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In Vitro Activity of a Novel Aminomethylcycline Antibacterial (KBP-7072) against Clinical Isolates with Molecularly Characterized Tetracycline Resistance Mechanisms

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Introduction

- KBP-7072 is a 3rd generation aminomethylcycline antibiotic being developed for the treatment of acute bacterial skin and skin structure infections, community-acquired bacterial pneumonia, and complicated intra-abdominal infections.
- This investigational drug has completed phase I clinical development and has shown potent *in vitro* activity against methicillin-resistant *Staphylococcus aureus*, penicillin-non-susceptible *Streptococcus pneumoniae*, ampicillin-resistant *Haemophilus influenzae*, vancomycin-resistant *Enterococcus faecalis*, carbapenem-resistant *Enterobacterales*, and carbapenem-resistant *Acinetobacter baumannii*.
- This study evaluated the *in vitro* activity of KBP-7072 against 413 contemporary surveillance isolates, including subsets with known acquired tetracycline resistance genes.

Materials and Methods

Bacterial isolates

- A global collection of 413 isolates recovered from documented infections during the SENTRY Surveillance Program for 2015–2019 were included in this study.
- Organisms tested in this study included 103 *S. aureus* (51 tetracycline-resistant), 102 *S. pneumoniae* (51 tetracycline-resistant), 103 *Escherichia coli* (52 tetracycline-resistant), and 105 *Klebsiella pneumoniae* (51 tetracycline-resistant) isolates (Table 1).
- Isolates were collected in 119 medical centers located in 34 countries: North America (the United States, 58 medical centers in 9 US census divisions; 183 isolates; 44.3% overall), Europe (19 countries, 36 medical centers; 162 isolates; 39.2% overall), Latin America (6 countries, 10 medical centers; 29 isolates; 7.0% overall), and the Asia-Pacific region (8 countries, 15 medical centers; 39 isolates; 9.4% overall).
- Bacterial isolate identification was confirmed by standard algorithms supported by matrix-assisted laser desorption ionization-time of flight mass spectrometry (Bruker Daltonics, Bremen, Germany) and genome sequencing.

Antimicrobial susceptibility testing

- Isolates were tested for susceptibility by broth microdilution following Clinical and Laboratory Standards Institute (CLSI) M07 (2018) guidelines.
- Frozen-form broth microdilution panels were manufactured by JMI Laboratories (North Liberty, Iowa) and contained fresh cation-adjusted Mueller-Hinton broth (2.5–5% lysed horse blood added for testing streptococci).
- Quality assurance was performed by concurrently testing CLSI-recommended quality control reference strains (*S. aureus* ATCC 29213, *E. faecalis* ATCC 29212, and *S. pneumoniae* ATCC 49619).

Characterization of resistance mechanisms by next-generation sequencing

- Tetracycline-resistant isolates had total genomic DNA extracted by the fully automated Thermo Scientific™ KingFisher™ Flex Magnetic Particle Processor (Cleveland, OH, USA), which was used as input material for library construction.
- DNA libraries were prepared using the Nextera™ library construction protocol (Illumina, San Diego, CA, USA) following the manufacturer's instructions and were sequenced on MiSeq Sequencer platforms at JMI Laboratories.
- FASTQ format sequencing files for each sample set were assembled independently using *de novo* assembler SPAdes 3.11.1. An in-house software was applied to align the assembled sequences against known tetracycline resistance genes.

Results

- KBP-7072 showed MIC_{50} and MIC_{90} values of 0.06 and 0.5 mg/L against *S. aureus*, respectively, and inhibited all isolates at ≤1 mg/L, including the tetracycline-resistant subset (Table 2).
- Overall, KBP-7072 had an MIC_{50} value of 0.06 mg/L against tetracycline-resistant *S. aureus*, whereas the MIC_{50} values for KBP-7072 varied between 0.25 mg/L against *tet*(M)-carrying isolates and 0.06 mg/L when tested against other genotypes (Table 2).
- A comparative analysis demonstrated that KBP-7072 (MIC_{50}, 0.06 mg/L), tigecycline (MIC_{50}, 0.12 and 0.25 mg/L), and omadacycline (MIC_{50}, 0.12 and 0.5 mg/L) showed similar MIC_{50} values against tetracycline-susceptible and -resistant *S. aureus*. Other tetracycline comparators had the MIC_{50} values increased 64- to 256-fold by *tet* genes (Table 3).
- Against *S. pneumoniae*, KBP-7072 ($MIC_{50/90}$, ≤0.015/0.03 mg/L) showed the lowest MIC results, which remained unchanged when comparing results obtained against tetracycline-susceptible and -resistant isolates (mostly *tet*(M)) (Tables 2 and 3).
- Similar MIC results were observed between omadacycline ($MIC_{50/90}$, 0.03-0.06/0.06 mg/L) and tigecycline ($MIC_{50/90}$, 0.03/0.03 mg/L) when tested against tetracycline-susceptible and -resistant *S. pneumoniae* populations (Table 3).
- KBP-7072 had MIC_{50} values of 0.12 mg/L and 0.25 mg/L when tested against tetracycline-susceptible and -resistant *E. coli*, respectively (Table 2).
  - KBP-7072 (MIC_{90}, 0.25 and 1 mg/L, respectively) and tigecycline (MIC_{90}, 0.25 and 0.5 mg/L) showed similar MIC_{90} values against these *E. coli* groups (Table 3).
- KBP-7072 ($MIC_{50/90}$, 0.25/0.5 mg/L) and tigecycline ($MIC_{50/90}$, 0.5 and 0.5 mg/L) had the lowest MIC values against tetracycline-susceptible *K. pneumoniae*. The MIC for KBP-7072 ($MIC_{50/90}$, 1/4 mg/L) and tigecycline ($MIC_{50/90}$, 1/2 mg/L) increased 2- to 8-fold against tetracycline-resistant *K. pneumoniae*, which mostly produced Tet(A) (Table 3).

Table 1. Geographic distribution of tetracycline-resistant isolates

Organism	Tetracycline resistance gene(s)	Number of isolates per region: North America	Europe	Latin America	Asia-W. Pacific
S. aureus	*tet*(K)	3	10	—	2
	tet(M)	5	3	—	20
	tet(K) + *tet*(M)	1	4	—	—
	tet(L)	—	—	1	—
	tet(L) + *tet*(M)	—	—	—	1
	tet(38)[a]	1			
S. pneumoniae	*tet*(M)	45	5		
	tet(32)	—	1		
E. coli	*tet*(A)	9	11		
	tet(A) + *tet*(B)	5	3		
	tet(B)	11	10		
	tet(D)	1	2		
K. pneumoniae	*tet*(A)	15	25		
	tet(A) + *tet*(B)	2	—		
	tet(A) + *tet*(G)	—	2		
	tet(D)	4	1		
	tet(G)	—	2		

[a] Acquired tetracycline resistance genes were not detected in this isolate.

Table 2. Antimicrobial activity of KBP-7072 tested against the main organism groups and characterized subsets

Organism/organism group (no. of isolates)	No. and cumulative % of isolates inhibited at MIC (mg/L) of: ≤0.015	0.03	0.06	0.12	0.25	0.5	1	2	4	8	MIC_{50}	MIC_{90}
S. aureus (103)		11 10.7	56 65.0	13 77.7	7 84.5	14 98.1	2 100.0				0.06	0.5
Tetracycline-S (52)		7 13.5	33 76.9	9 94.2	1 96.2	2 100.0					0.06	0.12
Tetracycline-R (51)		4 7.8	23 52.9	4 60.8	6 72.5	12 96.1	2 100.0				0.06	0.5
tet(M) (28)		2 7.1	5 25.0	1 28.6	6 50.0	12 92.9	2 100.0				0.25	0.5
tet(K) (15)		1 6.7	12 86.7	2 100.0							0.06	0.12
tet(K) + *tet*(M) (5)			5 100.0								0.06	—
Other[a] (3)		1 33.3	1 66.7	1 100							0.06	—
S. pneumoniae (102)	72 70.6	30 100.0									≤0.015	0.03
Tetracycline-S (51)	31 60.8	20 100.0									≤0.015	0.03
Tetracycline-R (51)	41 80.4	10 100.0									≤0.015	0.03
tet(M) (50)	40 80.0	10 100.0									≤0.015	0.03
tet(32) (1)	1 100.0										—	—
E. coli (103)			3 2.9	57 58.3	27 84.5	9 93.2	7 100.0				0.12	0.5
Tetracycline-S (51)			3 5.9	39 82.4	9 100.0						0.12	0.25
Tetracycline-R (52)				18 34.6	18 69.2	9 86.5	7 100.0				0.25	1
tet(A) (20)				5 25.0	7 60.0	3 100.0					0.25	0.5
tet(A) + *tet*(B) (8)				1 12.5	4 62.5	1 75.0	2 100.0				0.25	—
tet(B) (21)				9 42.9	9 85.7	1 90.5	2 100.0				0.25	0.5
tet(D) (3)				3 100.0							0.12	—
Klebsiella pneumoniae (105)			1 1.0	8 8.6	41 47.6	22 68.6	19 86.7	7 93.3	5 98.1	2 100.0	0.5	2
Tetracycline-S (54)			1 1.9	8 16.7	39 88.9	6 100.0					0.25	0.5
Tetracycline-R (51)					2 3.9	16 35.3	19 72.5	7 86.3	5 96.1	2 100.0	1	4
tet(A) (40)					2 5.0	14 40.0	16 80.0	4 90.0	2 95.0	2 100.0	1	2
tet(A) + *tet*(B) (2)								2 100.0			2	—
tet(A) + *tet*(G) (2)							1 50.0	1 100.0			1	—
tet(D) (5)						2 40.0	1 60.0	0 60.0	2 100.0		1	—
tet(G) (2)							1 50.0	0 50.0	1 100.0		1	—

[a] Includes 1 isolates each with *tet*(L) and *tet*(L) + *tet*(M), and 1 isolate with no acquired tetracycline resistance gene.
S, susceptible; R, resistant

Table 3. *In vitro* activity of old and new generation tetracycline agents

Organism (no.)	MIC_{50}/MIC_{90} (% Susceptible by CLSI/EUCAST[a]) KBP-7072	DOX	MIN	OMC	TET	TIG
TET-S *S. aureus* (52)	0.06/0.12	0.12/0.25 (100.0/100.0)	0.12/0.25 (100.0/98.1)	0.12/0.25 (94.2/-)	0.25/0.5 (100.0/96.2)	0.12/0.25 (98.1/98.1)
TET-R *S. aureus*[b] (51)	0.06/0.5	8/8 (29.4/11.8)	8/16 (43.1/29.4)	0.5/2 (58.8/-)	64/64 (0.0/0.0)	0.25/0.5 (90.2/90.2)
TET-S *S. pneumoniae* (51)	≤0.015/0.03	0.12/0.12 (100.0/100.0)	0.12/0.12 (-/100.0)	0.03/0.06 (100.0/-)	0.25/0.25 (100.0/100.0)	0.03/0.03 (100.0/-)
TET-R *S. pneumoniae*[c] (51)	≤0.015/0.03	4/16 (0.0/3.9)	8/16 (-/2.0)	0.06/0.06 (100.0/-)	32/64 (0.0/0.0)	0.03/0.03 (100.0/-)
TET-S *E. coli* (51)	0.12/0.25	1/2 (100.0/-)	1/1 (100.0/-)	0.5/1 (-/-)	1/2 (100.0/-)	0.12/0.25 (100.0/100.0)
TET-R *E. coli*[d] (52)	0.25/1	32/>32 (5.8/-)	8/32 (42.3/-)	1/4 (-/-)	>64/>64 (0.0/-)	0.25/0.5 (100.0/98.1)
TET-S *K. pneumoniae* (54)	0.25/0.5	1/2 (100.0/-)	1/2 (100.0/-)	1/2 (100.0/-)	1/2 (100.0/-)	0.5/0.5 (100.0/-)
TET-R *K. pneumoniae*[e] (51)	1/4	16/>32 (0.0/-)	4/>32 (52.9/-)	4/16 (54.9/-)	>64/>64 (0.0/-)	1/2 (92.2/-)

S, susceptible; R, resistant; DOX, doxycycline; MIN, minocycline; OMC, omadacycline; TET, tetracycline; TIG, tigecycline
[a] CLSI and EUCAST breakpoints were applied. FDA breakpoint interpretive criteria were used for tigecycline and omadacycline.
[b] Contains 15 tet(K), 5 tet(K)/tet(M), 1 tet(L), 28 tet(M), 1 tet(L)/tet(M), and 1 isolate with no acquired tetracycline resistance gene.
[c] All isolates carried tet(M), except for 1 strain with a tet(32).
[d] Contains 20 tet(A), 8 tet(A)/tet(B), 21 tet(B), and 3 tet(D).
[e] Contains 40 tet(A), 2 tet(A)/tet(B), 2 tet(A)/tet(G), 5 tet(D), and 2 tet(G).

Conclusions

- Based on the MIC results obtained in this study, KBP-7072 was in vitro active against wildtype *E. coli*, *K. pneumoniae*, *S. aureus*, and *S. pneumoniae* clinical isolates.
- KBP-7072 remained active against tetracycline-resistant isolates carrying a variety of acquired tetracycline resistance genes.
- These results indicate that KBP-7072 warrants further investigation as an option for treatment of infections, including those caused by pathogens resistant to older generation tetracycline.

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References

CLSI. M100Ed30. Performance standards for antimicrobial susceptibility testing; 30th informational supplement. Wayne, PA, Clinical and Laboratory Standards Institute, 2020.

CLSI. M07Ed11. Methods for dilution antimicrobial susceptibilty tests for bacteria that grow aerobically; approved standard—eleventh edition. Wayne, PA, Clinical and Laboratory Standards Institute, 2018.

EUCAST (2020). Breakpoint tables for interpretation of MIC's and zone diameters. Version 10.0, January 2020. Available at http://www.eucast.org/fileadmin/src/media/PDFs/EUCAST_files/Breakpoint_tables/v_10.0_Breakpoint_Tables.pdf. Accessed May 2020.

United States Food and Drug Administration (2020). Antibacterial susceptibility test interpretive criteria. Available at https://www.fda.gov/Drugs/DevelopmentApprovalProcess/DevelopmentResources/ucm575163.htm. Accessed May 2020.

Roberts MC. Update on acquired tetracycline resistance genes. 2005. *FEMS Microbiology Letters* 245,195-203

Mendes RM, Huband MD, Streit JM, Castanheira M, Flamm RK. Omadacycline *in vitro* activity against a molecularly characterized collection of clinical isolates with known tetracycline resistance mechanisms. 2020. *Diag Microbiol Infect Dis.* doi:10.1016/j.diagmicrobio.2020.115054.

Mendes RM, Jones RN, Woosley LN, Cattoir V, Castanheira M. Application of next generation sequencing for characterization of surveillance and clinical trial isolates: Analysis of the distribution of β-lactamase resistance genes and lineage background in the United States. 2019. Open Forum Infect Dis. doi:10.1093/ofid/ofz004.

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