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Comparative evaluation of Diatherix TEM-PCR™ and BioFire FilmArray® in the detection of viral and bacterial respiratory pathogens

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Background

The speed and efficiency of etiologic determination of influenza-like illness (ILI) has been greatly improved with multiplex diagnostic assays. We conducted a comparative evaluation of two multiplex PCR-based platforms with comparable coverage of viral and bacterial respiratory pathogens.

Methods

- Acute Respiratory Infection Consortium (ARIC) Natural History Study (NHS) is an observational, longitudinal study of influenza-like illness (ILI). Since 2009, we enrolled otherwise healthy military personnel and beneficiaries into ARIC at five military treatment facilities across the continental United States.
- Eligibility: patients presenting for care <72h after the onset of ILI, defined as fever (temperature of 100.4° F or greater at the time of evaluation, or by self-report) and sore throat or one of the following respiratory symptoms: cough, sputum production, shortness of breath, or chest pain. Patients with underlying medical conditions were excluded from participation.
- Nasal/oral specimens were collected by study coordinators using nylon flocked swabs (Copan Diagnostics, Corona, CA). at the time of enrollment and tested by two assays: [1] target-enriched multiplex PCR (TEM-PCR™; Diatherix Laboratories, Inc.; Huntsville, AL) and [2] BioFire FilmArray® Respiratory Panel (FilmArray®; BioFire Diagnostics, Salt Lake City, UT). Viral and bacterial pathogens covered by the two assays are listed in Table 1. Specimens (n=396) were selected based on availability, and tests were performed in separate laboratories.

Table 1. Respiratory pathogens covered by TEM-PCR™ and BioFire FilmArray®

Viral pathogens	Diatherix TEM-PCR™	BioFire FilmArray®
Human Rhinovirus and Enterovirus	✓ HRV, Coxackievirus/Echovirus	✓ HRV/Enterovirus
Influenza A (pH1N1, H1N1 and H3N2)	✓	✓
Influenza B	✓	✓
Coronavirus (HKU1, NL63, 229E, and OC43)	✓	✓
Respiratory Syncytial virus (A and B)	✓	✓
Parainfluenza (PIV1 -4)	✓	✓
Human Metapneumovirus	✓	✓
Adenovirus	✓	✓
Bocavirus	✓	
Bacterial pathogens		
Streptococcus pneumoniae	✓	
Haemophilus influenzae	✓	
Moraxella catarrhalis	✓	
Staphylococcus aureus (MSSA/MRSA)	✓	
Streptococcus pyogenes	✓	
Klebsiella pneumoniae	✓	
Mycoplasma pneumoniae	✓	✓
Acinetobacter baumannii	✓	
Pseudomonas aeruginosa	✓	
Neisseria meningitidis	✓	
Bordetella pertussis	✓	✓
Chlamydophila pneumoniae	✓	✓

- Specimens were also tested by single-plex PCR for influenza at the Naval Health Research Center (San Diego, CA) using the CDC human influenza virus real-time RT-PCR diagnostic panel.
- Kappa (κ) coefficients and 95% confidence intervals (CI) were computed. In addition, the sensitivity and specificity of each platform for detecting influenza virus was computed using single-plex PCR tests as a gold standard. Statistical analyses were performed using SAS software (Version 9.3; SAS Institute, Cary, NC).

Results

- Of the 396 specimens tested by both platforms, TEM-PCR™ and FilmArray® detected at least one viral pathogen among 256 (64.7%) and 262 (66.2%) specimens, respectively (Figure 1). The frequency of viral co-detection (i.e. two or more viral pathogens in a specimen) was 7.3% for TEM-PCR™ and 5.8% for BioFire FilmArray®.

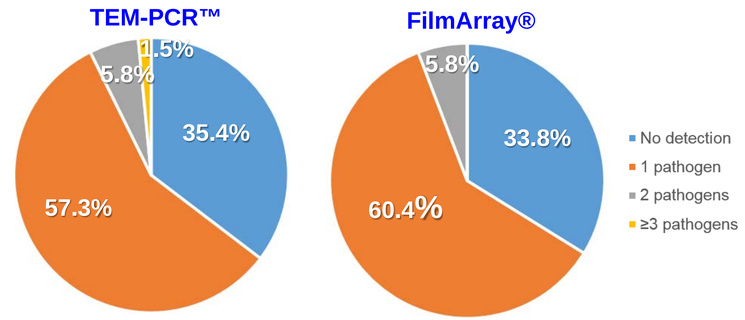

Figure 1. Number of viral pathogens detected among 396 ARIC NHS patients using TEM-PCR™ and FilmArray® Panels

- Viral pathogens: TEM-PCR™ had a higher frequency of detection of influenza A/B, coronavirus, human metapneumovirus, RSV, and parainfluenza virus, while FilmArray® more frequently detected adenovirus and enterovirus. There was substantial pathogen-specific agreement between the panels, with the exception of Adenovirus and influenza B (Table 2).

Table 2. Viral pathogen-specific agreement between TEM-PCR™ and FilmArray®

		FilmArray®		Agreement	
	TEM-PCR™	Negative	Positive	*K*	95%CI
Adenovirus	Negative	387	7	0.19	(-0.14, 0.52)
	Positive	1	1		
Influenza A	Negative	358	2	0.76	(0.63, 0.88)
	Positive	12	24		
A(H1N1)pdm09	Negative	374	2	0.80	(0.66, 0.94)
	Positive	5	15		
A(H3N2)	Negative	380	0	0.71	(0.51, 0.91)
	Positive	7	9		
Influenza B	Negative	384	2	0.58	(0.3, 0.86)
	Positive	5	5		
Enterovirus (including HRV)	Negative	289	21	0.74	(0.66, 0.82)
	Positive	15	71		
Coronavirus	Negative	329	2	0.91	(0.86, 0.97)
	Positive	7	58		
Human metapneumovirus (hMPV)	Negative	362	3	0.86	(0.76, 0.95)
	Positive	5	26		
RSV	Negative	344	5	0.85	(0.77, 0.93)
	Positive	7	40		
Parainfluenzavirus	Negative	377	0	0.97	(0.92, 1)
	Positive	1	18		

- Bacterial pathogens: TEM-PCR™ had a higher frequency of detection of M. pneumoniae. The prevalence of C. pneumoniae and B. pertussis was too low to allow a comparative evaluation.

Table 3. Bacterial pathogen-specific agreement between TEM-PCR and FilmArray®

		FilmArray®		Agreement	
	TEM-PCR™	Negative	Positive	*K*	95%CI
M. pneumoniae	Negative	389	0	0.72	(0.42, 1)
	Positive	3	4		

- Both assays were sensitive and specific in detecting influenza A and B, when using CDC human influenza virus real-time RT-PCR diagnostic panel as a gold standard (Table 4).

Table 4. Sensitivity and specificity of detecting influenza virus (A/B) using TEM-PCR™ and FilmArray®

	FilmArray® Single-plex PCR				TEM-PCR™ Single-plex PCR			
	Negative	Positive	Sen.(%)	Spe.(%)	Negative	Positive	Sen.(%)	Spe.(%)
Influenza A								
Negative	368	2	90.48	98.13	359	1	95.24	95.73
Positive	7	19			16	20		
A(H1N1)pdm09#								
Negative	377	0	100.0	98.18	374	0	100.0	97.4
Positive	7	10			10	10		
A(H3N2)#								
Negative	385	0	100.0	100.0	379	0	100.0	98.44
Positive	0	9			6	9		

Two specimens were tested positive for influenza A by NHRC, but no subtype was identified and were excluded from the analysis. Sen: sensitivity; Spe: specificity

Conclusions

These results reveal a high degree of concordance between Diatherix Laboratories TEM-PCR™ and BioFire FilmArray® in the detection of viral respiratory pathogens. Low correlation between the panels with respect to adenovirus may be due to differences in assay inclusivity for detection of the specific targets.

Acknowledgements and Disclaimer

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