

A comparison of Piperacillin-Tazobactam and Colistin ComASP™ (SensiTest) MIC to CLSI Broth Microdilution MIC for Gram Negative Challenge Isolates

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Abstract:

Background: The ComASP™ (previous product name was SensiTest) was recently developed by Liofilchem to provide a manual broth microdilution option for MIC testing of single antimicrobial agents using a 32-well dried (desiccated) panel. The ComASP Piperacillin-Tazobactam (P-T) includes a wide range of concentrations (0.008-128 µg/mL), which provides for testing of 2 isolates/panel. The ComASP Colistin is configured to test 4 isolates/panel (4 rows each containing 0.25-16 µg/mL). There have been challenges in testing piperacillin-tazobactam by some automated and gradient strip methods. There have also been difficulties in testing colistin by gradient strip methods and an overall lack of testing methods available for colistin. This study was performed as an initial evaluation at a single testing site using challenge organisms specific to each of the two agents.

Method: Each strain was tested once with frozen panels (CLSI reference) and by ComASP using the same inoculum in cation adjusted Mueller Hinton broth. Incubation conditions (ambient at 33-37°C for 16-20 hrs) and reading of the MIC were similar for both methods. The strains tested included *A. baumannii*, Enterobacteriaceae, *P. aeruginosa* and quality control strains. The colistin challenge set consisted of 52 strains, which were from the CDC resistant collections and included characterized strains with a range of colistin MIC results. The P-T challenge set consisted of 28 strains, which were from the LSI collection and were chosen because the majority of MIC results were near the breakpoints (between 16-≥128 µg/mL).



Results: Colistin and P-T agreement rates and error rates are shown in the table.

Antimicrobial Agent	n	EA	CA	Minor Error Rate
Colistin	52	98.1%	100%	0%
Piperacillin/tazobactam	28	100%	75%	25%

EA - essential agreement (within ± 1 dilution); CA - category agreement

Although essential agreement rate was 100% for P-T, the category agreement was 75% as a result of 7 strains with MICs that differed by one dilution and were near the breakpoints. **Conclusions:** There was excellent correlation of ComASP and reference broth microdilution MIC results for both piperacillin-tazobactam and colistin. Additional testing, with a larger number of isolates and testing sites, is warranted. The ComASP method is a simple broth microdilution MIC method that would provide an option, other than gradient diffusion, for testing of a single agent as a supplement to a clinical laboratory's automated system.

Methods

Piperacillin-Tazobactam	Colistin
Testing Site: Laboratory Specialists, Inc.	
MIC Method: Each study isolate and QC strain was tested once by Reference and Test Methods	
<p>Reference Method: Broth Microdilution (CLSI)¹</p> <p>Test Method: ComASP Piperacillin-Tazobactam³ (catalog no. 75003, Liofilchem (Waltham, MA))</p> <p>Contains piperacillin-tazobactam in concentrations of 0.008/4-128/4 µg/mL, 3.6 mL vials of CAMHB and sealing film. 2 Isolates can be tested/panel (each box contains 4 panels, 8 tests/box).</p>	<p>Reference Method: Broth Microdilution (CLSI)¹</p> <p>Test Methods: ComASP Colistin³ (catalog no. 75001, Liofilchem (Waltham, MA))</p> <p>Contains colistin in concentrations of 0.25-16 µg/mL, 3.6 mL vials of CAMHB and sealing film. 4 isolates can be tested/panel (each box contains 4 panels, 16 tests/box)</p>
	
<p>Clinical Isolates:</p> <p>28 Gram-negative bacteria: 2 <i>E. cloacae</i>, 9 <i>E. coli</i>, 2 <i>K. pneumoniae</i>, 1 <i>P. mirabilis</i>, 4 <i>A. baumannii</i>, 10 <i>P. aeruginosa</i>. LSI bank organisms were selected based on P-T MIC results and were biased to resistant isolates (13 isolates ≥128 µg/mL) and isolates with MIC results near the breakpoint (10 isolates 8-64 µg/mL). The remaining 5 isolates were susceptible to P-T.</p>	<p>52 Gram-negative bacteria: 1 <i>C. freundii</i>, 1 <i>C. koseri</i>, 4 <i>E. aerogenes</i>, 5 <i>E. cloacae</i>, 4 <i>E. coli</i>, <i>K. pneumoniae</i>, 2 <i>M. morgani</i>, 2 <i>P. mirabilis</i>, 1 <i>P. rettgeri</i>, 1 <i>P. stuartii</i>, 2 <i>S. marcescens</i>, 4 <i>A. baumannii</i>, 10 <i>P. aeruginosa</i></p> <p>CDC challenge organisms were selected based on species and range of colistin MIC results and the number of isolates with molecular characterized resistance mechanisms were: 3 OXA, 4 cAmpC, 8 KPC, 5 NDM, 4 VIM, 2 SME, 3 CTX-M, 2 mcr-1 and 2 OMP</p>
<p>Quality Control Isolates:</p> <ul style="list-style-type: none"> <i>E. coli</i> ATCC 25922 <i>Escherichia coli</i> ATCC 35218 <i>K. pneumoniae</i> ATCC 700603 <i>P. aeruginosa</i> ATCC 27853 	<ul style="list-style-type: none"> <i>E. coli</i> ATCC 25922 <i>P. aeruginosa</i> ATCC 27853

Results

- Quality Control:** All P-T and Colistin ComASP and Reference BMD MIC results were within established CLSI ranges
- Piperacillin/Tazobactam (Figure 1):** P-T ComASP MIC results were within +/- one doubling dilution for 28/28 isolates (100% essential agreement). The bias in selecting strains near the breakpoint, resulted in a category agreement of 75%, however, the excellent essential agreement is the best indication of performance in this study. There was no definitive trending of lower or higher MIC results.
- Colistin (Figure 2):** Colistin ComASP MIC results were within +/- one doubling dilution for 51/52 isolates (98.1% essential agreement). Category agreement was 100%. The trending that was observed (slightly higher ComASP results) occurred at lower MICs (≤0.25-1 µg/mL)

Figure 1. Piperacillin/tazobactam ComASP MIC compared to BMD MIC (µg/mL) for 28 Gram negative challenge isolates

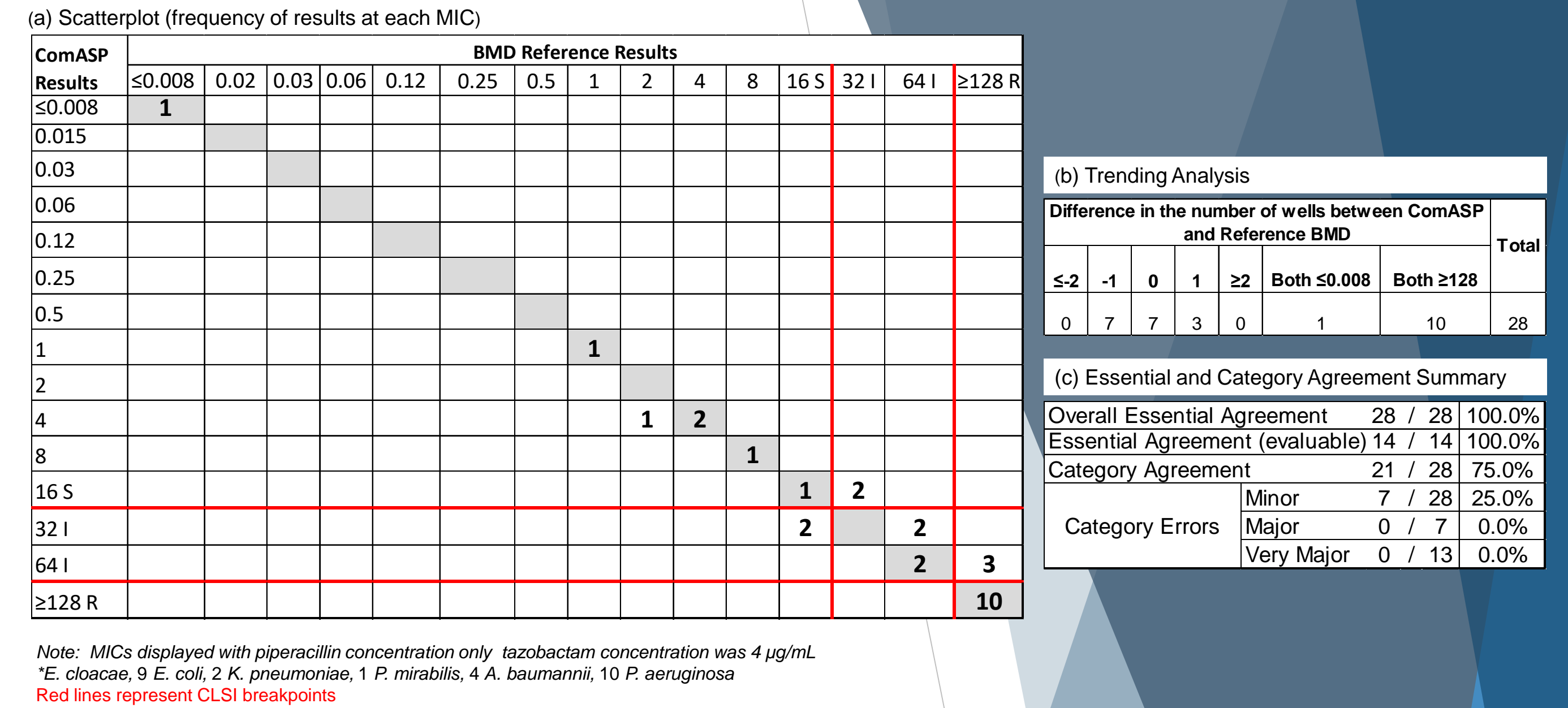
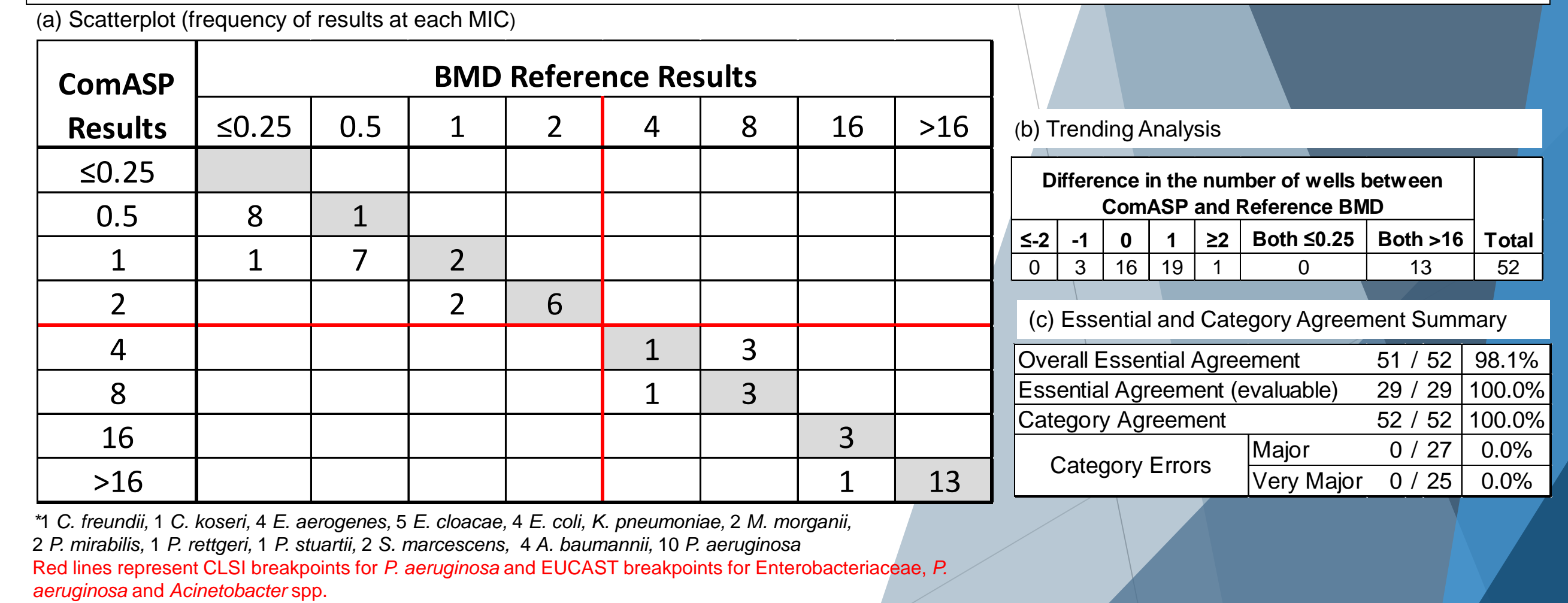


Figure 2. Colistin ComASP MIC compared to BMD MIC (µg/mL) for 52 Gram negative challenge isolates*



Conclusions

- The ComASP Piperacillin-Tazobactam compared similar to reference BMD against a challenge set of Enterobacteriaceae, *P. aeruginosa* and *A. baumannii* isolates. Additional multi-site studies and/or a 510(k) study is warranted.
- The ComASP Colistin compared similar to reference BMD against a challenge set of Enterobacteriaceae, *P. aeruginosa* and *A. baumannii* isolates. Additional multi-site studies and/or a 510(k) study is warranted, however, as a result of the lack of FDA colistin breakpoints, a 510(k) submission is not possible at this time.

References:

- Clinical and Laboratory Standards Institute. 2015. Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria that Grow Aerobically. 10th ed. Approved standard, CLSI M7-10, Wayne, PA.
- Clinical and Laboratory Standards Institute. 2017. Performance Standards for Antimicrobial Susceptibility Testing. Approved Standard – 27th Edition. CLSI document M100-276 Wayne, PA
- https://www.liofilchem.com/images/brochure/ComASP-range.pdf