Factors Associated with Diarrheagenic Escherichia Coli Infection among Children with Diarrhea in Kwali, Federal Capital Territory, Nigeria

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Abstract

Background: *Diarrhea caused by diarrheagenic Escherichia coli is a leading cause of illness among children in resource-poor communities. Several non-specific factors facilitate the continuous transmission of diarrheagenic E. coli infection. This study determined the factors associated with the disease among children residing in Kwali, a resource–poor community in the Federal Capital Territory Nigeria.*

Methods: *This study was a hospital–based unmatched 1:2 case–control study conducted among children with diarrhea (cases) and children without diarrhea (controls) aged between 0–15 years. Data on demographic, environmental and behavioral factors were collected using a structured questionnaire. Data was analyzed using STATA 15 and logistic regression was done to determine the factors that were associated with the disease at 0.05 significance level.*

Results: *Fifty nine cases of diarrhea and 118 controls were selected for this study. The adjusted multivariable logistic regression model showed that diarrheagenic E. coli infection was more likely among children who lived inside overcrowded rooms (aOR =2.58,95% CI : 0.77 – 8.56) and children with poor hand washing habits (aOR = 6.44, 95% CI: 1.87 – 22.02). Drinking stream water (aOR = 3.23, 95% CI: 1.07 – 9.69) and young age (aOR = 5.91, 95% CI: 1.98 – 17.68) were also associated with diarrheagenic E. coli infection.*

Conclusion*: Overcrowding, poor personal hygiene, young age and lack of access to improved water and sanitation facilities are some of the factors associated with the transmission and spread of diarrheagenic E. coli infection in Kwali.*

KEYWORDS: *Children, case-control study, diarrheagenic E.coli, factors, Kwali*

Introduction

Escherichia coli are the predominant facultative anaerobe commensals and micro biota in the mammalian gastrointestinal tract, yet some strains can cause severe diarrhea in humans.[1] Gastrointestinal colonization occurs within hours of life and the bacteria

will remain in a symbiotic relationship with the host.[2] Also, the pathogen remains harmlessly confined to the intestinal lumen until when gastrointestinal barriers are violated or when the host becomes immune-suppressed. Even non-pathogenic strains of *E. coli* can cause infection when immune suppression sets in.[2]

Diarrheagenic *E. coli* (DEC) are strains of *E. coli* that are pathogenic and cause diarrhea diseases.[3,4] Diarrhea caused by DEC is a leading cause of illness among children below the age of 15 years in resource – poor communities and it is transmitted through contaminated water and food especially in areas of low socio – economic status and sanitation challenges.[5,6,7] In Nigeria, more than a third of the hospital beds for children are occupied by patients with diarrhea.[8] Children are the most affected with diarrheagenic *Escherichia coli* infection in the Federal Capital Territory Nigeria.[9]

Despite several intervention strategies by the government, resource – poor communities continue to report cases of DEC infections in Nigeria which may be due to several non – specific factors that facilitates the continuous transmission of the disease such as demographic, environmental and behavioral factors.[5,9,10,11] The lack of access to prompt diagnostic tools is a major challenge in the diagnosis of DEC infection in developing countries.[10,12] A previous study showed high occurrence of diarrheal disease caused by DEC in patients from resource – poor communities within the Federal Capital Territory.[9]

There is also evidence that diarrheal disease can be caused by multiple causative agents including *Shigella Spp, Compylobacter jejuni, Salmonella Spp* and in some cases Vibrocholerae 01.[13,14,15,16] This calls for more research in understanding the epidemiology of the disease. There is currently no vaccine for DEC infection and the success of the control measures is in understanding the epidemiology of the disease, particularly the risk factors. This study assessed the factors associated with DEC infection among children with diarrhea in Kwali area council, a resource–poor community of the Federal Capital Territory, Nigeria.

Methodology

Study area

The Federal Capital Territory consists of six area councils among which Kwali area council has several resource – poor communities spread across the council.[17] These communities lack access to improved sanitary conditions, clean source of drinking water, proper waste disposal facilities and there is wide spread practice of open defecation amongst residents. A significant proportion of residents (about 25%) lack access to potable water and resort to streams as their main source of drinking water and other house hold chores.[17]

Study population

Children between ages 0–15 years who attended the outpatient clinic of General hospital Kwali and resident in Kwali area council.

Case definition

Diarrheal cases included in this study were children whose stool samples were watery, loosed with or without mucus and blood, and that had more than three episodes of stooling in the last twenty – four hours, and who attended the outpatient clinic of the hospital within the study period.

Confirmed cases

All DEC cases were determined after culture of diarrhea stool samples and biochemical identification of *E.coli* isolates was carried out as described by

Cheesbrough, 2006[18] (Table 1)
DNA of *E.coli* isolates were extracted and subjected to a multiplex polymerase chain reaction assay using specific primers for the detection of diarrheagenic *E.coli* virulence genes and the amplified genes were detected using agarose gel electrophoresis and the DNA bands were visualized and photographed under ultra – violet light.[4,19](Fig. 1)

Controls

Children between ages 0 – 15 years who did not have diarrhea, who attended the outpatient clinic of the hospital for other health consultations within the study period.

Study design

This study was a hospital–based unmatched 1:2 case control study conducted in Kwali area council of the Federal Capital Territory, Nigeria. The cases and controls were selected from children who attended the outpatient clinic of General hospital Kwali during the sample collection period. The cases were children who had diarrhea while the controls were selected from children who did not have diarrhea. The selection of control was done through balloting; all eligible controls were included in the balloting process. The number of controls subsequently selected was based on the number of cases identified in the outpatient clinic using a case to control ratio of 1:2. A questionnaire was then administered to both cases and controls to determine those who were exposed to cases of diarrhea in the one year preceding the study.

Sample size determination and data collection

The total sample size for the study was 177 (59 cases and 118 controls) calculated assuming a power of 80%.[20]At an alpha level of 0.05 using open Epi version 3.[21]An adapted structured questionnaire was used to collect data which captured the demographic, environmental and behavioral factors associated with transmission of diarrhea.[10]

Sampling technique

Medical laboratory officers of the hospital and the principal investigator identified cases from stool samples using the case definition. Controls were selected from the same outpatient clinic among children who did not have diarrhea. The selection was done through balloting, the number of controls selected was based on the number of cases identified using a case to control ratio of 1:2. The questionnaire was then administered to all the respondents (both cases and controls) to determine the history of exposure to cases of diarrhea in the past.

Data analysis

All data was analyzed using STATA 15 statistical software (StataCorp. 4905 Lake way drive station, Texas 77845 USA). Logistic regression was done to determine the factors associated with DEC infection. This was presented as crude odds ratios with a 95% confidence interval. The final age and gender adjusted multivariable logistic regression model fitted to determine the factors associated with DEC was done using backward stepwise approach. Statistical analysis was done at a 95% confidence level with P values < 0.05 considered statistically significant.

Ethical clearance

Ethical approval was obtained with reference number FHREC/20/1701/106 from the Health Research Ethics Committee of the Human and Health Services Secretariat of the Federal Capital Territory Administration.

Results

Demographic characteristics of patients

One hundred and seventy seven stool samples were collected, comprising of 59 cases and 118 controls. The age range of the children was between 0 and 15 years. Cases were younger than controls. The median age of cases was 8 years with an interquartile range between 1 to 13 years. The median age of controls was 9 years with an interquartile range of between 2 and 15 years. There was however no statistically significant difference between the ages of males and females (P >0.05). Majority of diarrhea cases were < 5years (Table 2).

Behavioral factors associated with DEC transmission

Open defecation practices (cOR = 3.08, 95% *CI*: 1.38 – 6.95) and drinking of non-potable water (cOR = 2.55, 95% *CI*: 1.34 – 4.78) were associated with DEC infection. A previous contact with someone who had diarrhea was associated with DEC infection as DEC cases had higher odds of previous contact with diarrhea patient (cOR = 2.68, 95% *CI:* 1.40 – 5.09). Comparatively, DEC cases were less likely to eat cooked food regularly (cOR = 2.74, 95% *CI*: 1.13 – 6.57). Good hand washing practices was less frequent among DEC cases compared to controls, {(cOR = 4.96, 95% *CI:* 2.40 – 10.25) and (cOR = 1.44, 95% *CI*: 1.63 – 10.20) respectively}. A greater proportion of cases compared to controls reported that they did not wash their hands regularly when required (Table 3).

Demographic factors associated with DEC transmission

There was a statistically significant association between gender and DEC infection. DEC cases were more likely to be females (cOR = 2.68, 95%*CI*: 1.36 – 5.24). Age (<5 years) was more likely to have DEC infection (cOR = 5.88, 95%*CI*: 1.95 – 17.65) compared with other age groups. There was also a statistically significant association between previous treatment for diarrhea and DEC infection (cOR = 5.25, 95%*CI*: 1.17 – 6.45) (Table 3).

Environmental factors associated with DEC transmission

DEC cases had higher odds of staying in a compound house compared to controls (cOR = 4.89, 95% *CI:* 1.51 – 5.48). Cases had a threefold higher odds of sharing a sleeping room with more than four persons compared to controls (cOR = 3.99, 95% *CI*: 1.69 – 6.39) or sharing a house with more than 8 persons (cOR = 6.76, 95% *CI*: 2.22 – 20.48) or without proper toilet facility (cOR = 4.70, 95%*CI*: 1.23 – 6.14)(Table 3).

The factors associated with DEC from the adjusted analysis were residence in a compound house (aOR = 25.42, 95% *Cl*: 6.13 – 105.07), age (aOR =5.91, 95% *Cl*: 1.98 – 17.68), female gender (aOR = 4.13, 95% *Cl*: 1.27 – 13.34), sharing a room with more than four persons (aOR = 2.58, 95% *CI*: 0.77 – 8.56) and poor hand washing habits (aOR = 6.44, 95% *CI*: 1.87 – 22.02) (Table 4).

Table 1: Culture and biochemical characteristics of *E.coli*

Culture		Morpohology	Biochemical Tests							Isolate
Mac Agar	EMB Agar	Gram Stain	IND*	MR*	VP*	CT*	GLU*	LAC*	SUC*	
Reddish	Greenish	Gram negative								
colonies	Metallic colonies	Short rods	+	+	-	-	+	+	+	*E.coli*

MAC – MacConkey agar, EMB – Eosine Methylene Blue agar, IND – Indole test, MR - Methyl red test, VP – Voges proskauer test, CT - Citrate utilization test, GLU – Glucose utilization test, LAC – Lactose utilization test, SUC - Sucrose utilization test.

Adapted from Cheesbrough,M.2006

Table 2: Sex and age distribution of study participants

Parameter	Controls (n=118)	Cases(n=59)	s*P*–value
Sex			
Male	45(38.1)	27(45.8)	0.003*
Female	73(61.9)	32(54.2)	
Age group			
<5 years	65(62.5)	39(37.5)	0.646
5 – 9 years	34(73.9)	12(26.1)	
10 – 15 years	19(70.4)	8(29.6)	
Age (median, IQR)	9(2,15)	8(1,13)	0.067

*P< 0.05 means statistically significant

Table 3: Factors significantly associated with DEC infection

	DEC cases		Controls	
Variables	cOR (95% *CI*)	*P* value	cOR (95%*CI*)	*P* value
Demographic factors				
Sex (female)	2.68(1.35 -5.24)	0.002	2.08(1.10– 6.47)	0.0025
Age(<5years)	5.88(1.95 -17.65)	0.032	2.70(1.38– 5.26)	0.004
Past treatment for diarrhea (yes)	5.25(1.17 -6.45)	0.023	3.46(1.75– 9.58)	0.014
Behavioral factors				
Bath everyday (No)	2.74(1.29 -5.78)	0.006	1.24(1.85 –10.82)	0.001
Frequency of bathing/day(once)	1.96(1.05-3.66)	0.01	3.48(1.47– 8.19)	0.004
Practice open defecation (yes)	3.08(1.38 -6.95)	0.003	1.48(1.15 –10.92)	0.025
Drinking of stream water (yes)	2 .55(1.34- 4.78)	0.001	1.08(1.36 – 5.24)	0.002
Contact with diarrhea patient (yes)	2.68(1.40 - 5.09)	0.001	1.21(2.24 –12.07)	<0.0001
Eating cooked food (not regularly)	2.74(1.13 -6.57)	0.021	1.10(1.40 – 6.97)	0.005
Hand washing (not regularly)	4.96(2.40-10.25)	<0.0001	1.44(1.63 –10.20)	0.001
Environmental factors				
Type of housing (compound)	4.89(1.51 -5.48)	0.001	1.72(1.88 –11.82)	0.001
Sleeping room capacity (>4 person)	3.99(1.69 -6.39)	<0.001	1.35(1.18 –10.47)	<0.0001
House occupancy (>8 person)	6.76(2.22 -20.48)	0.001	1.20(1.55 –24.61)	0.007
Toilet facility (open defecation)	4.70(1.23-6.14)	0.010	1.68(1.35 –10.02)	0.01

cOR crude odds ratio; *CI* confidence interval

Table 4: Multivariate analysis of factors associated with DEC in Kwali

Variables	aOR	95%*CI*	*P*-Value
Age (years)	5.91	1.98 – 17.68	0.002*
Sex (female)	4.13	1.27 – 13.34	0.037*
Hand washing (not regularly)	6.44	1.87 – 22.02	0.003*
Drinking stream water	3.23	1.07 – 9.69	0.033*
Type of toilet facility (open defecation)	3.57	0.97 – 12.97	<0.049*
Eating uncooked food	2.23	1.04 – 8.59	0.024*
Sleeping room capacity(>4 persons)	2.58	0.77 – 8.56	0.015*
Residents in compound houses	25.42	6.13 – 105.07	<0.0001*

*P<0.05 means statistically significant; aOR adjusted odds ratio; *CI* confidence interval

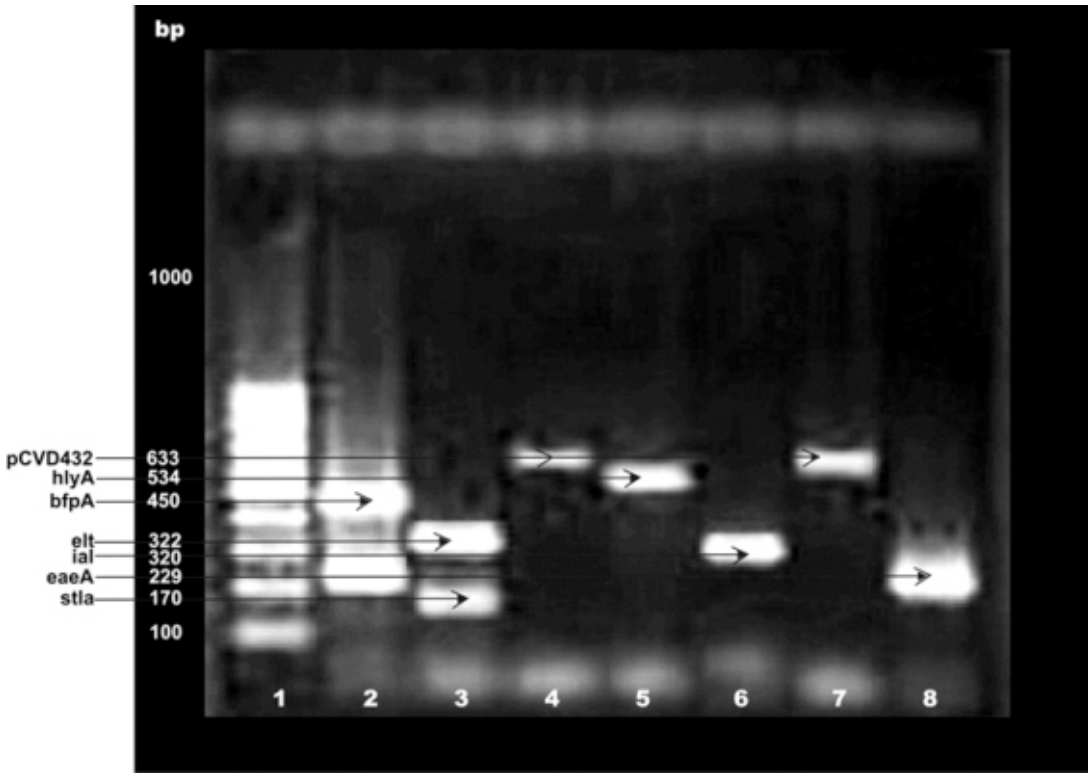

Fig.1: Agarose gel electrophoresis of 1% agarose of PCR product of virulence genes of Diarrheagenic *E. coli*.

Discussion

Lack of access to proper sanitary facilities, poor personal hygiene, overcrowding, drinking non-potable water and regular eating of uncooked food are the major factors that facilitate the transmission of DEC infection in Kwali, Federal Capital Territory Nigeria. This is mainly enhanced by the poor living standards in Kwali. Previous case – control studies conducted in Nigeria showed no significant association between age and DEC infection.[11,22] In this study however, the adjusted analysis showed that age was associated with higher odds of infection. This is similar to other studies which demonstrated that young age was significantly associated with DEC infection.[9,14] This reflects the combined effects of declining levels of maternally

acquired antibodies, the lack of active immunity in infants, the ingestion of contaminated foods and direct contact with human feces.

Previous infection with DEC does not result in long lasting immunity and individuals previously infected carry a significant risk of reinfection.[23]This could possibly be as a result of reactivation of latent DEC in a previously treated individual.[24]This study showed that DEC cases had a threefold higher odds of being previously infected. This confirms the fact that relapses of asymptomatic, infected persons is a major factor that drives the reemergence of DEC particularly in resource – poor communities.[10,25] In this study, DEC infected patients had higher odds of previous exposure to the disease. Transmission of DEC infection occurs through ingestion of contaminated foods and water.[9, 10, 27] This study showed that the risk of infection was higher among patients who practice open defecation and use rooms shared by more than four persons. Lack of access to proper sanitation such as potable drinking water, appropriate waste disposal facilities and toilet facilities enhance the transmission of DEC infection.

This study also observed an increased risk of DEC infection transmission among patient who did not wash their hands regularly. From the data collected, more than half of the patients admitted not washing their hands with soap and water. This shows the inadequacy of behavior change component in water sanitation and hygiene. The prevalence of substandard sanitary and personal hygiene conditions in resource – poor communities also facilitates the persistence of DEC infection in these communities.[27,28]

The observation that individuals living in overcrowded houses are at high risk of contracting communicable diseases is consistent with other studies.[27,29] It is well established that persons living in overcrowded houses belong largely to the low – socio economic groups.[27,28,29] This study has documented the connection between past exposure to diarrheagenic E. coli and subsequent development of diarrhea in children. A limitation of the study, however, is the choice of a case–control study design which carried the risk of a recall bias particularly among the controls. This was mitigated by limiting the period of recall to one year.

Conclusion

Lack of access to improved sanitary facilities, overcrowding and poor personal hygiene are the factors that facilitate the transmission and spread of DEC infection in Kwali, Federal Capital Territory Nigeria. Access to good sanitary facilities, drinking clean and potable water, encouraging good environmental and personal hygiene practices like regular hand washing and improving living conditions should be measures put in place to control DEC infection in Kwali communities. It is therefore important that public health strategies are implemented with deliberate behavioral change efforts.

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