510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY

I Background Information:

A 510(k) Number

K222073

B Applicant

bioMérieux, Inc

C Proprietary and Established Names

VITEK 2 AST-Gram Negative Cefazolin (≤1 - ≥32 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

II Submission/Device Overview:

A Purpose for Submission:

To obtain a substantial equivalence determination for the VITEK 2 AST-Gram Negative Cefazolin (≤ 1 - ≥ 32 µg/ml) assay used to define the *in vitro* antimicrobial susceptibility of *Escherchia coli* and *Proteus mirabilis* to Cefazolin on the VITEK 2 and VITEK 2 Compact Antimicrobial Susceptibility Test (AST) Systems.

B Measurand:

Cefazolin ≤ 1 - ≥ 32 µg/ml

C Type of Test:

Automated quantitative or qualitative antimicrobial susceptibility test

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

VITEK 2 AST-Gram Negative Cefazolin 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 Cefazolin is a quantitative test. Cefazolin has been shown to be active against most strains of the microorganisms listed below, according to the FDA label for this antimicrobial.

Active *in vitro* and in clinical infections:

Escherichia coli

Proteus mirabilis

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

D Special Instrument Requirements:

VITEK 2 and VITEK 2 Compact Systems using 9.04 software.

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-GN Cefazolin has the following concentrations in the card: 1, 2, and 8 µg/mL (equivalent standard method concentration by efficacy in µg/mL). The MIC result range for the VITEK 2 AST-GN Cefazolin is ≤1 - ≥32 µg/mL.

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 Omadacycline (≤0.25 - ≥16 µg/mL)

B Predicate 510(k) Number(s):
K213931

C Comparison with Predicate(s):

Device & Predicate Device(s):	**Device** K222073	**Predicate** K213931
Device Trade Name	VITEK 2 AST-Gram Negative Cefazolin (≤ 1 - ≥ 32 µg/ml)	VITEK 2 AST-Gram Negative Omadacycline (≤0.25 - ≥16 µg/mL)
General Device Characteristic Similarities		
Intended Use/Indications for Use	VITEK 2 AST-Gram Negative Cefazolin 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	VITEK 2 AST-Gram Negative Omadacycline 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

	determination of *in vitro* susceptibility to antimicrobial agents. VITEK 2 AST-Gram Negative Cefazolin is a quantitative test 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.	of *in vitro* susceptibility to antimicrobial agents. VITEK 2 AST-Gram Negative Omadacycline is a quantitative test 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.
Test Methodology	Automated quantitative antimicrobial susceptibility test for use with the VITEK 2 and VITEK 2 Compact Systems to determine the *in vitro* susceptibility of microorganisms.	Same
Inoculum	Saline suspension of organism	Same
Test Card	Gram Negative (AST-GN) Susceptibility Card	Same
Instrument	VITEK 2 and VITEK 2 Compact Systems	Same
Analysis Algorithm	Growth Pattern Analysis	Same
Type of Test	Quantitative	Same
General Device Characteristic Differences		
Antimicrobial Agent	Cefazolin	Omadacycline
Concentrations of antimicrobial on card	1, 2, 8 µg/ml	0.5, 2, 8, 16 µg/ml
Indications for Use	Cefazolin has been shown to be active against most strains of the microorganisms listed below,	Omadacycline has been shown to be active against most strains of the microorganisms listed below,

	according to the FDA label for this antimicrobial. Active *in vitro* and in clinical infections**:** *Escherichia coli* *Proteus mirabilis*	according to the FDA label for this antimicrobial. Active *in vitro* and in clinical infections: For ABSSSI: *Enterobacter cloacae* *Klebsiella pneumoniae* For CABP: *Klebsiella pneumoniae*

VI Standards/Guidance Documents Referenced:

- CLSI M07-A11: Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically; Approved Standard – 11th Edition (January 2018)
- CLSI M100-S32: Performance Standards for Antimicrobial Susceptibility Testing; 30th Informational Supplement (February 2022)
- FDA Class II Special Controls Guidance Document: Antimicrobial Susceptibility Test (AST) Systems; Guidance for Industry and FDA (Issued August 28, 2009)

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

Reproducibility testing for the VITEK 2 AST-Gram Negative Cefazolin was conducted at three sites (two external and one internal site) using a panel of ten Gram negative organisms consistent with the indications for use (i.e., three isolates of *Proteus mirabilis* and seven isolates of *Escherichia coli*). Each isolate was tested in triplicate, using separate inoculum, over three days for a total of 270 data points. Inocula were prepared using both the auto-dilution and manual dilution methods for testing with the VITEK 2 System. In addition, inocula were prepared by the manual dilution method for testing with the VITEK 2 Compact. The mode of MIC values was determined for each isolate and the reproducibility was calculated based on the number of MIC values that fell within ± one doubling dilution of the mode MIC value. The majority of data points were on-scale and within ± one doubling dilution agreement as compared to the mode MIC. The data was analyzed taking into consideration best-case and worst-case scenarios as described in the Class II Special Controls

Guidance Document: Antimicrobial Susceptibility Test (AST) Systems. The reproducibility performance is shown in Table 1.

Table 1: Reproducibility Performance

	VITEK 2		VITEK 2 Compact
	Manual Dilution	**Auto-Dilution**	**Manual Dilution**
Best case	99.6%	100%	100%
Worst case	99.3%	100%	99.6%

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):

   The CLSI recommended QC strain, namely *Escherichia coli* ATCC 25922, was tested a sufficient number of times (i.e., at least 20/site) at each testing site using both the VITEK 2 AST-GN Cefazolin and Broth Microdilution (BMD) reference method. Both the automatic dilution and manual dilution methods were used for the VITEK 2 and the manual dilution method was used for the VITEK 2 Compact. The results are summarized in **Table 2** below. Both the auto-dilution and the manual dilution methods for VITEK 2 and the manual dilution for VITEK 2 Compact QC results were within the expected range >95% of the time, which is acceptable.

   Because the CLSI recommended QC strain does not include the full CLSI/FDA-recommended dilution range required for QC testing, the sponsor included a footnote in labeling to indicate that the device does not include the full CLSI/FDA-recommended dilution range for QC testing.

Table 2: Quality Control Results for *Escherichia coli* ATCC 25922 for Cefazolin: VITEK 2 (Auto-Dilution and Manual Dilution Methods) and VITEK 2 Compact (Manual Dilution Method)

Organism	VITEK 2 Result Range	BMD Result Range (µg/mL)	VITEK 2 Auto-Dilution	BMD	VITEK 2 Manual Dilution	BMD	VITEK 2 Compact Manual Dilution	BMD
Escherichia coli ATCC 25922		≤0.125						
		0.25						
		0.5						
	≤1	1	0	53	0	29	0	30
	2	2	202	145	97	66	100	67

Organism	VITEK 2 Result Range	BMD Result Range (µg/mL)	VITEK 2 Auto-Dilution	BMD	VITEK 2 Manual Dilution	BMD	VITEK 2 Compact Manual Dilution	BMD
Expected Result: 1-4 µg/mL	4	4	1	5	0	2	0	2
	8	8	0	0	0	0	0	0
	16	16	0	0	1	0	0	0
	≥32	32	1	0	0	0	0	0
		≥64	0	1	0	1	0	1

BMD: broth microdilution

Because the CLSI recommended QC strain does not include the full CLSI/FDA-recommended dilution range required for QC testing the VITEK card reporting range (≤1 - ≥ 32 µg/mL), the sponsor verified *Escherichia coli* ATCC 35218 as additional QC. Both the auto-dilution and the manual dilution methods for VITEK 2 and the manual dilution for VITEK 2 Compact QC results were within the expected range >95% of the time, which is acceptable. The range finding study set the range for this organism at 2-8 µg/ml.

One ancillary quality control organism was tested throughout comparative testing by broth microdilution reference method only. This was done to perform further quality control of the broth microdilution panels. The organism tested was *Staphylococcus aureus* ATCC 29213. QC results for the broth microdilution method were within the expected result range >95% of the time. *S. aureus* ATCC 29213 was within range 100% (208/208).

Inoculum Density Control:
The DensiCHEK Plus was used to standardize the inoculum to a 0.5 McFarland standard. The instrument was standardized daily with all results recorded at each site. Calibration values were within the expected range.

Purity Check:
A purity check of all organisms was performed on the dilution tube used to prepare the VITEK 2 card inoculum. Only those cultures that were pure were evaluated in the study.

Device Failure:
During the performance of the comparative study, there were several instrument processing errors that resulted in loss of AST cards with the VITEK 2 System. All isolates affected by these errors were retested in accordance with the testing protocol. There were no device failures with the VITEK 2 Compact.

Study Protocol Changes:
During the course of the clinical trial, FDA updated the breakpoints for Cefazolin. For *Enterobacterales*, CLSI M-100 Ed 32 is recognized as of February 2022. Breakpoints changed from previous FDA-recognized breakpoints (≤1S, 2I, ≥4R) to the new FDA-recognized breakpoints (≤2S, 4I, ≥8R). The updated breakpoints were applied to the clinical trial data, and the performance was evaluated using the updated FDA breakpoints. This protocol change had no impact on testing or test results.

FDA does not recognize separate susceptibility test interpretative criteria for *Enterobacterales* for therapy of uncomplicated urinary tract infections. Therefore, the labeling for this device includes the following note:

> *Per the United States FDA-Recognized Susceptibility Test Interpretive Criteria (STIC) and CLSI, cefazolin should not be reported against Enterobacteriaceae recovered from uncomplicated urinary tract infections*.

Growth Failure Rate:
A total of 464 clinical and challenge isolates were tested by VITEK 2 AST-GN Cefazolin. One growth failure was recorded for a clinical isolate of *Escherichia coli*. Results for 463 isolates on the VITEK 2 AST were available.

6. Detection Limit:

   Not applicable.

7. Assay Cut-Off:

   Not applicable.

B Comparison Studies:

1. Method Comparison with Predicate Device:

   Testing of cefazolin on the VITEK 2 AST-Gram Negative card was performed at three external sites and one internal site. There were 387 clinical isolates and 76 challenge isolates tested for a total of 463 isolates tested. Results obtained with VITEK 2 AST-Gram Negative Cefazolin were compared to results obtained with the CLSI broth microdilution reference (BMD) panel. The MIC result range for the VITEK 2 AST-Gram Negative Cefazolin is ≤ 1 - ≥ 32 µg/mL for all species. The testing conditions for the reference method consisted of the following:

   - Medium: Cation Adjusted Mueller Hinton broth
   - Inoculum: 1 mL of organism suspension standardized to approximate a McFarland 0.5 standard
   - Incubation: 35 ± 2°C ambient air; 16-20 hours

   The VITEK 2 AST cards were inoculated with test organisms using the auto-dilution method (VITEK 2) and using the manual dilution method (VITEK 2 and VITEK 2 Compact). All test inocula used for the VITEK 2 AST cards and the reference method were standardized using the DensiCHEK Plus instrument.

   A total of 387 clinical isolates belonging to all genera were evaluated using auto-dilution and VITEK 2. Of these isolates, 64% were recent isolates (tested within one year of isolation) and 36% were stock isolates (no specific time from isolation). A total of 299 *Escherichia coli* clinical isolates and 45 *Proteus mirabilis* clinical isolates were tested as indicated species. In addition, isolates from non-indicated species were evaluated as clinical isolates:

Citrobacter freundii (1), *Citrobacter koseri (2)*, *Klebsiella pneumoniae (32)*, *Proteus vulgaris (1)*, *Enterobacter cloacae (1)*, *Enterobacter aerogenes (1)*, *Serratia marcescens (1)*, *Klebsiella oxytoca (1)*, *Morganella morganii (1)*, and *Providencia stuartii (1)*.

A total of 76 challenge isolates were evaluated, including 41 *E. coli* isolates and 35 *P. mirabilis* isolates. The challenge set was tested with the auto-dilution and manual dilution options of the VITEK 2 and with the manual dilution method on the VITEK 2 Compact.

Clinical and Challenge Data –VITEK 2 Auto-Dilution
The results obtained using the auto-dilution method of the VITEK 2 from the 463 total isolates (387 clinical isolates and 76 challenge isolates) are summarized in **Table 2**.

Table 3. Performance of All Clinical and Challenge Isolates for Cefazolin: VITEK 2 Auto-Dilution

	Tot	EA N	EA %	Eval Tot	Eval EA N	Eval EA %	CA Tot	CA %	No. R	No. S	min	maj	vmj
***Enterobacterales* [Breakpoints (µg/ml): <2 (S), 4 (I), >8 (R)]**													
Clinical[a]	387	376	97.2	262	252	96.2	313	80.9	91	216	70	3	1
Challenge[b]	76	75	98.7	61	60	98.4	74	97.4	31	14	2	0	0
Total	463	451	97.4	323	312	96.6	387	83.6	122	230	72	3	1

[a] includes 299 *E. coli*, 45 *P. mirabilis* and 43 other *Enterobacterales* isolates
[b] includes 41 *E. coli* and 35 *P. mirabilis*

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

Essential agreement (EA) occurs when the result of the reference method and that of the VITEK 2 AST-Gram Negative Cefazolin 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 VITEK 2 AST-Gram Negative Cefazolin or results 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 provided by the VITEK 2 AST-Gram Negative Cefazolin.

For all organisms evaluated using the auto-dilution method of the VITEK 2, EA was acceptable at 97.4% (**Table 3**). The CA of 83.6% was caused by 72 (15.6%) minor errors. Because the EA of evaluable results was 96.2%, and the error rates for major errors (1.3%) and very major errors (0.8%) were within acceptable limits, the CA of 83.6% was considered acceptable.

Challenge Data –VITEK 2 and VITEK 2 Compact Manual Dilution
The 76 challenge isolates were also tested at one site with the manual dilution option for the VITEK 2 and VITEK 2 Compact systems (summarized in **Table 4**).

Table 4. Performance of Challenge Isolates for Cefazolin: VITEK 2 Manual Dilution

	Tot	EA N	EA %	Eva Tot	Eval EA N	Eval EA %	CA Tot	CA %	No. R	No. S	min	maj	vmj
VITEK 2	76	73	96.1	60	57	95.0	72	94.7	31	14	4	0	0
VITEK 2 Compact	76	73	96.1	61	58	95.1	73	96.1	31	14	3	0	0

The overall performance of *Enterobacterales* is acceptable with an EA of 96.1%, a CA of 94.7%, and 4 minor errors (5.3%) for the VITEK 2 system.

The overall performance of *Enterobacterales* is acceptable with an EA of 96.1%, a CA of 96.1%, and three minor errors (3.9%) for the VITEK 2 Compact system.

As required under 511A(b)(2)(C)(ii)(I) of the Federal Food, Drug and Cosmetic Act, the following statement is included in the Precautions section of the device labeling to address testing and reporting 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.*

Trending

A trending analysis was conducted using the combined data (clinical and challenge) obtained from the VITEK 2 auto-dilution method. This trending calculation analyzes device MIC values that are determined to be one or more doubling dilutions lower or higher than the reference method. MIC values that are off-scale for both the reference and device are not considered in the trending analysis. Species for which the difference between the percentage of isolates with higher or lower MIC values was ≥ 30% with a statistically significant confidence interval were considered to have evidence of trending and is addressed in the labeling. There was no trending observed.

Table 5. Trending Analysis for *Enterobacterales* with VITEK 2 Auto-Dilution

Organism	Total Evaluable for Trending	≥ 1 Dilution lower No. (%)	Exact No. (%)	≥ 1 Dilution Higher No. (%)	Percent Difference (CI)	Trending Noted
E. coli	232	37 (15.9)	99 (42.7)	96 (41.4)	25.4% (17.3 to 33.1)	No
P. mirabilis	75	16 (21.3)	52 (69.3)	7 (9.33)	-12.0% (-23.6 to -0.35)	No

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 recognized susceptibility interpretative criteria for Cefazolin are listed in Table 6.

Table 6. FDA-Recognized Interpretative Criteria for Cefazolin

Organisms	Minimum Inhibitory Concentrations (µg/mL)[a]		
	S	I	R
Enterobacterales[b]	≤2	4	≥8

S = Susceptible; I = Intermediate; R = Resistant

Interpretative criteria are based on a dose of 2g every 8 hours

[a] FDA-Recognized Antimicrobial Susceptibility Test Interpretative Criteria Website https://www.fda.gov/Drugs/DevelopmentApprovalProcess/DevelopmentResources/ucm410971.htm

[b] Separate susceptibility test interpretative criteria for *Enterobacterales* for therapy of uncomplicated urinary tract infections are not recognized at this time.

VIII Proposed Labeling:

The labeling supports the finding of substantial equivalence for this device when evaluated with the current FDA-recognized Cefazolin breakpoints.

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 Cefazolin when revised breakpoints for Cefazolin 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 Cefazolin 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.