Solithromycin MIC Test Strip Technical Sheet

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 pre-defined 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.

Solithromycin (Solithera) is a fourth generation macrolide, the first fluoroketolide, which has potent activity against most macrolide-resistant bacteria including multidrug-resistant *Streptococcus pneumoniae*. It is also active against other organisms responsible for bacterial pneumonia such as *Haemophilus influenzae, Moraxella catarrhalis*, methicillin-susceptible *Staphylococcus aureus*, *Legionella pneumophila* and *Mycoplasma pneumoniae*. The proposed indication is the treatment of moderate to moderately-severe community acquired bacterial pneumonia (CABP).

Solithromycin MIC Test Strip generates a stable gradient of antibiotic giving an accurate MIC over the range 0.002-32 µg/mL. It is available in packages of 10, 30 and 100 tests:

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

TEST PROCEDURE

Before using Solithromycin 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:

- Agar plate medium, the choice depending on the organism under investigation, e.g. MH II agar (ref. 10031), MH II agar with 5% sheep blood (ref. 10131), Haemophilus Test agar (ref. 10080), MH-F agar (ref. 10132)
- Sterile saline (ref. 20095), Mueller Hinton broth (ref. 24107)
- Sterile loops, swabs (not too tightly spun), test tubes, pipettes and scissors
- Forceps
- McFarland turbidity standard set (ref. 80405)
- 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/broth to achieve the desired 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.

Inoculation

Dip a sterile swab in the broth culture or in a diluted form thereof and squeeze it on the wall of the test tube to eliminate excess liquid.
Alternatively, use a rotation plater to efficiently streak the inoculum over the agar surface. Allow excess moisture to be absorbed so that the surface is completely dry before applying MIC Test Strip.

Application

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 16-24 hours, in ambient air or CO_2-enriched atmosphere. Incubate nonfastidious organisms and *Streptococcus pneumoniae* in ambient air (WITHOUT CO_2). Other fastidious organisms like *Haemophilus influenzae* should be incubated with CO_2. As with other antibiotics in the macrolide group, the *in vitro* activity of solithromycin can be affected by the decrease in pH that results when incubating agar based tests in 5% carbon dioxide. 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 80% 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

Clinical MIC breakpoints for solithromycin have not been established yet. Before categorization always round up MIC Test Strip half dilution values to the next upper two-fold value. For example a *H. influenzae* solithromycin MIC of 1.5 µg/mL is reported as 2 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.

Quality Control MIC Range (µg/mL)	
S. aureus ATCC® 29213	0.03-0.12
E. faecalis ATCC® 29212	0.015-0.06
H. influenzae ATCC® 49247	1-4
S. pneumoniae ATCC® 49619	0.004-0.015[a]

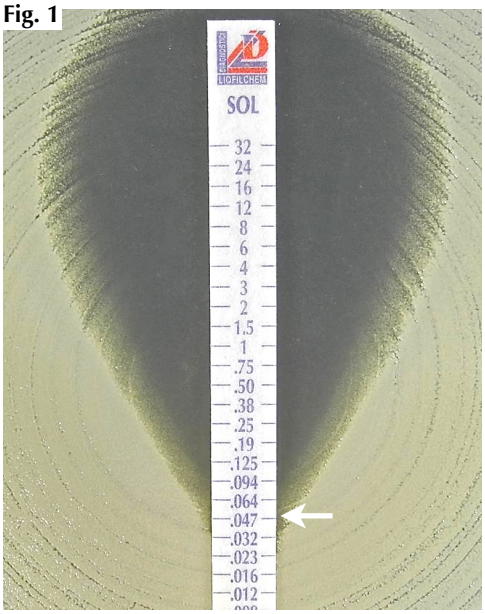
[a] QC range for ambient incubation

STORAGE

The unopened package of Solithromycin MIC Test Strip may be stored at temperature equal or below 8°C but is best 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.

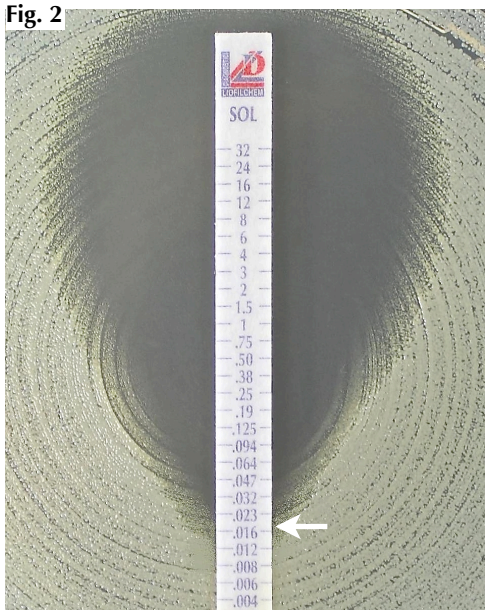
Solithromycin MIC Test Strip Reading Guide

Fig. 1



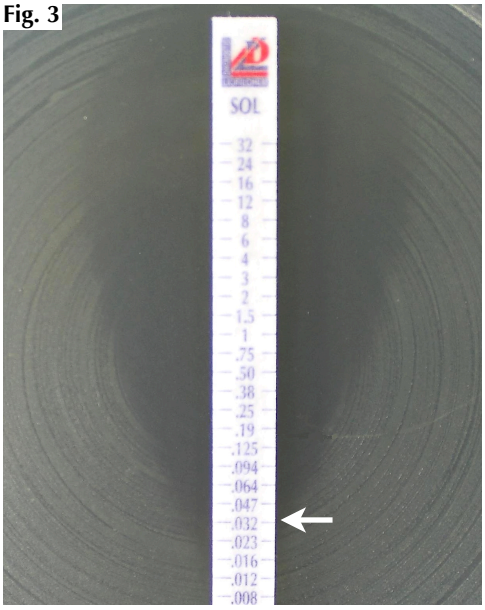
Read at 80% inhibition.
MIC 0.047 µg/mL, reported as 0.064 µg/mL

Fig. 2



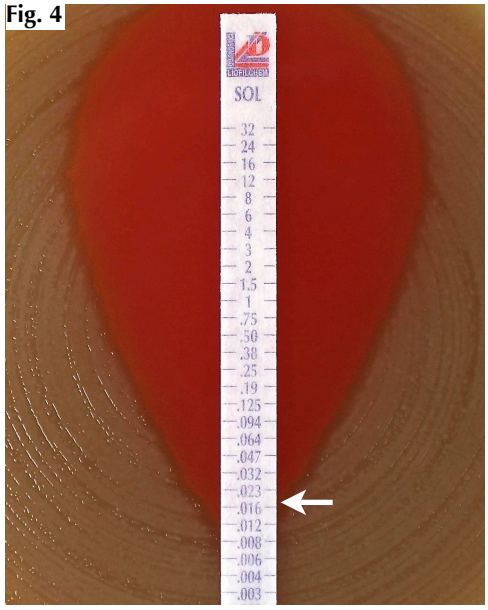
Read at 80% inhibition. MIC 0.016 µg/mL

Fig. 3



Read at 80% inhibition. MIC 0.032 µg/mL

Fig. 4



MIC 0.016 µg/mL

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- Barrera CM *et al*. (2016) Efficacy and safety of oral solithromycin versus oral moxifloxacin for treatment of community-acquired bacterial pneumonia: a global, double-blind, multicentre, randomised, active-controlled, non-inferiority trial (SOLITAIRE-ORAL). Lancet Infect Dis; 16:421-30.
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- CLSI M07-A10 (2015) Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria That Grow Aerobically: Approved Standard – Tenth Edition.
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- Waites KB *et al*. (2009) Comparative in vitro susceptibilities of human mycoplasmas and ureaplasmas to a new investigational ketolide, CEM-101. Antimicrob Agents Chemother; 53:2139-41.

PRESENTATION		µg/mL	Code	Packaging	Ref.
MIC Test Strip	Solithromycin	0.002-32	SOL	10 30 100	921431 92143 921430

MIC Test Strip, International Patent

LIOFILCHEM® s.r.l.

Via Scozia zona ind.le, 64026 Roseto degli Abruzzi (Te) Italy
Tel. +39 0858930745 Fax +39 0858930330 www.liofilchem.net liofilchem@liofilchem.net

Research **U**se **O**nly