



**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION
DECISION SUMMARY
ASSAY ONLY**

I Background Information:

A 510(k) Number

K252114

B Applicant

Liofilchem s. r. l.

C Proprietary and Established Names

MTS Aztreonam-Avibactam 0.016/4 - 256/4 µg/mL

D Regulatory Information

Product Code(s)	Classification	Regulation Section	Panel
JWY	Class II	21 CFR 866.1640 - Antimicrobial Susceptibility Test Powder	MI - Microbiology

II Submission/Device Overview:

A Purpose for Submission:

In this Traditional 510(k) submission, Liofilchem seeks the following:

1. To obtain a substantial equivalence determination for the Liofilchem MIC Test Strip (MTS) with aztreonam-avibactam at concentrations of 0.016/4 to 256/4 µg/mL.
2. To establish a Predetermined Change Control Plan (PCCP) to address future revisions to device labeling in response to breakpoint changes that are recognized on the FDA STIC webpage.

B Measurand:

Aztreonam-avibactam in the dilution range of 0.016/4 to 256/4 µg/mL

C Type of Test:

Quantitative antimicrobial susceptibility test (AST) growth-based detection

III Intended Use/Indications for Use:

A Intended Use(s):

See Indications for Use below.

B Indication(s) for Use:

The MTS (MIC Test Strip) Aztreonam-Avibactam 0.016/4-256/4 µg/mL is a quantitative method intended for *in vitro* determination of antimicrobial susceptibility of bacteria. MTS consists of specialized paper impregnated with a predefined concentration gradient of an antimicrobial agent, which is used to determine the minimum inhibitory concentration (MIC) in µg/mL of antimicrobial agents against bacteria as tested on agar media using overnight incubation and manual reading procedures. MTS Aztreonam-Avibactam at concentrations of 0.016/4-256/4 µg/mL should be interpreted at 16-20 hours of incubation.

Testing with MTS Aztreonam-Avibactam at concentrations of 0.016/4-256/4 µg/mL is indicated for Enterobacterales as recognized by the FDA Susceptibility Test Interpretive Criteria (STIC).

The MTS Aztreonam-Avibactam 0.016/4-256/4 µg/mL has demonstrated acceptable performance with the following organisms:

Enterobacterales (*Citrobacter freundii* complex, *Enterobacter cloacae* complex, *Escherichia coli*, *Klebsiella oxytoca*, *Klebsiella pneumoniae*, and *Serratia marcescens*)

C Special Conditions for Use Statement(s):

Rx - For Prescription Use Only

The ability of the MTS Aztreonam-Avibactam to detect the following resistant isolates is unknown because an insufficient number of resistant isolates were available at the time of comparative testing. If such a strain is observed, it should be submitted to a reference laboratory.

Aztreonam-Avibactam: K. pneumoniae, E. cloacae complex, C. freundii complex, K. oxytoca, and S. marcescens

D Special Instrument Requirements:

N/A – Manual reading only

IV Device/System Characteristics:

A Device Description:

MTS Aztreonam-Avibactam 0.016/4-256/4 µg/mL is made of special high-quality paper impregnated with avibactam at a fixed concentration of 4 µg/mL and a predefined concentration gradient of aztreonam across 15 two-fold dilutions like those used by conventional MIC methods. One side of the strip is labeled with the aztreonam-avibactam code (AZA) and the MIC

reading scale in µg/mL. MIC values are determined by identifying the drug concentration at which growth of the ellipse ends. The MIC Test Strip (MTS) is single use only.

B Principle of Operation:

MTS are made of specialized high-quality paper impregnated with a predefined concentration gradient of antibiotic, across 15 two-fold dilutions like those of a conventional MIC method. When the MTS is applied onto an inoculated agar surface, the preformed exponential gradient of antimicrobial agent diffuses into the agar. After 16-20 hours incubation, a symmetrical inhibition ellipse centered along the strip is formed. The MIC is read manually directly from the scale in terms of µg/mL at the point where the edge of the inhibition ellipse intersects the strip MTS.

Growth along the entire gradient (i.e., no inhibition ellipse) indicates that the MIC value is greater than or equal to (\geq) the highest value on the scale. An inhibition ellipse that intersects below the lower end of the scale is read as less than ($<$) the lowest value. An MIC of 0.125 µg/mL is considered to be the same as 0.12 µg/mL for reporting purposes.

An MTS MIC value which falls between standard two-fold dilutions must be rounded up to the next standard upper two-fold value before categorization.

V Substantial Equivalence Information:

A Predicate Device Name(s):

Liofilchem MIC Test Strip (MTS)-Vancomycin 0.016 -256 ug/mL

B Predicate 510(k) Number(s):

K153687

C Comparison with Predicate(s):

Device & Predicate Device(s):	Device: <u>K252114</u>	Predicate: <u>K153687</u>
Device Trade Name	MTS Aztreonam-Avibactam 0.016/4-256/4 µg/mL	Liofilchem MIC Test Strip (MTS)-Vancomycin 0.016-256 µg/mL
General Device Characteristic Similarities		
Plate Media	Mueller Hinton Agar	Same
MTS Strip Material	High quality paper impregnated with a predefined concentration of gradient antimicrobial agent	Same
Inoculation	Isolated colonies from culture in a suspension equivalent to 0.5 McFarland. Inoculum is applied to agar with swab manually.	Same
Reading	Manual; Interpret the MIC as	Same

Device & Predicate Device(s):	Device: <u>K252114</u>	Predicate: <u>K153687</u>
	100% inhibition	
Result	MIC in µg/mL	Same
General Device Characteristic Differences		
Intended Use/Indications for Use	Quantitative susceptibility to antimicrobial agents against specified gram-negative organisms	Quantitative susceptibility to antimicrobial agents against specified gram-positive organisms
Antimicrobial Agent	Aztreonam-Avibactam (AZA)	Vancomycin (VA)
Incubation	35°C ± 2°C for 16-20 hours	35°C ± 2°C for 24 hours

VI Standards/Guidance Documents Referenced:

- Class II Special Controls Guidance Document: *Antimicrobial Susceptibility Test (AST) Systems; Guidance for Industry and FDA* (August 2009).
- CLSI M07-Ed12. *Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria that Grow Aerobically* (March 2024).
- CLSI M100-Ed35. *Performance Standards for Antimicrobial Susceptibility Testing* (January 2025).

VII Performance Characteristics (if/when applicable):

A Analytical Performance:

1. Precision/Reproducibility:

Reproducibility testing of the Liofilchem MIC Test Strip (MTS) Aztreonam-Avibactam was performed using 10 Enterobacterales isolates (3 *K. pneumoniae*, 2 *E. coli*, 2 *E. cloacae*, 1 *C. freundii*, 1 *S. marcescens*, and 1 *K. oxytoca*). Testing was performed in triplicate at three sites on three separate days for a total of 270 data points (10 isolates x 3 replicates x 3 days of testing = 90 data points/site). Results were used to determine site to site and overall reproducibility.

The mode MIC value was pre-determined for each organism and the reproducibility was calculated based on the number of MIC values that fell within ± one doubling dilution of the mode. All MIC results were on scale. The results for overall reproducibility of MTS Aztreonam-Avibactam were 96.3% for all isolates of Enterobacterales that were within one doubling dilution of the mode MIC determined by the reference broth microdilution method. The results are 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):

Inoculum Density Check:

The inoculum is prepared from an overnight agar plate into saline to achieve turbidity equivalent to a 0.5 McFarland standard. The inoculum is applied to agar with a sterile swab manually. Colony counts are performed periodically at each site for all QC replicates.

Inoculum density checks were performed, and the colony counts obtained for each QC strain were within the recommended range of approximately 1×10^8 CFU/mL. Colony counts are also determined from one replicate of each reproducibility isolate on each of the three days of testing and from a minimum of 10% of the clinical strains tested and showed similar ranges.

Purity Checks:

Purity checks are performed on all isolates following MTS inoculation. All isolates were determined to be pure in both the broth microdilution reference panels and the MTS agar plates.

Growth Rate:

All clinical and challenge isolates grew in both the reference broth microdilution panels and the MTS agar plates.

Quality Control:

The CLSI-recommended quality control (QC) strain *Klebsiella pneumoniae* ATCC 700603 and CLSI QC strains *Escherichia coli* ATCC 25922 and *Escherichia coli* ATCC 35218 were tested at three sites for a minimum of 20 times at each site by both the MTS and the reference method. The results demonstrate that MTS Aztreonam-Avibactam can produce quality control results in the recommended range $\geq 95\%$ of the time which is acceptable (**Table 1**).

Table 1. QC Results for Aztreonam-Avibactam with CLSI Recommended QC Strains

QC Organism	Concentration (µg/mL)	Reference BMD (All Sites)	MTS (All Sites)
<i>E. coli</i> ATCC 25922 Expected Results: 0.03/4-0.12/4 µg/mL	0.015		
	0.03		15
	0.06	66	54
	0.12	3	
	0.25		
<i>E. coli</i> ATCC 35218	0.008		
	0.016	2	21

QC Organism	Concentration (µg/mL)	Reference BMD (All Sites)	MTS (All Sites)
Expected Results: 0.016/4-0.06/4 µg/mL	0.03	41	48
	0.06	26	
	0.12		
<i>K. pneumoniae</i> ATCC 700603 Expected Results: 0.06/4-0.5/4 µg/mL	0.03		
	0.06		
	0.12	14	21
	0.25	55	47
	0.5		1
	1		

ATCC=American Type Culture Collection

6. Detection Limit:

Not applicable.

7. Assay Cut-Off:

Not applicable.

B Comparison Studies:

1. Method Comparison with Predicate Device:

Results obtained with Liofilchem MTS with Aztreonam-Avibactam were compared to results obtained from frozen reference MIC panels. Reference MIC panels are prepared, tested, and interpreted as outlined in the CLSI document M07-Ed12.

Isolated colonies from an overnight agar plate were suspended in saline to achieve a 0.5 McFarland standard turbidity (approximately 10^8 CFU/mL). Testing conditions consisted of incubation of the inoculated Mueller Hinton agar plate in an inverted position at $35^\circ\text{C} \pm 2^\circ\text{C}$ for 16-20 hours. At the end of the appropriate incubation, the MIC value where the edge of the inhibition ellipse intersects the strip was compared to MIC results obtained with the CLSI reference broth microdilution method.

Clinical:

Clinical testing was performed at two external U.S. sites and one internal outside U.S. site with both MTS Aztreonam-Avibactam and the reference method. A total of 591 Enterobacterales clinical isolates were evaluated including: 227 *E. coli*, 219 *K. pneumoniae*, 55 *E. cloacae*, 30 *C. freundii*, 30 *K. oxytoca*, and 30 *S. marcescens* isolates. The clinical testing included 192 (32.5%) contemporary isolates (isolated no longer than 6 months prior to testing) and 399 (67.5%) stock isolates (isolated over 6 months prior to testing).

Challenge:

Challenge testing was performed at one external U.S. site. A total of 95 challenge isolates (25 *E. coli*, 25 *K. pneumoniae*, 15 *E. cloacae*, 10 *C. freundii*, 10 *K. oxytoca*, and 10 *S. marcescens*) were evaluated.

Results of MTS Aztreonam-Avibactam testing with clinical and challenge isolates are shown in **Table 2**.

Table 2. Overall Performance of MTS Aztreonam-Avibactam with Clinical and Challenge Isolates

	Tot	EA N	EA %	Eval Tot	Eval EA N	Eval EA %	CA Tot	CA %	No. R	No. S	min	major	vmj
Enterobacteriales [$\leq 4/4$ (S), $8/4$ (I), $\geq 16/4$ (R)]													
Clinical	591	570	96.5	555	534	96.2	583	98.7	10	578	7	1	0
Challenge	95	93	97.9	93	91	97.9	88	92.6	6	83	7	0	0
Total	686	663	96.7	648	625	96.5	671	97.8	16	661	14	1	0

EA – Essential Agreement

CA – Category Agreement

EVAL – Evaluable MIC results

S – Susceptible

min – Minor discrepancies

maj – Major discrepancies

vmj – Very major discrepancies

R – Resistant

Essential Agreement (EA) is when the Liofilchem MIC Test Strip (MTS) results agree exactly or within one doubling dilution of the reference broth microdilution results. Category Agreement (CA) is when the Liofilchem MIC Test Strip (MTS) result interpretation agrees exactly with the reference broth microdilution result interpretation.

For Enterobacteriales, the combined clinical and challenge results (686 isolates) were acceptable with an EA of 96.7% and CA of 97.8%. There were 14 minor errors, one major error (0.15%, 1/661), and no very major errors.

For clinical and challenge isolates tested with the Liofilchem MTS Aztreonam-Avibactam 0.016/4-256/4 µg/mL, the overall % EA and % CA meet the acceptance criteria.

MIC Trending Analysis:

Using the combined clinical and challenge data, an analysis of trending was conducted for all Enterobacteriales isolates (**Table 3**). 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.

Species for which the difference between the percentage of isolates with higher vs. lower MIC readings was $\geq 30\%$ 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.

No statistically significant trending with MTS Aztreonam-Avibactam (0.016/4-256/4 µg/mL) was observed when compared to the CLSI broth micro-dilution reference method, as summarized in **Table 3**.

Table 3. Observed Trending of Results Obtained with MTS Aztreonam-Avibactam

Organism Name	Total Evaluable for Trending	≥ 1 Dilution Lower No. (%)	Exact No.	≥ 1 Dilution Higher No. (%)	Percent Difference (95% CI)	Trending Noted
<i>C. freundii</i>	39	11, (28.2)	22	6, (15.4)	-13% (-30%, 6%)	No
<i>E. cloacae</i>	68	29, (42.7)	27	12, (17.7)	-25% (-39%, -10%)	No
<i>E. coli</i>	238	82, (34.5)	105	51, (21.4)	-13% (-21%, -5%)	No
<i>K. oxytoca</i>	39	12, (30.8)	19	8, (20.5)	-10% (-29%, 9%)	No
<i>K. pneumoniae</i>	241	65, (27.0)	101	75, (31.1)	4% (-4%, 12%)	No
<i>S. marcescens</i>	40	15, (37.5)	17	8, (20)	-18% (-36%, 2%)	No

Resistant Isolates:

A total of 686 clinical and challenge isolates were tested for Enterobacterales and 16 (2.3%) resistant isolates were available for testing. Due to the insufficient number of resistant *K. pneumoniae*, *E. cloacae*, *C. freundii*, *K. oxytoca*, and *S. marcescens* isolates evaluated, the following limitation is included in the device labeling:

The ability of the MTS Aztreonam-Avibactam to detect the following resistant isolates is unknown because an insufficient number of resistant isolates were available at the time of comparative testing. If such a strain is observed, it should be submitted to a reference laboratory.

Aztreonam-Avibactam: K. pneumoniae, E. cloacae complex, C. freundii complex, K. oxytoca, and S. marcescens

Resistance Mechanism in Challenge Isolates:

Challenge isolates of Enterobacterales harboring various molecular mechanisms of resistance were evaluated with MTS Aztreonam-Avibactam 0.016/4 - 256/4 µg/mL. The following mechanisms were evaluated: aac(3), aac(6'), aadA, aadB, ACT, AmpC, aph(3'), armA, ARR3, catA1, catB3, cmlA1, CMY, CTX-M, dfrA, DHA, ESBL, fosA, FOX-5, IMI-4, IMP, KPC, LEN, MBL, MIR-16, mph(A), msr(E), NDM, oqxAB, OXA, OXY, pbp3, rmtC, rmtF, SHV, SME, str, sul, TEM, tet, and VIM alleles.

Testing/Reporting Non-Indicated Species:

For this review, the interpretative criteria are applied to the organisms/organism groups according to the FDA STIC website. As required under 511A(2)(2)(B) of the Federal Food, Drug and Cosmetic Act, the following statement is added in the precautions section of labeling:

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 labelling for specific antimicrobial drugs provides the uses for which the antimicrobial drug is approved.

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 aztreonam-avibactam are listed in Table 4.

Table 4. FDA-Recognized Interpretive Criteria for Aztreonam-Avibactam

Organism	Minimum Inhibitory Concentration (µg/mL) ^a		
	Susceptible (S)	Intermediate (I)	Resistant (R)
Enterobacterales	≤ 4/4	8/4	≥ 16/4

^a According to the [FDA STIC Webpage, https://www.fda.gov/drugs/development-resources/fda-recognized-antimicrobial-susceptibility-test-interpretive-criteria](https://www.fda.gov/drugs/development-resources/fda-recognized-antimicrobial-susceptibility-test-interpretive-criteria)

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 Liofilchem intends to use to evaluate the MTS Aztreonam-Avibactam 0.016/4-256/4 µg/mL when revised breakpoints for aztreonam-avibactam are published on the FDA STIC webpage. The breakpoint change protocol included with the submission indicated that if specific criteria are met, Liofilchem will update the MTS Aztreonam-Avibactam 0.016/4-256/4 µg/mL 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.