Lysin CF-301 (exebacase) Administered in Addition to Vancomycin (VAN) Suppresses the Emergence of Reduced Susceptibilities to VAN Within Cardiac Vegetations in a Rabbit Model of MRSA Infective Endocarditis (IE)

SUNDAY-535

Jun Oh[1], Wessam Abdelhady[2], Yan Xiong[2,3], Steven Jones[1], Cara Cassino[1], Arnold S. Bayer[2,3], and Raymond Schuch[1]

[1]ContraFect Corporation, Yonkers, NY, USA; [2]LA Biomed, Torrance, CA, USA; [3]Geffen Sch. Of Med. at UCLA, Los Angeles, CA, USA

Contact Info:
Raymond Schuch
ContraFect Corporation
rschuch@contrafect.com

Abstract

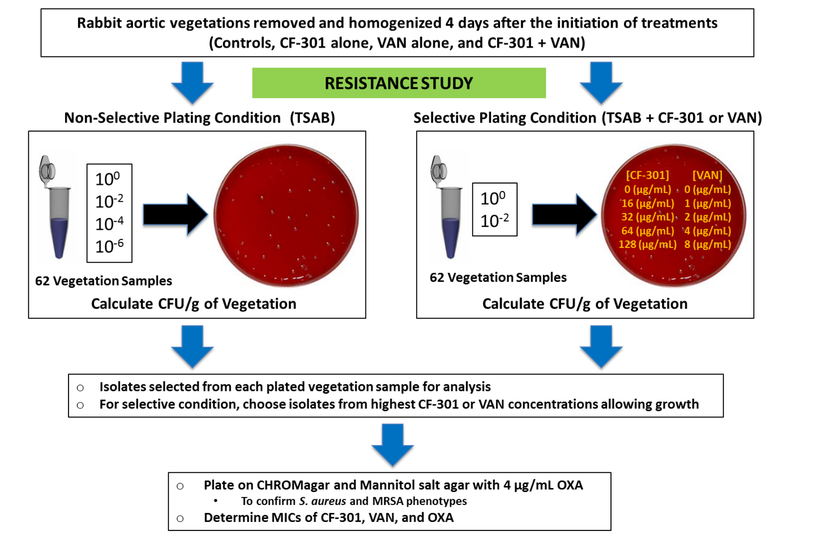
Background: CF-301 (exebacase) is a novel, recombinantly-produced, bacteriophage-derived lysin (cell wall hydrolase) which is in Phase 2 for the treatment of *S. aureus* endovascular infections, including bacteremia and IE, used in addition to standard- of-care antibiotics. We utilized the MRSA rabbit IE model to assess *in vivo* changes in the MIC of both CF-301 and VAN in animals treated with VAN alone, CF-301 alone or CF-301 + VAN.

Methods: The well-characterized indwelling catheter-induced model of aortic valve IE in rabbits due to MRSA was employed. Animals were treated with: **i**) VAN alone (7.5 mg/kg, IV BID x 4 d); **ii**) CF-301 alone (range = 0.35 mg/kg to 2.8 mg/kg; single-dose IV); **iii**) CF-301 + VAN; or **iv**) vehicle [controls]. At 24 h after the last dose of VAN, cardiac vegetations were sterilely removed from all groups and quantitatively cultured. To assess potential emergence of CF-301 resistance, vegetations from CF-301 treatment groups (alone or with VAN) were parallel-plated on TSAB (non-selective condition) or TSAB supplemented with CF-301 over a range of concentrations (selective condition). To study the potential emergence of VAN resistance, vegetations from the VAN treatment groups (alone and with CF-301), were also parallel-plated on TSAB ± VAN. Multiple colonies from each plating condition were subcultured and both CF-301 and VAN MICs for each colony were determined. MIC values for oxacillin (OXA) were also determined based on previous findings that treatments with CF-301 result in decreased OXA MICs, consistent with a "see-saw effect"(1).

Results: For animals treated with CF-301 alone, no CF-301 MIC change was observed in 100% of isolates (n=88) from non-selective media and a modest (2-fold) CF-301 MIC increase (i.e., from 1 to 2 µg/mL) was observed in 12.6% of isolates (n=88) from selective media. For animals treated with VAN alone, a 2-fold increase in VAN MIC was observed for 12.5, 2.5, and 15% of all isolates obtained from non-selective, CF-301 and VAN selective media conditions, respectively. Of note, irrespective of plating conditions, for all isolates obtained from animals treated with CF-301 + VAN, there were no shifts in VAN nor CF-301 MICs. Treatment with CF-301 regimens also resulted in significant OXA MIC decreases (64 to ≤2 µg/mL); interestingly, these decreases in OXA MIC were not always accompanied by increases in the CF-301 MIC.

Conclusions: CF-301 treatment of MRSA IE alone, and in the presence of VAN, demonstrated a low propensity for *in vivo* emergence of reduced susceptibility to CF-301. Importantly, combined CF-301 + VAN therapy prevented the development of increased VAN MICs among vegetation isolates. The suppression of increasing VAN MICs with CF-301 + VAN is consistent with previous *in vitro* findings, and is concordant with our prior findings of similar *ex vivo* and *in vitro* studies using daptomycin. The induction of the OXA "see-saw"-like effect is an added important feature of CF-301 treatment.

Study Outline



Non-Selective Plating Condition

Analysis of CF-301 (exebacase), VAN, and OXA MICs for isolates recovered on TSAB

Treatment Groups[a]	Log_{10} CFU/g of Vegetation[b]	n[c]	CF-301 MIC (µg/mL)[d]					VAN MIC (µg/mL)[e]				OXA MIC (µg/mL)[f]						
			0.25	0.5	1	2	4	0.5	1	2	4	<2	4	8	16	32	64	128
Pre-Treatment Control	5.46±1.78	41			41				41							5	36	
Buffer Treatment Control	7.26±1.54	32			31	1			32					2		25	5	
VAN (7.5)	6.17±2.42	32			31	1			28	4						16	16	
CF-301 (1.4)	8.24±0.02	24			24				24			7				7	10	
CF-301 (0.7)	7.93±0.12	24		1	23				24						1		23	
CF-301 (0.35)	8.17±0.58	24		3	21				24			6				7	11	
CF-301 (0.18)	8.65±0.58	16			16				16								16	
CF-301 (2.8) VAN (7.5)	<LOD																	
CF-301 (1.4) VAN (7.5)	2.92±1.73	12			12				12								12	
CF-301 (0.7) VAN (7.5)	4.23±2.92	24			24				24							10	14	
CF-301 (0.35) VAN (7.5)	3.34±0.91	34			34				34							10	24	

[a]Number in parenthesis represents dose in mg/kg
[b]LOD = limit of detection
[c]Number of isolates analyzed in MIC assays in each group
[d]The CF-301 MIC of *S. aureus* strain MW2 is 1 µg/mL
[e]The VAN MIC of *S. aureus* strain MW2 is 1 µg/mL
[f]The OXA MIC of *S. aureus* strain MW2 is 64 µg/mL

No decreased susceptibility to CF-301

CF-301 suppressed VAN MIC shift

CF-301 exposure resulted in decreased OXA MICs

- No decreased susceptibility to CF-301 was observed
- A 2-fold increase in the VAN MIC (from 1 to 2 µg/mL) was observed in VAN alone treatment
- CF-301 suppressed VAN MIC shift
- CF-301 treatment resulted in a decrease in OXA MICs (from 64 to ≤2 µg/mL) for a subset of isolates

Selective Plating on CF-301 (exebacase)

Analysis of CF-301 (exebacase), VAN, and OXA MIC of isolates recovered from TSAB supplemented with a range of CF-301 concentrations

Treatment Groups[a]	Log10 CFU/g of Vegetation[b]					n[c]	CF-301 MIC (µg/mL)[d]					VAN MIC (µg/mL)[e]				OXA MIC (µg/mL)[f]						
	0	16	32	64	128		0.25	0.5	1	2	4	0.5	1	2	4	<2	4	8	16	32	64	128
Pre-Treatment Control	5.46±1.78	4.33±1.75	3.91±1.48	2.53±0.31	<LOD	35			35				35							12	23	
Buffer Treatment Control	7.26±1.54	3.37±1.13	4.95±1.81	3.19±1.61	<LOD	44			44				44							35	9	
VAN (7.5)	6.17±2.42	4.85±1.88	4.41±1.71	3.82±1.37	<LOD	40			38	2			39	1						17	23	
CF-301 (1.4)	8.24±0.02	7.1±0.06	5.97±0.03	4.23±1.74	<LOD	24			20	4			24			3				8	13	
CF-301 (0.7)	7.93±0.12	6.7±0.14	5.78±1.09	3.81±1.84	<LOD	24		2	19	3			24						3	1	20	
CF-301 (0.35)	8.17±0.58	6.96±0.21	6.42±0.49	5.5±0.62	3.4±1.33	24			22	2			24			7					17	
CF-301 (2.8) VAN (7.5)	<LOD	<LOD	<LOD	<LOD	<LOD																	
CF-301 (1.4) VAN (7.5)	2.92±1.73	2.51±1.06	2.46±0.9	2.35±0.57	<LOD	12			12				12							1	11	
CF-301 (0.7) VAN (7.5)	4.23±2.92	3.78±2.16	3.73±2.10	2.86±1.31	<LOD	17			17				17							8	9	
CF-301 (0.35) VAN (7.5)	3.34±0.91	3.41±1.04	2.92±0.66	<LOD	<LOD	14			14				14								14	

[a]Number in parenthesis represents dose in mg/kg
[b]LOD = limit of detection
[c]Number of isolates analyzed in MIC assays in each group
[d]The CF-301 MIC of *S. aureus* strain MW2 is 1 µg/mL
[e]The VAN MIC of *S. aureus* strain MW2 is 1 µg/mL
[f]The OXA MIC of *S. aureus* strain MW2 is 64 µg/mL

≤2-fold shift increase

CF-301 exposure resulted in decreased OXA MICs

- A modest ≤2-fold increase in the CF-301 MIC observed
- CF-301 treatment resulted in a decrease in OXA MICs (from 64 to ≤2 µg/mL) for a subset of isolates

Selective Plating on VAN

Analysis of VAN MICs of isolates recovered from TSAB supplemented with a range of VAN concentrations

Groups[a]	Log10 CFU/g of Vegetation[b]				n[c]	VAN MIC (µg/mL)[d]				
	0	1	2	4		0.25	0.5	1	2	4
Pre-Treatment Control	5.46±1.78	3.16±0.71	<LOD	<LOD	42			42		
Buffer Treatment Control	7.26±1.54	5.39±1.66	<LOD	<LOD	31			31		
VAN (7.5)	6.17±2.42	4.42±2.11	2.72±0.5	2.46±0.4	46			39	7	
CF-301 (1.4)	8.24±0.02	6.53±0.58	<LOD	<LOD	24			24		
CF-301 (0.7)	7.93±0.12	6.34±0.10	<LOD	<LOD	24			24		
CF-301 (0.35)	8.17±0.58	6.54±0.51	<LOD	<LOD	24			24		
CF-301 (2.8) VAN (7.5)	<LOD	<LOD	<LOD	<LOD						
CF-301 (1.4) VAN (7.5)	2.92±1.73	2.61±0.44	<LOD	<LOD	16			16		
CF-301 (0.7) VAN (7.5)	4.23±2.92	3.09±1.29	<LOD	<LOD	29			29		
CF-301 (0.35) VAN (7.5)	3.34±0.91	2.68±0.35	<LOD	<LOD	25			25		

[a]Number in parenthesis represents dose in mg/kg
[b]LOD = limit of detection
[c]Number of isolates analyzed in MIC assays in each group
[d]The VAN MIC of *S. aureus* strain MW2 is 1 µg/mL

CF-301 suppressed VAN MIC shift

- No decreased susceptibility to CF-301 was observed (data not shown)
- A 2-fold increase in the VAN MIC (from 1 to 2 µg/mL) was observed in VAN alone group
- Suppression of VAN MIC shift at all doses tested

Summary

- CF-301 (exebacase) has a low propensity for resistance in this model, consistent previous in vitro findings and *ex vivo* IE model using DAP
- Emergent resistance to CF-301 (+/- VAN) was not observed in the rabbit IE model
- We expect that CF-301 variants with MIC ≤2 µg/mL will remain susceptible to the clinical CF-301 dose of 0.25 mg/kg, based on previous exposure target attainment animal studies and PK/PD modeling (2)
- CF-301 suppresses increases in VAN MICs when used in combination
- CF-301 treatment can resensitize MRSA strains to OXA

References

1. Oh et al., 2017 (ASM Poster: Friday 330)
2. Rotolo et al., 2016 (ASM Poster: Saturday LB-053)