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Cefepime-Taniborbactam Human Exposures Suppress the Emergence of Resistance among Cefepime-Nonsusceptible Enterobacterales and *Pseudomonas aeruginosa* in a Hollow Fiber Infection Model

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Abstract

Background Cefepime-taniborbactam (FTB) efficacy and safety were recently demonstrated in a Phase 3 study in adults with complicated urinary tract infections including pyelonephritis (CERTAIN-1). Taniborbactam is a boronic acid-based β-lactamase inhibitor (BLI) with broad activity against many serine β-lactamases (SBL) and metallo-β-lactamases (MBL). In this study, an in vitro hollow fiber infection model (HFM) was used to evaluate the potential for treatment-emergent resistance associated with the clinical FTB dose among *Escherichia coli* (*Eco*), *Klebsiella pneumoniae* (*Kpn*), and *Pseudomonas aeruginosa* (*Pae*).

Methods HFM validation was performed with a humanized dose of ceftazidime-avibactam (CZA, 2.5 g q8h, 2-h infusion) or meropenem-vaborbactam (MEV, 4 g q8h, 3-h infusion) against susceptible strains. Minimum inhibitory concentrations (MICs) of cefepime (FEP) and FTB among 9 total study strains (1 *Eco*, 5 *Kpn*, 3 *Pae*) ranged from 16 to >128 μg/mL and 0.25 to 8 μg/mL, respectively. Collectively, strains harbored a variety of clinically relevant cephalosporinases (CMY), extended-spectrum β-lactamases (ESBL; CTX-M, SHV, PER, VEB) and carbapenemases (SBL: KPC, OXA; MBL: NDM, VIM). Each HFM cartridge was inoculated with 10^7 to 10^8 CFU of a log phase culture. Syringe pumps infused humanized doses of FEP (2 g) or FTB (2.5 g) over 2 h q8h for 7 days. Samples were serially collected for quantitative culture. Subpopulations with elevated MIC (CZA, MEV, or FTB according to HFM treatment) were monitored with drug-supplemented agar.

Results Humanized exposures of CZA were bactericidal and prevented resistance for 7 days in the HFM against *Kpn* ATCC BAA-1705 (CZA MIC, 1 μg/mL). Against OXA-48-positive *Kpn* 752285 (MEV MIC, 2 μg/mL), regrowth to 10^{10} CFU/mL was observed by Day 1 of humanized MEV treatment; this result demonstrated the rigor of the HFM system in assessment of BLI activity as vaborbactam does not inhibit OXA-48, an SBL that confers low-level meropenem resistance. All 9 strains grew rapidly when treated with FEP alone, consistent with phenotypic resistance, whereas humanized FTB reduced the inoculum by 2 to 6 $\log_{10}$ CFU/mL following the first dose for all strains. FTB prevented regrowth for 7 days in each model, with ≥ 3 $\log_{10}$ CFU/mL reductions (i.e., bactericidal activity) achieved at Day 7 in all 9 strains. No CFU were recovered from a 1-mL sample volume at the final time point (i.e., cartridge sterilization) in 3 *Kpn* strains treated with FTB. Colonies with elevated FTB MIC (4x) were not detected from any FTB-treated model.

Conclusions Human FTB exposures demonstrated sustained bactericidal activity and suppressed the emergence of resistance in a validated 7-day HFM among ESBL- and/or carbapenemase-positive Enterobacterales and *P. aeruginosa* strains. These observations in a rigorous HFM support the clinical development of FTB and inform understanding of its potential role in SBL- and/or MBL-positive gram-negative infections.

Introduction

- **Cefepime-taniborbactam (FTB)**, an antipseudomonal cephalosporin-novel boronate β-lactamase inhibitor (BLI) combination, was safe and effective in adults with complicated urinary tract infections in the CERTAIN-1 Phase 3 clinical trial and is also under development for hospital-acquired bacterial pneumonia and ventilator-associated bacterial pneumonia (HABP/VABP).
- **Taniborbactam** (formerly VNRX-5133) acts solely as a BLI (lacking intrinsic antibacterial activity) to potentiate the in vitro activity of cefepime against Enterobacterales and *Pseudomonas aeruginosa* strains harboring serine (SBL)- and metallo (MBL)-β-lactamases (e.g., CTX-M, SHV, NDM, VIM, AmpC, OXA-48).[1]
- **Efficacy of FTB humanized exposures** has been demonstrated in multiple translational in vivo infection models.[2-4]
- **Hollow fiber infection models** (HFM, *see schematic at top right*) are dynamic in vitro systems preferred for assessments of antimicrobial resistance prevention.[5]
- **Objective.** The HFM was used to investigate the potential for treatment-emergent resistance to humanized exposures of FTB among SBL- and/or MBL-producing *Escherichia coli* (EC), *Klebsiella pneumoniae* (KP), and *P. aeruginosa* (PA) strains.

Methods

Bacterial strains & antimicrobial agents. Strains (**Table 1**) were acquired from the CDC & FDA Antibiotic Resistance (AR) Isolate Bank, American Type Culture Collection (ATCC), or the International Health Management Associates (Schaumburg, IL). Commercial vials of cefepime (FEP), ceftazidime, and meropenem were used in the HFM and chemical grade products (Sigma-Aldrich) were used in static susceptibility tests. Taniborbactam (Carbogen Amcis AG, batches CA19-1355 and CA20-0265), avibactam (Venatorx, NJL-159-054-4), and vaborbactam (MedChemExpress, lot HY-19930) were used throughout the study. Minimum inhibitory concentrations (MIC) of FEP, FTB, ceftazidime-avibactam (CZA), and meropenem-vaborbactam (MEV) were determined by broth microdilution.[6,7]

Hollow fiber infection model.[5] The extracapillary space (ECS) of each hollow fiber cartridge (C2011, FiberCell Systems) was inoculated with 20 mL of a log-phase bacterial suspension. Programmable syringe pumps infused all drugs (q8h for 7 days), recapitulating human plasma exposures. ECS samples were serially removed daily and antibacterial activity was assessed by quantitative culture with a lower limit of detection of 1.7 $\log_{10}$ colony forming units (CFU)/mL (or 1 $\log_{10}$ CFU/mL at 168 h). Resistant subpopulations were monitored using drug-supplemented (4x MIC) agar plates prepared daily. HFM validation studies were performed with marketed β-lactam-BLI (BL-BLI) combinations (**Figure 1**): Humanized CZA versus KP ATCC BAA-1705 (KP5, KPC-positive) was assessed to confirm the ability of the clinical dose to suppress resistance against a susceptible strain; humanized MEV versus KP 752285 (KP1, OXA-48-positive) tested the hypothesis that meropenem, unprotected from hydrolysis by OXA-48, would be ineffective in the HFM despite MEV susceptibility. FEP monotherapy served as growth control arms in FTB HFM studies (**Figure 2**).

Pharmacokinetic (PK) analysis. Target free (unbound) drug PK profiles for FTB were based on population parameters derived from healthy volunteers.[8] CZA and MEV profiles were informed by Prescribing Information[9] and published data,[10] respectively (**Table 2**). C2011 cartridge compatibility was confirmed for each test article using qualified LC-MS/MS methods; measured concentrations in the central circulation matched those in the ECS following administration of humanized doses (data not shown). In all pharmacodynamic HFM studies, concentrations of BLIs in central compartments were measured by qualified LC-MS/MS (Acquity HSS T3 or BEH C18, Waters™) methods to confirm clinical profiles were achieved in the HFM. Results for taniborbactam (**Figure 3**) are presented.

Results

Figure 1. HFM Validation using Marketed BL-BLIs vs. Susceptible Strains

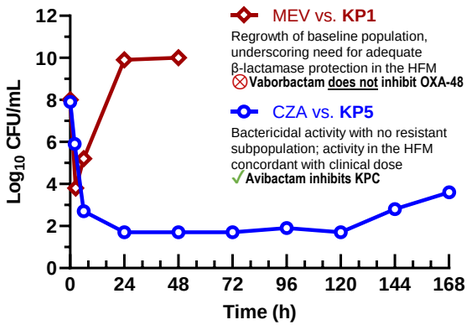

Table 1. Characterization of Strains Assessed in the Hollow Fiber Infection Model

Key	Bacterial isolate	Key β-Lactamases	Broth microdilution MIC (μg/mL)			
			FEP	FTB	CZA	MEV
EC1	*E. coli* AR 0055	CMY-6, **NDM-1**	>128	2	>128	64
KP1	*K. pneumoniae* 752285	CTX-M-15, OXA-48	>128	0.25	0.5	2
KP2	*K. pneumoniae* AR 0126	KPC-2	16	1	0.25	0.03
KP3	*K. pneumoniae* AR 0135	SHV-12, **VIM-1**	128	1	>128	8
KP4	*K. pneumoniae* AR 0145	CTX-M-15, **NDM-1**	128	0.25	>128	64
KP5	*K. pneumoniae* ATCC BAA-1705	KPC-2	32	0.25	1	0.016
PA1	*P. aeruginosa* 2235344	PER-1	32	2	8	16
PA2	*P. aeruginosa* AR 0054	**VIM-4**	128	8	128	>128
PA3	*P. aeruginosa* AR 0357	VEB-1	128	8	8	8

FEP, cefepime; FTB, cefepime-taniborbactam; CZA, ceftazidime-avibactam; MEV, meropenem-vaborbactam
Metallo-β-lactamases are bolded. MIC values are modes from at least three replicates.

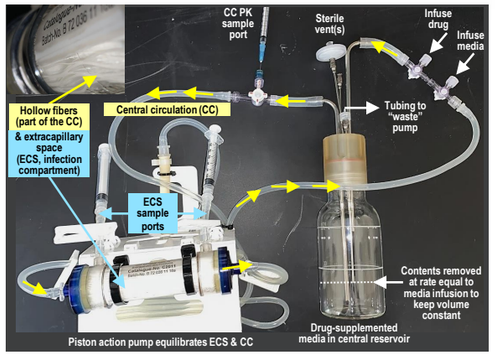

Figure 2. Activity of Cefepime-Taniborbactam against Enterobacterales (left) and *P. aeruginosa* (right) Strains Assessed in the Hollow Fiber Infection Model

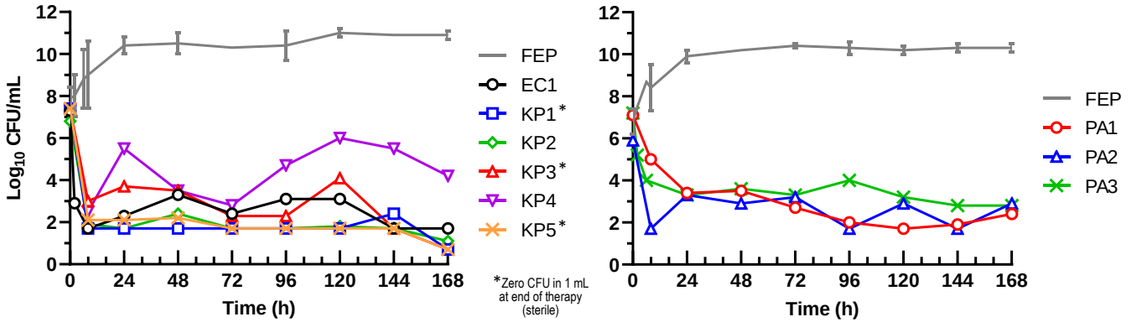

Figure 3. Taniborbactam Clinical Target Concentration-Time Profile and Measured Concentrations in Cefepime-Taniborbactam HFM Assessments

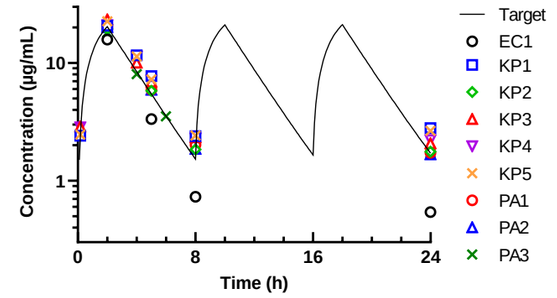

Table 2. Comparison of Human Pharmacokinetic Profiles to those Targeted in the Hollow Fiber Infection Model (HFM)

Clinical Dose Simulation		Time (%) above MIC for MIC of:							AUC_{0-24} (μg/mL*h)	$C_{max,0-24}$ (μg/mL)
		0.5	1	2	4	8	16	32		
Ceftazidime 2g q8h, 2 h infusion	HFM	100	100	100	100	84	62	38	676	68
	Human	100	100	100	100	93	70	44	771	81
Avibactam 0.5g q8h, 2 h infusion	HFM	100	91	71	48	20	0	0	110	11
	Human	92	77	61	44	22	0	0	106	13
Meropenem 2g q8h, 3 h infusion	HFM	100	100	100	100	80	58	31	550	52
	Human	100	100	100	97	77	57	32	551	51
Vaborbactam 2g q8h, 3 h infusion	HFM	100	100	100	97	77	55	25	494	46
	Human	100	100	100	100	83	58	23	494	42
Cefepime 2g q8h, 2 h infusion	HFM	100	100	100	100	85	61	34	620	62
	Human	100	100	100	100	85	59	35	688	78
Taniborbactam 0.5g q8h, 2 h infusion	HFM	100	100	94	71	46	15	0	208	21
	Human	100	100	92	70	46	17	0	208	21

AUC(0-24), area under the concentration-time curve from time 0 to 24 h; C(max,0-24), maximum concentration from time 0 to 24 h

Conclusions

- **In a 7-day hollow fiber infection model, human exposures of cefepime-taniborbactam (FTB) maintained bactericidal activity against 9 diverse and challenging clinical strains that produce serine β-lactamases, including ESBLs, and/or metallo-β-lactamases.**
- **Resistant subpopulations did not emerge on cefepime-taniborbactam treatment.**
- **These results support the clinical development of FTB and inform understanding of its potential role in SBL+ and/or MBL+ gram-negative bacterial infections.**

References

1. Hamrick et al. *Antimicrob Agents Chemother* 2020; 64(3):e01963-19.
2. Abdelraouf et al. *J Antimicrob Chemother* 2020; 75(12):3601-10.
3. Abdelraouf K, Nicolau DP. *J Antimicrob Chemother* 2023; 78(3):692-702.
4. Lasko et al. *J Antimicrob Chemother* 2022; 77(2):443-7.
5. Bulitta et al. *Antimicrob Agents Chemother* 2019; 63(5):e02307-02318.
6. Clinical and Laboratory Standards Institute. M100, 33rd edition (2023).
7. Clinical and Laboratory Standards Institute. M07, 11th edition (2018).
8. Dowell et al. *Antimicrob Agents Chemother* 2022; 66(9):e0025322.
9. AVYCAZ® Prescribing Information. Allergan USA, Inc.; 2019.
10. Sabet et al. *Antimicrob Agents Chemother* 2018; 62(2):e01969-17.

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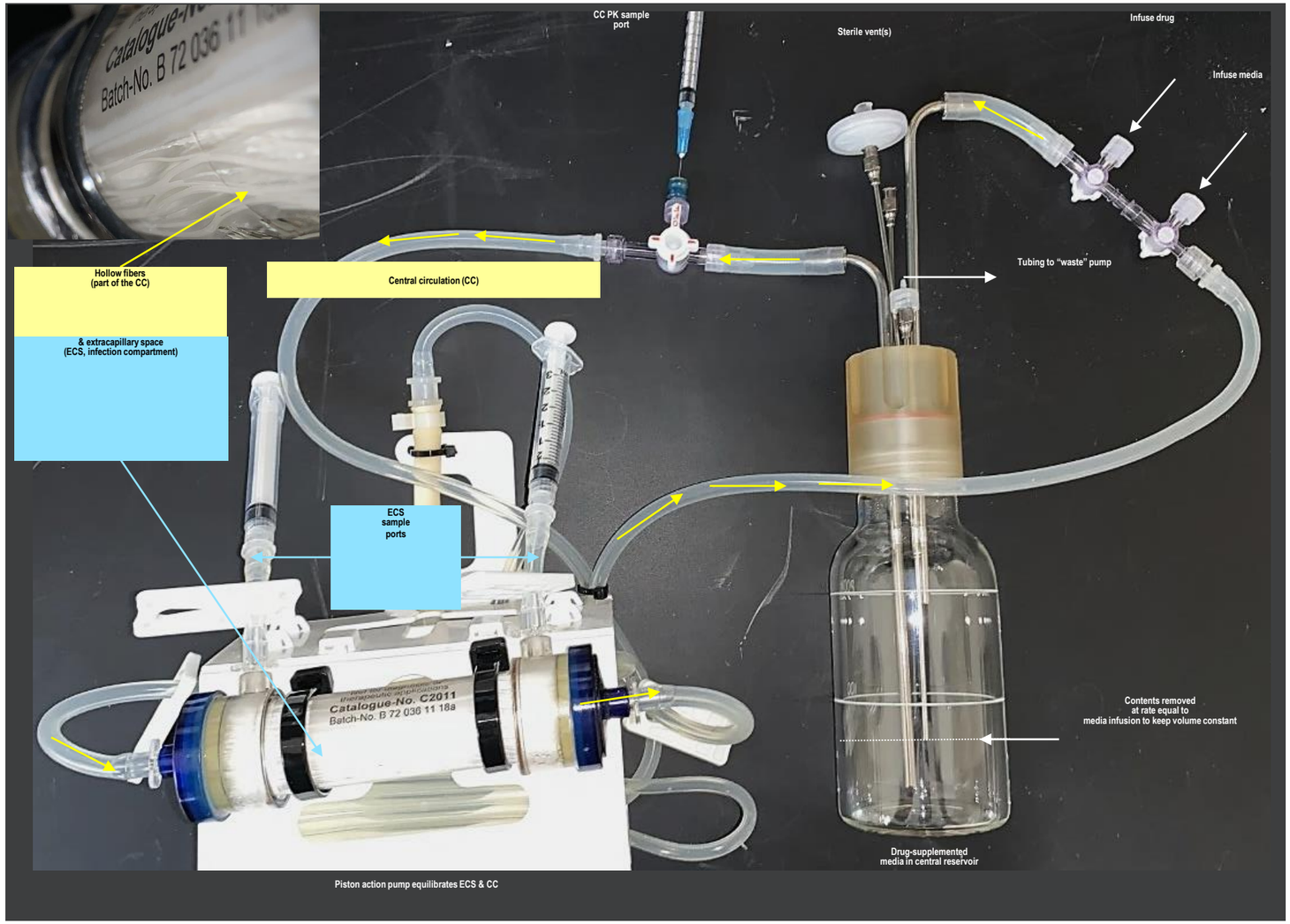
CC PK sample port
Sterile vent(s)
Infuse drug
Infuse media
Tubing to "waste" pump
Hollow fibers (part of the CC)
Central circulation (CC)
& extracapillary space (ECS, infection compartment)
ECS sample ports
Contents removed at rate equal to media infusion to keep volume constant
Drug-supplemented media in central reservoir
Piston action pump equilibrates ECS & CC