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ORIGINAL ARTICLE

The Distribution of Enteric Infections Utilizing Stool Microbial Polymerase Chain Reaction Testing in Clinical Practice

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Abstract

Background Gastrointestinal infection is a major cause of morbidity. We sought to characterize the pathogenic etiologies of gastrointestinal infection to identify seasonal patterns and predictors of specific infections utilizing a multiplex PCR assay in clinical practice.

Methods We performed a cross-sectional study of 9403 patients who underwent 13,231 stool tests with a FilmArray gastrointestinal pathogen PCR panel during an episode of diarrhea from March 2015 to May 2017. Our primary outcome was the presence of a positive panel. Logistic regression was used to test for associations between season and infections.

Results A positive result was found in 3426 tests (25.9%) in 2988 patients (31.8%), yielding 4667 pathogens consisting of 1469 viruses (31.5%), 2925 bacteria (62.7%), and 273 parasites (5.8%). Age less than 50 years was associated with a higher prevalence of pathogens compared to age ≥ 50 ($p < 0.0001$). The overall prevalence of a positive result for bacteria peaked in the summer (635, 29.2%), and the prevalence of viruses peaked in the winter (446, 31.8%). Compared to the winter, testing in the summer yielded a higher prevalence of bacteria (OR 1.52, 95% CI 1.33, 1.73, $p < 0.0001$) and lower odds of viruses (OR 0.69, 95% CI 0.58, 0.81, $p < 0.0001$), primarily driven by *E. coli* species and norovirus.

Conclusions Season was a major determinant in detecting specific pathogens. Our substantially lower positivity rate than previous reports in the literature on multiplex PCR assays may more accurately reflect true clinical practice. Recognizing the temporal distribution of enteric pathogens may help facilitate empiric treatment decisions in certain clinical situations.

Keywords Enteric infection · Multiplex PCR · Seasonality · Gastroenteritis · Diarrhea

Introduction

Acute gastrointestinal infection is a major cause of morbidity and mortality worldwide [1, 2]. The Centers for Disease Control and Prevention estimate nearly 48 million cases annually in the USA, accounting for a large number of hospitalizations and outpatient visits, and significant direct and indirect costs [2, 3]. Acute diarrheal illness may occur due to infection with viral, bacterial, or parasitic pathogens, typically resulting in diarrhea associated with enteric symptoms such as abdominal pain and/or cramping, fever, malaise, bloody stools, nausea, and vomiting, that generally lasts for less than 14 days [1, 4]. Though most acute enteric infections are self-limited, rarely such infections may result in more severe illness requiring hospitalization. In addition, sequelae can include Guillain–Barré syndrome, reactive arthritis, post-infection irritable bowel syndrome,

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post-infection malabsorption syndrome, or hemolytic uremic syndrome [4].

Several studies have investigated seasonal patterns in enteric infections [5–11]. Infection with *E. coli* O157, campylobacteriosis, salmonellosis, and shigellosis tends to peak in the summer months, whereas infection with other enteric pathogens, such as giardia, seems to have little seasonal variation [5–8, 11]. Viral pathogens, specifically norovirus, seem to peak during winter, and protozoan infection, specifically cryptosporidia, tends to peak in the spring [9, 10]. Despite these data, most studies examining seasonality of infection considered a limited number of pathogens or patients, focused on a narrow geographic distribution and ethnically homogenous population, and utilized diagnostic assays with variable accuracy [5–12]. A greater understanding of the temporal and spatial patterns in enteric infections may provide further information regarding the pathogenic etiology and the relationship between physical environment and risk of infection. Moreover, the rapid identification of specific pathogens is critical for appropriate patient management and surveillance to identify, monitor, and prevent outbreaks.

Recently, molecular multiplex assays have been started to replace conventional microbiological tests as a rapid and accurate means of approaching acute gastroenteritis [4, 13]. These assays allow for the identification of specific organisms not previously and readily diagnosable by the clinician. In the present study, we sought to characterize the pathogenic etiologies of acute gastrointestinal infection in a large, ethnically diverse setting to identify seasonal patterns and predictors of specific enteric infections utilizing an FDA-approved, multiplex PCR assay in clinical practice.

Methods

Study Population and Variables

We performed a cross-sectional study using the electronic medical records of inpatients and outpatients at NewYork-Presbyterian/Columbia University Medical Center, a quaternary care institution in New York City that serves patients from the surrounding urban area, the tristate region (NY, NJ, and CT), as well as people seeking quaternary care from more distant regions. We identified all outpatients and inpatients who underwent stool testing with a FilmArray gastrointestinal pathogen polymerase chain reaction (PCR) panel (BioFire Diagnostics, Salt Lake City, UT, USA) during an episode of diarrhea during the 26-month period spanning March 2015 through May 2017.

We recorded the following values from the medical record: date of PCR test, PCR results, date of birth, zip code, place of PCR test (e.g., emergency department, outpatient visit, inpatient hospitalization, endoscopy), sex, race, and ethnicity. Repeat gastrointestinal pathogen PCR tests within 6 months and any repeat positive gastrointestinal pathogen PCR tests on the same patient were excluded. A random sample of 100 patients was assessed to confirm that identified records had correct diagnoses codes, PCR test dates, and results. Of those sampled patients, all patients were correctly classified.

Enteric Pathogen Testing

The gastrointestinal pathogen panel PCR (BioFire FilmArray, Salt Lake City, Utah) tests for 22 analytes in stool including 13 bacteria, 5 viruses, and 4 parasites including *Campylobacter* (*jejuni, coli,* and *upsaliensis*), *Clostridium difficile* (Toxin A/B), *Plesiomonas shigelloides*, Salmonella, *Yersinia enterocolitica*, *Vibrio (parahaemolyticus, vulnificus*, and *cholerae*), enteroaggregative *E. coli* (EAEC), enteropathogenic *E. coli* (EPEC), enterotoxigenic *E. coli* (ETEC), Shiga-like toxin-producing *E. coli* (STEC), *E. coli* O157, Shigella/enteroinvasive *E. coli* (EIEC), *Cryptosporidium* spp., *Cyclospora cayetanensis*, *Entamoeba histolytica, Giardia lamblia*, adenovirus (AdV) F40/41, astrovirus, norovirus GI/GII, rotavirus A, and sapovirus (I, II, IV, and V). In our institution, an alternative PCR test is utilized for *Clostridium difficile*, and as such, these results are not reported with the gastrointestinal pathogen panel PCR and were not examined in this study. The gastrointestinal pathogen panel PCR is capable of the simultaneous detection and identification of nucleic acids from multiple bacteria, viruses, and parasites directly from stool samples in Cary Blair transport media. The multiplex PCR process takes approximately 1 h. The clinical sensitivity and specificity are 94.5–100% for all targets [4, 12].

Outcomes and Statistical Analyses

Our primary outcome was the presence of any positive gastrointestinal pathogen PCR panel according to seasonality. In accordance with prior studies, we divided the calendar year into winter (December 21–March 20), spring (March 21–June 20), summer (June 21–September 20), and fall (September 21–December 20) [14, 15]. Secondary analyses included predictors of bacterial, viral, or parasitic infections, and seasonal patterns in specific enteric infections, and by each of the 12 months. We measured associations between variables and PCR stool test results via the Chi-square test for categorical variables and the *t* test for continuous variables. We used the Pearson correlation coefficient to assess for changes in testing over time. We then used multiple logistic regression to test for the association between season and the presence of enteric infections, after adjusting for age, gender, location of the test (emergency department,

outpatient visit, inpatient hospitalization, endoscopy), race, ethnicity, and residential zip code (New York City, surrounding tristate area, other). All tests were considered significant at a 2-sided p value less than 0.05.

Results

During the data collection period, 9403 patients underwent 13,231 gastrointestinal pathogen PCR stool tests. The mean patient age was 42.9 years (range 0–102), 23.7% were younger than 18 years, 17.1% were older than 70 years, 48.8% were female, and 18% were Hispanic. Stool testing was most common in the spring (28.7%) and least common in the fall (22.7%, Table 1). As was previously reported in pilot data on a subset of 337 outpatients from January to December 2015 [16], a positive test was more likely in younger patients (38.8% age < 18, $p < 0.0001$; Supplementary Fig. 1), males (27.8%, $p < 0.0001$), Hispanics (29.2%, $p < 0.0001$), and residents of New York City (28.0%, $p < 0.0001$; Table 1).

A total of 4667 enteric pathogens were detected in 3426 tests (25.9%) representing 2988 patients (31.8%). Of the positive tests, more than one enteric pathogen was detected in 922 (26.9%). Of the positive tests, 1469 viruses (31.5%), 2925 bacteria (62.7%), and 273 parasites (5.8%) were identified (Table 2). The most commonly identified pathogens were enteropathogenic *E. coli* (1048, 22.5%), norovirus (808, 17.3%), and enteroaggregative *E. coli* (639, 13.7%). The least commonly identified organisms were *Entamoeba histolytica* (2, < 0.01%), *Vibrio cholera* (5, 0.1%), other *Vibrio* species (18, 0.4%), and *Cyclospora cayetanensis* (18, 0.4%). All pathogens were identified alone and in combination with other pathogens except *E. histolytica*, which only occurred in the presence of other pathogens.

There were several significant predictors of a positive test (Table 3). In terms of bacteria, age less than 50 ($p < 0.0001$), Hispanic ethnicity ($p = 0.002$), and residing in New York City ($p = 0.008$) were associated with a higher odds of bacterial enteric infection. In terms of viruses, age less than 50 ($p < 0.0001$) and residing in New York City ($p = 0.007$) were associated with a higher odds of viral enteric infection while testing in the emergency room ($p = 0.042$), whereas female sex ($p = 0.004$) predicted a lower odds of viral enteric infection. In terms of parasites, age less than 50 ($p < 0.0001$) was associated with a higher odds of parasitic enteric infection, whereas female sex ($p < 0.0001$) predicted a lower odds of parasitic enteric infection.

The overall monthly prevalence of a positive test varied significantly, with the highest prevalence of positive tests in September (319, 31.5%) and the lowest prevalence of positive tests in October (208, 20.9%; Table 1, Fig. 1). When grouped by season, in the winter 28.5% of tests were positive, while in the fall 24.1% of the tests were positive. There was a significant change in percent of positive tests over the data collection period from more than 35% in March of 2015 to less than 22% in April 2017 ($r = -0.795$, $p < 0.0001$, Fig. 1).

The overall prevalence of a positive result for bacteria peaked in the summer (635, 21.1%), and the seasonal prevalence of viruses peaked in the winter (446, 13.2%; Fig. 2). Compared to the winter, testing in the summer had a higher odds of bacterial infection (OR 1.52, 95% CI 1.33, 1.723, $p < 0.0001$) and lower odds of viral infection (OR 0.69, 95% CI 0.58, 0.81, $p < 0.0001$), and testing in the spring had a lower odds of viral infection (OR 0.62, 95% CI 0.53,0.73, $p < 0.0001$). More specifically, compared to December, testing in August (OR 1.51, 95% CI 1.21, 1.89, $p < 0.0001$) and September (OR 1.67, 95% CI 1.34, 2.08, $p < 0.0001$) had a higher odds of bacterial enteric infection, whereas testing in January (OR 1.35, 95% CI 1.05, 1.72, $p < 0.017$) had a higher odds of viral enteric infection (Table 3). There were no statistically significantly monthly or seasonal changes in parasitic enteric infection detection ($p > 0.05$). When we repeated the analysis excluding inpatients, the seasonal patterns were essentially unchanged except that the finding of lower odds of viral infections in the spring compared to the winter was no longer statistically significant (Supplementary Table 1).

The seasonal trends in enteric pathogen detection were primarily driven by *E. coli* species and norovirus (Fig. 3). Norovirus was less likely to be present in the spring (OR 0.72, 95% CI 0.60, 0.87, $p < 0.0001$), summer (OR 0.52, 95% CI 0.42, 0.65, $p < 0.0001$), and fall (OR 0.68, 95% CI 0.56, 0.83, $p < 0.0001$) compared to the winter (Supplementary Table 2). Both enteropathogenic *E. coli* (OR 1.59, 95% CI 1.33, 1.89, $p < 0.0001$) and enterotoxigenic *E. coli* (OR 1.63, 95% CI 1.11, 2.39, $p = 0.013$) were more likely to be present in the summer compared to the winter (Supplementary Table 2). Although there was seasonal variation in overall *E. coli* prevalence, there were no statistically significant changes in among *E. coli* species throughout the months ($p = 0.156$, Supplementary Fig. 2).

Discussion

In this cross-sectional analysis, nearly 26% of gastrointestinal pathogen PCR panels were positive for at least one pathogen during an episode of diarrhea. In addition, season was a major determinant in detecting pathogens. To our knowledge, this is the largest study to date examining seasonal variation of enteric pathogens using a multiplex PCR assay.

Our positivity rate is substantially lower than previous reports in the literature on the BioFire FilmArray multiplex PCR assay, ranging from 30 to more than 70% [4,

Table 1 Characteristics of 9403 patients who underwent 13,231 gastrointestinal pathogen PCR stool tests stratified by positivity

	Total $n = 13,231$ (n, % of total)	GI PCR negative $n = 9805$ (74.1) (n, % of subgroup)	GI PCR positive $n = 3426$ (25.9) (n, % of subgroup)	p value
Mean age at test (years)	42.9	46	34	
Median age at test (range)	46.8 (0–102)	51 (0–102)	31 (0–100)	0.0001
Age group				
< 18	3137 (23.7)	1889 (60.2)	1248 (39.8)	
18–29	1516 (11.5)	1093 (72.1)	423 (27.9)	
30–49	2395 (18.1)	1785 (74.5)	610 (25.5)	
50–69	3923 (29.7)	3171 (80.8)	752 (19.2)	
> 70	2260 (17.1)	1867 (82.6)	393 (17.4)	0.0001
Month of test				
January	1168 (8.8)	814 (69.7)	254 (30.3)	
February	1062 (8)	804 (75.7)	258 (24.3)	
March	1288 (9.7)	959 (74.5)	329 (25.5)	
April	1524 (11.5)	1137 (74.6)	387 (25.4)	
May	1125 (8.5)	862 (76.6)	263 (23.4)	
June	911 (6.9)	701 (76.9)	210 (23.1)	
July	999 (7.6)	736 (73.7)	263 (26.3)	
August	1101 (8.3)	784 (71.2)	317 (28.8)	
September	1012 (7.7)	693 (68.5)	319 (31.5)	
October	997 (7.5)	789 (79.1)	208 (20.9)	
November	981 (7.4)	748 (76.2)	233 (23.8)	
December	1063 (8)	778 (73.2)	285 (26.8)	0.0001
Season of test				
Summer	3389 (25.6)	2469 (72.9)	920 (27.1)	
Spring	3801 (28.7)	2883 (75.8)	918 (24.2)	
Fall	3008 (22.7)	2284 (75.9)	724 (24.1)	
Winter	3033 (22.9)	2169 (71.5)	864 (28.5)	0.0001
Place of test				
Inpatient	7787 (58.9)	5747 (73.8)	2040 (26.2)	
Emergency room	1416 (10.7)	1081 (76.3)	335 (23.7)	
Endoscopy unit	157 (1.2)	123 (78.3)	34 (21.7)	
Outpatient	3871 (29.3)	2854 (73.7)	1017 (26.3)	0.121
Sex				
Male	6777 (51.2)	4662 (72.2)	1792 (27.8)	
Female	6454 (48.8)	5143 (75.9)	1634 (24.1)	0.0001
Race				
Asian	581 (4.4)	433 (74.5)	148 (25.5)	
Non-Hispanic Black	1411 (10.7)	1126 (79.9)	285 (20.2)	
American Indian	20 (0.2)	10 (50.0)	10 (50.0)	
Other/unknown	6280 (47.5)	4417 (70.3)	1863 (29.7)	
Non-Hispanic White	4939 (37.3)	3819 (77.3)	1120 (22.7)	0.0001
Hispanic ethnicity				
Hispanic	2388 (18.1)	1691 (70.8)	697 (29.2)	
Non-Hispanic	4710 (35.6)	3738 (79.4)	972 (20.6)	
Unknown	6133 (46.4)	4376 (71.4)	1757 (28.6)	0.0001
Residential zip code				
New York City	8118 (61.4)	5843 (72.0)	2275 (28.0)	
Surrounding tristate area	4843 (36.6)	3741 (77.2)	1102 (22.8)	
Other	270 (2.0)	221 (81.9)	49 (18.1)	0.0001

Table 2 Types of enteric infections among those with a positive gastrointestinal pathogen panel PCR result

	Number	% of total positive
Viruses	1469	31.5
Adenovirus F 40/41	108	2.3
Astrovirus	116	2.5
Norovirus GI/GII	808	17.3
Rotavirus A	211	4.5
Sapovirus (I, II, IV, V)	226	4.8
Bacteria	2925	62.7
Campylobacter species	369	7.9
Plesiomonas shigelloides	33	0.7
Salmonella species	171	3.7
Yersinia enterocolitica	96	2.1
Vibrio species	13	0.3
Vibrio cholerae	5	0.1
Enteroaggregative *E. coli* (EAEC)	639	13.7
Enteropathogenic *E. coli* (EPEC)	1048	22.5
Enterotoxigenic *E. coli* (LT/ST)	194	4.2
Shiga-like toxin-producing *E. coli* STX/ST2	154	3.3
E. coli 0157	25	0.5
Shigella/Enteroinvasive *E. coli* (EIEC)	178	3.8
Parasites	273	5.8
Cryptosporidium	108	2.3
Cyclospora cayetanesis	18	0.4
Entamoeba histolytica	2	0
Giardia lamblia	145	3.1
Total	4667	100

12, 16–18]. In fact, previously reported pilot data from our institution demonstrated a 35% positivity rate in outpatients and a lower positivity rate for patients with underlying diarrheal diseases, such as inflammatory bowel disease and celiac disease [16, 19]. Given our large sample size, these results may more accurately reflect true clinical practice. Despite the lower positivity rate and the censor of *C. difficile* results from the BioFire FilmArray assay in our center, we found similar proportions of bacterial, viral, and parasitic pathogens to previous data in the literature. Moreover, we similarly found a greater proportion of infections in younger patients, particularly viruses. It is also possible that the lower positivity rate and decrease in this rate over the data collection period are due to our broad inclusion criteria, or more likely, increasing inappropriate utilization of the test, as our high number of inpatients suggests it may not have been clearly ordered for an episode of acute gastroenteritis.

In terms of seasonality, similar to previous reports using culture techniques and limited PCR analyses, we found bacterial pathogens, such as *Salmonella* and *E. coli* species, to be more common in the summer, and viral pathogens, such as norovirus and rotavirus, to be more common in the winter [5, 7, 9]. We are the first to report on seasonal variability in enteroaggregative *E. coli*, enteropathogenic *E. coli*, and enterotoxigenic *E. coli* infection, suggesting that the prevalence of these pathogens is increased during the summer. Given that all these infections are associated with travel, which occurs frequently during the summer, it is possible that travel-associated cases may explain these observations. We did not have information on recent travel in our study population to test this hypothesis.

Differing from previous reports using conventional culture data, we found no seasonal pattern to campylobacteriosis, cryptosporidiosis, and shigellosis, and we found giardiasis to be more common in the winter [5–11]. Given the large, diverse sample size, it is possible that employing this more sensitive PCR diagnostic assay may invalidate previously reported seasonal patterns and reveal new seasonality in specific enteric infections.

Seasonal patterns in enteric infection are thought to occur secondary to multiple pathogenic, human behavioral, and environmental mechanisms [5, 7–11, 14, 15, 20–24]. These include changes in the reproductive number and subsequent transmissibility of pathogens via effective contact rates, pathogen durability and survival, and population-level human susceptibility to disease [20, 23, 25]. Behaviors such as barbequing during the summer season, overseas travel, and the start of the school year are thought to increase contact rates, whereas relative immunosuppression and clustering within enclosed spaces during the winter season are thought to increase pathogen survival and human susceptibility to disease [14, 20]. These mechanisms may explain our findings of increased *E. coli* species prevalence during the summer months and increased norovirus prevalence during the winter months. Understanding seasonal trends may also provide a greater sense of pretest probability, yielding important information for empiric treatment decisions in certain clinical situations.

There are several limitations to the current study inherent to a retrospective design. Our analyses do not prove a cause-and-effect relationship between diarrhea and enteric infections. Individual patient information concerning precise presenting symptoms, medication exposures, recent travel, sexual behavior, other comorbid conditions, and management after stool testing was not available for analysis. Although the patient population was ethnically and geographically diverse, the majority of patients resided in the Northeast United States and generalizability may be limited. In addition, PCR testing fails to discriminate between active infection and asymptomatic colonization, and there is considerable uncertainty regarding clinical interpretation and cost-effectiveness of such multiplex assays [17]. The FilmArray gastrointestinal pathogen PCR panel does not assess for the presence of Listeria

Table 3 Monthly and seasonal distribution of bacterial, viral, and parasitic infections

Variable	Bacteria ($n=2174$)				Virus ($n=1404$)				Parasite ($n=267$)			
	Number (% positive)	p value	Multivariable odds ratio (95% CI)	p value	Number (% positive)	p value	Multivariable odds ratio (95% CI)	p value	Number (% positive)	p value	Multivariable odds ratio (95% CI)	p value
Age group		0.0001				0.0001				0.0001		
< 18	693 (31.9)		1.95 (1.67–2.27)	0.0001	624 (44.4)		3.89 (3.19–4.75)	0.0001	94 (35.2)		3.08 (1.91–4.98)	0.0001
18–29	265 (12.2)		1.48 (1.23–1.78)	0.0001	178 (12.7)		2.19 (1.73–2.79)	0.0001	47 (17.6)		3.42 (2.03–5.76)	0.0001
30–49	411 (18.9)		1.48 (1.25–1.74)	0.0001	207 (14.7)		1.54 (1.22–1.94)	0.0001	59 (22.1)		2.63 (1.59–4.35)	0.0001
50–69	527 (24.2)		1.12 (0.96–1.31)	0.154	266 (18.9)		1.19 (0.96–1.48)	0.123	46 (17.2)		1.23 (0.73–2.07)	0.435
> 70	278 (12.8)		Reference		129 (9.2)		Reference		21 (7.9)		Reference	
Month of test		0.0001				0.0001				0.738		
January	184 (8.5)		0.97 (0.77–1.22)	0.784	192 (13.7)		1.35 (1.05–1.72)	0.017	26 (9.7)		1.27 (0.69–2.34)	0.437
February	146 (6.7)		0.86 (0.68–1.10)	0.229	121 (8.6)		0.92 (0.70–1.20)	0.536	24 (9.0)		1.36 (0.73–2.52)	0.335
March	201 (9.2)		0.98 (0.78–1.22)	0.825	136 (9.7)		0.83 (0.64–1.07)	0.148	26 (9.7)		1.19 (0.65–2.20)	0.569
April	218 (10.0)		0.90 (0.72–1.12)	0.326	202 (14.4)		1.09 (0.86–1.39)	0.476	29 (10.9)		1.12 (0.62–2.03)	0.708
May	152 (7.0)		0.83 (0.65–1.05)	0.127	123 (8.8)		0.86 (0.66–1.12)	0.270	18 (6.7)		0.94 (0.49–1.83)	0.864
June	140 (6.4)		1.01 (0.79–1.29)	0.948	80 (5.7)		0.71 (0.53–0.96)	0.026	12 (4.5)		0.83 (0.40–1.73)	0.615
July	185 (8.5)		1.24 (0.98–1.57)	0.068	80 (5.7)		0.62 (0.46–0.83)	0.002	23 (8.6)		1.44 (0.77–2.70)	0.250
August	242 (11.1)		1.51 (1.21–1.89)	0.0001	89 (6.3)		0.61 (0.45–0.81)	0.001	25 (9.4)		1.34 (0.73–2.48)	0.350
September	242 (11.1)		1.67 (1.34–2.08)	0.0001	84 (6.0)		0.62 (0.46–0.82)	0.001	27 (10.1)		1.55 (0.85–2.85)	0.154
October	144 (6.6)		0.91 (0.71–1.16)	0.431	73 (5.2)		0.56 (0.41–0.76)	0.0001	18 (6.7)		1.08 (0.56–2.10)	0.813
November	152 (7.0)		0.99 (0.78–1.26)	0.927	90 (6.4)		0.74 (0.55–0.98)	0.036	21 (7.9)		1.31 (0.69–2.48)	0.411
December	168 (7.7)		Reference		134 (9.5)		Reference		18 (6.7)		Reference	
Season of test		0.0001				0.0001				0.571		
Spring	547 (25.2)		0.96 (0.84–1.09)	0.521	426 (30.3)		0.62 (0.53–0.73)	0.0001	67 (25.1)		1.07 (0.76–1.50)	0.694
Summer	635 (29.2)		1.52 (1.33–1.73)	0.0001	257 (18.3)		0.69 (0.58–0.81)	0.0001	67 (25.1)		0.99 (0.70–1.40)	0.944
Fall	476 (21.9)		1.07 (0.94–1.23)	0.319	275 (19.6)		0.86 (0.75–0.99)	0.044	61 (22.8)		0.85 (0.60–1.19)	0.337
Winter	516 (23.7)		Reference		446 (31.8)		Reference		72 (27)		Reference	
Place of test		0.516				0.018				0.633		
Inpatient	1271 (58.5)		0.96 (0.87–1.07)	0.471	864 (61.5)		1.09 (0.96–1.24)	0.177	158 (59.2)		0.97 (0.74–1.27)	0.817
Emergency room	218 (10.0)		0.85 (0.72–1.00)	0.055	127 (9)		0.80 (0.65–0.99)	0.042	27 (10.1)		0.92 (0.59–1.42)	0.692
Endoscopy unit	25 (1.1)		0.97 (0.63–1.50)	0.889	9 (0.6)		0.56 (0.28–1.10)	0.090	1 (0.4)		0.30 (0.04–2.17)	0.234
Outpatient	660 (30.4)		Reference		404 (28.8)		Reference		81 (30.3)		Reference	
Female	1063 (48.9)	0.18	0.91 (0.83–1.00)	0.052	647 (46.1)	0.0001	0.84 (0.75–0.95)	0.004	100 (37.5)	0.0001	0.58 (0.45–0.74)	0.0001
Race		0.0001				0.0001				0.0002		
Asian/Pac Islander	95 (4.4)		1.09 (0.86–1.38)	0.471	64 (4.6)		1.09 (0.82–1.44)	0.567	6 (2.2)		0.58 (0.25–1.33)	0.196

Table 3 (continued)

Variable	Bacteria ($n=2174$)				Virus ($n=1404$)				Parasite ($n=267$)			
	Number (% positive)	p value	Multivariable odds ratio (95% CI)	p value	Number (% positive)	p value	Multivariable odds ratio (95% CI)	p value	Number (% positive)	p value	Multivariable odds ratio (95% CI)	p value
Non-Hispanic Black	177 (8.1)		0.86 (0.72–1.03)	0.105	117 (8.3)		0.95 (0.77–1.19)	0.665	27 (10.1)		1.19 (0.76–1.86)	0.452
American Indian	3 (0.1)		0.87 (0.25–3.00)	0.825	7 (0.5)		5.15 (1.97–13.46)	0.0001	0		–	
Other/unknown	1178 (54.2)		1.19 (1.05–1.35)	0.009	774 (55.1)		1.24 (1.06–1.46)	0.008	158 (59.2)		1.22 (0.86–1.74)	0.257
Non-Hispanic White	721 (33.2)		Reference		442 (31.5)		Reference		76 (28.5)		Reference	
Hispanic ethnicity		0.0001				0.0001				0.0001		
Non-Hispanic	594 (27.3)		0.81 (0.70–0.93)	0.0001	405 (28.8)		0.88 (0.74–1.04)	0.124	69 (25.8)		0.75 (0.52–1.09)	0.129
Hispanic	491 (22.6)		1.30 (1.14–1.48)	0.002	260 (18.5)		0.95 (0.80–1.12)	0.513	41 (15.4)		0.75 (0.52–1.10)	0.138
Unknown	1089 (50.1)		Reference		739 (52.6)		Reference		157 (58.8)		Reference	
Residential zip code		0.0001				0.0001				0.538		
New York City	1438 (66.1)		1.67 (1.14–2.44)	0.008	949 (67.4)		1.99 (1.21–3.27)	0.007	163 (61)		1.81 (0.56–5.71)	0.311
Tristate area	705 (32.4)		1.31 (0.89–1.92)	0.168	441 (31.4)		1.48 (0.90–2.44)	0.126	101 (37.8)		1.89 (0.59–5.99)	0.282
Other	31 (1.4)		Reference		17 (1.2)		Reference		3 (1.1)		Reference	

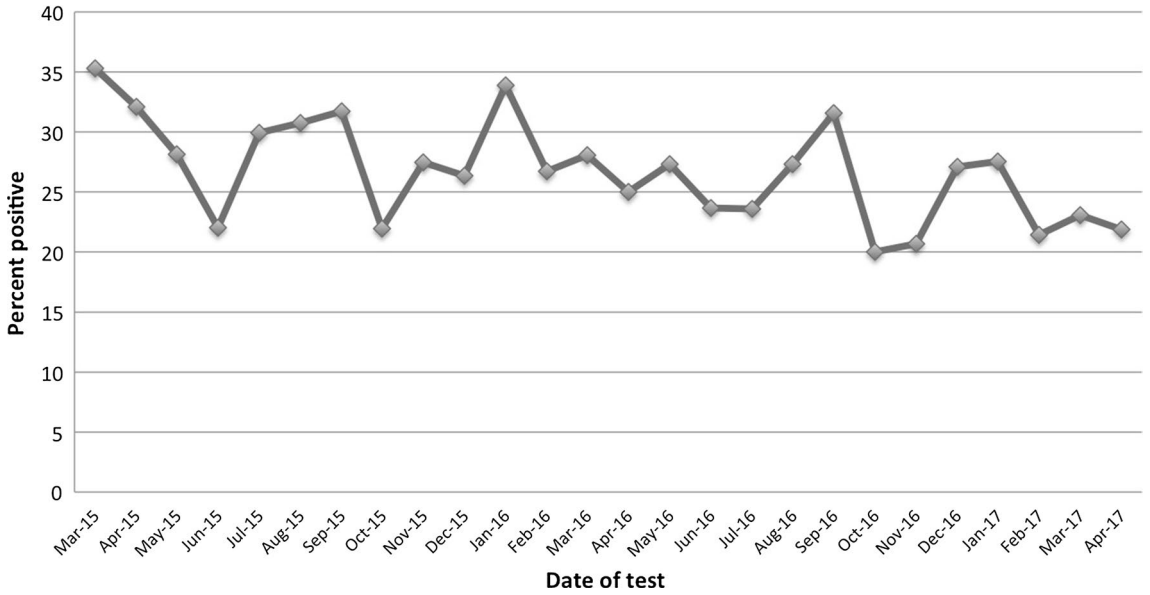

Fig. 1 Prevalence of enteric infections as detected by gastrointestinal pathogen panel PCR over the data collection period ($r = -0.795$, $p < 0.0001$)

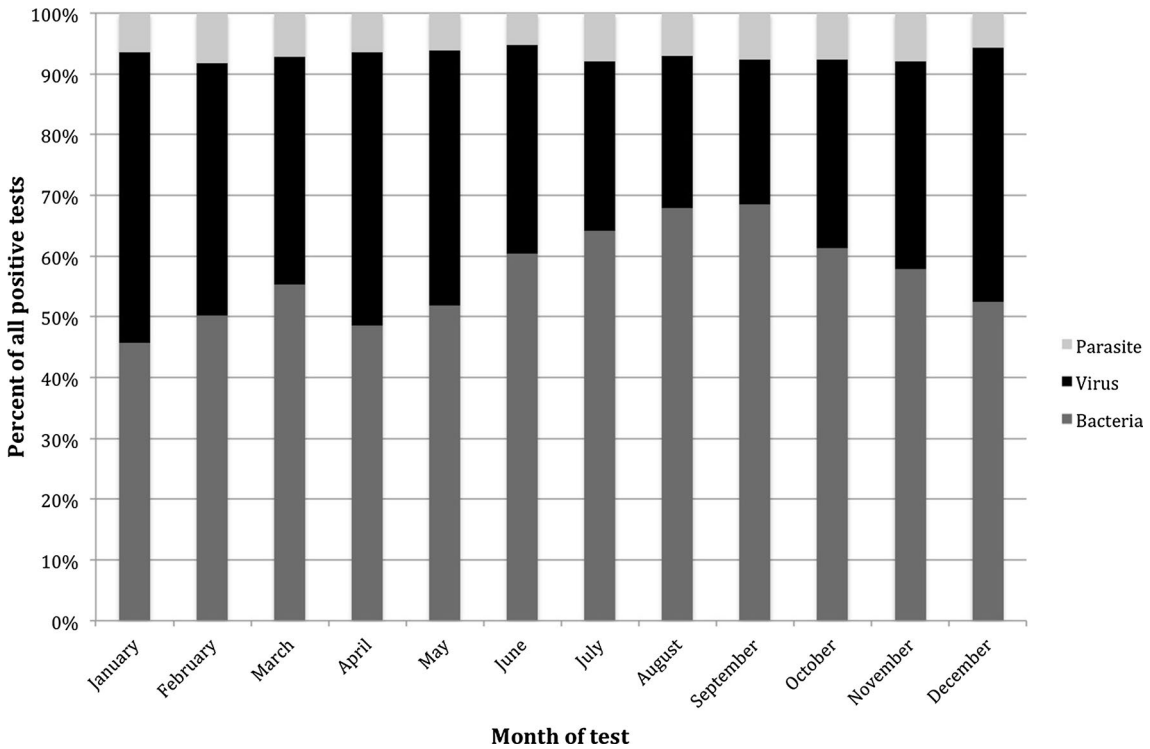

Fig. 2 Monthly distribution of bacteria, viruses, and parasites detected by gastrointestinal pathogen panel PCR, among those with a positive test result

monocytogenes or cytomegalovirus (CMV), pathogens of increasing importance, and given testing constraints in our institution, we were unable to analyze *Clostridium difficile* infection.

Despite these limitations, in this large analysis of patients with diarrhea, season and age were major determinants of enteric infection with bacterial, viral, and parasitic pathogens detected via stool PCR testing. In the

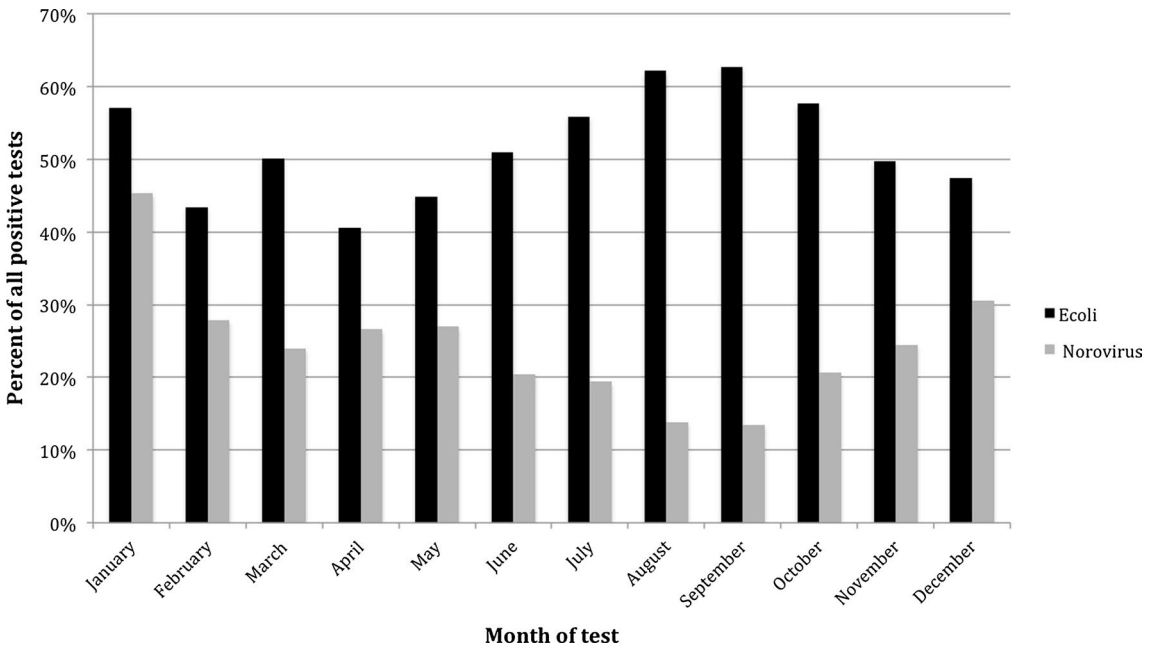

Fig. 3 Monthly distribution of the *E. coli* and norovirus, among those with a positive test result

setting of global ecological change, recognizing the temporal distribution of specific enteric infections is critically important for predicting the interaction between seasonal exposure and the physical environment as it relates to disease. Failure to identify these patterns may have deleterious effects on agricultural activities, human behavior, and public health. As the widespread availability of enteric PCR testing increases, further studies will examine specific clinical characteristics, presenting symptoms, and management decisions associated with enteric infection testing to inform consequent clinical management of patients. In addition, future study is needed to evaluate how well these rapid multiplex diagnostics impact patient care and outcomes.

Author's contribution JEA (Guarantor) was involved in study concept and design; acquisition of data; analysis and interpretation of data; drafting of the manuscript; and statistical analysis. AJ and YN contributed to study concept and design and acquisition of data. SW, GL, and MSR were involved in critical revision of the manuscript for important intellectual content.. PHRG and BL contributed to study concept and design; drafting of the manuscript; and critical revision of the manuscript for important intellectual content.. All authors approved the final version of the article, including the authorship list.

Compliance with ethical standards

Conflict of interest The authors disclose no potential conflicts relevant to this manuscript

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