

Original Research Article

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## *Escherichia coli* Gastroenteritis in Children Aged 0 To 5 in Three Hospitals in Chad

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### ABSTRACT

Introduction: Infectious gastroenteritis in children is a major public health concern. They are linked to the presence of certain pathogenic bacteria, particularly the *Escherichia coli* tribe. Enteropathogenic. The aim of the study was to determine the prevalence of *Escherichia coli* responsible for diarrhoea isolated from children aged 0-5 years. Material and Methods: This was a descriptive cross-sectional study conducted from 01 July to 31 December. 31 October 2021 at the Hôpital Régional de Sarh (HRS), the Centre Hospitalier Universitaire de la Mère and Child Hospital (CHU-ME) and the Notre Dame des Apôtres Hospital (HNDA) in N'Djamena. Stool analyses were carried out at the Bacteriology Laboratory of the Centre Hospitalier Universitaire de la Mère et de l'Enfant. The stools were collected in sterile jars containing Cary-Blair transport medium and then sent to the Bacteriology laboratory of the Centre Hospitalier Universitaire de la Mère et de l'Enfant for culturing on Eosine Bleu de Méthylène (EMB) agar and incubating in a bacteriological oven for 18 to 24 hours at 37°C. Colonies from this metallic sheen culture were subcultured onto Muller Hinton agar for purification and antigenic studies. Gram staining and oxidase tests were performed on the different strains. Biochemical identification of the isolates was carried out using the Le Minor slide and the API 20 E Gallery in accordance with Bio Mérieux recommendations. Results: A total of 296 stools were processed, and 36 strains of enteropathogenic *Escherichia coli* were isolated, giving an overall prevalence of 12.2%. Higher prevalences were obtained at HRS (16%), CHU-ME (11.3%) and HNDA (9.9%). The 12-59 month age group of children was the most infested (86.1%) in the three hospitals. Male children were infested at 58.3%. The following serotypes were identified: O111, O26, O86, O125, O126, O124, O114, O142 with a prevalence of (25%) for serotype O124 followed by serotype O126, O114 and O142 with 13.9%. Pathogenic *E. coli* strains were most isolated in July (58%) followed by August (27.8%). Households where children used boreholes as a source of drinking water were the most infected at 63.9%, followed by water from the STE at 25.8%. According to the results obtained, the infections were of food, environmental and hygienic origin. Conclusion: The study confirmed that *E. coli* remains the predominant species of gastroenteritis in diarrhoeal infections among children in Chad. There is a need to raise awareness of food and environmental hygiene among children. It would be important to broaden the scope of research into these enteropathogens in the food, environmental and clinical fields, to gain a better understanding of the origins and circulation patterns of these fearsome strains.

#### Keywords

Gastroenteritis,  
*Escherichia coli*,  
serotype, children  
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## Introduction

Infectious gastroenteritis in children is a major public health concern. They are linked to the presence of certain pathogenic bacteria, particularly the *Escherichia coli* tribe. Enteropathogenic (Titilawo *et al.*, 2015; Lanata *et al.*, 2013).

All age groups of the population are affected by gastroenteritis. However in infants and young children, enteric diseases are the second leading cause of death in children under 5, with an estimated 2.5 billion cases worldwide and 1.5 million deaths each year (Diawara, 2018). Mortality rates are particularly high in sub-Saharan Africa.

*Escherichia coli* is a bacterium commonly found in the digestive tract of humans and warm-blooded organisms (WHO, 2020). Most strains are harmless. Some, however, can cause food poisoning, leading to serious illness (WHO, 2020; Mariama *et al.*, 2022).

They are generally associated with malnutrition and other concomitant infections, particularly bacterial diarrhoea. In addition, many bacteria are implicated in the aetiology of gastroenteritis. Some have well-established enterovirulent properties (*Salmonella* spp, *Shigella* spp, *Campylobacter* spp, *Yersinia* spp, etc.). Other bacteria have become pathogenic after acquiring virulence factors. This is the case of *Escherichia coli* (*E. coli*), a species that accounts for 80% of the aerobic intestinal flora in humans.

In Africa, detection of this germ remains difficult in most developing countries because of the lack of laboratory equipment. The prevalence was 23.1% in Nigeria, 12% in Ethiopia and 3.9% in Senegal (Sylla, 2005). Among the most recent studies carried out in Mali on diarrhoea and urinary tract infections, the frequency of *Escherichia coli* was 43.9% and 45.3% respectively (Sylla, 2005). It is estimated that 15,900 children under the age of 5 die each year from diarrhoea in Chad. 90% of these deaths are directly linked to the consumption of unclean water and the lack of sanitation and hygiene (WSP, 2012).

In Chad, a study carried out in 2013 showed a prevalence of 34.73% of cases of diarrhoea linked to all enteropathogens (Bessimbaye, 2013).

Work carried out at the Hôpital de la Mère et de l'Enfant (HME) on the prevalence and sensitivity of *E. coli* strains to antimicrobials showed a 25.6% (Yandai, *et al.*, 2014).

With a view to comparing data with previous studies, this study was carried out to describe the epidemiological profile of enteropathogenic *Escherichia coli* strains isolated from the stools of children aged 0 to 5 years in hospitals in Chad. The aim was to gain a better understanding of how *E. coli* gastroenteritis manifests itself, so that it can be properly managed.

## Materials and Methods

### Study framework

The study was carried out in two towns in Chad: N'Djamena (Centre Hospitalier Universitaire de la Mère et de l'Enfant and Hôpital Notre Dame des Apôtres) and Sarh (Hôpital Régional). Analyses were carried out at the bacteriology unit of the Centre Hospitalier Universitaire de la Mère et de l'Enfant.

### Type, period and study population

This was a descriptive cross-sectional study from July to October 2021. It involved children of both sexes aged 0 to 5 years who were seen in paediatric wards where a bacteriological stool examination was requested.

### Inclusion and non-inclusion criteria

All children aged between 0 and 5 presenting with gastroenteritis and whose parents agreed to take part in the study were included. The study did not include children aged over 5 years, children without gastroenteritis or children with gastroenteritis whose parents refused to take part in the study.

## Sampling

An exhaustive and consecutive sample of all children aged 0-5 years hospitalised with diarrhoea or suspected diarrhoeal infections seen in paediatric wards was taken.

## Collection, culture and isolation of strains

Stool samples were collected from children aged 0 to 5 in sterile, single-use, labelled bottles containing Cary-Blair agar. The samples were transported to the laboratory of the Centre Hospitalier Universitaire de la Mère et de l'Enfant (CHUME) in compliance with the triple packaging transport conditions. Culture and isolation were carried out by plating the stools on EMB (Methylene Blue Eosin) agar and incubating at 37°C for 18 to 24 hours in a bacteriological oven. After 18 to 24 hours, colonies with a metallic sheen on EMB agar, characteristic of *E. Coli*, were transferred to Muller-Hinton (MH) agar for antigenic studies, Gram staining and the oxidase test.

## Biochemical identification of germs

Suspect *E. coli* colonies (green with a bright metallic sheen) were plated onto Muller Hinton (MH) agar after 24 hours incubation at 37°C. They were subjected to biochemical tests using the method of Poelma *et al.*, (1984). The oxidase-negative colonies were suspected of being *E. coli* strains. These suspected colonies were subjected to biochemical identification tests, namely the indole test for glucose and lactose metabolism and gas production. H<sub>2</sub> S was determined using Kligler Hajna medium. Mannitol mobility medium was used to determine mannitol metabolism and bacterial mobility. Simmons citrate medium was used to demonstrate the ability of the bacteria. Citrate was used as the sole carbon source. Readings were taken after 24 hours incubation at 37°C.

Biochemical identification of the incriminated strains was carried out after culture of the inoculum on the bioMérieux API20E gallery in accordance

with the instructions of the Société Française de Microbiologie (SFM). Revelation was carried out using TDA, JAMES and VP reagents.

## Statistical analysis

All the data collected was analysed using Statistical Package for Social Sciences (SPSS) version 2021. Statistical analysis used the  $\chi^2$  (chi2) test to compare two qualitative variables. Differences were considered significant when  $p < 0.05$ .

## Results and Discussion

### Sociodemographic characteristics of the study population Prevalence of *E. coli*

A total of 296 stools from children aged 0-5 years were received during the study period, from which 36 strains of *E.coli* were isolated, representing a prevalence of 12.2%.

The prevalence of enteropathogenic *Escherichia coli* isolated from 36 children from three hospitals was 12.2%: CHU-ME 11.3%, NDA9, 9% and HRS: 16%. Kenkoua *et al.*, (2008) at Cameroon found a prevalence of 15.3%.

Other similar studies in Burkina Faso had shown a high prevalence of *E. coli* of 25.3% by Konaté *et al.*, (2018) and 21.4% by Dembélé *et al.*, (2015). Other previous studies carried out in Burkina Faso reported a prevalence of 8% by Bonkougou *et al.*, (2013), in Senegal, (10%) by Fall-Niang *et al.*, (2013).

In the equatorial zone, a prevalence of 57.7% and 22% were respectively reported in China by O'Ryan *et al.*, (2005) and Chen *et al.*, (2014). In Chad, similar work carried out at the Hôpital Général de Référence Nationale (HGRN) in 2013 by Bessimbaye *et al.*, (2013) reported a prevalence of 34.73% of all enteropathogens.

These various prevalences testify to the geographical disparity of diarrhoeal diseases. The

results obtained in our study, with prevalences ranging from 9.9% to 16% in the three hospitals, could be explained by the increased awareness of hand hygiene observed during the SARS-COV-2 period. It would be important to continue observing barrier measures and hand and environmental hygiene.

Male children were more infected with a rate of 58.33% compared with 41.67% among female children. These results are comparable to those of Konaté *et al.*, (2018), who obtained a prevalence of 58.1% in male children, compared with 41.9% in female children. These results show that both boys and girls are susceptible to diarrhoeal diseases. The risk of incidence depends on the lifestyle and hygiene conditions in the area where the study was carried out, with regard to the pathogen responsible for diarrhoea in children.

Children in the 12-59 months age group were more infected at 86.12%, while children aged 0- 11 months had an infection rate of 14%. The results obtained are similar to those of other studies across Africa, which reported high infection rates in the 12 months and over age group: Coulybaly *et al.*, (1998) in Côte d'Ivoire and Luky *et al.*, (1998) in the DRC, with infection rates of 51% and 46% respectively.

It should be emphasised that at this age, the incidence of diarrhoea could also be the result of cross-contamination following the introduction of contaminated food and water into children's diets (Dillon, 2003), or of the household diet and the hygienic behaviour of parents. It has been observed that children aged between 12 and 59 months are particularly vulnerable.

It is at this age that children start to learn to walk, putting everything they find on the ground in their mouths and coming into contact with domestic animals. Pet faeces containing pathogenic bacteria could be responsible for contaminating children. The risk of diarrhoea occurring is very high when hygiene precautions are not taken into account when caring for the child (Djourdébbé *et al.*, 2015). The

persistence of these diarrhoeal diseases is often associated with the low level of hygiene observed by populations. This situation, favoured by poor nutrition, was reported by Barro *et al.*, in Burkina Faso (2005). The results of the present study show that age is a very important factor in the epidemiological surveillance of diseases, particularly childhood diarrhoeal diseases.

After serotyping 36 strains of enteropathogenic *E. coli*, eight (8) serogroups were identified: O111, O26, O86, O125, O126, O124, O114, O142. Serotype O124 was the most prevalent in the three hospitals in Chad, with a rate of 25.0%. These results confirm the data of Konaté *et al.*, (2018) and Fall-Niang *et al.*, (2013) who reported that serotypes O124 were the most circulating in Burkina Faso and Senegal.

During the present study, the July-August period was associated with a high prevalence of *E. coli* (58% in July and 28% in August.) There is usually a seasonal variation in the number of cases of diarrhoeal disease (Dembélé *et al.*, 2016).

Moreover, the high prevalence of diarrhoea cases during the June-August rainy season has also been observed in Burkina Faso by Bonkoukou *et al.*, (2013). However, Konaté *et al.*, (2018) obtained a high prevalence of *E. coli* in September-October. This difference could be explained by the climatic variations observed in recent years.

The results of this study show that most *E. coli* strains were isolated from children whose drinking water source was boreholes, with a rate of 63.90%. However, 27.80% of strains were isolated from households where the source of drinking water was the Société Tchadienne d'Eau (STE).

The results obtained are similar to those reported by Allhabo *et al.*, (2016) in Chad on the quality of water from manual boreholes in the peri-urban area of the city of N'Djamena, which confirmed that most borehole water was heavily contaminated with 44% *E. coli* and 84% *Enterobacteriaceae*.

**Table.1** Distribution of *E. coli* strains by hospitals

Hospital	Negative <i>E. coli</i>	Pathogenic <i>E. coli</i>
	N (%)	N(%)
<b>CHU-ME</b>	133(88,7)	17(11,3)
<b>HNDA</b>	64 (90,1)	07(9,9)
<b>HRS</b>	63 (84)	12 (16)
<b>Total</b>	260 (87,8)	36 (12,2%)

CHU-ME-University Hospital Center for Mother and Child; HNDA Hospital of Our Lady of the Apostles; HNDA; HRS- Sarh Regional Hospital

The prevalence of *E. coli* was 11.3% at CHU-ME, 9.9% at HNDA and 16% at HRS

**Table.2** Prevalence of the different serotypes identified.

Serotypes	N	%
<b>O111</b>	3	8.3
<b>O26</b>	3	8.3
<b>O86</b>	3	8.3
<b>O125</b>	3	8.3
<b>O126</b>	5	13.9
<b>O124</b>	9	25.0
<b>O114</b>	5	13.9
<b>O142</b>	5	13.9
<b>Total</b>	36	100%

Serotype 0124 represented 25% followed by serotype 0126, 0114, and 0142 with 13.9%.

**Table.3** Prevalence of *E. coli* strains isolated by month.

Month	N	%
<b>July</b>	21	58.3
<b>August</b>	10	27.8
<b>September</b>	04	11.1
<b>October</b>	1	2.8
<b>Total</b>	36	100

Pathogenic *E. coli* strains during the month of July represented 58.3% followed by the month of August with 27.8%.

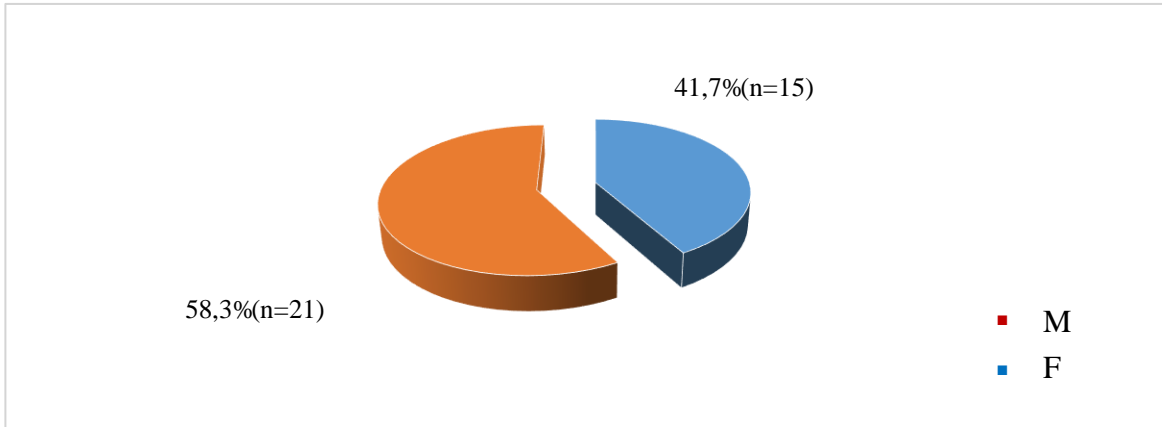
**Table.4** Prevalence of strains according to drinking water.

Water Source	N	%
<b>Forage</b>	23	63.9
<b>Mineral</b>	1	2.8
<b>River</b>	2	5.5
<b>STE</b>	10	27.8
<b>TOTAL</b>	36	100

STE = Chadian Water Company

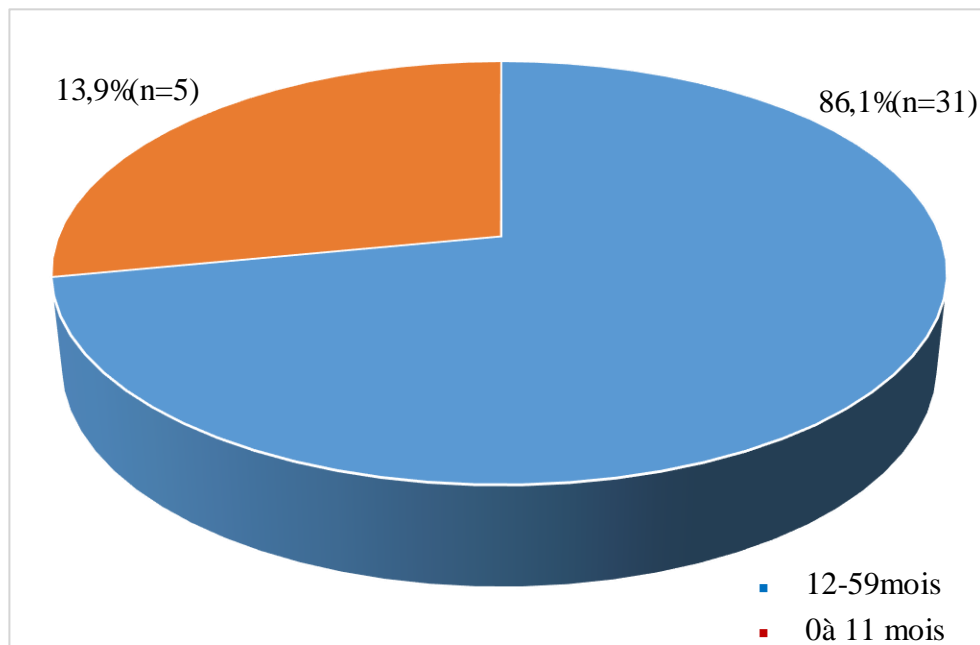
*E. coli* strains were isolated in households where children used as a source of drinking water from boreholes 63.9%, followed by water from the STE with 25.8%.

**Fig.1** Prevalence of *E. coli* strains according to sex



Boys were more infected than girls with a rate of 58.33%.

**Fig.2** Prevalence of strains according to age group Children aged 12-59 months accounted for 86.1%.



In a study carried out in Chad on the bacteriological quality of well, borehole and river water consumed in the Doba oil basin, Maoudoumbaye *et al.*, (2016) found 74 strains of *E. coli* in borehole water, 137 in well water and 36 in river water. Bengaly *et al.*, (2016) found that the bacteriological quality of drinking water in the district of Bamako was unsatisfactory, with the majority of samples having levels of total and/or faecal coliforms above the Malian standard. It is therefore important and urgent

to carry out studies on the quality of drinking water and to run awareness campaigns on drilling techniques and their treatment in order to avoid the proliferation of pathogenic strains in drinking water, which would be a public health problem.

Pathogenic bacteria, particularly enteropathogenic *E. coli*, are a major cause of paediatric diarrhoea in developing countries. Epidemiological surveillance of diarrhoeal diseases necessarily involves proper

identification of the bacteria involved. The prevalence of *E. coli* infections during the present study was high in the health facilities studied. Drinking water is contaminated at different levels. Age, hygiene and diet were factors linked to infection. It seems important and urgent to broaden the scope of research into these enteropathogens in the food and clinical fields, in order to gain a better understanding of the origin and mode of circulation of these fearsome strains.

### Conflicts of Interest

The authors declare that there are no conflicts of interest in this work.

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