



Original Research Article

Bacteria agents of Diarrhoea in Children Aged 0-5 Years, in Minna, Niger State, Nigeria

M.Galadima and O.O.Kolo*

Department of Microbiology, Federal University of Technology Minna, Niger State

*Corresponding author

ABSTRACT

Keywords

Diarrhoea,
Dehydration,
Children,
Antibiotics
and Bacteria

Stool samples from children with diarrhoea attending the General Hospital Minna, Nigeria were analysed for the presence of different types of bacteria using standard bacteriological methods. Isolates were subjected to antimicrobial susceptibility using the disc diffusion method. The bacteria identified were as follows: *Escherichia coli* 84 (47.734%), *Shigella* species 34(19.32%), *Salmonella* species 29 (16.6532%). Others were *Citrobacter* species 8 (4.55%), *Enterobacter* species 11 (6.65%), *Vibro cholerae* 4 (2.23%). Ten (5.68%) samples yielded no bacteria growth. The highest number of isolates, (51%), belonged to those in the 0157 serogroup of *E.coli* 75% of *Salmonella* species belonged to *Salmonella* paratyphi A; 19% were positive for *Salmonella* paratyphi B and 8% for *Salmonella* typhi. 66% of the examined samples yielded *Shigella dysenteriae* while 30% were positive for *Shigella flexneri*, This study has shown that *Escherichia coli* accounts for most cases of infantile diarrhoea in Minna, Nigeria. All isolates were resistant to chloramphenicol (30ug) and streptomycin (30ug) and were highly sensitive to amoxicillin and ciprofloxacin.

Introduction

Diarrhoea is the most common cause of gastroenteritis both in children and adult. It is a condition of having three or more loose or liquid bowel movement per day (WHO, 2008). It is a common cause of infant deaths in developing countries, especially where safe drinking water and adequate sanitation hygiene is unavailable and the second most common cause of deaths worldwide. Diarrhoea is preventable and treatable. Diarrhoea which is usually due to bacterial, viral, or

parasitic infection is a common problem that usually lasts 1 or 2 days and resolves on its own without special treatment (Chen *et al.*, 2010). The loss of fluids through diarrhoea can cause electrolyte imbalances and dehydration. Dehydration alters the child's natural balance of water and electrolytes (sodium, potassium, chloride) and can be serious if not treated promptly. Diarrhoea is not a disease, but is a symptom of a number of illnesses (Kandakai-Oluyemi, *et al.*, 2009). Reports

in the literature indicate that in 2009 diarrhoea was estimated to have caused 1.1 million deaths in people aged 5 years and over and 1.5 million deaths in children under the age of 5 years (Odukoya, 1998). Studies have shown that bacterial agents are important causes of infantile diarrhoea in many developing countries. Prolonged diarrhoea persisting for more than 2 days may be a sign of a more serious problem and poses the risk of dehydration (Katribe *et al.*, 2008). It has been demonstrated in previous studies that diarrhoea infection is more severe in younger children and incidence of occurrence is highest in the six month period following child birth. This study was undertaken to identify the species of bacteria that cause diarrhoea in children in Minna Nigeria, and their antibiotic susceptibility.

Materials and Methods

Study Base

The study was based at General Hospital, Minna.

Study Population

The study population included infants and young children between 0 – 5 years attending the Paediatric unit and Out Patient Department (OPD) of the General Hospital in Minna, Nigeria. Informed consent was obtained from patients' mothers, hospital authorities, laboratory technician and Clinicians involved in the management of the patients.

Sample Collection

Stool samples were collected from the patients in clean, clear, transparent, wide mouthed bottles. The population size was determined by standard statistical formula with prevalence rate of 9%, Confidence

level at 95% (standard value of 1.96) and Margin of error at 5% (Standard value of 0.05)

Study Procedure

The specimens were processed according to the guidelines provided by Cheesbrough (2009) for the laboratory diagnosis of enteric pathogens. These include: macroscopy, microscopy, culture, gram stain, motility testing, and biochemical tests. The stool samples were examined macroscopically and the appearances, consistency and colour were recorded.

A loopful of liquid stool or fecal suspension was enriched in selenite F broth for 24hrs at 37°C and then subcultured on MacConkey agar (MAC), Salmonella Shigella agar (SSA) and Sorbitol MacConkey Agar. The plates were incubated at 37°C for 24hrs. The resultant isolates were purified for further tests.

Identification of the isolates.

The isolates were grouped into lactose fermenting and non lactose fermenting colonies which were then characterised based on the following standard biochemical tests by (Cheesbrough, 2009). Citrate Utilisation Test, Indole Test, hydrogen sulphide production and gas production test (using triple sugar iron agar), Motility test, Urease test, Carbohydrate fermentation test, Voges-proskauer test, lysine decarboxylase test. The identified colonies were grouped using of *Salmonella paratyphi* group A and B polyvalent antiserum and *Shigella dysenteriae* and *Shigella flexneri*.

Polyvalent antisera. The sensitivity of the isolates to antimicrobial agents was determined on nutrient agar plate using disc diffusion method and interpreted

based on diameter of zone of inhibition (mm). All isolates were tested for sensitivity to the following antibiotics: Septrin (30ug), sparfloxacin (25ug), amoxicillin (25ug), augmentin (30ug), gentamycin (25ug), pefloxacin (25ug), tarivid (30ug), streptomycin (10ug), and chloramphenicol (30ug).

Results and Discussion

253 isolates were examined, 139 (54.94%) were identified as *Escherichia coli*, 49(19.37%) *Shigella* species, 42 (16.60%) *Salmonella* species, and 8 (3.16%) *Citrobacter* species. Others were *Enterobacter* species 11 (4.34%), and *Vibro cholerae* 4 (1.58%). 38 (15%) samples did not yield any growth.

Figure 1 shows the frequency of occurrence of enterobacteriaceae in examined samples. The highest number of isolates was observed for *E. coli*, 139(54.94%), There was no bacterial growth in 38 (15%) of the samples which suggested that the cause of the diarrhoea might be due to other causes other than bacteria.

The age group 6 –12 months had the highest percentage prevalence 59 (88%) for *E.coli* and 21 (31.34%) for *Shigella* species, was followed by the 12 – 24 month age group which had 34 (80.04%) *E.coli*. The age group 24-48 months had the prevalence of 75% while those aged 0-6 months had the lowest number of *E.coli* isolates. The difference between these groups was found to be statistically significant ($P>0.05$).

The age group 12-24 months had highest percentage prevalence 18 (42.86%) for *Salmonella* species. The 6 – 12 month age group had prevalence of 18 (26.87%). No isolate was recovered from 0- 6 months age group.

The age group 24-48 months had highest percentage prevalence 3 (12.50%) for *Enterobacter* species. The 6 – 12 month age group which produced 5 (7.46%) isolates, and no isolate was recovered from 0- 6 months age group.

The age group 6-12 months had the highest prevalence percentage 5 (11.90%) for *Citrobacter* species with the female having slightly higher percentage than the male. The 12 – 24 months age group which produced 1 (4.16%) isolates. No isolate was recovered from 0- 6 and 48 -60 months age group.

The age group that had highest percentage 1 (4.76%) of positive isolates of *Vibro cholerae* species was 48 -60 month age group, and was followed by the 6 – 12 month age group which had 2 (2.99%) isolates, and followed by age 12-24 months which had 1(2.38%) of the isolates. No isolate was recovered from 0-6 and 24 -48 months age group.

51% of the isolates were *E. coli* 0157. 66% of the examined samples yielded *Shigella dysenteriae* while 30% for *Shigella flexneri*. 72% of *Salmonella* species isolates were positive for *Salmonella paratyphi* A 19% were *Salmonella paratyphi* B and 8% *Salmonella typhi*. All isolates of *E.coli* were moderately sensitive to septrin, sparfloxacin amoxicillin, augmentin, gentamycin, pefloxacin, and tarivid. About 89% of the isolates were resistant to chloramphenicol and streptomycin and 91% were highly sensitive to amoxicillin and ciprofloxacin. All isolates of *Salmonella* species were resistant to chloramphenicol streptomycin, and ciprofloxacin. 70% were highly sensitive to septrin, sparfloxacin, augmentin, gentamycin, and pefloxacin. 86% of *Salmonella* isolates are moderately

sensitive to amoxicillin and tarivid. All *Shigella species* isolates were moderately sensitive to sprafloxacin, chloramphenicol, gentamycin, pefloxacin, and tarivid, 56% were highly sensitive to amoxicillin and ciprofloxacin, and 45% were resistant to septrin and streptomycin.

The prevalence rate of *E.coli* was 2.5 times higher than that of *Shigella species* which is the major bacteriological causes of diarrhoea in children below five years of age. This is higher than the 26% prevalence rate document by Olanipekun (1996) for children with diarrhoea attending the Jos University Teaching Hospital in Jos, Nigeria and 15% by kandakai- Oluyemi for children attending the Abuja national hospital but similar to report by Rotimi *et al.*(1994) for children attending Obafemi Awolowo University Teaching Hospital in Ile-ife and several other international authors who reported that the prevalence rate of *E.coli* is 2.5 higher than that of *Shigella* which are the major bacteriological causes of diarrhoea in children below two years (King *et al.*, 2003, Longstreth *et al.*, 2006).

The age prevalence is similar to reports published by Olanipekun (1996) and Rotimi *et al.*, (1994) and several authors (King *et al.*, 2003, Longstreth *et al.*, 2006) who observed that the highest incidence of gastroenteritis in children was found within the age range of 7 – 12 months. During this period, children are weaned and start moving around putting few contaminated toys and unguided things into their mouths. Figure 5 shows the distribution of *Enterobacter* species in relation to the age of the children examined. The highest isolation rates were found in children between the ages of 24 - 48 months followed by age group 0-6 months.

Figure 5 shows the distribution of *Citrobacter* species in relation to the age of the children examined. The highest isolation rates were found in children between the ages of 12 -24 months followed by age group 24-48 months. Figure 6 shows distribution of *Vibro cholerae* in relation to the age of the children examined. The highest isolation rates were found in children between the ages of 48- 60 months followed by age group 6- 12 months. Most cases of diarrhoea are not sex specific. Females had a higher occurrence in some cases while in some males had higher percentage prevalence.

The low isolation rate of *E.coli* in children older than 12 months may be associated with the development of immunity or the loss of receptors for some specific adhesion molecules.

The highest number of *E.coli* isolated belonged to the 0157 H7 sero-group. 0157 sero-group has been recognized by the World Health Organization (WHO, 2008) to be one of the major causes of diarrhoea. However, due to the unavailability of the monovalent typing sera, further specific typing was not carried out to show which serotype occurs more frequently in infantile diarrhoea within the study's geographic area.

The antimicrobial susceptibility profile shows that high rates of resistance were recorded for Streptomycin, Septrin and Chloramphenicol . Several reports have indicated that these drugs are also less effective against other bacterial agents isolated, largely because they are inexpensive and can be obtained easily without a doctor's prescription (Chatkaeomorakot *et al.*, 1987).

This could result in plasmid mediated antibiotic resistance found to be common in *Escherichia coli*. All isolates were highly sensitive to ciprofloxacin, amoxicillin, and pefloxacin. These antimicrobial agents are expensive, cannot be easily purchased without prescription. These are few of the reasons why these drugs are highly effective in treating these infections. Fortunately, diarrhoea is

usually self-limited and rehydration is the most effective treatment.

The use of antibiotics in general is of minor importance and has been criticized on the grounds of drug toxicity and the risk of increased wide-spread antimicrobial resistance.

Figure.1 Prevalence of enteropathogenic bacteria in children

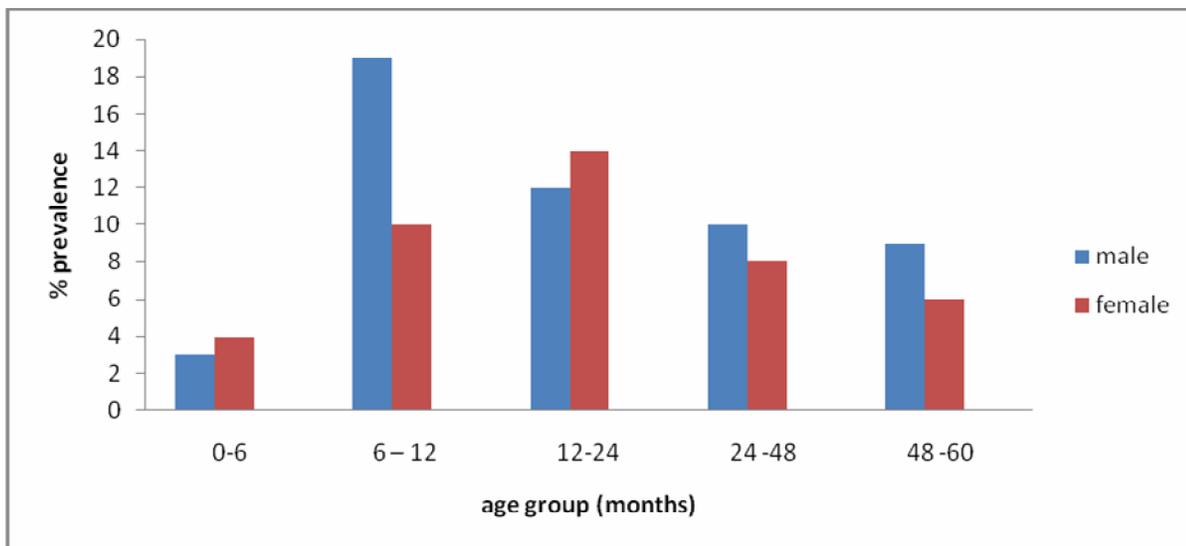


Figure.2 Prevalence of *E.coli* in relation to age and sex of the children

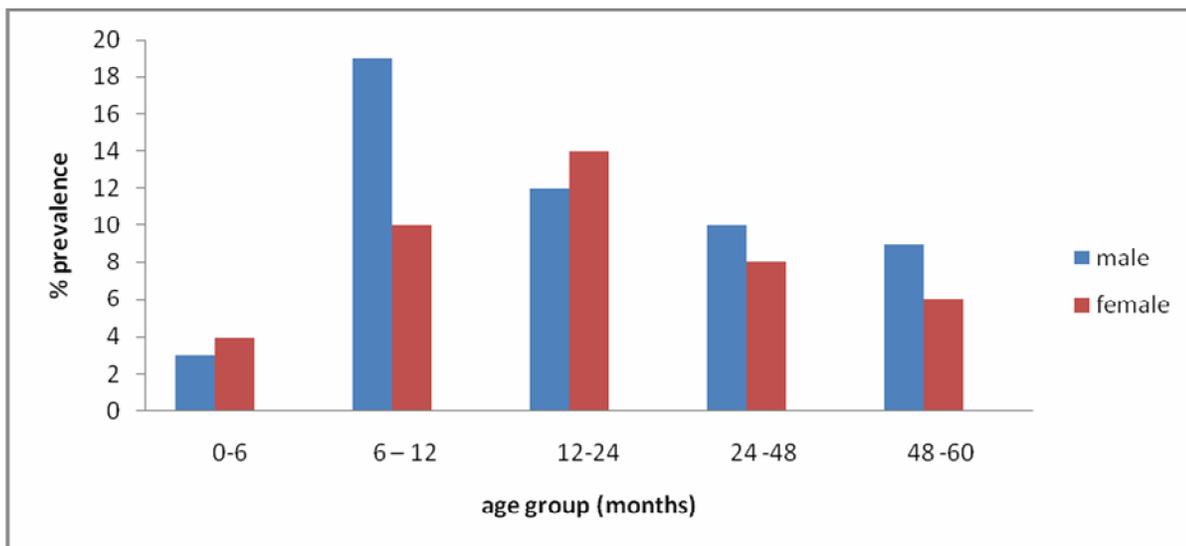


Figure.3 Prevalence of *Shigella* species in relation to age and sex of the children

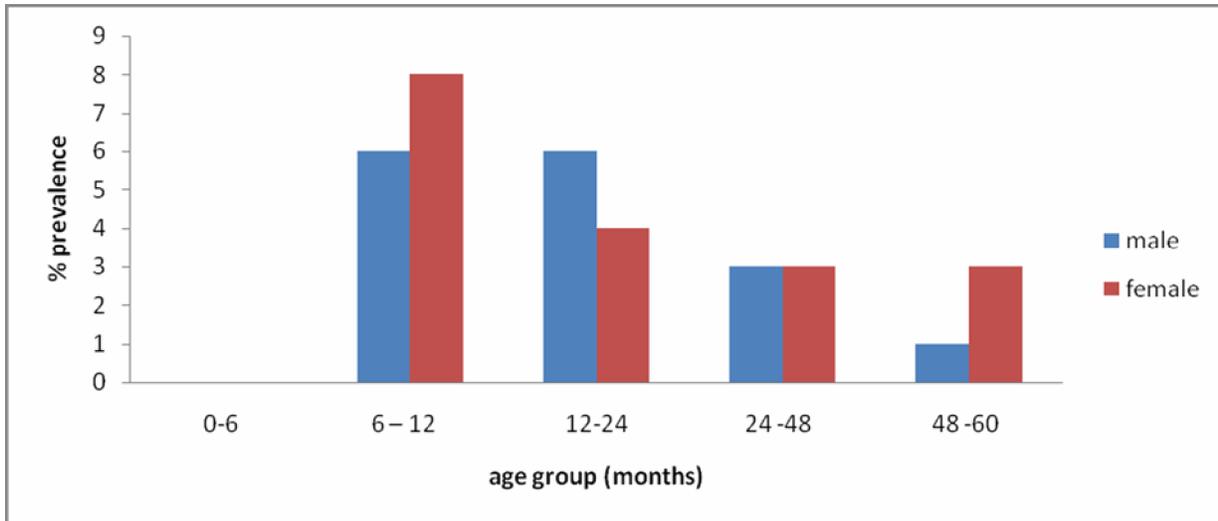
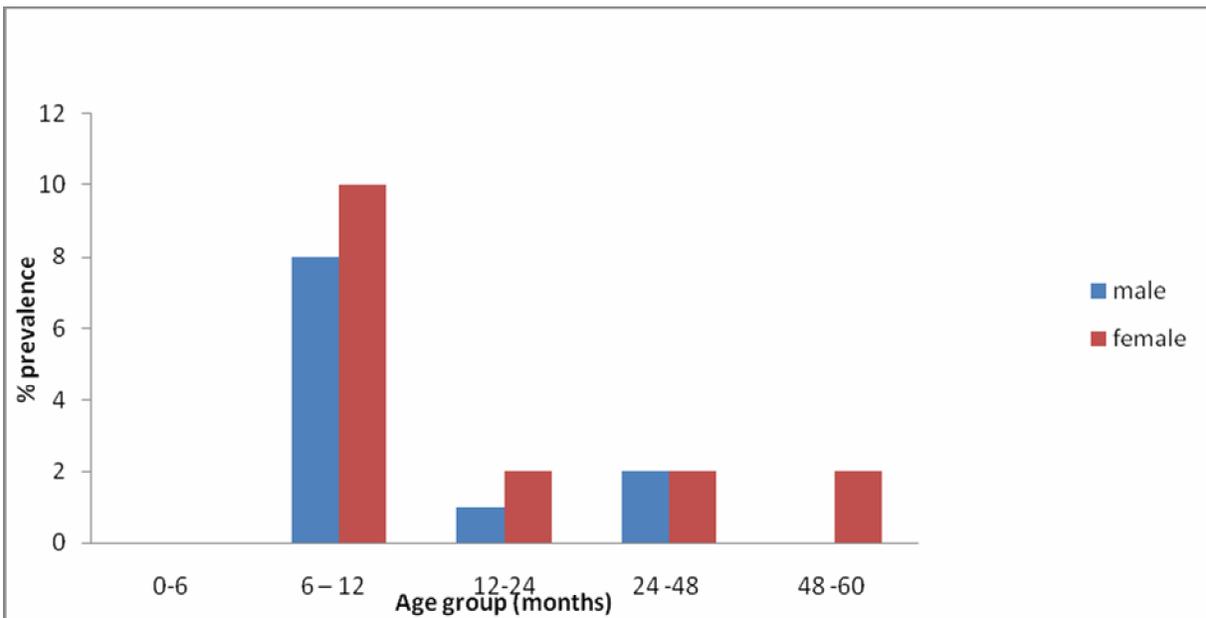


Figure.4 Prevalence of *Salmonella* species in relation to age and sex of the children



This study has shown that *Escherichia coli* 0157: H7 is the commonest cause of infantile diarrhoea in Minna, Nigeria, and that the species isolated were moderately sensitive to septrin, sprafloxacin, amoxicillin, augmentin, gentamycin, pefloxacin, and tarivid. All isolates were

resistant to chloramphenicol and streptomycin but were highly sensitive to amoxicillin and ciprofloxacin. Therefore, amoxicillin and ciprofloxacin should be used in the treatment of *E.coli* infections in the study area.

References

- Chatkaeomorakot, A., Echeverria, P., Taylor, D.N. (1987). Hela cell – adherent *E. coli* in children with diarrhoea in Thailand. *Journal of Infectious Diseases*; 160: 243–247.
- Cheesbrough, M. (2009). District Laboratory practice in Tropical countries Part 2. Cambridge University Press, Cambridge: 178–180.
- Chen, C.Y., Chen, W.C., Chin, S.C., Lai, Y.H., Tung, K.C., Chiou, C.S., Hsu, Y.M., Chang, C.C. (2010). Prevalence and antimicrobial susceptibility of salmonellae isolates from reptiles in Taiwan. 44-50
- Kandakai-Olukemi, Y.T., Mawak, J.D., Ochai I.J., Olukemi, M.A. (2009). Isolation of Enteropathogenic *Escherichia coli* from Children with Diarrhoea Attending the National Hospital in Abuja, Nigeria. *Shiraz E-Medical Journal*; 10(3): 99-106
- Katiribe, E., Bogomolnaya, L. M., Wingert, H., Andrews-Polymenis, H. (2008). Subspecies IIIa and IIIb *Salmonellae* Are Defective for Colonization of Murine Models of Salmonellosis Compared to *Salmonella enterica* subsp. I Serovar Typhimurium. *Journal of Bacteriology*. 191: 2843-2850
- King, C.K., Glass, R., Bresee, J.S., Duggan, C. (2003). "Managing acute gastroenteritis among children: oral rehydration, maintenance, and nutritional therapy". *MMWR Recommendation Rep* 52 (RR-16): 1–16. PMID 14627948.
- King, C.K., Glass, R., Bresee, J.S., Duggan, C. (2003). Managing acute gastroenteritis among children: oral rehydration, maintenance, and nutritional therapy. *MMWR Recomm Rep*; 52:1.
- Longstreth, G.F., Thompson, W.G., Chey, W.D., Houghton, L.A., Mearin, F., Spiller, R.C. (2006). "Functional bowel disorders". *Gastroenterology* 130 (5): 1480–91.
- Mahmood, D.A .and Feachern, R.G. (1987). Clinical and epidemiological characteristics of Rota virus and EPEC – associated hospital infantile diarrhoea in Basrah, Iraq. *Journal of Tropical Paediatrics* ; 33: 319-322.
- Odukoya, D.K. and Olasupo, N.A. (1998). Drug resistance and plasmid profile of diarrhoeagenic bacteria isolated in Nigeria. *Journal of Hospital Medicine*; 7(1): 29-31.
- Olanipekun, O.O. (1996). Prevalence of Enteropathogenic *Escherichia coli* in children with diarrhoea attending Jos University Teaching Hospital (MSc Thesis). Jos: University of Jos.
- Viswanatha, V.K. Hodges, K. and Hecht, G. (2009). "Enteric infection meets intestinal function: how bacterial pathogens cause diarrhoea". *Nature Reviews. Microbiology* 7 (2): 110–9.
- World Health Organization (2009). "Diarrhoea" <http://www.who.int/topics/diarrhoea/en/>. World Health Organization.