Clinical Profile and Epidemiology of Campylobacter Associated Diarrhea Among Children in New Delhi, India

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Abstract

Background: Campylobacter, a well-known enteropathogen among children shows variable clinical presentations. Age groups and seasonal distribution is dependent on geographical position.

Objectives: To explore clinical manifestations and seasonal variation of Campylobacter infection and to study its importance as enteric pathogen among children.

Patients and Methods: Two hundred five children (≤12 years age) having acute diarrhea as cases and 100 children without from diarrhea were taken as control. All the fecal samples were processed for Campylobacter species by culture on to modified charcoal cefoperazone deoxycholate agar and Skirrow’s Columbia blood agar media. Detection of Campylobacter specific antigen in faecal samples was also done by enzyme-immuno assay.

Results: A total of 32 (15.61%) faecal samples of children with diarrhea had positive results for Campylobacter spp. Among them 31.25% cases had polymicrobial infections. Children below 1 year were most commonly (18.96%) affected by the infection. The organism was isolated throughout the year with a higher isolation rates during summer and monsoon months. Watery diarrhea was significantly more common in the Campylobacter infected cases.

Conclusions: Application of antigen assay increases detection rate of Campylobacter enteritis cases, which was significantly higher than the control group (P<.05). Specific clinical profile could not be associated with this infection which, indicates need of microbiological diagnosis of this pathogen for antibiotic therapy.

Keywords: Campylobacter, Diarrhea, Polymicrobial infection, Pediatrics

Background

Campylobacter species are primarily zoonotic, with a wide variety of wild and domestic animals, especially birds implicated as reservoir. They mainly cause foodborne gastroenteritis following ingestion of chicken, raw milk, untreated water and contact with pets, especially household live chickens. Increasing trend of Campylobacter infections have been seen in developed countries for years and it accounts for one of the most common bacterial causes of diarrhea, with an incidence ranging from 10% to 46%.

In recent times there are many reports from developing countries describing Campylobacter jejuni and Campylobacter coli as important enteropathogen during first 5 years of life with isolation rate ranging from 10% to 46%.

Though, the epidemiology, clinical presentations and microbial profile are not similar in these two economical world territories. Application of newer, sensitive molecular diagnostic methods beside culture might play role behind the increasing detection rate of this fastidious, microaerophilic organism. It is present as colonized gastrointestinal flora and also as asymptomatic carrier commonly in children, but it causes severe gastroenteritis among young children <2 years old, elderly or immunocompromised patients and may require antibiotic therapy. Thus, various virulence markers had been studied to prove its pathogenicity.

Clinically, Campylobacter infection is indistinguishable from acute gastrointestinal infections produced by other bacterial pathogens. In some patients, the diarrhea is minimal and abdominal cramps and pains are the predominant features; this can lead to a mistaken diagnosis of acute abdomen and unnecessary laparotomy. Fluoroquinolones and macrolides are main parts of treatment used for this infection; however, marked increase in fluoroquinolone resistance and presence of high level azithromycin resistance in Campylobacter isolates are becoming threat in In-
The most important post-infectious complication of *C. jejuni* infection is the Guillain-Barré syndrome which affects 1–2 persons per 100 000 populations in the United States each year.¹⁰,¹¹

**Objectives**

The present study was designed to investigate clinical manifestations and epidemiology of Campylobacter infection and to study the importance of *C. jejuni* as an enteric pathogen among children.

**Patients and Methods**

The study was conducted with ethical permission in the Department of Microbiology, Maulana Azad Medical College and Department of Pediatrics, LN hospital, New Delhi for 2 consecutive years. The study group included 205 patients aged 12 years or below having acute diarrhea (<14 days duration) admitted in diarrhea ward of the hospital. A total of 100 age and sex matched children without any gastrointestinal complaints were taken as control. After proper counselling, an informed consent was taken from the parents/guardians/person attending the study subject. Detailed personal history, diarrheal episode and associated signs and symptoms were recorded on a pre-designed pro forma.

**Exclusion Criteria**

Children on antimicrobial therapy were excluded from the study.

**Sample Collection and Transport**

Stool samples were requested from all patients and controls who fulfilled the inclusion criteria. Proper instructions were given regarding collection of specimen i.e. freshly passed faeces to be collected in a clean, wide mouth, screw capped plastic container and transported to microbiology laboratory within 2 hours of collection. In case of delay of more than two hours, samples were transported in Cary Blair medium/ buffered glycerol saline.

**Examination of Sample**

The stool specimen was processed as follows:

**Culture:** All fecal samples were processed for *Campylobacter* species by direct inoculation and after enrichment in BHI broth on modified charcoal cefoperazone deoxycholate agar (CCDA) (Oxoid) and Skirrow’s Columbia blood agar media with *Campylobacter* growth supplement and *Campylobacter* selective supplement (Butzler) (Oxoid) containing bacitracin (12,500 IU), cycloheximide (25 mg), colistin sulfate (5000 IU), cephalothin sodium (7.5 mg) and novobiocin (2.5 mg). The plates were incubated along with control strain of *C. jejuni* for 48 hours at 42°C under microaerophilic conditions (5% O₂, 5% CO₂, 2% H₂, and 88% N₂ by volume) generated by ANOXOMAT AN2OP system. Plates were examined after 48 hours and in case of no growth re-examined after 72 hours and then again after 7 days of incubation.¹²

Suspected colonies of *Campylobacter* grown were confirmed by oxidase test, catalase test, hippurate hydrolysis, hydrolysis of indoxyl acetate, growth on 1% glycine and 1.5% NaCl and susceptibility to cefoperazone (30 μg), nalidixic acid (30 μg) and cephalothin (30 μg) as per standard techniques.¹³

All samples were examined by wet mount for the presence of parasites and inoculated on several diagnostic media such as MacConkey’s agar, xylose lysine deoxycholate agar, blood agar and bile salt agar directly and after enrichment in selenite F broth and alkaline peptone water for the isolation of conventional enteropathogens. Characteristic colonies were identified by based on colony characteristics, biochemical reactions and agglutination test with respective antisera.

**Detection of Campylobacter Antigen in Stool Samples**

ProSpecT™ Campylobacter Microplate Assay (Oxoid Ltd, UK) was used for qualitative detection of *Campylobacter* specific antigen in faecal samples as per manufacturers’ instructions.

**Statistical Analysis**

All data obtained was analyzed using SPSS statistical software. Chi-square test with Yates correction, Fisher exact test were used to compare the results, wherever applicable.

**Results**

A total of 32 (15.61%) children with diarrhea had positive results for *Campylobacter* antigen among which 15 samples yielded growths on culture media. The detection rate from the controls without diarrhea was 4%. The difference between the isolation rates was statistically significant (P<0.05). All the isolates were identified as *C. jejuni*. Among 32 positive cases, 22 (68.75%) children were infected with *C. jejuni* as a sole pathogen, whereas 10 (31.25%) cases had polymicrobial infections. Most common pathogen isolated along with *C. jejuni* was *Vibrio cholerae* O1 Ogawa (15.62%) followed by enteropathogenic *Escherichia coli* (EPEC) 9.4% and rotavirus in 3.12% cases. One case had triple infection with *C. jejuni*, EPEC and rotavirus. Mean age of children with *Campylobacter* infection was 9 months with peak incidence (18.96%) in children below 1 year (Table 1). Males were more frequently (1.5:1) infected than females.

Seasonal distribution of *Campylobacter* infection is presented in Figure 1. A total of 71.87% isolates were detected during the summer and monsoon months of which highest recovery was in the month of July (25%).

Watery diarrhea was significantly more common than inflammatory diarrhea in the *Campylobacter* infected cases (Table 2). Fever was observed in 59% cases infected with *Campylobacter* alone, but it was significantly more (90%) associated with mixed infection, P = .0402 (Table 3). Abdominal pain and vomiting were equally common in both the groups (75% and 90.62%). In contrast, dehydration was observed in 59% cases infected with *Campylobacter* as sole pathogen, but was much more common among mixed infection cases (90%), P < .05, which was
Table 1. Age Distribution of Campylobacter Isolates Both From Cases and Control

<table>
<thead>
<tr>
<th>Age (y)</th>
<th>No. of Patients Studied</th>
<th>Positive for Campylobacter, No. (%)</th>
<th>No. of Controls Studied</th>
<th>Positive for Campylobacter, No. (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;1</td>
<td>116</td>
<td>22 (18.96)</td>
<td>58</td>
<td>0 (0)</td>
</tr>
<tr>
<td>1-2</td>
<td>37</td>
<td>6 (16.22)</td>
<td>19</td>
<td>1 (5.26)</td>
</tr>
<tr>
<td>2-5</td>
<td>27</td>
<td>3 (11.11)</td>
<td>13</td>
<td>2 (15.38)</td>
</tr>
<tr>
<td>&gt;5</td>
<td>25</td>
<td>1 (4)</td>
<td>10</td>
<td>1 (10)</td>
</tr>
<tr>
<td>Total</td>
<td>205</td>
<td>32 (15.61)*</td>
<td>100</td>
<td>4 (4)*</td>
</tr>
</tbody>
</table>

*There was a significant difference between the isolation rates of Campylobacter spp. in cases and controls in total was significant (P=.0023) but between each age group was not significant (P>.05).

Table 2. Clinical Presentation of Campylobacter Infection

<table>
<thead>
<tr>
<th>Findings</th>
<th>Campylobacter Positive Cases, n=32 (%)</th>
<th>Campylobacter Negative Cases, n=173 (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Watery diarrhea*</td>
<td>25 (78.12)</td>
<td>66 (38.15)</td>
</tr>
<tr>
<td>Inflammatory diarrhea</td>
<td>7 (21.87)</td>
<td>35 (20.23)</td>
</tr>
<tr>
<td>Fever (&gt;100°F)*</td>
<td>22 (68.75)</td>
<td>89 (49.13)</td>
</tr>
<tr>
<td>Abdominal pain*</td>
<td>24 (75)</td>
<td>40 (23.12)</td>
</tr>
<tr>
<td>Vomiting*</td>
<td>29 (90.62)</td>
<td>53 (30.63)</td>
</tr>
<tr>
<td>Dehydration*</td>
<td>22 (68.75)</td>
<td>52 (30.05)</td>
</tr>
<tr>
<td>Cough and coryza</td>
<td>3 (9.37)</td>
<td>27 (15.61)</td>
</tr>
<tr>
<td>Convulsion</td>
<td>2 (6.25)</td>
<td>19 (10.98)</td>
</tr>
</tbody>
</table>

*P<.0001.
ly associated than inflammatory diarrhea in the Campylobacter infected cases (71.12% vs. 21.87%; P < 0.0001). Though a study from Pakistan found blood and mucus in 90% of diarrheal stools that yielded C. jejuni, Bhadra et al. noticed watery diarrhea in 97.6% of C. jejuni/coli infected cases.\textsuperscript{1,7}

In comparative analysis to investigate the difference in clinical presentation between the patients infected with Campylobacter alone (n = 22) and those infected with multiple pathogens (n = 10), fever and dehydration was found to be more common in mixed infections. Though Tribble et al. evaluated sensitivity and specificity of various clinical presentations and stool characteristics as modality to diagnose Campylobacter infection, we could not associate any clinical pattern specific for Campylobacter enteritis.\textsuperscript{24}

In the present study Campylobacter comprised a significant percentage of enteropathogens among children in India, which indicates requirement of routine identification of this pathogen. Moreover, clinical features could not be used to diagnose of campylobacteriosis per se because of the non-specific nature of the symptoms.

Conflict of Interest Disclosures
None.

References


