510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY

I Background Information:

A 510(k) Number

K220805

B Applicant

bioMérieux, Inc

C Proprietary and Established Names

VITEK 2 AST-Gram Positive Cefoxitin Screen

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
LTW	Class II	21 CFR 866.1640 - Antimicrobial susceptibility test powder	MI - Microbiology
LTT	Class II	21 CFR 866.1640 - Antimicrobial susceptibility test powder	MI - Microbiology

II Submission/Device Overview:

A Purpose for Submission:

To obtain a substantial equivalence determination for Cefoxitin Screen for predicting *mecA*-mediated oxacillin resistance in *Staphylococcus* spp. on the VITEK 2 and VITEK 2 Compact Antimicrobial Susceptibility Test (AST) Systems

B Measurand:

Cefoxitin 4, 5 µg/mL

C Type of Test:

Automated qualitative test designed to predict *mecA*-mediated oxacillin resistance in *Staphylococcus* spp.

III Intended Use/Indications for Use:

A Intended Use(s):
The VITEK 2 Gram-Positive Susceptibility Card is intended for use with the VITEK 2 Systems in clinical laboratories as an *in vitro* test to determine the susceptibility of *Staphylococcus* spp., *Enterococcus* spp., and *Streptococcus agalactiae* to antimicrobial agents when used as instructed.

B Indication(s) for Use:
VITEK 2 AST-Gram Positive Cefoxitin Screen test is designed to predict *mecA*-mediated oxacillin resistance in *Staphylococcus* spp. It 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.

The VITEK 2 Gram-Positive Susceptibility Card is intended for use with the VITEK 2 Systems in clinical laboratories as an *in vitro* test to determine the susceptibility of *Staphylococcus* spp., *Enterococcus* spp., and *Streptococcus agalactiae* to antimicrobial agents when used as instructed.

C Special Conditions for Use Statement(s):
Rx - For Prescription Use Only

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

- Cefoxitin Screen Test (oxsf02n): *Staphylococcus simulans*

Perform an alternative method of testing prior to reporting results when a Positive (+) result is obtained with the following antibiotic/organism combination(s):

- Cefoxitin Screen Test (oxsf02n): *Staphylococcus capitis*

D Special Instrument Requirements:

VITEK 2 and VITEK 2 Compact Systems, VITEK 2 Systems (PC) version 9.04

IV Device/System Characteristics:

A Device Description:

VITEK 2 AST-GP Cefoxitin Screen is designed to predict *mecA*-mediated oxacillin resistance in *Staphylococcus* spp. The cefoxitin screen and oxacillin work in combination to determine the final interpretation reported for oxacillin. The VITEK 2 AST-GP Cefoxitin Screen is a qualitative test based on the CLSI, "Disk Diffusion Test for Prediction of *mecA* mediated resistance in *Staphylococci*." The VITEK 2 AST-GP Cefoxitin Screen test 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.

The VITEK 2 card is inoculated with a standardized organism suspension, and growth inside the card is optically monitored throughout the incubation cycle. Results are automatically calculated once a predetermined growth threshold is reached and a report is generated that contains the MIC result and the interpretive category result.

VITEK 2 AST-GP Cefoxitin Screen has the following concentrations in the card: 4 and 5 µg/mL (equivalent standard method concentration by efficacy in µg/mL).

B Principle of Operation:

The VITEK 2 AST card is essentially a miniaturized, abbreviated and automated version of the doubling dilution technique. 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 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 (up to 24 hours for *Streptococcus* species). 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. For the VITEK 2 Cefoxitin Screen, the report will list either a positive or negative result. The VITEK 2 Cefoxitin Screen and oxacillin work in combination to determine the final oxacillin interpretation based on the CLSI recommendations.

V Substantial Equivalence Information:

A Predicate Device Name(s):

Vitek 2 Gram Positive Cefoxitin Screen

B Predicate 510(k) Number(s):

K053097

C Comparison with Predicate(s):

Device & Predicate Device(s):	Device: K220805	Predicate: K053097
Device Trade Name	VITEK 2 AST-Gram Positive Cefoxitin Screen	VITEK 2 AST Gram Positive Cefoxitin Screen
General Device Characteristic Similarities		
Intended Use/ Indications for Use	VITEK 2 AST-Gram Positive Cefoxitin Screen is designed to predict *mecA*-mediated oxacillin resistance in *Staphylococcus* spp. It 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. The VITEK 2 AST Gram-Positive Susceptibility Card is intended for use with the VITEK 2 Systems in clinical laboratories as an *in vitro* test to determine the susceptibility of *Staphylococcus* spp., *Enterococcus* spp., and *Streptococcus agalactiae* to antimicrobial agents when used as instructed.	VITEK 2 AST-Gram Positive Cefoxitin Screen is designed to predict *mecA*-mediated resistance in Staphylococci. It 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. The VITEK 2 AST Gram-Positive Susceptibility Card is intended for use with the VITEK 2 Systems in clinical laboratories as an *in vitro* test to determine the susceptibility of *Staphylococcus* spp., *Enterococcus* spp., and *Streptococcus agalactiae* to antimicrobial agents when used as instructed.
Test Methodology	Automated qualitative antimicrobial susceptibility test for use with VITEK 2 and VITEK 2 Compact Systems to determine the *in vitro* susceptibility of microorganisms	Same
Inoculum	Saline suspension of organisms	Same
Test Card	Gram Positive (AST-GP) Susceptibility Card	Same
Instrument	VITEK 2 and VITEK 2 Compact Systems	Same
Analysis Algorithm	Discriminant Analysis	Same
Antimicrobial Agent	Cefoxitin	Same
General Device Characteristic Differences		
Concentrations	4, 5	6

VI Standards/Guidance Documents Referenced:

1. MacLowry, J.D. and Marsh, H.H., Semi-automatic Microtechnique for Serial Dilution Antibiotic Sensitivity Testing in the Clinical laboratory, Journal of Laboratory Clinical Medicine, 72:685-687, 1968.

2. Gerlach, E .H., Microdilution 1 : A Comparative Study, p . 6 3-76. Current Techniques for Antibiotic Susceptibility Testing. A. Balows (ed.), Charles C. Thomas, Springfield, IL, 1974.

3. Barry, A.L., The Antimicrobic Susceptibility Test, Principles and Practices, Lea and Febiger, Philadelphia, PA, 1976.

4. CLSI Document M02, Performance Standards for Antimicrobial Disk Susceptibility Tests, Approved Standard -13th Edition (January 2018)

5. CLSI Document M100, Performance Standards for Antimicrobial Susceptibility Testing; 31st Edition, (March 2021).

6. 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-GP Cefoxitin Screen was conducted at three clinical sites using a panel of ten *Staphylococcus* spp. (i.e., five *S. aureus*, three *S. epidermidis,* one *S. haemolyticus* and one *S. saprophyticus* isolates). Each isolate was tested in triplicate over three days for a total of 270 data points. Inocula were prepared using both the auto-dilution and manual dilution methods for testing in the VITEK 2 System. In addition, the inocula were prepared by the manual dilution method for use with the VITEK 2 Compact. The data was analyzed as described in the Class II Special Controls Guidance Document: Antimicrobial Susceptibility Test (AST) Systems. The testing resulted in overall reproducibility of 100% for each dilution method and VITEK 2 system, which is acceptable.

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

The QC strains, namely *Staphylococcus aureus* BAA-1026 and *Staphylococcus aureus* ATCC 29213, are not CLSI-recommended for routine QC for detection of methicillin (oxacillin) resistance but have been evaluated and determined to be acceptable. Each QC strain was tested a sufficient number of times (i.e., at least 20/site) at each testing site using both the VITEK 2 AST-GP Cefoxitin Screen Test and the CLSI test for detecting methicillin (oxacillin) resistance with cefoxitin disk diffusion. 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 1** below.

Table 1: Quality Control Results for Cefoxitin Screen: VITEK 2 (Auto-Dilution and Manual Dilution Methods) and VITEK 2 Compact (Manual Dilution Method)

Organism	Expected DD Result*	Test Results	VITEK 2 Auto-Dilution	DD	VITEK 2 Manual Dilution	DD	VITEK 2 Compact Manual Dilution	DD
Staphylococcus aureus **BAA-1026**	**≤ 21 mm**	**NEG**	**1**	**0**	**0**	**0**	**0**	**0**
	POS	**POS**	**135**	**136**	**62**	**62**	**62**	**62**
Staphylococcus aureus **ATCC 29213**	**≥ 22 mm**	**NEG**	**138**	**138**	**61**	**61**	**61**	**61**
	NEG	**POS**	**0**	**0**	**0**	**0**	**0**	**0**

*Expected DD result is based on breakpoints according to FDA STIC Website
DD, 30 µg cefoxitin disk diffusion as surrogate test for oxacillin
POS, Positive; NEG, Negative

Both the auto-dilution and the manual dilution methods for VITEK 2 and the manual dilution for VITEK 2 Compact generated expected QC results >95% of the time, which is acceptable.

One ancillary quality control organism (*Staphylococcus aureus* ATCC 25923) was tested throughout the comparative testing by Cefoxitin disk diffusion to perform further quality control of the Cefoxitin disk. Expected QC results for the Cefoxitin disk diffusion method were generated 100% (147/147) of the time.

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.

Growth Failure Rate
A total of 818 clinical and challenge isolates were tested by VITEK 2 AST-GP Cefoxitin Screen Test. Four isolates failed to grow and a total of 814 isolates have VITEK 2 AST results available.

6. Detection Limit:

   Not applicable

7. Assay Cut-Off:

   Not applicable

B Comparison Studies:

1. Method Comparison with Predicate Device:
   A clinical study was conducted at five clinical sites using the VITEK 2 AST-GP Cefoxitin Screen. There were 739 clinical isolates and 75 challenge isolates tested for a total of 814 isolates. Results obtained with the VITEK 2 AST-GP Cefoxitin Screen Test were compared to the qualitative values determined by the CLSI test for detecting methicillin (oxacillin) resistance using cefoxitin disk diffusion as a surrogate test. The testing conditions for the cefoxitin disk diffusion method consisted of the following:

   - Medium – Mueller Hinton agar with Cefoxitin 30 µg disks
   - Inoculum – Direct colony suspension
   - Incubation – *S. aureus* and *S. lugdunensis*: 35°C ± 2°C; 16-18 hours. Other *Staphylococcus* spp. (excluding *S. pseudintermedius* and *S. scheiferi*: 35°C ± 2°C; 24 hours (may be reported after 18 hours, if resistant)

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

   A total of 743 clinical *Staphylococcus* spp. isolates were evaluated at five sites: 66.2% were considered contemporary isolates (isolated from clinical specimen and tested within 6 months) and 33.8% were stock isolates. Complete test results are available for 739 clinical isolates (453 *S. aureus*, 13 *S. capitis*, 185 *S. epidermidis*, 19 *S. haemolyticus*, 20 *S. lugdunensis*, 20 *S. saprophyticus*, 14 *S. simulans* and 15 *S. warneri*). All clinical isolates were tested with the auto-dilution option of the VITEK 2.

   A total of 75 challenge *Staphylococcus* spp. isolates (1 Coagulase negative staphylococcus, 55 *S. aureus*, 10 *S. epidermidis*, 4 *S. haemolyticus*, 4 *S. lugdunensis*, and 1 *S. simulans*) were evaluated at one site. 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

VITEK 2 AST-GP Cefoxitin Screen performance was determined with 814 isolates (739 clinical isolates and 75 challenge isolates) by comparing VITEK 2 results with cefoxitin disk diffusion results that were evaluated based on susceptibility testing interpretive criteria (breakpoints) established for *Staphylococcus* spp. A final VITEK 2 result for oxacillin was determined by comparison of the Cefoxitin Screen Test card result and the Oxacillin card result using forcing rules. Oxacillin is forced susceptible for coagulase-negative staphylococci (CoNS) other than *S. lugdunensis* and *S. epidermidis* when the oxacillin MIC result is 0.5 – 2 µg/mL and the cefoxitin screen result is negative. This rule was used in the final analysis of the result. After forcing, category agreement and error rates were calculated using Cefoxitin disk diffusion as the comparator method.

Table 2: Performance of All Clinical and Challenge Isolates for Cefoxitin Screen After Forcing Rules Compared to the Cefoxitin Disk Diffusion Method: VITEK 2 Auto-Dilution

Organism Type	Total	Number Neg (S)	Number Pos (R)	CA	%CA	maj	vmj
Staphylococcus aureus* and *Staphylococcus lugdunensis							
Clinical	473	322	151	467	98.7	5	1
Challenge	59	33	26	58	98.3	1	0
Combined	532	355	177	525	98.7	6	1
Other Staphylococci spp.*							
Clinical	252	139	113	246	97.6	6	0
Challenge	15	3	12	15	100	0	0
Combined	267	142	125	261	97.8	6	0

CA – Category Agreement
R – Resistant isolates
S – Susceptible isolates
maj – major errors
vmj – very major errors

*Fifteen *Staphylococcus simulans* isolates are not included in performance calculation for other Staphylococci spp.

The overall performance of *S. aureus* and *S. lugdunensis* is acceptable with a CA of 98.7%. There were six major errors (6/355 = ~1.7%) and one very major error (1/177 = ~0.6%). The overall performance of other Staphylococci spp. is acceptable with a CA of 97.8%. There were no very major errors and six major errors due to an increased major error rate observed with *S. capitis.*

To address the performance issue noted above for *S. capitis*, the sponsor included the following limitation in the device labeling:

Perform an alternative method of testing prior to reporting results when a Positive (+) result is obtained with the following antibiotic/organism combination(s):

- *Cefoxitin Screen Test (oxsf02n): Staphylococcus capitis*

In addition to evaluating results obtained with *Staphylococcus* spp., 15 isolates of *S. simulans* were tested with the auto-dilution method. However, performance of cefoxitin screen with this species was not acceptable. The following limitation was added to the device labeling:

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

- *Cefoxitin Screen Test (oxsf02n): Staphylococcus simulans*

Challenge Data – VITEK 2 and VITEK 2 Compact Manual Dilution

The 75 challenge isolates were also tested at one site with the manual dilution option for the VITEK 2 and VITEK 2 Compact systems (**Table 3**). A final result for oxacillin was determined by comparison of the Cefoxitin Screen Test card result and the Oxacillin card result using forcing rules. After forcing, category agreement and error rates were calculated using Cefoxitin disk diffusion as the comparator method.

Table 3: Performance of Cefoxitin Screen Test + Oxacillin After Forcing Rules Compared to the Cefoxitin Disk Diffusion Method – VITEK 2 Manual Dilution

	Total*	Number Neg (S)	Number Pos (R)	CA	%CA	maj	vmj
VITEK 2	74	36	38	73	98.6	1	0
VITEK 2 Compact	74	36	38	73	98.6	1	0

CA – Category Agreement
S – Susceptible isolates
R – Resistant isolates
maj – major errors
vmj – very major errors

*One *S. simulans* isolate was not included in performance calculation for VITEK 2 and VITEK 2 Compact

The overall performance of *Staphyloccocus* spp. is acceptable with a CA of 98.6% when using the manual dilution with both the VITEK 2 and VITEK 2 Compact systems. There was one major error (1/36 = ~2.8%) and no very 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:

The FDA-recognized susceptibility interpretive criteria for Cefoxitin (surrogate test for oxacillin) is listed in Table 4.

Table 4: FDA-Recognized Interpretive Criteria for Cefoxitin (zone diameter in mm)[a,b]

Organism	Susceptible	Intermediate	Resistant
Staphylococcus aureus complex *Staphylococcus lugdunensis*	≥22	-	≤21
Staphylococcus epidermidis	≥25	-	≤24
Other Staphylococci spp.	≥25	-	≤24

[a] According to FDA STIC Website
[b] 30 µg cefoxitin disk as surrogate test for oxacillin

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.