



**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY AND INSTRUMENT**

I Background Information:

A 510(k) Number

K241324

B Applicant

Affinity Biosensors, LLC

C Proprietary and Established Names

LifeScale Gram Negative Kit (LSGN) with the LifeScale AST system

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
SAN	Class II	21 CFR 866.1650 - A Cellular Analysis System For Multiplexed Antimicrobial Susceptibility Testing	MI-Microbiology
LON	Class II	21 CFR 866.1645 - Fully automated short-term incubation cycle antimicrobial susceptibility system	MI-Microbiology

II Submission/Device Overview:

A Purpose for Submission:

- To obtain a substantial equivalence determination for the addition of the following antimicrobial agents to the LifeScale Gram Negative Kit (LSGN): Amikacin, Cefepime, Ceftazidime-avibactam, Gentamicin, Levofloxacin, Meropenem, Meropenem-vaborbactam and Piperacillin-tazobactam. The LSGN Kit is used with the LifeScale AST system for testing positive blood culture samples containing gram-negative bacilli.
- Removal of limitations included in the cleared LSGN with the LifeScale AST system (K211815) for Aztreonam, Ceftazidime, Ertapenem, and Cefazolin.

B Measurand:

Antimicrobial		Range µg/mL	
		Min (≤)	Max (>)
Amikacin	AMI	4	256
*Ampicillin	AMP	2	64
*Aztreonam	AZT	1	64
*Cefazolin	FAZ	0.25	16
Cefepime	FEP	0.5	64
*Ceftazidime	TAZ	1	64
Ceftazidime-avibactam	CZA	2/4	32/4
*Ertapenem	ETP	0.12	8
Gentamicin	GEN	1	32
Levofloxacin	LEVO	0.25	16
Meropenem	MERO	0.12	16
Meropenem-vaborbactam	MEV	0.5/8	16/8
Piperacillin-tazobactam	P/T	4/4	256/4
*Trimethoprim-sulfamethoxazole	SXT	0.25	8

*Antimicrobics cleared in K211815

C Type of Test:

The LifeScale Gram Negative Kit (LSGN) with the LifeScale AST system is a quantitative antimicrobial susceptibility test system that determines the minimum inhibitory concentration of specific organisms from positive blood culture samples.

III Intended Use/Indications for Use:

A Intended Use(s):

The LifeScale AST system is a multiplexed *in vitro* diagnostic test that uses a microfluidic sensor and resonant frequency to calculate organism concentration and/or mass distribution for quantitative antimicrobial susceptibility testing (AST). Testing is performed directly on blood cultures signaled as positive by a continuous monitoring blood culture system and confirmed by Gram stain. The LifeScale AST system does not provide organism identification and is not indicated for use with polymicrobial samples. Interpretive results (Susceptible/Intermediate/Susceptible-dose dependent/Resistant) are provided for specific drug/organism combinations. Results are intended to be used in conjunction with other clinical and laboratory findings. Standard laboratory protocols for processing positive blood cultures should be followed to ensure availability of isolates for supplemental testing as needed. Additionally, subculture of positive blood culture is necessary for the susceptibility testing of organisms present in polymicrobial samples, for testing antimicrobial agents and species not indicated for testing with the device, for epidemiologic testing and for recovery of organisms present in microbial samples.

B Indication(s) for Use:

The LifeScale Gram Negative Kit (LSGN) is indicated for use with the LifeScale AST system for *in vitro* testing of positive blood culture samples confirmed by Gram stain as containing gram-negative bacilli for the antimicrobial agents and specific target organisms identified below:

- Ampicillin: *Escherichia coli*
- Aztreonam: *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella aerogenes*, *Klebsiella oxytoca*

- Cefazolin: *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella variicola*
- Ceftazidime: *Acinetobacter* spp. (other than *Acinetobacter ursingii*), *Escherichia coli*, *Klebsiella aerogenes*, *Klebsiella oxytoca*, *Klebsiella variicola*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*
- Ertapenem: *Escherichia coli*, *Klebsiella aerogenes*, *Klebsiella pneumoniae*, *Klebsiella oxytoca*
- Trimethoprim-Sulfamethoxazole: *Escherichia coli*, *Klebsiella aerogenes*, *Klebsiella oxytoca*, *Klebsiella variicola*
- Amikacin: *Acinetobacter* spp., *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella aerogenes*, *Klebsiella oxytoca*, *Klebsiella variicola*, *Pseudomonas aeruginosa*
- Cefepime: *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella aerogenes*, *Klebsiella oxytoca*, *Pseudomonas aeruginosa*
- Ceftazidime-avibactam: *Escherichia coli*, *Klebsiella aerogenes*, *Klebsiella oxytoca*, *Klebsiella variicola*, *Pseudomonas aeruginosa*
- Gentamicin: *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella aerogenes*, *Klebsiella oxytoca*, *Klebsiella variicola*, *Pseudomonas aeruginosa*
- Levofloxacin: *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella aerogenes*, *Klebsiella oxytoca*, *Pseudomonas aeruginosa*
- Meropenem: *Acinetobacter* spp., *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella oxytoca*, *Pseudomonas aeruginosa*
- Meropenem-vaborbactam: *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella aerogenes*, *Klebsiella oxytoca*
- Piperacillin-tazobactam: *Acinetobacter* spp., *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

Limitations

The following notable limitations are provided in the package insert. This is not an exhaustive list. Please refer to the Instructions for Use for all limitations.

1. Only the following drug/organism combinations have been shown to provide acceptable results with the LifeScale LSGN Kit:
 - Amikacin: *Acinetobacter* spp., *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella aerogenes*, *Klebsiella oxytoca*, *Klebsiella variicola*, *Pseudomonas aeruginosa*
 - Ampicillin: *Escherichia coli*
 - Aztreonam: *Escherichia coli*, *Klebsiella aerogenes*, *Klebsiella pneumoniae*, *Klebsiella oxytoca*
 - Cefazolin: *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella variicola*
 - Cefepime: *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella aerogenes*, *Klebsiella oxytoca*, *Pseudomonas aeruginosa*
 - Ceftazidime: *Acinetobacter* spp. (other than *Acinetobacter ursingii*), *Escherichia coli*, *Klebsiella aerogenes*, *Klebsiella oxytoca*, *Klebsiella variicola*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*
 - Ceftazidime-avibactam: *Escherichia coli*, *Klebsiella aerogenes*, *Klebsiella oxytoca*
 - Ertapenem: *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella aerogenes*, *Klebsiella oxytoca*

- Gentamicin: *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella aerogenes*, *Klebsiella oxytoca*, *Klebsiella variicola*, *Pseudomonas aeruginosa*
 - Levofloxacin: *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella aerogenes*, *Klebsiella oxytoca*, *Pseudomonas aeruginosa*
 - Meropenem: *Acinetobacter* spp., *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella oxytoca*, *Pseudomonas aeruginosa*
 - Meropenem-vaborbactam: *Escherichia coli*, *Klebsiella pneumoniae*, *Klebsiella aerogenes*, *Klebsiella oxytoca*
 - Piperacillin-tazobactam: *Acinetobacter* spp., *Escherichia coli*, *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*
 - Trimethoprim-sulfamethoxazole: *Escherichia coli*, *Klebsiella aerogenes*, *Klebsiella oxytoca*, *Klebsiella variicola*
2. Perform an alternative method of testing prior to reporting results for the following antimicrobial/organism combination(s):
 - Aztreonam: *P. aeruginosa*
 - Ceftazidime-avibactam: *P. aeruginosa*, *K. pneumoniae*
 - Ceftazidime: *Acinetobacter ursingii*
 - Trimethoprim-sulfamethoxazole: *K. pneumoniae*
 3. Perform an alternative method of testing prior to reporting results for:
 - Ceftazidime: *P. aeruginosa* at MIC value of 16 µg/mL due to the occurrence of major errors (5/61 susceptible isolates, (8.2%) adjusted to 2 major errors (3.3%) due to a lack of an intermediate breakpoint).
 - Ertapenem: *K. oxytoca* at MIC value of 0.5 µg/mL due to the occurrence of very major errors (2/10 resistant isolates, 20%).
 - Meropenem: *K. oxytoca* when the LifeScale MIC is 0.5 µg/mL due to two very major discrepancies.
 - Cefepime: *P. aeruginosa* when the MIC is 4 µg/mL due to the occurrence of very major errors (3/29 resistant isolates, 10.34%).
 - Piperacillin/tazobactam: *K. pneumoniae* when the MIC is 16 µg/mL due to the occurrence of minor errors, that were in essential agreement, resulting in a category agreement below 90%.
 4. Testing with the LifeScale AST system should not be performed on polymicrobial samples.
 5. The LifeScale LSGN Kit showed unacceptable performance for:
 - *E. coli* with Ertapenem when tested using the following blood culture bottle types: bioMérieux Standard Anaerobic, BACTEC Standard Anaerobic and VersaTREK REDOX 1 Aerobic. Use alternate bottle types for determining Ertapenem MICs for *E. coli*.
 - *A. baumannii* with Amikacin when tested using the following blood culture bottle types: bioMérieux Standard Aerobic and VersaTREK REDOX 1 Aerobic. Use alternate bottle types for determining Amikacin MICs for *A. baumannii*.
 6. Media equivalency was not determined for Trimethoprim-sulfamethoxazole/*E. coli* due to a lack of available isolates with on-scale MICs.

7. Interference has not been established for the following drug/organism combinations:
 - All interferents: Ceftazidime/*A. baumannii*
 - Platelets and heparin: Cefazolin/*K. pneumoniae*
8. Potential interference by antimicrobial agents that may be present in a patient blood specimen has not been established with the Affinity LifeScale LSGN Kit. Please use caution when interpreting results if information is available about the patient treatment with antimicrobial agents.
9. The performance of the LifeScale LSGN panel has been evaluated using the following blood culture bottles:
 - BD BACTEC: Standard Aerobic, Standard Anaerobic, Plus Aerobic, Lytic Anaerobic
 - BacT/ALERT: Standard Aerobic, Standard Anaerobic
 - VersaTREK: REDOX 1 Aerobic media, REDOX 2 Anaerobic media
10. The use of the LifeScale AST system does not eliminate the need for subculture of the positive blood culture.
11. If the subculture (purity) plate indicates the sample is polymicrobial, the AST results should be voided, and susceptibility testing on each isolate using an alternative method with standard inoculum preparation should be performed.
12. Positive blood cultures must be processed immediately on the LifeScale AST system or within 12 hours of blood culture bottle positivity should delays be unavoidable.

D Special Instrument Requirements:

The LifeScale Gram Negative Kit (LSGN) assay is performed on the LifeScale instrument with Software version 2.3.58.

IV Device/System Characteristics:

A Device Description:

Refer to the [K211815](#) Decision Summary.

Table 1 provides the reportable MIC ranges and breakpoints for antimicrobial agents claimed for testing with the LifeScale System.

Table 1. Reportable MIC Ranges and Species-Specific Breakpoints for Antimicrobials Included in the LifeScale System.

Antimicrobial	Indicated Species	LifeScale Reportable Range (µg/mL)	FDA-Recognized Breakpoints			
			S	SDD	I	R
Ampicillin	<i>E. coli</i>	≤2 - >64	≤8		16	≥32
Aztreonam	<i>E. coli</i> , <i>K. pneumoniae</i> , <i>K. aerogenes</i> , <i>K. oxytoca</i>	≤1 - >64	≤4		8	≥16
Cefazolin	<i>E. coli</i> , <i>K. pneumoniae</i> , <i>K. variicola</i>	≤0.25 - >16	≤2		4	≥8

Antimicrobial	Indicated Species	LifeScale Reportable Range (µg/mL)	FDA-Recognized Breakpoints			
			S	SDD	I	R
Ceftazidime	<i>Acinetobacter</i> spp. (other than <i>Acinetobacter ursingii</i>)	≤1 - >64	≤8		16	≥32
	<i>E. coli</i> , <i>K. aerogenes</i> , <i>K. oxytoca</i> , <i>K. variicola</i>		≤4		8	≥16
	<i>P. aeruginosa</i>		≤8		-*	≥16
Ertapenem	<i>E. coli</i> , <i>K. aerogenes</i> , <i>K. pneumoniae</i> , <i>K. oxytoca</i>	≤0.125 - >8	≤0.5		1	≥2
Trimethoprim/ Sulfamethoxazole	<i>E. coli</i> , <i>K. aerogenes</i> , <i>K. oxytoca</i> , <i>K. variicola</i>	≤0.25 - >8	≤2/38		-*	≥4/76
Amikacin	<i>Acinetobacter</i> spp., <i>E. coli</i> , <i>K. pneumoniae</i> , <i>K. aerogenes</i> , <i>K. oxytoca</i> , <i>K. variicola</i> , <i>P. aeruginosa</i>	≤4 - >256	≤16		32	≥64
Cefepime	<i>E. coli</i> , <i>K. pneumoniae</i> , <i>K. aerogenes</i> , <i>K. oxytoca</i>	≤0.5 - >64	≤2	4-8	-*	≥16
	<i>P. aeruginosa</i>		≤8		-*	≥16
Ceftazidime/ avibactam	<i>E. coli</i> , <i>K. aerogenes</i> , <i>K. oxytoca</i>	≤2/4 - >32/4	≤8/4		-*	≥16/4
Gentamicin	<i>E. coli</i> , <i>K. pneumoniae</i> , <i>K. aerogenes</i> , <i>K. oxytoca</i> , <i>K. variicola</i> , <i>P. aeruginosa</i>	≤1 - >32	≤4		8	≥16
Levofloxacin	<i>E. coli</i> , <i>K. pneumoniae</i> , <i>K. aerogenes</i> , <i>K. oxytoca</i>	≤0.25 - >16	≤0.5		1	≥2
	<i>P. aeruginosa</i>		≤1		2	≥4
Meropenem	<i>E. coli</i> , <i>K. pneumoniae</i> , <i>K. aerogenes</i>	≤0.12 - >16	≤1		2	≥4
	<i>Acinetobacter</i> spp., <i>P. aeruginosa</i>		≤2		4	≥8
Meropenem/ vaborbactam	<i>E. coli</i> , <i>K. pneumoniae</i> , <i>K. aerogenes</i> , <i>K. oxytoca</i>	≤0.5 - >16	≤4/8		8/8	≥16/8
Piperacillin/ tazobactam	<i>E. coli</i> , <i>K. pneumoniae</i>	≤4 - >256	≤8/4		16/4	≥32/4
	<i>P. aeruginosa</i>		≤8/4	16/4	-*	≥32/4
	<i>Acinetobacter</i> spp.		≤16/4		32/4- 64/4	≥128/ 4

S = Susceptible; SDD = Susceptible-dose dependent; I = Intermediate; R = Resistant

*No intermediate category is defined for these drug/organism combinations. For some drug organism combinations, the SDD is defined instead of an intermediate category.

B Principle of Operation:

Refer to the [K211815](#) Decision Summary.

C Instrument Description Information:

Refer to the [K211815](#) Decision Summary.

Quality Control:

Quality control (QC) is performed by the operator using manufacturer-specified organisms (*Escherichia coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853, *Klebsiella pneumoniae*

ATCC 700603, and *Enterobacter cloacae* ABGNQC1.) appropriate for each antimicrobial agent. The QC AST reports “out of range” for any MIC results that are not consistent with expected MIC QC ranges.

V Substantial Equivalence Information:

A Predicate Device Name(s):

LifeScale Gram Negative Kit (LSGN) with the LifeScale AST System

B Predicate 510(k) Number(s):

K211815

C Comparison with Predicate(s):

Device & Predicate Device(s):	<u>K241324</u> (New Device)	<u>K211815</u> (Predicate Device)
Device Trade Name	Lifescale Gram Negative Kit (LSGN) with the LifeScale AST system	Same
General Device Characteristic Similarities		
Intended Use/Indications For Use	The LifeScale AST system is a multiplexed <i>in vitro</i> diagnostic test that uses a microfluidic sensor and resonant frequency to calculate organism concentration and/or mass distribution for quantitative antimicrobial susceptibility testing (AST). Testing is performed directly on blood cultures signaled as positive by a continuous monitoring blood culture system and confirmed by Gram stain. The LifeScale AST system does not provide organism identification and is not indicated for use with polymicrobial samples. Interpretive results (Susceptible/Intermediate/Susceptible-dose dependent/Resistant) are provided for specific drug/organism combinations. Results are intended to be used in conjunction with other clinical and laboratory findings. Standard laboratory protocols for processing positive blood cultures should be followed to ensure availability of isolates for supplemental testing as needed. Additionally, subculture of positive blood culture is necessary for the susceptibility testing of organisms present in polymicrobial samples, for testing antimicrobial agents and species not indicated for testing with the device, for epidemiologic testing and for recovery of organisms present in microbial samples. The LifeScale Gram Negative Kit (LSGN) is intended for use with the LifeScale AST system for <i>in vitro</i> testing of positive blood	Same

	culture samples confirmed by Gram stain as containing gram-negative bacilli.	
Sample	Blood cultures signaled as positive by a continuous monitoring blood culture system.	Same
Inoculation Method	Automated	Same
Read Method	Automated	Same
Result Report	Report results as a minimum inhibitory concentration (MIC) and categorical interpretation (S, I/SDD, R)	Same
Sample Preparation	Centrifugation and pipetting of sample	Same
IVD Functions	AST	Same
Instrument	LifeScale AST system	Same
Technology	Microfluidic and resonant frequency to calculate organism concentration and/or mass distribution	Same
Organisms Tested	Gram-negative bacilli	Same
General Device Characteristic Differences		
Antimicrobial Agents	Amikacin Cefepime Ceftazidime/avibactam Gentamicin Levofloxacin Meropenem Meropenem/vaborbactam Piperacillin/tazobactam	Ampicillin Aztreonam Cefazolin Ceftazidime Ertapenem Trimethoprim/sulfamethoxazole

VI Standards/Guidance Documents Referenced:

- FDA Class II Special Controls Guidance Document: Antimicrobial Susceptibility (AST) Systems; Guidance for Industry and FDA (Issued August 28, 2009).
- CLSI M100-Ed 34, *Performance Standards for Antimicrobial Susceptibility Testing*; 2024.
- CLSI M100-Ed 33, *Performance Standards for Antimicrobial Susceptibility Testing*; 2023.
- CLSI M100-Ed 32, *Performance Standards for Antimicrobial Susceptibility Testing*; 2022.
- CLSI M07. 11th ed. *Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically*. 2018.
- CLSI M07. 12th ed. *Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically*. 2024.
- FDA STIC Website FDA-Recognized Antimicrobial Susceptibility Test Interpretive Criteria
- IEC 60601-1-2 Edition 4.0 2014-02, Medical electrical equipment - Part 1-2: General requirements for basic safety and essential performance
- IEC 61010-1 Edition 3.1 2017-01, Safety requirements for electrical equipment for measurement, control, and laboratory use

- IEC 62304 Edition 1.1 2015-06 Consolidated Version, Medical device software - Software life cycle processes

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

The reproducibility study data were provided to support the addition of antimicrobials, such as Amikacin, Cefepime, Ceftazidime/avibactam, Gentamicin, Levofloxacin, Meropenem, Meropenem/vaborbactam and Piperacillin/tazobactam with the LifeScale LSGN Kit on the LifeScale AST system. The study was conducted in two phases: Initial and Supplement. Initial study was conducted at three sites (TR1, TR2 and TR3) testing 26 isolates (strains) with all eight antimicrobials. Since an insufficient number of isolates were evaluated for Cefepime, Ceftazidime/avibactam, and Meropenem, and best-case and worst-case reproducibility for Piperacillin/tazobactam were <95% and <90%, respectively, a supplemental study was performed using appropriate, on-scale isolates. The supplement study was conducted at three sites (AB1, AB2, and AB3) testing 18 isolates (strains) with Cefepime, Ceftazidime/avibactam, Meropenem, and Piperacillin/tazobactam. Data from the supplemental testing were combined with the data from the initial study.

Best-case ($\geq 95\%$) and worst-case ($\geq 89\%$) reproducibility were acceptable for all antibiotics tested (Amikacin, Cefepime, Ceftazidime/avibactam, Gentamicin, Levofloxacin, Meropenem, Meropenem/vaborbactam, and Piperacillin/tazobactam) for the combined datasets. Results of the reproducibility are summarized in Table 2.

Table 2. Summary of results by antibiotic with best and worst-case reproducibility across all sites.

Antimicrobial	Best case #	Best case %	Worst case #	Worst case %
Amikacin	468/481	97.3%	450/481	93.6%
Cefepime	504/509	99.0%	459/509	90.2%
Ceftazidime/avibactam	345/350	98.6%	339/350	96.9%
Gentamicin	503/509	98.8%	466/509	91.6%
Levofloxacin	373/373	100%	361/373	96.8%
Meropenem	508/534	95.1%	477/534	89.3%
Meropenem/vaborbactam	274/287	95.5%	261/287	90.9%
Piperacillin/tazobactam	555/566	98.1%	521/566	92.0%

2. Inoculum Density Study:

An inoculum density study was performed to determine the effect of organism concentration in a positive blood culture on the performance of the LifeScale LSGN Kit and the ability of the LifeScale AST instrument to prepare an appropriate inoculum regardless of the starting concentration. By design, the LifeScale AST instrument automatically determines the concentration of cells in the suspension and will calculate the dilution needed to inoculate the LSGN plate at a target concentration of 4.5×10^5 CFU/mL. The organism concentration in each contrived sample was confirmed by colony count; all colony counts corresponded to the

target organism concentration ranges of 10^4 (for a subset of *P. aeruginosa* samples), 10^6 and 10^9 CFU/mL. Contrived samples containing very low (10^4 CFU/mL) concentrations of *P. aeruginosa* were tested to confirm that the LifeScale AST system will terminate testing and generate an invalid result if sufficient growth is not detected. All samples were tested in triplicate with the LifeScale LSGN Kit and the LifeScale system. AST results were compared to the mode MIC result determined by the reference method.

A minimum of 9 isolates (strains) were tested for each antimicrobial to cover each claimed drug/organism combination. Contrived samples were prepared with target organism concentrations at 10^6 CFU/mL and 10^9 CFU/mL of representative species. To address the observed poor performance issues for Piperacillin-tazobactam/Enterobacterales, a supplemental study was conducted testing 7 isolates (strains) at 10^6 CFU/mL for Piperacillin/tazobactam. MIC results for samples containing 10^6 and 10^9 CFU/mL showed good agreement (EA \geq 90%) for claimed drug/organism combinations for the LifeScale LSGN Kit as compared to the reference method results regardless of the starting organism concentration. No results were obtained for samples containing 10^4 CFU/mL of *P. aeruginosa*; all tests were terminated due to insufficient growth, confirming that low organism concentrations in the sample will result in an invalid test.

Analysis of trending at each tested concentration showed a few instances of trending (e.g., high or low) as compared to the reference method for the following antimicrobial/organism combinations at the specified organism concentrations of the positive blood culture samples:

The following drug/organism combinations showed high trending:

- Amikacin/*E. coli* at 10^9 CFU/mL
- Cefepime/*E. coli* at 10^6 CFU/mL and 10^9 CFU/mL
- Gentamicin/*E. coli* at 10^9 CFU/mL
- Gentamicin/*P. aeruginosa* at 10^9 CFU/mL
- Piperacillin-tazobactam/*E. coli* at 10^9 CFU/mL

The following drug/organism combinations showed low trending:

- Ceftazidime-avibactam/*E. coli* at 10^6 CFU/mL

To address the overall trending observed in the clinical and analytical studies, the following general statement was added to the device labeling.

“In the clinical study or in the Inoculum Density analytical study, the majority of drug/organism combinations tested with the LifeScale LSGN kit showed MIC values equal to or at least one doubling dilution higher/lower than the reference method. Use caution when reporting drug resistance for any antimicrobial.”

The following drug/organism combinations showed high trending:

- Amikacin: *E. coli*, *K. aerogenes*, *K. oxytoca*, *K. pneumoniae*, *K. variicola*, *Acinetobacter* spp., *P. aeruginosa*
- Cefepime: *E. coli*, *K. aerogenes*, *K. oxytoca*, *K. pneumoniae*
- Ceftazidime-avibactam: *K. aerogenes*, *K. oxytoca*

- *Gentamicin: E. coli, K. aerogenes, K. oxytoca, K. pneumoniae, P. aeruginosa*
- *Levofloxacin: K. pneumoniae, K. oxytoca, K. aerogenes, P. aeruginosa*
- *Meropenem: K. oxytoca, P. aeruginosa*
- *Meropenem-vaborbactam: K. pneumoniae, K. oxytoca, K. aerogenes*
- *Piperacillin-tazobactam: E. coli, K. pneumoniae*

The following drug/organism combinations showed low trending:

- *Ceftazidime-avibactam: E. coli*
- *Meropenem-vaborbactam: E. coli*
- *Piperacillin-tazobactam: A. baumannii*

3. Linearity:

Not applicable

4. Analytical Specificity/Interference:

An interfering substances study was performed to determine the effect of potential interferents in blood cultures on results obtained with the LifeScale LSGN Kit. BACTEC Standard Aerobic Media was seeded with strains of *E. coli*, *K. pneumoniae*, *A. baumannii* and *P. aeruginosa*; samples included: 1) bottles seeded with organism and the potential interferent and 2) control bottles seeded with organism and no interferent. Endogenous substances were evaluated as possible interferents in concentrations equivalent to concentrations found in human blood (Table 3). Testing of seeded bottles with and without interferent was performed in triplicate. AST performance in the presence of potential interferents was determined by comparison of the EA of MIC results obtained from samples with and without interferents. Comparisons were performed by analysis of MIC mode of the control tests compared with the MIC of each replicate containing interferent.

Table 3. Endogenous Interferents, Concentration Tested

Interfering substances	Concentration Tested	Reference Range in Human Blood
Conjugated bilirubin	0.003 mg/mL	0-0.002 mg/mL
Unconjugated bilirubin	0.003 – 0.012 mg/mL	0.002 mg/mL
Gamma globulin	6 - 13 mg/mL	6-13 mg/mL
Hemoglobin	100 mg/mL	100-200 mg/mL
Triglycerides	5 mg/mL	1.5-5 mg/ml
White blood cells	4.5 x10 ⁶ -1.0 x10 ⁷ cells/mL	4.5 x10 ⁶ -1.0 x10 ⁷ cells/mL
Platelets	≥450,000 platelets/μL	150,000 -400, 000/μl
Heparin	330 units/dL	0.35 – 1 U/mL

For all the drug/organism combinations tested, no interference was noted for any potential interferent as determined by LifeScale MIC comparison with reference results.

The effect of antimicrobial agents as interferent was not evaluated. The following limitation included in the original 510(k) pre-market submission (K211815), should remain in the device labeling:

Potential interference by antimicrobial agents that may be present in a patient blood specimen has not been established with the Affinity LifeScale LSGN kit. Please use caution

when interpreting results if information is available about the patient treatment with antimicrobial agents.

5. Media Equivalency Study:

A media equivalency study was conducted to assess compatibility of the LifeScale LSGN Kit with various commonly used blood culture bottle types. Eight blood culture bottle types from three different blood culture manufacturers [bioMérieux, Inc. (BacT/Alert: Standard Aerobic, Standard Anaerobic), Becton Dickinson (BACTEC: Standard Aerobic, Standard Anaerobic, Lytic Anaerobic, Plus Aerobic), and ThermoFisher (VersaTREK: REDOX 1 Aerobic, REDOX 2 Anaerobic)] were evaluated analytically with the LifeScale LSGN Kit. A total of 23 well-characterized isolates (strains) of *E. coli*, *K. pneumoniae*, *A. baumannii* and *P. aeruginosa* were inoculated into each blood culture bottle type; ten replicates of each resulting positive blood culture were tested using the LifeScale LSGN Kit at the time of positivity. BACTEC bottles were incubated in a continuous monitoring blood culture instrument; other bottle types were incubated in a conventional incubator with or without mixing per the bottle instructions for use. The time to positivity was estimated for the BacT/Alert and VersaTREK blood culture bottles based on time to positivity for BACTEC media and confirmed by LifeScale AST system cell counts.

In the original 510(k) pre-market submission (K211815), the MIC results for each bottle type with each antimicrobial/organism combination were compared to the mode MIC value obtained with the broth microdilution reference method. However, the current 510(k) pre-market submission follows a different comparison approach. Since all other analytical studies used a single bottle type (i.e., BD BACTEC Standard Aerobic Media), and all bottle types were tested in the clinical study, the individual results from other bottle types were directly compared to the LifeScale AST mode MIC for this specific bottle type (i.e., the “comparator”) directly. The acceptance criteria of EA \geq 95% for each antimicrobial/bottle type was applied to this direct comparison approach.

Results for the majority of drug/organism combinations for claimed species showed good performance with all blood culture bottles with the following exceptions:

- Results obtained with the combination of Amikacin/*A. baumannii* with bioMérieux Standard Aerobic and VersaTREK REDOX 1 Aerobic Media showed low EA when compared to the comparator, BD BACTEC Standard Aerobic Media.

To address the results obtained with Amikacin/*A. baumannii* the following limitation was included in the device labeling:

The LifeScale LSGN kit showed unacceptable performance for A. baumannii with Amikacin when tested using the following blood culture bottle types: bioMérieux Standard Aerobic and VersaTREK REDOX 1 Aerobic. Use alternative bottle types for determining Amikacin MICs for A. baumannii.

6. Assay Reportable Range:

Not Applicable

7. Traceability, Stability, Expected Values (Controls, Calibrators, or Methods):

Quality control testing was performed each day that testing was conducted during the clinical and analytical studies. Quality control samples were prepared from isolated colonies as described in Section IVA above (Device Description). Gram-negative CLSI-recommended quality control strains were tested a minimum of 20 times using both the LifeScale LSGN Kit and the reference broth microdilution method at all sites during the clinical studies.

For Cefepime and Meropenem, the LifeScale LSGN panel did not include the full CLSI expected range for *P. aeruginosa* ATCC 27853. To address the use of this strain in QC testing the following footnote was added to the QC table:

Cefepime and Meropenem: Does not include the full CLSI expected range

For all antimicrobials, greater than 95% of results obtained during the clinical study were within the expected range; quality control results were acceptable (Table 4 below).

Table 4. QC Results for the LifeScale LSGN Kit, Clinical Study

Antimicrobial	QC Strain	Expected Range µg/mL	No. in Range/No. Tested (%)	
			Reference	LifeScale
Amikacin	<i>E. cloacae</i> ABGNQC1 ^a	8 - 32	23/23 (100)	117/117 (100)
Cefepime	<i>P. aeruginosa</i> ATCC 27853 ^b	≤0.5 - 4	111/111 (100)	241/241 (100)
Ceftazidime/avibactam	<i>E. cloacae</i> ABGNQC1 ^{a, b}	≤2/4 - 8/4	23/23 (100)	117/117 (100)
Gentamicin	<i>E. cloacae</i> ABGNQC1 ^a	8 - 32	23/23 (100)	116/116 (100)
Levofloxacin	<i>P. aeruginosa</i> ATCC 27853	0.5 - 4	107/111 (96.4)	241/242 (99.6)
Meropenem	<i>P. aeruginosa</i> ATCC 27853	≤0.12 - 1	112/112 (100)	238/242 (98.3)
Meropenem/vaborbactam	<i>E. cloacae</i> ABGNQC1 ^a	4/8 - 16/8	23/23 (100)	115/117 (98.3)
Piperacillin/tazobactam	<i>K. pneumoniae</i> ATCC 700603	8/4 - 32/4	135/135 (100)	418/418 (100)

^a Validation performed for non-CLSI recommended QC strain.

^b Does not include full CLSI expected range. Results were considered acceptable.

Validation of *Enterobacter cloacae* ABGNQC1 as a new QC Organism

To validate the use of *E. cloacae* ABGNQC1 as a QC organism with the LifeScale LSGN Kit on the LifeScale AST system, a total of 40 replicates were tested by at least two operators per media lot, spanning over a period of at least three days. A minimum of two media lots were tested, resulting in at least 80 data points using the LifeScale AST and the CLSI Reference Broth Microdilution Method. This validation study was done to establish expected QC MIC ranges for Amikacin, Ceftazidime/avibactam, Gentamicin, and Meropenem/vaborbactam. In

the clinical study, an additional 92 data points were tested by BMD for a total of 467 data points with >95% in-range, which supported the expected QC ranges of *E. cloacae* ABGNQC1 for Amikacin, Ceftazidime/avibactam, Gentamicin, and Meropenem/vaborbactam in the LifeScale AST clinical study. Taken together, these data are acceptable to support the use of *E. cloacae* ABGNQC1 as a QC organism when testing Amikacin, Ceftazidime/avibactam, Gentamicin, and Meropenem/vaborbactam on the LifeScale AST.

Inoculum Density Check: Organism concentration determined automatically by the instrument. Refer the Inoculum Density study above.

LifeScale LSGN Tests Initiated and Failed to Report a Result. Overall, 2.38% of tests initiated during the analytical and clinical studies failed to provide a result (Table 5).

Table 5. LifeScale LSGN Tests Initiated and Failed to Report a Result

Reason for Exclusion/Incomplete Test	No. Exclusion or Incomplete Test/Total No of Tests (%)			
	Clinical Study	Analytical Studies	QC	Overall
Plate Failures^a	5/986 (0.51)	15/3307 (0.45)	0/1871 (0.00)	20/6164 (0.32)
Growth Failures	6/986 (0.61)	17/3307 (0.51)	0/1871 (0.00)	23/6164 (0.37)
LifeScale Failures^b	4/986 (0.41)	50/3307 (1.51)	15/1871 (0.80)	69/6164 (1.12)
Other Reasons^c	4/986 (0.41)	31/3307 (0.94)	0/1871 (0.00)	35/6164 (0.57)
Total Excluded/Incomplete Tests	19/986 (1.93)	113/3307 (3.42)	15/1871 (0.80)	147/6164 (2.38)

^a Plate failures include: unable to verify positive controls, sensor clog detected, system unable to calculate MIC.

^b LifeScale failures include: LifeScale system software and hardware failures

^c Other Reasons include: operator error, incubation time greater than 8 hours, user canceled, protocol error

Purity Check: Purity checks were performed for all clinical and analytical tests. Testing was not performed on samples with mixed growth or contamination.

8. Sample Stability Study:

A sample stability study was conducted to demonstrate that positive blood cultures tested at 12 hours post positivity (T₁₂) and held at incubation temperature (35 °C) are equivalent to results obtained at the time of positivity (T₀).

Contrived positive blood culture specimens (prepared in BD Standard Aerobic blood culture bottles) containing the recommended blood volume were tested within one hour of bottle ring (T₀) and at 13 ± 0.5 hours post bottle ring (T₁₃). Testing at 13 hours was performed to support the stability at 12 hours post bottle ring as listed in the device labeling. Testing was performed in triplicate for each sample at each timepoint. Resulting MICs were compared to the modal LifeScale MIC determined at T₀. The stability acceptance criterion was ≥ 95% agreement for EA. QC was performed each day of testing.

A total of thirteen (13) gram-negative organisms with known MICs as determined by broth microdilution testing were evaluated. Species tested included *A. baumannii* (2), *E. coli* (6), *K. pneumoniae* (3), and *P. aeruginosa* (2). Table 6 shows the EA for each organism/antimicrobial combination and the overall EA for all species tested for each antimicrobial in the sample stability study. Results for Cefepime, Ceftazidime/avibactam, Gentamicin, Levofloxacin and Piperacillin/tazobactam were acceptable, indicating that results obtained at T₁₃ were equivalent to MIC results obtained at T₀. The EA for Amikacin, Meropenem, Meropenem/vaborbactam were not acceptable (EA <95%; Table 6). When comparing the LifeScale MICs at T₀ and T₁₃ to the BMD reference mode MIC, the overall EA for these antimicrobials resulted was 100%. This is noted in a footnote in the device labeling.

Table 6. Summary of sample stability results (MIC T=13 vs. Mode MIC T=0)

Antibiotic	No. Samples Tested	Data Points		LifeScale T ₁₃ to LifeScale Mode T ₀
		T=0	T=13	No. EA/Total Tested (EA%)
Amikacin	13	39	39	36/39* (92.3%)
Cefepime	11	33	33	31/32 (96.9%)
Ceftazidime/avibactam	11	33	33	33/33 (100.0%)
Gentamicin	11	33	33	33/33 (100.0%)
Levofloxacin	11	33	33	33/33 (100.0%)
Meropenem	13	39	39	36/39* (92.3%)
Meropenem/vaborbactam	9	27	27	24/27* (88.9%)
Piperacillin/tazobactam	13	39	39	38/39 (97.4%)
Total	92	276	276	264/275 (96.0%)

* Comparison to the BMD reference mode MIC resulted in an EA of 100%.

The study demonstrates that results from samples tested within one hour after bottle ring provide equivalent results to those tested at 13 hours after bottle ring when incubated in the blood culture monitoring system at 35 °C.

To address the timing of positive blood culture processing, the following limitation included in the original 510(k) pre-market submission (K211815), should remain in the device labeling:

Positive blood cultures must be processed immediately on the LifeScale AST system or within 12 hours of blood culture bottle positivity should delays be unavoidable.

9. Off-line and On-line Incubation Study:

An off-line and on-line incubation study was conducted to demonstrate that results obtained for the LifeScale LSGN plates incubated in an off-line incubator (a non-CO₂ laboratory incubator with a temperature of 35°C ± 2° C) are accurate (as determined by comparison to the reference method) to results obtained for LSGN plates incubated on the integrated thermal stage in the LifeScale AST instrument (on-line). In addition, the study evaluated the effect of an 8-hour incubation on the final MIC (for plates held in an off-line incubator for an additional 2 hours after the maximum incubation time prior to measurement). The LifeScale instrument has the capacity for incubating a single LSGN plate on-line; additional plates processed within that plate's incubation period must be incubated in an off-line incubator. For

plates incubated on-line, the system monitors growth in the positive control well through measurements of cell concentration after a minimum of three hours of incubation. For plates incubated in an off-line incubator, measurement of the control well takes place at baseline (time 0) and again when replaced in the instrument after 3 hours of incubation. The system tracks the inoculated plates and alerts the user when a plate must be retrieved from the offline incubator and placed on the instrument stage for measurement.

For this study a set of challenge strains (including *E. coli*, *K. pneumoniae*, *A. baumannii* and *P. aeruginosa*) were inoculated into a BD BACTEC blood culture bottle and incubated until positivity. At positivity, the blood culture was processed in accordance with the LifeScale AST system's instructions for use, inoculated to LifeScale LSGN plates and tested in triplicate under the following incubation conditions:

- Three hours incubation on-line
- Three hours incubation off-line
- Six hours incubation off-line
- Eight hours incubation off-line

LifeScale LSGN Kit results were compared to results obtained with the reference method. LifeScale LSGN Kit results as compared to the reference method from samples evaluated in the clinical study (which included samples incubated both on- and off-line) were also evaluated.

Results of the study indicate that on-line and off-line incubation under the above conditions provided acceptable results for Amikacin, Cefepime, Ceftazidime/avibactam, Gentamicin, Levofloxacin, Meropenem, Meropenem/vaborbactam and Piperacillin/tazobactam.

Additionally, the current submission provided additional data for both on-line and off-line incubation options, for ertapenem. The performance for both incubation options was acceptable, and the limitation above for off-line incubation of Ertapenem/*E. coli* was removed from the device labeling.

10. Detection Limit:

Not Applicable

11. Assay Cut-Off:

Not Applicable

12. Accuracy (Instrument):

Not Applicable

13. Cross Contamination/Carry-Over:

Please refer to K211815 for the cross contamination/carry-over study results for the LifeScale AST device.

B Comparison Studies:

1. Method Comparison with Predicate Device:

Clinical performance testing with the LifeScale AST system with the LifeScale LSGN Kit was conducted in two phases: Initial and Supplement. The initial testing was performed at six geographically diverse U.S. clinical sites testing fresh (prospective) positive blood cultures as well as positive blood cultures contrived with clinical isolates (seeded clinical). Positive blood cultures contrived with challenge organisms, including resistant isolates and isolates with on-scale MIC results, were also tested at the clinical sites. The supplement testing aimed to 1) supplement the initial testing data to support the claims of adding additional antimicrobials (i.e., Amikacin, Cefepime, Ceftazidime/avibactam, Gentamicin, Levofloxacin, Meropenem, Meropenem/vaborbactam and Piperacillin/tazobactam) with the LifeScale AST system; 2) the use of a validated new Gram-Negative Quality Control strain (*Enterobacter cloacae* ABGNQC1); and 3) support the removal of limitations included in the cleared LifeScale LSGN Kit with the LifeScale AST system (K211815) for Aztreonam, Ceftazidime, Ertapenem, and Cefazolin. The supplement testing was performed at six U.S. clinical sites testing a limited number of prospective positive clinical blood cultures as well as positive blood cultures contrived with clinical isolates (seeded clinical) and challenge isolates. In total, 1644 positive blood cultures were tested across both phases: 307 fresh (prospective), 958 seeded with fresh or stock isolates, and 379 seeded with challenge isolates, including resistant isolates and isolates with on-scale MIC results.

LifeScale LSGN Kit testing was performed per instructions in the device labeling. Plates were incubated on-line (in the LifeScale instrument) or off-line (in an auxiliary incubator). Incubation times for all drug/organism combinations averaged 3.5 hours.

Pure bacterial isolates used for contriving blood cultures as well as those isolates subcultured from prospective positive blood cultures were sent to a central reference laboratory in the U.S. for identification verification using an FDA-cleared MALDI-TOF system. AST testing was performed using the reference broth microdilution (BMD) method, as described in the CLSI document *Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria that Grow Aerobically*, M07, 11th and 12th editions. Reference BMD panels were run in triplicate for each isolate and an MIC mode was determined for comparison with the LifeScale LSGN Kit MIC result. If an MIC mode could not be established from the first three BMD results, a second set of BMD assays was run in triplicate and the MIC mode across all six tests was determined. If a mode still could not be established, the median MIC was used for comparison with the LifeScale LSGN Kit MIC result.

LifeScale LSGN Kit performance was based on criteria outlined in the *Class II Special Controls Guidance Document: Antimicrobial Susceptibility Test (AST) Systems*. Performance criteria included essential agreement (EA) and categorical agreement (CA) and the number and percent of categorical errors (minor, major and very major errors). EA was calculated as the percentage of MIC results that fell within ± 1 doubling dilution of the reference result; CA was calculated as the percentage of LifeScale LSGN Kit interpretive results (S/I/SDD/R) that were identical to the interpretive categories of the reference result. For drug/organism group combinations where the susceptible-dose dependent category is recognized in place of

the intermediate category, any errors that were observed with this category were designated as minor errors.

Acceptable results were obtained with the drug/organism combinations outlined below. Results for drug/organism combinations not listed were not acceptable. To address the use of the device for drug/organism combinations that provided acceptable results, the following limitation was included in the device labeling:

Only the following drug/organism combinations have been shown to provide acceptable results with the LifeScale LSGN Kit:

- *Amikacin: Acinetobacter spp., E. coli, K. pneumoniae, K. aerogenes, K. oxytoca, K. variicola, P. aeruginosa*
- *Cefepime: E. coli, K. pneumoniae, K.aerogenes, K. oxytoca*
- *Ceftazidime/avibactam: E. coli, K.aerogenes, K. oxytoca*
- *Gentamicin: E. coli, K. pneumoniae, K.aerogenes, K. oxytoca, K. variicola, P. aeruginosa*
- *Levofloxacin: E. coli, K. pneumoniae, K.aerogenes, K. oxytoca, P. aeruginosa*
- *Meropenem: Acinetobacter spp., E. coli, K. pneumoniae, K. oxytoca, P. aeruginosa*
- *Meropenem/vaborbactam: E. coli, K. pneumoniae, K.aerogenes, K. oxytoca*
- *Piperacillin/tazobactam: Acinetobacter spp., E. coli, K. pneumoniae, P. aeruginosa*

Perform an alternative method of testing prior to reporting results for the following antimicrobial/organism combination(s):

- *Cefepime: P. aeruginosa when the MIC is 4 µg/mL due the occurrence of very major errors (3/29 resistant isolates, 10.34%)*
- *Ceftazidime/avibactam: K. pneumoniae, P. aeruginosa*
- *Meropenem: K. oxytoca when the LifeScale MIC is 0.5 µg/mL due to two very major discrepancies*
- *Piperacillin/tazobactam: K. pneumoniae when the MIC is 16 µg/mL due to the occurrence of minor errors, that were in essential agreement, resulting in a category agreement below 90%*

Positive blood culture samples containing more than one organism should not be tested with the LifeScale AST system. The following limitations are included in the device labeling:

Testing with the LifeScale AST system should not be performed on polymicrobial samples.

If the subculture (purity) plate indicates the sample is polymicrobial, the AST results should be voided, and susceptibility testing on each isolate using an alternative method with standard inoculum preparation should be performed.

A summary of the performance of LifeScale LSGN Kit is described below for each antimicrobial agent with indicated species. Complete details including EA, CA and error rate analysis per organism group are summarized in Table 7.

Amikacin/Enterobacterales. A total of 480 Enterobacterales samples were evaluated with Amikacin [*E. coli* (140), *K. aerogenes* (52), *K. pneumoniae* (142), *K. oxytoca* (111), *K. variicola* (35)]. The combined results from clinical and challenge sample testing demonstrated an EA of 97.3% and CA of 95.8%. There were 3 major errors (0.8%) and no very major errors.

Amikacin/*Acinetobacter* spp. A total of 77 *Acinetobacter* spp. samples were evaluated with Amikacin [*A. baumannii* (21), *A. baumannii/nosocomialis* group (37) and other rare *Acinetobacter* species (19) including *A. lwoffii*, *A. pittii*, *A. calcoaceticus* species, *A. radioresistens* and *A. ursingii*]. The combined results from clinical and challenge sample testing demonstrated an EA of 100% and CA of 97.4%. There were no major or very major errors.

Amikacin/*P. aeruginosa*. A total of 59 *P. aeruginosa* samples were evaluated with Amikacin. The combined results from clinical and challenge sample testing demonstrated an EA of 98.3% and CA of 94.9%. There were no major or very major errors.

Cefepime/Enterobacterales. A total of 445 Enterobacterales samples were evaluated with Cefepime [*E. coli* (139), *K. aerogenes* (53), *K. pneumoniae* (142), *K. oxytoca* (111)]. The combined results from clinical and challenge sample testing demonstrated an EA of 92.6% and CA of 95.7%. There were 3 major errors (1.1%) and no very major errors.

Cefepime/*P. aeruginosa*. A total of 101 *P. aeruginosa* samples were evaluated with Cefepime. The combined results from clinical and challenge sample testing demonstrated an EA of 93.1% and CA of 84.2%. Though it showed the CA below 90%, it is acceptable as it demonstrated an Evaluable EA of 91.0%. There were 13 major errors (18.1%) and 3 very major errors (10.3%). Due to a lack of an intermediate breakpoint, further analysis of the errors was performed, and adjustments were made by considering the MIC values of the errors compared to the reference MIC value. Eleven of the major errors were in essential agreement with the reference MIC value. Therefore, the major error rate and very major error rate were adjusted to 2.8% (2/72) and 10.3% (3/29), respectively, which was still unacceptable. To address this unacceptable very major error rate, the following limitation is included in the device labeling:

Perform an alternative method of testing prior to reporting results for:

- *Cefepime: P. aeruginosa when the LifeScale MIC is 4 µg/mL due to the occurrence of very major errors (3/29 resistant isolates, 10.34%)*

Ceftazidime-avibactam/Enterobacterales. A total of 303 Enterobacterales samples were evaluated with Ceftazidime/avibactam [*E. coli* (139), *K. aerogenes* (53), *K. oxytoca* (111)]. The combined results from clinical and challenge sample testing demonstrated an EA of 99.0% and CA of 99.0%. There were 3 major errors (1.2%) and 0 very major error. Although the EA and CA of *K. pneumoniae* with Ceftazidime-avibactam were acceptable at 98.6% and 98.6%, respectively, the very major error rate (5.0%) was not acceptable. Due to a lack of an intermediate breakpoint, further analysis of the errors was performed, and adjustments were made by considering the MIC values of the errors compared to the reference MIC value. The adjusted very major error rate for *K. pneumoniae* was 5.0% (1/20), which was still unacceptable.

Additionally, although the EA and CA of *P. aeruginosa* with Ceftazidime-avibactam were acceptable at 94.9% and 91.5%, respectively, and the very major errors (33.3%) was not acceptable. Due to a lack of an intermediate breakpoint, further analysis of the errors was performed, and adjustments were made by considering the MIC values of the errors compared to the reference MIC value. Two of the very major errors were in essential agreement with the reference MIC value. Therefore, the very major error rate was adjusted to 16.7% (2/12), which was still unacceptable. The [general limitation](#) listed above addresses the species that have shown good performance with LifeScale LSGN kit testing with Ceftazidime-avibactam and for which testing with LifeScale LSGN kit may be performed.

Gentamicin/Enterobacterales. A total of 480 Enterobacterales samples were evaluated with Gentamicin [*E. coli* (139), *K. aerogenes* (53), *K. pneumoniae* (142), *K. oxytoca* (111), *K. variicola* (35)]. The combined results from clinical and challenge sample testing demonstrated an EA of 98.8% and CA of 97.7%. There were 2 major errors (0.6%) and 1 very major error (0.8%).

Gentamicin/*P. aeruginosa*. A total of 59 *P. aeruginosa* samples were evaluated with Gentamicin. The combined results from clinical and challenge sample testing demonstrated an EA of 94.9% and CA of 93.2%. There were no major or very major errors.

Levofloxacin/Enterobacterales. A total of 437 Enterobacterales samples were evaluated with Levofloxacin [*E. coli* (137), *K. aerogenes* (53), *K. pneumoniae* (139), *K. oxytoca* (108)]. The combined results from clinical and challenge sample testing demonstrated an EA of 98.2% and CA of 96.6%. There were no major or very major errors.

Levofloxacin/*P. aeruginosa*. A total of 101 *P. aeruginosa* samples were evaluated with Levofloxacin. The combined results from clinical and challenge sample testing demonstrated an EA of 96.0% and CA of 88.1%, which was considered acceptable since most of the categorical errors were minor and the EA of evaluable results was 90.0%.

Meropenem/Enterobacterales. A total of 392 Enterobacterales samples were evaluated with Meropenem [*E. coli* (139), *K. pneumoniae* (142), *K. oxytoca* (111)]. The combined results from clinical and challenge sample testing demonstrated an EA of 91.6% and CA of 96.7%. There were 4 very major errors (3.3%) and no major errors, which was not acceptable. When evaluating results by individual species, *K. oxytoca* had 2 very major errors (2/12 = 16.7%), which is not acceptable. To address this unacceptable very major error rate, the following limitation is included in the device labeling:

Perform an alternative method of testing prior to reporting results for:

- *Meropenem: K. oxytoca when the LifeScale MIC is 0.5 µg/mL due to two very major discrepancies*

Meropenem/*Acinetobacter* spp. A total of 78 *Acinetobacter* spp. samples were evaluated with Meropenem [*A. baumannii* (22), *A. baumannii/nosocomialis* group (37) and other rare *Acinetobacter* species (19) including *A. lwoffii*, *A. pittii*, *A. calcoaceticus* species, *A. radioresistens* and *A. ursingii*]. The combined results from clinical and challenge sample

testing demonstrated an EA of 96.2% and CA of 97.4%. There were no major or very major errors.

Meropenem/*P. aeruginosa*. A total of 59 *P. aeruginosa* samples were evaluated with Meropenem. The combined results from clinical and challenge sample testing demonstrated an EA of 96.6% and CA of 89.8%, which was considered acceptable since most of the categorical errors were minor and the EA of evaluable results was good (>90%). There were no major or very major errors.

Meropenem-vaborbactam/Enterobacterales. A total of 442 Enterobacterales samples were evaluated with Meropenem/vaborbactam [*E. coli* (139), *K. aerogenes* (53), *K. pneumoniae* (140), *K. oxytoca* (110)]. The combined results from clinical and challenge sample testing demonstrated an EA of 95.0% and CA of 94.6%. There were 2 major errors (0.6%) and no very major error.

Piperacillin-tazobactam/Enterobacterales. A total of 391 Enterobacterales samples were evaluated with Piperacillin/tazobactam [*E. coli* (210), *K. pneumoniae* (181)]. The combined results from clinical and challenge sample testing demonstrated an EA of 90.8% and CA of 92.1%. There were 3 major errors (1.6%) and 3 very major errors (1.5%). When evaluating results by individual species, *K. pneumoniae* demonstrated an CA of 89.0%, with the EA of evaluable results was 52.9%, which is not acceptable. To address this unacceptable CA, the following limitation is included in the device labeling:

Perform an alternative method of testing prior to reporting results for:

- *Piperacillin/tazobactam: K. pneumoniae when the MIC is 16 µg/mL due to the occurrence of minor errors, that were in essential agreement, resulting in a category agreement below 90%*

Piperacillin-tazobactam/*Acinetobacter* spp. A total of 89 *Acinetobacter* spp. samples were evaluated with Piperacillin/tazobactam [*A. baumannii* (51), *A. baumannii/nosocomialis* group (38)]. The combined results from clinical and challenge sample testing demonstrated an EA of 92.1% and CA of 94.4%. There were no major or very major errors.

Piperacillin-tazobactam/*P. aeruginosa*. A total of 185 *P. aeruginosa* samples were evaluated with Piperacillin/tazobactam. The combined results from clinical and challenge sample testing demonstrated an EA of 94.1% and CA of 93.5%. There were 1 major error (0.9%) and 1 very major error (1.5%).

Table 7. Performance for All Antimicrobial Agents with All Organisms Groups

	Tot	No. EA	EA %	Eval EA Tot	No. Eval EA	Eval EA %	No. CA	CA %	No. R	No. S	min	maj	vmj
Amikacin– Enterobacterales: <i>E. coli</i>, <i>K. pneumoniae</i>, <i>K. aerogenes</i>, <i>K. oxytoca</i>, <i>K. variicola</i> [Breakpoints (µg/mL): 16 (S), 32 (I), 64 (R)]													
Seeded Challenge	118	113	95.8	18	13	72.2	111	94.1	56	58	6	1	0
Prospective	68	67	98.5	2	1	50	67	98.5	0	68	0	1	0
Seeded Clinical	294	287	97.6	34	27	79.4	282	95.9	6	274	11	1	0

	Tot	No. EA	EA %	Eval EA Tot	No. Eval EA	Eval EA %	No. CA	CA %	No. R	No. S	min	maj	vmj
Combined	480	467	97.3	54	41	75.9	460	95.8	62	400	17	3	0
Amikacin– <i>Acinetobacter</i> spp.: <i>A. baumannii</i> , <i>A. baumannii nosocomialis</i> group, <i>A. lwoffii</i> , <i>A. pittii</i> , <i>A. calcoaceticus</i> species, <i>A. radioresistens</i> and <i>A. ursingii</i> [Breakpoints (µg/mL): 16 (S), 32 (I), 64 (R)]													
Seeded Challenge	19	19	100	4	4	100	18	94.7	6	12	1	0	0
Seeded Clinical	58	58	100	7	7	100	57	98.3	11	46	1	0	0
Combined	77	77	100	11	11	100	75	97.4	17	58	2	0	0
Amikacin– <i>P. aeruginosa</i> [Breakpoints (µg/mL): 16 (S), 32 (I), 64 (R)]													
Seeded Challenge	15	15	100	6	6	100	14	93.3	6	8	1	0	0
Prospective	15	15	100	2	2	100	15	100	0	15	0	0	0
Seeded Clinical	29	28	96.6	7	6	85.7	27	93.1	1	27	2	0	0
Combined	59	58	98.3	15	14	93.3	56	94.9	7	50	3	0	0
Cefepime– Enterobacterales: <i>E. coli</i>, <i>K. pneumoniae</i>, <i>K. aerogenes</i>, <i>K. oxytoca</i> [Breakpoints (µg/mL): 2 (S), 4-8 (SDD*), 16 (R)]													
Seeded Challenge	112	100	89.3	19	7	36.8	106	94.6	90	19	5	1	0
Prospective	65	63	96.9	5	3	60	63	96.9	2	62	2	0	0
Seeded Clinical	268	249	92.9	40	21	52.5	257	95.9	62	200	9	2	0
Combined	445	412	92.6	64	31	48.4	426	95.7	154	281	16	3	0
Cefepime– <i>P. aeruginosa</i> [Breakpoints (µg/mL): 8 (S), 16 (R)]													
Seeded Challenge	46	44	95.7	26	24	92.3	45	97.8	16	30	0	1	0
Prospective	15	15	100	13	13	100	12	80	1	14	0	3	0
Seeded Clinical	40	35	87.5	39	34	87.2	28	70.0	12	28	0	9	3
Combined	101	94	93.1	78	71	91.0	85	84.2	29	72	0	13	3
Ceftazidime/avibactam– Enterobacterales: <i>E. coli</i>, <i>K. aerogenes</i>, <i>K. oxytoca</i> [Breakpoints (µg/mL): 8 (S), 16 (R)]													
Seeded Challenge	71	68	97.8	7	4	57.1	68	95.8	44	27	0	3	0
Prospective	30	30	100	0	0	0	30	100	0	30	0	0	0
Seeded Clinical	202	202	100	0	0	0	202	100	3	199	0	0	0
Combined	303	300	99.0	7	4	57.1	300	99.0	47	256	0	3	0
Gentamicin– Enterobacterales: <i>E. coli</i>, <i>K. pneumoniae</i>, <i>K. aerogenes</i>, <i>K. oxytoca</i>, <i>K. variicola</i> [Breakpoints (µg/mL): 4 (S), 8 (I), 16 (R)]													
Seeded Challenge	118	117	99.2	14	13	92.9	115	97.5	80	37	2	0	1
Prospective	68	66	97.1	5	3	60	68	100	2	66	0	0	0
Seeded Clinical	294	291	99.0	36	33	91.7	286	97.3	44	249	6	2	0

	Tot	No. EA	EA %	Eval EA Tot	No. Eval EA	Eval EA %	No. CA	CA %	No. R	No. S	min	maj	vmj
Combined	480	474	98.8	55	49	89.1	469	97.7	126	352	8	2	1
Gentamicin– <i>P. aeruginosa</i> [Breakpoints (µg/mL): 4 (S), 8 (I), 16 (R)]													
Seeded Challenge	15	15	100	1	1	100	15	100	8	7	0	0	0
Prospective	15	14	93.3	2	1	50	14	93.3	0	15	1	0	0
Seeded Clinical	29	27	93.1	15	13	86.7	26	89.7	2	25	3	0	0
Combined	59	56	94.9	18	15	83.3	55	93.2	10	47	4	0	0
Levofloxacin– Enterobacterales: <i>E. coli</i>, <i>K. pneumoniae</i>, <i>K. aerogenes</i>, <i>K. oxytoca</i> [Breakpoints (µg/mL): 0.5 (S), 1 (I), 2 (R)]													
Seeded Challenge	112	111	99.1	16	15	93.8	110	98.2	93	17	2	0	0
Prospective	62	60	96.8	8	6	75	57	91.9	3	55	5	0	0
Seeded Clinical	263	258	98.1	34	29	85.3	255	97.0	58	195	8	0	0
Combined	437	429	98.2	58	50	86.2	422	96.6	154	267	15	0	0
Levofloxacin– <i>P. aeruginosa</i> [Breakpoints (µg/mL): 1 (S), 2 (I), 4 (R)]													
Seeded Challenge	46	46	100	13	13	100	46	100	17	29	0	0	0
Prospective	15	14	93.3	4	3	75	12	80	1	13	3	0	0
Seeded Clinical	40	37	92.5	23	20	87.0	31	77.5	8	27	9	0	0
Combined	101	97	96.0	40	36	90.0	89	88.1	26	69	12	0	0
Meropenem – Enterobacterales: <i>E. coli</i>, <i>K. pneumoniae</i>, <i>K. oxytoca</i> [Breakpoints (µg/mL): 1 (S), 2 (I), 4 (R)]													
Seeded Challenge	103	87	84.5	26	10	38.5	102	99.0 3	83	19	1	0	0
Prospective	63	62	98.4	1	0	0	63	100	0	63	0	0	0
Seeded Clinical	226	210	92.9	35	19	54.3	214	94.7	40	182	7	1	4
Combined	392	359	91.6	62	29	46.8	379	96.7	123	264	8	1	4
Meropenem– <i>Acinetobacter</i> spp.: <i>A. baumannii</i>, <i>A. baumannii</i> nosocomialis group, <i>A. lwoffii</i>, <i>A. pittii</i>, <i>A. calcoaceticus</i> species, <i>A. radioresistens</i> and <i>A. ursingii</i> [Breakpoints (µg/mL): 2 (S), 4 (I), 8 (R)]													
Seeded Challenge	19	19	100	10	10	100	19	100	8	11	0	0	0
Seeded Clinical	59	56	94.9	32	29	90.6	57	96.6	29	30	2	0	0
Combined	78	75	96.2	42	39	92.9	76	97.4	37	41	2	0	0
Meropenem– <i>P. aeruginosa</i> [Breakpoints (µg/mL): 2 (S), 4 (I), 8 (R)]													
Seeded Challenge	15	15	100	7	7	100	14	93.3	8	7	1	0	0
Prospective	15	15	100	10	10	100	13	86.7	2	12	2	0	0
Seeded Clinical	29	27	93.1	22	20	90.9	26	89.7	15	13	3	0	0
Combined	59	57	96.6	39	37	94.9	53	89.8	25	32	6	0	0

	Tot	No. EA	EA %	Eval EA Tot	No. Eval EA	Eval EA %	No. CA	CA %	No. R	No. S	min	maj	vmj
Meropenem/vaborbactam– Enterobacterales: <i>E. coli</i>, <i>K. aerogenes</i>, <i>K. oxytoca</i>, <i>K. pneumoniae</i> [Breakpoints (µg/mL): 4/8 (S), 8/8 (I), 16/8 (R)]													
Seeded Challenge	111	94	84.7	31	14	45.2	91	82.0	72	37	19	1	0
Prospective	65	65	100	0	0	NA	65	100	0	65	0	0	0
Seeded Clinical	266	261	98.1	8	3	37.5	262	98.5	5	260	3	1	0
Combined	442	420	95.0	39	17	43.6	418	94.6	77	362	22	2	0
Piperacillin/tazobactam– Enterobacterales: <i>E. coli</i>, <i>K. pneumoniae</i> [Breakpoints (µg/mL): 8/4 (S), 16/4 (I), 32/4 (R)]													
Seeded Challenge	162	150	92.6	26	14	53.9	155	95.7	139	21	7	0	0
Prospective	58	52	89.7	8	2	25	52	89.7	4	54	4	1	1
Seeded Clinical	171	153	89.5	33	15	45.5	153	89.5	55	114	14	2	2
Combined	391	355	90.8	67	31	46.3	360	92.1	198	189	25	3	3
Piperacillin/tazobactam– <i>Acinetobacter</i> spp.: <i>A. baumannii</i>, <i>A. baumannii</i> nosocomialis group [Breakpoints (µg/mL): 16/4 (S), 32/4 – 64/4 (I), 128/4 (R)]													
Seeded Challenge	35	34	97.1	7	6	85.7	33	94.3	27	8	2	0	0
Seeded Clinical	54	48	88.9	16	10	62.5	51	94.4	39	14	3	0	0
Combined	89	82	92.1	23	16	69.6	84	94.4	66	22	5	0	0
Piperacillin/tazobactam– <i>P. aeruginosa</i> [Breakpoints (µg/mL): 8/4 (S), 16/4 (SDD*), 32/4 (R)]													
Seeded Challenge	114	110	96.5	28	24	85.7	112	98.3	45	69	2	0	0
Prospective	15	15	100	5	5	100	12	80	2	11	3	0	0
Seeded Clinical	56	49	87.5	26	19	73.1	49	87.5	20	32	5	1	1
Combined	185	174	94.1	59	48	81.4	173	93.5	67	112	10	1	1

*SDD- Susceptible-dose dependent.

EA - Essential Agreement

CA - Category Agreement

Eval - Evaluable isolates

R - Resistant isolates

S - Susceptible isolates

min - minor errors

maj - major errors

vmj - very major errors

Trending

A trending analysis using combined clinical and challenge sample results was conducted to evaluate antimicrobial-organism combinations for which LifeScale LSGN MIC results were determined to be one or more doubling dilutions lower or higher than the reference result (Table 8). MIC results that were off-scale for both the reference and LifeScale LSGN Kit were not considered in the trending analysis. Antimicrobial-organism combinations for which the difference between the percentage of isolates with higher or lower MIC values was $\geq 30\%$ with a statistically significant confidence interval were considered to have evidence of trending and is addressed in the labeling.

Table 8. LifeScale LSGN Kit – Analysis of Trending in the Clinical Study

Drug	Organism	Total Evaluable for Trending	≥ 1 Dilution lower No. (%)	Exact No. (%)	≥ 1 Dilution Higher No. (%)	Percent Difference (CI)	Trending Noted in the Clinical Study
Amikacin	<i>Escherichia coli</i> ^a	53	11, (20.75)	6	36, (67.92)	47%, (29%, 61%)	Yes
	<i>Klebsiella aerogenes</i>	38	0, (0)	1	37, (97.37)	97%, (83%, 100%)	Yes
	<i>Klebsiella oxytoca</i>	97	0, (0)	5	92, (94.85)	95%, (87%, 98%)	Yes
	<i>Klebsiella pneumoniae</i>	112	8, (7.14)	12	92, (82.14)	75%, (65%, 82%)	Yes
	<i>Klebsiella variicola</i>	28	0, (0)	0	28, (100)	100%, (83%, 100%)	Yes
	<i>Acinetobacter baumannii</i>	14	0, (0)	3	11, (78.57)	79%, (45%, 92%)	Yes
	<i>Acinetobacter baumannii/nosocomialis group</i>	27	4, (14.81)	4	19, (70.37)	56%, (30%, 72%)	Yes
	<i>Acinetobacter calcoaceticus species</i>	1	0, (0)	0	1, (100)	100%, (- 12%, 100%)	No
	<i>Acinetobacter lwoffii</i>	1	0, (0)	0	1, (100)	100%, (- 12%, 100%)	No
	<i>Acinetobacter pittii</i>	5	0, (0)	0	5, (100)	100%, (39%, 100%)	Yes
	<i>Acinetobacter radioresistens</i>	2	0, (0)	0	2, (100)	100%, (7%, 100%)	Yes
	<i>Acinetobacter ursingii</i>	1	0, (0)	0	1, (100)	100%, (- 12%, 100%)	No
	<i>Pseudomonas aeruginosa</i>	41	7, (17.07)	6	28, (68.29)	51%, (30%, 66%)	Yes
Cefepime	<i>Escherichia coli</i> ^b	65	16, (24.62)	2	47, (72.31)	48%, (31%, 61%)	Yes
	<i>Klebsiella aerogenes</i>	26	4, (15.38)	1	21, (80.77)	65%, (39%, 80%)	Yes
	<i>Klebsiella oxytoca</i>	74	10, (13.51)	1	63, (85.14)	72%, (58%, 80%)	Yes
	<i>Klebsiella pneumoniae</i>	65	17, (26.15)	7	41, (63.08)	37%, (20%, 51%)	Yes
	<i>Pseudomonas aeruginosa</i>	90	27, (30)	36	27, (30)	0%, (- 13%, 13%)	No
Ceftazidime/ avibactam	<i>Escherichia coli</i> ^c	5	5, (100)	0	0, (0)	-100%, (- 100%, - 39%)	Yes
	<i>Klebsiella aerogenes</i>	7	1, (14.29)	0	6, (85.71)	71%,	Yes

Drug	Organism	Total Evaluable for Trending	≥ 1 Dilution lower No. (%)	Exact No. (%)	≥ 1 Dilution Higher No. (%)	Percent Difference (CI)	Trending Noted in the Clinical Study
						(19%, 88%)	
	<i>Klebsiella oxytoca</i>	10	1, (10)	0	9, (90)	80%, (37%, 92%)	Yes
Gentamicin	<i>Escherichia coli</i> ^a	109	20, (18.35)	22	67, (61.47)	43%, (31%, 54%)	Yes
	<i>Klebsiella aerogenes</i>	20	1, (5)	1	18, (90)	85%, (58%, 93%)	Yes
	<i>Klebsiella oxytoca</i>	40	6, (15)	3	31, (77.5)	62%, (42%, 75%)	Yes
	<i>Klebsiella pneumoniae</i>	61	14, (22.95)	10	37, (60.66)	38%, (20%, 52%)	Yes
	<i>Klebsiella variicola</i>	5	1, (20)	1	3, (60)	40%, (- 16%, 73%)	No
	<i>Pseudomonas aeruginosa</i> ^a	35	11, (31.43)	8	16, (45.71)	14%, (- 8%, 35%)	No
Levofloxacin	<i>Escherichia coli</i>	33	7, (21.21)	15	11, (33.33)	12%, (- 9%, 32%)	No
	<i>Klebsiella aerogenes</i>	19	1, (5.26)	3	15, (78.95)	74%, (44%, 87%)	Yes
	<i>Klebsiella oxytoca</i>	47	4, (8.51)	7	36, (76.6)	68%, (50%, 79%)	Yes
	<i>Klebsiella pneumoniae</i>	53	5, (9.43)	9	39, (73.58)	64%, (47%, 75%)	Yes
	<i>Pseudomonas aeruginosa</i>	64	15, (23.44)	24	25, (39.06)	16%, (0%, 31%)	No
Meropenem	<i>Escherichia coli</i>	72	34, (47.22)	6	32, (44.44)	-3%, (- 19%, 13%)	No
	<i>Klebsiella oxytoca</i>	42	12, (28.57)	1	29, (69.05)	40%, (19%, 57%)	Yes
	<i>Klebsiella pneumoniae</i>	60	24, (40)	10	26, (43.33)	3%, (- 14%, 20%)	No
	<i>Acinetobacter baumannii</i>	14	6, (42.86)	4	4, (28.57)	-14%, (- 44%, 19%)	No
	<i>Acinetobacter baumannii/nosocomialis group</i>	16	4, (25)	7	5, (31.25)	6%, (- 24%, 35%)	No
	<i>Acinetobacter calcoaceticus species</i>	1	1, (100)	0	0, (0)	-100%, (- 100%, 12%)	No
	<i>Acinetobacter lwoffii</i>	1	0, (0)	0	1, (100)	100%, (- 12%, 100%)	No
	<i>Acinetobacter pittii</i>	6	3, (50)	1	2, (33.33)	-17%, (- 56%, 32%)	No

Drug	Organism	Total Evaluable for Trending	≥ 1 Dilution lower No. (%)	Exact No. (%)	≥ 1 Dilution Higher No. (%)	Percent Difference (CI)	Trending Noted in the Clinical Study
	<i>Acinetobacter radioresistens</i>	3	1, (33.33)	1	1, (33.33)	0%, (-53%, 53%)	No
	<i>Acinetobacter ursingii</i>	1	0, (0)	1	0, (0)	0%, (-79%, 79%)	No
	<i>Pseudomonas aeruginosa</i>	48	8, (16.67)	16	24, (50)	33%, (15%, 49%)	Yes
Meropenem/ vaborbactam	<i>Escherichia coli</i>	44	33, (75)	3	8, (18.18)	-57%, (-70%, -37%)	Yes
	<i>Klebsiella aerogenes</i>	25	0, (0)	2	23, (92)	92%, (70%, 98%)	Yes
	<i>Klebsiella oxytoca</i>	110	8, (7.27)	0	102, (92.73)	85%, (76%, 90%)	Yes
	<i>Klebsiella pneumoniae</i>	54	6, (11.11)	5	43, (79.63)	69%, (52%, 79%)	Yes
Piperacillin/ tazobactam	<i>Escherichia coli</i> ^a	70	28, (40)	5	37, (52.86)	13%, (-4%, 28%)	No
	<i>Klebsiella pneumoniae</i>	111	21, (18.92)	9	81, (72.97)	54%, (42%, 64%)	Yes
	<i>Acinetobacter baumannii</i> ^c	32	19, (59.38)	6	7, (21.88)	-38%, (-56%, -13%)	Yes
	<i>Acinetobacter baumannii/nosocomialis group</i>	22	9, (40.91)	6	7, (31.82)	-9%, (-35%, 18%)	No
	<i>Pseudomonas aeruginosa</i>	110	50, (45.45)	30	30, (27.27)	-18%, (-30%, -5%)	No

^a High trending noted in the Inoculum Density study for positive blood culture samples with an organism concentration of 10⁹ CFU/mL.

^b High trending noted in the Inoculum Density study for positive blood culture samples with an organism concentration of 10⁶ and 10⁹ CFU/mL.

^c Low trending noted in the Inoculum Density study for positive blood culture samples with an organism concentration of 10⁶ CFU/mL.

Analysis of trending in the clinical study indicated that LifeScale LSGN MIC values for the following antimicrobial/organism combinations tended to be at least one doubling dilution higher than the reference MIC value:

- Amikacin: *E. coli*, *K. aerogenes*, *K. oxytoca*, *K. pneumoniae*, *K. variicola*, *Acinetobacter spp.*, *P. aeruginosa*
- Cefepime: *E. coli*, *K. aerogenes*, *K. oxytoca*, *K. pneumoniae*
- Ceftazidime-avibactam: *K. aerogenes*, *K. oxytoca*
- Gentamicin: *E. coli*, *K. aerogenes*, *K. oxytoca*, *K. pneumoniae*
- Levofloxacin: *K. pneumoniae*, *K. oxytoca*, *K. aerogenes*
- Meropenem: *K. oxytoca*, *P. aeruginosa*

- Meropenem-vaborbactam: *K. pneumoniae*, *K. oxytoca*, *K. aerogenes*
- Piperacillin-tazobactam: *K. pneumoniae*

LifeScale LSGN MIC values for the following antimicrobial/organism combinations tended to be at least one doubling dilution lower than the reference MIC value:

- Ceftazidime-avibactam: *E. coli*
- Meropenem-vaborbactam: *E. coli*
- Piperacillin-tazobactam: *A. baumannii*

To address the overall observed trending with the LifeScale LSGN Kit in both the clinical study and inoculum density study, the following statement was added to the device labeling:

In the clinical study or in the Inoculum Density analytical study, the majority of drug/organism combinations tested with the LifeScale LSGN Kit showed MIC values equal to or at least one doubling dilution higher than the reference method. Use caution when reporting drug resistance for any antimicrobial. The following drug/organism combinations showed high trending:

The following drug/organism combinations showed high trending:

- Amikacin: *E. coli*, *K. aerogenes*, *K. oxytoca*, *K. pneumoniae*, *K. variicola*, *Acinetobacter spp.*, *P. aeruginosa*
- Cefepime: *E. coli*, *K. aerogenes*, *K. oxytoca*, *K. pneumoniae*
- Ceftazidime-avibactam: *K. aerogenes*, *K. oxytoca*
- Gentamicin: *E. coli*, *K. aerogenes*, *K. oxytoca*, *K. pneumoniae*, *P. aeruginosa*
- Levofloxacin: *K. pneumoniae*, *K. oxytoca*, *K. aerogenes*
- Meropenem: *K. oxytoca*, *P. aeruginosa*
- Meropenem-vaborbactam: *K. pneumoniae*, *K. oxytoca*, *K. aerogenes*
- Piperacillin-tazobactam: *E. coli*, *K. pneumoniae*

The following drug/organism combinations showed low trending:

- Ceftazidime-avibactam: *E. coli*
- Meropenem-vaborbactam: *E. coli*
- Piperacillin-tazobactam: *A. baumannii*

Removal of limitations included in K211815.

In the current 510(k) pre-market submission, additional clinical evaluation testing was performed to support the removal of limitations included in the cleared LifeScale LSGN Kit with the LifeScale AST system (K211815) for Aztreonam, Ceftazidime, Ertapenem, and Cefazolin.

Ertapenem/*E. coli* – Removal of Off-line Limitation

In K211815, evaluation of the performance of on-line and off-line incubation of panels testing Ertapenem/*E. coli* showed unacceptable performance for this drug/organism

combination when incubated offline. The following limitation was included in the device labeling:

Due to unacceptable performance of Ertapenem/E. coli with incubation in an off-line incubator, perform an alternative method of testing prior to reporting results for Ertapenem/E. coli when panels are incubated in an off-line incubator.

The current submission provided additional data for both incubation options, with the following performance:

Table 9. Ertapenem/E.coli On-line/Off-line Performance

	Tot	No. EA	EA %	Eval EA Tot	No. Eval EA	Eval EA %	No. CA	CA %	No. R	No. S	min	maj	vmj
Ertapenem E. coli													
Off-line Combined	94	85	90.4	12	3	25	85	90.4	20	74	8	1	0
On-line Combined	108	105	97.2	5	2	40	104	96.3	60	48	3	1	0

The performance for both on-line and off-line incubation is acceptable, and the above limitation was removed from the device labeling.

Aztreonam/K. pneumoniae

In K211815, the sponsor provided data for testing Aztreonam/*K. pneumoniae* which showed unacceptable performance (low EA and increased major errors). The following limitation was included in the device labeling:

Perform an alternative method of testing prior to reporting results for the following antimicrobial/organism combination(s): Aztreonam/K. pneumoniae

The current submission included data from testing 54 additional *K. pneumoniae* isolates (seeded clinical and seeded challenge), with the following performance for the initial (K211815) and additional data combined:

Table 10. Aztreonam Overall Performance

	Tot	No. EA	EA %	Eval EA Tot	No. Eval EA	Eval EA %	No. CA	CA %	No. R	No. S	min	maj	vmj
Aztreonam K. pneumoniae [Breakpoints (µg/mL): ≤4 (S), 8 (I), ≥16 (R)]													
Seeded Challenge	50	45	90	6	1	16.7	47	94	34	15	1	2	0
Prospective	34	29	85.3	5	0	0	31	91.2	3	31	2	0	1
Seeded Clinical	76	70	92.1	10	4	40	73	96.1	39	37	3	0	0
Combined	160	144	90	21	5	23.8	151	94.4	76	83	6	2	1

The performance for Aztreonam/*K. pneumoniae* is acceptable, and the above limitation was removed from the device labeling.

Ceftazidime/ *K. pneumoniae*

In K211815, the sponsor provided data for testing Ceftazidime/*K. pneumoniae* which showed unacceptable performance (increased major errors). The following limitation was included in the device labeling:

*Perform an alternative method of testing prior to reporting results for the following antimicrobial/organism combination(s): Ceftazidime/*K. pneumoniae**

The current submission included data from testing 54 additional *K. pneumoniae* isolates (seeded clinical and seeded challenge), with the following performance for the initial (K211815) and additional data combined:

Table 11. Ceftazidime/*K. pneumoniae* Overall Performance

	Tot	No. EA	EA %	Eval EA Tot	No. Eval EA	Eval EA %	No. CA	CA %	No. R	No. S	min	maj	vmj
Ceftazidime <i>K. pneumoniae</i> [Breakpoints (µg/mL): ≤4 (S), 8 (I), ≥ 16 (R)]													
Seeded Challenge	49	48	98.0	4	3	75	48	98.0	36	13	0	1	0
Prospective	35	32	91.4	6	3	50	32	91.4	3	30	2	0	1
Seeded Clinical	76	74	97.4	12	10	83.3	75	98.7	40	36	0	1	0
Combined	160	154	96.3	22	16	72.7	155	96.9	79	79	2	2	1

The performance for Ceftazidime/*K. pneumoniae* is acceptable, and the above limitation was removed from the device labeling.

Ceftazidime/*Acinetobacter* spp.

In K211815, the sponsor provided data for testing Ceftazidime with a variety of *Acinetobacter* spp. Performance for *A. baumannii* and *A. baumannii/A. nosocomialis* group was acceptable but combined testing of other *Acinetobacter* spp. showed unacceptable performance (increased very major errors). The following limitation was included in the device labeling:

*Perform an alternative method of testing prior to reporting results for the following antimicrobial/organism combination(s): Ceftazidime/*Acinetobacter* spp. (other than *A. baumannii* and *A. baumannii/nosocomialis* group).*

The current submission indicates that the unacceptable percentage of very major errors was due to very major errors observed only with *Acinetobacter ursignii*. The limitation was modified to limit reporting to *Acinetobacter* spp. other than *A. ursignii*.

Table 12. Performance Ceftazidime/*Acinetobacter* spp.

	Tot	No. EA	EA %	Eval EA Tot	No. Eval EA	Eval EA %	No. CA	CA %	No. R	No. S	min	maj	vmj
Ceftazidime <i>A. baumannii</i> , <i>A. baumannii/A. nosocomialis</i> group, <i>A. calcoaceticus</i> , <i>A. lwoffii</i> , <i>A. pittii</i> , <i>A. radioresistens</i> , and <i>Acinetobacter</i> spp. [Breakpoints (µg/mL): 8 (S), 16 (I), 32 (R)]													
Seeded Challenge	46	45	97.8	23	22	95.7	45	97.8	28	18	1	0	0
Seeded Clinical	72	67	93.1	35	30	85.7	71	98.6	35	34	1	0	0
Combined	118	112	94.9	58	52	89.7	116	98.3	63	52	2	0	0

The performance data for all *Acinetobacter* spp. including *A. baumannii*, *A. baumannii/A. nosocomialis* group, *A. calcoaceticus*, *A. lwoffii*, *A. pittii*, *A. radioresistens*, and *Acinetobacter* spp. showed acceptable performance (Table 12). The combining and inclusion of all above species is considered acceptable as the drug label for ceftazidime lists “*Acinetobacter* species” as Indicated species List 2. Thus, the revised limitation as proposed by the sponsor is acceptable:

Perform an alternative method of testing prior to reporting results for the following antimicrobial/organism combination(s): Ceftazidime/Acinetobacter ursingii

Cefazolin/*E. coli*

In K211815, the sponsor provided data for testing Cefazolin/*E. coli* which showed unacceptable performance. The following limitation was included in the device labeling:

Perform an alternative method of testing prior to reporting results for the following antimicrobial/organism combination(s): Cefazolin/E. coli

The current submission included data from testing 66 additional *E. coli* isolates (seeded clinical and seeded challenge), with the following performance for the initial (K211815) and additional data combined:

Table 13. Overall Performance Cefazolin/*E. coli*

	Tot	No. EA	EA %	Eval EA Tot	No. Eval EA	Eval EA %	No. CA	CA %	No. R	No. S	min	maj	vmj
Cefazolin <i>E. coli</i> [Breakpoints (µg/mL): ≤2 (S), 4 (I), ≥8 (R)]													
Seeded Challenge	86	85	98.8	13	12	92.3	80	93.0	78	8	6	0	0
Prospective	23	23	100	21	21	100	20	87.0	4	15	3	0	0
Seeded Clinical	94	89	94.7	68	63	92.7	78	83.0	35	50	13	1	2
Combined	203	197	97.0	102	96	94.1	178	87.7	117	73	22	1	2

The performance for Cefazolin/*E. coli* is acceptable, and the above limitation was removed from the device labeling.

Ertapenem/*K. pneumoniae*

In K211815, the sponsor provided data for testing Ertapenem/*K. pneumoniae* which showed unacceptable performance (due to increased very major errors). The following limitation was included in the device labeling:

*Perform an alternative method of testing prior to reporting results for the following antimicrobial/organism combination(s): Ertapenem/*K. pneumoniae**

The current submission included data from testing 54 additional *K. pneumoniae* isolates (seeded clinical and seeded challenge), with the following performance for the initial (K211815) and additional data combined:

Table 14. Overall Performance Ertapenem/*K. pneumoniae*

	Tot	No. EA	EA %	Eval EA Tot	No. Eval EA	Eval EA %	No. CA	CA %	No. R	No. S	min	maj	vmj
Ertapenem <i>K. pneumoniae</i> [Breakpoints (µg/mL): 0.5 (S), 1 (I), 2 (R)]													
Seeded Challenge	48	48	100	2	2	100	47	97.9	39	7	1	0	0
Prospective	35	34	97.1	1	0	0	35	100	0	35	0	0	0
Seeded Clinical	75	72	96	10	7	70	72	96	32	43	2	0	1
Combined	158	154	97.5	13	9	69.2	154	97.5	71	85	3	0	1

The performance for ertapenem/*K. pneumoniae* is acceptable, and the above limitation was removed from the device labeling.

Analysis of Trending

Drug	Organism	Total Evaluable for Trending	≥ 1 Dilution lower No. (%)	Exact No. (%)	≥ 1 Dilution Higher No. (%)	Percent Difference (CI)	Trending Noted
Aztreonam	<i>K. pneumoniae</i>	31	12 (38.7)	2	17 (54.84)	16% (-8% to 38%)	No
Ceftazidime	<i>K. pneumoniae</i>	59	16 (27.1)	10	33 (55.9)	29% (11%, 44%)	No
Ceftazidime	<i>Acinetobacter</i> spp.	69	15 (21.7)	26	28 (40.6)	19% (3% to 33%)	No
Cefazolin	<i>E. coli</i>	105	22 (21.2)	44	39 (37.1)	16% (4% to 28%)	No
Ertapenem	<i>K. pneumoniae</i>	39	9 (23.1)	5	25 (64.1)	41% (19%, to 58%)	Yes High

The following footnote should be added to the performance table in the device labeling:

K. pneumoniae tested with Ertapenem showed MIC values equal to or at least one doubling dilution higher than the reference method.

On-line vs. Off-line Incubation

Table 15. Summary of Off-line vs. On-line incubation

Drug	Organism	Acceptable Performance		Trending	
		Off-line	On-line	Off-line	On-line
Aztreonam	<i>K. pneumoniae</i>	yes	yes	no	Yes High ^a
Ceftazidime	<i>K. pneumoniae</i>	yes	Increased vmj errors ^b	no	Yes High ^a
Ceftazidime	<i>Acinetobacter</i> spp.	yes	yes	no	no
Cefazolin	<i>E. coli</i>	Increased vmj errors ^b	yes	no	no
Ertapenem	<i>K. pneumoniae</i>	Increased vmj errors ^b	yes	Yes High ^a	Yes High ^a

^a **Trending.** The following trending footnote should be added to the performance table in the device labeling:

K. pneumoniae tested with Aztreonam, Ceftazidime and Ertapenem and incubated on-line and *K. pneumoniae* tested with Ertapenem and incubated off-line showed MIC values equal to or at least one doubling dilution higher than the reference method.

^b **Performance.** The following footnote should be added to the performance table in the device labeling:

The very major error rate was increased for the following:

- *Ceftazidime/K. pneumoniae* incubated on-line (2.6%)
- *Cefazolin/E. coli* incubated off-line (2.3%)
- *Ertapenem/K. pneumoniae* incubated off-line (2.9%)

2. Matrix Comparison:

Not Applicable

C Clinical Studies:

1. Clinical Sensitivity:

Not Applicable

2. Clinical Specificity:

Not Applicable

3. Other Clinical Supportive Data (When 1. and 2. Are Not Applicable):

Not Applicable

D Clinical Cut-Off:

Not Applicable

E Expected Values/Reference Range:

Antimicrobial	Organism	Interpretive Criteria ^a			
		S	SDD	I	R
Amikacin	Enterobacterales	≤16	-	32	≥64
	<i>P. aeruginosa</i>	≤16	-	32	≥64
	<i>Acinetobacter</i> spp.	≤16	-	32	≥64
Cefepime	Enterobacterales	≤2	4-8	-	≥16
	<i>P. aeruginosa</i>	≤8	-	-	≥16
Ceftazidime/avibactam	Enterobacterales	≤8/4	-	-	≥16/4
Gentamicin	Enterobacterales	≤4	-	8	≥16
	<i>P. aeruginosa</i>	≤4	-	8	≥16
Levofloxacin	Enterobacterales	≤0.5	-	1	≥2
	<i>P. aeruginosa</i>	≤1	-	2	≥4
Meropenem	Enterobacterales	≤1	-	2	≥4
	<i>P. aeruginosa</i>	≤2	-	4	≥8
	<i>Acinetobacter</i> spp.	≤2	-	4	≥8
Meropenem/vaborbactam	Enterobacterales	≤4/8	-	8/8	≥16/8
Piperacillin/tazobactam	Enterobacterales	≤8/4	-	16/4	≥32/4
	<i>P. aeruginosa</i>	≤8/4	16/4	-	≥32/4
	<i>Acinetobacter</i> spp.	≤16/4	-	32/4-64/4	≥128/4

S = Susceptible; SDD = Susceptible-dose dependent; I = Intermediate; R = Resistant

^a FDA-Recognized Antimicrobial Susceptibility Test Interpretive Criteria Website

<https://www.fda.gov/Drugs/DevelopmentApprovalProcess/DevelopmentResources/ucm410971.htm>

F Other Supportive Instrument Performance Characteristics Data:

Not Applicable

VIII Proposed Labeling:

The labeling supports the finding of substantial equivalence for this device.

IX Conclusion:

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

To support the implementation of changes to FDA-recognized susceptibility test interpretive criteria (i.e., breakpoints), this submission included a predetermined change control plan (PCCP) that was reviewed and accepted by FDA as described in the [Antimicrobial Susceptibility Test](#)

[\(AST\) System Devices – Updating Breakpoints in Device Labeling guidance](https://www.fda.gov/Drugs/DevelopmentApprovalProcess/DevelopmentResources/ucm410971.htm). This PCCP addresses future revisions to device labeling in response to breakpoint changes that are recognized on the FDA STIC webpage (<https://www.fda.gov/Drugs/DevelopmentApprovalProcess/DevelopmentResources/ucm410971.htm>). The protocol outlined the specific procedures and acceptance criteria that Affinity Biosensors intends to use to evaluate the LifeScale AST system when revised breakpoints for indicated drugs are published on the FDA STIC webpage. The PCCP included with the submission indicated that if specific criteria are met, Affinity Biosensors will update the LifeScale AST system label to include (1) the new breakpoints, (2) an updated performance section after re-evaluation of data in this premarket notification with the new breakpoints, and (3) any new limitations as determined by their evaluation.