Utility of a Multiplex Gastrointestinal Panel to Aid in Epidemiological Outbreak Investigation of Shigella

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ABSTRACT

Background

Shigella is estimated to cause 400,000 infections annually in the US. Novel multiplex PCR gastrointestinal (GI) pathogen panels have the ability to identify pathogens rapidly, increase detection rates due to sensitivity, as well as clarify outbreaks in the community. The objective of this study was to identify and clarify the epidemiological findings of a recent Shigella outbreak in Rhode Island (RI) by combining the RI Department of Health (DOH) culture and epidemiological data, with the multiplex GI panel being investigated at Rhode Island Hospital (RIH) during the same time period.

Methods

Stool samples were prospectively evaluated from patients with suspected GI illness using the IUO FilmArray® GI Panel* (Biofire Diagnostics, Salt Lake city, UT) (*FDA cleared on 5/4/2014): a multiplex PCR assay that simultaneously detects multiple bacterial, viral, toxin gene, and parasitic targets, including Shigella/Enteroinvasive *E. coli* (EIEC). GI panel results for Shigella/EIEC were compared to those from standard stool culture and PCR. Culture isolates submitted to RI DOH had additional confirmation and Pulsed field gel electrophoresis (PFGE) typing to clarify epidemiology.

Results

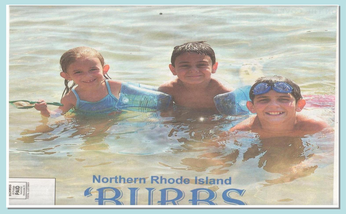
For specimens during the suspected outbreak period, where both a study sample and standard lab specimen were analyzed, the multiplex GI panel detected 10 cases of Shigella/EIEC compared to only 6 cases identified by culture. Cases positive by multiplex assay only were confirmed by an independent PCR with sequencing. The six culture positive shigella had identical pulse types based on PFGE typing and were concordant with other pulse types prevalent in RI during the outbreak.

Conclusions

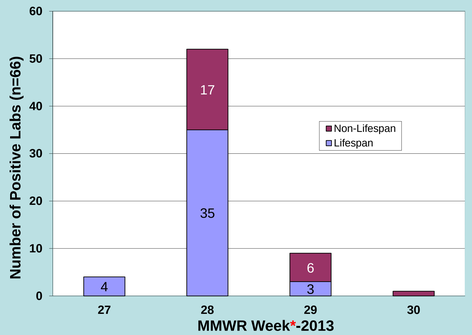
More shigella cases were identified by the multiplex GI assay compared to culture for patients presenting with diarrhea, but were unable to be confirmed as part of the suspected outbreak. Because most patient specimens were submitted from outpatient sites, it is possible that sensitivity of culture was compromised in transit with failure to identify true shigella positive patients. While having a more sensitive diagnostic tool, such as a GI multiplex could have added benefit in early reporting for public safety and epidemiological investigations, alternate technology will have to aid current culture isolation with PFGE for confirmation of these discrepant results.

RESULTS

Spring Lake, RI: Source of Shigella Outbreak during Rhode Island Summer- July 2013



Graph 1- Culture Positive Shigella Tests Associated with an Outbreak in Rhode Island July, 2013- RI DOH



*MMWR Week 27- 06/30-07/06; Week 28- 07/07- 07/13
MMWR Week 29- 07/14-07/20; Week 30- 07/21-07/27

Table 1- Patient and Specimen Information for Shigella Outbreak

Sample ID	Sample Collection Date	Filmarray testing Date	Patient and Specimen Information												Shigella/ EIEC		
			Demographics			Reason for Specimen Submission											
			Age	Sex	Patient Status	Stool Culture	*Vibrio*	*Yersinia*	Shiga Toxin	O&P Exam	Crypt/Gia- DFA	Virology	Tox. *C. difficile*	Other	FA[c]	Cx[d]	PCR[e]
140	07/08/13	07/11/13	1-5	M	OP[a]	Y	N	N	N	N	N	N	N	N	+	+	+
146	07/08/13	07/11/13	13-21	F	OP	Y	N	Y	N	N	N	N	N	N	+	+	+
153	07/08/13	07/11/13	1-5	M	ED[b]	Y	N	Y	N	N	N	N	N	N	+	+	+
155	07/09/13	07/11/13	22-64	F	OP	Y	N	N	N	N	Y	N	Y	N	+	-	+
156	07/10/13	07/11/13	1-5	F	OP	Y	N	N	N	N	Y	N	Y	N	+	-	+
158	07/10/13	07/11/13	1-5	F	OP	Y	N	Y	N	N	Y	N	N	N	+	+	+
160	07/10/13	07/11/13	1-5	M	OP	Y	N	Y	N	N	Y	N	Y	N	+	-	+
178	07/15/13	07/15/13	6-12	F	ED	Y	N	N	N	N	N	N	N	N	+	+	+
184	07/15/13	07/16/13	1-5	M	ED	Y	N	N	N	N	N	N	N	N	+	-	+
185	07/15/13	07/16/13	1-5	F	OP	Y	N	N	N	N	N	N	N	N	+	+	+

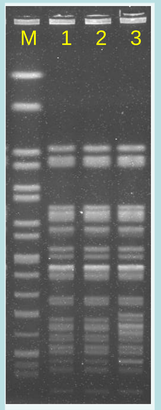
[a]Outpatient site, [b]Emergency Department, [c]Filmarray GI multiplex test
[d]Stool Culture, [e]Comparator PCR

RESULTS

Table 2- RI DOH PFGE patterns of the Outbreak isolates from Lifespan laboratories

Sample ID	Sample Collection Date	Filmarray testing Date	RI DOH PFGE upload Date	Demographics			Shigella/EIEC			RI DOH PFGE pattern
				Age	Sex	Patient Status	FA	Cx	PCR	
140	07/08/13	07/11/13	7/15/2013	1-5	M	OP	+	+	+	RIJ16X01.073
146	07/08/13	07/11/13	7/15/2013	13-21	F	OP	+	+	+	RIJ16X01.073
153	07/08/13	07/11/13	7/18/2013	1-5	M	ED	+	+	+	RIJ16X01.073
155	07/09/13	07/11/13		22-64	F	OP	+	-	+	
156	07/10/13	07/11/13		1-5	F	OP	+	-	+	
158	07/10/13	07/11/13	7/22/2013	1-5	F	OP	+	+	+	RIJ16X01.073
160	07/10/13	07/11/13		1-5	M	OP	+	-	+	
178	07/15/13	07/15/13	7/22/2013	6-12	F	ED	+	+	+	RIJ16X01.073
184	07/15/13	07/16/13		1-5	M	ED	+	-	+	
185	07/15/13	07/16/13	7/22/2013	1-5	F	OP	+	+	+	RIJ16X01.073

Representative PFGE pattern of outbreak isolates



- Stool DNA Samples from Culture negative and positive patients tested with a spp. specific PCR for Shigella/EIEC turned positive
- Bidirectional sequencing and subsequent BLAST search identified both Culture positive and negative samples as Shigella/EIEC
- While PFGE helped track the epidemiology of the Culture positive outbreak isolates, the multiplex Filmarray GI panel testing proves to be a promising complementary culture independent technology for early and rapid public health response to contain outbreaks.

CONCLUSION

- Appropriate and timely transport of stool specimen is critical for recovery of enteric pathogens in standard stool culture.
- Failure to culture and identify Shigella isolates indicates that sensitivity might have been compromised in transit since 70% samples were submitted to the lab from outpatient sites (Table-1).
- Having a more sensitive diagnostic tool, such as a GI multiplex could have added benefit in early reporting for public safety and epidemiological investigations.
- With advent of genomics, future add-on panels could be designed with additional strain specific markers/targets for Reflexive testing for strain typing/discrimination that could prove to be invaluable during outbreaks as a Culture independent diagnostic test.

Acknowledgements

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