E-101, a novel first in class topical anti-infective, has potent activity against clinical isolates of important pathogens in Europe collected from 2008-2010

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ABSTRACT

Objectives: E-101 is a novel topical antimicrobial developed in Europe for the prevention of surgical site infections (SSI). Its unique mechanism of action utilizes myeloperoxidase (MPO) to produce reactive oxygen species that kill bacteria locally. As with any new agent, it is important to understand its current activity profile and to monitor for changes in that profile that may indicate the emergence of resistance. This study reports the *in vitro* activity of E-101 against European isolates of SSI pathogens alongside key comparators (currently utilized agents and important phenotypic markers).

Methods: 260 non-duplicate clinical isolates of *S. aureus*, coagulase-negative staphylococci (CoNS), enterococci, enterics (*E. coli*, *K. pneumoniae*, *P. mirabilis*, and *E. cloacae*), and *P. aeruginosa* collected from 27 sites across 8 countries in Europe were evaluated. E-101 was evaluated using a modified broth microdilution method based on CLSI guidelines (M7) in which MPO was serially diluted and inocula were delivered in enzyme substrate. E-101 MICs represent mg/L of MPO. Comparator agents were evaluated in accordance with CLSI M7 and M100.

Results: E-101 had an MIC50 and MIC90 of 0.015 mg/L against *S. aureus* and CoNS. Among staphylococci, 18% of *S. aureus* and 77% of CoNS were methicillin resistant, while 100% were susceptible to vancomycin, linezolid, and daptomycin. E-101 had an MIC50/MIC90 of 0.06/0.12 mg/L against *E. faecium* (13% vancomycin resistant) and 0.25/0.25 mg/L against *E. faecalis* (no vancomycin resistance). Against enterics, E-101 had similar activity by MIC50/MIC90 (mg/L) across evaluated species (*E. coli*: 0.06/0.06, *K. pneumoniae*: 0.12/0.12, *P. mirabilis*: 0.03/0.06, *E. cloacae*: 0.06/0.12). Among enterics, resistance to imipenem was not observed and resistance to ceftazidime varied by species (3% for *E. coli* and *P. mirabilis*, 14% for *K. pneumoniae*, and 20% for *E. cloacae*). Of the evaluated *P. aeruginosa*, resistance to most comparators (gentamicin, imipenem, levofloxacin, piperacillin/tazobactam) was 10-20%, and E-101 had an MIC50/MIC90 of 0.03/0.06 mg/L.

Conclusions: Based on the *in vitro* activity profile, E-101 has potent activity against European clinical isolates of both Gram-positive and Gram-negative pathogens commonly associated with SSI. There is no apparent impact of current resistance common among these species on the activity profile of E-101. These data illustrate the potential of E-101 for the prevention of SSI.

BACKGROUND

- E-101 is a novel myeloperoxidase (MPO) mediated therapeutic currently undergoing clinical development for the prevention of surgical site infections (SSI) in Europe and the US
- Upon activation and in the presence of halide and H_2O_2, MPO generates reactive oxygen species which damage bacterial cells
- As part of its development, it is important to understand the *in vitro* activity profile of E-101 against pathogens for which it is expected to be active (SSI pathogens)
- This study evaluates the *in vitro* activity of E-101 against a variety of recent and clinically relevant Gram-positive and Gram-negative pathogens collected across Europe

METHODS

- Non-duplicate, non-consecutive, clinically relevant isolates of the indicated pathogens (Tables 1 and 2) were collected from 35 sites across 9 European countries from 2008-2010 and chosen at random for this study
- Susceptibility of isolates to E-101 was determined using a modified broth microdilution method based on CLSI M7 guidelines. Modifications included diluting E-101 enzyme solution containing MPO in 2x cation-adjusted Mueller-Hinton broth in the panel, and delivering the inoculum at 2x final concentration in 2x substrate solution to achieve a final concentration of 1x E-101, 1x substrate solution, and 5 x 10^5 CFU/mL. Immediately post-inoculation, E-101 begins to generate reactive oxygen species. MICs are reported based on mg/L MPO in E-101.
- Concurrent inocula were tested against relevant comparators by broth microdilution in accordance with CLSI M7

RESULTS

Staphylococci

- E-101 had potent MICs against both *S. aureus* and coagulase-negative Staphylococci (CoNS) with an MIC_{50} and MIC_{90} of 0.015 mg/L
- Among evaluated staphylococci, 18% of *S. aureus* were MRSA and 77% of CoNS were MRCoNS
- Methicillin resistance among evaluated Staphylococci had no impact on E-101 activity

Enterococci

- E-101 was active against both *E. faecalis* and *E. faecium*, but by MIC_{50}/MIC_{90} (mg/L) was slightly more active against *E. faecium* (0.06/0.12) than *E. faecalis* (0.25/0.25)
- Vancomycin non-susceptible (VAN NS) enterococci were only detected among *E. faecium* (31% VAN NS)
- Reduced vancomycin susceptibility among evaluated *E. faecium* had no impact on E-101 activity

Enterobacteriaceae

- By MIC_{90} (mg/L), E-101 activity ranged from 0.06 against *E. coli* and *P. mirabilis* to 0.12 against *K. pneumoniae* and *E. cloacae*
- Among evaluated isolates, 10% were ceftazidime resistant (CAZ R), and CAZ R rates were variable by species
- Resistance to ceftazidime among evaluated Enterobacteriaceae had no impact on E-101 activity

P. aeruginosa

- E-101 had an MIC_{50} of 0.03 mg/L and MIC_{90} of 0.06 mg/L against evaluated isolates
- The evaluated isolates were 10-25% resistant to the majority of the evaluated comparator agents, and 23% were multi-drug resistant (MDR)
- E-101 had identical activity against MDR isolates relative to non-MDR isolates

TABLE 1. Activity of E-101 and comparators against European Gram-positive clinical isolates

Organism	Drug	MIC (mg/L) MIC_{50}	MIC_{90}	%S	%R
S. aureus (n=50)	E-101	0.015	0.015	⁜	⁜
	Oxacillin	0.25	>4	82.0	18.0
	Cefazolin	0.5	2	94.0	6.0
	Ciprofloxacin	0.25	>4	78.0	18.0
	Clindamycin	0.12	0.12	94.0	6.0
	Erythromycin	0.25	>8	84.0	16.0
	Daptomycin	0.25	0.5	100.0	⁜
	Vancomycin	0.5	0.5	100.0	0.0
	Linezolid	2	2	100.0	0.0
	Gentamicin	0.25	0.5	94.0	4.0
	Tetracycline	0.25	1	90.0	10.0
	Trimeth/ Sulfa	<=0.5	<=0.5	100.0	0.0
CoNS (n=30)	E-101	0.015	0.015	⁜	⁜
	Oxacillin	>4	>4	23.3	76.7
	Cefazolin	2	16	80.0	10.0
	Ciprofloxacin	4	>4	46.7	53.3
	Clindamycin	0.06	>4	63.3	36.7
	Erythromycin	>8	>8	30.0	70.0
	Daptomycin	0.5	0.5	100.0	0.0
	Vancomycin	1	1	100.0	0.0
	Linezolid	1	1	100.0	⁜
	Gentamicin	0.12	>16	53.3	43.3
	Tetracycline	2	>32	63.3	36.7
	Trimeth/ Sulfa	<=0.5	>4	66.7	33.3
E. faecalis (n=14)	E-101	0.25	0.25	⁜	⁜
	Ampicillin	1	1	100.0	0.0
	Cefazolin	32	32	⁜	⁜
	Ciprofloxacin	1	>4	50.0	50.0
	Erythromycin	>8	>8	14.3	71.4
	Daptomycin	1	1	100.0	⁜
	Vancomycin	1	1	100.0	0.0
	Linezolid	1	2	100.0	0.0
	Tetracycline	>32	>32	21.4	78.6
E. faecium (n=16)	E-101	0.06	0.12	⁜	⁜
	Ampicillin	>16	>16	18.8	81.3
	Cefazolin	>64	>64	⁜	⁜
	Ciprofloxacin	>4	>4	0.0	81.3
	Erythromycin	>8	>8	0.0	75.0
	Daptomycin	2	2	100.0	⁜
	Vancomycin	0.5	>32	68.8	12.5
	Linezolid	1	2	100.0	0.0
	Tetracycline	0.25	>32	81.3	18.8
	Trimeth/ Sulfa	>4	>4	⁜	⁜

FIGURE 1. E-101 MICs against European Gram-positive clinical isolates

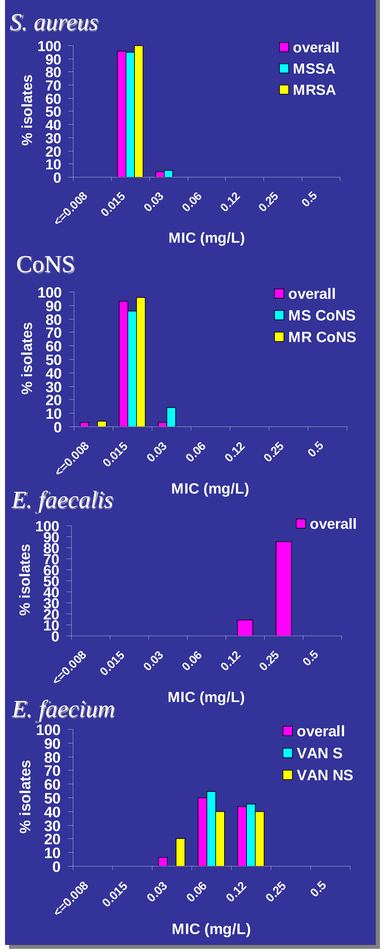
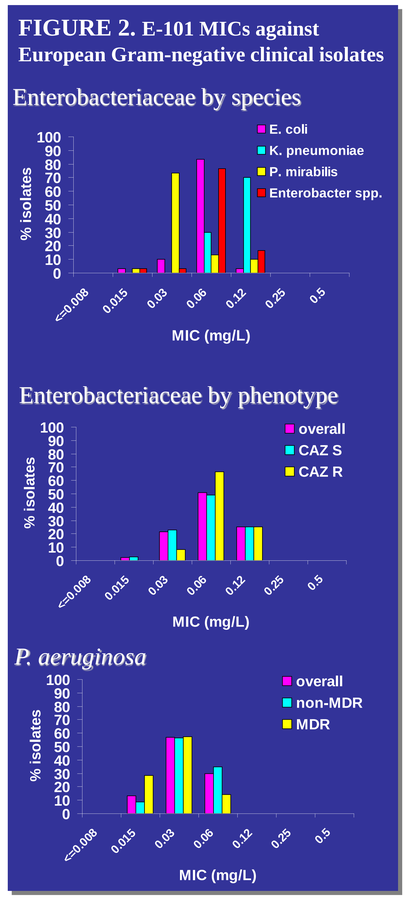


TABLE 2. Activity of E-101 and comparators against European Gram-negative clinical isolates

Organism	Drug	MIC (mg/mL) MIC_{50}	MIC_{90}	%S	%R
E. coli (n=30)	E-101	0.06	0.06	⁜	⁜
	Ampicillin	>32	>32	26.7	73.3
	Aztreonam	0.06	0.25	93.3	6.7
	Cefazolin	2	16	23.3	46.7
	Ceftazidime	0.12	0.25	96.7	3.3
	Imipenem	0.12	0.25	100.0	0.0
	Pip/Tazo	2	8	100.0	0.0
	Levofloxacin	0.03	>8	73.3	26.7
	Gentamicin	0.5	4	90.0	10.0
	Tetracycline	2	>32	53.3	46.7
	Trimeth/ Sulfa	<=0.25	>32	60.0	40.0
K. pneumoniae (n=30)	E-101	0.12	0.12	⁜	⁜
	Ampicillin	>32	>32	6.7	83.3
	Aztreonam	0.06	16	86.7	13.3
	Cefazolin	1	>32	60.0	33.3
	Ceftazidime	0.12	16	86.7	13.3
	Imipenem	0.12	0.25	100.0	0.0
	Pip/Tazo	4	64	83.3	6.7
	Levofloxacin	0.06	8	86.7	13.3
	Gentamicin	0.25	0.5	93.3	6.7
	Tetracycline	1	>32	66.7	30.0
	Trimeth/ Sulfa	<=0.25	>32	76.7	23.3
P. mirabilis (n=30)	E-101	0.03	0.06	⁜	⁜
	Ampicillin	>32	>32	40.0	60.0
	Aztreonam	<=0.03	<=0.03	100.0	0.0
	Cefazolin	4	32	6.7	83.3
	Ceftazidime	<=0.03	0.12	96.7	3.3
	Imipenem	2	4	43.3	20.0
	Pip/Tazo	<=0.5	1	100.0	0.0
	Levofloxacin	0.06	4	86.7	6.7
	Gentamicin	1	>16	80.0	16.7
	Tetracycline	32	>32	6.7	90.0
	Trimeth/ Sulfa	>32	>32	40.0	60.0
Enterobacter spp. (n=30)	E-101	0.06	0.12	⁜	⁜
	Ampicillin	>32	>32	6.7	90.0
	Aztreonam	0.12	>32	80.0	16.7
	Cefazolin	>32	>32	3.3	93.3
	Ceftazidime	0.25	>32	80.0	20.0
	Imipenem	0.5	1	96.7	3.3
	Pip/Tazo	2	>128	83.3	16.7
	Levofloxacin	0.06	2	90.0	6.7
	Gentamicin	0.5	0.5	96.7	3.3
	Tetracycline	2	4	93.3	3.3
	Trimeth/ Sulfa	<=0.25	0.5	90.0	10.0
P. aeruginosa (n=30)	E-101	0.03	0.06	⁜	⁜
	Ampicillin	>32	>32	⁜	⁜
	Aztreonam	8	>32	53.3	26.7
	Cefazolin	>32	>32	⁜	⁜
	Ceftazidime	4	>32	76.7	16.7
	Imipenem	2	16	70.0	20.0
	Pip/Tazo	4	64	90.0	10.0
	Levofloxacin	0.5	>8	80.0	20.0
	Gentamicin	2	>16	83.3	16.7
	Tetracycline	16	>32	6.7	86.7
	Trimeth/ Sulfa	8	>32	3.3	96.7

FIGURE 2. E-101 MICs against European Gram-negative clinical isolates



CONCLUSIONS

- E-101 had potent activity *in vitro* against target clinical pathogens from Europe
- The activity of E-101 was not impacted by common resistances encountered among the target bacterial species
- These data illustrate the potential of E-101 when used topically to prevent surgical site infection

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