Pattern of Acute Parasitic Diarrhea in Children under Five Years of Age in Kathmandu, Nepal

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ABSTRACT

Diarrheal diseases are major problem of developing countries. Though precise data on childhood mortality associated with diarrheal diseases in Nepal is not available, it has been estimated that approximately 25% of child death are associated with diarrheal disease, particularly acute diarrhea. The purpose of this study was to assess the incidence of parasitic pathogens causing acute diarrhea in children under 5 years of age. A total of 525 children with acute diarrhea in a children’s hospital of Kathmandu, Nepal were enrolled between April 2011 and September 2011. The higher prevalence of diarrhea was in the age group of less than 2 years. Out of total 525 enrolled cases, protozoal parasites were found in 10.7% (56/525) of cases and helminthic parasites were found in 1.3% (7/525) of cases. Highest prevalence of 60.3% (38/63) of parasitic infection was found in the age group of 6 - 24 months followed by 7.9% (5/63) in the age group of less than 6 months. Of the total enrolled cases E. histolytica were 6.7% (35/525) followed by G. lamblia 3.4% (18/525) and the least frequency was due to A. lumbricoides constituting 0.6% (3/525). The aim of this study was to know the parasitic agents causing acute diarrhea in children.

Keywords: Parasitic Diarrhea; Protozoa; Helminthes; TUTH; Nepal

1. Introduction

Diarrhea is the condition of having three or more loose or liquid bowel movement per day. Diarrhea is defined by World Health Organization (WHO) as having 3 or more loose or liquid stools per day or as having more stools than is normal for that person [1]. Acute diarrhea, defined as an increased frequency of defecation (three or more times/day or at least 200 g of stool/day) lasting less than 14 days, may be accompanied by nausea, vomiting, abdominal cramping, clinically significant systemic symptoms, or malnutrition [2]. In 2009 diarrhea was estimated to have caused 1.5 million deaths in children under the age of 5 years [3]. Nepal being a developing country, diarrheal diseases are major problem. Though precise data on childhood mortality associated with diarrheal diseases in Nepal is not available, it has been estimated that approximately 25% of child death are associated with diarrheal diseases, particularly acute diarrhea [4]. The WHO Child Health Epidemiology Reference Group estimates that 16% of deaths in African children younger than five years are directly attributable to diarrheal diseases [5].

The incidence of pathogens causing diarrhea varies between developed and developing world setting. In developed countries about 70% of diarrheal cases are of viral (40% rotavirus), 10% - 20% of bacterial and <10% of protozoal origin [6-9]. In developing countries 50% - 60% cases are of bacterial (Enteropathogenic Esherichia coli 25%, Campylobacter jejuni 10% - 18%, Salmonella spp. and Shigella spp. 5% each), 35% of viral (15% - 25% rotavirus) origin, and in many the cause is unidentified or mixed [6-10]. Diarrhea, including that of parasitic origin, remains one of the most common illnesses among children and, as reported by the World Health Organization, is one of the major causes of infant and childhood mortality in developing countries [11]. Intestinal opportunistic parasitic infections are important causes of diarrhea which is a serious health problem in tropical regions. Giardia spp. and Cryptosporidium spp. are common parasitic causes of human diarrhea with the prevalence rate of 1% - 3% in the industrialized world and 4% - 17% in developing countries [12].

Many species of protozoan parasites live in the gastrointestinal tract, infecting some 3.5 billion individuals.
worldwide. Three species are of particular importance: Entamoeba histolytica, Giardia lamblia, and Cryptosporidium parvum [13]. The coccidian parasite Cyclospora cayetanensis is a newly recognized enteric pathogen causing prolonged diarrhea in humans [14,15]. It has been implicated as an important cause of diarrheal illness in the context of Nepal [16-19].

2. Methodology
This study was conducted at Tribhuvan University Teaching Hospital, Department of Microbiology-Public Health Research Laboratory. A total of 525 stool samples were collected from the children under 5 years of age visiting Kanti Children’s Hospital, Kathmandu, Nepal with acute diarrhea in the periods between April 2011 and September 2011. Written informed consent was obtained from the children’s parents or guardian before enrollment. Ethical improvement was taken from the Institutional Review Board (IRB), Institute of Medicine, Tribhuvan University Teaching Hospital, Kathmandu, Nepal.

2.1. Sampling
From each participating children, clinical data were obtained and stool sample were collected in a Clean, leak proof capped plastic container. About 5 ml of fresh stool specimen was collected from babies’ diapers or clean bed pans with a clean plastic spatula in specimen containers. Specimens were transported to the laboratory within half an hour for investigation following the standard laboratory protocol (WHO protocol) in the Public Health Research laboratory, Institute of medicine, Tribhuvan University Teaching Hospital, Maharajganj, Kathmandu.

2.2. Macroscopic Examination
The colour, consistency, presence of blood and mucus and any other abnormalities were observed macroscopically. A direct wet mount of faecal material, particularly with liquid or unformed stool, is the fastest method for detection of motile trophozoites of E. histolytica, Giardia and other intestinal parasites.

2.3. Microscopic Examination
A direct wet mount of liquid stool specimen was prepared in a drop of normal saline for the observation of pus cells, ova, cyst and trophozoites of any parasites. The presence of pus cells is one factor suggestive of invasive infection in cases of community acquired gastroenteritis. Iodine preparations of liquid stools were prepared in 5 times diluted Lugol’s iodine. 1 - 2 ml fecal suspension was placed in 12 ml conical centrifuge tube. Then Sheather’s sugar solution was added until tube was three-quarters filled. It was stirred vigorously with applicator stick. Tube was filled with sugar solution to 1 or 2 cm from the top. Then it was centrifuged at 5000 rpm for 10 minutes. Surface material was transferred to microscopic slide by means of a wire loop. It was covered with coverslip and observed with high power. Smear was also prepared for the modified Kinyoun’s acid-fast staining [20].

2.4. Analysis
Differences in proportions were assessed by Chi-square test. P values < 0.05 were considered statistically significant.

3. Result
A total of 525 diarrheal stools of the children under five years of age visiting Kanti Children’s Hospital Kathmandu, from April 2011 to September 2011 were enrolled for the study. The samples were collected from the children with acute diarrheal with or without abdominal pain, nausea, vomiting, fever, and with or without mucus and/or blood in stools. Among total enrolled cases in the study, 64.2% (337/525) were male and 35.8% (188/525) were female. Out of total 525 enrolled cases, 61.5% (323/525) were from IPD and 38.5% (202/525) were from OPD.

The higher prevalence of diarrhea was in the age group of less than 2 years among which the prevalence was highest 69.9% (367/525) in the age group of 6 - 24 months, 19.2% (101/525) in the age group of less than 6 months and the least prevalence of 2.7% (14/525) was found in the age group of 49 - 60 months (Table 1). The prevalence of diarrhea in less than 2 years of age was found to be statistically significant (P < 0.01).

Out of total 525 enrolled cases, protozoal parasites were found in 10.7% (56/525) of cases and helminthic parasites were found in 1.3% (7/525) of cases while 88% (462/525) of cases were due to other than parasites (Figure 1). Parasites infected cases were 77.8% (49/63) among male while 22.2% (14/63) were among female. Highest prevalence of 60.3% (38/63) of parasitic infection was found in the age group of 6 - 24 months followed by 7.9% (5/63) in the age group of less than 6 months.

Table 1. Age and gender wise distribution of diarrheal cases.

<table>
<thead>
<tr>
<th>Age groups in months</th>
<th>Male (%)</th>
<th>Female (%)</th>
<th>Total (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Less than 6</td>
<td>60 (17.8%)</td>
<td>41 (21.8%)</td>
<td>101 (19.2%)</td>
</tr>
<tr>
<td>6 - 24</td>
<td>238 (70.6%)</td>
<td>129 (68.6%)</td>
<td>367 (69.9%)</td>
</tr>
<tr>
<td>25 - 36</td>
<td>18 (5.3%)</td>
<td>9 (4.8%)</td>
<td>27 (5.2%)</td>
</tr>
<tr>
<td>37 - 48</td>
<td>10 (3.0%)</td>
<td>6 (3.2%)</td>
<td>16 (3.0%)</td>
</tr>
<tr>
<td>49 - 60</td>
<td>11 (3.3%)</td>
<td>3 (1.6%)</td>
<td>14 (2.7%)</td>
</tr>
<tr>
<td>Total</td>
<td>337 (100%)</td>
<td>188 (100%)</td>
<td>525 (100%)</td>
</tr>
</tbody>
</table>
months and the least prevalence of 6.3% (4/63) were in the age group of 49 - 60 months as shown in Figure 2. Occurrence of entero-parasites in children below 2 years of age was statistically significant than in those above 2 years of age (P < 0.01).

Of the total enrolled cases E. histolytica were 6.7% (35/525) followed by Giardial lamblia 3.4% (18/525), H. nana 0.7% (4/525) and Cyclospora cayetanensis and A. lumbricoides each constituting 0.6% (3/525) as depicted in Figure 3. The protozoal infected cases over helminthic infected cases was found to be statistically significant (P < 0.01).

4. Discussion

Diarrheal disease remains one of the largest health problems in many parts of the world. The disease is often mild and self-limiting but, particularly in the elderly and young children, the symptoms may be very severe. Studies in developing countries have shown that children in the first 2 years of the life may have up to 10 episodes of diarrheal disease, often with significant mortality [21]. Diarrheal disease occupied the second place among the top ten diseases in Nepal [22]. This study was conducted to find out the current trend of entero-parasites causing acute diarrhea in children under 5 years of age. In this study, 525 cases with acute diarrhea were processed for this study who visited Kanti Children’s Hospital between April 2011 and September 2011. 337 cases were male and 188 were female with male to female ratio being 1.79:1. Children below 5 years of age were enrolled in this study. In present study, among the total cases of 525, 61.5% (323/525) were from IPD and 38.5% (202/525) were from OPD.

The maximum number of samples were from the age group of less than 2 years in which the age group of 6 - 24 months constitutes maximum number (69.9%) followed by the age group of less than 6 months (19.2%). This indicates that the prevalence of diarrhea is higher in children below 2 years of age. Among the total cases enrolled, the prevalence of protozoal parasite was 10.7% (56/525) and helminthic parasite was 1.3% (7/525) and 88% (280/525) of cases were due to other than parasitic causes in this study.

Higher frequency of diarrhea was seen in male which was 64.2% (337/525) than female 35.8% (188/525). Similarly Shariff et al. in 2003 in children under 5 years of age in eastern Nepal found the prevalence of 67.5% of diarrhea in males [23]. Higher prevalence of diarrhea in male was also reported to be 56.4% by J. B. Sherchand et al. in Nepal [24], 57% by Eileen J. Klein et al. in Missouri [25] and 61.4% by Sabrina J. Moyo et al. in Tanzania [26]. Similarly the frequency of diarrhea was more in the age group less than 2 years of age among which the age group of 6 - 24 months was highest 69.9% (367/525) than the age group of less than 6 months 19.2% (101/525). The frequency of diarrhea in age less than 2 years was found to be statistically significant (P < 0.01). Similar study by M. Shariff et al. (2003) in children under 5 years of age in eastern Nepal showed the majority of diarrheal cases (70.9%) were observed in patients between 6 months and 2 years of age [23].

Higher frequency of parasites infected cases were male (77.8%) and 22.2% were female. Ogunlesi Tinuade et al. (2006) from Nigeria also found the higher prevalence of 57.1% in male whereas 42.9% of parasite positive cases were in female [27], which is consistent with our findings.

In this study the children less than 2 years were more infected than other age group among which the age between 6 - 24 months were the most infected age group followed by less than 6 months of age. The incidence of high entero-parasites among the age less than 2 years was found to be statistically significant (P < 0.01). It appeared
that infants below 6 months of age were initially protected to some extent by maternal antibodies against severe diarrhea, and they seem to have acquired adequate immunity between 12 and 16 months of age. The greater risk of infants and young children in the period between 6 - 12 months with declined levels of maternal antibodies to infection have been documented [24].

Despite the government policy for anti-helminthic program and health education program launched at community level, the intestinal parasite infestation still remains the major cause of diarrheal diseases in children [24]. In present study too the parasitic infestation was the major etiologic agent that causes diarrhea in children. The protozoa accounted for 10.7% whereas helminthes accounted for 1.3%. The most constituted parasite causing acute diarrhea was *E. histolytica* constituting 6.7% (35/525) followed by *Giardia lamblia* 3.4% (18/525), *H. nana* 0.7% (4/525) and *Cyclospora cayetanensis* and *A. lumbricoïdes* each constituting 0.6% (3/525). Ogbonnaya Ogbu *et al*. in 2006 [28], found *E. histolytica* from 3.3% cases and *G. lamblia* from 2.7% of cases which concurs with our study. Similarly Mohammad Youssef *et al*. (1994) reported *E. histolytica* from 4.9% of cases [29] and Nazek Al-Gallas *et al*. detected 2.6% of *G. lamblia* [30]. Inacio M. Mandomando *et al*. found 9.3% *A. lumbricoïdes* and 2.5% *G. lamblia* [31] and S. Das *et al*. found 4.8% *E. hystolytica*, 6.3% *G. intestinalis*, 3.6% hookworm and 4.3% roundworm as the major agents of gastrointestinal disturbance especially seen in school age children [32].

The low positivity with helminthic parasites in our study may be due to the government policy for anti-helminthic program launched at community level. Apart from this, primary school children in selected districts are being provided with deworming tablets twice yearly by World Food Program, Plan Nepal and Save the Children, US [33].

5. Conclusion

The frequency of parasitic diarrhea was higher in male children compared to female children. The study showed that the age group 6 - 24 months were found to be the most infected group among children. The frequency of *E.
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