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Full Length Research Paper

Spectrum of dysentery in children presenting to a tertiary level teaching hospital in New Delhi

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Dysentery accounts for significant morbidity in pediatric population with a high case fatality rate, if left untreated. Further, the easy availability of antibiotics has led to widespread emergence of resistant strains. The aims of this study were: (1) to study the clinical spectrum of dysentery in children, and (2) to determine various enteropathogens causing dysentery in children. 60 children in the age group 1 month to 12 years, presenting with dysentery (defined as loose stools with visible blood), were enrolled. The stool samples were cultured to determine various enteropathogens and their antibiotic sensitivity pattern. About 61.7% of children were in the age group of 6 months to 2 years. 71.7% had no dehydration at presentation. No complication was documented in our study. 80% of stool samples were grossly bloody and 58.3% were grossly mucoid. Enteropathogens were identified in 44 cases (73.3%). Leading isolates were *Shigella* in 23 cases (38.3%), *Escherichia coli* in 18 (30%). *Salmonella* were seen in 2 patients, accounting for 3.3% and *Aeromonas* in one patient. Among the *Shigella*, *Shigella flexneri* was the most frequent isolate (73.9%). Majority of *Shigella* were resistant to nalidixic acid (95.7%), norfloxacin (87%), and amoxicillin (56.5%). Most isolates were sensitive to cefotaxime, gentamycin and amikacin (95.6% each). Among the *E. coli*, EHEC were seen in 9 out of 18 (50%) cases, followed by ETEC and EPEC in 22.2% patients each. EIEC were seen in 5.6% of cases. Majority of *E. coli* were resistant to amoxicillin (95%), nalidixic acid (88.9%), norfloxacin (66.7%), and cefotaxime (56%). However, most strains were sensitive to gentamycin (88.8%) and amikacin (100%). We conclude that enteropathogen resistance to commonly used antibiotics is rapidly rising however, resistance to extended spectrum cephalosporins is still rare. Thus, local susceptibility patterns should be assessed periodically to guide antimicrobial therapy.

Key words: Dysentery, enteropathogens, antibiotic resistance.

INTRODUCTION

Acute diarrheal diseases rank 2nd among all infectious diseases, as a killer in 0 to 5 years age group. India alone loses 0.6 million children each year due to diarrhea. Morbidity due to diarrheal diseases is also very high, amounting to 6 to 12 episodes of diarrhea/year/child (Sur and Bhattacharya, 2006). Dysentery accounts for a significant proportion of all diarrhea cases. It is characterised by the passage of loose stools mixed with blood and mucus, fever, abdominal cramps and tenesmus. Worldwide, the incidence of dysentery is

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estimated to be 164.7 million cases per year, of which163.2 million were in developing countries, where 1.1million deaths occurred. About 60% of all episodes and 61% of all deaths attributable to shigellosis involved children younger than 5 years (Ashkenazi, 2004).

There is increasing recognition of wide array of enteric pathogens associated with dysentery namely, *Shigellae*, EIEC, EHEC, *Salmonella* species, *Vibrio parahaemolyticus*, *Campylobacter*, *Yersinia* and *Entamoeba histolytica*. *Shigellae* alone cause 10 to 15% of acute diarrhea and more than 50% of all the dysentery cases in less than 5 years of age and a high case fatality rate if left untreated. For the Indian subcontinent, *Shigella flexneri* continues to be the most common serogroup isolated, in contrast to the developed world where *Shigella sonnei* is common (Kotloff et al., 1999; von Seidlin et al., 2006; Naik, 2006). *E. coli* and *Campylobacter jejuni* are responsible for 25% and 5 to 15% of the dysentery cases, respectively (Sur and Bhattacharya, 2006).

Effective antimicrobial therapy can reduce both the duration and severity of dysentery. Emergence of resistance to ampicillin and co-trimoxazole in the 1980s led to the use of nalidixic acid as the first line drug for shigellosis. However, increasing number of isolates are showing resistance to nalidixic acid and quinolones, leading to therapeutic problem which needs to be studied in detail (Pazhani et al., 2005; Mamatha et al., 2007; Taneja, 2007; Dhodapkar et al., 2008; Srinivasa et al., 2009).

There is a paucity of data from north India, especially Delhi, regarding exact incidence of various enteropathogens, causing dysentery. In this study, we present the clinico-etiologic spectrum of dysentery in children presenting to our hospital with an attempt to define the causative organisms and their sensitivity pattern to various antimicrobials.

MATERIALS AND METHODS

Sixty children were enrolled in a prospective observational study conducted in diarrhea ward of Department of Paediatrics, Maulana Azad Medical College and associated Lok Nayak Hospital, New Delhi.

Inclusion criteria

Children aged between 1 month to 12 years presenting to diarrhea ward of Department of Paediatrics, Maulana Azad Medical College and associated Lok Nayak Hospital presenting with dysentery (defined as loose stools with visible blood), during the years 2009 to 2010 were enrolled in the study. These children were assessed by a detailed history and complete physical examination using a pre designed proforma. The children were managed as per WHO protocol for management of dehydration and stool samples were cultured to determine various enteropathogens causing dysentery in children. The current antibiotic sensitivity pattern of isolated pathogens was studied.

Exclusion criteria

Children with known causes of blood in stools like rectal polyps, inflammatory bowel diseases, and bleeding diathesis were excluded.

Sample collection and transportation

Stool samples were collected directly in clean containers while the child passed stools. The sample collected was transported to microbiology lab within two hours of collection. In case of delay of more than two hours, samples were transported in Cary Blair medium/buffered glycerol saline and inoculated in selenite F broth medium for enrichment.

Stool examination

Stools were examined macroscopically for colour, consistency, presence of blood, mucus, worms, and microscopically by saline preparation, iodine and Gram staining for ova, cyst, and bacteria. Stool samples were cultured directly and after enrichment in selenite F broth and alkaline peptone water (APW) onto blood agar medium, MacConkey's agar, Xylose lysine desoxycholate agar, bile salt agar and sorbitol MacConkey agar. For *C. jejuni* isolation, stool samples were cultured on Skirrow's/Butzler's/charcoal cefoperazone desoxycholate agar (CCDA) media and incubated under microaerophilic conditions at 42°C for 48 h. For isolation of *Yersinia enterocolitica*, stool samples were inoculated onto *Yersinia* selective media. The organisms were identified on the basis of colony characteristics and biochemical tests and confirmed serologically by agglutination with specific antisera. All bacterial isolates were subjected to antibiotic sensitivity using disc diffusion method. Clostridium difficile toxin A was detected using commercially available Enzyme-linked immunosorbent assay (ELISA) kit. Other investigations wherever clinically indicated include:

(a) Complete blood counts;
(b) Serum electrolytes;
(c) Kidney function tests;
(d) Blood gas analysis;
(e) Blood culture and sensitivity;
(f) Chest X-ray

Data was analyzed using chi square, Fisher exact test and t-test using statistical software Statistical package for social sciences (SPSS) version 12.0.

RESULTS

Of the sixty children recruited, 21 (35%) were male and 39 (65%) were females. The male to female ratio is 0.53. Majority of cases (61.7%) were from 6 months to 2 years of age. Lowest incidence of dysentery was seen in age group 0 to 6 months [4 out of 60 (6.7%)]. Of the fathers of 53 children, 88.3% were educated till primary school, 10% were illiterates and only one father was educated till college. Similar trend was seen in mothers' education as well. 46 out of 60 (76.7%) of mothers were educated till primary school. A higher proportion (21.7%) was illiterate

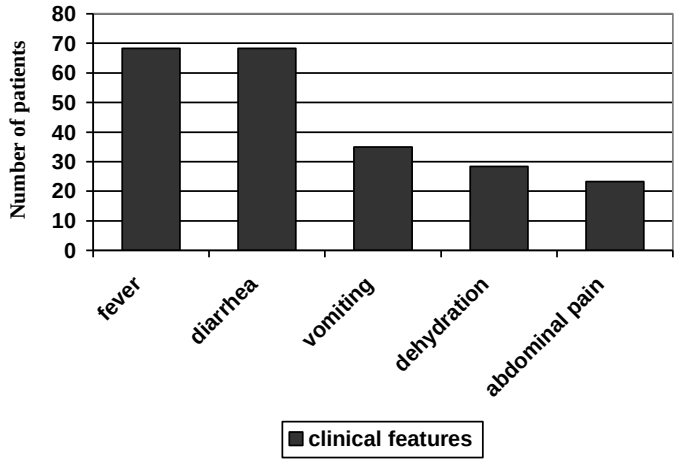

Figure 1. Clinical features of patients with dysentery.

and only a mother of only one child was educated till college.

The main presenting complaints were fever, diarrhea, vomiting and abdominal pain. Fever and diarrhea were observed in 68.3% of patients (41 out 60). Vomiting was observed in 21 (35%) children. Abdominal pain was noted in 23.3% of patients (Figure 1). In our study, majority of patients [43 out of 60 (71.7%)] had no dehydration at presentation. Some dehydration was observed in 28.3% of patients. Dehydrated patients required longer duration of hospital stay. The difference was statistically significant with p = 0.05. We found malnutrition in 44 patients (73.3%). However, severe malnutrition was only seen in 10% of children. Median time to recovery was 24 h; 51.7% (31 out of 60) recovered within 24 h. 58 patients out of 60 (96.7%) recovered within 72 h.

On gross examination of stools, blood was present in 80% (48 out of 60) of the samples. 35 out of 60 stools (58.3%) were mucoid. In microscopic examination, pus cells were seen in 90% of patients. Microscopically, RBCs were identified in 48 (80%) of cases. Presence of RBC correlated significantly with culture positive cases (p = 0.006). Cysts of *E. histolytica* were not isolated.

In hematological investigations, anemia was found in 12 (20%) patients of whom 5 (8.4%) had hemoglobin level of < 6 g/dl. Total leucocyte count was raised in 19 (31.7%) patients. All the patients had normal serum electrolytes and kidney function tests. Metabolic acidosis was not seen in anyone. 34 patients (56.7%) required only antibiotics, whereas 26 patients (43.3%) required intravenous fluids in addition to antibiotics. No complication was seen in any child.

Enteropathogens were identified in 44 cases (73.3%). Leading isolates were *Shigella* in 23 cases (38.3%), followed by *E. coli* in 18 (30%). *Salmonella* were seen in 2 patients, accounting for 3.3%. *Aeromonas* was isolated in one case (Table 1). *S. flexneri* was the most frequent isolate detected in 17 patients (73.9%) followed by *Shigella dysenteriae* in 3 (13.1%), *Shigella boydii* in 2 patients and *Shigella sonnei* was isolated in one patient. We documented a longer duration of diarrhea for cases of *Shigella* than that seen in patients of *E. coli*, p < 0.05. Majority (95.6%) of *Shigella* strains were sensitive to cefotaxime, gentamycin and amikacin. 56.5% of strains were resistant to amoxicillin, 87% were resistant to norfloxacin and 95.7% of strains were resistant to nalidixic acid (Table 2).

On serotypic analysis of *E. coli*, EHEC were seen in 9 out of 18 (50%) cases, followed by ETEC and EPEC in 22.2% patients each. EIEC were seen in 5.6% of cases. A greater degree of resistance to amoxycillin was noted (95%) and cefotaxime (56%). Majority of strains were sensitive to gentamycin and amikacin. 66.7% of strains were resistant to norfloxacin and 88.9% were resistant to nalidixic acid (Table 2).

Table 1. Enteropathogens isolated.

Parameter	N (%)
Aeromonas	1 (1.7)
E. coli	18 (30)
No growth	16 (26.7)
Salmonella	2 (3.3)
Shigella	23 (38.3)
Total	60 (100)

Table 2. Antibiotic sensitivity patterns of *Shigella* and *E. coli*.

Antibiotic	Shigella [N (%)]	E. coli [N (%)]
Amoxycillin	10 (43.5)	1 (5)
Cefotaxime	22 (95.6)	8 (44)
Gentamycin	22 (95.6)	16 (88.8)
Amikacin	22 (95.6)	18 (100)
Norfloxacin	3 (13)	6 (33.3)
Nalidixic acid	1 (4.3)	2 (11.1)

DISCUSSION

Sixty children with blood in stools were recruited. Majority of cases (61.7%) were 6 months to 2 years of age due to unhygienic introduction of complementary feeds. Lowest incidence of dysentery was seen in age group 0 to 6 months [4 out of 60 (6.7%)]. This is attributed to predominant breastfeeding practice in this age group. Male

preponderance was noted in shigellosis cases with M: F ratio being 2.2:1. Similar findings were reported by Naik (2006) from Africa, Taneja (2007) from India and Ghaemi et al. (2007) from Iran.

A relation was observed between educational status of parents and dysentery in children, with majority of parents being educated till primary school, a finding similar to that reported by Rustom et al. (2006). The main presenting complaints were fever, diarrhea, (68.3% each), vomiting (35%) and abdominal pain (23.3%). The findings are consistent with that observed by von Seidlin et al. (2006) who reported watery diarrhea in 65% of dysentery cases, fever in 45%, vomiting and abdominal pain in 20% cases. They also reported mucoid diarrhea and abdominal pain more frequently in culture positive cases, and vomiting and watery diarrhea were more often seen in culture negative cases. However no such association was seen in our study.

Dutta et al. (2003) also documented fever in 63.8% and abdominal pain in 20.4% of children presenting with bloody diarrhea. In our study, majority of patients [43 out of 60 (71.7%)] had no dehydration at presentation. Some dehydration was observed in 28.3% of patients. Dehydrated patients required longer duration of hospital stay. The difference was statistically significant, with $p = 0.05$. On the contrary, Dutta et al. (2003) documented moderate dehydration in 87.8% and severe dehydration in 10% of dysentery cases. We found malnutrition in 44 patients (73.3%). However, severe malnutrition was only seen in 10% of children. Malnutrition and dehydration have not been reported as significant problem in literature from the West.

On gross examination of stools, blood and mucus were found in 80% (48 out of 60) and 58.3% (35 out of 60) stool samples, respectively. In microscopic examination, pus cells were seen in 90% of patients. Rustam et al. (2006) documented mucoid stools in 96.3% of patients and pus cells in 100% of stool samples. Microscopically, WBCs were found in 73.3% and RBCs in 80% of samples. Presence of RBCs correlated significantly with culture positive cases, $p = 0.006$. Patwari et al. (1993) also observed the presence of RBC and WBC as significant predictor of dysentery. No complication was seen in any child. However, about 10 years back, Thapa et al. (1995) reported central nervous system (CNS) manifestations in 45% of patients, renal failure in 25% and subacute intestinal obstruction in 5% of cases. This difference might be due to change in health service seeking attitudes, leading to earlier treatment and better management of cases.

In our study, enteropathogens were identified in 44 cases (73.3%). Leading isolates were *Shigella* in 23 cases (38.3%), followed by *E. coli* in 18 (30%). Very few studies have documented spectrum of enteropathogens in dysentery cases. Our findings are similar to those of Dutta et al. (2003) who enrolled 100 patients with bloody diarrhea, among whom *Shigella* was isolated in 39% of cases. Among the cases due to *Shigella*, *S. flexneri* was the most frequent isolate detected in 17 patients (73.9%), followed by *S. dysenteriae* in 3 (13.1%). The findings are consistent with those of Pazhani et al. (2005) in Kolkata, who documented *S. flexneri* as the most common isolate (60%).

Mamatha et al. (2007) (Manipal) also reported *S. flexneri* in majority of cases (45%). Uppal et al. (2004) from New Delhi also isolated *S. flexneri* in 78.5% of cases. This is in accordance with the fact that *S. flexneri* is more common in developing countries in contrast to developed countries where *S. sonnei* is most common (Kotloff et al., 1999).

Among the *Shigella* spp., majority were resistant to nalidixic acid (95.7%), norfloxacin (87%), and amoxicillin (56.5%). Most isolates were sensitive to cefotaxime, gentamycin and amikacin (95.6% each). Similar resistance patterns have been reported by Pazhani et al. (2005) and Dutta et al. (2003) from east, Dhodapkar et al. (2008) and Srinivasa et al. (2009) from south and Taneja (2007) from north India. None of the strains were resistant to ceftriaxone. Our study confirms that amoxicillin and nalidixic acid have no role in dysentery now. Nalidixic acid was recommended by the WHO as the first-line treatment against shigellosis until 2004, when it was replaced by ciprofloxacin (von Seidlin et al., 2006). However there is growing concern regarding resistance to quinolones as documented by this study and by other authors (Pazhani et al., 2005; Mamatha et al., 2007; Taneja, 2007; Uppal and Arora, 2004).

A total of 18 isolates of *E. coli* were documented (30% of total dysentery cases) which was higher that that reported from other parts of India (Das et al., 2007). On serotypic analysis, EHEC were seen in 9 out of 18 (50%) cases, followed by ETEC and EPEC in 22.2% patients each. EIEC were seen in 5.6% of cases. Serotypic prevalence of *E. coli* has not been studied extensively in bloody diarrhea. A study in India by Bhan et al. (1989) found no STEC in children with diarrhoea in Delhi. In low and middle income countries, prevalence of STEC in childhood diarrhea has been reported to be lower than that of ETEC, EPEC, or EAEC by Presterl et al. (2003), however our study documents the growing significance of STEC in childhood dysentery. There is paucity of studies reporting antibiotic susceptibility of *E. coli* strains from childhood dysentery.

In our study, 95% of isolates were resistant to commonly used drug amoxicillin, 88.9% of strains were resistant to nalidixic acid, resistance to norfloxacin was observed in 66.7% cases. 56% isolates were resistant to cefotaxime. However, majority of strains were susceptible to aminoglycosides, gentamycin (88.8%) and Amikacin (100%). High degree of resistance to commonly used antibiotics was also noted by Das et al. (2007) who reported

33% of *E. coli* strains to be resistant to norfloxacin, 33% were resistant to gentamicin, 44% of the strains were resistant to cefotaxime and 77% were resistant to nalidixic acid. Thus local susceptibility patterns should be assessed periodically to guide antimicrobial therapy.

Abbreviations: EAEC, Enteroaggregative *Escherichia coli*; **EHEC,** enterohemorrhagic *Escherichia coli*; **EIEC,** Enteroinvasive *Escherichia coli*; **EPEC,** Enteropathogenic *Escherichia coli*; **ETEC,** Enterotoxigenic *Escherichia coli*; **RBC,** red blood cell; **STEC,** Shiga toxin producing *E. coli*; **WBC,** white blood cells; **WHO,** World Health Organization.

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