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Detection of bacterial pneumonia pathogens in patients with COVID-19 using a Commercial Microarray in comparison with a commercial multiplex PCR and culture

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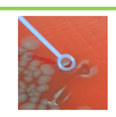
Background

Bacterial pneumonia is a challenging COVID-19 complication. Early detection of pathogens and timely initiation of appropriate treatment is important. We compared a commercial microarray assay with a commercial multiplex PCR and with culture.

Methods

100 respiratory samples (mostly BAL, n=88) from 100 patients with COVID-19 cared for on ICU were analysed using three methods. For culture, blood, chocolate and MacConkey agar were cultivated for 48h at 36±1°C, colony identification was performed by Vitek-MS, Vitek 2 or MicroScan WalkAway. For HB analysis, 20µl respiratory specimen was transferred to LINA buffer, vortexed and 20µl of BAL-LINA mixture was added to the PCR master mix. Only bacteria were taken into account for this study. For UY analysis, 180µl respiratory specimen was automatically extracted within the device.

Figure 1. Overview: applied methods in this study.

	Hyborg RED2; Cube Dx **(HB)**	Unyvero HPN; Curetis **(UY)**	Culture **(CU)**
System			
Method	Microarray	Multiplex PCR with array detection	
Target	Bacteria: 16S	20 bacterial pathogens	Bacteria capable of dividing
Time-to-result	2 h	4.5 h	> 24h

Results

Table 1. Detected bacterial species by three approaches. *

	Species	HB	UY	CU
Gram positive bacteria	*S. aureus*	5	14	8
	S. pneumoniae	7	2	0
Gram negative bacteria	E. coli	7	6	5
	K. pneumoniae / oxytoca / variicola / aerogenes	7	7	7
	E. cloacae complex	2	0	0
	P. mirabilis / spp.	2	1	1
	S. marcescens	4	4	4
	M. morganii	0	1	1
	S. maltophilia	0	2	0
	P. aeruginosa	6	5	3
	Burkholderia spp.	0	0	1
	A. baumannii / calcoaceticus complex	4	1	1
	H. influenzae	20	3	0
	M. catarrhalis	1	1	0
	Total	**65**	**47**	**31**

* Enterococci and normal constituents of the oral cavity/ respiratory tract are not displayed.

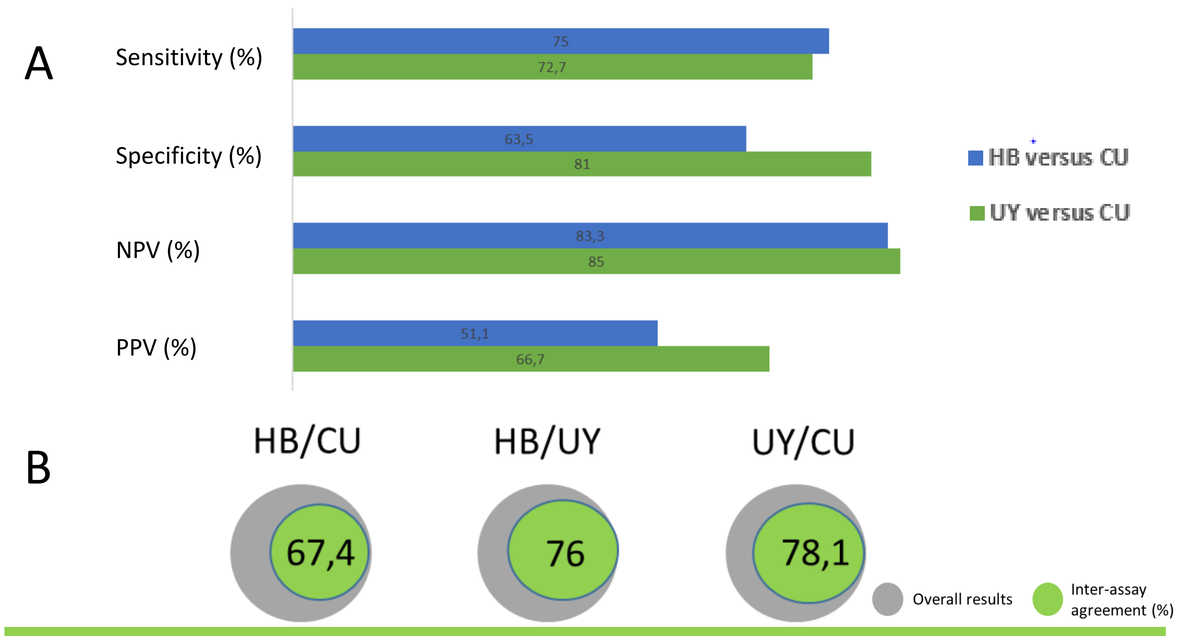


Figure 2. Performance (A) and agreement (B) of molecular tests versus culture approach.

Table 2. Patients' characteristics dependent on *H. influenza* detection.

	H. influenzae detected by HB or UY	Without detection of *H.influenzae*
Total	n = 20	n = 76
Median age (years)	56	59,5
Gender		
"-" male	16 (80)	56 (73,7)
"-"female	4 (20)	20 (26,3)
Material		
"-"Bronchoalveolar lavage	18 (90)	70 (92,1)
"-"Trachela secretion	2 (10)	4 (5,3)
"-"Bronchial secretion	0	1 (1,3)
"-"Sputum	0	1 (1,3)
C-reactive protein (CRP; mg/dl)	15,7 (± 6,44)	19,64 (± 9,96)
Leucocytes (per nl)	15,97 (± 11,35)	15,26 (±8,67)
Procalcitonin (PCT; ng/ml)	3,21 (± 5,26)	8,49 (± 29,83)
Invasive ventilation	20 (100)	74 (97,4) (1 unknown)
Extracorporeal membrane oxygenation (ECMO)	14 (70) (6 unknown)	35 (46,1) (32 unknown)
Dexamethason (6 mg IV for 6 to 10 days)	16 (80) (4 unknown)	41 (53,9) (35 unknown)
In-house mortality	14 (70)	48 (63,2)
***H. influenzae* effective antibiotic therapy started within 5 days before sampling**	14 (70)	59 (77,6)
Underlying conditions	(14 unknown)	(22 unknown)
"-" Chronic respiratory disease	1 (5)	5 (6,6)
"-" Cancer	0	2 (2,6)
"-" Diabetes	4 (20)	5 (6,6)
"-" Solid Organ transplant	1 (5)	0
"-" Cardiovascular disease	8 (40)	10 (13,2)

Data are presented as n (%) or as mean ± standard deviation

- 4 samples were excluded due to invalid performance (HB)
- In samples with *H. influenzae*, also *S. aureus* (1x) and *S.pneumonia* (2x) were detected
- *H. influenzae* detection was best by HB, followed by UY and worst by CU

Conclusion

- HB assay can be a valuable and time-saving technology for the direct detection for respiratory pathogens
- HB assay requires minimal hands-on-time

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