510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY

I Background Information:

A 510(k) Number

K213241

B Applicant

BioMerieux, Inc.

C Proprietary and Established Names

VITEK 2 AST-Yeast Fluconazole (≤ 0.5-≥ 64 µg/mL), VITEK 2 AST-YS Fluconazole (≤0.5-≥ 64 µg/mL), VITEK 2 AST-YS Fluconazole

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 a modification to Fluconazole testing on the VITEK 2 and VITEK 2 Compact Systems Antimicrobial Susceptibility Test (AST) Systems using revised, species-specific interpretative criteria that are currently recognized.

B Measurand:

Fluconazole concentrations on VITEK 2 AST Yeast Fluconazole card: 2, 4,8,16, 32 and 64 µg/mL. The MIC result range for the card is ≤ 0.5 - ≥ 64 µg/mL.

C Type of Test:

Automated quantitative antifungal susceptibility test of *Candida* species to fluconazole.

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

VITEK 2 AST-Yeast Fluconazole is designed for antifungal susceptibility testing of Candida 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 antifungal agents. VITEK 2 AST-Yeast Fluconazole is a quantitative test. Fluconazole has been shown to be active against most strains of the microorganisms listed below, according to the FDA label for this antifungal.

Active in vitro and in clinical infections:

Candida albicans

Candida parapsilosis

Candida tropicalis

The VITEK 2 Fungal 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 yeasts to antifungal agents when used as instructed.

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

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

- Fluconazole (flu02n): *Candida glabrata*
- Fluconazole (flu02n): *Candida parapsilosis* when the MIC is 1 or 2 µg/mL

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

- Fluconazole (flu02n): *Candida tropicalis*

D Special Instrument Requirements:

For use with the VITEK 2 and VITEK 2 Compact Systems

IV Device/System Characteristics:

A Device Description:

VITEK 2 AST-YS Fluconazole has the following concentrations in the card: 2, 4, 8, 16, 32, and 64 µg/mL (equivalent standard method concentration by efficacy in µg/mL).

The MIC result range for VITEK 2 AST-YS Fluconazole on the VITEK 2 card for is ≤0.5 – ≥64 µg/mL. For all species, the MIC result range indicates that the VITEK 2 system is capable of producing the following MIC results ≤0.5, 1, 2, 4, 8, 16, 32, ≥64 µg/mL for AST-YS Fluconazole test. This means the VITEK 2 systems does not provide results lower than 0.5 µg/mL or greater than 64 µg/mL for the AST-YS Fluconazole test.

The MIC interpretive criteria and equivalent concentrations are as follows:

VITEK 2 AST-YST	Equivalent Standard Method Concentration by Efficacy in µg/mL	MIC Ranges and FDA Categories* (MIC in µg/mL)		
		S	I	R
Fluconazole	2, 4, 8, 16, 32, 64	*Candida spp.*		
		≤ 2	4	≥ 8

B Principle of Operation:

The principle of the VITEK 2 AST cards is based on the microdilution minimum inhibitory concentration (MIC) technique reported by MacLowry and Marsh and Gerlach. 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 which only contains microbiological culture media is resident on all cards. The remaining wells contain premeasured portions of a specific antibiotic combined with culture media. The bacterial or yeast isolate to be tested is diluted to a standardized concentration with 0.45 – 0.5% saline before being used to rehydrate the antimicrobial medium within the card. The VITEK 2 System automatically fills, seals and places 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. At the completion of the incubation cycle, a report is generated that contains the MIC value along with the interpretive category result for each antibiotic contained on the card.

V Substantial Equivalence Information:

A Predicate Device Name(s):
Vitek 2 Ast - Yeast Fluconazole

B Predicate 510(k) Number(s):
K133817

C Comparison with Predicate(s):

Device & Predicate Device(s):	Device: K213241	Predicate: K133817
Device Trade Name	VITEK 2 AST-YS Fluconazole	VITEK 2 AST-Yeast Fluconazole
General Device Characteristic Similarities		
Intended Use	VITEK 2 AST-Yeast Fluconazole is designed for antifungal susceptibility testing of Candida 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 antifungal agents. VITEK 2 AST-Yeast Fluconazole is a quantitative test. Fluconazole has been shown to be active against most strains of the microorganisms listed below, according to the FDA label for this antifungal. Active in vitro and in clinical infections: *Candida albicans* *Candida parapsilosis* *Candida tropicalis* The VITEK 2 Fungal Susceptibility Card is intended for use with the	VITEK 2 AST-Yeast Fluconazole is designed for antifungal susceptibility testing of Candida 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 antifungal agents. VITEK 2 AST-Yeast Fluconazole is a quantitative test. Fluconazole has been shown to be active against most strains of the microorganisms listed below, according to the FDA label for this antifungal. Active in vitro and in clinical infections: *Candida albicans* *Candida parapsilosis* *Candida tropicalis* The VITEK 2 Fungal Susceptibility Card is intended for use with the VITEK 2 Systems for the

	VITEK 2 Systems in clinical laboratories as an in vitro test to determine the susceptibility of clinically significant yeasts to antifungal agents when used as instructed.	automated quantitative or qualitative susceptibility testing of isolated colonies for most clinically significant aerobic Gram-negative bacilli, Staphylococcus spp., Enterococcus spp., Streptococcus spp., and clinical significant yeast.
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 yeast.	Same
Antimicrobial Agent	Fluconazole	Same
Inoculum	Saline suspension of organism	Same
Test Card	VITEK 2 Yeast (AST) Susceptibility Test Card	Same
Analysis Algorithms	Discriminant Analysis	Same
Instrument	VITEK 2 and VITEK 2 Compact Systems	Same
Concentrations	2, 4, 8, 16, 32, 64	Same
General Device Characteristic Differences		
Claimed species	*Candida albicans* *Candida parapsilosis* *Candida tropicalis*	*Candida albicans* *Candida parapsilosis* *Candida tropicalis* *Candida dubiniensis* *Candida guilliermondii* *Candida lusitaniae*
Breakpoints for *Candida* spp.	*Candida albicans:* ≤2 (S), 4 (I), ≥8 (R) *Candida parapsilosis:* ≤2 (S), 4 (I), ≥8 (R) *Candida tropicalis:* ≤2 (S), 4 (I), ≥8 (R)	*Candida* spp.: ≤8 (S), 16-32 (I), ≥64 (R)

VI Standards/Guidance Documents Referenced:

CLSI Document M27, "*Reference Method for Broth Dilution Antifungal Susceptibility Testing of Yeasts*"; Fourth Edition (November 2017)

CLSI Document M27-S4, "*Reference Method for Broth Dilution Antifungal Susceptibility Testing of Yeasts*"; Fourth Informational Supplement (December 2012)

CLSI Document M60, "*Performance Standards for Antifungal Susceptibility Testing of Yeast*"; Second Edition (November 2017)

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

   Reproducibility studies were performed using the ten isolate study design in triplicate at three external clinical sites on three separate days. The studies included both the automated and manual dilution methods with the VITEK 2 instrument system and the manual dilution method with the VITEK 2 Compact instrument system. Greater than 95% reproducibility was demonstrated with both the VITEK 2 and VITEK 2 Compact Systems. There are no concerns.

2. Linearity:

   N/A

3. Analytical Specificity/Interference:

   N/A

4. Assay Reportable Range:

   N/A

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

   Inoculum Density Check: Inoculum density control was monitored using the DensiCHEK Plus instrument.

   Purity Check: Verification of isolate purity was conducted on all clinical, challenge and reproducibility organism suspensions.

   Growth or Device Failure: The VITEK 2 AST-YS Fluconazole trial had several device failures including isolated mechanical failures that did not repeat (host communication error, mechanical jam, failure to eject, and an optics failure) and user errors. These errors did not raise concerns since they were isolated and did not reflect systematic issues.

   Quality Control Testing: The CLSI recommended QC strains *Candida krusei* (ATCC 6258) and *Candida parapsilosis* (ATCC 22019) were tested a minimum of 20 times at each site by the reference method, the VITEK 2 instrument platform using both auto- and manual dilution

methods, and the VITEK 2 Compact instrument platform using the manual dilution method. A summary of the QC performance is provided in **Table 1**.

The quality control results for the VITEK 2 AST-YS Fluconazole were within the recommended range ≥95% which is acceptable.

Table 1. Quality Control Results for VITEK 2 Fluconazole

QC Organisms (All sites)	Expected Range (Fluconazole, µg/mL)	Concentration (µg/mL)	VITEK 2 Manual Dilution Frequency		VITEK 2 Auto Dilution Frequency		VITEK 2 Compact Manual Dilution	
			Test	Reference	Test	Reference	Test	Reference
Candida parapsilosis ATCC 22019	0.5-4	≤0.25						
		0.5		20		20		20
		1		104	8	103	9	104
		2	142	24	147	24	147	24
		4	13	8		8		8
		8	1					
		16						
		32						
		64						
		≥128						
Candida krusei ATCC 6258	8-64	≤0.25						
		0.5						
		1						
		2						
		4						
		8	21	39	26	39	27	38
		16	19	103	36	102	35	103
		32	109	14	93	14	93	14
		64	7					
		≥128						

6. Detection Limit:

   N/A

7. Assay Cut-Off:
   N/A

B Comparison Studies:

1. Method Comparison with Predicate Device:

   The performance of the VITEK 2 System was established for *Candida* spp. based on a previously conducted clinical study at three external sites and one internal site. Performance data for the three claimed *Candida* species was reanalyzed using updated STIC breakpoint criteria for 367 clinical isolates and 64 challenge isolates of *C. candida, C. parapsilosis,* and *C. tropicalis* that were previously tested. Due to the updated STIC recognized organisms for Fluconazole, the challenge isolates fell to 64 isolates from the previous submission. To compensate for the lack of challenge isolates and meet FDAs recommended criteria of 75

challenge organisms, the sponsor tested 11 new *C. albicans* challenge isolates and combined the analysis with the reanalyzed clinical and challenge data. All comparisons were made to the 24-hour broth microdilution reference method as described in CLSI documents M27-A3 and M27-S4. The clinical isolate specimens were composed of 282 (76.8%) fresh and 85 (23.2%) stock samples. Both automated and manual dilution methods were tested on the VITEK 2 System. Challenge isolates were also tested with the manual dilution method on the VITEK 2 Compact.

Essential agreement (EA) was calculated for when the VITEK 2 system results were within +/- two doubling dilutions of the reference method results. Evaluable results were defined as when both the reference method results and the VITEK 2 system results were on-scale. Evaluable results were also defined as when the reference method results were on-scale and off-scale VITEK 2 system results clearly did not agree within the accepted +/- two doubling dilutions. Category agreement was calculated for when the VITEK 2 system result interpretations agreed exactly with the reference method result qualitative interpretations. Results are summarized in Table 2 below.

Table 2. Overall Performance of Clinical and Challenge Isolates for VITEK 2 Fluconazole.

	Tot	# EA	% EA	Eval Tot	# Eval EA	% Eval EA	CA Tot	% CA	# R	# Min	# Maj	# Vmj
VITEK 2 Automatic Dilution- Clinical and Challenge												
***C. albicans* (Breakpoints (µg/mL): ≤2 S, 4 I, ≥8 R)**												
Clinical	174	165	94.8	21	12	57.1	164	94.3	3	5	5	0
Challenge	52	51	98.1	5	4	80.0	50	96.2	3	1	1	0
Total	226	216	95.6	26	16	61.5	214	94.7	6	6	6	0
***C. parapsilosis* (Breakpoints (µg/mL): ≤2 S, 4 I, ≥8 R)**												
Clinical	99	98	99.0	30	29	96.7	93	93.9	14	3	1	2
Challenge	10	10	100.0	2	2	100.0	10	100.0	0	0	0	0
Total	109	108	99.1	32	31	96.9	103	94.5	14	3	1	2
***C. tropicalis* (Breakpoints (µg/mL): ≤2 S, 4 I, ≥8 R)**												
Clinical	94	89	94.7	33	28	84.9	86	91.5	5	4	4	0
Challenge	13	12	92.3	7	6	85.7	11	84.6	1	1	1	0
Total	107	101	94.4	40	34	85.0	97	90.7	6	5	5	0
***Candida* spp. (all)**												
Clinical	367	352	95.9	84	78	92.3	343	93.5	22	12	10	2
Challenge	75	73	97.3	14	12	85.7	71	94.7	4	2	2	0
Total	**442**	**425**	**96.2**	**98**	**81**	**82.7**	**414**	**93.7**	**26**	**14**	**12**	**2**

EA – Essential Agreement maj – major discrepancies
CA – Category Agreement vmj – very major discrepancies
R – resistant isolates min – minor discrepancies

Clinical performance was acceptable. Due to performances obtained with *C. parapsilosis*, the following limitation statement was added to the package insert:

> "Perform an alternative method of testing prior to reporting of results for the following antibiotic/organism combination(s):
> - Fluconazole: *Candida parapsilosis* when the MIC is 1 or 2 µg/mL"

To offer a better understanding of this issue by the end-user, FDA also agreed with the sponsor to add the following statement as a footnote to the performance table in the device labeling to address the Very Major Errors for *C. parapsilosis*:

"An overall essential agreement rate of 99.1% and an overall category agreement rate of 94.5% were observed for *Candida parapsilosis* when tested with VITEK 2 Fluconazole. Compared to the reference broth microdilution, two of 14 results for *Candida parapsilosis* (one of which was in essential agreement) resulted in very major errors."

The sponsor added a limitation in their labeling addressing the major errors observed for *C. tropicalis:*

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

- Fluconazole (flu02n): *Candida tropicalis"*

MIC Trends:
An analysis of trending was conducted using the combined clinical and challenge data. This trending calculation considers MIC values that are determined to be one or more doubling dilutions lower or higher compared to the reference method irrespective of whether the device MIC values are on-scale or not. Results that are not clearly at least one dilution lower, at least one dilution higher or in exact agreement with the CLSI reference method are not considered in the trending analysis.

Species for which the difference between the percentage of isolates with higher vs. lower MIC reading was ≥ 30% and for which the confidence interval was determined to be statistically significant were considered to show evidence of trending. Trending that provides higher or lower MIC values compared to the reference is addressed in labeling.
A trend toward lower MIC readings was observed for all *C. albicans, C parapsilosis, and C. tropicalis* when compared to the CLSI broth microdilution reference method, as summarized in Table 3. The following statement is included as a footnote to the performance table in the device labeling to address the observed trending:

"VITEK 2 Fluconazole MIC values tended to be in exact agreement or at least one doubling dilution higher when testing *C. albicans, C parapsilosis, and C. tropicalis* compared to the broth microdilution reference method."

Table 3. Trending Analysis for VITEK 2 Fluconazole.

Trending Analysis VITEK 2 Fluconazole (Clinical and Challenge Isolates) Automatic Dilution						
Organism	**Total Evaluable for Trending**	**≥ 1 Dilution lower No. (%)**	**Exact No. (%)**	**≥ 1 Dilution Higher No. (%)**	**Percent Difference (CI)**	**Trending Noted**
C. albicans	52	13 (25.0%)	3 (5.8%)	36 (69.2%)	44.2% (25.4%, 58.8%)	Yes
C. parapsilosis	45	7 (15.6%)	4 (8.9%)	34 (75.6%)	60.0% (40.6%, 72.6%)	Yes
C. tropicalis	71	6 (8.5%)	5 (7.0%)	60 (77.4%)	7601% (62.6%, 84.1%)	Yes
All three *Candida* species.	168	26 (15.5%)	12 (7.1%)	130 (77.4%)	61.9% (52.6%, 69.3%)	Yes

2. Matrix Comparison:

N/A

C Clinical Studies:

1. Clinical Sensitivity:

   N/A

2. Clinical Specificity:

   N/A

3. Other Clinical Supportive Data (When 1. and 2. Are Not Applicable):

   N/A

D Clinical Cut-Off:

N/A

E Expected Values/Reference Range:

VITEK 2 AST-YST	Equivalent Standard Method Concentration by Efficacy in µg/mL	MIC Ranges and FDA Categories* (MIC in µg/mL)		
		S	I	R
Fluconazole	2, 4, 8, 16, 32, 64	*Candida spp.*		
		≤ 2	4	≥ 8

VIII Proposed Labeling:

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

IX Conclusion:

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.
To support the implementation of changes to FDA-recognized susceptibility test interpretive criteria (i.e., breakpoints), this submission 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 BioMerieux VITEK2 AST-Yeast Fluconazole (≤0.5 - ≥64 µg/mL) when revised breakpoints for Fluconazole are published on the FDA STIC webpage. The breakpoint change protocol included with the submission indicated that if specific criteria are met, BioMerieux will update the Fluconazole 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.