Enhancing Pathogen Identification Using a Comprehensive PCR System in Adult and Pediatric patients with Meningitis and a negative Gram stain

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Abstract

Background: Meningitis with a negative CSF Gram stain represents a diagnostic and therapeutic challenge, as the majority of the causative organisms are unknown. Novel and fast molecular techniques may increase the detection of the etiological agents.

Methods: Patients admitted to the 4 hospitals in Houston, TX between Nov. 2008 - Mar. 2014 with community-acquired meningitis (fever, headache, vomiting, photophobia, stiff neck, focal neurological symptoms), CSF cell count > 5 cells/mm³ and a negative CSF Gram stain were eligible. Residual patient CSF underwent additional testing by a research use only version of the Film ArrayTM Meningitis / Encephalitis panel (FA ME, BioFire Diagnostics, LLC). The panel requires 200 µL CSF and simultaneously tests for 6 bacteria (*S. pneumoniae, N. meningitidis, S. agalactiae, H. influenzae, L. monocytogenes, E. coli* K1), 8 viruses (Herpes simplex types 1, 2, 6, Cytomegalovirus, Epstein-Barr virus [**EBV**], Enterovirus, Parechovirus, Varicella zoster virus [**VZV**]) and 2 fungi (*Cryptococcus neoformans* and *gattii*).

Results: Of the 149 patients enrolled, 48 (32.2%) had residual CSF (38 adults, 10 children < age 18) available for FA ME testing. Pathogens were identified in 14 (29.2%) of 48 samples by standard evaluation and 15 (31.2%) by FA ME. Among FA ME results, viral pathogens were most common [EBV (8), HSV2 (3), VZV (3), HSV1 (1), enterovirus (1)], followed by bacterial [*S. pneumoniae* (2)] and fungal [*C. neoformans* (1)]. Co-detections were present in 6 patients (12.5%); EBV was present in all (6) along with VZV (2), HSV1 (1), HSV2 (1), *C. neoformans* (1), and *S. pneumoniae* (1). In 8 (16.6%) patients, FA ME identified pathogens not previously identified. Standard evaluations identified pathogens in 5 (15.2%) of 33 FA ME negative samples [West Nile Virus (**WNV**) (4), *Histoplasma capsulatum* (1)].

Conclusion:Testing with the FA ME panel resulted in pathogen detections not previously recognized and for which treatment is recommended. The FA ME panel did not detect, however, some pathogens identified by standard techniques; assays for WNV and *Histoplasma* are not contained on the panel. Rapid, comprehensive testing for the most common pathogens causing meningitis will aid in the diagnosis and treatment of patients with negative CSF Gram stains.

Background

- The capacity to rapidly identify the organisms is critical for the accurate diagnosis and treatment.
- Standard techniques require several days for initial identification of a pathogenic organism, and some organisms are not recognized using the standard techniques.
- The use of a comprehensive Polymerase chain reaction (PCR) system have expanded the range of pathogens that can be identified in clinical laboratories.



Results

			SPUTUM	URINE		STOOL			NW			FAME
etssia /IgG	Leptospira	West Nile V. IgG/IgM	Culture	Culture	Urinary Antigen	Сх	Enterovirus	Adenovirus	Viral Cx	Rapid RSV	Rapid Flu	PCR
					Histoplasma							EBV
				Candida albicans								EBV
					Legionella, Pneumococcus							Pneumococcus
												vzv
				Staphylococcus aureus	Pneumococcus							Pneumococcus
												EBV, Cryptococcus
			Pneumococcus									Pneumococcus
												HSV2
												HSV1, EBV
				Enterobacter aerogenes								VZV, EBV
												VZV, EBV
			Aspergillosis									EBV
												HSV2
												EBV, Pneumococcus
												Enterovirus

Patient Characteristics with Film Array™ Meningitis / Encephalitis panel

	n (%)				
Median age, years (mean-range)	38.3 (3 months –				
	82 years)				
Gender					
Male	21 (43.7%)				
Female	27 (56.2%)				
Adults	38 (79.1%)				
Children	10 (20.8%)				
Presentation Diagnosis					
Meningitis	25 (52.0%)				
Encephalitis	23 (47.9%)				
Presenting symptoms					
Fever	25 (52.0%)				
Headache	24 (50.0%)				
Stiff Neck	20 (41.6%)				
Photophobia	14 (29.1%)				
Seizure	6 (12.5%)				
CSF					
WBC (mean-range)	456 (7 –4850)				
Lymphocytes	56 (1-99)				
Protein	98 (9-460)				
Glucose	56 (1-139)				

Conclusions

- Testing with the FA ME panel resulted in pathogen detections not previously recognized and for which treatment is recommended.
- Rapid, comprehensive testing for the most common pathogens causing meningitis will aid in the diagnosis and treatment of patients with negative CSF Gram stains.

This poster contains information regarding assays that have not been cleared by the FDA for in vitro diagnostic use.

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