Prevalence of enteric pathogen-associated community gastroenteritis among kindergarten children in Gaza

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Abstract

Gastroenteritis is considered as one of the leading causes of illness and death in children under 5 years age, especially in developing countries. It is one of the major public health problems among childhood in Gaza strip, Palestine. This study was conducted to determine the prevalence of enteric pathogen-associated community gastroenteritis among kindergarten children in Gaza. A total of 150 stool samples were collected and investigated for parasitic, viral and bacterial pathogens at Al Azhar microbiology laboratories by using standard microbiological and serological procedures. Out of the 150 study samples, the overall percentage of positive stool samples with a known enteric pathogen was 60.6%. The prevalence of different enteric pathogens causing community gastroenteritis among symptomatic cases (88.5%) was significantly higher than the prevalence in asymptomatic carriage (11.1%). The most prevalent isolated enteric pathogens were *Entamoeba histolytica* (28.0%) and *Giardia lamblia* (26.7%). Rotavirus was found in 3.1% of symptomatic cases but not detected in asymptomatic carriage. However, adenovirus type 40 and 41 were not detected in any of the study samples. The bacterial enteric pathogens *Shigella* and Enterohemorrhagic *Escherichia coli* O157:H7 (EHEC) have comparable occurrence as rotavirus (3.1%), meanwhile, *Salmonella* was not isolated. Mixed infection with more than 1 pathogen was found (11.4%) only among symptomatic cases. Children aged 3-year-old showed the highest prevalence of community gastroenteritis. This study demonstrates a high prevalence of parasitic enteropathogens and a relatively low prevalence of bacterial and viral enteropathogens among kindergarten children living in Gaza city, moreover, children aged 3 years old showed the highest prevalence of isolated enteropathogens.

Keywords: community gastroenteritis, enteropathogens, Gaza, kindergarten

Introduction

Worldwide, diarrheal diseases are continued to be a common health problem that increase the financial burden on the health systems particularly in developing countries. Acute gastroenteritis is considered as one of the leading causes of illness and death in children under the age of 5 years⁴¹⁻⁴³, and is characterized by acute onset of diarrhea, which may or may not be accompanied by nausea, emesis, fever, abdominal pain and dehydration. Acute gastroenteritis is prevalent worldwide and associated with high rates of morbidity and mortality in developing countries⁴⁴⁻⁴⁷. In Gaza, acute gastroenteritis is considered as a common infection...
among children and is known to cause high morbidity and mortality rates if they do not receive proper treatment on time ²,³,⁴.

Gastroenteritis transmission is spread by person to person or ingestion of contaminated food and drink. Ingestion of food containing toxins produced by bacterial contaminants causes rapid onset of vomiting and/or diarrhea. Water is also contaminated by bacteria, viruses, or protozoa that cause gastroenteritis ⁵,⁶. A person continues to excrete enteropathogens after an episode of acute gastroenteritis or even be an asymptomatic carrier without previous disease. Asymptomatic carriers of parasitic, viral or bacterial pathogens have epidemiological importance due to their influence as a source of infection ⁷,⁸.

The etiologic agents include a wide variety of bacteria, viruses and parasites. The commonest are rotavirus, Salmonella, Shigella, Campylobacter jejuni, enteropathogenic Escherichia coli (EPEC), enterotoxicogenic E. coli (ETEC) and Yersinia enterocolitica. Parasitic agents, especially Cryptosporidium, Entamoeba histolytica, and Giardia lamblia are also etiological agents of diarrhea in children ⁹. Severe clinical diseases and mortality are associated with these intestinal pathogens which are known to cause malnutrition and impairment of physical development in children and affect their growth and learning ¹⁰.

Gaza Strip (geographic coordinates 31°25' N, 34°20' E) is a narrow region (365 km²) of land along the Mediterranean coast, just 41 km long and 6–12 km wide. It is one of the most heavily populated areas (4,383 persons/km²) of the world with a total population about 1.6 million people ¹¹. In the present work, we attempted to investigate the prevalence of different enteric pathogens causing community gastroenteritis among kindergarten children in Gaza.

**Subjects and methods**

**Study population and sample collection**

This study was performed from the beginning of January to the end of June 2011. One hundred fifteen written requests were sent to the parents or guardians of the symptomatic cases to have their approval to involve their child in the study. Thankfully, 96 (response rate 83.5%) were agreed to participate, and they represent 15 kindergartens of the different localities and population clusters. Also a response rate of 72.0% (54/75) was achieved for the asymptomatic carriers (apparently healthy controls) of same kindergartens. The study protocol and data handling were approved by the ethical committee at the Biology Department, Al Azhar University-Gaza. The study was performed in accordance with the ethical standards established in the 1964 and 1975 Declaration of Helsinki, and the modifications thereafter. A simple questionnaire that included personal, demographic and past history of diseases and treatment was constructed and conducted in Arabic language. The questionnaire was distributed to the parents who agree to include their children in this study prior to sample collection for filling and return with collected sample. Fresh stool samples were collected from cases and controls in labeled plastic vials without preservatives. Diarrhea was defined as the excretion of at least 2 loose stools in a 24-hour period ¹². Any child suffering from any chronic disease or on antibiotics treatment was excluded from participation. Stool samples were transferred to the microbiology laboratory in ice box and processed within 4 hours of collection.

**Microbiological methods**

Standard microbiological procedures and guidelines as described by the American Clinical Laboratory Institute (CLSI) were used. For the identification of parasites, fresh stool specimens prepared with saline and/or iodine were directly examined. Briefly, wet mount preparation of stool in 0.9% saline and/or iodine were directly examined. Briefly, wet mount preparation of stool in 0.9% saline and/or iodine was examined microscopically for the presence of protozoa cysts and trophozoites or helminthes ova or larva and adult stage. Moreover, flotation and sedimentation techniques were performed on all samples that gave negative result using direct microscopy examination.

**Detection of bacterial agents**

Fresh stool samples were directly plated onto salmonella-shigella (SS), Heckett enteric (HE), and Sorbitol MacConkey agar (SMAC) agars (HiMedia, India) and incubated for 18 to 24 hours at 37 °C. Moreover, approximately 1 g of each sample was inoculated into 10 mL of selenite cysteine broth (SCB) (HiMedia, India), and incubated for 18 to 24 hours at 37 °C. Approximately 0.5 mL of inoculated SCB was subcultured onto SS and HE agars and incubated as mentioned above. Suspected colonies on the primary and subculture plates were further identified by standard laboratory procedures as colony morphology, Gram staining, and biochemical tests including oxidase, urease and API20E system. Polyvalent antisera against Shigella and E. coli O157:H7 were used for confirmation.

**Detection of viral agents**

Enzyme immunoassay was used for the detection of group A rotavirus and adenovirus serotype 40 and 41.
According to the instructions of the manufacturer. A total of 100 mL of stool specimen was diluted in appropriate amounts of sample buffer included in the commercial GastroVir-Stripcolor kit (Cori BioConcept, Belgium). The GastroVir-Stripcolor kit was performed to detect group A rotaviruses and the most human adenovirus serotypes.

Statistical analyses
Data were tabulated, encoded and statistically analyzed using the Statistical Package for the Social Sciences (SPSS) version 15 software (IBM Corporation, Somers, NY). Discrete variables were expressed as percentages. Data were compared using Pearson Chi-square and the rank-sum test (Wilcoxon-Mann-Whitney test) as appropriate. The level of statistical significance was set at \( P < 0.05 \).

Results
The present study was a cross-sectional descriptive study that included 150 kindergarten children from Gaza city. The stool samples (Table 1) were collected from children from both genders (56% male and 44% female). The ages of children enrolled in this study were 3 (8%), 4 (34%) and 5 (58%) years old. According to the study protocol, 96 diarrheal stool samples were collected from 51 boys (53.1%) and 45 girls (46.9%) and they considered as symptomatic cases. On the other hand the asymptomatic carriage (controls group) included 33 boys (61.1%) and 21 (38.9%) girls.

According to the rank-sum test (Wilcoxon–Mann–Whitney test) significant difference was reported between cases and controls in terms of the age of the children. Cases are significantly younger than the control; mean ranks were 68.50 and 87.94 respectively, \( P = 0.003 \).

The highest percentage of diarrheal stool samples (10/12, 83.3%) was collected from the 3-year-old group, followed by the 4- and 5-year-old group (39/51, 67% and 47/87, 54%, respectively). There was a significant difference between the percentage of diarrhea in the 3-year-old group and other age groups (\( P = 0.01 \)).

Overall (Table 2), there are 91 samples (60.7%) positive for parasitic, bacterial, and/or viral enteric pathogens. The parasitic etiologic agents had the highest prevalence rate (54.7%) in comparison to the bacterial (4%) and viral (2%) etiologic agents. More than half of samples collected were positive for one or more parasitic agent. The highest isolated enteric pathogen was \( E.\ histolytica \) (28%), followed by \( G.\ lamblia \) (26.7%). However, adenovirus and \( Salmonella \) were not isolated from all tested samples. The overall prevalence of positive stools from symptomatic cases was significantly higher (88.5%) than that isolated from asymptomatic controls (11.1%). \( E.\ histolytica \) was the most prevalent in diarrhea cases (43.8%), followed by \( G.\ lamblia \) (35.4%). Rotavirus, \( Shigella\) spp. and \( E.\ coli\) O157:H7 has 3.1% for each (Table 2, and Fig. 1). All types of enteric pathogens including viral, bacterial and parasitic agents were isolated from

| Table 1 | Sex and age characteristics of the study samples |
|---------|-----------------------------------------------|
|         | Cases | Control | Total | \( \chi^2 \) | \( P \)-value |
| Sex     |       |         |       |              |               |
| Boys    | 51 (53.1) | 33 (61.1) | 84 (56) | 0.99         |               |
| Girls   | 45 (46.9) | 21 (38.9) | 66 (44) |               | 0.39          |
| Age     |       |         |       | \( \chi^2 \) |               |
| 3 years | 10 (10.4) | 2 (3.7)   | 12 (8.0) | 0.15*        |               |
| 4 years | 39 (40.6) | 12 (22.2) | 51 (34.0) |               | 0.01          |
| 5 years | 47 (49.0) | 40 (71.4) | 87 (58.0) |               |               |

*Corrected \( \chi^2 \).

| Table 2 | Enteric pathogens isolated from symptomatic cases and asymptomatic carriage (controls) |
|---------|-----------------------------------------------|
| Microorganism | Cases (96) | Controls (54) | Overall (150) |
|            | No. | %     | No. | No. | No. | %     |
| Rotavirus  | 3   | 3.10% | 0   | 0%  | 3   | 2.00% |
| Adenovirus | 0   | 0%    | 0   | 0%  | 0   | 0.00% |
| Salmonella spp. | 0 | 0%    | 0   | 0%  | 0   | 0.00% |
| Shigella spp. | 3   | 3.10% | 0   | 0%  | 3   | 2.00% |
| E. coli O157:H7 | 3   | 3.10% | 0   | 0%  | 3   | 2.00% |
| E. histolytica | 42  | 43.80% | 0   | 0%  | 42  | 28.00% |
| G. lamblia | 34  | 35.40% | 6   | 11.10% | 40  | 26.70% |
| Total     | 85  | 88.50% | 6   | 11.10% | 91  | 60.70% |
symptomatic cases samples, while only parasitic agent G. lamblia was isolated from asymptomatic controls (Table 2).

Table 3 shows the number and percentage of positive samples among different age groups. In 3 years age group children, E. histolytica was the most prevalent (75%) isolated pathogen. Rotavirus and Shigella were found in only 8.3%, while E. coli O157:H7 and G. lamblia were not detected in this age group. Children from the 4-year-old group also have E. histolytica as the commonest (27%) isolated enteric pathogen followed by G. lamblia (27%). However, Rotavirus was isolated from 3.9% of the children and Shigella spp. was recovered from 1.9%, which represents the lowest isolated enteric pathogen in the 4-year-old group. Whereas, the enteric pathogen E. coli O157:H7 was not detected. Interestingly, in the 5-year-old group, the prevalence profile was different where G. lamblia infection was the most frequent (29.9%) isolated enteric pathogen followed by E. histolytica (18.4%), E. coli O157:H7 (3.4%) and Shigella (1.1%). Nevertheless, rotavirus was not detected.

Parasitic pathogens had the highest prevalence in comparison to the other enteric pathogens, where it accounted for more than half of the screened samples (89/150; 59.3%). Overall [34 (85.0%) from symptomatic cases and 6 (15.0%) from asymptomatic controls]. E. histolytica was detected in 42 (28%) samples out of the total grand, where all of them (100%) were from cases (Table 4). Statistically significant prevalences were reported for G. lamblia and E. histolytica among symptomatic cases as compared to asymptomatic controls (P < 0.0002 and P < 0.0001, respectively).

Viral pathogens had the lowest prevalence (2%) among enteropathogens that were isolated in this study. Most of the cases (93/96; 96.9%) and all control samples (54/54; 100%) were negative. Rotavirus was isolated from 3 (2%) samples; all of them were reported in symptomatic cases. The difference in the presence and absence of rotavirus in cases and controls was not significant (P = 0.189). However, the second viral pathogen adenovirus was not isolated from any sample.

Bacterial pathogens had also lower prevalence in comparison to the parasitic pathogens (6/150, 4%). Regarding the prevalence of bacterial pathogens in cases and control samples, Shigella spp. and E. coli O157:H7 were isolated from 3 (2%) samples; all of them were reported in symptomatic cases. The difference in the presence and absence of Shigella spp. and E. coli O157:H7 in cases and controls was not significant (P = 0.189). Finally, Salmonella was not isolated from any sample (Table 4).

Table 5 shows that E. histolytica was detected in 90%, 43.6% and 34% of cases in the 3-, 4- and 5-year-old groups, respectively. The prevalence of E. histolytica in the 3-year-old group was significantly higher than that of the other age groups. G. lamblia was detected in 0.0%, 33.3% and 44.7% of cases in the 3-, 4- and 5-year-old groups, respectively. However, G. lamblia was found in 8.3% and 12.5% of control samples (asymptomatic) collected from the 4- and 5-year-old groups, respectively. A statistical difference was found between the prevalence of parasitic infection among cases and control samples in all age groups with P value of 0.007, < 0.0001 and < 0.0001 in the 3-, 4- and 5-year-old groups, respectively. Rotavirus was isolated from 10%, 5.1% and

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**Table 3** Distribution of enteric pathogens isolated from stools according to age

| Age (years) | Number of tested samples | Rotavirus | Shigella spp. | E. coli O157:H7 | E. histolytica | G. lamblia |
|-------------|--------------------------|-----------|---------------|-----------------|---------------|-----------|
| 3           | 12 (8%)                  | 1 (8.3%)  | 1 (8.3%)      | 0 (0%)          | 9 (75%)       | 0 (0%)    |
| 4           | 51 (34%)                 | 2 (3.9%)  | 1 (1.9%)      | 0 (0%)          | 17 (33.3%)    | 14 (27%)  |
| 5           | 87 (58%)                 | 0 (0%)    | 1 (1.1%)      | 3 (3.4%)        | 16 (18.4%)    | 26 (29.9%)|
| Total       | 150 (100%)               | 3 (2%)    | 3 (2%)        | 3 (2%)          | 42 (28%)      | 40 (26%)  |

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![Fig. 1 Prevalence of different parasitic, viral and bacterial enteropathogens among symptomatic cases.](image-url)
The prevalence of rotavirus in the 3-year-old group was higher than that of the other age groups. Meanwhile, rotavirus was not detected in controls. There was no significant difference in the prevalence of rotavirus infection between cases and controls in the 3- and 4-year-old groups ($P > 0.05$). *Shigella* spp. was isolated from 10%, 2.6% and 2.1% of cases in the 3-, 4- and 5-year-old groups, respectively.

The prevalence of *Shigella* spp. in the 3-year-old group was higher than that of the other age groups. There was no statistical difference in the prevalence of *Shigella* spp. infection between cases and controls in all age groups ($P > 0.05$).

### Table 4 Prevalence of parasitic, viral and bacterial pathogens among symptomatic cases and asymptomatic carriage (controls)

| Pathogen                        | Cases (diarrhea) | Controls | Total | $P$-value |
|---------------------------------|------------------|----------|-------|-----------|
| **Prevalence of parasitic agents** |                  |          |       |           |
| *E. histolytica*                |                  |          |       |           |
| Negative                        | 54 (56.2%)       | 54 (100%)| 108 (72%)| $< 0.0001^*$ |
| Positive                        | 42 (43.8%)       | 0 (0%)  | 42 (28%)|           |
| *G. lamblia*                    |                  |          |       |           |
| Negative                        | 62 (64.6%)       | 48 (88.8%)| 110 (73.3%)| 0.0002$^*$ |
| Positive                        | 34 (35.4%)       | 6 (11.2%)| 40 (26.7%)|           |
| **Prevalence of viral agents**  |                  |          |       |           |
| Rotavirus                       |                  |          |       |           |
| Negative                        | 93 (96.9%)       | 54 (100%)| 147 (98%)| 0.189     |
| Positive                        | 3 (3.1%)         | 0 (0%)  | 3 (2%)  |           |
| Adenovirus                      |                  |          |       |           |
| Negative                        | 96 (100%)        | 54 (100%)| 150 (100%)| No statistical analysis is computed |
| Positive                        | 0 (0%)           | 0 (0%)  | 0 (0%)  |           |
| **Prevalence of bacterial agents** |                  |          |       |           |
| *Salmonella* spp.               |                  |          |       |           |
| Negative                        | 93 (96.9%)       | 54 (100%)| 147 | 0.189     |
| Positive                        | 3 (3.1%)         | 0 (0%)  | 3     |           |
| *E. coli* O157:H7               |                  |          |       |           |
| Negative                        | 93 (96.9%)       | 54 (100%)| 147 | 0.189     |
| Positive                        | 3 (3.1%)         | 0 (0%)  | 3     |           |
| Shigella spp.                   |                  |          |       |           |
| Negative                        | 96 (100%)        | 54 (100%)| 150 | No statistical analysis is computed |
| Positive                        | 0 (0%)           | 0 (0%)  | 0     |           |

*Significant at $P < 0.05$. 

### Table 5 Prevalence of different enteric pathogens among symptomatic cases and asymptomatic carriage (controls) according to age group

| Type of pathogen | 3 years |        | 4 years |        | 5 years |        |
|------------------|---------|--------|---------|--------|---------|--------|
|                  | Cases (10) | Controls (2) | Cases (39) | Controls (12) | Cases (47) | Controls (40) |
| *E. histolytica* | 9 (90%)   | 0 (0%)  | 17 (43.6%)| 0 (0%)  | 16 (34%) | 0 (0%)  |
| *G. lamblia*     | 0 (0%)    | 0 (0%)  | 13 (33.3%)| 1 (8.3%)| 21 (44.7%)| 5 (12.5%)|
| *E. histolytica* + *G. lamblia* | 0 (0%)    | 0 (0%)  | 4 (10.3%)| 0 (0%)  | 3 (6.4%) | 0 (0%)  |
| $P$ value        | 0.007*    |        | 0.0001* |        | 0.0001* |        |
| Rotavirus        | 1 (10%)   | 0 (0%)  | 2 (5.1%) | 0 (0%) | 0 (0%)  | 0 (0%)  |
| $P$ value        | 0.64      | 0.42   | No statistical analysis is computed | No statistical analysis is computed | No statistical analysis is computed |
| *Shigella* spp.  | 1 (10%)   | 0 (0%)  | 1 (2.6%) | 0 (0%) | 1 (2.1%) | 0 (0%)  |
| $P$ value        | 0.64      | 0.57   | 0.35    |        |        | 0.10    |

| *E. coli* O157:H7 | 0 (0%)    | 0 (0%)  | 0 (0%)  | 0 (0%)  | 3 (6.4%) |
| $P$ value         | No statistical analysis is computed | No statistical analysis is computed |        |        | 0.10    |

*Significant at $P$ value < 0.05.
Table 6 Prevalence of mixed infections among symptomatic cases*

| Pattern of infection | Number | Percentage |
|----------------------|--------|------------|
| E. histolytica + G. lamblia | 7 (96) | 7.30% |
| G. lamblia + Shigella spp. | 3 (96) | 3.10% |
| G. lamblia + E. histolytica + E. coli O157:H7 | 1 (96) | 1.00% |
| Total | 11 (96) | 11.40% |

*No mixed infections were detected among asymptomatic carriage (controls).

age groups (P > 0.05). E. coli O157:H7 was only isolated (6.4%) from cases in the 5-year-old group. Therefore, all the asymptomatic control samples (100%) in all age groups were negative for bacterial infections.

Mixed infection with more than one enteric pathogen was detected in 11.4% (11/96) of the symptomatic cases (Table 6). Mixed infection with E. histolytica and G. lamblia; G. lamblia and Shigella; or G. lamblia, E. histolytica and E. coli O157:H7 were detected in 7 (3.7%), 3 (3.1%) and 1 (1.0%) samples, respectively. Nevertheless, all mixed infections were only isolated from the symptomatic cases.

Discussion

Diarrheal diseases are a major public health problem for children in Gaza strip, as in other developing communities around the world. The WHO reported that diarrheal diseases caused by viruses, parasites and enteropathogenic bacteria are a leading cause of morbidity and mortality in children; with 1.5–2.5 million deaths estimated to occur annually among children aged less than 5 years and mostly in developing countries[35]. This study was aimed to determine the prevalence of common enteropathogens including parasitic, viral and bacterial agents among kindergarten children suffering community gastroenteritis in Gaza. Out of the 150 stool samples tested, 60.7% were positive for enteropathogens. Comparing our findings with other studies conducted in Gaza strip and our region, it is in accordance, higher or sometimes lower than what was reported. In Gaza strip, Abu Elamreen et al. and Elmanama et al. found 51.5% and 66.7% positive stools for common enteropathogens in children aged less than 5 years with acute gastroenteritis and diarrhea[9,16]. In a study conducted in Jordan by Youssef et al., the prevalence of enteric pathogens in hospitalized children (aged 1–5 years) is in agreement with our findings (66.4%). However, Nimri and Meqdam in Jordan reported a higher prevalence than our study where they found 77.8% of their samples had a potential enteric pathogen[11,17]. In Saudi Arabia, El-Sheikh and El-Assouli in Jeddah reported a lower prevalence of enteropathogens (45.6%) than our findings in fecal samples collected from children (aged 0–5 years) suffering from acute diarrhea. In a recent study by Johargy et al. in Saudi Arabia, the authors reported enteropathogens in 39% of tested stool samples collected from pediatric patients aged less than 5 years[18,19].

The overall prevalence of enteropathogens in cases (diarrhea) and the asymptomatic control group (asymptomatic) was 88.5% and 11.1%, respectively. Comparing our findings with some published studies, there is consistency with some and discrepancy with others. For example, in Vietnam in a case–control study, Hien et al. identified enteric pathogens in 66% and 36% of cases and controls, respectively[20]. Albert et al. in Dhaka, Bangladesh, reported 74.8% and 43.9% potential enteric pathogen in cases with diarrhea and controls without diarrhea in children aged less than 5 years, respectively[21]. In Northwest Germany, Karsten et al. studied the incidence of community gastroenteritis of rural and urban population and found 35.8% of diarrheal samples (cases) were positive for enteropathogens and 8.7% were positive in the control group[10]. These variations could be explained by the differences in the study population, endemicity of enteropathogens, socioeconomic situation, seasonal variations, laboratory techniques used and specific practices prevalent among the different populations.

In this study, 59.2% of the examined children suffered from parasitic infection with one or more parasites. Several studies conducted in Gaza strip investigated the prevalence of parasitic infection among different age groups in different places and times around the year. Some of these studies are in accordance with our findings and others had higher or lower prevalence levels. Our findings (59.2%) were higher than that reported by Yassin et al. and Shubair et al. in Gaza city (27.6% and 24.5%, respectively). However, in another study by AL-Zain et al. in Gaza strip, a higher prevalence than our findings was reported (72.9%). Abu Elsoud et al. in Egypt studied the predictors of the intestinal parasitic infection among pre-school children living in the rural areas and found that 27% of them had intestinal parasitic infection[22–25]. The most common enteric pathogen found was E. histolytica (28%). Other studies conducted in Gaza showed that E. histolytica was the most isolated pathogen from children[9,23,26]. The prevalence rate of infection with G. lamblia was 26%. Many studies in the area reported lower or higher prevalence of
Community gastroenteritis among kindergarten children in Gaza

67

dthis parasite depending upon the characteristics of each study. In 4 different studies all conducted in Gaza strip, the prevalence of *G. lamblia* was 1.33%, 8%, 10.3% and 62%, respectively. These differences could be primarily explained by the differences in target population, time and duration of each study, seasonal variation, size of sample, and place of each study. In addition, it could be due to the influence of environmental conditions, hygiene practice, level of sanitation and laboratory techniques.

Rotavirus was isolated from 3.1% of cases. In some previous studies around the world, rotavirus was found as one of the most frequently detected enteropathogens, with high isolation rate as in Gaza, Tunisia, Saudi Arabia, Jordan, Europe and South America. These differences could be explained by the difference of the age groups that were investigated in each study and also the laboratory methods used such as ELISA or PCR techniques. The target population in most of these studies was infants and very young children who, according to the literature, have rotavirus as the commonest causative agent of gastroenteritis. In fact, our study focused on children aged 3–5 years where less prevalence of rotavirus could be present. When we compared our findings to a study conducted in Gaza on similar age groups, almost the same prevalence of rotavirus was detected. Moreover, in Bangladesh, Albert et al. studied 814 children with diarrhea. The children were up to 5 years of age. The isolation rate of rotavirus was 4.2%, 2.4%, and 1.2% from the 3-, 4- and 5-year-old groups, respectively. These findings are in accordance with our findings in the three age groups (3, 4 and 5 years old). Our study indicated that there was a tendency of decreasing rates of rotavirus infection in older children. This might partly be explained by the fact that older children acquired protective immunity during previous, probably sub-clinical exposures to rotavirus, and as a result become more resistant to infection with this agent.

Regarding the prevalence of bacterial pathogens, *Shigella* spp. and *E. coli* O157:H7 were isolated from 3.1% of symptomatic cases. Al Jarousha et al. studied bacterial enteropathogens associated with childhood diarrhea and found that 1.6% and 2.3% of cases were affected by *Shigella* spp. and *E. coli* O157:H7, respectively. However, Abu El-Amreen et al. reported 4% of *Shigella* spp. and 0.24% of *E. coli* O157:H7, although Yousif et al. reported prevalence of 4.9% and 5.7% of *Shigella* spp. and *E. coli* O157:H7, respectively. These results are largely in agreement with our findings in the same age groups.

Finally, we noticed that the infection rate was decreased as the age increased. These findings are similar to other findings published in the country and our region where the highest prevalence of enteric pathogens was mostly detected in younger children. However, as a limitation of this study, we could not exclude all other causative agents such as anaerobic and/or fastidious growing bacteria and some viruses like norovirus which require special procedures and equipment that are not available in Gaza.

In conclusion, this study demonstrates a high prevalence of parasitic enteropathogens and a relatively low prevalence of bacterial and viral enteropathogens among kindergarten children in Gaza. No difference in the prevalence of diarrhea or infection was detected between males and females. The 3-year-old group showed a significant higher positive rate of enteropathogens in comparison to the 4- and 5-year-old groups.

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