510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY

I Background Information:

A 510(k) Number

K214023

B Applicant

bioMérieux, Inc

C Proprietary and Established Names

VITEK 2 AST-Gram Negative Ciprofloxacin (≤0.06 - ≥4 µg/mL)

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
LON	Class II	21 CFR 866.1645 - Fully Automated Short-Term Incubation Cycle Antimicrobial Susceptibility System	MI - Microbiology
LTT	Class II	21 CFR 866.1640 - Antimicrobial susceptibility test powder	MI - Microbiology
LTW	Class II	21 CFR 866.1640 - Antimicrobial susceptibility test powder	MI - Microbiology

II Submission/Device Overview:

A Purpose for Submission:

To update the VITEK 2 AST Gram Negative Ciprofloxacin device labeling to include updated FDA-recognized breakpoints for *Enterobacterales* and *Pseudomonas aeruginosa*, as published in the FDA STIC website.

Breakpoints for *Salmonella* spp. (also indicated for use with this device) remain unchanged.

Previously obtained QC and reproducibility data is applicable to this reevaluation.

B Measurand:

Ciprofloxacin (≤0.06 – ≥4 µg/ml)

C Type of Test:

Automated quantitative or qualitative antimicrobial susceptibility test

III Intended Use/Indications for Use:

A Intended Use(s):
The VITEK 2 Gram-Negative Susceptibility Card is intended for use with the VITEK 2 Systems in clinical laboratories as an *in vitro* test to determine the susceptibility of clinically significant aerobic Gram-negative bacilli to antimicrobial agents when used as instructed.

B Indication(s) for Use:
VITEK 2 AST-Gram Negative Ciprofloxacin is designed for antimicrobial susceptibility testing of Gram negative bacilli and is intended for use with the VITEK 2 and VITEK 2 Compact Systems as a laboratory aid in the determination of *in vitro* susceptibility to antimicrobial agents. VITEK 2 AST-Gram Negative Ciprofloxacin is a quantitative test.

Ciprofloxacin has been shown to be active against most strains of the microorganisms listed below, according to the FDA label for this antimicrobial.

Active both *in vitro* and in clinical infections:
Citrobacter koseri
Citrobacter freundii
Enterobacter cloacae
Escherichia coli
Klebsiella pneumoniae
Morganella morganii
Proteus mirabilis
Proteus vulgaris
Providencia rettgeri
Providencia stuartii
Pseudomonas aeruginosa
Salmonella typhi
Serratia marcescens
Shigella sonnei

In vitro data available but clinical significance is unknown:
Klebsiella aerogenes
Klebsiella oxytoca
Salmonella enteritidis

The VITEK 2 Gram-Negative Susceptibility Card is intended for use with the VITEK 2 Systems in clinical laboratories as an *in vitro* test to determine the susceptibility of clinically significant aerobic Gram-negative bacilli to antimicrobial agents when used as instructed.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

Due to the occurrence of minor errors (resulting in a category agreement below 90% for the following species), perform an alternative method of testing prior to reporting of results for the following antibiotic/organism combination:
Ciprofloxacin (cip02): MICs of 0.25µg/mL or 0.5 µg/mL for *P. rettgeri*

Due to the occurrence of minor errors (resulting in a categorical agreement below 90% for the manual dilution option), perform an alternative method of testing prior to reporting of results generated with the manual dilution option for the following antibiotic/organism combinations:
Ciprofloxacin (cip02): MIC of 0.5 µg/mL for *S. marcescens, K. pneumoniae*

The ability of the AST card to detect resistance with the following combination(s) is unknown because resitant strains were not available at the time of comparative testing:
Ciprofloxacin: *Salmonella enteridis, Salmonella typhi* and *Shigella sonnei*

D Special Instrument Requirements:

VITEK 2 and VITEK 2 Compact Systems using VITEK 2 Systems 8.01 software (or later)

IV Device/System Characteristics:

A Device Description:

The VITEK 2 AST card is a miniaturized, abbreviated and automated version of the doubling dilution technique for determining the minimum inhibitory concentration (MIC). Each VITEK 2 AST card contains 64 wells. A control well(s) which contain only nutrient medium is resident on all cards. The remaining wells contain premeasured portions of antimicrobials combined with the nutrient media. The isolate to be tested is diluted to a standardized concentration with 0.45% to 0.50% saline before being used to rehydrate the antimicrobial medium within the card. The VITEK 2 System will automatically (or allow operator to manually) dilute the bacterial suspension to prepare an inoculum for susceptibility cards. Then, the VITEK 2 will fill, seal and place the card into the incubator/reader. The VITEK 2 Compact has a manual filling, sealing, and loading operation. The VITEK 2 Systems monitor the growth of each well in the card over a defined period of time. The analysis program determines when a well demonstrates growth based on attenuation of light measured by an optical scanner. This data is used to determine the minimum inhibitory concentration or "MIC" values for the antimicrobial agent. At the completion of the incubation cycle, a report is generated that contains the MIC value along with the interpretive category result for each antimicrobial contained on the card.

VITEK 2 AST-Gram Negative Ciprofloxacin has the following concentrations in the card: 0.06, 0.12, 0.5 and 1 µg/mL (equivalent standard method concentration by efficacy in µg/mL). The ciprofloxacin MIC result range for the VITEK 2 is ≤0.06 to ≥4 µg/mL. For all species, the VITEK 2 system is capable of reporting the following MIC results: ≤0.06, 0.12, 0.25, 0.5, 1, 2, and ≥4 µg/mL for the AST-Gram Negative Ciprofloxacin test.

B Principle of Operation:

The VITEK 2 and VITEK 2 Compact Systems utilize automated growth-based detection using attenuation of light measured by an optical scanner. The optics in the systems use visible light to directly measure organism growth within each of the 64 micro-wells. Transmittance optics is based on an initial light reading of a well before significant growth has begun. Every 15 minutes throughout the incubation cycle (defined period of time based on the VITEK 2 card), light transmittance readings of each well determine organism growth by the amount of light that is prevented from passing through the well. At the completion of the incubation period, the MIC values and their associated interpretive category results for each antimicrobial on the test card are displayed in an automatically generated report.

V Substantial Equivalence Information:

A Predicate Device Name(s):
VITEK 2 AST-Gram Negative Eravacycline (≤0.12 - ≥4µg/mL)

B Predicate 510(k) Number(s):
K191766

C Comparison with Predicate(s):

	Device: **K214023**	**Predicate:** **K191766**
Device Trade Name	VITEK 2 AST-Gram Negative Ciprofloxacin (≤0.06 - ≥4 µg/mL)	VITEK 2 AST-Gram Negative Eravacycline (≤0.12 - ≥4µg/mL)
General Device Characteristic Similarities		
Intended Use	The VITEK 2 Gram-Negative Susceptibility Card is intended for use with the VITEK 2 Systems in clinical laboratories as an *in vitro* test to determine the susceptibility of clinically significant aerobic Gram-negative bacilli to antimicrobial agents when used as instructed.	Same
Test Method	Automated quantitative antimicrobial susceptibility test for use with the VITEK 2 and VITEK 2 Compact Systems to determine the *in vitro* susceptibility of Gram-negative bacilli	Same
Inoculum	Standardized saline suspension of test organism	Same
Test Card	VITEK 2 Gram Negative Susceptibility Test Card	Same
Instrument	VITEK 2 and VITEK 2 Compact Systems	Same

	Device: K214023	Predicate: K191766
Analysis Algorithm	Growth pattern analysis	Same
General Device Characteristic Differences		
Antimicrobial Agent	Ciprofloxacin	Eravacycline
Antimicrobial Concentrations	0.06, 0.12, 0.5 and 1 µg/mL	0.25, 1, 2 and 4 µg/mL
Reporting Range	≤0.06 to ≥4 µg/mL	≤0.12 to ≥4 µg/mL
Indicated Organisms	Active *in vitro* and in clinical infections: *Citrobacter koseri, Citrobacter freundii, Enterobacter cloacae* *Escherichia coli, Klebsiella pneumoniae, Morganella morganii, Proteus mirabilis* *Proteus vulgaris, Providencia rettgeri, Providencia stuartii, Pseudomonas aeruginosa, Salmonella typhi, Serratia marcescens, Shigella sonnei* *In vitro* data available but clinical significance is unknown: *Klebsiella aerogenes, Klebsiella oxytoca, Salmonella enteritidis*	Active *in vitro* and in clinical infections: *Citrobacter freundii, Enterobacter cloacae, Escherichia coli, Klebsiella oxytoca, Klebsiella pneumoniae* *In vitro* data available but clinical significance is unknown: *Citrobacter koseri, Klebsiella (Enterobacter) aerogenes*

VI Standards/Guidance Documents Referenced:

Data that was reanalyzed to support this submission was obtained from a previously completed clinical study. The clinical study was reviewed and cleared in K162737. The CLSI Standards and FDA guidance document listed below were effective during the original clearance:

- CLSI M100-S24: Performance Standards for Antimicrobial Susceptibility Testing; Twenty-fourth Informational Supplement, Vol. 34 No. 1 (January 2014)
- CLSI M07-A9: Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically; Approved Standard-Ninth Edition" Vol. 32 No, 2 (January 2012)
- Class II Special Controls Guidance Document: Antimicrobial Susceptibility Test (AST) Systems; Guidance for Industry and FDA (Issued August 28, 2009)

Additional testing (described below) was performed and included in support of this submission. The CLSI Standards and FDA guidance document listed below were effective during the additional testing:

- CLSI M100-S29: Performance Standards for Antimicrobial Susceptibility Testing; Twenty-ninth Informational Supplement, Vol. 39 No. 1 (January 2019)

- CLSI M07-A10: Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically; Approved Standard-Tenth Edition" Vol. 35 No, 2 (January 2015)
- Class II Special Controls Guidance Document: Antimicrobial Susceptibility Test (AST) Systems; Guidance for Industry and FDA (Issued August 28, 2009)

A revised edition of the CLSI M100 document became available after receipt of this submission by FDA. As noted below, the revised version updated the recommended quality control reporting range for *P. aeruginosa* ATCC 27853. The CLSI Standard listed below was effective during the clearance of this submission:

- CLSI M100-S31: Performance Standards for Antimicrobial Susceptibility Testing; Thirty-first Informational Supplement, Vol. 41 No. 3 (March 2021)

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:
   Reproducibility of the VITEK 2 AST-GN Ciprofloxacin was previously evaluated during review of K162737 and was determined to be acceptable.

   In summary, a 10-isolate panel composed of species consistent with the intended use were tested at three sites using both inoculation methods (manual and automatic) for testing with the VITEK 2 and the manual inoculation method for testing with the VITEK 2 Compact.

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):
   Quality Control (QC) testing, Inoculum and Growth failure for the VITEK 2 GN-AST Ciprofloxacin were previously evaluated during review of K162737 and were determined to be acceptable. QC specific to the change in breakpoints was not performed since the QC ranges used in K162737 were either unchanged (*E. coli* ATCC 25922) or expanded (*P. aeruginosa* ATCC 27853; range expanded from 0.25 – 1 µg/mL to 0.12 – 1 µg/mL) and therefore would not negatively impact QC performance. QC results with unchanged and expanded QC ranges are shown in **Table 1**.

   In summary, QC organisms recommended by both the FDA and CLSI, namely *E. coli* ATCC 25922 and *P. aeruginosa* ATCC 27853, were tested using the reference broth microdilution (BMD) method and the VITEK 2 GN-AST Ciprofloxacin during the clinical study. The overall QC results for both the reference and VITEK 2 were acceptable. Both the BMD dilution range (≤0.008 - ≥8 µg/mL) and the VITEK 2 Ciprofloxacin card (≤0.06 - ≥4 µg/mL) covered the full expected result range of *P. aeruginosa* ATCC 27853 (expected range of

0.12-1 µg/mL); however, resulted in off-scale MIC values for *E. coli* ATCC 25922 (expected range of 0.004-0.016 µg/mL). An *E. coli* ATCC 25922 MIC value of ≤0.008 µg/mL (with BMD) and ≤0.06 µg/mL (with VITEK 2) indicated that the quality control test results were acceptable. To address the expected off-scale QC results when testing *E. coli* ATCC 25922, the device labeling reviewed in the current submission includes the following footnote under the Quality Control Table:
"*Does not include the full CLSI/FDA recommended dilution range for QC testing with this organism*".

Table 1. Quality Control Result Frequencies for VITEK 2 (Auto-Dilution and Manual Dilution Methods) and VITEK 2 Compact (Manual Dilution Method)

Organism	VITEK 2 Result Range[1] (µg/mL)	BMD[2] Result Range (µg/mL)	VITEK 2 Auto-Dilution	BMD	VITEK 2 Manual Dilution	BMD	VITEK 2 Compact Manual Dilution	BMD
E. coli (ATCC 25922) Expected Range: 0.004-0.016 µg/mL		≤0.008		200		102		102
		0.016		21		7		7
		0.03						
	≤0.06	0.06	222		109		109	
	0.125	0.12						
	0.25	0.25						
	0.5	0.5						
	1	1						
	2	2						
	≥4	4						
		≥8						
P. aeruginosa (ATCC 27853) Expected Range: 0.12-1 µg/mL		≤0.008						
		0.016						
		0.03						
	≤0.06	0.06	1					
	0.12	0.12	1	1		1		1
	0.25	0.25	18	98	9	52	13	52
	0.5	0.5	200	119	98	52	93	52
	1	1		2		2	1	2
	2	2						
	≥4	4						
		≥8						

[1]VITEK 2 Card range for Ciprofloxacin is ≤0.06 - ≥4 µg/mL and does not include the full CLSI/FDA-recommended dilution range for QC testing of *E. coli* ATCC 25922. The lowest concentration of the VITEK 2 Ciprofloxacin MIC range is 0.06 µg/mL. Obtaining a value of ≤0.06 µg/mL was considered an indication that the quality control test results were acceptable. Similarly, the lowest concentration of the BMD Ciprofloxacin MIC range is 0.008 µg/mL. Obtaining a value of ≤0.008 µg/mL was considered an indication that the quality control test results were acceptable.
[2]BMD: CLSI reference broth microdilution.

Since the full QC range was covered for *P. aeruginosa* ATCC 27853, the QC performance data is acceptable for the purpose of re-evaluating *Enterobacterales* and *P. aeruginosa* performance when the updated breakpoints are applied.

6. Detection Limit:
Not applicable

7. Assay Cut-Off:
Not applicable

B Comparison Studies:

1. Method Comparison with Reference Method:
Performance after reanalysis and additional testing
The VITEK 2 AST-GN Ciprofloxacin (≤0.06 - ≥4 µg/mL) was originally cleared in premarket submission K162737. Since there were no changes to the design or dilution range of the VITEK 2 AST card, performance evaluation was achieved via reanalysis of the MIC data provided in the original 510(k) submission (K162737) using revised interpretive criteria currently recognized by FDA for ciprofloxacin against *Enterobacterales* and *P. aeruginosa*. An additional 88 isolates were added to the original 890 *Enterobacteriaceae* (*Enterobacterales*) data set for evaluation of the VITEK 2/automatic dilution method to address a lack of resistant isolates and potential categorical errors as a consequence of the revised interpretive criteria (43 *Serratia marcescens*, 6 *Enterobacter cloacae*, 2 *Enterobacter cloacae cloacae*, 22 *Enterobacter cloacae* complex, and 15 *Providencia rettgeri* isolates). In addition, one *Klebsiella pneumonia pneumoniae* isolate was removed from the reanalysis due to a prior misidentification; this isolate has now been identified as *Acinetobacter baumannii* complex, a species for which FDA does not have recognized breakpoints, resulting in a total of 977 (908 clinical and 69 challenge) *Enterobacterales* isolates for analysis using revised interpretive criteria for *Enterobacterales*. No additional testing was performed for *P. aeruginosa* and the original 236 (146 clinical and 90 challenge) isolate data set was reanalyzed using the revised interpretive criteria for *P. aeruginosa.*

The VITEK 2 with automatic dilution method results are shown in **Table 2** for the reanalysis of data based on the updated interpretive criteria for *Enterobacterales* and *P. aeruginosa*. For *Enterobacterales*, this included the original clinical and challenge data combined with the data after additional testing. For *P. aeruginosa*, this was based only on the original clinical and challenge data combined since no additional testing was performed.

The VITEK 2 and VITEK 2 Compact systems with manual dilution method results of the reanalyzed challenge data with updated *Enterobacterales* and *P. aeruginosa* interpretive criteria are summarized in **Table 3**. Original data obtained with *Salmonella* spp. was not reanalyzed since the interpretive criteria remain unchanged since the original submission.

Table 2. Reanalysis of Performance with the Original and Expanded *Enterobacterales* Isolates and Original *P. aeruginosa* Isolates with the VITEK 2 (Auto Dilution)[a]

	Tot	No.EA	EA %	Eval EA Tot	No. Eval EA	Eval EA %	No. CA	CA %	No. R	No. S	min	maj	vmj
***Enterobacterales*[b], ≤0.25 (S), 0.5 (I), ≥1 (R)**													
Clinical	908	888	97.8	110	90	81.8	874	96.3	216	681	32	1	1
Challenge	69	69	100	37	37	100	56	81.2	25	33	13	0	0
Combined	977	957	97.9	147	127	86.4	930	95.2	241	714	45	1	1

	Tot	No.EA	EA %	Eval EA Tot	No. Eval EA	Eval EA %	No. CA	CA %	No. R	No. S	min	maj	vmj
***Pseudomonas aeruginosa*, ≤0.5 (S), 1 (I), ≥2 (R)**													
Clinical	146	132	90.4	95	81	85.3	140	95.9	32	107	6	0	0
Challenge	90	89	98.9	24	23	95.8	88	97.8	67	22	2	0	0
Combined	236	221	93.6	119	104	87.4	228	96.6	99	129	8	0	0
Organism Group: *Salmonella*[a], ≤0.0625 (S), 0.12-0.5 (I), ≥1 (R)													
Clinical	11	11	100	4	4	100	11	100	3	6	0	0	0
Challenge	10	10	100	2	2	100	10	100	0	8	0	0	0
Combined	21	21	100	6	6	100	21	100	3	14	0	0	0

[a]Performance with *Salmonella* spp. was not reanalyzed since breakpoints remain unchanged.
[b]*Enterobacterales* isolates evaluated include: 54 *C. freundii*, 45 *C. koseri*, 47 *E. aerogenes*, 38 *E. cloacae,* 43 *E. cloacae* complex, 26 *E. cloacae* ssp. *cloacae*, 152 *E. coli*, 51 *K. oxytoca*, 53 *K. pneumoniae*, 112 *K. pneumoniae* ssp. *pneumoniae*, 12 *M. morganii*, 18 *M. morganii* ssp. *morganii*, 59 *P. mirabilis*, 29 *P. vulgaris*, 57 *P. rettgeri*, 26 *P. stuartii*, 134 *S. marcescens*, 21 *Shigella* ssp.

EA – Essential Agreement
CA – Category Agreement
EVAL – Evaluable isolates
R – Resistant isolates
min – minor errors
maj – major errors
vmj – very major errors

Essential Agreement (EA) occurs when there is agreement between the MIC result of the reference method and that of the VITEK 2 test card within plus or minus one serial two-fold dilution of the antibiotic. Evaluable results are those that are on scale for both the VITEK 2 test card and the reference method or those in which an off scale result is at least two doubling dilutions from the on scale result. Category Agreement (CA) occurs when the interpretation of the result of the reference method agrees exactly with the interpretation of the VITEK 2 test card.

The combined Essential Agreement (EA) for *Enterobacterales* is >90% which is acceptable, as described in the "Class II Special Control Guidance Document: Antimicrobial Susceptibility Test (AST) Systems; Guidance for Industry and FDA, August 2009" (AST guidance).

The updated breakpoints for *Enterobacterales* lowered the susceptible, intermediate and resistant breakpoints by two two-fold dilutions. Applying the updated breakpoints for *Enterobacterales* reduced the combined Category Agreement (CA) from 97.8% in K162737 to 95.2% in the current submission, due to an increase in minor errors from 18 (2.0%) to 45 (4.6%) and very major errors from 0 (0%) to 1 (0.4%). These results are acceptable.

The combined EA for *P. aeruginosa* is >90% which is acceptable, as described in the AST guidance. The updated breakpoints for *P. aeruginosa* lowered the susceptible, intermediate and resistant breakpoints by one two-fold dilution. Applying the updated breakpoints for *P. aeruginosa* increased the combined CA from 94.1% in K162737 to 96.6% in the current submission, due to a decrease in minor errors from 12 (5.1%) to 8 (3.4%), major errors from 1 (0.7%) to 0 (0%), and very major errors from 1 (1.1%) to 0 (0%).

Table 3. Reanalysis of Performance with the Original *Enterobacterales* and *P. aeruginosa* Challenge Isolates with the VITEK 2 and VITEK 2 Compact Systems (Manual Dilution)[a]

	Tot	No.EA	EA %	Eval EA Tot	No. Eval EA	Eval EA %	No. CA	CA %	No. R	No. S	min	maj	vmj
***Enterobacterales*, ≤0.25 (S), 0.5 (I), ≥1 (R)**													
VITEK 2	69	69	100	38	38	100	57	82.61	25	33	12	0	0
VITEK 2 Compact	69	67	97.1	37	35	94.59	54	78.26	25	33	15	0	0
***Pseudomonas aeruginosa*, ≤0.5 (S), 1 (I), ≥2 (R)**													
VITEK 2	90	90	100	23	23	100	87	96.67	67	22	3	0	0
VITEK 2 Compact	90	89	98.89	24	23	95.83	87	96.67	67	22	3	0	0
***Salmonella* [a], ≤0.0625 (S), 0.12-0.5 (I), ≥1 (R)**													
VITEK 2	10	10	100	2	2	100	10	100	0	8	0	0	0
VITEK 2 Compact	10	10	100	2	2	100	10	100	0	8	0	0	0

[a]Performance with *Salmonella* spp. was not reanalyzed since breakpoints remain unchanged.

EA – Essential Agreement
CA – Category Agreement
EVAL – Evaluable isolates
R – Resistant isolates
min – minor errors
maj – major errors
vmj – very major errors

Essential Agreement (EA) occurs when there is agreement between the MIC result of the reference method and that of the VITEK 2 test card within plus or minus one serial two-fold dilution of the antibiotic. Evaluable results are those that are on scale for both the VITEK 2 test card and the reference method or those in which an off scale result is at least two doubling dilutions from the on scale result. Category Agreement (CA) occurs when the interpretation of the result of the reference method agrees exactly with the interpretation of the VITEK 2 test card.

The EA of *Enterobacterales* challenge isolates is >90% with the manual dilution option of the VITEK 2 and VITEK 2 Compact instruments, which is acceptable. However, applying the updated breakpoints for *Enterobacterales* resulted in low CA for both VITEK 2 and VITEK 2 Compact, which was caused by the large number of minor errors. Since the EA of evaluable results is >90% and the majority of the categorical errors were minor errors, the following limitations are added to the package insert to address the low CA with the manual dilution option and the number of minor errors generated with the automatic and manual dilution options:

Due to the occurrence of minor errors (resulting in a category agreement below 90% for the following species), perform an alternative method of testing prior to reporting of results for the following antibiotic/organism combination:
Ciprofloxacin (cip02): MICs of 0.25µg/mL or 0.5 µg/mL for P. rettgeri

Due to the occurrence of minor errors (resulting in a categorical agreement below 90% for the manual dilution option), perform an alternative method of testing prior to reporting of results generated with the manual dilution option for the following antibiotic/organism combinations:
Ciprofloxacin (cip02): MIC of 0.5 µg/mL for S. marcescens, K. pneumoniae

The EA of *P. aeruginosa* challenge isolates is >90% with the manual dilution option of the VITEK 2 and VITEK 2 Compact instruments, which is acceptable. Applying the updated breakpoints for *P. aeruginosa* resulted in >90% CA, three minor errors (3.3%), and zero major or very major errors, which is acceptable.

As required under 511A(b)(2)(C)(ii)(I) of the Federal Food, Drug and Cosmetic Act, the following statement is included in the device labeling to address testing of non-indicated species:

> *Per the FDA-Recognized Susceptibility Test Interpretive Criteria website, the safety and efficacy of antimicrobial drugs, for which antimicrobial susceptibility is tested by this AST device, may or may not have been established in adequate and well-controlled clinical trials for treating clinical infections due to microorganisms outside of those found in the indications and usage in the drug label. The clinical significance of susceptibility information in those instances is unknown. The approved labeling for specific antimicrobial drugs provides the uses for which the antimicrobial drug is approved.*

Resistant Organisms

A total of 129 (14.5%) *Enterobacteriaceae*, 3 (14.2%) *Salmonella* spp., and 90 (38.1%) *P. aeruginosa* isolates) were originally found to be resistant to ciprofloxacin by the reference method (K162737). After applying the updated breakpoints for *Enterobacterales* and *P. aeruginosa* and including data from the additional testing with *Enterobacterales*, a total of 241 (24.7%) *Enterobacterales* isolates and 99 (41.9%) of *P. aeruginosa* isolates were determined to be resistant to ciprofloxacin by the reference method. As an impact from the change in breakpoints and the testing of additional challenge *Enterobacterales* isolates the resistance rates have slightly increased compared to the original clearance providing more datapoints for assessing CA and error rates. Based on the time of initial testing, the resistant strains evaluated in the original study likely represent the current resistance population.

Additional testing with 28 *Enterobacter cloacae* spp. and 20 *Providencia rettgeri* resistant isolates resulted in the removal of these species from the original limitation statement regarding a lack of testing with resistant organisms. As such, the limitation statement has been updated as follows:

> *The ability of the AST card to detect resistance with the following combination(s) is unknown because resistant strains were not available at the time of comparative testing: Ciprofloxacin: Salmonella enteritidis, Salmonella typhi and Shigella sonnei*

MIC Trending Analysis

A trending reanalysis was performed using the combined (clinical and challenge) *Enterobacterales* and *P. aeruginosa* data obtained in K162737 to align with current trending analysis methods utilized and to also assess the data from additional testing with *Enterobacterales* isolates with the VITEK 2 auto-dilution method.

This trending calculation takes into account MIC values that are determined to be one or more doubling dilutions lower or higher than 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.

Organisms in which the difference between the percentage of isolates with higher vs. lower MIC values was ≥ 30% and for which the confidence interval was determined to be statistically significant were considered to show evidence of trending. Trending that showed higher or lower MIC values compared to the reference is addressed in the labeling.

Table 4. Trending analysis of the VITEK 2 Gram Negative Ciprofloxacin

Organism Group	Total Evaluable for Trending	>= 1 Dilution Lower No. (%)	Exact No. (%)	>= 1 Dilution Higher No. (%)	Percent Difference (95% CI)	Trending Noted
Enterobacterales	**198**	**52 (26.26)**	**55 (27.78)**	**91 (45.96)**	**19.7% (10.26 to 28.65)**	**No**
Citrobacter freundii	16	3 (18.75)	8 (50.00)	5 (31.25)	12.5% (-17.17 to 39.71)	No
Citrobacter koseri	0	0	0	00	0	NA
Enterobacter aerogenes	3	1 (33.33)	0	2 (66.67)	33.33% (-31.58 to 71.78)	Yes*
Enterobacter cloacae[1]	18	2 (11.11)	5 (27.78)	11 (61.11)	50% (18.75 to 70.24%)	Yes
Escherichia coli	17	0	1 (5.88)	16 (94.12)	94.12% (66.10 to 98.95)	Yes
Klebsiella oxytoca	9	1 (11.11)	4 (44.44)	4 (44.44)	33.33% (-7.93 to 63.63)	Yes*
Klebsiella pneumoniae[2]	22	2 (9.09)	11 (50.00)	9 (40.91)	31.82% (6.08 to 53.21)	Yes
Morganella morganii[3]	4	0	2 (50.00)	2 (50.00)	50.00% (-10.21 to 85.00)	Yes*
Proteus mirabilis	6	1 (16.67)	1 (16.67)	4 (66.67)	50% (-4.04 to 77.32)	Yes*
Proteus vulgaris	5	0	0	5 (100)	100% (38.55 to 100)	Yes
Providencia rettgeri	28	9 (32.14)	7 (25.00)	12 (42.86)	10.71% (-13.99 to 33.70)	No
Providencia stuartii	4	0	0	4 (100)	100% (30.72 to 100)	Yes
Serratia marcescens	64	33 (51.56)	15 (23.44)	16 (25.00)	-26.56% (-41.40 to -9.73)	No
Shigella sonnei	2	0	1 (50.00)	1 (50.00)	50.00% (-27.26 to 90.55)	Yes*
Shigella spp	0	0	0	0	0	NA
Pseudomonas aeruginosa	**134**	**19 (14.18)**	**41 (30.60)**	**74 (55.22)**	**41.04% (30.13 to 50.57)**	**Yes**
***Salmonella* spp.**	**6**	**0**	**2 (33.33)**	**4 (66.67)**	**66.67% (13.11 to 90.32)**	**Yes**
Salmonella ser. *Enteritidis*	2	0	1 (50.00)	1 (50.00)	50.00% (-27.26 to 90.55)	Yes*
Salmonella ser. *Typhi*	4	0	1 (25.00)	3 (75.00)	75.00% (8.52 to 95.44)	Yes

*95% Confidence interval not statistically significant.
[1]Includes *Enterobacter cloacae* (5), *Enterobacter cloacae* complex (8), and *Enterobacter cloacae* ssp. *cloacae* (5) isolates.
[2]Includes *Klebsiella pneumoniae* (4) and *Klebsiella pneumoniae* ssp. *pneumoniae* (18) isolates.
[3]Includes *Morganella morganii* (1) and *Morganella morganii* ssp. *morganii* (3) isolates.

A statistically significant trend toward higher MIC values was observed *Enterobacter cloacae, Escherichia coli, Klebsiella pneumoniae, Proteus vulgaris, Providencia stuartii, Pseudomonas aeruginosa* and *Salmonella* species. The following footnote to the performance table is included in the package insert to address the trending observed for VITEK 2 AST-GN Ciprofloxacin:

Ciprofloxacin MIC values for Enterobacter cloacae, Escherichia coli, Klebsiella pneumoniae, Proteus vulgaris, Providencia stuartii, Pseudomonas aeruginosa and Salmonella species tended to be in exact agreement or at least one doubling dilution higher than the reference method.

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:

The FDA and CLSI susceptibility interpretive criteria for Ciprofloxacin are as listed in **Table 5**.

Table 5. FDA Recognized Interpretive Criteria for Ciprofloxacin

	Minimum Inhibitory Concentrations (µg/mL)[a]		
Organisms	**S**	**I**	**R**
Enterobacteriaceae	≤0.25	0.5	≥1
Salmonella spp.	≤0.06	0.12 - 0.5	≥1
Pseudomonas aeruginosa	≤0.5	1	≥2

S = Susceptible; I = Intermediate; R = Resistant
[a]According to CLSI M100-Ed31 and FDA STIC Website https://www.fda.gov/Drugs/DevelopmentApprovalProcess/DevelopmentResources/ucm410971.htm

VIII Proposed Labeling:

The labeling was updated as follows:

- Inclusion of a statement to address testing of non-indicated species, as required by the 21st Century Cures Act legislation
- Updated breakpoints for *Enterobacterales* and *Pseudomonas aeruginosa* with Ciprofloxacin
- Updated performance data using the currently recognized breakpoints, along with limitation statements to reflect results obtained from this evaluation
- Inclusion of a footnote to the performance table to reflect results obtained from a trending analysis
- Update of the previous limitation statement regarding lack of testing with resistant organisms to only include *Salmonella enteritidis, Salmonella typhi* and *Shigella sonnei*

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

IX Conclusion:

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

To support the implementation of changes to FDA-recognized susceptibility test interpretive criteria (i.e., breakpoints), this submission included a breakpoint change protocol that was reviewed and accepted by FDA. 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). The protocol outlined the specific procedures and acceptance criteria that bioMérieux intends to use to evaluate the VITEK 2 System with Ciprofloxacin when revised breakpoints for Ciprofloxacin are published on the FDA STIC webpage. The breakpoint change protocol included with the submission indicated that if specific criteria are met, BioMérieux will update the Ciprofloxacin 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.