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Delafloxacin: activity against fastidious organisms tested by EUCAST vs CLSI methodology

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ABSTRACT updated

Background: Delafloxacin (DLX) is a new fluoroquinolone with activity against a broad spectrum of Gram-negative and Gram-positive pathogens, including methicillin-resistant *Staphylococcus aureus* (MRSA), atypicals and anaerobes.
DLX is FDA approved and under evaluation by EMA for the treatment of acute bacterial skin and skin structure infections. For fastidious organisms, CLSI and EUCAST MIC methods use different media. The study assessed the DLX susceptibility of 899 isolates of nine fastidious species from DLX Surveillance within the SENTRY Antimicrobial Surveillance Program using EUCAST method and compared the data with those previously obtained with CLSI method.

Methods: MICs were determined at three different laboratories using Mueller-Hinton Fastidious (MH-F) broth panels manufactured at each laboratory according to the EUCAST Media Preparation v5.0, 2017. As no quality control (QC) ranges are available from EUCAST for DLX, Levofloxacin (LVX) was tested for QC. For quality assurance purposes, *Streptococcus pneumoniae* ATCC 49619, *Haemophilus influenzae* ATCC 49247, and *Haemophilus influenzae* ATCC 49766 were tested across the laboratories.

Results: DLX MIC results were overall similar by EUCAST and CLSI methods, with differences usually within 1 doubling dilution.
With most species, DLX MIC results by the CLSI method were slightly higher than those by the EUCAST method.
All tested species were highly susceptible to DLX. No significant differences were observed between the results obtained by different laboratories with QC strains.

Conclusions: The results of this study showed an overall good correlation between the two methods for the nine species studied.

INTRODUCTION

DLX has been approved by FDA.

The data from DLX preclinical profiling studies and surveillances studies for fastidious pathogens were previously generated according to CLSI guidelines for MIC determination, that use a different medium from that recommended by EUCAST.

Since DLX is now under evaluation by EMA we compared the MIC values obtained using the EUCAST methodology with those previously obtained using CLSI methodology on a large collection of fastidious pathogens.

The aim of this exercise was to compare the MIC values obtained using EUCAST methodology with the MIC values previously obtained, on the same strains, using CLSI methodology.

REFERENCES

1. EUCAST Media for MIC Determination by the Broth Microdilution Method. Version 5.0, January, 2017
2. CLSI M100-S27, 2017. Performance Standards for Antimicrobial Susceptibility Testing.
3. ISO 20776-1, Clinical laboratory testing and *in vitro* diagnostic test systems- Part 1: Reference method for testing the *in vitro* activity of antimicrobial agents against rapidly growing aerobic bacteria involved in infectious diseases
4. EUCAST routine and extended internal quality control for MIC determination and disk diffusion as recommended by EUCAST. Version 7.0, 2017.

METHODS

Testing Sites: Department of Laboratory Sciences and Infectious Diseases Università Cattolica del S. Cuore, Rome, Italy (UCSC); Department of Experimental Clinical Medicine – Laboratory of Clinical Microbiology University of Florence (UNIFI); JMI Laboratories IA, USA (JMI).

Isolates: 899 isolates of nine fastidious species were randomly chosen from DLX Surveillance Program and/or SENTRY Antimicrobial Surveillance Program.

MIC method: Mueller-Hinton Fastidious (MH-F) broth panels were manufactured at each laboratory according to the EUCAST Media Preparation v5.0, 2017.
Procedure for reference MIC panel preparation, method of panel inoculation, incubation and reading were according to ISO 20776-1 standards.

LVX was tested as QC antibiotic, as no quality control (QC) ranges are available from EUCAST for DLX. *Streptococcus pneumoniae* ATCC 49619, *Haemophilus influenzae* ATCC 49247, and *Haemophilus influenzae* ATCC 49766 were tested across the laboratories, for quality assurance purposes.

Bridging study: considering the three different panel production processes, a selection of 12 strains of three different species (*Streptococcus pneumoniae*, *Haemophilus influenzae* and *Moraxella catarrhalis*) were tested across all 3 different labs.

Table1: Delafloxacin results by used methods

Species	Isolates (N)	MIC_{90} (mg/mL)		MIC_{50} (mg/mL)		Range (mg/mL)	
		EUCAST	CLSI	EUCAST	CLSI	EUCAST	CLSI
H. influenzae	101	0.001	0.002	0.00025	0.001	0.00025-0.002	0.000125-0.004
H. parainfluenzae	98	0.125	0.25	0.004	0.008	0.0005->0.25	0.0005->0.25
M. catharralis	98	0.004	0.008	0.002	0.004	0.00025-0.008	≤0.001-0.016
S. agalactiae	100	0.03	0.03	0.008	0.016	≤0.002-0.5	≤0.004-0.5
S. anginosus group	105	0.008	0.03	0.004	0.008	0.0005-0.125	≤0.002-0.06
S. dysgalactiae	100	0.016	0.03	0.008	0.016	≤0.002-0.125	≤0.004-0.25
S. mitis group	101	0.06	0.06	0.016	0.03	0.004-1	0.008-0.5
S. pneumoniae	97	0.016	0.03	0.004	0.016	≤0.002-0.5	≤0.004-0.5
S. pyogenes	99	0.016	0.03	0.008	0.008	≤0.002-0.03	≤0.004->0.03

Figure 1: *S. pyogenes, S. pneumoniae, H. influenzae* Delafloxacin MIC distributions

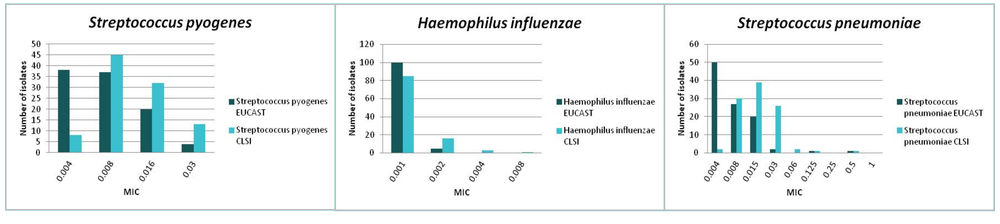

Table 2: Protocol results for ATCC in 3 laboratories JMI, UNIFI and UCSC – Quality control

QC Strains	Species	LAB	Delafloxacin EUCAST	Levofloxacin EUCAST
ATCC 49247	*Haemophilus influenzae*	JMI	0.00025 - 0.0005	0.015 - 0.03
		UNIFI	0.00025	0.008
		UCSC	0.00025	0.008 - 0.015
ATCC 49619	*Streptococcus pneumoniae*	JMI	0.008	1
		UNIFI	0.004	0.5
		UCSC	0.004 - 0.008	0.5
ATCC 49766	*Haemophilus influenzae*	JMI	0.00025 - 0.0005	0.015 - 0.03
		UNIFI	0.00025	0.008
		UCSC	0.00025 - 0.0005	0.008

Table 3: Bridge study- results of common strains tested

Strains	Organism	DLX JMI	DLX UNIFI	DLX UCSC	LVX JMI	LVX UNIFI	LVX UCSC
898823	*Streptococcus pneumoniae*	0.015	0.004	0.004	1	1	1
917146	*Streptococcus pneumoniae*	0.008	0.004	0.004	1	1	1
948424	*Streptococcus pneumoniae*	0.008	0.004	0.004	1	0.5	0.5
956829	*Haemophilus influenzae*	0.00025	≤ 0.0001	0.00025	0.015	0.008	0.008
967559	*Moraxella catarrhalis*	0.001	0.002	0.002	0.015	0.015	0.03
970251	*Haemophilus influenzae*	0.002	0.0005	0.0005	0.03	0.015	0.008
970259	*Haemophilus influenzae*	0.002	0.0005	0.0005	0.03	0.015	0.008
972070	*Moraxella catarrhalis*	0.004	0.001	0.002	0.06	0.03	0.03
975800	*Haemophilus influenzae*	0.0005	0.0002	0.00025	0.015	0.008	0.008
976041	*Moraxella catarrhalis*	0.06	0.008	0.015	2	1	1
980411	*Streptococcus pneumoniae*	0.008	0.004	0.004	1	0.5	0.5
985507	*Moraxella catarrhalis*	0.002	0.001	0.001	0.03	0.015	0.015

RESULTS

DLX MIC results were overall similar by EUCAST and CLSI methods, with differences usually within 1 doubling dilution and a trend of MICs obtained by the CLSI methodology at being slightly higher than those obtained by the EUCAST methodology (Table 1). Should be noted that the comparison with CLSI data was carried out retrospectively and not generated using the same inoculum.

All tested species were highly susceptible to DLX (Table 1).

All QC results for LVX were within the accepted QC ranges (Table2).

The bridging study showed an overall good interlaboratory agreement (Table 3).

CONCLUSIONS

Delafloxacin showed potent in vitro antibacterial activity against the 899 strains of nine fastidious species.
The results showed an overall good correlation between the two methods for the nine species studied. Usually, the MIC values obtained with the EUCAST methodology were equal or lower than those obtained with the CLSI methodology.