510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY

I Background Information:

A 510(k) Number

K192665

B Applicant

Accelerate Diagnostics, Inc.

C Proprietary and Established Names

Accelerate Pheno System, Accelerate PhenoTest BC Kit

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
PRH, NSU, PEO, PAM, PEN, LON	Class II	21 CFR 866.1650 -	MI - Microbiology

II Submission/Device Overview:

A Purpose for Submission:

To obtain 510(k) clearance for a modified assay for antimicrobial susceptibility testing of with the following antimicrobial agents: ceftazidime, cefepime, meropenem and piperacillin/tazobactam for testing *Pseudomonas aeruginosa*. The submission also provides data in support of the addition of a claim for susceptibility testing of *P. aeruginosa* with aztreonam. In addition, the submission provided results of validation studies performed with an automated quality control method and various improvements made for the organism identification assay.

B Measurand:

Azithromycin	reporting range: 2 – 64 µg/mL
Ceftazidime	reporting range: 2 – 64 µg/mL
Cefepime	reporting range: 2 – 64 µg/mL
Meropenem	reporting range: 1 - 16 µg/mL
Piperacillin/Tazobactam	reporting range: 4 – 256 µg/mL

C Type of Test:

The Accelerate PhenoTest BC is a multiplexed in vitro diagnostic test utilizing both qualitative nucleic acid fluorescence in situ hybridization (FISH) identification and quantitative antimicrobial susceptibility methods and is intended for use with the Accelerate Pheno System. The PhenoTest BC assay is performed directly on positive blood culture samples identified as positive by a continuous monitoring blood culture system.

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

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 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.

The Accelerate PhenoTest BC kit identifies the following Gram-positive and Gram-negative bacteria and yeasts utilizing FISH probes targeting organism-specific ribosomal RNA sequences:

Staphylococcus aureus, Staphylococcus lugdunensis, Coagulase-negative *Staphylococcus* species (i.e., *Staphylococcus epidermidis, Staphylococcus haemolyticus, Staphylococcus hominis, Staphylococcus capitis, Staphylococcus lugdunensis, Staphylococcus warneri,* not differentiated), *Enterococcus faecalis, Enterococcus faecium, Streptococcus* spp. (i.e., *Streptococcus mitis, Streptococcus oralis, Streptococcus gallolyticus, Streptococcus agalactiae, Streptococcus pneumoniae*, not differentiated), *Pseudomonas aeruginosa, Acinetobacter baumannii, Klebsiella* spp. (i.e., *Klebsiella pneumoniae, Klebsiella oxytoca,* not differentiated), *Escherichia coli, Enterobacter* spp. (i.e., *Enterobacter cloacae, Enterobacter (Klebsiella) aerogenes,* not differentiated), *Proteus* spp. (i.e., *Proteus mirabilis, Proteus vulgaris,* not differentiated), *Citrobacter* spp. (i.e., *Citrobacter freundii, Citrobacter koseri,* not differentiated), *Serratia marcescens, Candida albicans* and *Candida glabrata*.

The Accelerate PhenoTest BC kit tests the following antimicrobial agents with the specific target organisms identified below:

Amikacin: *Pseudomonas aeruginosa, Acinetobacter baumannii, Klebsiella* spp. (i.e., *Klebsiella pneumoniae, Klebsiella oxytoca,* not differentiated), *Escherichia coli, Enterobacter* spp. (i.e., *Enterobacter cloacae, Enterobacter (Klebsiella) aerogenes,* not differentiated), *Proteus* spp. (i.e., *Proteus mirabilis, Proteus vulgaris,* not differentiated), *Citrobacter* spp. (i.e., *Citrobacter freundii, Citrobacter koseri,* not differentiated) and *Serratia marcescens*

Ampicillin: *Enterococcus faecalis* and *Enterococcus faecium*

Ampicillin/Sulbactam: *Escherichia coli, Klebsiella* spp. (i.e., *Klebsiella pneumoniae, Klebsiella oxytoca*, not differentiated), and *Proteus* spp. (i.e., *Proteus mirabilis, Proteus vulgaris*, not differentiated)

Aztreonam: *Pseudomonas aeruginosa, Klebsiella* spp. (i.e., *Klebsiella pneumoniae, Klebsiella oxytoca*, not differentiated), *Escherichia coli, Enterobacter* spp. (i.e., *Enterobacter cloacae, Enterobacter (Klebsiella) aerogenes*, not differentiated), *Proteus* spp. (i.e., *Proteus mirabilis, Proteus vulgaris*, not differentiated), *Citrobacter* spp. (i.e., *Citrobacter freundii, Citrobacter koseri*, not differentiated) and *Serratia marcescens*

Ceftazidime: *Pseudomonas aeruginosa, Klebsiella* spp. (i.e., *Klebsiella pneumoniae, Klebsiella oxytoca*, not differentiated), *Escherichia coli, Enterobacter* spp. (i.e., *Enterobacter cloacae, Enterobacter (Klebsiella) aerogenes*, not differentiated), *Proteus* spp. (i.e., *Proteus mirabilis, Proteus vulgaris*, not differentiated), *Citrobacter* spp. (i.e., *Citrobacter freundii, Citrobacter koseri*, not differentiated) and *Serratia marcescens*

Ceftaroline: *Staphylococcus aureus*

Cefepime: *Pseudomonas aeruginosa, Klebsiella* spp. (i.e., *Klebsiella pneumoniae, Klebsiella oxytoca*, not differentiated), *Escherichia coli, Enterobacter* spp. (i.e., *Enterobacter cloacae, Enterobacter (Klebsiella) aerogenes*, not differentiated), *Proteus* spp. (i.e., *Proteus mirabilis, Proteus vulgaris*, not differentiated) ,*Citrobacter* spp. (i.e., *Citrobacter freundii, Citrobacter koseri*, not differentiated) and *Serratia marcescens*

Ceftriaxone: *Klebsiella* spp. (i.e., *Klebsiella pneumoniae, Klebsiella oxytoca,* not differentiated), *Escherichia coli, Enterobacter* spp. (i.e., *Enterobacter cloacae, Enterobacter (Klebsiella) aerogenes*, not differentiated), *Proteus* spp. (i.e., *Proteus mirabilis, Proteus vulgaris*, not differentiated), *Citrobacter* spp. (i.e., *Citrobacter freundii, Citrobacter koseri*, not differentiated) and *Serratia marcescens*

Ciprofloxacin: *Pseudomonas aeruginosa, Klebsiella* spp. (i.e., *Klebsiella pneumoniae, Klebsiella oxytoca*, not differentiated), *Escherichia coli, Enterobacter* spp. (i.e., *Enterobacter cloacae, Enterobacter (Klebsiella) aerogenes*, not differentiated), *Proteus* spp. (i.e., *Proteus mirabilis, Proteus vulgaris*, not differentiated), *Citrobacter* spp. (i.e., *Citrobacter freundii, Citrobacter koseri*, not differentiated) and *Serratia marcescens*

Daptomycin: *Staphylococcus aureus*, Coagulase-negative *Staphylococcus* species (i.e., *Staphylococcus epidermidis, Staphylococcus haemolyticus, Staphylococcus hominis, Staphylococcus capitis, Staphylococcus lugdunensis, Staphylococcus warneri*, not differentiated), *Enterococcus faecalis* and *Enterococcus faecium*

Ertapenem: *Klebsiella* spp. (i.e., *Klebsiella pneumoniae, Klebsiella oxytoca*, not differentiated), *Escherichia coli, Enterobacter* spp. (i.e., *Enterobacter cloacae, Enterobacter (Klebsiella) aerogenes*, not differentiated), *Proteus* spp. (i.e., *Proteus mirabilis, Proteus vulgaris*, not differentiated), *Citrobacter* spp. (i.e., *Citrobacter freundii, Citrobacter koseri*, not differentiated) and *Serratia marcescens*

Gentamicin: *Pseudomonas* aeruginosa, Klebsiella spp. (i.e., Klebsiella pneumoniae, Klebsiella oxytoca, not differentiated), *Escherichia coli, Enterobacter* spp. (i.e., *Enterobacter cloacae, Enterobacter (Klebsiella) aerogenes*, not differentiated), *Proteus* spp. (i.e., *Proteus mirabilis,*

Proteus vulgaris, not differentiated), *Citrobacter* spp. (i.e., *Citrobacter freundii, Citrobacter koseri*, not differentiated) and *Serratia marcescens*

Linezolid: *Staphylococcus aureus, Enterococcus faecalis* and *Enterococcus faecium*

Meropenem: *Pseudomonas aeruginosa, Klebsiella* spp. (i.e., *Klebsiella pneumoniae, Klebsiella oxytoca*, not differentiated), *Escherichia coli, Enterobacter* spp. (i.e., *Enterobacter cloacae, Enterobacter (Klebsiella) aerogenes*, not differentiated), *Proteus* spp. (i.e., *Proteus mirabilis, Proteus vulgaris*, not differentiated), *Citrobacter* spp. (i.e., *Citrobacter freundii, Citrobacter koseri*, not differentiated) and *Serratia marcescens*

Piperacillin/Tazobactam: *Pseudomonas aeruginosa, Acinetobacter baumannii, Klebsiella* spp. (i.e., *Klebsiella pneumoniae, Klebsiella oxytoca*, not differentiated), *Escherichia coli, Enterobacter* spp. (i.e., *Enterobacter cloacae, Enterobacter (Klebsiella) aerogenes*, not differentiated), *Proteus* spp. (i.e., *Proteus mirabilis, Proteus vulgaris*, not differentiated), *Citrobacter* spp. (i.e., *Citrobacter freundii, Citrobacter koseri*, not differentiated) and *Serratia marcescens*

Tobramycin: *Pseudomonas aeruginosa, Klebsiella* spp. (i.e., *Klebsiella pneumoniae, Klebsiella oxytoca*, not differentiated), *Escherichia coli, Enterobacter* spp. (i.e., *Enterobacter cloacae, Enterobacter (Klebsiella) aerogenes*, not differentiated), *Proteus* spp. (i.e., *Proteus mirabilis, Proteus vulgaris*, not differentiated), *Citrobacter* spp. (i.e., *Citrobacter freundii, Citrobacter koseri*, not differentiated) and *Serratia marcescens*

Vancomycin: *Staphylococcus aureus, Staphylococcus lugdunensis*, Coagulase-negative *Staphylococcus* species (i.e., *Staphylococcus epidermidis, Staphylococcus haemolyticus, Staphylococcus hominis, Staphylococcus capitis, Staphylococcus lugdunensis, Staphylococcus warneri*, not differentiated), *Enterococcus faecalis* and *Enterococcus faecium*

The following resistance phenotype is reported based on qualitative tests: Methicillin-resistance (*S. aureus, S. lugdunensis*, coagulase negative staphylococci).

The Accelerate PhenoTest BC kit is indicated as an aid in the diagnosis of bacteremia and fungemia. It is also indicated for susceptibility testing of specific pathogenic bacteria as identified above commonly associated with or causing bacteremia. 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 identification and susceptibility testing of: organisms not identified by the Accelerate PhenoTestBC kit, organisms present in polymicrobial samples, organisms for which species identification is critical for patient care (e.g. speciation of Streptococcus spp.), samples for which an "indeterminate" result for any probe was obtained, for testing antimicrobial agents not included on the Accelerate panel and for epidemiologic testing.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

The following additional limitations were added to the device labeling based on performance demonstrated in the current submission:

Due to the occurrence of very major errors with ceftazidime, isolates of P. aeruginosa that provide an MIC of 8µg/mL should be retested using an alternative/reference method.

A trend towards high MIC values was noted for meropenem with P. aeruginosa, along with a low category agreement mainly due to minor errors. To avoid potential for major errors and if critical to patient care, isolates of P. aeruginosa that provide an MIC of ≥8 µg/mL should be retested using an alternate method.

D Special Instrument Requirements:

Accelerate PhenoTest System

IV Device/System Characteristics:

A Device Description:

The Accelerate Pheno system is comprised of the Accelerate Pheno instrument, software, host computer, analysis computer, and the Accelerate PhenoTest BC kit. The Accelerate PhenoTest BC Kit contains a sample vial, a 48-channel disposable test cassette and a reagent cartridge used to test samples from a blood culture bottle that has been flagged as positive by a continuous monitoring blood culture system. All identification (ID) and antimicrobial susceptibility testing (AST) is performed in individual flowcells of the test cassette. The reagent cartridge contains gel electrofiltration (GEF) stations, fluorescence *in situ* hybridization (FISH) probes, antibiotics, and reagents for automated sample preparation, identification of bacterial and fungal target organisms (Table 1), and antimicrobial susceptibility testing and phenotypic resistance detection testing for bacterial target organisms (Tables 2 and 3). The user loads an aliquot of the positive blood culture into the sample vial, places the test cassette, reagent cartridge and sample vial into an Accelerate Pheno System module, and then presses the module button to close the module door and start the run.

Table 1. Probe Sets and Species Identified by Each Probe Set

Probe Set	On-panel Species
ABA	*Acinetobacter baumannii*
CAL	*Candia albicans*
CGL	*Candida glabrata*
CIT	*Citrobacter freundii* *Citrobacter koseri*
CNS	*Staphylococcus epidermidis* *Staphylococcus haemolyticus* *Staphylococcus hominis* *Staphylococcus capitis* *Staphylococcus lugdunensis* *Staphylococcus warneri*
ECO	*Escherichia coli*
EFM	*Enterococcus faecium*
EFS	*Enterococcus faecalis*
ENT	*Klebsiella aerogenes* *Enterobacter cloacae*
KLE	*Klebsiella oxytoca* *Klebsiella pneumoniae*
PAE	*Pseudomonas aeruginosa*

Probe Set	On-panel Species
PRO	*Proteus mirabilis* *Proteus vulgaris*
SAU	*Staphylococcus aureus*
SLU	*Staphylococcus lugdunensis*
SMA	*Serratia marcescens*
STR	*Streptococcus mitis* *Streptococcus oralis* *Streptococcus gallolyticus* *Streptococcus agalactiae* *Streptococcus pneumoniae*

Table 2. Antimicrobial Agents Tested*, Gram-Positive Organisms

Organism	Ampicillin	Ceftaroline	Daptomycin	Linezolid	Vancomycin	Cefoxitin
S. aureus	-	X	X	X	X	X
S. lugdunensis	-	-	-	-	X	X
Coagulase negative *Staphylococcus*	-	-	X	-	X	X
Enterococcus faecalis	X	-	X	X	X	-
Enterococcus faecium	X	-	X	X	X	-

*(X) = Tested, (-) = Not tested

Table 3. Antimicrobial Agents Tested*, Gram-Negative Organisms

Organism	Ampicillin-sulbactam	Piperacillin-tazobactam	Cefepime	Ceftazidime	Ceftriaxone	Ertapenem	Meropenem	Amikacin	Gentamicin	Tobramycin	Ciprofloxacin	Aztreonam
E. coli	X	X	X	X	X	X	X	X	X	X	X	X
***Klebsiella* spp.**	X	X	X	X	X	X	X	X	X	X	X	X
***Enterobacter* spp.**	-	X	X	X	X	X	X	X	X	X	X	X
***Proteus* spp.**	X	X	X	X	X	X	X	X	X	X	X	X
***Citrobacter* spp.**	-	X	X	X	X	X	X	X	X	X	X	X
S. marcescens	-	X	X	X	X	X	X	X	X	X	X	X
P. aeruginosa	-	X	X	X	-	-	X	X	X	X	X	X
A. baumannii	-	X	-	-	-	-	-	X	-	-	-	-

*(X) = Tested, (-) = Not tested

B Principle of Operation:

The Accelerate Pheno system uses an automated sample preparation and surface immobilization method to enable microscopy-based, single-cell analysis for ID and AST. Identification is accomplished via fluorescence *in situ* hybridization (FISH). Antimicrobial susceptibility testing uses microscopic observation of individual, live, growing bacterial cells in near real time (approximately every 10 minutes) in the presence of antimicrobial agents.

The Accelerate Pheno system employs automated sample and reagent pipetting, temperature controlled incubation, digital microscopy, image acquisition and analysis in an integrated and fully automated system.

V Substantial Equivalence Information:

A Predicate Device Name(s):

Accelerate Pheno system, Accelerate Phenotest BC Kit

B Predicate 510(k) Number(s):

DEN160032

C Comparison with Predicate(s):

Table 4. Comparison with the Predicate Device

Device & Predicate Device(s):	**K192665 Device**	**DEN160032 Predicate**
Device Trade Name	PhenoTest BC Kit	Same
General Device Characteristic Similarities		
Intended Use/Indications For Use	Qualitative identification of microorganisms in positive blood culture and AST testing of a single species present in a positive blood culture	Same
Species identified	*S. aureus*, *S. lugdunensis*, coagulase negative *Staphylococcus* spp. (*S. epidermidis, S. haemolyticus, S. hominis, S. capitus, S. lugdunensis, S. warneri*, not differentiated), *E. faecalis*, *E. faecium*,	Same

Device & Predicate Device(s):	**K192665 Device**	**DEN160032 Predicate**
	Streptococcus spp. (*S. oralis, S. gallolyticus, S. agalactiae, S. pneumoniae*, not differentiated), *P. aeruginosa*, *A. baumannii*, *Klebsiella* spp. (*K. pneumoniae, K. oxytoca*, not differentiated), *E. coli*, *Enterobacter* spp. *(E. cloacae, K. aerogenes*, not differentiated), *Proteus* spp. (*P. mirabilis, P. vulgaris*, not differentiated), *Citrobacter* spp. (*C. freundii, C. koseri*, not differentiated), *S. marcescens*, *C. albicans*, *C. glabrata*	
Sample	Positive blood culture as identified by a continuous monitoring blood culture system	Same
Reagent Cartridge	Accelerate PhenoTest BC Kit	Same
General Device Characteristic Differences		
Instrument	Accelerate Pheno System with addition of a far red filter that distinguishes between bacterial cells and debris for the identification assay	Original filter system
Test Kit	Enhanced wet reagent well to eliminate bubble formation	Original wet reagent well
Antimicrobial Agents	Uses double	Uses a single

Device & Predicate Device(s):	**K192665 Device**	**DEN160032 Predicate**
	concentrations of cefepime, ceftazidime, and piperacillin/tazobactam for *P. aeruginosa* testing only	concentration of cefepime, ceftazidime, and piperacillin/tazobactam for *P. aeruginosa* testing
	Uses double dilution of aztreonam for testing *P. aeruginosa*	Aztreonam not tested with *P. aeruginosa*
	Removal of inducible resistance determination (MLSb) and Erythromycin testing	Included inducible resistance determination (MLSb) and Erythromycin testing
External Quality Control Assay	Automated QC tests only – no user QC organism preparation	Manual QC tests only – user QC organism preparation required
AST Software and algorithms	Modified software and interpretive algorithms for susceptibility testing to determine MICs from double concentrations of antibiotic.	Original software for susceptibility testing with single concentrations of antibiotic. Original interpretive algorithms
	Meropenem interpretive algorithm changes were introduced to improve accuracy of MIC determinations	Original meropenem interpretive algorithm
	Modified expert rules	Original expert rules
Identification software modification	Modification of interpretive rules to decrease ambiguous calls caused by debris or noise objects	Original interpretive rules
	Addition of the AO Bright Rule to decrease the occurrence of invalid identification results	Original interpretive rules
	Improved ID target detection thresholds	Original target detection thresholds
	Noise rejection analysis to decrease false positives	Original analysis

Device & Predicate Device(s):	K192665 Device	DEN160032 Predicate
Time to Result	ID results – approximately 2 hours AST results – Approximately 7 hours	ID results – approximately 1.5 hours AST results – approximately 6.5 hours
Computing System	Add interface PC/Analysis module	Control PC/Analysis PC

VI Standards/Guidance Documents Referenced:

IEC 61010-1:2010, Safety requirements for electrical equipment for measurement, control, and laboratory use- Part 1: General requirements

IEC 60601-1-2:2014, Medical electrical equipment – Part 1-2: General requirements for basic safety and essential performance – Collateral Standard: Electromagnetic disturbances – Requirements and tests.

IEC 60825-1, Safety of laser products - Part 1: Equipment classification, and requirements, Edition 3.0

IEC 62304:2006+A1:2015, Medical device software – Software life cycle processes

ISO 14971:2007, Medical devices – Application of risk management to medical devices

CLSI M100-S29, Performance Standards for Antimicrobial Susceptibility Testing, January 2019

CLSI AUTO11-A2E, Information Technology Security of In Vitro Diagnostic Instruments and Software Systems, Second Edition, October 31, 2014

Guidance for Industry and FDA: Class II Special Controls Guidance Document: Antimicrobial Susceptibility Test (AST) Systems, August 28, 2009

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

A reproducibility study was conducted at the internal site using 15 *P. aeruginosa* isolates. The β-lactam antibiotics (aztreonam, ceftazidime, cefepime and piperacillin/tazobactam) were tested with the modified AST method/algorithm (using two concentrations of the antimicrobial agent in two different flow cells to determine a final single MIC value and interpreted using a modified algorithm). Meropenem was tested using the current AST method (a single concentration of antibiotic) and a modified interpretive algorithm. Isolates were spiked into blood culture bottles containing human blood and incubated until positive

on a continuous blood culture monitoring system. Each isolate was tested on at least five different days using three different Accelerate Pheno Systems for a total of 15 data points for each isolate. Samples were tested within eight hours of positivity. Quality control was performed each day of testing.

Reproducibility was analyzed to determine best case and worst case; best case assumed that results that were off-scale were within ± one doubling dilution of the mode and worst case assumed that results that were off-scale were outside of ± one doubling dilution of the mode. Best case results for aztreonam, ceftazidime, cefepime and meropenem were acceptable with >95% of results falling within ± 1 doubling dilution of the mode (Table 5).

For piperacillin/tazobactam the best case reproducibility was 92.3%, less than the 95% reproducibility recommended in the AST Special Controls document. Review of the line data for the on-scale isolates indicated that for the very few isolates for which reproducibility provided MICs greater than ± 1 doubling dilution from the mode, variability was not due to the Accelerate module or the kit used for testing; variability was due to the highly variable response of some individual isolates to the drug. The sponsor added the following footnote to the reproducibility table in the device labeling:

> *Two P. aeruginosa isolates were found to be highly variable in their response to piperacillin/tazobactam; removal of these isolates provided the following reproducibility results: best case 96.4%, worst case 93.3%*

Table 5. Reproducibility Results for *P. aeruginosa* with Aztreonam, Ceftazidime, Cefepime, Piperacillin/Tazobactam Tested with the Modified Method/Interpretive Algorithm and for Meropenem Tested with a Modified Interpretive Algorithm

Antimicrobial Agent	No. isolates Tested	Total No. Tests	Best Case		Worst Case	
			No. Within ± 1 Dilution/Total	%	No. Within ± 1 Dilution/Total	%
Aztreonam	12	180	171/180	95.0	165/180	91.7
Ceftazidime	10	150	143/150	95.3	134/150	89.3
Cefepime	12	180	175/180	97.2	175/180	97.2
Piperacillin/Tazobactam	13	195	180/195	92.3	175/195	89.7
Meropenem	8	120	115/120	95.8	105/120	87.5

2. Linearity:

   Not applicable

3. Analytical Specificity/Interference:

   Not applicable

4. Assay Reportable Range:

Not applicable

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

CLSI-recommended QC strains were tested for each antimicrobial agent evaluated using the automated QC methodology. The QC strains tested were *P. aeruginosa* ATCC 27853 for aztreonam, cefepime and meropenem, *E. coli* ATCC 25922 for ceftazidime, and *E. coli* ATCC 35218 for piperacillin/tazobactam. For all QC strains, quality control results were within the acceptable range. Results of QC testing are shown in Table 6 below and demonstrate that acceptable QC results can be obtained with this device for > 95% of tests.

Table 6. Quality Control Results for Aztreonam, Ceftazidime, Cefepime, Piperacillin/Tazobactam and Meropenem

Drug	QC Strain/Expected MIC Range	Drug Concentrations	No (%) of results at each concentration
Aztreonam	*P. aeruginosa* ATCC 27853 2-8 µg/mL	2	0
		4	30 (100)
		8	0
Ceftazidime	*E. coli* ATCC 25922 0.06-0.5 µg/mL	0.06	0
		0.12	0
		0.25	30 (100)
		0.5	0
Cefepime	*P. aeruginosa* ATCC 27853 0.5 – 4 µg/mL	0.5	0
		1	0
		2	30 (100)
		4	0
Piperacillin/Tazobactam	*E. coli* ATCC 35218 0.5 -2 µg/mL	0.5	0
		1	30 (100)
		2	0
Meropenem	*P. aeruginosa* ATCC 27853 0.25-1 µg/mL	0.25	0
		0.5	30 (100)
		1	0

The automated QC method utilizes a single dilution of each antimicrobial agent to derive the MIC result. In order to demonstrate that the QC result obtained using the single drug concentration provides adequate quality control of AST tests performed using the double drug concentration used for patient organism testing, a QC equivalency study was performed. Sixty replicates of a single strain of *P. aeruginosa* that had on-scale MIC values for aztreonam, ceftazidime, cefepime and piperacillin/tazobactam were tested. Equivalency was not evaluated for meropenem as only a single drug concentration is used for testing patient isolates. Results showed that >95% of replicates tested using the single drug combination had

mode MIC values within ± one doubling dilution of the replicates tested using the double drug dilution. Results demonstrated that the single drug concentration QC testing methods provides reliable quality control of the drug assays that utilize double drug concentrations to provide a single MIC result.

6. Detection Limit:

   Not Applicable

7. Assay Cut-Off:

   Not applicable

B Comparison Studies:

1. Method Comparison with Predicate Device:

   The purpose of this submission was first to evaluate a modified method for the testing of *P. aeruginosa* isolates with β-lactamase antimicrobial agents (ceftazidime, cefepime and piperacillin/tazobactam). The modified method uses two concentrations of the antimicrobial agent in two different flow cells to determine a final single MIC value instead of the current method which uses a single concentration of the agent in a single flow cell. The modified interpretive algorithm requires information from both flow cells (each with a different antibiotic concentration) in order to determine the final MIC; if information from both flow cells is not available, no MIC result will be reported. These modifications were made to improve device performance and to remove current limitations in the device labeling.

   In this submission the two-concentration testing method was also used to add a new claim for testing *P. aeruginosa* with aztreonam to the intended use of the device. The earlier version of aztreonam testing cleared through the de novo pathway evaluated the performance of aztreonam with members of the Enterobacterales and did not include testing *P. aeruginosa.*

   An additional purpose of the submission was to evaluate a modification of the interpretive algorithm for meropenem designed to improve the minor error rate observed for *P. aeruginosa* MIC results evaluated using the current interpretive algorithm.

   P. aeruginosa challenge isolates utilized in the study were cultured from frozen stocks and used to spike blood culture bottles containing healthy donor blood. Bottles were introduced to the continuously monitored blood culture system and incubated until flagged as positive. Samples from the positive blood cultures were Gram-stained, sub-cultured for purity and run on the Accelerate Pheno System as described in the device labeling. Results were obtained from at least three different Accelerate Pheno system modules at the internal site. AST results were compared to historical results obtained using replicate testing of the broth microdilution reference method.

Aztreonam

For the purpose of adding a new claim for aztreonam testing for *P. aeruginosa* to the intended use of the device, results obtained from a comparative study of *P. aeruginosa* challenge isolates tested with two concentrations of aztreonam were evaluated. Information from the two flow cells were analyzed using an interpretive algorithm modified from the algorithm for aztreonam used for isolates belonging to members of the *Enterobacterales*. A total of 144 *P. aeruginosa* isolates were evaluated using the 2-concentration method/algorithm. Results from the 2-concentration method/algorithm were compared to results obtained by the CLSI reference broth microdilution method to determine performance.

The reporting range for aztreonam on the PhenoTest BC kit is ≤2 to ≥64 µg/mL. Results using the two-concentration method/algorithm showed acceptable performance for *P. aeruginosa* for all performance parameters (EA, EA evaluable, CA and percent errors) when compared to an expanded reporting range for the CLSI reference method (≤0.125 to ≥128 µg/mL) (Table 7). The sponsor also evaluated results using a truncated reference method reporting range (identical to the PhenoTest BC kit reporting range, ≤2 to ≥64 µg/mL) which provided an improved EA over the untruncated reporting range (Table 7); isolates providing very high or very low MICs (≤2 or ≥64 µg/mL) by the reference method which were out of EA using the expanded reporting range were within EA using the truncated range. Performance results for the untruncated and truncated reference method ranges are shown in Table 7 below.

Evaluation of trending of aztreonam with *P. aeruginosa* showed no significant trending (Table 10).

Table 7. Results for Aztreonam with *P. aeruginosa* with the 2-Concentration Method/Algorithm (Untruncated and Truncated Reference Method Reporting Range)

	Tot	EA N	E %	Eval Tot	Eval EA N	Eval EA %	CA Tot	CA %	No. R	No. S	min	maj	vmj
Untruncated	144	131	91.0	87	74	85.1	134	93.1	35	105	9	1	0
Truncated	144	135	93.8	73	64	87.7	134	93.1	35	105	9	1	0

EA – Essential Agreement
Eval – Evaluable Results
CA – Categorical Agreement
R – Resistant isolates
S – Susceptible isolates
min – Minor discrepancies
maj – Major discrepancies
vmj – Very major discrepancies

Essential agreement (EA) occurs when the result of the reference method and that of the PhenoTest BC are within plus or minus one serial two-fold dilution of the antibiotic. Evaluable results are those that are on scale for both the reference method and the PhenoTest BC. Category agreement (CA) occurs when the interpretation of the result of the reference method agrees exactly with the interpretation provided by the PhenoTest BC.

Results obtained were acceptable with the new method/algorithm and support the addition of a claim for testing *P. aeruginosa* with aztreonam with the 2-concentration method/algorithm.

Ceftazidime

For the purpose of removing the current limitation related to the occurrence of major errors with *P. aeruginosa* and ceftazidime, a comparative study using challenge isolates of *P. aeruginosa* isolates was performed. (The limitation applied to DEN160032 indicates that *P. aeruginosa* isolates tested with ceftazidime may show resistant results that are susceptible by the reference method (i.e., major errors). The limitation further states that *P. aeruginosa* isolates that provide an MIC of > 16 µg/mL should be tested with an alternate method.) For the current study, results obtained from a total of 144 *P. aeruginosa* isolates tested with the two-concentration method/revised algorithm were compared to results obtained with the CLSI broth microdilution reference method using an expanded reporting range of ≤0.25 to ≥128 µg/mL; the reporting range for ceftazidime on the PhenoTest BC Kit is ≤2 to ≥64 µg/mL. Three isolates (2.1%) provided no MIC value with the modified method/algorithm due to a lack of information from both antibiotic-containing flow cells.

Results obtained using the two-concentration method/revised algorithm showed acceptable performance for EA (92.2%) and CA (96.5%) as compared to the reference method expanded reporting range. Testing with the two-concentration method/revised algorithm also showed low and acceptable percent of major errors (1.9%) supporting the removal of current limitation related to the occurrence of major errors. However, the percent of very major errors was increased to 7.9% (3 very major errors/38 resistant isolates). Two of the three very major errors were within essential agreement with the reference method; final determination of the major error rate takes into consideration the lack of an intermediate breakpoint for *P. aeruginosa* with ceftazidime and an adjustment is made to the error rate. The adjusted very major error rate for this drug/organism combination was 2.6%, still higher than acceptable. To address the adjusted very major error rate the sponsor added the following footnote to the performance table:

> *The observed very major error rate for ceftazidime when testing P. aeruginosa is 7.9%. Based on the essential agreement and lack of an intermediate breakpoint for ceftazidime, the adjusted very major error rate is 2.6%.*

To address the occurrence of very major errors, the sponsor included the following limitation in the device labeling:

> *Due to the occurrence of very major errors with ceftazidime, isolates of P. aeruginosa that provide an MIC of 8µg/mL should be retested using an alternative/reference method.*

Trending results obtained for ceftazidime with *P. aeruginosa* showed no significant trending toward lower MIC values (Table 10).

The sponsor also evaluated PhenoTest BC Kit results compared to a truncated reference method reporting range of ≤2 to ≥64 µg/mL, which provided an improved EA; isolates providing very low MICs (≤2 , 9 isolates) by the reference method which were out of EA using the expanded reference method reporting range were within EA using the truncated range. Performance results for the truncated range are shown in Table 8 below. The sponsor reported the untruncated and truncated performance in the device labeling.

The modification of the PhenoTest BC AST assay with the addition of a second concentration of ceftazidime in an additional channel has improved the EA, CA and

occurrence of major errors with ceftazidime testing on this device (Table 8). Based on the acceptable results, the sponsor removed the limitation related to major errors for ceftazidime with *P. aeruginosa* from the device labeling, provided an update to the ceftazidime/*P. aeruginosa* performance and included a limitation related to occurrence of very major errors.

Cefepime

For the purpose of removing the current limitation related to the occurrence of major errors with *P. aeruginosa* and cefepime, a comparative study using challenge isolates of *P. aeruginosa* isolates was performed. (The limitation applied to DEN160032 indicates that *P. aeruginosa* isolates tested with cefepime can produce a resistant result for isolates that are susceptible by the reference method (i.e., major error) and that results should be confirmed with an alternate method if critical to patient care.) For the current study, results obtained from a total of 143 *P. aeruginosa* isolates tested with the two-concentration method/revised algorithm were compared to results obtained with the CLSI broth microdilution reference method using an expanded reporting range of ≤0.25 to ≥128 µg/mL; the reporting range for cefepime on the PhenoTest BC kit is ≤2 to ≥64µg/mL. One isolate (0.7%) provided no final MIC value with the modified method/algorithm due to a lack of information from both antibiotic-containing flow cells.

Results obtained using the two-concentration method/revised algorithm showed acceptable performance for CA (92.3%). In addition the number/percent of major errors was higher than considered acceptable (10 major errors/107 susceptible isolates, 9.3%). Eight of the ten major errors were within essential agreement with the reference method; because there is no intermediate breakpoint for cefepime with *P. aeruginosa* the very major rate was adjusted to 1.9% which is acceptable.

A single very major error was observed (1 very major error/36 resistant isolates, 2.8%). The very major error was within essential agreement of the reference method; because there is no intermediate breakpoint for cefepime with *P. aeruginosa*, the very major error rate was adjusted to 0%. To address the actual and adjusted error rates, the following footnote was added to the performance table:

> *The observed major and very major error rates for cefepime when testing P. aeruginosa are 9.3% and 2.8%, respectively. Based on the essential agreement and lack of an intermediate breakpoint for cefepime, the adjusted major error rate is 1.9% and the adjusted very major error rate is 0%.*

An analysis of trending for cefepime with *P. aeruginosa* shows a trend toward higher MIC results, which may result in the occurrence of major errors (see Table 10, Trending, below). The sponsor included a footnote to the performance table indicating the potential for trending (see Trending below).

Using the untruncated reference method reporting range, a total of 24/143 (16.8%) isolates were out of essential agreement giving an EA of 83.2%. To address the low essential agreement the sponsor also evaluated results using a truncated reporting range for the reference method (≤2 to ≥64 µg/mL) which provided an improved EA of 95.1%; isolates providing very low MICs (≤2 µg/mL, 20/24 isolates) or very high MICs (≥64 µg/mL, 2/24 isolates) by the reference method which were out of EA using an expanded reference method

reporting range were within EA using the truncated range. Performance results for the untruncated range are shown in Table 8 below.

The modification of the PhenoTest BC AST assay with the addition of a second dilution of cefepime in an additional channel and evaluated using the truncated reference method reporting range shows acceptable performance for cefepime testing on this device (Table 8). The sponsor reported the untruncated and truncated performance in the device labeling. Based on these results, the sponsor removed the limitation related to major errors for cefepime with *P. aeruginosa* from the device labeling and provided an update to the cefepime/*P. aeruginosa* performance.

Piperacillin/Tazobactam

For the purpose of addressing post-market observations of increased major errors with piperacillin/tazobactam and the removal of a recently added limitation related to the occurrence of major errors with *P. aeruginosa* and piperacillin/tazobactam, a comparative study using challenge isolates of *P. aeruginosa* isolates was performed. (The limitation applied to the labeling of the PhenoTest BC Kit indicates that *P. aeruginosa* isolates can produce a resistant result for isolates that are susceptible to piperacillin/tazobactam by the reference method (i.e., major error) and that results should be confirmed with an alternate method if critical to patient care). For the current study results obtained from a total of 144 isolates tested with the two-concentration method/revised algorithm were compared to results obtained with the CLSI reference broth microdilution method using an expanded reporting range of ≤0.25 to ≥512 µg/mL; the reporting range for piperacillin/tazobactam on the PhenoTest BC Kit is ≤4 to ≥256 µg/mL. Six isolates (4.2%) provided no final MIC value with the modified method/algorithm due to a lack of information from both antibiotic-containing flow cells.

Results obtained using the two-concentration method/revised algorithm showed acceptable performance for EA (93.5%), CA (94.2%) and percent of major (0%) and very major (0%) errors (Table 8). Results obtained support the removal of the current limitation related to the occurrence of major errors.

The sponsor also evaluated PhenoTest BC Kit results compared to a truncated reference reporting range of ≤4 to ≥256 µg/mL which provided an improved EA; isolates providing very low MICs (≤4 µg/mL, 1 isolate) or very high MICs (≥256 µg/mL, 5 isolates) by the reference method which were out of EA using an expanded reporting range were within EA using the truncated range. Performance results for the truncated range are shown in Table 8 below. The sponsor reported the untruncated and truncated performance in the device labeling.

Evaluation of trending of piperacillin/tazobactam with *P. aeruginosa* showed no significant trending (Table 10).

The modification of the PhenoTest BC AST assay with the addition of a second concentration of piperacillin/tazobactam has provided acceptable performance of piperacillin/tazobactam testing on this device (Table 8). Based on the acceptable results, the sponsor removed the limitation related to major errors for piperacillin/tazobactam with *P. aeruginosa* from the device labeling and provided an update to the piperacillin/tazobactam/*P. aeruginosa* performance.

Table 8. Results for Ceftazidime, Cefepime, and Piperacillin/Tazobactam with *P. aeruginosa* with the 2-Concentration Method/Algorithm (Untruncated and Truncated Reference Method Reporting Range)

	Tot	EA N	E %	Eval Tot	Eval EA N	Eval EA %	CA Tot	CA %	No. R	No.S	min	maj	vmj
Ceftazidime													
Untruncated	141	130	92.2	58	47	81.0	136	96.5	38	103	NA[a]	2	3[b]
Truncated	141	136	96.5	30	25	83.3	136	96.5	38	103	NA[a]	2	3[b]
Cefepime													
Untruncated	143	119	83.2	66	42	63.6	132	92.3	36	107	NA[a]	10[c]	1[d]
Truncated	143	136	95.1	40	33	82.5	132	92.3	36	107	NA[a]	10[c]	1[d]
Piperacillin/Tazobactam													
Untruncated	138	129	93.5	52	43	82.7	130	94.2	30	101	8	0	0
Truncated	138	133	96.4	27	22	81.5	130	94.2	30	101	8	0	0

[a] NA, Not Applicable due to lack of an intermediate breakpoint for this drug

[b] The very error rate is 7.9% adjusted to 2.6%. Adjustment made due to a lack of an intermediate breakpoint and 2 of 3 very major results in EA with the reference method.

[c] The major error rate is 9.3% adjusted to 1.9%. Adjustment made due to a lack of an intermediate breakpoint and 8 results in EA with the reference method.

[d] Very major error rate is 2.8% adjusted to 0%. Adjustment made due to a lack of an intermediate breakpoint and the single result in EA with the reference method.

Meropenem

For the purpose of improving performance and reducing the occurrence of minor errors, a comparative study using challenge isolates of *P. aeruginosa* isolates was performed using a single concentration of meropenem and a modified interpretive algorithm. Results obtained from a total of 144 *P. aeruginosa* isolates were compared to results obtained with the CLSI broth microdilution reference method using an expanded reporting range of ≤0.03 to ≥256 µg/mL; the reporting range for meropenem on the PhenoTest BC Kit is ≤1 to ≥16 µg/mL.

Results obtained using the modified interpretive algorithm showed acceptable performance for EA (93.1%) and the occurrence of major (2.0%) and very major (0%) errors (Table 9). The CA was lower than acceptable at 88.2%, mainly due to the occurrence of minor errors. According to the AST Special Controls document, a CA <90.0% caused by a high number of minor errors is acceptable when very good EA of evaluable results are observed. In the current study the EA of evaluable results for meropenem with *P. aeruginosa* was low at 64.3% (untruncated) and 68.0% (truncated). The data shows that the majority of minor errors obtained with the new interpretive algorithm were obtained with isolates that showed MICs of 8 µg/mL and 16 µg/mL (resistant interpretive category). For these isolates, the reference method was 4 µg/mL (intermediate interpretive category). To address the low CA, the sponsor included the following footnote to the performance table:

> *The observed low category agreement was due to the occurrence of a high number of minor errors.*

In addition, the sponsor included the following limitation related to the potential for minor errors to the device labeling:

A trend towards high MIC values was noted for meropenem with P. aeruginosa, along with a low category agreement mainly due to minor errors. To avoid potential for major errors and if critical to patient care, isolates of P. aeruginosa that provide an MIC of ≥8 µg/mL should be retested using an alternate method.

An analysis of trending for meropenem with *P. aeruginosa* shows a trend toward higher MIC results (see Table 10, Trending, below) and is likely responsible for the increased rate of minor errors. The sponsor included a footnote to the performance table indicating the possibility of trending (see "Trending" below).

The sponsor also evaluated results using a truncated reporting range of ≤1 to ≥16 µg/mL which provided an improved EA; isolates providing very low or very high MICs (≤1 or ≥16 µg/mL) by the reference method which were out of EA using an expanded reporting range were within EA using the truncated range. The sponsor reported the untruncated and truncated performance in the device labeling. Performance results for the untruncated and truncated reference method reporting range are shown in Table 9 below.

The algorithm modification of the PhenoTest BC AST assay for meropenem has improved the performance of meropenem testing on this device (Table 9). The sponsor had previously included a limitation in the device labeling related to the occurrence of major errors which did not accurately reflect the type of errors observed. The data provided in this submission supports the removal of the limitation related to major errors for meropenem with *P. aeruginosa* from the device labeling and provided an update to the meropenem/*P. aeruginosa* performance.

Table 9. Results for Meropenem with *P. aeruginosa* with the Revised Interpretive Algorithm (Untruncated and Truncated Reference Method Reporting Range)

	Tot	EA N	EA %	Eval Tot[a]	Eval EA N	Eval EA %	CA Tot	CA %	No. R	No. S	min	maj	vmj
Meropenem													
Untruncated	144	134	93.1	28	18	64.3	127	88.2	25	102	15	2	0
Truncated	144	136	94.4	25	17	68.0	127	88.2	25	102	15	2	0

Reporting Range

For all antimicrobial agents performance was calculated using both the truncated and untruncated reporting range for the reference method. Performance in the device labeling was based on the truncated reference range results but to provide transparency regarding reduced EA for isolates with MICs at very high or very low dilutions, the sponsor added the following footnote to the performance table:

Using an expanded reference range, EA was reduced as follows due to lack of EA for very high or very low MICs: Aztreonam 91.0%, Ceftazidime 92.2%, Cefepime 83.2%, Piperacillin/Tazobactam 93.5%, Meropenem 93.1%

Trending

An analysis of trending was conducted using the challenge data for *P. aeruginosa* with each antimicrobial agent. This trending calculation takes into account MIC values that are determined to be one or more doubling dilution lower or higher compared to the reference method irrespective of whether the device MIC values are on scale or not. Results that are not clearly at least one dilution lower, at least one dilution higher or in exact agreement with the CLSI reference method are not considered in the trending analysis.

For *P. aeruginosa* with the various antimicrobial agents for which the difference between the percentage of isolates with higher vs. lower readings was >30% and for which the confidence interval was determined to be statistically significant were considered to show evidence of trending. Trending that provides higher or lower MIC values compared to the reference is addressed in labeling. Trending calculations for *P. aeruginosa* with aztreonam, ceftazidime, cefepime, meropenem, and piperacillin/tazobactam are shown in Table 10 below.

Table 10. Observed Trending for Aztreonam, Ceftazidime, Cefepime, Piperacillin/Tazobactam and Meropenem with *P. aeruginosa*

Organism	Total Evaluable for Trending	≥ 1 Dilution lower No. (%)	Exact No. (%)	≥ 1 Dilution Higher No. (%)	Percent Difference (CI)	Trending Noted
Aztreonam	122	57 (46.7)	31 (25.4)	34 (27.9)	-18.9 (-30.2 to -6.7)	No
Ceftazidime	73	21 (28.8)	17 (23.3)	35 (48.0)	19.2 (3.4 to 33.7)	No
Cefepime	83	20 (24.1)	11 (13.3)	52 (62.7)	38.6 (23.7 to 51.0)	Yes
Piperacillin/Tazobactam	76	41 (54.0)	12 (15.8)	23 (30.3)	-23.7 (-37.8 to -8.0)	No
Meropenem	34	1 (2.9)	8 (23.5%)	25 (73.5%)	70.6% (50.1 to 82.7)	Yes

To address the observed trending the sponsor included the following footnote to the performance table in the device labeling:

> *When not in exact agreement, cefepime and meropenem MIC results for P. aeruginosa tended to be at least one doubling dilution higher than the reference method which increases the potential for major errors.*

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:

Table 11. FDA-Recognized Interpretive Criteria for *P. aeruginosa*

Antimicrobial Agent	Interpretive Criteria for *P. aeruginosa* (µg/mL)[a]		
	Susceptible	Intermediate	Resistant
Aztreonam	**≤ 8**	**16**	**≥ 32**
Ceftazidime	**≤ 8**	**-**	**≥ 16**
Cefepime	**≤ 8**	**-**	**≥ 16**
Meropenem	**≤ 2**	**4**	**≥ 8**
Piperacillin/Tazobactam	**≤ 16/4**	**32/4 - 64/4**	**≥ 128/4**

[a] FDA STIC Webpage
https://www.fda.gov/Drugs/DevelopmentApprovalProcess/DevelopmentResources/ucm410971.htm

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 referred to the breakpoint change protocol that was reviewed and accepted by FDA in K192665. This protocol 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) for aztreonam, ceftazidime, cefepime, piperacillin/tazobactam and meropenem. The protocol outlined the specific procedures and acceptance criteria that Accelerate Diagnostics intends to use to evaluate the PhenoTest BC kit when revised breakpoints published on the FDA STIC webpage for the previously stated antimicrobial agents. The breakpoint change protocol indicated that if specific criteria are met, Accelerate Diagnostics will update the PhenoTest BC kit device 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.