

Shigellae – associated diarrhoea in children in Baghdad – Iraq

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Abstract

Background: *shigella* spp. reported to be the second commonest bacterial agent responsible for childhood diarrhea after *E.coli*. Currently, isolation of the bacterium and confirmation of the diagnosis by bacteriological and biochemical methods remains the "gold standard".

Objective: To determine the prevalence of *shigella* spp. among children below 3 years with acute diarrhoea and susceptibility of the isolates to commonly used antimicrobials.

Methods: This study was carried out in the outpatient's clinic of Children Central Teaching Hospital in Baghdad from May 2007 – April 2008. One hundred and fifty children below 3 years with acute diarrhea were the source of stool specimens to detect *shigella* spp. All isolates were diagnosed according to bacteriological and biochemical standard methods. Available antimicrobial were used to determine the susceptibility of isolates to antibiotics

Results: *Shigella flexneri* type 2 was the predominate serotype out of 9 isolates. The prevalence of *shigellae* isolates was significantly higher in children older than one year. All patients were on artificial feeding, 78% were using untreated water for drinking. All isolates were sensitive to ceftriaxone, ciprofloxacin, nalidixic acid, norfloxacin and gentamicin. Drug resistant to 3 or more drugs was found in 56% of the isolates.

Conclusion: *Shigella flexneri* type 2 was the predominate serotype and most isolates were resistant to trimethoprim-sulphamethoxazole (89%).

Keywords: *Shigellae*, Diarrhoea, Antibiotic, Children, Iraq.

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Introduction

Diarrhoeal diseases are one of the main causes of death especially among young children which leads to at least five million deaths each year ⁽¹⁾. Endemic bacillary dysentery accounts for about 10% of the disease in children aged less than five years ^(2, 3). Among the different pathogens responsible for diarrhoea, *shigella* spp. play an important role in causing inflammatory diarrhoea and dysentery ^(4, 5), with a significant morbidity and mortality in developing countries ⁽⁶⁾.

Shigellosis is an acute diarrhoeal disease caused by *shigella* spp.

It has caused and continued to be responsible for morbidity and / or mortality in high risk populations such as children under 5 years of age ⁽⁷⁾. Spread of shigellosis is through contaminated water, poor sanitation and overcrowded areas ⁽⁸⁾. Over the past decades, *shigella* spp. has become progressively resistant to the most widely used and inexpensive antimicrobials ^(4, 9, 10, 11). Moreover changes in the virulence of *shigella* spp. make it difficult to formulate a drug of choice for the treatment of shigellosis ⁽¹²⁾.

Patients and methods

Patients:

This study was carried out in the outpatient Clinic of Children Central Teaching Hospital in Baghdad during the period from May 2007 - April 2008. One hundred and fifty children with acute diarrhoea age rang from few

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days to 3 years.(Most children (84%) were less than one).

Specimens collection:

Stool specimens were collected in sterile wide mouth containers.

Methodology:

All isolates were diagnosed according to well known microbiological methods⁽¹³⁾. For optimal isolation, three different media and an enrichment medium were used. The samples were inoculated directly on MacConkey agar, xylose – lysine deoxycholate agar and *Salmonella-Shigella* agar. Enrichment was done in Na – Tetra thionat broth and incubated at 37C overnight. Biochemical identification of *shigella* spp. has been performed according to standard methods⁽¹⁴⁾. Confirmation of diagnosis was through slide agglutination test using commercially available antisera (Wellcome Diagnosis, UK)⁽¹⁴⁾.

Antimicrobial susceptibility tests:

Shigella isolates were examined for their susceptibilities to ampicillin , chloramphenicol, ceftriaxone, ciprofloxacin, gentamicin, kanamycin, nalidixic acid, norfloxacin and co - trimoxazole by the standard disc - diffusion method⁽¹⁵⁾.

Results

Table (1) shows that *shigella flexneri* type 2 was the predominate serotype out of 9 isolates. Of children with diarrhoea , *shigellae* were isolated equally from both sexes (5 from 80 males and 4 from 70 females) . A total of 6 *shigella flexneri* and 3 *shigella sonnei* were isolated. Of the 6 strains

of *shigella flexneri* 4 were *shigella flexneri* type 2 , 1 of *shigella flexneri* type 1 and 1 of *shigella flexneri* type 3. Out of 9 *shigella* strains , 5 (56%) were isolated in summer , 1 (11%) in autumn , 2 (22%) in winter , 1 (11%) in spring. Three *shigella* strains were isolated from 50 children with diarrhoea aged 6 months to one year , 6 strains out of 24 patients aged more than one year and none were isolated from 76 children less than 6 months old . All patients with *shigella* isolates (100%) were on artificial feeding, 7 (78%) were using untreated water for drinking, 1 (11%) took Bactrim 2 days prior to stool collection and none travelled abroad in the last 30 days before stool collection . Blood and mucus were seen in the stool of 5 (56%) patients with diarrhoea . Clinical examination showed 6 (67%) with fever and 3 (33%) with vomiting , 1 (11%) was diagnosed to have septicemia and 1(11%) had generalized convulsion. Clinical findings are shown in Table - 1 . The drug susceptibility patterns of 9 isolates of *shigella* strains were determined . Resistant strains to ampicillin was found to be 78% , 67% to chloramphenicol , 67% to kanamycin , 56% to streptomycin and 89% to trimethoprim-sulphamethoxazole . All isolates were sensitive to ceftriaxone, ciprofloxacin, nalidixic acid, norfloxacin and gentamicin as compared to Table-2. Drug resistance to 3 or more drugs was shown by 56% of strains.

Table 1 :Information about the children with diarrhoea and their *shigella* species isolates.

Patients number	Gender	Age	Month of Occurrence	Use of untreated drinking water	Stool with mucus	Stool with blood	Fever	Vomiting	Duration of diarrhea in (days)	Frequency	Species and Serotype of <i>Shigella</i>
1	F	11 mo.	May	No	+	+	-	-	1	5	<i>Sh. sonnei</i>
2	F	30 mo.	June	Yes	-	-	-	-	2	3	<i>Sh. flexneri</i> type 2
3	M	27 mo.	July	Yes	-	-	+	-	1	6	<i>Sh. sonnei</i>
4	F	18 mo	July	Yes	-	-	-	-	1	8	<i>Sh. flexneri</i> type 2
5	F	3 yrs.	August	No	+	+	+	-	2	5 - 7	<i>Sh. flexneri</i> type 2
6	M	6 mo.	September	Yes	-	-	+	-	7	7 - 8	<i>Sh. flexneri</i> type 1
7	M	6 mo.	November	Yes	+	+	+	+	1	10	<i>Sh. flexneri</i> type 2
8	M	13 mo.	December	Yes	+	+	+	+	10	6 - 7	<i>Sh. flexneri</i> type 3
9	M	32 mo.	March	Yes	+	+	+	+	1	4	<i>Sh. sonnei</i>

Table 2: Antimicrobial susceptibility patterns of *shigella* spp. isolates from outpatients presenting at Hospital in Baghdad - Iraq.

Antimicrobial agent	Resistant			Intermediate			Susceptible		
	Zone size(mm)	No.	%	Zone size(mm)	No.	%	Zone size(mm)	No.	%
Ampicillin	≤ 13	7	78	14 - 16	0	0.0	≥ 17	2	22
Chloramphenicol	≤ 12	6	67	13 - 17	1	11	≥ 18	2	22
Ceftriaxone	≤ 14	1	11	15 - 16	0	0.0	≥ 17	8	89
Ciprofloxacin	≤ 12	1	11	13 - 14	1	11	≥ 15	7	78
Gentamicin	≤ 15	2	22	16 - 19	2	22	≥ 20	5	56
Kanamycin	≤ 13	6	67	14-17	1	11	≥ 18	2	22
Nalidixic acid	≤ 14	2	22	15 - 18	1	11	≥ 19	6	67
Norfloxacin	≤ 13	3	33	14 - 17	0	0.0	≥ 18	6	67
Streptomycin	≤ 11	5	56	12 - 14	2	22	≥ 15	2	22
Trimethoprim-Sulphamethoxazole	≤ 10	8	89	11 - 15	0	0.0	≥ 16	1	11

In total 9 *shigella* strains were tested.

Discussions

In the present study *shigella flexneri* was found to be the most frequent isolate in children with *shigellae* associated diarrhea in our study, rate in Baghdad with type 2 being the predominating serotype. These findings conform with several studies^(16,17,18), and other developing countries^(19,20,21), but they are in contrast with studies in developed countries where *shigella sonnei* is dominant and *shigella flexneri* is the second most prevalent isolate^(4,22,23). In some countries, *shigellae* were isolated more often from children older than 2 years of age than from younger children^(24,25,26). In our experience *shigella* spp. were isolated only from children who were not breast - fed and significantly more frequently from children older than one year of age⁽²⁷⁾. Human milk has been shown to protect against severe shigellosis in children up to 35 months of age⁽²⁸⁾. Studies showed that all milk samples obtained from mothers contained antibodies to antigens encoded by the large virulence plasmid in strains of *shigella*⁽²⁹⁾. In addition to this, children more than one year old are capable of moving around and come into more direct contact with other children and adults which expose them under certain circumstances, to infection with these organisms. Food and waterborne outbreak of shigellosis have been reported from different parts of the world^(8, 30, 31). Most (78%) of our patients with stools positive for *shigella* have used untreated drinking water. We found blood and mucus in the stools of 56% and fever in 67% of our patients. These clinical findings are in line with those reported by other investigators in under - developed countries^(19,20). Several investigators reported cases of generalized convulsions associated with shigellosis^(16,32,33). Of our patients, only

one (11%) had generalized convulsions and we believe this to be the first time such a case has been reported from Iraq which conform by Daoud *et al.*⁽¹⁶⁾, who studied 93 children with shigellosis and found 15% of the patients developed generalized convulsions. They reported that neither specific diagnostic procedures nor drug therapy were usually necessary due to benign and self - limiting nature of convulsions associated with shigellosis.

Antibiotics can be useful in the treatment of *shigella* - associated diarrhoea, however trimethoprim - sulphamethoxazole no longer to be considered the drug of choice in our hospital as the majority (89%) of the local isolates were resistant to this antibiotic. Several studies reported the same findings^(16, 17). AL - Eissa *et al.*⁽¹⁷⁾, from Saudi Arabia reported 74% of their *shigella* isolates were resistant to trimethoprim - sulphamethoxazole. All our isolates were sensitive to ceftriaxone, ciprofloxacin, nalidixic acid, norfloxacin and most of them to gentamicin, therefore they should be considered the drugs of choice for treatment of diarrhea due to *shigellae*, however, susceptibility testing to antibiotics before administering the drug is recommended.

The present study shows that *shigellae* associated diarrhea in children is still a public health problem.

Shigella flexneri type 2 was the predominate serotype and most isolates were resistant to trimethoprim - sulphamethoxazole (89%), thus it no longer to be considered the drug of choice in the treatment of *shigellae* associated diarrhea in children below 3 years in our study.

Since all isolates were sensitive to ceftriaxone, ciprofloxacin, nalidixic

acid, so they can be considered as the drugs of choice.

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