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Antimicrobial activity of iclaprim tested against recent *S. aureus* clinical isolates: Results from the International Study of Iclaprim Susceptibility (ISIS).

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ABSTRACT

Background: Iclaprim (ICL) is a novel dihydrofolate reductase inhibitor with broad antibacterial activity. We evaluated the comparative potency and bactericidal activity (MBC) of ICL tested against a large collection of clinical *Staphylococcus aureus* strains.

Methods: 4,516 clinical isolates (2,359 from the USA and 2,157 Europe [EU]) collected in 2004–2006 were susceptibility (S) tested by CLSI broth microdilution method against ICL, trimethoprim (TMP), co-trimoxazole (TMP/SMX) and various comparators. MBC values for ICL, TMP and VAN were assessed for a subset of 41 strains.

Results: ICL was generally 16-fold more potent than TMP and VAN and was equally potent to TMP/SMX (Table). 95% of strains were inhibited at ≤0.5 µg/mL of ICL. ICL was equally active against methicillin-S (MSSA) and -resistant (MRSA) strains from the USA (MIC_{90}, 0.12 µg/mL for both). In EU, MRSA showed ICL MIC values slightly higher than MSSA (MIC_{90}, 1 vs. 0.12 µg/mL), but the difference for TMP was more pronounced (MIC_{90}, 16 vs. 1 µg/mL). ICL exhibited bactericidal activity against the majority of isolates tested. ICL showed superior bactericidal activity to VAN. ICL MBC/MIC rates were ≤2 for 63.4% and ≤4 for 85.4% of strains tested. Only 2 strains (4.9%; both MSSA) had elevated (≥32) MBC/MIC ratios. 5 strains (3 MRSA) had VAN MBC/MIC at ≥32.

Antimicrobial/organism (n)	MIC_{50}	MIC_{90}	% S
S. aureus (4,516)			
ICL	0.06	0.12	-
TMP	1	2	94.8
TMP/SMX	0.06	0.12	97.2
Erythromycin	>4	>4	35.8
Clindamycin	≤0.12	>4	67.4
Levofloxacin	4	>4	42.8
Oxacillin	>2	>2	33.5
Linezolid	2	2	100.0
Vancomycin	1	1	100.0

Conclusions: ICL was very active and highly bactericidal against recent clinical *S. aureus* strains and its activity was not adversely affected by resistance to oxacillin or other antimicrobials tested. Notably, ICL showed superior bactericidal activity than VAN against MRSA.

INTRODUCTION

Iclalclaprim is a novel investigational drug, which is being developed for serious Gram-positive bacterial infections. The compound has been granted fast-track product designation and its intravenous formulation has recently completed two pivotal Phase III clinical studies in complicated skin and skin structures infections (cSSSI). In addition, a recent bronchial alveolar lavage study has shown that in healthy subjects the concentrations of iclaprim in the epithelial lining fluid and alveolar macrophages were about 20 and 40 times higher than in plasma, respectively. Iclaprim is also orally bioavailable and several Phase I clinical studies have been conducted with its oral form.

Iclaprim is a novel dihydrofolate reductase (DFHR) inhibitor belonging to the diaminopyrimidine class of antibiotics for which trimethoprim (TMP) is the most well-known representative. TMP is frequently used in combination with sulfamethoxazole (SMX) and this combination has been used in clinical practice for almost five decades. SMX is highly synergistic with TMP as a result of inhibition of two sequential enzymes in the folate pathway, thereby enhancing the potency and bactericidal activity as well as reducing the potential for resistance development. However, SMX is often associated with allergic reactions. In contrast, iclaprim by itself shows a potent activity against a variety of pathogens and exhibits a potent bactericidal action against methicillin-susceptible and -resistant *Staphylococcus aureus, Streptococcus pneumoniae and* important RTI pathogens.

In the present study, we evaluated the comparative potency and bactericidal activity of iclaprim tested against a large collection of contemporary clinical *S. aureus* strains.

METHODS

Bacterial isolates: A total of 4,516 clinical *S. aureus* isolates, 1,503 oxacillin- (methicillin-) susceptible (MSSA) and 3,003 MRSA, were selected for the in-vitro trial. All organisms were collected from patients in North American (2,359) and European (2,157) medical centers between 2004 and 2006. Sources of infection consisted of bloodstream, skin and soft tissue, respiratory and patients hospitalized from pneumonia. MBC values for iclaprim and vancomycin were assessed for a subset of 41 clinical strains.

Susceptibility testing: MIC values were evaluated by the Clinical and Laboratory Standards Institute (CLSI) broth microdilution method per M7-A7 (2006). Quality control ranges and interpretive criteria for iclaprim and comparator compounds were used as published in CLSI M100-S17. Quality control ranges for iclaprim were used as published in M100-S17 (2007).

Bactericidal activity: MBC values were assessed by plating the broth onto appropriate growth media from the MIC microdilution tray well and those from at least five log_2 dilutions at and above the MIC for each organism. Quantitative colony counts were also performed on the starting inoculum. The lowest concentration of antimicrobial agent that killed ≥99.9% of the initial inoculum was defined as the MBC endpoint.

RESULTS

- Iclaprim was very active against *S. aureus* (MIC_{50} 0.06 µg/mL; MIC_{90} 0.12 µg/mL) (Table 1) with over 95% of the strains being inhibited at MICs ≤0.5 µg/mL. Iclaprim was 16-fold more potent than TMP (MIC_{50} 1 µg/mL; MIC_{90} 2 µg/mL) and showed in-vitro activity similar to that of the TMP/SMX combination (MIC_{50} 0.06 µg/mL; MIC_{90} 0.12 µg/mL).
- The activity of iclaprim against MSSA (1,513 strains) and MRSA (3,003 strains) was similar with $MIC_{50/90}$ values of 0.06/0.12 µg/mL for both isolates (Table 1). In contrast, both TMP and TMP/SMX were slightly less active against MRSA isolates compared with MSSA strains (Table 1). MIC_{90}s for all three drugs were somewhat lower against MRSA isolates from the USA than against isolates from Europe (Table 1).

Table 1. Activity of iclaprim and comparator agents tested against *S. aureus* from the USA and Europe.

Antimicrobial agent (no. tested)	MIC_{50}	MIC_{90}	Range	% susceptible/ resistant[a]
Oxacillin-susceptible *S. aureus* (MSSA)				
USA (760)				
Iclaprim	0.06	0.12	0.008–8	- / -[b]
Trimethoprim	1	2	0.06–>64	98.7 / 1.3
Trimethoprim-sulfamethoxazole	0.06	0.06	0.015–>8	98.9 / 1.1
Erythromycin	0.25	>4	≤0.12–>4	70.7 / 27.8
Clindamycin	≤0.12	≤0.12	≤0.12–>4	95.8 / 4.2
Tetracycline	≤0.5	≤0.5	≤0.5–>16	97.1 / 2.5
Levofloxacin	0.25	0.5	≤0.12–>4	93.7 / 6.3
Vancomycin	1	1	≤0.5–2	100.0 / 0.0
Europe (753)				
Iclaprim	0.06	0.12	0.015–4	- / -[b]
Trimethoprim	1	1	0.12–>64	98.7 / 1.3
Trimethoprim-sulfamethoxazole	0.06	0.06	0.015–>8	99.7 / 0.3
Erythromycin	0.25	>4	≤0.12–>4	85.3 / 14.3
Clindamycin	≤0.12	≤0.12	≤0.12–>4	97.1 / 2.7
Tetracycline	≤0.5	≤0.5	≤0.5–>16	94.4 / 5.6
Levofloxacin	0.25	0.25	≤0.12–>4	94.8 / 4.9
Vancomycin	1	1	≤0.5–2	100.0 / 0.0
All (1,513)				
Iclaprim	0.06	0.12	0.008–8	- / -[b]
Trimethoprim	1	1	0.06–>64	98.7 / 1.3
Trimethoprim-sulfamethoxazole	0.06	0.06	0.015–>8	99.3 / 0.7
Erythromycin	0.25	>4	≤0.12–>4	77.9 / 21.1
Clindamycin	≤0.12	≤0.12	≤0.12–>4	96.4 / 3.4
Tetracycline	≤0.5	≤0.5	≤0.5–>16	95.8 / 4.0
Ciprofloxacin	0.5	1	≤0.12–>4	95.8 / 5.9
Levofloxacin	0.25	0.5	≤0.12–>4	94.2 / 5.6
Vancomycin	1	1	≤0.5–2	100.0 / 0.0
Oxacillin-resistant *S. aureus*				
USA (1,599)				
Iclaprim	0.06	0.12	≤0.004–8	- / -[b]
Trimethoprim	0.5	1	0.06–>64	95.7 / 4.3
Trimethoprim-sulfamethoxazole	0.06	0.06	0.015–>8	97.9 / 2.1
Erythromycin	>4	>4	0.25–>4	5.1 / 94.3
Clindamycin	≤0.12	>4	≤0.12–>4	53.9 / 46.0
Tetracycline	≤0.5	2	≤0.5–>16	93.8 / 5.9
Levofloxacin	>4	>4	≤0.12–>4	25.2 / 74.5
Vancomycin	1	1	≤0.5–2	100.0 / 0.0
Europe (1,404)				
Iclaprim	0.06	1	0.008–8	- / -[b]
Trimethoprim	1	16	0.12–>64	89.7 / 10.3
Trimethoprim-sulfamethoxazole	0.06	0.25	0.015–>8	94.0 / 6.0
Erythromycin	>4	>4	≤0.12–>4	25.4 / 72.9
Clindamycin	0.25	>4	≤0.12–>4	51.4 / 48.2
Tetracycline	≤0.5	>16	≤0.5–>16	78.3 / 20.6
Levofloxacin	>4	>4	≤0.12–>4	7.3 / 91.3
Vancomycin	1	1	≤0.5–2	100.0 / 0.0
All (3,003)				
Iclaprim	0.06	0.12	≤0.004–8	- / -[b]
Trimethoprim	1	2	0.06–>64	92.9 / 7.1
Trimethoprim-sulfamethoxazole	0.06	0.25	0.015–>8	96.1 / 3.9
Erythromycin	>4	>4	≤0.12–>4	14.6 / 84.3
Clindamycin	≤0.12	>4	≤0.12–>4	52.7 / 47.0
Tetracycline	≤0.5	>16	≤0.5–>16	86.5 / 12.8
Ciprofloxacin	>4	>4	≤0.12–>4	16.3 / 83.1
Levofloxacin	>4	>4	≤0.12–>4	16.8 / 82.4
Vancomycin	1	1	≤0.5–2	100.0 / 0.0

[a] Criteria as published by the CLSI, β-lactam susceptibility should be directed by the oxacillin test results.
[b] No criteria have been established by the CLSI.

- Iclaprim exhibited bactericidal activity against the vast majority of staphylococcal strains tested, with MBCs ≤0.25 µg/mL against all MRSA and ≤0.5 µg/mL against 18 of the 21 MSSA isolates (Table 2).

Table 2. Distribution of iclaprim MIC and MBC results.

Organism (no. of isolates)	No. of isolates at MIC or MBC value (µg/mL) of:									
	0.008	0.015	0.03	0.06	0.12	0.25	0.5	1	2	>2
MSSA (21)										
MIC	0	0	0	1	15	5	0	0	0	0
MBC	0	0	0	0	0	8	10	1	0	2
MRSA (20)										
MIC	0	0	3	7	10	0	0	0	0	0
MBC	0	0	0	1	12	7	0	0	0	0

- Iclaprim exhibited MBC/MIC ratios ≤4 against all MRSA strains and against 18 of the 21 MSSA isolates (Table 3). By contrast, vancomycin MBC/MIC were ≤4 for only 12 of the 20 MRSA isolates tested (Table 3).

Table 3. MBC/MIC ratio results for iclaprim and vancomycin tested against a selected group of 41 organisms.

MBC/MIC ratio/organism (no.)	No. of isolates at MBC/MIC	
	Iclaprim	Vancomycin
MSSA (21)		
1	2	14
2	7	2
4	9	2
8	1	1
16	0	0
≥32	2	2
MRSA (20)		
1	4	8
2	13	1
4	3	3
8	0	4
16	0	1
≥32	0	3

CONCLUSIONS

- **Iclaprim was highly active against MSSA and MRSA (MIC_{90} 0.06 µg/mL and MIC_{90} 0.12 µg/mL) and over 95% of the isolates tested in the study exhibited MIC values ≤0.5 µg/mL.**
- **Iclaprim showed similar activities against MSSA (1,513 strains) and MRSA (3,003 strains) with $MIC_{50/90}$ values of 0.06/0.12 µg/mL against both isolates.**
- **Iclaprim was bactericidal against all MRSA and against the vast majority MSSA isolates tested in this study.**

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