Multi-Center Evaluation of the VITEK®MS System for Mass Spectrometric Identification of Non-*Enterobacteriaceae* Gram Negative Bacilli

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BACKGROUND AND OBJECTIVES

The accurate, rapid identification of the non-*Enterobacteriaceae* Gram negative bacilli (NEGNB) is critical for antibiotic selection, especially for multidrug resistant NEGNB such as *Acinetobacter* sp., *Pseudomonas* sp. and *Stenotrophomonas* sp. NEGNB also cause nosocomial infections and identification triggers prompt initiation of infection control measures. Matrix-assisted laser desorption ionization time-of-flight (MALDI-TOF) mass spectrometry (MS) can potentially provide NEGNB colony identification within minutes.

This multi-center study assessed the performance of VITEK®MS and version 2.0 of the knowledge base (bioMérieux, Marcy l'Etoile, France) for identifying NEGNB clinical isolates.

MATERIALS AND METHODS

I. Clinical Trial Sites: This study was performed at five geographically diverse US sites (California, Ohio, Missouri, New York and Massachusetts).

II. Strains Tested: Unique NEGNB clinical isolates (n=558), comprising 18 genera and 33 species (*Achromobacter*: n=41, *Acinetobacter*: n=108, *Aeromonas*: n=35, *Alcaligenes*: n=12, *Bordetella*: n=25, *Brevundimonas*: n=7, *Burkholderia*: n=40, *Chryseobacterium*: n=8, *Elizabethkingia*: n=10, *Ochrobactrum*: n=10, *Pasteurella*: n=14, *Pseudomonas*: n=109, *Ralstonia*: n=10, *Rhizobium*: n=14, *Sphingobacterium*: n=15, *Sphingomonas*: n=9, *Stenotrophomonas*: n=53, *Vibrio*: n=38).
A. baumannii complex included *A. baumannii* (n=19), *A. baumannii* complex (n=34), *A. calcoaceticus* (n=8),*A. nosocomialis* (n=4).
Isolates were obtained from test sites or, for rare isolates, provided by bioMérieux.

III. Preparation and Testing: NEGNB, calibrant (*E. coli* ATCC 8739) and controls (*E. aerogenes* [ATCC 13048], *K. oxytoca* [ATCC 13182], *P. aeruginosa* [ATCC 10145], and *S. aureus* [ATCC 29213] were grown on appropriate media for <72 hr prior to testing (see below). Repeat testing of an isolate with an acceptable identification was not permitted.

IV. Interpretation: NEGNB identification and confidence values were determined by comparison of spectra to VITEK®MS v2.0 knowledge base. VITEK®MS results were compared to 16s rRNA DNA sequencing.

RESULTS: Table 1: VITEK MS Identification of NEGNB Isolates

Species	Isolates Tested n	Correct to Genus and Species n (%)[a]	Correct to Genus only n (%)	Total Correct Results[b] n (%)	Miss-ID to Genus and Species[c] n (%)	Miss-ID to Single Species only n (%)	Total Miss-ID[d] n (%)	Total no ID[e] n (%)
Achromobacter denitrificans	17	0	15 (88.2)	15 (88.2)	0	0	0	2 (11.8)
Achromobacter xyloxoxidans	24	0	22 (91.7)	22 (91.7)	0	0	0	2 (8.3)
Acinetobacter baumannii complex[g]	65	56 (86.2)	0	56 (86.2)	0	0	0	9 (13.8)
Acinetobacter haemolyticus	6	6 (100)	0	6 (100)	0	0	0	0
Acinetobacter junii	11	6 (54.5)	3 (27.3)	9 (81.8)	0	1 (9.1)	1	2 (18.2)
Acinetobacter lwoffii	26	22 (84.6)	1 (3.8)	23 (88.5)	0	0	0	3 (11.5)
Aeromonas hydrophila/caviae[h]	25	16 (64.0)	8 (32.0)	24 (96.0)	0	2 (8.0)	2 (8.0)	1 (4.0)
Aeromonas sobria	10	4 (40.0)	6 (60.0)	10 (100)	0	1 (10.0)	1 (10.0)	0
Alcaligenes faecalis ssp. *faecalis*	12	11 (91.7)	0 (0)	11 (91.7)	1 (8.3)	0	1 (8.3)	0
Bordetella bronchiseptica	10	2 (20.0)	3 (30.0)	5 (50.0)	0	0	0	5 (50.0)
Bordetella parapertussis	6	6 (100.0)	0 (0)	6 (100)	0	0	0	0
Bordetella pertussis	9	6 (66.7)	3 (33.3)	9 (100)	0	0	0	0
Brevundimonas diminuta	7	7 (100)	0	7 (100)	0	0	0	0
Burkholderia cepacia	9	3 (33.3)	5 (55.6)	8 (88.9)	0	0	0	1 (11.1)
Burkholderia multivorans	25	24 (96.0)	0	24 (96.0)	0	0	0	1 (4.0)
Burkholderia stabilis	6	0 (0)	6 (100)	6 (100)	0	0	0	0 (0)
Chryseobacterium indologenes	8	7 (87.5)	0	7 (87.5)	0	0	0	1 (12.5)
Elizabethkingia meningoseptica	10	10 (100)	0	10 (100)	0	0	0	0
Ochrobactrum anthropi	10	10 (100)	0	10 (100)	0	0	0	0
Pasteurella multocida	14	14 (100)	0	14 (100)	0	0	0	0
Pseudomonas aeruginosa	57	55 (96.5)	0	55 (96.5)	0	0	0	2 (3.5)
Pseudomonas fluorescens	19	15 (78.9)	3 (15.8)	18 (94.7)	0	0	0	1 (5.3)
Pseudomonas putida	25	20 (80.0)	2 (8.0)	22 (88.0)	0	1 (4.0)	1 (4.0)	3 (12.0)
Pseudomonas stutzeri	8	8 (100)	0	8 (100)	0	0	0	0
Ralstonia pickettii	10	8 (80.0)	0	8 (80.0)	0	0	0	2 (20.0)
Rhizobium radiobacter	14	10 (71.4)	0	10 (71.4)	2 (14.3)	0	2 (14.3)	2 (14.3)
Sphingobacterium multivorum	5	4 (80.0)	0	4 (80.0)	0	0	0	1 (20.0)
Sphingobacterium spiritivorum	10	10 (100)	0	10 (100)	0	0	0	0
Sphingomonas paucimobilis	9	9 (100)	0	9 (100)	0	0	0	0
Stenotrophomonas maltophilia	53	51 (96.2)	0	51 (96.2)	1 (1.9)	0	1 (1.9)	1 (1.9)
Vibrio cholerae	11	10 (90.9)	0	10 (90.9)	0	0	0	1 (9.1)
Vibrio parahaemolyticus	16	14 (87.5)	1 (6.3)	15 (93.8)	0	0	0	1 (6.3)
Vibrio vulnificus	11	10 (90.9)	0	10 (90.9)	0	0	0	1 (9.1)

Total Isolates Tested	Correct to Genus and Species[a]	Correct to Genus only	Total Correct Results[b]	MisID to Genus and Species[c]	MisID to Single Species only	Total MisID[d]	Total No ID[e]
N= 558	434	78	512	4	5	9	42
%	77.8%	14.0%	91.8%	0.7%	0.9%	1.6%	7.5%

MisID: incorrect identification; ID: identification

[a] Correct ID to genus level with either 2 or 3 species listed including correct species (n=73) or without correct species (n=5); or single ID correct to genus but incorrect to species (n=5).
[b] Total IDs correct to genus level only (n=78) or genus and species (n=434).
[c] Single ID correct to genus level but incorrect to species level (n=5). Results are included in correct to genus level category.
[d] Total incorrect ID to genus and species levels (n=4) or single ID correct to genus level but incorrect to species level (n=5).
[e] Total with no ID due to either mixed genera (n=14) or 'no ID in knowledge base' (n=28)

RESULTS: Table 3: Discordant Results

Number Isolates	Organism	VITEK MS Identification(s) Incorrect at Species Level[a]
1	*Acinetobacter junii*	*Acinetobacter haemolyticus*
2	*Aeromonas caviae*	*Aeromonas sobrias*
1	*Aeromonas sobria*	*Aeromonas hydrophila/caviae*
1	*Pseudomonas putida*	*Pseudomonas viridiflava*
Number Isolates	**Organism**	**VITEK MS Identification(s) Incorrect at Genus Level**
1	*Alcaligenes faecalis* ssp *faecalis*	*Staphylococcus aureus*
1	*Rhizobium radiobacter*	*Obesumbacterium proteus*
1	*Rhizobium radiobacter*	*Achromobacter denitrificans* and *Achromobacter xylosoxidans*
1	*Stenotrophomonas maltophilia*	*Ochrobactrum anthropi*

SUMMARY AND CONCLUSIONS

Summary

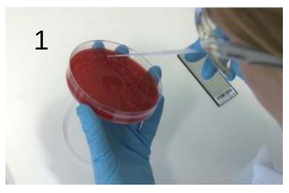
- Overall, VITEK®MS identified 91.8% of NEGNB correct to species-level (77.8%) or genus-level (14.0%).
- Overall there were 4 isolates (0.7%) misidentified to genus-level and 5 isolates (0.9%) misidentified to species-level.
- VITEK®MS called 14 NEGNB (2.5%) mixed genera and no ID was provided for the remaining 28 NEGNB (5%).

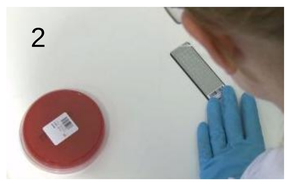
Conclusions

- VITEK®MS provides a highly accurate and rapid identification of NEGNB.
- Time to results in minutes versus hours or days for traditional microbiologic testing allows the Laboratory to provide clinically relevant results in a time frame that can have a major impact on patient care and outcome**.**

VITEK MS SET-UP

A 1 µl loop was used to apply the bacterial isolates to wells on a disposable bar-coded target slide (Figures 1 and 2).





VITEK MS SET-UP, DATA ANALYSIS AND INTERPRETATION

Matrix solution (α-cyano-4-hydroxycinnamic acid) was added to each well and dried (Figure 3). Slides were inserted into VITEK MS (Figure 4).

Mass spectra were generated using the VITEK®MS and mass spectra interpreted by VITEK®MS v2.0 knowledge base.

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