf-Life Stability

A real time stability study was performed to evaluate the stability of the ARIES® MRSA Assay Cassette in order to establish a shelf life. This was assessed by testing six replicates of positive target and six replicates of negative target on

three different lots of ARIES® MRSA Assay Cassettes stored at two different temperatures (4°C and 25°C/room temperature) at ten different time points extending out to 19 months. ARIES® MRSA Assay Cassettes generated the expected MRSA positive calls for all positive replicates and MRSA negative calls for all negative replicates stored at both 4˚C and 25˚C at all time points across all cassette lots.

Cassette open box stability evaluated the performance of the ARIES® MRSA Assay Cassettes after removal from the cassette pouch and exposed to ambient temperatures, humidity and light for 10 hours. The results showed that all MRSA positive and MRSA negative samples through the 10 hour study duration generated 100% expected results at all 5 time points. Three lots of cassettes were used to assess open box stability. Over the course of 10 hours, all three lots of cassettes produced expected results, showing that ARIES® MRSA Assay Cassettes are stable in ambient temperatures for up to 10 hours after they have been removed from the storage pouch.

d. Detection Limit:

A Limit of Detection (LoD) study was performed to evaluate the analytical sensitivity of the ARIES MRSA Assay using one strain of MRSA mecA+ (NR-46232) and one strain of MRSA mecC+ (BAA-2312). The preliminary LoD for each strain was determined by performing a six point, five-fold dilution series in natural nasal matrix (NNM). The preliminary LoD was identified as the lowest concentration that had 100% positivity (out of at least 5 replicates). Concentrations in colony forming units per milliliter (CFU/mL) and CFU per cassette were determined for dilutions of each strain by plating the dilutions used in the preliminary study and colony counting.

The LoD concentrations determined in the preliminary study were confirmed with the same strains diluted in NNM to concentrations at, below, and above the LoD and tested with 20 replicates. The confirmed LoD concentrations, identified as the lowest concentration detected in ≥ 95% of replicates, for each strain are presented in Table 5.

Table 5: LoD Results for MRSA strains

Species	Strain	Matrix Type	LoD (CFU/mL)	LoD (CFU/cassette)	ARIES MRSA Positivity	95% C.I.
MRSA *mecA*+	**NR-46232**	NNM	1.95E+04	3.89E+03	100% (20/20)	83.9% - 100%
		SNM+LA	1.95E+04	3.89E+03	95.0% (19/20)	76.4% - 99.1%
MRSA *mecC*+	**BAA-2312**	NNM	7.75E+04	1.55E+04	100% (20/20)	83.9% - 100%

e. Inclusivity (Analytical Reactivity)

Inclusivity (Analytical Reactivity) of ARIES® MRSA Assay was evaluated by testing fifty-five (55) methicillin-resistant *Staphylococcus aureus* (MRSA) strains including the two strains tested in the Limit of Detection (LoD) study. These strains are representative SCC*mec*, Pulse-Field Gel Electrophoresis (PFGE), and MREJ variants from diverse geographic locations. Each strain was diluted in simulated nasal matrix in Modified Liquid Amies (SNM+LA) to a final concentration of 3X LoD and tested in triplicate. The results demonstrated that all 55 strains generated 100% (3/3 replicates) MRSA positivity as expected. The results of the analytical reactivity (inclusivity) study are presented in Table 6.

Table 6: Inclusivity (Analytical Reactivity) Results

Strain Description	Source	Catalog #	Strain/ Location	PFGE Type	SCC*mec* Type	MRSA Positivity (%)
MRSA *mecA+*	BEI	NR-46232 (LoD strain)	NRS703/ Minnesota	USA300	IV	100% (3/3)
	ATCC	BAA-38	N/A	N/A	I	100% (3/3)
	ATCC	BAA-1686	N/A	N/A	II	100% (3/3)
	ATCC	BAA-1687[1]	N/A	N/A	II	100% (3/3)
	ATCC	BAA-1692	N/A	USA100	II	100% (3/3)
	ATCC	BAA-1681	N/A	USA100	II	100% (3/3)
	ATCC	BAA-1682	N/A	USA100	II	100% (3/3)
	ATCC	BAA-1699	N/A	USA100	II	100% (3/3)
	BEI	NR-46250	NRS721/ Oregon	USA100	II	100% (3/3)
	ATCC	BAA-1761	N/A	USA100	II	100% (3/3)
	ATCC	BAA-1750	N/A	USA200	II	100% (3/3)
	BEI	NR-46251	NRS722/ Oregon	USA200	II	100% (3/3)
	ATCC	BAA-39	N/A	N/A	III	100% (3/3)
	ATCC	BAA-40	N/A	N/A	IIIa	100% (3/3)
	ATCC	BAA-1717	N/A	USA300	IVa	100% (3/3)
	ATCC	BAA-1762	N/A	USA300	IVb	100% (3/3)
	ATCC	BAA-1680	N/A	USA300	IVa	100% (3/3)
	BEI	NR-46070	NRS384/ Mississippi	USA300	IV	100% (3/3)
	ATCC	BAA-1683	N/A	USA400	IVa	100% (3/3)
	ATCC	BAA-1707	N/A	USA400	IV	100% (3/3)
	ATCC	BAA-1752	N/A	USA400	IV	100% (3/3)
	ATCC	BAA-1757	N/A	USA400	IV	100% (3/3)
	ATCC	BAA-1696	N/A	USA400	IVa	100% (3/3)

Strain Description	Source	Catalog #	Strain/ Location	PFGE Type	SCC*mec* Type	MRSA Positivity (%)
	BEI	NR-46207	NRS678/ Connecticut	USA500	IV	100% (3/3)
	ATCC	BAA-1689	N/A	USA500	IV	100% (3/3)
	BEI	NR-46220	NRS691/ Georgia	USA500	IV	100% (3/3)
	ATCC	BAA-1688	N/A	N/A	V	100% (3/3)
	ATCC	BAA-42	N/A	N/A	VI	100% (3/3)
	BEI	NR-46177	NRS648/ California	USA600	II	100% (3/3)
	ATCC	BAA-1751	N/A	USA600	II	100% (3/3)
	BEI	NR-46218	NRS689/ Georgia	USA700	IV	100% (3/3)
	ATCC	BAA-1766	N/A	USA700	V	100% (3/3)
	BEI	NR-46221	NRS692/ Georgia	USA800	IV	100% (3/3)
	ATCC	BAA-1771	N/A	USA800	IV	100% (3/3)
	BEI	NR-46197	NRS668/ Colorado	USA800	N/A	100% (3/3)
	ATCC	BAA-1747	N/A	USA1000	IV	100% (3/3)
	ATCC	BAA-1769	N/A	USA1000	IV	100% (3/3)
	ATCC	BAA-1764	N/A	USA1100	IV	100% (3/3)
	ATCC	BAA-1767	N/A	USA1100	IV	100% (3/3)
	ATCC	33592	N/A	N/A	III	100% (3/3)
	ATCC	33593	N/A	N/A	III	100% (3/3)
	ATCC	43300	N/A	N/A	II	100% (3/3)
	ATCC	700698	N/A	N/A	II	100% (3/3)
	ATCC	700699	N/A	N/A	II	100% (3/3)
	ATCC	700787	N/A	N/A	II	100% (3/3)
	ATCC	700788	N/A	N/A	II	100% (3/3)
	ATCC	700789	N/A	N/A	II	100% (3/3)
	ATCC	BAA-41	N/A	N/A	II	100% (3/3)
	ATCC	BAA-43	N/A	N/A	IIIa	100% (3/3)
	ATCC	BAA-44	N/A	N/A	I	100% (3/3)
	ATCC	BAA-1720	N/A	N/A	II	100% (3/3)
	ATCC	BAA-2094	N/A	N/A	V	100% (3/3)
	ATCC	BAA-2096	N/A	N/A	IV	100% (3/3)
MRSA *mecC+*	ATCC	BAA-2313 (LoD strain)	N/A	N/A	XI	100% (3/3)
	ATCC	BAA-2312	N/A	N/A	XI	100% (3/3)

[1]One replicate was reported as Invalid on initial testing; reported as Positive upon repeat testing.

f. Challenge Study

An additional analytical study was carried out to evaluate the analytical performance of the ARIES® MRSA Assay using a panel of challenge strains. The challenge panel included 16 methicillin-resistant *Staphylococcus aureus* (MRSA) strains with high minimum inhibitory concentration (MIC) values of ≥ 16 µg/mL oxacillin and 17 MRSA strains with low MIC values of ≤ 8 µg/mL oxacillin, four borderline oxacillin-resistant *Staphylococcus aureus* (BORSA) strains, 16 empty cassette variants of *Staphylococcus aureus* strains, and one methicillin-resistant *Staphylococcus epidermidis* (MRSE) strain. The MRSA strains were tested at 3X LoD and all other strains were tested at a concentration of 1E+06 CFU/mL. Each strain was tested in triplicate. Fourteen of the sixteen (14/16) MRSA with high MICs and 16/17 of the MRSA with low MICs generated the expected MRSA positive results at 3X LoD (100% MRSA Positive). Two strains of MRSA with high MIC values and one strain of MRSA with low MIC value that did not generate the expected MRSA positive results when tested at 3X LoD were re-tested at 5X LoD and generated the expected MRSA positive results (100% MRSA Positive). The four BORSA strains, the MRSE strain, and all empty cassette variants of *Staphylococcus aureus* generated the expected 0% MRSA positive results (100% MRSA Negative). Results of the challenge study are presented in Table 7.

Table 7: Challenge Study Results

Target Description	Source	Strain ID	Test Concentration	ARIES® MRSA Positivity
MRSA with High Oxacillin MIC	ARLG	ARLG-1643	3X LoD	100% (3/3)
	ARLG	ARLG-1644	3X LoD	100% (3/3)
	ARLG	ARLG-1645	3X LoD	100% (3/3)
	ARLG	ARLG-1646	3X LoD	100% (3/3)
	ARLG	ARLG-1662	3X LoD	100% (3/3)
	ARLG	ARLG-1669[1]	3X LoD	66.7% (2/3)
	ARLG	ARLG-1604	3X LoD	100% (3/3)
	ARLG	ARLG-1613[1]	3X LoD	66.7% (2/3)
	CDC AR Bank	AR-0215	3X LoD	100% (3/3)
	CDC AR Bank	AR-0218	3X LoD	100% (3/3)
	CDC AR Bank	AR-0219	3X LoD	100% (3/3)
	CDC AR Bank	AR-0220	3X LoD	100% (3/3)
	CDC AR Bank	AR-0223	3X LoD	100% (3/3)
	CDC AR Bank	AR-0224	3X LoD	100% (3/3)
	CDC AR Bank	AR-0227	3X LoD	100% (3/3)
	CDC AR Bank	AR-0228	3X LoD	100% (3/3)
MRSA with Low Oxacillin MIC	ARLG	ARLG-1642[1]	3X LoD	66.7% (2/3)
	Lyon University	20130237	3X LoD	100% (3/3)
	Lyon University	20130524	3X LoD	100% (3/3)

Target Description	Source	Strain ID	Test Concentration	ARIES® MRSA Positivity
	CDC AR Bank	AR-0216	3X LoD	100% (3/3)
	CDC AR Bank	AR-0217	3X LoD	100% (3/3)
	CDC AR Bank	AR-0221	3X LoD	100% (3/3)
	CDC AR Bank	AR-0225	3X LoD	100% (3/3)
	CDC AR Bank	AR-0226	3X LoD	100% (3/3)
	ATCC	BAA-1688	3X LoD	100% (3/3)
	CDC AR Bank	AR-472	3X LoD	100% (3/3)
	CDC AR Bank	AR-473	3X LoD	100% (3/3)
	CDC AR Bank	AR-474	3X LoD	100% (3/3)
	CDC AR Bank	AR-475	3X LoD	100% (3/3)
	CDC AR Bank	AR-476	3X LoD	100% (3/3)
	CDC AR Bank	AR-477	3X LoD	100% (3/3)
	CDC AR Bank	AR-478	3X LoD	100% (3/3)
	CDC AR Bank	AR-479	3X LoD	100% (3/3)
BORSA	CDC AR Bank	AR-0489	1E+06 CFU/mL	0% (0/3)
	CDC AR Bank	AR-0490	1E+06 CFU/mL	0% (0/3)
	CDC AR Bank	AR-0491	1E+06 CFU/mL	0% (0/3)
	CDC AR Bank	AR-0492	1E+06 CFU/mL	0% (0/3)
Empty Cassette Variant of SA	Lyon University	20101270	1E+06 CFU/mL	0% (0/3)
	Lyon University	20112896	1E+06 CFU/mL	0% (0/3)
	Lyon University	20112911	1E+06 CFU/mL	0% (0/3)
	Lyon University	20120556	1E+06 CFU/mL	0% (0/3)
	Lyon University	20120844	1E+06 CFU/mL	0% (0/3)
	Lyon University	20120871[2]	1E+06 CFU/mL	0% (0/3)
	Lyon University	20120984	1E+06 CFU/mL	0% (0/3)
	Lyon University	20121469	1E+06 CFU/mL	0% (0/3)
	Lyon University	20121544[2]	1E+06 CFU/mL	0% (0/3)
	Lyon University	20121635	1E+06 CFU/mL	0% (0/3)
	Lyon University	20121891	1E+06 CFU/mL	0% (0/3)
	Lyon University	20130769	1E+06 CFU/mL	0% (0/3)
	Lyon University	20131190	1E+06 CFU/mL	0% (0/3)
	Lyon University	20131273	1E+06 CFU/mL	0% (0/3)
	Lyon University	20131727	1E+06 CFU/mL	0% (0/3)
	Lyon University	20140852	1E+06 CFU/mL	0% (0/3)
MRSE	ATCC	51625	1E+06 CFU/mL	0% (0/3)

[1] Two strains of MRSA with high MIC values (ARLG-1669, ARLG-1613) and one strain of MRSA with low MIC value (ARLG-1642) that did not generate the expected MRSA positive results (100% MRSA positive) when tested at 3X LoD were re-tested at 5X LoD and generated the expected MRSA positive results (100% MRSA positive; 3/3).

[2] One replicate was reported as Invalid on initial testing; reported as Negative upon repeat testing.

g. Analytical specificity

Cross-Reactivity and Microbial Interference:

Analytical specificity of the ARIES® MRSA Assay in the presence of potentially cross-reacting or interfering organisms was evaluated. Ninety-nine (99) organisms commonly present in nasal specimen collection sites were tested at concentrations of 1E+06 CFU/mL for non-viral organisms, 1E+05 TCID50/mL for viruses and 5 µg/mL for human genomic DNA. The study was performed using one strain of *mecA+* methicillin-resistant *Staphylococcus aureus* (MRSA) (NR-46232) and one strain of *mecC+* MRSA (BAA-2313) at 3x Limit of Detection (LoD), as well as a negative sample consisting of simulated nasal matrix in Modified Liquid Amies (SNM+LA). Target samples at an intermediate concentration prepared in SNM+LA were spiked with potentially cross-reactive organisms prepared in SNM+LA before being tested on the ARIES® MRSA Assay in triplicates. The test results demonstrated that all replicates of both MRSA strains in absence and presence of the potentially cross-reactive organisms generated 100% expected MRSA positive results; and all replicates of SNM+LA tested in absence and presence of the cross-reacting organisms generated 100% expected MRSA negative results (0% MRSA positivity). A summary of all samples tested are presented in Table 8.

Table 8: Cross-reactivity/Microbial Interference Study

Non-Staphylococcal organisms	
Acinetobacter baumannii[3]	*Listeria monocytogenes*
Acinetobacter haemolyticus	*Legionella pneumophila*
Bacillus cereus	*Moraxella catarrhalis*
Bordetella pertussis	*Micrococcus luteus*
Candida albicans	*Mycoplasma pneumoniae*
Citrobacter freundii	*Mycobacterium tuberculosis* avirulent
Candida glabrata	*Neisseria meningitidis*
Citrobacter koseri	*Listeria monocytogenes*
Chlamydia pneumoniae	*Pasteurella aerogenes*
Corynebacterium bovis	*Pseudomonas aeruginosa*
Corynebacterium flavescens	*Pseudomonas fluorescens*[1]
Corynebacterium genitalium	*Proteus mirabilis*
Cryptococcus neoformans	*Providencia stuartii*
Enterobacter aerogenes	*Proteus vulgaris*
Enterobacter cloacae[1]	*Salmonella enterica subsp. Enterica*
Enterococcus faecalis	*Serratia marcescens*
Enterococcus faecium	*Streptococcus agalactiae*
Escherichia coli (O157:H7)	*Streptococcus anginosus*
Enterococcus flavescens	*Streptococcus mitis*
Enterococcus gallinarum	*Streptococcus mutans*
Enterococcus hirae	*Streptococcus pneumoniae*

Haemophilus influenzae	*Shigella sonnei*
Klebsiella oxytoca	*Streptococcus pyogenes*
Klebsiella pneumoniae (ESBL-producing)	*Streptococcus salivarius*
Klebsiella pneumoniae (KPC-producing)	*Streptococcus sanguinis*
Lactobacillus crispatus	*Streptococcus suis*
	Yersinia enterocolitica
Viruses	
Adenovirus Type 40	Measles virus
Adenovirus Type 7	Mumps virus
Coronavirus 229E	Parainfluenza 1
Coronavirus OC43	Parainfluenza 2
Cytomegalovirus	Parainfluenza 3
Epstein Barr Virus	Rhinovirus type 1A[3]
Influenza A	RSV A
Influenza B	RSV B
Human metapneumovirus	
Coagulase Negative Staphylococci (CoNS)	
Staphylococcus arlettae	*Staphylococcus equorum*
Staphylococcus captis	*Staphylococcus felis*
Staphylococcus carnosus	*Staphylococcus gallinarum*[2]
Staphylococcus chromogenes	*Staphylococcus haemolyticus* Z067
Staphylococcus epidermidis (255-01B)	*Staphylococcus kloosii*
Staphylococcus epidermidis (RP12 CIP 106510)	*Staphylococcus lentus*
Staphylococcus epidermidis (MRSE;RP62A)	*Staphylococcus pulvereri*
Staphylococcus epidermidis (CCF 15990)	*Staphylococcus simulans* Z032
Staphylococcus epidermidis (CCF 16471)	*Staphylococcus warneri* Z113[3]
Coagulase Positive Staphylococci	
Staphylococcus pseudointermedius	*Staphylococcus delphini*
Staphylococcus scheleiferi subsp. Coagulans	*Staphylococcus intermedius*
Staphylococcus scheleiferi subsp. Schleiferi	*Staphylococcus lutrae*
Methicillin-susceptible *Staphylococcus aureus* (MSSA)	
Staphylococcus aureus BAA-1749	*Staphylococcus aureus* 29213
Staphylococcus aureus BAA-1765	*Staphylococcus aureus* 0801675
Staphylococcus aureus BAA-1718	
Other	
Human genomic DNA	

[1] One replicate of SNM+LA negative matrix was reported as Invalid on initial testing; reported as Negative upon repeat testing.

[2] One replicate containing *mecA*+ MRSA was reported as Invalid on initial testing; reported as Positive upon repeat testing.

[3] One replicate containing *mecC*+ MRSA strain was reported as Invalid on initial testing; reported as Positive upon repeat testing.

Potentially Interfering Substances:

Twenty-four (24) potentially interfering non-microbial substances that are commonly present at the site of specimen collection were tested with the ARIES® MRSA Assay. The study was performed using one strain of *mecA+* methicillin-resistant *Staphylococcus aureus* (MRSA) (NR-46232) and one strain of *mecC+* MRSA (BAA-2313) at 3x LoD prepared in simulated nasal matrix in Modified Liquid Amies (SNM+LA), as well as SNM+LA alone as a negative control. Samples at intermediate concentrations prepared in SNM+LA were spiked with the non-microbial substances prepared in the appropriate diluent to reach the final concentrations and tested in triplicate. All replicates of both MRSA strains tested in the absence and presence of the potentially interfering substances generated 100% MRSA positivity as expected; and all replicates of SNM+LA alone generated 0% MRSA positivity (100% negativity) as expected. No interference was detected for any of the substances at the concentration tested. Potentially interfering substances that were tested are presented in Table 9.

Table 9: Potentially Interfering Substances

Substance	Test Concentration
Whole Blood	5% (v/v)
Mucin	5 mg/mL
Phenylephrine	0.03 µg/mL
Drixoral (Oxymetazoline)	10% (v/v)
Benzalkonium chloride	0.12%
Propylene glycol	20% (v/v)
Sorbitol[1]	6.45%
Benzyl alcohol[1]	0.5% (v/v)
Hypromellose	0.10%
Phosphoric acid	1.282 mg/mL
Beclomethasone	8.4 µg/mL
Dexamethasone	12 µg/mL
Flunisolide	5 µg/mL
Triamcinolone	22 µg/mL
Budesonide	6.30E-03 µg/mL
Mometasone	4.50E-04 µg/mL
Flonase (Fluticasone)	1.26E-03 µg/mL
ZICAM[2] (Galphimia glauca, Histaminum hydrochloricum)	10% (v/v)
Benzocaine	75 µg/mL
Menthol	0.5 mg/mL
Zanamivir[3]	1 mg/mL
Mupirocin	50 µg/mL

Tobramycin	33 µg/mL
FluMist© (Live intranasal influenza virus vaccine)	10% (v/v)

[1] One replicate of SNM+LA negative matrix was reported as Invalid on initial testing; reported as Negative upon repeat testing.

[2] One replicate containing *mecA*+ MRSA strain was reported as Invalid on initial testing; reported as Positive upon repeat testing.

[3] One replicate containing *mecC*+ MRSA strain was reported as Invalid on initial testing; reported as Positive upon repeat testing.

Competitive Interference:

Competitive interference was tested with methicillin-resistant *Staphylococcus aureus* (MRSA) at 1x limit of detection (LoD) and the co-infecting agent, methicillin-susceptible *Staphylococcus aureus* (MSSA) or methicillin-resistant coagulase-negative staphylococci (MRCoNS), at increasing concentrations. Each combination was tested in triplicate. The results showed that the ARIES® MRSA Assay detected MRSA at 1x LoD in the presence of increasing concentrations of MSSA or MRCoNS. No competitive interference in the ARIES® MRSA Assay was observed for co-infections of MRSA with MSSA or MRCoNS.

Carry-Over/Cross-Contamination:

Carry-over and cross contamination for the ARIES® MRSA Assay was evaluated by testing sixty (60) high positive (1.0E+07 CFU/mL) MRSA samples prepared in simulated nasal matrix in modified Liquid Amies (SNM+LA) and sixty (60) MRSA negative samples consisting of SNM+LA alone. The high positive samples were run adjacent to negative samples in an alternating pattern across twenty (20) consecutive module runs using two ARIES® instruments. Overall percent agreement with expected results was 100% for both high positive and negative samples. One replicate was reported Invalid on initial testing; reported as Positive upon repeat testing. No carry-over or cross contamination was observed in the study.

h. Assay cut-off

For the ARIES® MRSA Assay, each target (*mecA/mecC*, *orfX*, and SCC*mec*) has a Ct cut-off, Tm window, and Tm Peak Threshold. In addition, the internal sample processing control (SPC) also has a corresponding Ct cut-off, Tm window, and Tm Peak Threshold. Collectively, the cut-off values compose the assay protocol file parameters, which are used to determine the assay result for the detection of the target as Positive, Negative, or Invalid. These values are hard-coded into the ARIES® MRSA Assay Protocol File and are not modifiable. The Assay Protocol File parameters were determined, and their performance in the ARIES® MRSA Assay were evaluated according to the following general procedure:

- Initial Assay Protocol File parameters were set during internal optimization and benchmarking studies.
- The final Assay Protocol File parameters were then established during internal verification studies using data from optimization, benchmarking and verification.
- The selected Assay Protocol File parameter values were utilized in the determination of assay performance in the multi-site clinical trial conducted for the ARIES MRSA Assay.

The specific assay parameters for the ARIES® MRSA Assay are considered **confidential and proprietary**.

2. Comparison Studies

a. Method comparison with predicate device:

Not applicable.

b. Nasal Swab Comparison:

A nasal swab equivalency study was performed to evaluate the reproducibility of the ARIES® MRSA Assay with two different nasal swab types, Regular Nylon Flocked Swab (Copan Catalog Number: 480C) and Flexible Minitip Nylon Flocked Swab (Copan Catalog Number: 482C). The swabs were evaluated using one strain of Methicillin-resistant *Staphylococcus aureus* (MRSA) *mecA+* (NR-46232) at three concentrations, as well as a negative sample (Simulated nasal matrix in Modified Liquid Amies, SNM+LA). Samples at intermediate concentrations prepared in SNM+LA were transferred to Modified Liquid Amies using each of the two nasal swab types to reach the final testing concentrations at 3x LoD, 5x LoD and 10x LoD, respectively, and then tested on the ARIES® MRSA Assay. The test results demonstrated that both swab types generated 100% expected MRSA positivity for each strain of the concentrations tested. Both swab types also generated 0% positivity (100% negativity) for negative samples. Swab equivalency study results are presented in Table 10.

Table 10: ARIES® MRSA Assay Nasal Swab Equivalency Results

Assay Target	Part Number	Test Concentration (CFU/mL)	MRSA Positivity	
			Regular swab (Copan 480C)	Flexible minitip swab (Copan 482C)
MRSA *mecA+*	NR-46232	3x LoD (5.85E+04)	100% (6/6)	100% (6/6)
		5x LoD (9.75E+04)	100% (6/6)	100% (6/6)
		10x LoD (1.95E+05)	100% (6/6)	100% (6/6)
Negative (SNM+LA)	N/A	N/A	0% (0/6)	0% (0/6)

3. Clinical Performance:

Clinical performance of the ARIES® MRSA Assay for nasal swab specimens collected from patients at risk for methicillin-resistant *Staphylococcus aureus* (MRSA) colonization was established through a clinical study.

Performance of the ARIES® MRSA Assay was evaluated prospectively from August 2018 to February 2019 at four (4) geographically distinct clinical sites within the United States using the ARIES® System. Specimens included in the clinical study consisted of excess leftover de-identified, nasal clinical specimens collected using the Liquid Amies Elution Swab (Eswab™) Collection and Transport system, or equivalent, from patients at risk for nasal colonization. All eligible clinical specimens were tested by both the reference method (direct and enriched bacterial culture) and ARIES® MRSA Assay and the results compared. Reference method testing was performed at a centralized testing facility while ARIES® MRSA Assay testing was performed at each clinical site on their own clinical specimens.

A total of 2254 nasal swab specimens from subjects at risk for MRSA nasal colonization were collected. Of these 2254 specimens, 472 were excluded from the study based on inclusion/exclusion criteria leaving a total of 1,782 unique specimens that met the pre-determined eligibility criteria. Of the 472 excluded specimens, 275 specimens were from a subject who had taken a systemic or topical nasal antibiotics within 48 hours to 1 week prior to specimen collection, 93 specimens were not tested by the reference method due to either a delay in shipment or because testing could not start within 48 hours of collection, 27 specimens had an Indeterminate culture result, 24 specimens had insufficient volume for testing by both or either the ARIES MRSA Assay or reference method, 20 specimens were from a subject that had a known positive lab result for MRSA in the past 90 days of specimen collection, 9 specimens were not tested according to the reference method protocol, 8 specimens had *Proteus mirabilis* swarming on the blood agar plate for the reference method, 6 specimens did not have passing external controls for ARIES MRSA/SA Assay testing, 4 specimens were processed using the incorrect ARIES MRSA/SA Assay reagents, 3 specimens were from subjects previously enrolled in the study, 3 specimens were tested by the culture method after 48 hours from collection.

1,782 specimens were enrolled in the study and tested for methicillin-resistant *Staphylococcus aureus* by both the reference method and the ARIES® MRSA Assay. There were 20 specimens (20/1,782, 1.1%) that when tested with ARIES® MRSA Assay yielded an invalid result due to run failure or instrument error. None of these specimens were re-tested due to insufficient specimen volume.

For the 1,762 eligible specimens that were included in the device performance calculations, clinical sensitivity of the ARIES® MRSA Assay was 93.3% (97/104) with a lower bound 95% confidence interval of 87%, when compared to direct and enriched

bacterial culture. Clinical specificity of the ARIES® MRSA Assay was 93.5% (1550/1658) with a lower bound 95% confidence interval of 92%, when compared to direct and enriched bacterial culture. Of the 108 specimens that were MRSA-negative by culture but MRSA-positive by the ARIES MRSA Assay, culture showed that 63 specimens were *S. aureus* and 45 were negative (no growth). Positive percent agreement of the ARIES® MRSA Assay against direct bacterial culture was 93.5% (87/93) with a lower bound 95% confidence interval of 87%. Negative percent agreement of the ARIES® MRSA Assay against direct bacterial culture was 92.9% (1551/1669) with a lower bound 95% confidence interval of 92%.

Table 11: ARIES® MRSA Assay Performance Compared to Direct and Enriched Culture

ARIES® MRSA Assay	Direct and Enriched Bacterial Culture for MRSA		
	Positive	Negative	TOTAL
Positive	97	108	205
Negative	7	1550	1557
TOTAL	104	1658	1762
Sensitivity (95% CI)	93.3% (87% - 97%)		
Specificity (95% CI)	93.5% (92% - 95%)		

Table 12: ARIES® MRSA Assay Performance Compared to Direct Culture

ARIES® MRSA Assay	Direct Bacterial Culture for MRSA		
	Positive	Negative	TOTAL
Positive	87	118	205
Negative	6	1551	1557
TOTAL	93	1669	1762
Positive Percent Agreement (95% CI)	93.5% (87% - 97%)		
Negative Percent Agreement (95% CI)	92.9% (92% - 94%)		

The study results demonstrate that the diagnostic accuracy of the ARIES® MRSA Assay is acceptable for the detection of methicillin-resistant *Staphylococcus aureus* (MRSA) DNA from nasal swabs in patients at risk for nasal colonization.

4. Expected values/Reference range:

Table 13 provides a summary of the expected values of the 1762 prospectively collected nasal swab specimens that were included in the prospective analysis.

Table 13: ARIES® MRSA Assay Expected Values of Clinical Study Population

	Number of Subjects	% Distribution
Gender		
Male	122	12.8% (122/954)
Female	83	10.3% (83/808)
Overall	205	11.6% (205/1762)
Age (yrs)		
<2	21	5.2% (21/405)
2 - 11	21	8.4% (21/251)
12 - 21	19	10.1% (19/189)
22 - 59	74	15.4% (74/481)
≥60	70	16.1% (70/436)
Overall	**205**	**11.6% (205/1762)**

M. Proposed Labeling:

The labeling provided in the submission satisfies the requirements of 21 CFR 809.10.

N. Conclusion:

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