**INTRODUCTION**

Liofilchem® MIC Test Strip is a quantitative method intended for the *in vitro* determination of antimicrobial susceptibility of non-fastidious Gram negative and Gram positive aerobic bacteria (for example, Enterobacteriaceae, *Pseudomonas, Enterococcus* and *Staphylococcus* species) and fastidious bacteria (for example, anaerobes, *Haemophilus* and *Streptococcus* species and *N. gonorrhoeae*). MIC Test Strip 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.

**Ceftolozane-tazobactam** (Zerbaxa) is a combination product consisting of a cephalosporin-class antibacterial drug and a β-lactamase inhibitor indicated for the treatment of the following infections caused by designated susceptible microorganisms:

- Complicated Intra-abdominal Infections, used in combination with metronidazole;
- Complicated Urinary Tract Infections, including Pyelonephritis.

**Ceftolozane-tazobactam MIC Test Strip** generates a stable concentration gradient for ceftolozane (0.016-256 µg/mL) in the presence of a fixed concentration of tazobactam (4 µg/mL). It is available in packages of 10, 30 and 100 tests:

- The 10-test box contains 10 strips individually packed in desiccant envelopes and an instruction sheet
- The 30-test box contains 30 strips individually packed in desiccant envelopes and an instruction sheet
- The 100-test box contains 10 desiccant envelopes, each containing 10 strips, and an instruction sheet; this pack contains a storage tube as well.

**TEST PROCEDURE**

Before using Ceftolozane-tazobactam MIC Test Strip from an unopened package, visually inspect to ensure the package is intact. Do not use the strips if the package has been damaged.

When removed from the refrigerator or freezer, allow the package or storage container to reach room temperature for about 30 minutes. Moisture condensing on the outer surface must evaporate completely before opening the package.

**Materials required but not provided:**

- Mueller Hinton II Agar, 90 (ref. 10031) or 140 mm (ref. 10231) plates
- Sterile saline (0.85% NaCl) (ref. 20095)
- Sterile loops, swabs (not too tightly spun), test tubes, pipettes and scissors
- Forceps
- 0.5 McFarland turbidity standard (ref. 80400)
- Incubator (35 ± 2°C)
- Quality control organisms
- Additional technical information from www.liofilchem.net

**Inoculum preparation**

Suspend well-isolated colonies from an overnight agar plate into saline to achieve a 0.5 McFarland standard turbidity. A confluent or almost confluent lawn of growth will be obtained after incubation, if the inoculum is correct.

In order to verify that your procedure gives the correct inoculum density in terms of CFU/mL, performing regular colony counts is recommended. A confluent or almost confluent lawn of growth will be obtained after incubation, if the inoculum is correct.

**Inoculation**

Apply the strip to the agar surface with the scale facing upwards and code of the strip to the outside of the plate, pressing it with a sterile forceps on the surface of the agar and ensure that whole length of the antibiotic gradient is in complete contact with the agar surface. Once applied, do not move the strip.

**Incubation**

Incubate the agar plates in an inverted position at 35 ± 2°C for 16-20 hours in ambient atmosphere. Extend the incubation for up to 48 hours in case of slow growing organisms.

**EVALUATING THE RESULTS**

**Reading**

Observe where the relevant inhibition ellipse intersects the strip and read the MIC at complete inhibition. Growth along the entire gradient i.e. no inhibition ellipse indicates that the value is greater than or equal to (≥) 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.

**Interpretation**

The susceptibility interpretative criteria recommended by the CLSI and EUCAST are shown below. Always round up MIC Test Strip half dilution values to the next upper two-fold value before categorization. For example a *E. coli* ceftolozane-tazobactam MIC of 0.19 µg/mL is reported as 0.25 µg/mL.

See page 2 for example of results. Also consult the MIC Test Strip Photographic Guide.

**QUALITY CONTROL**

CLSI-recommended quality control strains are used as outlined under TEST PROCEDURE. Repeat the test if any MIC result is out of the quality control range. If three consecutive failures occur, please, contact the Manufacturer at liofilchem@liofilchem.net

<table>
<thead>
<tr>
<th>Organism</th>
<th>Breakpoint (µg/mL)</th>
<th>Quality Control MIC Range (µg/mL)</th>
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<td>EUCAST</td>
</tr>
<tr>
<td></td>
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<td>I</td>
</tr>
<tr>
<td>Enterobacteriaceae</td>
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<td>4</td>
</tr>
<tr>
<td>Pseudomonas aeruginosa</td>
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<td>Streptococcus spp Viridans Group</td>
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<td>16</td>
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</table>

**STORAGE**

The unopened package of Ceftolozane-tazobactam MIC Test Strip should be stored at −20°C until the given expiry date. Leftover MIC Test Strip from an opened package must be stored at 2-8°C in the airtight tube, containing desiccant, provided in the pack for no more than 7 days. Do not store near sources of heat and do not expose to excessive temperature variations.
Ceftolozane-tazobactam MIC Test Strip Reading Guide

**Fig. 1**

**MIC 0.25 μg/mL**

**Fig. 2**

**MIC 0.5 μg/mL**

**Fig. 3**

**MIC 0.75 μg/mL, reported as 1 μg/mL**

**Fig. 4**

**MIC 32 μg/mL**

**REFERENCES**


<table>
<thead>
<tr>
<th>PRESENTATION</th>
<th>µg/mL</th>
<th>Code</th>
<th>Packaging</th>
<th>Ref.</th>
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<tbody>
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