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

I Background Information:

A 510(k) Number

K231536

B Applicant

Avails Medical, Inc.

C Proprietary and Established Names

eQUANT System

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
QZX	Class II	21 CFR 866.1650 - A Cellular Analysis System For Multiplexed Antimicrobial Susceptibility Testing	MI - Microbiology
JTN	Class II	21 CFR 866.1620 - Antimicrobial susceptibility test disc	MI - Microbiology

II Submission/Device Overview:

A Purpose for Submission:

To obtain a substantial equivalence determination for the preparation of a 0.5 McFarland Standard equivalent from positive blood culture samples using the eQUANT System for downstream susceptibility testing with the agar disk diffusion method (Kirby-Bauer).

B Measurand:

Standardized suspension of Gram negative bacteria equivalent to a 0.5 McFarland standard prepared from positive blood culture samples.

C Type of Test:

Positive blood culture processor that prepares an inoculum for antimicrobial susceptibility testing.

III Intended Use/Indications for Use:

A Intended Use(s):

The eQUANT System is an automated inoculum preparation system that uses potentiometric sensing of oxidation-reduction potential changes due to pathogen metabolism to generate a 0.5 McFarland-equivalent suspension (the eMcFarland or eMcF) from positive blood culture samples. Samples are processed directly from blood culture samples identified as positive by a continuous monitoring blood culture system and confirmed as Gram-negative rods by Gram stain. Organism identification, as determined by an FDA cleared device for direct testing from positive blood culture, must be available before processing samples on the eQUANT System.

B Indication(s) for Use:

The eQUANT System is an automated inoculum preparation system that uses potentiometric sensing of oxidation-reduction potential changes due to pathogen metabolism to generate a 0.5 McFarland-equivalent suspension (the eMcFarland or eMcF) from positive blood culture samples that can be used for direct, qualitative in vitro susceptibility testing by the agar disk diffusion test method (Kirby-Bauer). Samples are processed directly from blood culture samples identified as positive by a continuous monitoring blood culture system and confirmed as Gram-negative rods by Gram stain. Organism identification must be confirmed by an FDA cleared device for testing from positive blood culture before processing samples on the eQUANT System.

Evaluation of the eQUANT System's inoculum preparation was conducted for use with agar disk diffusion susceptibility testing and performance was demonstrated for the following antimicrobial agents with Enterobacterales species, *Acinetobacter* species and *Pseudomonas aeruginosa* as identified below:

Amoxicillin/clavulanate- *Escherichia coli, Klebsiella oxytoca, Klebsiella pneumoniae, Proteus mirabilis*

Ampicillin- *Escherichia coli*

Aztreonam- *Citrobacter freundii, Enterobacter cloacae, Escherichia coli, Klebsiella aerogenes, Klebsiella oxytoca, Klebsiella pneumoniae, Proteus mirabilis, Proteus vulgaris, Serratia marcescens* and *Pseudomonas aeruginosa*

Cefazolin- *Klebsiella pneumoniae*

Cefepime- *Enterobacter cloacae, Escherichia coli, Klebsiella oxytoca, Klebsiella pneumoniae, Proteus mirabilis, Proteus vulgaris, Serratia marcescens,* and *Pseudomonas aeruginosa*

Ceftriaxone- *Citrobacter freundii, Enterobacter cloacae, Escherichia coli, Klebsiella aerogenes, Klebsiella oxytoca, Klebsiella pneumoniae, Proteus mirabilis, Proteus vulgaris, Serratia marcescens*

Ertapenem- *Citrobacter freundii, Enterobacter cloacae, Escherichia coli, Klebsiella aerogenes, Klebsiella oxytoca, Klebsiella pneumoniae, Proteus mirabilis, Proteus vulgaris, Serratia marcescens*

Gentamicin- *Citrobacter freundii, Enterobacter cloacae, Escherichia coli, Klebsiella aerogenes, Klebsiella oxytoca, Klebsiella pneumoniae, Proteus mirabilis, Proteus vulgaris, Serratia marcescens,* and *Pseudomonas aeruginosa*

Levofloxacin- *Citrobacter freundii, Enterobacter cloacae, Escherichia coli, Klebsiella aerogenes, Klebsiella oxytoca, Klebsiella pneumoniae, Proteus mirabilis, Proteus vulgaris, Serratia marcescens*, and *Pseudomonas aeruginosa*

Meropenem- *Acinetobacter* spp., *Citrobacter freundii, Enterobacter cloacae, Escherichia coli, Klebsiella oxytoca, Klebsiella pneumoniae, Proteus mirabilis, Proteus vulgaris, Serratia marcescens*, and *Pseudomonas aeruginosa*

Piperacillin/tazobactam- *Acinetobacter* spp., *Escherichia coli, Klebsiella pneumoniae, Proteus mirabilis, Proteus vulgaris, Serratia marcescens*, and *Pseudomonas aeruginosa*

Tobramycin- *Citrobacter freundii, Enterobacter cloacae, Escherichia coli, Klebsiella aerogenes, Klebsiella oxytoca, Klebsiella pneumoniae, Proteus mirabilis, Proteus vulgaris, Serratia marcescens*, and *Pseudomonas aeruginosa*

Susceptibility test 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.

C Special Conditions for Use Statement(s):

- Rx - For Prescription Use Only
- The eQUANT system should not be used for any clinical specimens other than monomicrobial positive blood cultures containing indicated Gram-negative bacteria.
- Polymicrobial samples should not be processed on the eQUANT system.
- The use of the eQUANT system does not eliminate the need for subculture of the positive blood culture.
- If the subculture (purity) plate indicates the sample is polymicrobial, the AST results should be voided, and susceptibility testing on each isolate using a standard inoculum preparation should be performed.

- The performance of the eQUANT system has only been evaluated with the following blood culture bottles:
    - bioMérieux BacT/ALERT FA Plus Aerobic
    - bioMérieux BacT/ALERT FN Plus Anaerobic
    - bioMérieux BacT/ALERT SA Standard Aerobic
    - bioMérieux BacT/ALERT SN Standard Anaerobic
    - BD BACTEC Plus Aerobic
    - BD BACTEC Plus Anaerobic
    - BD BACTEC Standard Aerobic
    - BD BACTEC Standard Anaerobic
    - BD BACTEC Lytic Anaerobic

- Positive blood cultures must be processed immediately on the eQUANT instrument or within 12 hours of blood culture bottle positivity should delays be unavoidable.

- The eMcFarland must be removed from with eQUANT Instrument within one (1) hour of run completion. Do not use an eMcFarland that has been incubating in the eQUANT instrument for longer than one (1) hour.

- The eMcFarland must be used within ten (10) minutes after it is removed from the eQUANT Instrument to maintain the appropriate organism concentration.

- *Pseudomonas aeruginosa* and *Acinetobacter* spp. isolated from anaerobic blood culture bottles should not be run on the eQUANT Instrument.

- High concentrations of hemoglobin (>2 g/dL) and platelets (>900,000 µL) may increase the number of Aeration Blockage Errors observed when testing *Acinetobacter* spp. or *P. aeruginosa*, resulting in aborted eQUANT runs.

- Performance of the eMcFarland for use in downstream Disk Diffusion AST has only been established using the antibiotics listed in the Indications for Use.

- Qualitative interpretation of susceptibility results should be performed in accordance with the disk manufacturer's instructions.

- Disk Diffusion AST zone diameter results generated from an eMcFarland inoculum tend to be smaller when compared to disk diffusion zone diameters results generated from a colony inoculum. If zone diameters fall near the low end of the intermediate breakpoint, consider alternative testing for the following drug/organism combinations:
    - Amoxacillin/Clavulanate: *Proteus vulgaris*
    - Aztreonam: *Citrobacter freundii, Escherichia coli, Proteus mirabilis, Proteus vulgaris*
    - Cefepime: *Escherichia coli, Klebsiella aerogenes, Proteus mirabilis, Proteus vulgaris*
    - Ceftriaxone: *Escherichia coli, Klebsiella aerogenes, Proteus mirabilis, Proteus vulgaris*
    - Ertapenem: *Escherichia coli, Klebsiella oxytoca, Proteus mirabilis, Proteus vulgaris, Serratia marcescens*
    - Gentamicin: *Klebsiella aerogenes, Proteus vulgaris*

- Levofloxacin: *Klebsiella aerogenes, Klebsiella oxytoca*
- Meropenem: *Proteus mirabilis, Proteus vulgaris, Serratia marcescens*
- Piperacillin/Tazobactam: *Proteus mirabilis, Proteus vulgaris*
- Tobramycin: *Proteus vulgaris*

- Perform an alternative method of testing prior to reporting results for the following antibiotic/organism combination(s):
  - Aztreonam: *Proteus mirabilis* when the disk zone diameter from an eMcFarland inoculum produces a resistant result due to one major error.
  - Cefazolin: *Escherichia coli, Proteus mirabilis*
  - Cefepime: *Citrobacter freundii, Klebsiella aerogenes*
  - Cefepime: *Proteus mirabilis* when the disk zone diameter from an eMcFarland inoculum produces a resistant result due to one major error.

- The ability of the eQUANT system to generate an inoculum to detect resistance with the following combinations is unknown because an insufficient number of resistant isolates were encountered at the time of comparative testing:
  - Aztreonam: *Proteus mirabilis, Proteus vulgaris*
  - Cefepime: *Citrobacter freundii, Klebsiella aerogenes, Proteus mirabilis*
  - Ceftriaxone: *Proteus vulgaris*
  - Ertapenem: *Klebsiella oxytoca, Proteus vulgaris*
  - Gentamicin: *Klebsiella oxytoca, Citrobacter freundii, Proteus vulgaris, Serratia marcescens*
  - Levofloxacin: *Klebsiella oxytoca, Klebsiella aerogenes, Proteus vulgaris, Serratia marcescens*
  - Piperacillin/Tazobactam: *Proteus vulgaris*
  - Tobramycin: *Klebsiella aerogenes, Proteus vulgaris*

D Special Instrument Requirements:

eQUANT Instrument, Software Version 1.21.0

IV Device/System Characteristics:

A Device Description:

The eQUANT system is an automated instrument that uses potentiometric sensing of changes in oxidation-reduction potential (ORP) during pathogen metabolism to prepare an organism suspension equivalent to a 0.5 McFarland standard (1-2e8 CFU/ml ± 0.6 log) directly from a positive blood culture. The eQUANT system consists of four components: the eQUANT instrument, a single use eTube disposable, a single use eQUANT Reagent tube (CAMHB with antifoam), and a workflow tray.

The eQUANT system processes a single positive blood culture sample at a time. Before processing on the eQUANT instrument, the positive blood culture is confirmed as Gram-negative rods followed by organism identification (ID) using an ID method FDA-cleared for use with positive blood culture. Mixed cultures or organisms identified that are not included in the eQUANT intended use must not be processed on the eQUANT system. Positive blood cultures

must be processed immediately on the eQUANT instrument or within 12 hours of blood culture bottle positivity should delays be unavoidable. Once the organism ID is determined, 1mL of eQUANT Reagent (cation-adjusted Mueller Hinton broth (CAMHB) supplemented with antifoam (0.0015%) to reduce air bubble formation) is added to the eTube disposable, followed by the addition of 34 µL of the positive blood culture. The eTube disposable with diluted sample is vortexed and then placed in the eQUANT instrument for incubation.

Once inserted, the eTube disposable sits in a thermal module which is heated to 37°C ± 2°C to grow the bacteria to a concentration equivalent to a 0.5 McFarland (eMcFarland, or eMcF). The eQUANT sensor located in the eTube disposable is an ORP sensor consisting of two electrode components, which both come into direct contact with the diluted positive blood culture sample. The eQUANT ORP sensor responds to changes in the ORP during pathogen growth/metabolism. As the concentration of microorganisms in the sample increases, the growth media becomes reduced, and the voltage measured by the ORP sensor becomes more negative. With the organism ID of the tested sample and the blood culture bottle type as inputs to the system, the algorithm is applied to the real-time voltage measurements to determine the point in time at which the organism concentration reaches a level equivalent to a standard 0.5 McFarland. At the endpoint, the sample immediately starts to cool down to 15°C ± 2°C to inhibit further growth. The sample can be held for up to one hour on the instrument, before being used for downstream agar disk diffusion susceptibility testing.

The eQUANT System can determine the susceptibility of specific organisms when tested against various antimicrobials (**Table 1**).

Table 1. Organism-Specific Breakpoints for Antimicrobials for use with the eQUANT System

Antimicrobial	Organism Group	FDA-Recognized/Approved Breakpoints* (zone diameter in mm)		
		S	I	R
Amoxicillin/ Clavulanate	Enterobacterales	≥ 18	14-17	≤ 13
Ampicillin	Enterobacterales	≥ 17	14-16	≤ 13
Aztreonam	Enterobacterales	≥ 21	18-20	≤ 17
	P. aeruginosa	≥ 22	16-21	≤ 15
Cefazolin	Enterobacterales	≥ 23	20-22	≤ 19
Cefepime	Enterobacterales	≥ 25	19-24	≤ 18
	P. aeruginosa	≥ 18	-	≤ 17
Ceftriaxone	Enterobacterales	≥ 23	20-22	≤ 19
Ertapenem	Enterobacterales	≥ 22	19-21	≤ 18
Gentamicin	Enterobacterales, *P. aeruginosa*	≥ 15	13-14	≤ 12
Levofloxacin	Enterobacterales	≥ 21	17-20	≤ 16
	P. aeruginosa	≥ 22	15-21	≤ 14

Antimicrobial	Organism Group	FDA-Recognized/Approved Breakpoints* (zone diameter in mm)		
		S	I	R
Meropenem	Enterobacterales	≥ 23	20-22	≤ 19
	P. aeruginosa	≥ 19	16-18	≤ 15
	Acinetobacter spp.	≥ 18	15-17	≤ 14
Piperacillin/ Tazobactam	Enterobacterales	≥ 25	21-24	≤ 20
	P. aeruginosa	≥ 21	15-20	≤ 14
	Acinetobacter spp.	≥ 21	18-20	≤ 17
Tobramycin	Enterobacterales, *P. aeruginosa*	≥ 15	13-14	≤ 12

* FDA STIC Website https://www.fda.gov/drugs/development-resources/fda-recognized-antimicrobial-susceptibility-test-interpretive-criteria

(-) Indicates that a corresponding zone diameter range is not defined for that category.

B Principle of Operation:

See Device Description.

C Instrument Description Information:

1. Instrument Name:

   eQUANT Instrument

2. Specimen Identification:

   Gram stain analysis and identification (ID) using a method FDA-cleared for use with positive blood cultures are performed prior to initiating the eQUANT Instrument. The eQUANT Instrument does not track patient samples internally. Sample traceability is up to the user to label the eQUANT eTube disposable with sample information. A Label Tag area has been provided on all eTube disposables to either write sample information or attach a (LIS) label sticker. This Label Tag is visible even when the eQUANT Instrument lid is closed. The user then selects the organism ID and blood culture bottle type from the dropdown menu on the touchscreen user interface of the eQUANT Instrument.

3. Specimen Sampling and Handling:

   Positive blood culture (PBC) samples must be processed immediately or within 12 hours of positivity should delays be unavoidable. The user places the eTube disposable and eQUANT Reagent tube in designated slots in the provided workflow tray. PBC bottles are vortexed and the septum of the PBC bottle is sterilized prior to collecting 500 µL using a syringe and transferring to a sterile tube. One mL of the eQUANT Reagent and 34µL of the PBC sample are aliquoted into the eTube disposable and then vortexed. After the user ensures bubbles have been eliminated, the eTube disposable is placed back into the workflow tray. The eTube sample will be loaded into the eQUANT following instrument prompts.

4. Calibration:

The eQUANT Instrument is equipped with an internal Quality Control that is automatically performed with each run. Once the eTube disposable with sample (diluted PBC) is inserted into the eQUANT Instrument and the run is started, a series of built-in checks are conducted to ensure that essential Instrument and eTube disposable performance specifications are met enabling the generation of an eMcFarland within the target range of 1-2e8 ± 0.6 log CFU/mL.

5. Quality Control:

Quality controls are performed to ensure that the eQUANT Instrument and eTube disposable work according to their intended use and performance specifications. The eQUANT Instrument has a validated built-in Quality Control (Internal QC) that is designed to check the eTube disposable and instrument performance parameters during every run that is performed on the instrument. Good laboratory practices recommend performing additional external QC testing to verify appropriate colony counts and acceptable AST results. Colony counts may be QC tested using contrived or clinical positive blood culture samples to confirm results are in the expected range ($2.51x10^7$ to $7.96x10^8$ CFU/mL). AST may be QC tested using positive blood culture samples contrived with CLSI-recommended QC strains. Alternatively, AST may be QC tested per the disk manufacturer's instructions.

V Substantial Equivalence Information:

A Predicate Device Name(s):
Accelerate Pheno System, Accelerate PhenoTest BC Kit

B Predicate 510(k) Number(s):
K192665

C Comparison with Predicate(s):

Device & Predicate Device(s):	Device: K231536	Predicate: K192665
Device Trade Name	eQUANT System	Accelerate Pheno System, Accelerate PhenoTest BC Kit
General Device Characteristic Similarities		
Intended Use	The eQUANT System is an automated inoculum preparation system that uses potentiometric sensing of oxidation-reduction potential changes due to pathogen metabolism to generate a 0.5 McFarland-equivalent suspension (the eMcFarland or eMcF) from positive blood culture samples. Samples are processed directly from blood culture samples	The Accelerate PhenoTest BC kit is a multiplexed in vitro diagnostic test utilizing both qualitative nucleic acid fluorescence in situ hybridization (FISH) identification and quantitative, antimicrobial susceptibility testing (AST) methods and is intended for use with the Accelerate Pheno system. The Accelerate PhenoTest BC kit is capable of simultaneous detection and identification of multiple microbial targets followed by susceptibility testing of the appropriate detected bacterial

Device & Predicate Device(s):	Device: K231536	Predicate: K192665
	identified as positive by a continuous monitoring blood culture system and confirmed as Gram-negative rods by Gram stain. Organism identification, as determined by an FDA cleared device for direct testing from positive blood culture, must be available before processing samples on the eQUANT System.	organisms. The Accelerate PhenoTest BC kit is performed directly on blood culture samples identified as positive by a continuous monitoring blood culture system. Results are intended to be interpreted in conjunction with Gram stain results.
Antibiotics	Amoxicillin/clavulanate Ampicillin Aztreonam Cefazolin Cefepime Ceftriaxone Ertapenem Gentamicin Levofloxacin Meropenem Piperacillin/Tazobactam Tobramycin	Amikacin Ampicillin Ampicillin/Sulbactam Aztreonam Ceftazidime Ceftaroline Cefepime Ceftriaxone Ciprofloxacin Daptomycin Ertapenem Gentamicin Linezolid Meropenem Piperacillin/Tazobactam Tobramycin Vancomycin
Indicated Organisms	*Acinetobacter* spp. *Citrobacter freundii* *Enterobacter cloacae* *Escherichia coli* *Klebsiella aerogenes* *Klebsiella pneumoniae* *Klebsiella oxytoca* *Proteus mirabilis* *Proteus vulgaris* *Pseudomonas aeruginosa* *Serratia marcescens*	Gram-negative species: *Acinetobacter baumannii* *Citrobacter* spp. *Enterobacter* spp. *Escherichia coli* *Klebsiella* spp. *Proteus* spp. *Pseudomonas aeruginosa* *Serratia marcescens* Additional Gram-positive bacteria and yeast are also included on the Accelerate PhenoTest BC kit.
Sample Type	Positive blood culture aliquot	Same
General Device Characteristic Differences		
Technology	Measure pathogen concentration via potentiometric sensing of changes in oxidation-reduction potential (ORP) during pathogen metabolism. Uses species-specific	Microscopy-based, single cell analysis. Identification via fluorescence *in situ* hybridization (FISH); AST via microscopic observation of individual growing bacterial cells in the presence

Device & Predicate Device(s):	Device: K231536	Predicate: K192665
	and blood culture bottle specific algorithms to determine when a 0.5 McFarland equivalent concentration is reached.	of antimicrobial agents.
Output/Results Reporting	Liquid suspension of bacterial (0.5 McFarland equivalent) suitable for Disk Diffusion susceptibility testing; no results reported	Microbial identification and MIC-based susceptibility test results

VI Standards/Guidance Documents Referenced:

- FDA Class II Special Controls Guidance Document: Antimicrobial Susceptibility Test (AST) Systems; Guidance for Industry and FDA (Issued August 28, 2009)
- CLSI M100-Ed33. *Performance Standards for Antimicrobial Susceptibility Testing*; 33rd Edition (March 2023)
- CLSI. *Performance Standards for Antimicrobial Disk Susceptibility Tests.* 13th ed. CLSI standard M02, Edition 13. Clinical and Laboratory Standards Institute; 2018.

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

The Reproducibility Study was performed to demonstrate that the eQUANT System reproducibly prepares an 0.5 McFarland equivalent inoculum, the eMcFarland, at an organism concentration of $1x10^8$ to $2x10^8$ ± 0.6 log ($2.51x10^7$ to $7.96x10^8$) CFU/mL from a positive blood culture (PBC). The reproducibility of the eQUANT System was assessed across sites (one internal and two external), with six operators, six runs, 12 instruments and consumable lots. A panel of six organisms was contrived in blood culture bottles with human blood added and incubated on a blood culture monitoring system until positivity. From each positive blood culture, initial eQUANT System testing was performed in duplicate by two operators at each site for a total of four eMcFarlands per PBC. The resulting eMcFarlands were plated to confirm that colony counts met the defined concentration specifications.

Reproducibility for all species was acceptable (≥95%) except for *A. baumannii*. Overall colony counts of *A. baumannii* were <95% in-range in the initial study (67/72 = 93.1%) and were further evaluated in a supplemental in-house study in which testing was performed with three instruments and two operators for five days (1 site x 5 days x 1 organism x 3 replicates x 2 operators = 30 eQUANT runs). Data from both studies are collated and summarized in **Table 2**. Performance is summarized for each species tested. Overall reproducibility of the eQUANT System was >95% in-range colony counts and determined to be acceptable.

Table 2. eQUANT System Reproducibility Results Summary

Organism	In-Range Test Results	Reproducibility %
Escherichia coli	72/72	100.0
Pseudomonas aeruginosa	72/72	100.0
Acinetobacter baumannii	97/102	95.1
Proteus vulgaris	72/72	100.0
Serratia marcescens	72/72	100.0
Overall Agreement	457/462	98.9

2. Linearity:

   Not applicable.

3. Analytical Specificity/Interference:

   Endogenous/Exogenous Interfering Substances

   An interfering substances study was performed to evaluate if substances naturally present or artificially introduced into blood culture bottles affect the eQUANT System performance. A panel of nine different bacterial isolates (four isolates of *A. baumannii,* two isolates of *E. coli* and one isolate each of *K. pneumoniae*, *P. vulgaris*, and *P. aeruginosa*) were individually mixed with human whole blood, seeded directly into a BacT/ALERT SA Standard Aerobic blood culture bottle and incubated for growth in a continuous monitoring blood culture system until indicated as positive for growth (i.e., bottle ring). Each potential interfering substance (**Table 3**), and individual antibiotics from each drug class (**Table 4**), were tested at the concentrations listed below. Each test isolate had one control bottle replicate without the interferent added and three test bottle replicates with the interferent added at appropriate concentrations according to the test procedure. For blood culture bottles containing interferents that could not achieve a high concentration (positivity), the organism was grown to positivity without the interferent, and the interferent was spiked directly into the eTube. When acceptance criteria were not met, repeat testing was performed in triplicate. Colony counts and Kirby-Bauer disk diffusion with indicated antimicrobials were performed for all indicated species. Expected AST results (>95% overall CA) were obtained for all organism samples containing endogenous and exogenous interfering substances tested; all categorical errors were minor and less than 3 mm difference when compared to the control replicate (i.e., no potential interferent) AST results (**Table 3**). However, high concentrations of two substances, hemoglobin (*A. baumannii, P. aeruginosa*) and platelets (*P. aeruginosa*), resulted in aborted eQUANT Instrument runs due to aeration blockage errors detected by the instrument. At decreased interferent concentration, valid eMcFarlands were successfully generated and AST results were as expected. The following limitation is included in the device labeling,

   > High concentrations of hemoglobin (>2 g/dL) and platelets (>900,000 µL) may increase the number of Aeration Blockage Errors observed when testing *Acinetobacter* spp. or *P. aeruginosa*, resulting in aborted eQUANT runs.

   In addition, 100% of all colony counts were within the expected range of $1x10^8$ to $2x10^8$ ± 0.6 log ($2.51x10^7$ to $7.96x10^8$) CFU/mL.

Table 3. Endogenous and Exogenous Interfering Substances Study AST Results

Substance	Concentration	Enterobacterales		*P. aeruginosa*		*A. baumannii*	
		No. CA/ Total[a]	% CA	No. CA/ Total[b]	% CA	No. CA/ Total[c]	% CA
Hemoglobin*	20 g/dL 2 g/dL – 8 g/dL[#]	130/132	98.5	21/21	100.0	23/24	95.8
SPS	0.1% w/v	132/132	100.0	20/21	95.2	24/24	100.0
Heparin	3 units/mL	129/132	97.8	21/21	100.0	24/24	100.0
Triglyceride	37 mmol/L	129/132	97.8	21/21	100.0	23/24	95.8
Platelets	1e6/µL 9e5/ µL (*P. aeruginosa*)	132/132	100.0	21/21	100.0	24/24	100.0
WBCs (Buffy Coat)*	12,000/µL	132/132	100.0	21/21	100.0	24/24	100.0
Hematocrit*	50%	129/132	97.8	21/21	100.0	24/24	100.0
Conj. Bilirubin	40 mg/dL	128/132	97.0	21/21	100.0	24/24	100.0
Gamma-Globulin*	50 mg/mL	132/132	100.0	21/21	100.0	24/24	100.0
Unconjug. Bilirubin	40 mg/dL	129/132	97.8	21/21	100.0	24/24	100.0

CA – categorical agreement

[a]Disk diffusion results from amoxicillin/clavulanic acid, ampicillin, aztreonam, cefazolin, cefepime, ceftriaxone, ertapenem, gentamicin, levofloxacin, meropenem, piperacillin/tazobactam, and tobramycin

[b]Disk diffusion results from aztreonam, cefepime, gentamicin, levofloxacin, meropenem, piperacillin/tazobactam and tobramycin

[c]Meropenem and piperacillin/tazobactam disk diffusion results

*Endogenous substances added directly into the eTube as acceptable organism concentrations in the blood culture bottle were not achieved.

#Concentration assessed for *A. baumannii* and *P. aeruginosa*

In a limited study with four resistant isolates, expected AST results (>95% overall CA) were obtained for all antibiotic interfering substances tested except for *A. baumannii* in the presence of chloramphenicol (2.41e2 µmol/L) with a CA of 75% (9/12). Since all three categorical errors were minor with all zone diameter errors having a one mm difference when compared to the control replicate zone diameter result, performance was considered acceptable.

Table 4. Antibiotic Interfering Substances Study AST Results

Substance	Concentration	Enterobacterales		*P. aeruginosa*		*A. baumannii*[d]	
		No. CA/ Total	% CA	No. CA/ Total	% CA	No. CA/ Total	% CA
Ampicillin	2.15e2 µmol/L	60/63[a]	95.2	NA	NA	NA	NA
Ceftriaxone	1.51e3 µmol/L	36/36[b]	100.0	NA	NA	NA	NA
Cefazolin	2.643 mmol/L	33/33[c]	100.0	NA	NA	NA	NA
Chloramphenicol	2.41e2 µmol/L	NA	NA	21/21	100.0	9/12	75.0*
Ciprofloxacin	3.62 µmol/L	36/36[b]	100.0	NA	NA	NA	NA
Gentamicin	6.28 µmol/L	NA	NA	NA	NA	6/6	100.0
Cefepime	492 µg/mL	36/36[b]	100.0	NA	NA	NA	NA
Trimethoprim/ Sulfamethoxazole	1.4e-4 M/ 1.5e-3 M	33/33[c]	100.0	NA	NA	NA	NA
Tetracycline	5.40 µmol/L	NA	NA	21/21	100.0	6/6	100.0
Piperacillin/ Tazobactam	2.13 µmol/L/ 1.02e2 µmol/L	36/36[b]	100.0	NA	NA	NA	NA

CA, categorical agreement; NA, Not applicable.

[a]*E. coli,* one isolate; *P. vulgaris*, one isolate
[b]*E. coli*, one isolate
[c]*K. pneumoniae*, one isolate
[d]*A. baumannii*, one isolate was assessed for each antibiotic except for chloramphenicol, which tested two isolates
[*]Acceptable performance since 3/3 minor errors were ≤ 3mm difference in zone diameter when compared to the control replicate (i.e., no potential interferent).

4. Assay Reportable Range:

Not applicable.

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

Quality Control Testing. Quality control testing was performed each day that testing was conducted. CLSI recommended QC strains for each antimicrobial were tested a sufficient number of times (i.e., at least 20 times/site) at each testing site using the eQUANT System. Quality Control testing was performed using contrived positive blood cultures spiked with one of three CLSI recommended QC strains (*E. coli* ATCC 25922, *K. pneumoniae* ATCC 700603, and *P. aeruginosa* ATCC 27853), covering each of the antimicrobials tested. QC with at least one strain was performed on each day of testing at each site on a rotating basis. QC acceptance criteria was based on eQUANT eMcFarland colony counts and on expected Disk Diffusion AST results.

For the eMcFarland concentration, QC pass rates were based on eMcFarland colony counts within the expected range of 2.51e7 and 7.96e8 CFU/mL. If outside the expected range, QC was repeated. Overall, eMcFarland colony counts were within the expected colony range for 99.2% of samples, with 98.8% in-range for *E. coli* ATCC 25922, 100% in-range for *K. pneumoniae* ATCC 700603, and 98.9% in-range for *P. aeruginosa* ATCC 27853.

QC sample eMcFarlands were also tested using the panel of Disk Diffusion antimicrobials indicated with the QC strain being tested. QC expected ranges and results for the eQUANT System are summarized in **Table 5**. For all antimicrobials, except for meropenem, greater than 95% of results were within the expected range, which is acceptable. Eight of the *P. aeruginosa* ATCC 27853 QC failures with meropenem occurred at Site 2 and were due to a suspected defective set of meropenem antimicrobial disks. The disks were replaced, and meropenem QC at this site subsequently passed. Sample meropenem Disk Diffusion results impacted by the Meropenem QC failures were excluded from performance analysis.

Table 5. QC Expected Ranges and Results for the eQUANT System

Antimicrobial	QC Organism	Expected Range (mm)	No. in Range (%)
			eQUANT
Amoxicillin/Clavulanic Acid	*E. coli* ATCC 25922	18-24	79/79 (100)
Ampicillin	*E. coli* ATCC 25922	15-22	80/80 (100)
Aztreonam	*E. coli* ATCC 25922	28-36	80/80 (100)
	K. pneumoniae ATCC 700603	10-16	82/82 (100)
	P. aeruginosa ATCC 27853	23-29	88/88 (100)
Cefazolin	*E. coli* ATCC 25922	21-27	79/80 (98.8)
Cefepime	*E. coli* ATCC 25922	31-37	79/80 (98.8)
	K. pneumoniae ATCC 700603	23-29	80/81 (98.8)
	P. aeruginosa ATCC 27853	25-31	90/90 (100)
Ceftriaxone	*E. coli* ATCC 25922	29-35	80/80 (100)

Antimicrobial	QC Organism	Expected Range (mm)	No. in Range (%) eQUANT
	K. pneumoniae ATCC 700603	16-24	81/82 (98.8)
	P. aeruginosa ATCC 27853	17-23	89/90 (98.9)
Ertapenem	*E. coli* ATCC 25922	29-36	80/80 (100)
	P. aeruginosa ATCC 27853	13-21	86/88 (97.7)
Gentamicin	*E. coli* ATCC 25922	19-26	80/80 (100)
	P. aeruginosa ATCC 27853	17-23	88/88 (100)
Levofloxacin	*E. coli* ATCC 25922	29-37	80/80 (100)
	P. aeruginosa ATCC 27853	19-26	88/88 (100)
Meropenem	*E. coli* ATCC 25922	28-35	80/80 (100)
	P. aeruginosa ATCC 27853	27-33	76/88 (86.4)*
Piperacillin/Tazobactam	*E. coli* ATCC 25922	24-30	80/80 (100)
	P. aeruginosa ATCC 27853	25-33	87/88 (98.9)
Tobramycin	*E. coli* ATCC 25922	18-26	80/80 (100)
	P. aeruginosa ATCC 27853	20-26	162/162 (100)

*8/12 QC failures with *P. aeruginosa* ATCC 27853 were due to a defective set of meropenem disks. Any PBC clinical samples tested on those days were excluded from the final analysis.

Device Failure. The eQUANT Instrument is equipped with a self-checking mechanism to identify run errors. There were six instrument aborted runs (6/578 = 1.0%) that were observed in the original and supplemental clinical studies. All were detected at the time of failure by the instrument and resulted in excluded samples, and samples were repeated per device labeling. Three aborted runs were due to aeration blockage detection, two aborted runs were due to exceeding maximum eQUANT runtime, and one aborted run was due to an ORP signal error detected. Four of the six runs resolved upon repeat testing. No additional follow-up or instrument changes were performed.

6. Detection Limit:
   Not applicable.

7. Assay Cut-Off:
   Not applicable.

8. Accuracy (Instrument):
   Not applicable.

9. Carry-Over:
   A Carryover Study was performed to evaluate if bacterial carryover occurs between runs on the eQUANT System. Two organisms with distinct colony morphologies, *E. coli* and *P. aeruginosa,* were individually mixed into human whole blood, seeded directly into a BacT/ALERT SA Standard Aerobic blood culture bottle and incubated for growth in a continuous monitoring blood culture system until indicated as positive for growth (i.e., bottle ring). The resulting PBC samples were run on the same eQUANT System within 12 hours of positivity in an alternating pattern for a total of three (3) runs per species. The resulting eMcFarlands were subcultured to assess whether carryover occurred between runs. All colony counts were within the expected range (2.51e7 to 7.96e8 CFU/mL). No carryover was observed, as evidenced by monomicrobial cultures of the expected organism, which was acceptable.

B Comparison Studies:

1. Method Comparison with Predicate Device:
   The purpose of the method comparison study was to demonstrate the clinical performance of the eQUANT System in providing in-range colony counts equivalent to a 0.5 McFarland (2.51e7 to 7.96e8 CFU/mL) and qualitative disk diffusion (DD) AST results direct from positive blood culture (PBC) containing Gram-negative bacteria. AST DD results obtained with inoculum prepared with the eQUANT System were compared to DD results obtained with inoculum prepared from isolated colonies (i.e., standard), according to CLSI M02.

   Positive blood cultures included fresh, leftover samples from patients with suspected bacteremia along with positive blood cultures contrived with clinical stock isolates from the clinical sites or challenge isolates. Clinical stock isolates and challenge isolates were enrolled to supplement fresh positive blood cultures due to low prevalence of certain species and antimicrobial resistance expected during prospective collection. Isolates were subcultured on appropriate media (Tryptic Soy Agar with 5% sheep blood), spiked into blood culture bottles at a concentration of approximately 10^2 CFU per bottle containing fresh human donor blood, and incubated on the appropriate blood culture system until indicated as positive for growth (i.e., bottle ring).

   Testing with the eQUANT System was performed within 12 hours of blood culture positivity with approximately 53% of PBC samples tested within four hours of flagging positive, 28% of samples tested within 4-8 hours of positivity, and the remaining 19% of samples tested within 8-12 hours of positivity. Organism identification obtained from an FDA-cleared identification method was required input into the eQUANT System. An aliquot from all positive blood cultures was subcultured onto Tryptic Soy Agar with 5% Sheep Blood and incubated for 18-24 hours to check for purity and for comparator testing by standard disk diffusion, per CLSI guidelines.

   Since the PBC sample type is different from the sample type evaluated to support FDA-clearance for disk diffusion and testing direct from PBC presents additional risks, clinical studies were performed to evaluate and support individual antimicrobial/organism claims.

   Clinical performance testing on the eQUANT System was initially performed at three U.S. test sites (2 external, 1 internal). For instances in which additional testing was required to supplement existing data from the original study and support specific claims, testing was performed at one internal site.

   In the original clinical study, a total of 165 positive blood culture samples (PBCs) were enrolled with 155 included in performance analysis. Of the 155 PBCs, 42 were fresh, prospective samples (27.1%), 103 were contrived with stock isolates (66.5%) and 10 were contrived with challenge isolates (6.5%). In the supplemental study, a total of 413 PBC samples contrived from stock isolates were enrolled with 412 PBC samples included in performance analysis. A total of 578 samples were enrolled in the original and supplemental study, which included fresh, prospective PBC and contrived PBC spiked with either clinical stock or challenge isolates. Eleven samples were excluded due to off-panel organisms, polymicrobial samples, insufficient growth, sample mix-up, and protocol deviations. In total, 567 samples were included in the performance analysis including 42 fresh, positive blood

cultures, 515 contrived blood cultures with clinical stock isolates and 10 contrived blood cultures with challenge isolates.

Clinical stock and challenge isolates were collected for testing. In the selection of clinical stock isolates, on-panel organisms (i.e., Gram-negative bacteria species included in the eQUANT System indications for use) available from isolate banks at the external and internal clinical sites were selected to supplement species and antimicrobial resistance recommendations. These isolates were originally sourced from clinical specimens from patients admitted at the clinical site or supplied by the sponsor. Pure isolates were cultured and contrived in blood cultures using healthy human donor blood and tested upon indicated as positive for growth (i.e., bottle ring). A panel of challenge isolates was provided for potential testing at the three original clinical sites; however, the results were only included from one external clinical site. These challenge isolates include well-characterized isolates with known resistance profiles that were obtained from isolate repositories, including the CDC/FDA AR Bank.

Disk diffusion performance using eQUANT-prepared inocula was evaluated in comparison to the standard DD. Categorical agreement (CA) was defined as DD interpretation results (S/I/R) that were the same between the eQUANT-prepared and standard-prepared inocula. Very major errors were defined as false susceptible results from the eQUANT-prepared inocula, major errors were defined as false resistance results from the eQUANT-prepared inocula, and minor errors were defined as results with minor discrepancies (i.e., an intermediate result reported as either resistant or susceptible, or vice versa). Since this is a method-to-method comparison, results were considered acceptable if the CA was ≥95% with ≤1% very major errors and ≤1.5% major errors. Drug/organism combinations with CA performance of 90-95% were considered acceptable due to minor errors when the eQUANT zone diameters of a significant number of the errors were ≤3 mm difference compared to the standard disk diffusion zone diameters.

A high-level summary of the eQUANT System performance is described below for each antimicrobial and indicated species. Complete details and results including CA and error rate analyses are summarized in **Table 6**.

Amoxicillin/Clavulanic Acid. A total of 223 Enterobacterales isolates (146 *Escherichia coli,* 15 *Klebsiella oxytoca,* 21 *Klebsiella pneumoniae,* and 41 *Proteus mirabilis*) were evaluated with amoxicillin/clavulanic acid. The combined results from clinical and challenge isolate testing demonstrated a CA of 94.2%. There were 12 minor, 1 major (1/150 = 0.7%) and 0 very major errors. When evaluating results by individual species, *E. coli* had a CA of 93.8% with 8 minor, 1 major (1/93 = 1.1%) and 0 very major errors. Since the eQUANT zone diameter of 6 of the 8 minor errors were ≤3 mm difference compared to the zone diameter of the standard DD method, the CA performance was considered acceptable and a footnote was included in the device labeling to address the CA performance. *P. mirabilis* had a CA of 92.7% with 3 minor, 0 major and 0 very major errors. Since the eQUANT zone diameter of 2 of the 3 minor errors were ≤3 mm difference compared to the zone diameter of the standard DD method, the CA performance was considered acceptable and a footnote was included in the device labeling to address the 90-95% CA performance. The following footnote was included in the device labeling to address the 90-95% CA performance when testing amoxicillin/clavulanic acid:

Categorical agreement was 90-95% due to minor errors for the following drug/organism combinations and considered acceptable since the eQUANT zone diameters of a significant number of the errors were ≤3 mm difference compared to the standard disk diffusion zone diameters:
Amoxicillin/Clavulanic acid: *Escherichia coli*, *Proteus mirabilis*

Ampicillin. A total of 33 *E. coli* isolates were evaluated with ampicillin. The combined results from prospective and stock isolate testing demonstrated a CA of 100% with no errors. Overall, performance is acceptable.

Aztreonam. A total of 189 Enterobacterales isolates (60 *E. coli*, 22 *K. aerogenes*, 21 *K. pneumoniae*, 15 *K. oxytoca*, 8 *E. cloacae*, 21 *P. mirabilis*, 16 *P. vulgaris*, 13 *S. marcescens*, 11 *C. freundii*, and 2 *Citrobacter* spp.) were evaluated with aztreonam. The combined results from clinical and challenge isolate testing demonstrated a CA of 96.8% with 5 minor, 1 major error (1/141 = 0.7%) and 0 very major errors. When evaluating results by individual species, *P. mirabilis* had a CA of 95% with 1 major error (1/20 = 5.0%). The following limitation is included in the device labeling to address the high major error rate:

Perform an alternative method of testing prior to reporting results for the following antibiotic/organism combination(s):
Aztreonam: *Proteus mirabilis* when the disk zone diameter from an eMcFarland inoculum produces a resistant result due to one major error.

A limitation statement is included in the device labeling to address the lack of testing with resistant *Proteus mirabilis* and *Proteus vulgaris* isolates.

A total of 25 *P. aeruginosa* isolates were evaluated with aztreonam. The combined results from clinical and challenge isolate testing demonstrated a CA of 96% with 1 minor error and 0 major and 0 very major errors. Overall, performance was acceptable.

Cefazolin. A total of 213 Enterobacterales isolates (131 *E. coli*, 41 *K. pneumoniae* and 41 *P. mirabilis* isolates) were evaluated with cefazolin. The combined results from clinical and challenge isolate testing demonstrated a CA of 85% with 31 minor errors, 1 major error (1/71 = 1.4%) and 0 very major errors. When evaluating results by individual species, *E. coli* had a CA of 82.4% with 23 minor, 0 major and 0 very major errors. The performance is not acceptable. *P. mirabilis* had a CA of 85.4% with 5 minor, 1 major (1/71 = 1.4%), and 0 very major errors. The performance is not acceptable. *K. pneumoniae* had a CA of 92.7% with three minor, 0 major and 0 very major errors. Since the eQUANT zone diameter of the three minor errors were ≤3 mm difference compared to the zone diameter of the standard DD method, the CA performance was considered acceptable and the following footnote was included in the device labeling to address the 90-95% CA performance:

Categorical agreement was 90-95% due to minor errors for the following drug/organism combinations and considered acceptable since the eQUANT zone diameters of a significant number of the errors were ≤3 mm difference compared to the standard disk diffusion zone diameters:
Cefazolin: *Klebsiella pneumoniae*

Due to the unacceptable performance for *E. coli* and *P. mirabilis* these drug/organism combinations are not indicated for use with the eQUANT System. The following limitation is included in the device labeling to restrict reporting of *E. coli* and *P. mirabilis* due to unacceptable performance:

> Perform an alternative method of testing prior to reporting results for the following antibiotic/organism combination(s):
> - Cefazolin: *Escherichia coli, Proteus mirabilis*

Cefepime. A total of 237 Enterobacterales isolates (40 *E. coli*, 21 *S. marcescens*, 21 *K. pneumoniae*, 15 *K. oxytoca*, 21 *K. aerogenes*, 41 *E. cloacae*, 21 *C. freundii*, 16 *P. vulgaris*, and 41 *P. mirabilis* isolates) were evaluated with cefepime. The combined results from clinical and challenge isolate testing demonstrated a CA of 91.2% with 18 minor errors, 1 major error (1/162 = 0.6%) and 0 very major errors. When evaluating results by individual species, *C. freundii* had a CA of 81% with 4 minor errors, 0 major and 0 very major errors. The performance was not acceptable. *K. aerogenes* had a CA of 85.7% with 3 minor errors, 0 major and 0 very major errors. The performance was not acceptable. *E. cloacae* had a CA of 90.2% with 4 minor errors, 0 major and 0 very major errors. Since the eQUANT zone diameter of 2 of the 4 minor errors were ≤3 mm difference compared to the zone diameter of the standard DD method, the CA performance was considered acceptable and a footnote was included in the device labeling to address the CA performance. *K. oxytoca* had a CA of 93.3% with one minor error, 0 major and 0 very major errors. Since the eQUANT zone diameter of the minor error was ≤3 mm difference compared to the zone diameter of the standard DD method, the CA performance was considered acceptable and a footnote was included in the device labeling to address the CA performance. *P. mirabilis* had a CA of 92.7% with 2 minor errors, 1 major error (1/35 = 2.9%), and 0 very major errors. Since the eQUANT zone diameter of 2 of the 4 minor errors were ≤3 mm difference compared to the zone diameter of the standard DD method, the CA performance was considered acceptable and a footnote was included in the device labeling to address the 90-95% CA performance. The following footnote was included in the device labeling to address the 90-95% CA performance when testing cefepime.

> Categorical agreement was 90-95% due to minor errors for the following drug/organism combinations and considered acceptable since the eQUANT zone diameters of a significant number of the errors were ≤3 mm difference compared to the standard disk diffusion zone diameters:
> Cefepime: *Enterobacter cloacae, Klebsiella oxytoca*, *Proteus mirabilis*

Due to the unacceptable performance for *C. freundii* and *K. aerogenes*, these drug/organism combinations are not indicated for use with the eQUANT System. The following limitation is included in the device labeling to restrict reporting of *C. freundii* and *K. aerogenes* due to unacceptable performance and to address the high major error rate for *P. mirabilis*:

> Perform an alternative method of testing prior to reporting results for the following antibiotic/organism combination(s):
> Cefepime: *Citrobacter freundii, Klebsiella aerogenes*
> Cefepime: *Proteus mirabilis* when the disk zone diameter from an eMcFarland inoculum produces a resistant result due to one major error.

A limitation statement is included in the device labeling to address the lack of testing with resistant *Klebsiella oxytoca*, *Proteus mirabilis*, *Proteus vulgaris,* and *Serratia marcescens* isolates.

A total of 41 *P. aeruginosa* isolates were evaluated with cefepime. The combined results from clinical and challenge isolate testing demonstrated a CA of 95.1% with 0 minor errors and two major errors (2/21= 9.5%) and 0 very major errors. Due to the lack of an intermediate interpretive criterion and since the eQUANT zone diameter of the two major errors were ≤3 mm difference compared to the zone diameter of the standard DD method, the major error rate was considered acceptable and the following footnote was included in the device labeling to address the high major error rate:

> The major error rate was ≥1.5% and considered acceptable since the eQUANT zone diameters for the major errors were ≤3 mm difference compared to the standard disk diffusion zone diameters.

Ceftriaxone. A total of 223 Enterobacterales isolates (33 *E. coli*, 13 *S. marcescens*, 21 *K. pneumoniae*, 15 *K. oxytoca*, 42 *K. aerogenes*, 21 *E. cloacae*, 21 *C. freundii*, 16 *P. vulgaris*, and 41 *P. mirabilis* isolates) were evaluated with ceftriaxone. The combined results from clinical and challenge isolate testing demonstrated a CA of 96.9% with 5 minor errors, two major errors (2/134 = 1.5%) and 0 very major errors. When evaluating results by individual species, *K. aerogenes* had a CA of 92.9% with 2 minor errors, 1 major error (1/21 = 4.8%), and 0 very major errors. Since the eQUANT zone diameter of 1 of the 2 minor errors were ≤3 mm difference compared to the zone diameter of the standard DD method, the CA performance was considered acceptable and is addressed in a footnote included in the device labeling. In general, the eQUANT zone diameter for ceftriaxone/*K. aerogenes* results tended to be smaller (i.e., more resistant) compared to the standard DD method, which may contribute to the CA and cause the single major error, which is addressed in a footnote included in the device labeling. *P. mirabilis* had a CA of 92.7% with two minor errors, one major (1/32 = 3.1%) and 0 very major errors. Since the eQUANT zone diameter of 1 of the 2 minor errors were ≤3 mm difference compared to the zone diameter of the standard DD method, the CA performance was considered acceptable and is addressed in a footnote included in the device labeling. In general, the eQUANT zone diameter for ceftriaxone/*P. mirabilis* results tended to be smaller (i.e., more resistant) compared to the standard DD method, which may contribute to the CA and cause the single major error, which is addressed in a footnote included in the device labeling. To address the 90-95% CA performance and high major error rate for *K. aerogenes* and *P. mirabilis* when testing ceftriaxone, the following statement is included as a footnote to the performance table in the device labeling:

> A single major error was observed for the following drug/organism combinations which may be due to eQUANT zone diameters tending to be smaller than the standard disk diffusion zone diameters:
> Ceftriaxone: *Klebsiella aerogenes, Proteus mirabilis*

In addition, the following footnote was included in the device labeling to address the 90-95% CA performance for *K. aerogenes* and *P. mirabilis*:

Categorical agreement was 90-95% due to minor errors for the following drug/organism combinations and considered acceptable since the eQUANT zone diameters of a significant number of the errors were ≤3 mm difference compared to the standard disk diffusion zone diameters:
Ceftriaxone: *Klebsiella aerogenes*, *Proteus mirabilis*

A limitation statement is included in the device labeling to address the lack of testing with resistant *Proteus vulgaris* isolates.

Ertapenem. A total of 187 Enterobacterales isolates (40 *E. coli*, 41 *S. marcescens*, 21 *K. pneumoniae*, 16 *K. oxytoca*, 13 *K. aerogenes*, 8 *E. cloacae*, 21 *C. freundii*, 16 *P. vulgaris*, and 11 *P. mirabilis* isolates) were evaluated with ertapenem. The combined results from clinical and challenge isolate testing demonstrated a CA of 97.9% with 4 minor errors, 0 major and 0 very major errors. Overall, performance is acceptable.

A limitation statement is included in the device labeling to address the lack of testing with resistant *Klebsiella oxytoca* and *Proteus vulgaris* isolates.

Gentamicin. A total of 255 Enterobacterales isolates (80 *E. coli*, 22 *S. marcescens*, 21 *K. pneumoniae*, 16. *K. oxytoca*, 13 *K. aerogenes*, 21 *E. cloacae*, 41 *C. freundii*, 2 *Citrobacter* spp., 16 *P. vulgaris*, 22 *P. mirabilis* and one *Proteus* spp. isolates) were evaluated with gentamicin. The combined results from clinical and challenge isolate testing demonstrated a CA of 97.3% with 5 minor errors, 1 major error (1/227 = 0.4%) and 1 very major errors (1/26 = 3.8%). When evaluating results by individual species, *E. coli* had a CA of 97.5% with 0 minor, 1 major (1/75 = 1.3%), and 1 very major errors (1/4 = 25%). The one very major error (1/4 = 25%) was considered a random error due to the limited number of resistant isolates tested. Therefore, the performance was considered acceptable.

A limitation statement is included in the device labeling to address the lack of testing with resistant *Klebsiella oxytoca*, *Citrobacter freundii, Proteus vulgaris,* and *Serratia marcescens* isolates.

A total of 41 *P. aeruginosa* isolates were evaluated with gentamicin. The combined results from clinical and challenge isolate testing demonstrated a CA of 95.1% with two minor errors, 0 major and 0 very major errors. Overall, performance was acceptable.

Levofloxacin. A total of 206 Enterobacterales isolates (40 *E. coli*, 21 *S. marcescens*, 41 *K. pneumoniae*, 16. *K. oxytoca*, 13 *K. aerogenes*, 8 *E. cloacae*, 11 *C. freundii*, 15 *P. vulgaris*, and 41 *P. mirabilis* isolates) were evaluated with levofloxacin. The combined results from clinical and challenge isolate testing demonstrated a CA of 96.1% with 8 minor errors, 0 major and 0 very major errors. When evaluating results by individual species, *P. mirabilis* had a CA of 92.7% with 3 minor errors, 0 major and 0 very major errors. Since the eQUANT zone diameter of the 3 minor errors were ≤3 mm difference compared to the zone diameter of the standard DD method, the CA performance was considered acceptable and the following footnote was included in the device labeling to address the 90-95% CA performance of *P. mirabilis*:

Categorical agreement was 90-95% due to minor errors for the following drug/organism combinations and considered acceptable since the eQUANT zone

diameters of a significant number of the errors were ≤3 mm difference compared to the standard disk diffusion zone diameters:
Levofloxacin: *Proteus mirabilis*

A limitation statement is included in the device labeling to address the lack of testing with resistant *Klebsiella oxytoca*, *Klebsiella aerogenes*, *Proteus vulgaris,* and *Serratia marcescens* isolates.

A total of 25 *P. aeruginosa* isolates were evaluated with levofloxacin. The combined results from clinical and challenge isolate testing demonstrated a CA of 96% with 1 minor, 0 major and 0 very major errors. Overall, performance was acceptable.

Meropenem. A total of 206 Enterobacterales isolates (76 *E. coli*, 40 *S. marcescens*, 19 *K. pneumoniae*, 16. *K. oxytoca*, 8 *E. cloacae*, 21 *C. freundii*, 16 *P. vulgaris*, and 10 *P. mirabilis* isolates) were evaluated with meropenem. The combined results from clinical and challenge isolate testing demonstrated a CA of 97.1% with 4 minor errors, 2 major error (2/180 = 1.1%) and 0 very major errors. When evaluating results by individual species, *S. marcescens* had a CA of 95% with 1 minor, 1 major (1/36 = 2.78%) and 0 very major errors. In general, the eQUANT zone diameter for meropenem/*S. marcescens* results tended to be smaller (i.e., more resistant) compared to the standard DD method, which may have caused the single major error. To address the high major error rate for *S. marcescens*, the following statement is included as a footnote to the performance table in the device labeling:

The categorical agreement was ≥95% for the following drug/organism combinations; however, a single major error was observed which may be due to eQUANT zone diameters tending to be smaller than the standard disk diffusion zone diameters:
Meropenem: *Serratia marcescens*

A total of 25 *P. aeruginosa* isolates were evaluated with meropenem. The combined results from clinical and challenge isolate testing demonstrated a CA of 100% with 0 minor, 0 major and 0 very major errors. Overall, performance was acceptable.

A total of 16 *Acinetobacter* spp. isolates were evaluated with meropenem. The combined results from clinical and challenge isolate testing demonstrated a CA of 100% with 0 minor, major and very major errors. Overall, performance was acceptable.

Piperacillin/Tazobactam. A total of 239 Enterobacterales isolates (157 *E. coli*, 23 *S. marcescens*, 21 *K. pneumoniae*, 16 *P. vulgaris*, and 22 *P. mirabilis* isolates) were evaluated with piperacillin/tazobactam. The combined results from clinical and challenge isolate testing demonstrated a CA of 94.1% with 14 minor errors, and 0 major error or 0 very major errors. When evaluating results by individual species, *E. coli* had a CA of 93.0% with 11 minor, 0 major and 0 very major errors. Since the eQUANT zone diameter of 7 of the 11 minor errors were ≤3 mm difference compared to the zone diameter of the standard DD method, the CA performance was considered acceptable and the following footnote was included in the device labeling to address the 90-95% CA performance:

Categorical agreement was 90-95% due to minor errors for the following drug/organism combinations and considered acceptable since the eQUANT zone

diameters of a significant number of the errors were ≤3 mm difference compared to the standard disk diffusion zone diameters:
Piperacillin/Tazobactam: *Escherichia coli*

A limitation statement is included in the device labeling to address the lack of testing with resistant *Proteus vulgaris* isolates.

A total of 25 *P. aeruginosa* isolates were evaluated with piperacillin/tazobactam. The combined results from clinical and challenge isolate testing demonstrated a CA of 96% with 1 minor, 0 major and 0 very major errors. Overall, performance was acceptable.

A total of 41 *Acinetobacter* spp. isolates were evaluated with piperacillin/tazobactam. The combined results from clinical and challenge isolate testing demonstrated a CA of 95.1% with two minor errors, 0 major and 0 very major errors. Overall, performance was acceptable.

Tobramycin. A total of 203 Enterobacterales isolates (60 *E. coli*, 13 *S. marcescens*, 21 *K. pneumoniae*, 16. *K. oxytoca*, 22 *K. aerogenes*, 21 *E. cloacae*, 11 *C. freundii*, 2 *Citrobacter* spp., 16 *P. vulgaris*, and 21 *P. mirabilis* isolates) were evaluated with tobramycin. The combined results from clinical and challenge isolate testing demonstrated a CA of 96.6% with 7 minor errors, 0 major and 0 very major errors. Overall, the performance was acceptable.

A limitation statement is included in the device labeling to address the lack of testing with resistant *Klebsiella aerogenes* and *Proteus vulgaris* isolates.

A total of 25 *P. aeruginosa* isolates were evaluated with tobramycin. The combined results from clinical and challenge isolate testing demonstrated a CA of 100% with 0 minor, 0 major and 0 very major errors. Overall, performance was acceptable.

Table 6. eQUANT System Clinical Performance Compared to Standard DD

	Tot	**No. CA**	**CA%**	**No. R or NS**	**No. S**	**min**	**maj**	**vmj**
Amoxicillin/Clavulanic Acid-Enterobacterales [Breakpoints (mm): ≥18(S), 14-17(I), ≤13(R)]								
Challenge Seeded	3	3	100.0	1	2	0	0	0
Fresh	37	33	89.1	6	25	3	1	0
Clinical Seeded	183	174	95.1	53	123	9	0	0
Total	**223**	**210**	**94.2**[1]	**60**	**150**	**12**	**1**	**0**
Ampicillin-*Escherichia coli* [Breakpoints (mm): ≥17(S), 14-16(I), ≤13(R)]								
Challenge Seeded	-	-	-	-	-	-	-	-
Fresh	25	25	100.0	15	9	0	0	0
Clinical Seeded	8	8	100.0	8	0	0	0	0
Total	**33**	**33**	**100.0**	**23**	**9**	**0**	**0**	**0**
Aztreonam-Enterobacterales [Breakpoints (mm): ≥21(S), 18-20(I), ≤17(R)]								
Challenge Seeded	7	7	100.0	1	5	0	0	0
Fresh	40	38	95.0	4	36	2	0	0
Clinical Seeded	142	138	97.2	40	100	3	1	0
Total	**189**	**183**	**96.8**	**45**	**141**	**5**	**1**	**0**
Aztreonam-*Pseudomonas aeruginosa* [Breakpoints (mm): ≥22(S), 16-21(I), ≤15(R)]								
Challenge Seeded	1	1	100.0	0	1	0	0	0
Fresh	1	1	100.0	0	1	0	0	0

	Tot	No. CA	CA%	No. R or NS	No. S	min	maj	vmj
Clinical Seeded	23	22	95.7	11	8	1	0	0
Total	**25**	**24**	**96.0**	**11**	**10**	**1**	**0**	**0**
Cefazolin-Enterobacterales [Breakpoints (mm): ≥23(S), 20-22(I), ≤19(R)]								
Challenge Seeded	2	1	50.0	1	1	1	0	0
Fresh	37	28	75.7	12	19	9	0	0
Clinical Seeded	174	152	87.4	85	51	21	1	0
Total	**213**	**181**	**85.0[1]**	**98**	**71**	**31**	**1**	**0**
Cefepime-Enterobacterales [Breakpoints (mm): ≥25(S), 19-24(I), ≤18(R)]								
Challenge Seeded	7	7	100.0	0	6	0	0	0
Fresh	40	38	95.0	4	33	2	0	0
Clinical Seeded	190	173	91.1	38	123	16	1	0
Total	**237**	**218**	**92.0[1]**	**42**	**162**	**18**	**1**	**0**
Cefepime-*Pseudomonas aeruginosa* [Breakpoints (mm): ≥18(S), (-)(I), ≤17(R)]								
Challenge Seeded	1	1	100.0	0	1	0	0	0
Fresh	1	1	100.0	0	1	0	0	0
Clinical Seeded	39	37	94.9	20	19	0	2	0
Total	**41**	**39**	**95.1**	**20**	**21**	**0**	**2**	**0**
Ceftriaxone-Enterobacterales [Breakpoints (mm): ≥23(S), 20-22(I), ≤19(R)]								
Challenge Seeded	7	7	100.0	2	5	0	0	0
Fresh	40	39	97.5	7	33	0	1	0
Clinical Seeded	176	170	97.0	75	96	5	1	0
Total	**223**	**216**	**96.9**	**84**	**134**	**5**	**2**	**0**
Ertapenem-Enterobacterales Breakpoints (mm): ≥22(S), 19-21(I), ≤18(R)]								
Challenge Seeded	7	7	100.0	0	7	0	0	0
Fresh	40	40	100.0	0	40	0	0	0
Clinical Seeded	140	136	97.1	29	106	4	0	0
Total	**187**	**183**	**97.9**	**29**	**153**	**4**	**0**	**0**
Gentamicin-Enterobacterales [Breakpoints (mm): ≥15(S), 13-14(I), ≤12(R)]								
Challenge Seeded	7	7	100.0	1	6	0	0	0
Fresh	41	39	95.1	4	37	0	1	1
Clinical Seeded	207	202	97.6	21	184	5	0	0
Total	**255**	**248**	**96.9**	**26**	**227**	**5**	**1**	**1**
Gentamicin-*Pseudomonas aeruginosa* [Breakpoints (mm): ≥15(S), 13-14(I), ≤12(R)]								
Challenge Seeded	1	1	100.0	0	1	0	0	0
Fresh	1	1	100.0	0	1	0	0	0
Clinical Seeded	39	37	94.9	17	21	2	0	0
Total	**41**	**39**	**95.1**	**17**	**23**	**2**	**0**	**0**
Levofloxacin-Enterobacterales [Breakpoints (mm): ≥21(S), 17-20(I), ≤16(R)]								
Challenge Seeded	7	7	100.0	1	5	0	0	0
Fresh	40	37	92.5	7	33	3	0	0
Clinical Seeded	159	154	96.9	41	108	5	0	0
Total	**206**	**198**	**96.1**	**49**	**146**	**8**	**0**	**0**
Levofloxacin-*Pseudomonas aeruginosa* [Breakpoints (mm): ≥22(S), 15-21(I), ≤14(R)]								
Challenge Seeded	1	1	100.0	0	1	0	0	0
Fresh	1	1	100.0	0	1	0	0	0
Clinical Seeded	23	22	95.7	11	6	1	0	0
Total	**25**	**24**	**96.0**	**11**	**8**	**1**	**0**	**0**
Meropenem-*Acinetobacter* spp.[Breakpoints (mm): ≥18(S), 15-17(I), ≤14(R)]								
Challenge Seeded	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**
Fresh	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**
Clinical Seeded	16	16	100.0	9	6	0	0	0
Total	**16**	**16**	**100.0**	**9**	**6**	**0**	**0**	**0**
Meropenem-Enterobacterales [Breakpoints (mm): ≥23(S), 20-22(I), ≤19R)]								
Challenge Seeded	6	6	100.0	0	6	0	0	0

	Tot	No. CA	CA%	No. R or NS	No. S	min	maj	vmj
Fresh	34	34	100.0	0	34	0	0	0
Clinical Seeded	166	160	96.4	23	140	4	2	0
Total	**206**	**200**	**97.1**	**23**	**180**	**4**	**2**	**0**
Meropenem-*Pseudomonas aeruginosa* [Breakpoints (mm): ≥19(S), 16-18(I), ≤15R)]								
Challenge Seeded	1	1	100.0	0	1	0	0	0
Fresh	1	1	100.0	0	1	0	0	0
Clinical Seeded	23	23	100.0	17	6	0	0	0
Total	**25**	**25**	**100.0**	**17**	**8**	**0**	**0**	**0**
Piperacillin-Tazobactam-*Acinetobacter* spp. [Breakpoints (mm): ≥21(S), 18-20(I), ≤17R)]								
Challenge Seeded	2	2	100.0	1	1	0	0	0
Fresh	**-**	**-**	**-**	**-**	**-**	**-**	**-**	**-**
Clinical Seeded	39	37	94.9	24	15	2	0	0
Total	**41**	**39**	**95.1**	**25**	**16**	**2**	**0**	**0**
Piperacillin-Tazobactam-Enterobacterales [Breakpoints (mm): ≥25(S), 21-24(I), ≤20R)]								
Challenge Seeded	4	4	100.0	1	3	0	0	0
Fresh	38	33	86.8	1	30	5	0	0
Clinical Seeded	197	188	95.4	44	133	9	0	0
Total	**239**	**225**	**94.1**[1]	**46**	**166**	**14**	**0**	**0**
Piperacillin-Tazobactam - *Pseudomonas aeruginosa* [Breakpoints (mm): ≥21(S), 15-20(I), ≤14R)]								
Challenge Seeded	1	1	100.0	0	1	0	0	0
Fresh	1	1	100.0	0	1	0	0	0
Clinical Seeded	23	22	95.7	9	12	1	0	0
Total	**25**	**24**	**96.0**	**9**	**14**	**1**	**0**	**0**
Tobramycin-Enterobacterales [Breakpoints (mm): ≥15(S), 13-14(I), ≤12R)]								
Challenge Seeded	7	7	100.0	1	6	0	0	0
Fresh	40	37	92.5	3	36	3	0	0
Clinical Seeded	156	152	97.4	29	122	4	0	0
Total	**203**	**196**	**96.6**	**33**	**164**	**7**	**0**	**0**
Tobramycin-*Pseudomonas aeruginosa* [Breakpoints (mm): ≥15(S), 13-14(I), ≤12R)]								
Challenge Seeded	1	1	100.0	0	1	0	0	0
Fresh	1	1	100.0	0	1	0	0	0
Clinical Seeded	23	23	100.0	8	14	0	0	0
Total	**25**	**25**	**100.0**	**8**	**16**	**0**	**0**	**0**

CA – Category Agreement
R – Resistant isolates
NS – Non-susceptible isolates
S – Susceptible isolates
min – minor errors
maj – major errors
vmj – very major errors

[1]CA Performance (<95%) is described above and addressed in device labeling.

Trending

A trending analysis was conducted using the combined data (fresh, clinical seeded and challenge seeded) to evaluate antimicrobial-organism combinations for which DD zone diameters obtained from eQUANT inoculum tended to be ≥ 3 mm smaller or larger than DD zone diameters obtained from the standard DD method (colony inoculum).

Trending results were stratified by species to determine if species-related trends were observed (**Table 7**). Species for which the difference between the percentage of isolates with larger or smaller zone diameter sizes was ≥15% with a statistically significant confidence interval were considered to have evidence of trending.

Table 7. Trending by Species (fresh, clinical seeded and challenge seeded samples)

	eQUANT System						
Drug	**Organism**	**Total**	**≥3 mm Smaller Zone Size # (%)**	**Equivalent Zone Size (-2 to +2 mm)**	**≥3mm Larger Zone Size # (%)**	**Percent Difference (95% CI)**	**Trending Noted (≥15%)**
Amoxicillin-Clavulanate	*E. coli*	146	5 (3.4)	138	3 (2.1)	+1.3% (-2.9%, 5.9%)	No
Amoxicillin-Clavulanate	*K. oxytoca*	15	2 (13.3)	12	1 (6.7)	+6.6% (-18.4%, 31.8%)	No
Amoxicillin-Clavulanate	*K. pneumoniae*	21	1 (4.8)	20	0 (0)	+4.8% (-11.2%, 22.7%)	No
Amoxicillin-Clavulanate	***P. mirabilis***	**41**	**14 (34.1)**	**27**	**0 (0)**	**+34.1% (18.9%, 49.5%)**	**Yes**
Ampicillin	*E. coli*	33	1 (3.2)	31	1 (3.2)	0.0% (-12.5%, 12.5%)	No
Aztreonam	***C. freundii***	**11**	**2 (18.2)**	**9**	**0 (0.0)**	**+18.2% (-10.8%, 47.7%)**	**Yes**
Aztreonam	*Citrobacter* spp.	2	0 (0.0)	2	0 (0.0)	0.0% (-65.8%, 65.8%)	No
Aztreonam	*E. cloacae*	8	0 (0.0)	8	0 (0.0)	0.0% (-32.4%, 32.4%)	No
Aztreonam	***E. coli***	**60**	**14 (23.3)**	**44**	**2 (3.3)**	**+20.0 (8.0%, 32.3%)**	**Yes**
Aztreonam	*K. aerogenes*	22	3 (13.6)	19	0 (0.0)	+13.6 (-3.7%, 33.3%)	No
Aztreonam	*K. oxytoca*	15	1 (6.7)	14	0 (0.0)	+6.7% (-14.5%, 29.8%)	No
Aztreonam	*K. pneumoniae*	21	0 (0.0)	21	0 (0.0)	0.0% (-15.5%, 15.5%)	No
Aztreonam	***P. mirabilis***	**21**	**6 (28.6)**	**15**	**0 (0.0)**	**+28.6% (7.2%, 50.0%)**	**Yes**
Aztreonam	***P. vulgaris***	**16**	**7 (43.8)**	**8**	**1 (6.3)**	**+37.5% (7.3%, 61.1%)**	**Yes**
Aztreonam	*S. marcescens*	13	2 (15.4)	10	1 (7.7)	+7.7% (-20.2%, 35.3%)	No
Aztreonam	*P. aeruginosa*	25	0 (0.0)	25	0 (0.0)	+0.0% (-13.3%, 13.3%)	No
Cefazolin	*E. coli*	131	8 (6.1)	118	5 (3.8)	+2.3% (-3.4%, 8.2%)	No
Cefazolin	*K. pneumoniae*	41	0 (0)	38	3 (7.3)	-7.3% (-19.4%, 2.5%)	No
Cefazolin	*P. mirabilis*	41	4 (9.8)	37	0 (0.0)	+9.8% (-0.6%, 22.6%)	No
Cefepime	*C. freundii*	21	2 (9.5)	14	5 (23.8)	-14.3% (-36.7%, 9.2%)	No
Cefepime	*E. cloacae*	41	2 (4.9)	31	8 (19.5)	-14.6% (-29.6%, 0.0%)	No
Cefepime	***E. coli***	**40**	**10 (25.0)**	**27**	**3 (7.5)**	**+17.5% (1.1%, 33.5%)**	**Yes**
Cefepime	***K. aerogenes***	**21**	**5 (23.8)**	**16**	**0 (0.0)**	**+23.8% (3.5%, 45.1%)**	**Yes**
Cefepime	*K. oxytoca*	15	2 (13.3)	12	1 (6.7)	+6.6% (-18.4%, 31.8%)	No
Cefepime	*K. pneumoniae*	21	2 (9.5)	18	1 (4.8)	+4.7% (-14.4%, 24.5%)	No
Cefepime	***P. mirabilis***	**41**	**15 (36.6)**	**26**	**0 (0.0)**	**+36.6%**	**Yes**

	eQUANT System						
Drug	**Organism**	**Total**	**≥3 mm Smaller Zone Size # (%)**	**Equivalent Zone Size (-2 to +2 mm)**	**≥3mm Larger Zone Size # (%)**	**Percent Difference (95% CI)**	**Trending Noted (≥15%)**
						(21.0%, 51.9%)	
Cefepime	***P. vulgaris***	**16**	**8 (50.0)**	**7**	**1 (6.3)**	**+43.5% (12.6%, 66.3%)**	**Yes**
Cefepime	*S. marcescens*	21	3 (14.3)	18	0 (0.0)	+14.3% (-3.8%, 34.6%)	No
Cefepime	*P. aeruginosa*	41	6 (14.6)	35	0 (0.0)	+14.6% (3.1%, 28.4%)	No
Ceftriaxone	*C. freundii*	21	1 (4.8)	17	3 (14.3)	-9.5% (-30.3%, 10.7%)	No
Ceftriaxone	*E. cloacae*	21	1 (4.8)	17	3 (14.3)	-9.5% (-30.3%, 10.7%)	No
Ceftriaxone	***E. coli***	**33**	**8 (24.2)**	**25**	**0 (0.0)**	**+24.2% (8.8%, 41.0%)**	**Yes**
Ceftriaxone	***K. aerogenes***	**42**	**13 (31.0)**	**29**	**0 (0.0)**	**+31.0% (16.4%, 46.0%)**	**Yes**
Ceftriaxone	*K. oxytoca*	15	1 (6.7)	12	2 (13.3)	-6.7% (-31.8%, 18.4%)	No
Ceftriaxone	*K. pneumoniae*	21	1 (4.8)	20	0 (0.0)	+4.8% (-11.2%, 22.7%)	No
Ceftriaxone	***P. mirabilis***	**41**	**15 (36.6)**	**26**	**0 (0.0)**	**+36.6% (21.0%, 51.9%)**	**Yes**
Ceftriaxone	***P. vulgaris***	**16**	**5 (31.3)**	**11**	**0 (0.0)**	**+31.3% (5.4%, 55.6%)**	**Yes**
Ceftriaxone	*S. marcescens*	13	0 (0.0)	13	0 (0.0)	0.0% (-22.8%, 22.8%)	No
Ertapenem	*C. freundii*	21	3 (14.3)	14	4 (19.0)	-4.8% (-27.7%, 18.6%)	No
Ertapenem	*E. cloacae*	8	1 (12.5)	7	0 (0.0)	+12.5% (-21.5%, 47.1%)	No
Ertapenem	***E. coli***	**40**	**8 (20.0)**	**30**	**2 (5.0)**	**+15.0% (0.1%, 30.2%)**	**Yes**
Ertapenem	*K. aerogenes*	13	1 (7.7)	12	0 (0.0)	+7.7% (-16.0%, 33.3%)	No
Ertapenem	***K. oxytoca***	**16**	**4 (25.0)**	**11**	**1 (6.3)**	**+18.8% (-7.8%, 43.8%)**	**Yes**
Ertapenem	*K. pneumoniae*	21	0 (0.0)	21	0 (0.0)	0.0% (-15.5%, 15.5%)	No
Ertapenem	***P. mirabilis***	**11**	**2 (18.2)**	**9**	**0 (0.0)**	**+18.2% (-10.8%, 47.7%)**	**Yes**
Ertapenem	***P. vulgaris***	**16**	**7 (43.8)**	**9**	**0 (0.0)**	**+43.8% (15.4%, 66.8%)**	**Yes**
Ertapenem	***S. marcescens***	**41**	**15 (36.6)**	**26**	**0 (0.0)**	**+36.6% (21.0%, 51.9%)**	**Yes**
Gentamicin	*C. freundii*	41	5 (12.2)	34	2 (4.9)	+7.3% (-5.9%, 21.1%)	No
Gentamicin	*Citrobacter spp.*	2	0 (0.0)	2	0 (0.0)	0.0% (-65.8%, 65.8%)	No
Gentamicin	*E. cloacae*	21	0 (0.0)	21	0 (0.0)	0.0% (-15.5%, 15.5%)	No
Gentamicin	*E. coli*	80	4 (5.0)	75	1 (1.3)	+3.8% (-2.5%, 11.0%)	No
Gentamicin	***K. aerogenes***	**13**	**2 (15.4)**	**11**	**0 (0.0)**	**+15.4%**	**Yes**

	eQUANT System						
Drug	**Organism**	**Total**	**≥3 mm Smaller Zone Size # (%)**	**Equivalent Zone Size (-2 to +2 mm)**	**≥3mm Larger Zone Size # (%)**	**Percent Difference (95% CI)**	**Trending Noted (≥15%)**
						(-42.2%, 10.0%)	
Gentamicin	*K. oxytoca*	16	0 (0.0)	16	0 (0.0)	0.0% (-19.4%, 19.4%)	No
Gentamicin	*K. pneumoniae*	21	0 (0.0)	21	0 (0.0)	0.0% (-15.5%, 15.5%)	No
Gentamicin	*P. mirabilis*	22	3 (13.6)	19	0 (0.0)	+13.6% (-3.7%, 33.3%)	No
Gentamicin	*Proteus* spp.	1	0 (0.0)	1	0 (0.0)	0.0% (-79.4%, 79.4%)	No
Gentamicin	***P. vulgaris***	**16**	**3 (18.8)**	**13**	**0 (0.0)**	**+18.8% (-4.1%, 43.0%)**	**Yes**
Gentamicin	*S. marcescens*	22	0 (0.0)	22	0 (0.0)	0.0% (-14.9%, 14.9%)	No
Gentamicin	*P. aeruginosa*	41	0 (0.0)	41	0 (0.0)	0.0% (-8.6%, 8.6%)	No
Levofloxacin	*C. freundii*	11	0 (0.0)	11	0 (0.0)	0.0% (-25.9%, 25.9%)	No
Levofloxacin	*E. cloacae*	8	0 (0.0)	8	0 (0.0)	0.0% (-32.4%, 32.4%)	No
Levofloxacin	*E. coli*	40	2 (5.0)	35	3 (7.5)	-2.5% (-15.4%, 10.0%)	No
Levofloxacin	***K. aerogenes***	**13**	**2 (15.4)**	**11**	**0 (0.0)**	**+15.4% (-10.0%, 42.2%)**	**Yes**
Levofloxacin	***K. oxytoca***	**16**	**0 (0.0)**	**13**	**3 (18.8)**	**-18.8% (-43.0%, 4.1%)**	**Yes**
Levofloxacin	*K. pneumoniae*	41	1 (2.4)	36	4 (9.8)	-7.3% (-20.3%, 4.4%)	No
Levofloxacin	*P. mirabilis*	41	5 (12.2)	34	2 (4.9)	+7.3% (-5.9%, 21.1%)	No
Levofloxacin	*P. vulgaris*	15	3 (20.0)	10	2 (13.3)	+6.7% (-21.1%, 33.6%)	No
Levofloxacin	*S. marcescens*	21	1 (4.8)	20	0 (0.0)	+4.8% (-11.2%, 22.7%)	No
Levofloxacin	*P. aeruginosa*	25	0 (0.0)	25	0 (0.0)	0.0% (-13.3%, 13.3%)	No
Meropenem	*Acinetobacter* spp.	16	1 (6.3)	14	1 (6.3)	0.0% (-22.7%, 22.7%)	No
Meropenem	*C. freundii*	21	5 (23.8)	11	5 (23.8)	0.0% (-25.0%, 25.0%)	No
Meropenem	*E. cloacae*	8	1 (12.5)	7	0 (0.0)	+12.5% (-21.5%, 47.1%)	No
Meropenem	*E. coli*	76	14 (18.4)	55	7 (9.2)	+9.2% (-2.0%, 20.4%)	No
Meropenem	*K. oxytoca*	16	2 (12.5)	12	2 (12.5)	0.0% (-25.2%, 25.2%)	No
Meropenem	*K. pneumoniae*	19	1 (5.3)	18	0 (0.0)	+5.3% (-12.1%, 24.6%)	No
Meropenem	***P. mirabilis***	**10**	**2 (20.0)**	**8**	**0 (0.0)**	**+20.0% (-11.2%, 51.0%)**	**Yes**
Meropenem	***P. vulgaris***	**16**	**3 (18.8)**	**13**	**0 (0.0)**	**+18.8% (-4.1%, 43.0%)**	**Yes**
Meropenem	***S. marcescens***	**40**	**15 (37.5)**	**25**	**0 (0.0)**	**+37.5%**	**Yes**

	eQUANT System						
Drug	**Organism**	**Total**	**≥3 mm Smaller Zone Size # (%)**	**Equivalent Zone Size (-2 to +2 mm)**	**≥3mm Larger Zone Size # (%)**	**Percent Difference (95% CI)**	**Trending Noted (≥15%)**
						(21.6%, 53.0%)	
Meropenem	*P. aeruginosa*	25	0 (0.0)	25	0 (0.0)	0.0% (-13.3%, 13.3%)	No
Piperacillin-Tazobactam	*Acinetobacter* spp.	41	3 (7.3)	38	0 (0.0)	+7.3% (-2.5%, 19.4%)	No
Piperacillin-Tazobactam	*E. coli*	157	19 (12.5)	137	1 (5.7)	+11.5% (6.4%, 17.5%)	No
Piperacillin-Tazobactam	*K. pneumoniae*	21	2 (9.5)	19	0 (0.0)	+9.5% (-7.4%, 28.9%)	No
Piperacillin-Tazobactam	***P. mirabilis***	**22**	**7 (31.8)**	**15**	**0 (0.0)**	**+31.8% (10.4%, 52.7%)**	**Yes**
Piperacillin-Tazobactam	***P. vulgaris***	**16**	**6 (37.5)**	**10**	**0 (0.0)**	**+37.5% (10.4%, 61.4%)**	**Yes**
Piperacillin-Tazobactam	*S. marcescens*	23	2 (8.7)	21	0 (0.0)	+8.7% (-6.9%, 26.8%)	No
Piperacillin-Tazobactam	*P. aeruginosa*	25	1 (4.0)	24	0 (0.0)	+4.0% (-9.7%, 19.5%)	No
Tobramycin	*C. freundii*	11	1 (9.1)	9	1 (9.1)	0.0% (-29.6%, 29.6%)	No
Tobramycin	*Citrobacter spp.*	2	0 (0.0)	2	0 (0.0)	0.0% (-65.8%, 65.8%)	No
Tobramycin	*E. cloacae*	21	0 (0.0)	21	0 (0.0)	0.0% (-15.5%, 15.5%)	No
Tobramycin	*E. coli*	60	6 (10.0)	52	2 (3.3)	+6.7% (-3.0%, 17.1%)	No
Tobramycin	*K. aerogenes*	22	2 (9.1)	20	0 (0.0)	+9.1% (-7.2%, 27.8%)	No
Tobramycin	*K. oxytoca*	16	0 (0.0)	15	1 (6.3)	-6.3% (-28.3%, 13.8%)	No
Tobramycin	*K. pneumoniae*	21	0 (0.0)	20	1 (4.8)	-4.8% (-22.7%, 11.2%)	No
Tobramycin	*P. mirabilis*	21	1 (4.8)	20	0 (0.0)	+4.8% (-11.2%, 22.7%)	No
Tobramycin	***P. vulgaris***	**16**	**4 (25.0)**	**12**	**0 (0.0)**	**+25.0% (0.6%, 49.5%)**	**Yes**
Tobramycin	*S. marcescens*	13	0 (0.0)	13	0 (0.0)	0.0% (-22.8%, 22.8%)	No
Tobramycin	*P. aeruginosa*	25	1 (4.0)	23	1 (4.0)	0.0% (-15.9%, 15.9%)	No

A trend toward lower zone diameters was observed for nearly all drug/organism combinations when testing using eQUANT compared to zones obtained when testing by standard inoculum. This trend was determined to be statistically significant for several drug/organism combinations. The following limitation is included in the device labeling to address the trending observed for the eQUANT System:

> Disk Diffusion AST zone diameter results generated from an eMcFarland inoculum tend to be smaller when compared to disk diffusion zone diameter results generated from a colony inoculum. If zone diameters fall near the low end

of the intermediate breakpoint, consider alternative testing for the following drug/organism combinations:

- Amoxacillin/Clavulanate: *Proteus vulgaris*
- Aztreonam: *Citrobacter freundii*, *Escherichia coli, Proteus mirabilis, Proteus vulgaris*
- Cefepime: *Escherichia coli, Klebsiella aerogenes, Proteus mirabilis, Proteus vulgaris*
- Ceftriaxone: *Escherichia coli, Klebsiella aerogenes, Proteus mirabilis, Proteus vulgaris*
- Ertapenem: *Escherichia coli, Klebsiella oxytoca, Proteus mirabilis, Proteus vulgaris, Serratia marcescens*
- Gentamicin: *Klebsiella aerogenes, Proteus vulgaris*
- Levofloxacin: *Klebsiella aerogenes, Klebsiella oxytoca*
- Meropenem: *Proteus mirabilis, Proteus vulgaris, Serratia marcescens*
- Piperacillin/Tazobactam: *Proteus mirabilis, Proteus vulgaris*
- Tobramycin: *Proteus vulgaris*

Microbial Suspension Accuracy

In addition to assessment of AST performance by disk diffusion, colony counts were performed from the eQUANT-prepared eMcFarland for 100% of original clinical PBC samples and 10% of supplemental clinical PBC samples. The eMcFarland colony counts from eQUANT samples were considered acceptable if they were within the expected colony count range of 2.51e7 and 7.96e8 CFU/mL (a suspension range based on the turbidity equivalent to 0.5 McFarland which contains approximately 1-2x10^8 CFU/mL for *E. coli* ± 0.6 log difference). The summary of colony count accuracy for eMcFarlands of PBC samples processed with the eQUANT System is shown in **Table 8**.

Table 8. Percentage of eMcFarland Suspensions with Acceptable Microbial Concentration

	eMcFarland Colony Counts # in Expected Range/Total (%)				
	Site #1	**Site #2**	**Site #3**	**Site #4**	**Overall**
Enterobacterales	50/50 (100)	36/36 (100)	25/25 (100)	54/54 (100)	165/165 (100)
***Acinetobacter* spp.**	7/8 (87.5)	8/8 (100)	3/3 (100)	6/6 (100)	24/25 (96.0)
P. aeruginosa	4/4 (100)	12/13 (92.3)	8/8 (100)	6/6 (100)	30/31 (96.8)
Total	61/62 (98.4)	56/57 (98.2)	36/36 (100)	66/66 (100)	219/221 (99.1)

The colony counts of the overall PBC clinical samples was 99.1% in-range and therefore met the defined acceptance criteria of ≥95%. The two samples outside the expected colony count range were one contrived *Acinetobacter* spp. sample and one contrived *P. aeruginosa* sample in the original study. For these two samples, there were no errors observed in Disk Diffusion AST results.

2. Matrix Comparison:

Blood Culture Compatibility

The eQUANT System was tested for compatibility with nine different blood culture bottles. A panel of eight different bacterial isolates (four isolates of *A. baumannii* and one isolate each of *E. coli*, *K. pneumoniae*, *P. vulgaris*, and *P. aeruginosa*) were mixed with human whole blood, seeded directly into a blood culture bottle and incubated for growth in a continuous monitoring blood culture system until indicated as positive for growth (i.e., bottle ring). Performance was evaluated by verifying the colony counts of the resulting eMcFarland and by comparing Disk Diffusion obtained with the eMcFarland to the standard DD method results obtained using a 0.5 McFarland organism concentration prepared using isolated colonies. 100% of all colony counts were within the expected range of $1x10^8$ to $2x10^8 \pm 0.6$ log ($2.51x10^7$ and $7.96x10^8$) CFU/mL. AST results from all indicated antimicrobials were stratified by bottle types and organism group (**Table 9**).

Table 9. Compatibility of the eQUANT System with Different Blood Culture Bottle Types

Org Group	Bottle System	Bottle Type	Total	CA #	CA %	vmj	maj	min
Enterobacterales[a]	BD BACTEC	Aero Plus	96	87	90.6*	0	0	9
		STD AERO	96	88	91.7*	0	0	8
		STD ANA	96	89	91.7*	0	0	7
		LYTIC	96	89	92.7*	0	0	7
		ANA PLUS	96	88	91.7*	0	0	8
	BACT/ALERT	SA	96	89	92.7*	0	0	7
		FA	96	89	92.7*	0	0	7
		FN	96	88	91.7*	0	0	8
		SN	96	93	96.9	0	0	3
P. aeruginosa[b]	BD BACTEC	Aero Plus	21	19	90.5*	0	0	2
		STD AERO	21	20	95.2	0	0	1
	BACT/ALERT	SA	21	21	100	0	0	0
		FA	21	20	95.2	0	0	1
A. baumannii[c]	BD BACTEC	Aero Plus	24	24	100	0	0	0
		STD AERO	24	24	100	0	0	0
	BACT/ALERT	SA	24	24	100	0	0	0
		FA	24	24	100	0	0	0

CA – Category Agreement; min – minor errors; maj – major errors; vmj – very major errors

[a]Disk diffusion results with amoxicillin/clavulanic acid, ampicillin, aztreonam, cefazolin, cefepime, ceftriaxone, ertapenem, gentamicin, levofloxacin, meropenem, piperacillin/tazobactam, and tobramycin
[b]Disk diffusion results with aztreonam, cefepime, gentamicin, levofloxacin, meropenem, piperacillin/tazobactam and tobramycin
[c]Meropenem and piperacillin/tazobactam disk diffusion results
*<95% CA due to minor errors of which the majority were from eQUANT zone diameters that were ≤3mm difference compared to the zone diameters obtained by the standard method

For *A. baumannii*, disk diffusion was performed and expected AST results (>95% CA) were obtained for all bottle types assessed.

For *P. aeruginosa*, disk diffusion was performed and >95% CA was obtained for all bottle types except for BD BACTEC Aero Plus which demonstrated performance of 90.5% CA with two minor errors with levofloxacin. However, the two minor errors zone diameters were less than 3 mm difference when compared to the control replicate AST result which was considered acceptable.

For Enterobacterales, >95% CA was only obtained for the bottle type BacT/ALERT SN; the remaining bottle types demonstrated a CA performance of 90-93%. The minor errors were spread across all bottle types. The eQUANT zone diameter of the majority (60/64=93.8%) of the error results were ≤3 mm difference compared to the standard method zone diameter which was considered acceptable. In addition, 63/64 of the minor errors were due to a single isolate of *K. pneumoniae* with minor errors detected across bottle types and among 6 of the 12 tested drugs (i.e., levofloxacin, piperacillin/tazobactam, cefepime, ceftriaxone, gentamicin, tobramycin). Taken together, the blood culture compatibility performance data were considered acceptable and eQUANT System can be used with these positive blood culture bottle types from BacT/ALERT and BACTEC blood culture systems.

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:

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.