02032

Comparison of iron-depleted (ID) versus cation-adjusted Mueller-Hinton broth (CAMHB) media for cefiderocol susceptibility among carbapenemase-producing Enterobacterales (CPEs)

03. Bacterial susceptibility & resistance

3c. Susceptibility testing methods (incl. assay validation, phenotypic assays and comparative studies, excl. TB)

Likely attendance

Onsite

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Background

Cefiderocol is the first siderophore-conjugated cephalosporin that displays activity against Gram-negative bacteria, including CPEs. As cefiderocol requires low iron levels for optimal activity, CLSI and EUCAST recommend that MIC determination must be performed in ID-CAMHB. There are concerns about the ability of routine laboratories to prepare this broth. Additionally, commercial methods have been flagged with accuracy and reproducibility issues. We aimed to compare cefiderocol MICs obtained by ID-CAMHB and with regular CAMHB.

Methods

Clinical isolates (one per patient) from a multicenter and prospective study carried out in November 2021 (183 hospitals) were included. Isolates had a positive PCR (multiplex) for *bla*$_{KPC}$, *bla*$_{NDM}$, *bla*$_{OXA\text{-}48\text{-}like}$, *bla*$_{VIM}$ and/or *bla*$_{IMP}$. ID-CAMHB was obtained from a unique batch of CAMHB (BD DIfco) after chelation (Chelex-100, BioRad) and subsequently cations re-supplementation. Calcium, magnesium and iron levels were verified by automated spectrophotometric methods (ARCHITECT-c4100, US). For MIC determination, the same bacterial inoculum was used across methods. MIC was defined as the first well in which the reduction of growth corresponds to a button of <1mm or is replaced by the presence of light haze/faint turbidity. Strains displaying skipped wells (<0.3%) were excluded. Cefiderocol was interpreted using the EUCAST (S<=2-R>2mg/L) and CLSI (S<=4-R>=16mg/L) breakpoints. Categorical (CA) and essential (EA) agreements were calculated.

Results

Iron levels were <1.3µg/dl for ID-CAMHB (below limit of detection) and 24µg/dl for regular broth. A total of 435 CPEs were included (80.2% *Klebsiella pneumoniae*): 185 *bla*$_{KPC}$, 207

bla$_{NDM}$, 29 *bla*$_{KPC}$+*bla*$_{NDM}$, 12 *bla*$_{NDM}$+*bla*$_{OXA-163}$, and 2 *bla*$_{KPC}$+*bla*$_{NDM}$+*bla*$_{OXA-163}$. Overall, MIC50/90 was 1/>8 mg/L and 4/>16 for ID-CAMHB and CAMHB, respectively. MIC50/90, EA and CA are depicted in Fig-1 (total sample), Fig-2 (*bla*$_{KPC}$) and Fig-3 (*bla*$_{NDM}$ alone or in combinations).

Conclusions

The methods evaluated in this work showed high dispersion in the MIC values, especially among KPC producers. Contrary, among enzymes that confer reduced sensitivity to cefiderocol, as NDM, 83.2% of MIC values were within +/- 2log$_2$. Both methods were more homogeneous in the reporting of the interpretation categories, with variations according to the selected standard. It is urgently required to have in routine labs a simple, reliable, accurate and clinically validated methodology to assess cefiderocol susceptibility.

Scatterplot comparing MICs obtained in different broth media											
Fig 1 - Total sample (n 435)											
IRON DEPLETED CAMHB	>8							4	8	15	21
	8							4	15	6	17
	4						5	10	16	10	13
	2					1	5	15	10	19	18
	1				1	4	12	18	5	5	6
	0,5			1		5	29	16	5	2	
	0,25			1		12	10	10			
	0,12				1	12	11	2	2		
	<=0,06		1	3	2	26	17	4			
		<=0,06	0,12	0,25	0,5	1	2	4	8	16	>16
		Regular CAMHB									

	Iron depleted CAMHB	Regular CAMHB
MIC50	1	4
MIC90	>8	>16
CA%	CLSI:67.3%. EUCAST: 66.9%	
EA%	33.6%%	

Fig 2 - KPC producers (n 185)											
IRON DEPLETED CAMHB	>8							1	1		2
	8										3
	4						1			1	7
	2					1		1	3	15	11
	1				1		1	2	3	4	6
	0,5			1		2	11	11	5	2	
	0,25			1		4	8	8			
	0,12				1	7	7	1	1		
	<=0,06		1	3	2	26	15	4			
		<=0,06	0,12	0,25	0,5	1	2	4	8	16	>16
		Regular CAMHB									

	Iron depleted CAMHB	Regular CAMHB
MIC50	0,5	2
MIC90	2	>16
CA%	CLSI:72.4 %. EUCAST: 57.8 %	
EA%	6%	

Fig 3 - MBL and carbapenemase combinations (n 250)											
IRON DEPLETED CAMHB	>8							3	7	15	19
	8							4	15	6	14
	4						4	10	16	9	6
	2						5	14	7	4	7
	1					4	11	16	2	1	
	0,5					3	18	5			
	0,25					8	2	2			
	0,12					5	4	1	1		
	<=0,06						2				
		<=0,06	0,12	0,25	0,5	1	2	4	8	16	>16
		Regular CAMHB									

	Iron depleted CAMHB	Regular CAMHB
MIC50	4	8
MIC90	>8	>16
CA%	CLSI:65%. EUCAST: 74%	
EA%	53%	
MICs in mg/L	+/- 0 dils	+/- 1 dils / +/- 2 dils

Keyword 1
Antimicrobial susceptibility testing (AST)
Keyword 2
Antimicrobial resistance (AMR)
Keyword 3
cefiderocol

Conflicts of interest

Do you have any conflicts of interest to declare?
No