510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY

I Background Information:

A 510(k) Number

K232201

B Applicant

bioMérieux, Inc

C Proprietary and Established Names

VITEK 2 AST-Streptococcus Penicillin (≤0.06 - ≥8 µg/mL), VITEK 2 Streptococcus Penicillin (≤0.06 - ≥8 µg/mL), VITEK 2 Streptococcus Penicillin

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 update VITEK 2 AST-Streptococcus Penicillin device labeling to include FDA-recognized oral penicillin (Penicillin V) breakpoints for *Streptococcus pneumoniae*. Performance data originally obtained and evaluated using parenteral (injection) penicillin breakpoints for *S. pneumoniae* (meningitis and non-meningitis) in K112000 was reanalyzed with oral penicillin breakpoints in this submission.

Breakpoints and performance data for β-hemolytic Streptococci and Viridians species remain unchanged since the original clearance (K112000).

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

B Measurand:

Penicillin concentrations of 0.06, 0.12, 0.5, and 2 µg/mL. The MIC result range for the VITEK 2 AST-Streptococcus Penicillin test is ≤0.06 - ≥8 µg/mL

C Type of Test:

Automated quantitative or qualitative antimicrobial susceptibility test for penicillin.

III Intended Use/Indications for Use:

A Intended Use(s):

The VITEK 2 *Streptococcus* Susceptibility Card is intended for use with the VITEK 2 Systems in clinical laboratories as an *in vitro* test to deterine the susceptibility of *Streptococcus pneumoniae,* beta-hemolytic *Streptococcus*, and Viridans *Streptococcus* to antimicrobial agents when used as instructed.

Indication(s) for Use:

VITEK 2 Streptococcus Penicillin is designed for antimicrobial susceptibility testing of *Streptococcus* species 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 Streptococcus Penicillin is a quantitative test. Penicillin 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:

Beta hemolytic streptococci groups C and G
Streptococcus pyogenes
Streptococcus agalactiae
Streptococcus viridans group
Streptococcus pneumoniae

The VITEK 2 *Streptococcus* Susceptibility Card is intended for use with the VITEK 2 Systems in clinical laboratories as an *in vitro* test to deterine the susceptibility of *Streptococcus pneumoniae,* beta-hemolytic *Streptococcus*, and Viridans *Streptococcus* to antimicrobial agents when used as instructed.

B Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

The ability of the AST card to detect resistance with the following combination(s) is unknown because resistant strains were not available or an insufficient number were encountered at the

time of comparative testing: Benzylpenicillin (p01n): *Streptococcus pneumoniae* (When applying breakpoints of ≤2 (S), 4 (I), ≥8 (R)), Beta-Hemolytic *Streptococcus*

C Special Instrument Requirements:

For use with the VITEK 2 and VITEK 2 Compact Systems. VITEK 2 and VITEK 2 Compact Systems (VITEK 2 Systems version 5.02 was used in K11200. New breakpoints will be incorporated into a future VITEK 2 Systems software release.)

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 which contains 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. The data are 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.

The VITEK 2 AST-Streptococcus Penicillin has the following concentrations in the card: 0.06, 0.12, 0.5, and 2 µg/mL (equivalent standard method concentration by efficacy in µg/mL). The MIC result range for the VITEK 2 AST-Streptococcus Penicillin test is ≤0.06 - ≥8 µg/mL. For all species, the MIC result range indicates that the VITEK 2 system is capable of producing the following MIC results ≤0.06, 0.125, 0.25, 0.5, 1, 2, 4 and ≥8 for AST-Streptococcus Penicillin test. This means the VITEK 2 systems does not provide results lower than 0.06µg/mL or greater than 8 µg/mL for the AST-Streptococcus Penicillin 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 Streptococcus Penicillin

B Predicate 510(k) Number(s):
K112000

C Comparison with Predicate(s):

Device & Predicate Device(s):	Device **K232201**	Predicate **K112000**
Device Trade Name	VITEK 2 AST-Streptococcus Penicillin (≤0.06 - ≥8 µg/mL)	VITEK 2 AST-ST Penicillin (≤0.06 - ≥8 µg/mL)
General Device Characteristic Similarities		
Intended Use/Indications for Use	VITEK 2 Streptococcus Penicillin is designed for antimicrobial susceptibility testing of *Streptococcus* species 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 Streptococcus Penicillin is a quantitative test. Penicillin 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: Beta hemolytic Streptococci groups C and G *Streptococcus pyogenes* *Streptococcus agalactiae* *Streptococcus* viridans group *Streptococcus pneumoniae*	Same
Test Methodology	Automated quantitative antimicrobial	Same

Device & Predicate Device(s):	Device K232201	Predicate K112000
	susceptibility test for use with the VITEK 2 and VITEK 2 Compact Systems to determine the *in vitro* susceptibility of microorganisms	
Antimicrobial Agent	Penicillin	Same
Inoculum	Saline suspension of organism	Same
Test Card	Streptococcus (AST-ST) Susceptibility Card	Same
Analysis Algorithms	Discriminant Analysis	Same
Concentrations	0.06, 0.12, 0.5, 2	Same
General Device Characteristic Differences		
Breakpoints (S/I/R)	• *S. pneumoniae* (non-meningitis) [≤2 / 4 / ≥8] • *S. pneumoniae* (meningitis) [≤0.06/ - / ≥0.12] • Viridans Streptococci [≤0.12/ 0.25-2 / ≥4] • Beta-hemolytic *Streptococcus* [≤0.12/ - / -] • *S. pneumoniae* (oral/non-meningitis) [≤0.06/ 0.12-1/ ≥2]	• *S. pneumoniae* (non-meningitis) [≤2 / 4 / ≥8] • *S. pneumoniae* (meningitis) [≤0.06/ - / ≥0.12] • Viridans Streptococci [≤0.12/ 0.25-2 / ≥4] • Beta-hemolytic *Streptococcus* [≤0.12/ - / -] • *S. pneumoniae* (oral) [none]

VI Standards/Guidance Documents Referenced:

1. FDA Class II Special Controls Guidance Document: Antimicrobial Susceptibility Test (AST) Systems; Guidance for Industry and FDA (Issued August 28, 2009)
2. CLSI M7-A5, "Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically; Approved Standard - Fifth Edition", (January 2000)
3. CLSI M07, “Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically; Approved Standard – Seventh Edition”, (January 2006)
4. CLSI M07, "Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically; Approved Standard **-** Eleventh Edition", (January 2018)
5. CLSI M100, "Performance Standards for Antimicrobial Susceptibility Testing; 33rd Edition", (March 2023)

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

   Reproducibility testing for the VITEK 2 AST-Streptococcus Penicillin was previously evaluated during review of K112000 and was determined to be 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, Inoculum and Growth failure for the VITEK 2 AST-Streptococcus Penicillin were performed in support of clearance of K112000 and were determined acceptable. QC testing specific to the additional oral penicillin breakpoints was not performed in the current submission since the penicillin QC range of 0.25 – 1 µg/mL used in K112000 was unchanged for *S. pneumoniae* ATCC 49619. The QC results with the unchanged QC range are shown in **Table 1**.

   In summary, the QC organism recommended by both the FDA and CLSI, namely *S. pneumoniae* ATCC 49619 was tested using the reference broth microdilution (BMD) method as well as the VITEK 2 AST-Streptococcus Penicillin by the automatic and the manual dilution methods during the clinical study a minimum 20 times/site. QC results for the VITEK 2 AST-Streptococcus Penicillin were within the expected results range >95% of the time for both automatic and manual dilution modes of the VITEK 2 (**Table 1**), which is acceptable. In addition, a similar QC study conducted to evaluate the VITEK 2 Compact System demonstrated QC results within the expected results range >95% of the time, which is acceptable.

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

Organism	VITEK 2 Result Range (µg/mL)	BMD Result Range (µg/mL)	VITEK 2 Auto-Dilution	BMD	VITEK 2 Manual Dilution	BMD
S. pneumoniae **ATCC 49619**		≤0.03				
	≤0.06	0.06				
	0.12	0.12				
	0.25	0.25	22	79	12	78
Expected Result	0.5	0.5	154	86	154	94
0.25 – 1 µg/mL	1	1	2	3	9	3
	2	2				
	4	4				

Organism	VITEK 2 Result Range (µg/mL)	BMD Result Range (µg/mL)	VITEK 2 Auto-Dilution	BMD	VITEK 2 Manual Dilution	BMD
	≥8	8				
		16				
		≥32				

6. Detection Limit:

   Not applicable

7. Assay Cut-Off:

   Not applicable

B Comparison Studies:

1. Method Comparison with Predicate Device:

   The VITEK 2 AST-ST Penicillin was originally cleared in premarket submission K112000 and included indications for Beta-hemolytic streptococci groups C and G, *Streptococcus pyogenes*, *Streptococcus agalactiae*, *Streptococcus* viridans group, and *Streptococcus pneumoniae* for testing with the VITEK 2 auto dilution, the VITEK 2 manual dilution, and the VITEK 2 Compact manual dilution. VITEK 2 AST-ST Penicillin results were compared to the CLSI broth microdilution reference method.

   The performance of the VITEK 2 AST-Streptococcus Penicillin with *S. pneumoniae* using the current FDA-recognized oral penicillin breakpoints was evaluated by reanalyzing the data obtained for *S. pneumoniae* in support of K112000. *S. pneumoniae* performance when using both the oral penicillin breakpoints as well as the unchanged parenteral (injection) penicillin (meningitis and non-meningitis) breakpoints are summarized in this submission. Breakpoints for β-hemolytic Streptococci and Viridians species also remain unchanged; performance data are not shown here.

   Since there was no change in the design or the penicillin dilution range of the VITEK 2 AST card, the performance evaluation of the VITEK 2 AST-Streptococcus Penicillin with *S. pneumoniae* for the oral penicillin breakpoints was achieved by reanalyzing the penicillin MIC data of the original 510(k) submission (K112000). For this review, the current penicillin (oral and parenteral) breakpoints are applied to *S. pneumoniae* according to the FDA STIC webpage.

   A total of 350 (clinical and challenge) *S. pneumoniae* isolates were tested using the VITEK 2 with the auto dilution. *S. pneumoniae* results of the original data using the unchanged parenteral breakpoints and re-analyzed data using the oral breakpoints are summarized in **Table 2**.

   The Essential Agreement (EA) and Categorical Agreement (CA) when using each of the parenteral breakpoints 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). When using the oral breakpoints, the EA is >90% and the CA is 89.1%, which is acceptable since the majority of errors is minor and the EA of evaluable results is very good, as described in the AST guidance. There were no major or very major errors observed with the oral and parenteral non-meningitis breakpoints.

When analyzing the performance data with the **parenteral non-meningitis penicillin breakpoints**, the Essential Agreement (EA) and Categorical Agreement (CA) 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). There were no major or very major errors observed.

When analyzing the performance data with the **parental meningitis penicillin breakpoints**, the EA and CA is >90%, which is acceptable, as described in the AST guidance. As previously reported in K112000, there were 6 major errors (3.2% major error rate) and 2 very major errors (1.2% very major error rate). The major error rate was considered acceptable given the very high overall CA agreement and lack of an intermediate meningitis interpretive criterion for penicillin. During this review, further analysis of the categorical errors was performed and adjustments made by considering the MIC values where the errors occurred. Five of the six major errors and one of the two very major errors was one doubling dilution from the reference and thus in essential agreement. Therefore, the adjusted major error rate is 0.5% (1/189) and the adjusted very major error rate is 0.6% (1/161), which is acceptable. To address the adjustment of the major error, the following performance footnote was included in the device labeling:

> "*The overall major error rate for penicillin when testing S. pneumoniae and using the meningitis breakpoints is 3.2% (6/189). Based on the essential agreement and lack of an intermediate meningitis breakpoint for penicillin, the overall adjusted major error rate for S. pneumoniae clinical and challenge isolates is 0.5% (1/189).*"

When analyzing the performance data with the **oral non-meningitis penicillin breakpoint**s, the EA is >90% and the CA is 89.1%, which is acceptable since the majority of errors are minor and the EA of evaluable results is very good, as described in the AST guidance. There were no major or very major errors observed.

Table 2. Analysis of the Original Performance for Penicillin with *Streptococcus pneumoniae* Using Parenteral Non-meningitis, Parenteral Meningitis and Oral Non-meningitis Breakpoints with the VITEK 2 (Auto Dilution)

	Total	#EA	%EA	Eval EA Total	Eval EA #	Eval EA %	#CA	%CA	#R	#S	min	Maj	Vmj
***Streptococcus pneumoniae* Parenteral Non-meningitis Breakpoints ≤2 (S), 4 (I), ≥8 (R)**													
Clinical	300	293	97.7	117	110	94.0	276	92.0	0	266	24	0	0
Challenge	50	49	98	49	48	98	47	94.0	0	47	3	0	0
Combined	350	342	97.7	166	158	95.2	323	92.3	0	303	27	0	0
***Streptococcus pneumoniae* Parenteral Meningitis Breakpoints ≤0.06 (S), ≥0.12 (R)**													
Clinical	300	293	97.7	117	110	94.0	292	97.3	112	188	N/A	6	2
Challenge	50	49	98	49	48	98	50	100	49	1	N/A	0	0
Combined	350	342	97.7	166	158	95.2	342	97.7	161	189	N/A	6	2
***Streptococcus pneumoniae* Oral Non-meningitis Breakpoints ≤0.06 (S), 0.12-1 (I), ≥2 (R)**													

	Total	#EA	%EA	Eval EA Total	Eval EA #	Eval EA %	#CA	%CA	#R	#S	min	Maj	Vmj
Clinical	300	293	97.7	117	110	94.0	276	92.0	61	188	24	0	0
Challenge	50	49	98	49	48	98	38	76	31	1	14	0	0
Combined	350	342	97.7	166	158	95.2	314	89.7	92	189	38	0	0

CA – Category Agreement
Eval – Evaluable isolates
R – Resistant isolates
S – Susceptible 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.

Performance of the VITEK 2 and the VITEK 2 Compact was also evaluated with the same 50 challenge organisms using the manual dilution method. Comparisons to the reference method are shown below in **Table 3.**

Table 3. Analysis of the Original Performance for Penicillin with *Streptococcus pneumoniae* Using Parenteral Non-meningitis, Parenteral Meningitis and Oral Non-meningitis Breakpoints with the VITEK 2 Systems (Manual Dilution)

	Total	#EA	%EA	Eval EA Total	Eval EA #	Eval EA %	#CA	%CA	#R	#S	min	Maj	Vmj
***Streptococcus pneumoniae* Parenteral Non-meningitis Breakpoints ≤2 (S), 4 (I), ≥8 (R)**													
VITEK 2	50	48	96	48	46	95.8	46	92	0	40	4	0	0
VITEK 2 Compact	50	48	96	49	47	95.9	47	97	0	39	3	0	0
***Streptococcus pneumoniae* Parenteral Meningitis Breakpoints ≤0.06 (S), ≥0.12 (R)**													
VITEK 2	50	48	96	48	46	95.8	50	100	49	1	N/A	0	0
VITEK 2 Compact	50	48	96	49	47	95.9	50	100	49	1	N/A	0	0
***Streptococcus pneumoniae* Oral Non-meningitis Breakpoints ≤0.06 (S), 0.12-1 (I), ≥2 (R)**													
VITEK 2	50	48	96	48	46	95.8	38	76	31	1	12	0	0
VITEK 2 Compact	50	48	96	49	47	95.9	40	80	33	1	10	0	0

Resistant Organisms

The ability of the AST card to detect resistance with the following combination(s) is unknown because resistant strains were either not available or an insufficient number were encountered at the time of comparative testing:

- Benzylpenicillin (p01n): *Streptococcus pneumoniae* (When applying breakpoints of ≤2 (S), 4 (I), ≥8 (R)), Beta-Hemolytic *Streptococcus*

MIC Trending Analysis

A trending reanalysis was performed using the combined (clinical and challenge) data obtained in K112000 to align with current trending analyses. 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. No trending was observed.

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 penicillin when tested against streptococci are listed in **Table 4.**

Table 4. FDA Recognized Interpretive Criteria for Penicillin [a]

Pathogen	Minimum Inhibitory Concentrations (µg/mL)		
	S	I	R
S. pneumoniae (oral, non-meningitis)	≤0.06	0.12 - 1	≥2
S. pneumoniae (non-meningitis)	≤2	4	≥8
S. pneumoniae (meningitis)	≤0.06	-	≥0.12
Viridans Streptococci	≤0.12	0.25 - 2	≥4
Beta hemolytic *Streptococcus*	≤0.12	-	-

S = Susceptible; I = Intermediate; R = Resistant

[a] According to FDA STIC Website

VIII Proposed Labeling:

The labeling was updated to include:

- Addition of oral penicillin non-meningitis breakpoints for testing *S. pneumoniae*.
- Performance data using the unchanged FDA-recognized parenteral penicillin breakpoints for *S. pneumoniae* (meningitis and non-meningitis) and additional performance data using the FDA-recognized oral penicillin breakpoints
- Inclusion of a footnote to the performance table to describe the adjusted major error rate when using parental meningitis penicillin breakpoints.

The labeling supports the finding of substantial equivalence for this device when evaluated with the current FDA-recognized penicillin breakpoints for tests with *Streptococcus pneumoniae.*

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. 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 penicillin when revised breakpoints for penicillin 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 penicillin 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.