Prevalence of Intestinal Parasites among Children with Diarrhea in Abeokuta, Ogun State, Nigeria

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Abstract: Intestinal parasitic infections have always been an important public health problem in the tropics, particularly in developing countries. In this study intestinal parasites were investigated in 120 diarrheal stool samples collected from children aged under 5years in Abeokuta, Ogun State, Nigeria. Samples were collected from Sacred Heart Hospital, Lantoro, Abeokuta, Ogun State, Nigeria. The intestinal parasites were detected using saline and iodine wet mount preparations. Of the 120 stool samples examined, 31(25.8%) had intestinal parasites. It showed that the prevalence of intestinal parasites was higher in females (26.6%) than males (25.0%). However, this difference is statistically significant (P>0.05). It showed that the prevalence of intestinal parasites was higher among children ages 4 to 5 years old (38.2%) than their counterparts in age group <1 to 3 years (15.4%). There was significant difference (P<0.05) between enteric parasitosis and age. A total of three parasite species were observed (two protozoa and one helminth) from the diarrhea stool samples. The frequency of occurrence of intestinal parasites from the diarrheal stools showed *Ascaris lumbricoides* was the most predominant (54.8%). This was followed by *Entamoeba coli* (25.8%) while *Entamoeba histolytica* was least predominant (19.4%). Mixed infection was not observed. This study indicates that parasites are important cause of diarrhea, especially among children. Though usually not life threatening, chronic parasitic infestation can impair physical and mental growth and general development of children. This study has shown that intestinal parasites are still highly prevalent among children under 5 years of age in Nigeria. The presence of these three parasitic intestinal parasites among children with diarrhea in Abeokuta, Ogun State, Nigeria supports the earlier observations that parasitic infections constitute a major public health problem in the country. The study also provides data for understanding the epidemiological status of the human gastrointestinal parasites in Abeokuta, Ogun State, Nigeria. Therefore, there is need for regular awareness programs on sanitary and good hygiene among children.
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1. INTRODUCTION

Diarrhea is the passage of unusually loose or watery stools, usually at least three times within 24 hour period. It is the consistency of the stools rather than the number that is most important (Sinclair *et al.*, 2003). It is frequent in poor populations (Khan *et al*, 1990) and in immunocompromised individuals (Ferreira and Borges, 2002). Emerging diarrheagenic enteric parasites have been reported (Gianotti, 1990; Hoge *et al*, 1995; Sherchand and Shrestha, 1996; Ono *et al.*, 2001). Diarrhea is produced by a variety of etiological agents, in which intestinal parasitic infection contributes to some extent (Khan *et al*, 1990). Enteric protozoa are associated with traveler's diarrhea (Okhuysen, 2001). In the case of helminth parasites, there is little agreement on which worm definitely causes diarrhea (Genta, 1993).

Gastro-intestinal parasites are identified as a cause of morbidity and mortality throughout the world particularly in the under developed countries (Odu et al., 2011a; Odu et al., 2013). They are one of the most common infections in humans especially in tropical and sub-tropical countries (Awolaju and Morenikeji, 2009; Odu et al., 2011a; Odu et al., 2013). Intestinal parasitic diseases remain a serious public health problem in many developing countries especially due to fecal contamination of water and food (Jimenez-Gonzalez et al., 2009; Odu et al., 2011a; Odu et al., 2013).

Transmission of agents that cause diarrhea are usually by the faecal oral route, which include the ingestion of faecal contaminated water or food, person to person contact and direct contact with infected faeces (Andu *et al.,* 2002). Epidemiological studies of diarrhoea have been reported from several African countries including South Africa (Household *et al.*, 1988), Gabon (Presterl *et al.*, 2003), Egypt (Rao et al., 2003) and Kenya (Sang *et al.*, 1996). It ranks second only to respiratory diseases and is a major cause of morbidity among notifiable diseases in some part of the world (Coker et al., 1998).

The main cause of death from acute diarrhea is dehydration, which result from loss of fluid and electrolyte in stool. Another important cause of death is dysentery and under nutrition. Diarrhea is an important cause of under nutrition because patients eat less during diarrhea and their ability to absorb nutrients is reduced. Moreover, nutrient requirement is increased as a result of infection (Sinclair *et al.*, 2003). Risk factors that predispose children to diarrhea include poor sanitation, poor social and economic status and malnutrition (Andu *et al.,* 2002). Laboratory information is particularly useful to help distinguish invasive enteropathogens (which may require antimicrobial therapy) from non-invasive agents, such as viruses (rotavirus, adenovirus, calicivirus, and astrovirus) and parasites (*Giardia lamblia*, *Entamoeba histolytica* and *Cryptosporium* sp).

Previous studies elsewhere in Nigeria have been along these lines. Among such studies are those of Mordi and Ngwodo (2007), Ajero et al. (2008), Okolie et al. (2008), Tohon et al. (2008), Chukwuma et al. (2009), Awolaju and Morenikeji (2009), Alli et al. (2011a,b,c) and Odu et al. (2010, 2011a,b, 2013). This study was conducted to elucidate the association of parasites with diarrhea among the children in Abeokuta, Ogun State, Nigeria.

2. MATERIAL AND METHODS

2.1. Study Population

The study population was diarrhea patients under 5years attending the Children Emergency Paediatric Unit (CHER) and Out Patient Department (OPD) of the Sacred Heart Hospital Lantoro, Abeokuta, Ogun State, Nigeria. This study was approved by the Ethical committee of the hospital.

2.2. Sample Collection

A total of 120 stool samples were collected from children presenting with diarrhea. The stool samples were collected into a clear, transparent, wide mouthed bottle. The name, age and sex of the patients were properly labeled on the universal bottles containing the samples.

2.3. Processing of Specimens

Stool samples were inspected for the presence of parasitic forms. Saline and Iodine wet mount preparations were made and examined using x10 and x40 objectives (Cheesbrough, 2006).

2.4. Macroscopic Examination of Stool Samples

This describes the appearance of the stool i.e. the physical appearance such as colour, to know whether the stool is formed, semi-formed, unformed or watery, presence of blood/mucus, or pus. When a stool is unformed, containing pus and mucus the possible cause is shigella (shigellosis). When a stool sample is semi-formed and black hookworm disease is suspected. Unformed with blood and mucus stool possible cause is schistosomiasis. There are many appearance of faecal sample: Bloody diarrhea, watering stools, Rice water stools with mucous flakes etc. Blood can also be found in the stools of an individual suffering from haemorrhoids, ulcerative colitis, or tumours of the intestinal tract. A normal stool sample appears brown and formed or semi-formed. While for infants are yellowish – green and semi-formed. In this work the sample analyzed were without mucous or blood. But there are samples that are black and semi-formed, watery stool, and some appeared brown, formed or semi-formed.

2.5. Parasitological Analysis

Among the different parasitological techniques for stool analysis, formol - ether concentration technique as described by Cheesbrough (2006) was employed in this study. The procedure involved emulsifying about one gram (1g) of faeces with an applicator stick in a test tube containing 7ml of formalin solution it was well mixed, 3ml of ether was then added and mixed properly the tube was corked with cotton wool and shook vigorously in an inverted position and the stopper is removed with care. Each sample was made in this same way and the test tubes were balanced in the centrifuge (Model: MINOR 35 from MST Ltd) and centrifuged at 1500 r.p.m for 5 minutes. At the end of centrifugation, the following layer were observed in the test tube: ether at the top (colourless clear liquid); a plug of debris (dark coloured thick); formal solution (a colourful liquid with suspended debris) and a sediment (solid deposit at the bottom of tubes). The plug of debris was then removed from sides of the tube with an applicator stick. The first three layers were decanted down the sediment with a few drops allowed to drain back from the sides of the tube. A cotton swab was used to remove any debris adhering to the sides of the tube. The remaining sediments and the fluid that drained back were mixed properly by flicking the test tube. After which a smear preparation was made using a drop of iodine solution on a slide and the sediment

was added and properly emulsified also on the left side of the slide a smear was made using normal saline covered with a over slip for microscopic examination. The X 10 and X 40 objective was used to examine the whole area under the cover slip for parasite ova, cyst and larvae. Slowly to the other end of the slide, iodine solution decolorized the parasite and making it more visible. In cases were debris were still found in the sample during examination the samples were subjected to the same procedure (formol – ether technique) describe above until it becomes much clearer.

2.6. Identification of Parasites (Ova, Larvae and Adult)

Positive specimens were identified on the basis of microscopy. Using standard methods (CDC, 2007), a trained laboratory scientist at Department of Medical Microbiology, Federal Medical Centre, Abeokuta, Nigeria interpreted the microscopic slides of stool specimens. Several criteria were employed in recognizing the worms: *Ascaris lumbricoides* eggs were recognized on the basis of being round, ova or elliptical with rough membrane (fertilized) or they were a bit elongated and also has rough membrane (unfertilized).

2.7. Enumeration of helminthes eggs

The procedure for counting helminthes eggs in stool sample involves making a wet preparation of the sediment on a clean slide and covering the drop with a cover slip. Starting at one corner of the cover slip, the preparation was systematically examined under a light microscope, using X40 lens moving it back and forth across and noting the number of egg found.

2.8. Data analysis

The prevalence (P), defined as the percentage of infected individuals (NP) among the total number of individuals examined (N) (P= (NP/N) x 100). The helminthes density is the mean number of eggs per gram of stool of each subject. The incidence rate (IN) which is defined as the ratio of the number of new positive samples detected one year after treatment to the number of negative samples obtained before treatment and during the control phases expressed as a percentage (Nkengazong et al., 2009).

3. RESULTS

Of the 120 stool samples examined, 31(25.8%) had intestinal parasites detected. Table 1 shows the prevalence of intestinal parasites in relation to sex. It showed that the prevalence of intestinal parasites was higher in females (26.6%) than males (25.0%). However, this difference is statistically significant (P>0.05).

Table 1: Prevalence of intestinal parasites in relation to sex

Sex	No. tested (%)	No. positive (%)
Females	64(53.3)	17(26.6)
Males	56(46.7)	14(25.0)
Total	**120(100.0)**	**31(25.8)**

Table 2 shows the prevalence of intestinal parasites in relation to age. It showed that the prevalence of intestinal parasites was higher among children ages 4 to 5 years old (38.2%) than their counterparts in age group <1 to 3 years (15.4%). There was significant difference (P<0.05) between enteric parasitosis and age.

Table 2: Prevalence of intestinal parasites in relation to age

Age (years)	No. tested (%)	No. positive (%)
≤1-3	65(54.0)	10(15.4)
4-5	55(46.0)	21(38.2)
Total	**120(100.0)**	**31(25.8)**

A total of three parasite species were observed (two protozoa and one helminth) from the diarrhea stool samples. Table 3 shows the frequency of occurrence of intestinal parasites from the diarrheal stool. *Ascaris lumbricoides* was the most predominant (54.8%) while *Entamoeba histolytica* was least predominant (19.4%). Mixed infection was not observed.

Table 3: Frequency of occurrence of intestinal parasites

Parasites	No. (%)
Entamoeba histolytica	6(19.4)
Entamoeba coli	8(25.8)
Ascaris lumbricoides	17(54.8)
Total	**31(100.0)**

4. DISCUSSION

Morbidity due to intestinal parasites has always been an important public health problem in the tropics, but the incidence and severity may vary depending on the location and period of time (Sethi *et al.,* 2000). In this present study, intestinal parasites were detected in 25.8% of the stool samples examined. Several Indian studies have reported prevalence ranging from 7.5-15.5% in Chandigarh and 16.8% in Delhi (Sethi *et al.*, 2000). The 25.8% overall prevalence of intestinal parasites reported in this study is lower than the 47.1% reported in Andhra Pradesh (Das *et al.*, 1981); the 73.4% in Tamil Nadu (Ganga and Ravichandran, 1995); the 70.8% and 60.0% in Gujarat (Das *et al.*, 1981).

Contrary to our findings, some workers in Nigeria and overseas had earlier on reported higher prevalence rates. Wariso and Ibe (1994) reported 46.0% prevalence rate of intestinal parasite within some parts of Port Harcourt, Nigeria. Egwunyenga *et al.* (2001) reported 33.3% in Nigeria. Alison et al. (2004) reported 17.0% in Uganda. Mordi and Ngwodo (2007) reported a value of 0.7% in all the eighteen local government areas of Edo State, Nigeria. Okolie et al. (2008) reported a prevalence value of 75% among patients with appendicitis in Oguta, Imo State, Nigeria. Chukwuma et al. (2009) in their study on the prevalence of parasitic geohelminth infection of primary school children in Ebenebe Town, Anambra State, reported a prevalence value of 53.6% in soil and 87.7% in stool. Awolaju and Morenikeji (2009) reported a value of 48.4% among primary and post-primary schools children Ilesa West, Osun State and 50.80% among school children in Ilaje, Osun State, Nigeria. Chukwuma et al. (2009) also reported prevalence of geohelminth eggs/larvae in soil with respect to schools to be Umuji primary school 52.5%, Umuogbuefi primary school 83.3% and Obuno primary school 32.5% and overall prevalence in stool samples in the three schools to be 87.7% with distribution as follows; Umuji primary school, 87.5%, Umuogbuefi primary school, 97.5% and Obuno primary school, 75%. Jimenez-Gonzalez et al. (2009) reported a value of 34.0% among inhabitants of a rural community in Mexico. Odu et al. (2011a) reported an overall prevalence of 30.7% among school children in rural and urban communities in Rivers State, Nigeria. Alli et al. (2011a) reported 49.4% in Ibadan, Oyo State, Nigeria. Odu et al. (2013) reported an overall prevalence of 15.7% among primary school children in Rivers State, Nigeria.

The 25.8% prevalence of intestinal parasites from this study is a pointer to the fact that one-quarter of the diarrheal diseases in children in Abeokuta, Nigeria might be associated with enteric parasites. Though females showed a marginally higher prevalence (26.6%) compared with males (25.0%), there was no significant difference ($p>0.05$) between the enteric parasitosis and sex. This suggested that parasitic diarrheal diseases were independent of sex in Abeokuta, Nigeria. This is comparable to what has been reported previously by other workers. Ono et al. (2001) also reported no significant difference in the prevalence of diarrheal diseases in the two sexes caused by parasites. Saathof et al. (2004) in KwaZulu-Natal/South Africa and Tohon et al. (2008) in Nigeria also claimed that parasitic infections were not sex dependent. It agrees favourably with Awolaju and Morenikeji (2009) who reported no significant among primary and post-primary schools children Ilesa West, Osun State, Nigeria. It also agrees with Nkengazong et al. (2009) who also reported that differences in prevalence values of parasites between the sexes in Kotto Barombi and in Marumba II were not statistically significant. Also, Mafiana et al. (1998) and Agbolade et al. (2004) that helminthic infections were not sex dependent.

Similar findings have also been reported in the general population and school children in Nepal (Rai *et al.*, 1986, 1995, 2001a,b, 2002; Ishiyama *et al.*, 2001). These findings indicated that both sexes were equally exposed to enteric parasites, particularly diarrheagenic protozoa. This can be attributed to unplanned urbanization, which results in poor sanitary and hygienic conditions and contamination of drinking water with fecal matter (Adhikari *et al.*, 1986; Ono *et al.*, 2001). Our findings also disagrees with that of Anosike et al. (2004) who reported that parasitic infections were significantly higher in males than females. Adeyeba and Akinlabi (2002) and Baldo et al. (2004) showed that infection rates for intestinal parasites were higher in males than females. Chukwuma et al. (2009) reported prevalence of parasitic infection to be higher in females than in males. Alli et al. (2011b) reported a significant relationship between intestinal parasites among palm wine drinkers and sex; males were found to be positive to all the parasites encountered, whereas no female was positive. Okonko et al. (2009) reported that gastro-intestinal parasite infections from 2002 to 2004 were significantly higher in males than females ($p < 0.05$).

The study revealed a significantly higher prevalence (38.7%) of enteric parasitosis in children 4 to 5 years old. High prevalence of enteric parasitosis observed in children 4 to 5 years old in this study was also in agreement with the findings of Ishiyama *et al.* (2001). This disagrees with Odu et al. (2013) who reported that prevalence of intestinal parasites were not age dependent. This might be due to habits as well as poor or lack of environmental sanitation especially where people eat or drink. Also, low body immune system especially as concerned children might be responsible for high infection rate reported in this study (Sorensen *et al*., 1996; Alli et al., 2011b).

In this study, the intestinal parasites detected include *Ascaris lumbricoides*, *Entamoeba histolytica* and *Entamoeba coli.* Mixed infection was not observed. These intestinal parasites have been reported in various parts of Nigeria (Mordi and Ngwodo, 2007; Ajero et al., 2008; Okolie et al., 2008; Tohon et al., 2008; Chukwuma et al., 2009; Awolaju and Morenikeji, 2009; Okonko et al., 2009;

Odu et al., 2010, 2011a,b, Alli et al., 2011a,b; Odu et al., 2013).

Ascaris lumbricoides was the most predominant (54.8%). This was in agreement to most findings reported from Nepal in which *A. lumbricoides* topped the list of detected parasites from children diarrheal (Rai and Gurung, 1986; Rai *et al.*, 1995, 2000a,b, 2001a,b, 2002). The higher prevalence reported for *Ascaris lumbricoides* in this study agreed with previous report by Adeyeba and Akinlabi (2002), Agbolade et al. (2004); Alli et al. (2011a) and Odu et al. (2013). Okolie et al. (2008); Okonko et al. (2009); Alli et al. (2011a) and Odu et al. (2013) also reported *A. lumbricoides* to be most predominant in their studies. Infection by *Ascaris lumbricoides* is spread through eggs, which are swallowed as a result of ingestion of contaminated soil or contact between the mouth and the various objects carrying the adherent eggs. Contamination of food or drink by dust or handling is another source of infection. Ascaris ova are spread through the agents of flood and coprophagous animals, and can thus be transported to locations far from the defecation sites (Obiamiwe and Nmorsi, 1990; Mordi and Ngwodo, 2007). The eggs are passed unaltered through the intestine of coprophagous animals. The well-protected eggs can withstand drying and can survive for very lengthy periods. Soil pollution is thus a major factor in the epidemiology of human ascariasis (Mordi and Ngwodo, 2007).

The 54.8% prevalence reported *Ascaris lumbricoides* in our study is comparable to the prevalence reported by some researchers in other parts of Nigeria. A prevalence of 57.4% was reported by Odu et al. (2013) among school children. Egwunyenga et al. (2004) reported a prevalence of 55.0% in Eku, Delta State of Nigeria. Nwosu et al. (2004) reported a prevalence of 52.0% in school children in Abia and Imo States of Nigeria. Odikamnoro and Ikeh (2004) reported a prevalence of 51.5% among the Kpiri-kpiri community of Abakiliki of Ebonyi State, Nigeria. The 54.8% however, is high when compared with what has been previously reported in other areas. Mordi and Ngwodo (2007) reported a value of 30.0% in all the eighteen local government areas of Edo State, Nigeria. Human ascariasis is spread through faecal pollution of soil, and so the intensity of infection depends on the degree of soil pollution (Mordi and Ngwodo, 2007).

Entaemoeba coli (25.8%) is the second predominant intestinal parasite in this study. This also contrasted the findings of Egwari *et al*. (2005) in Lagos southwest Nigeria who in their study of sachet water found no *Entamoeba coli*. Human amoebiasis is a disease caused by a Protozoan amoeba of the genera *Entamoeba*, of which *E. histolytica* is the most medically important. Infection occurs when man ingests foodstuffs, vegetables or drink water faecally contaminated by cysts of the parasite (Ajero et al., 2008). Egwari *et al*. (2005) in Lagos, Nigeria also noted that *Entaemoeba coli* and other enteric pathogens formed a significant part of the isolates on the outside sachet surfaces of samples collected from cooling receptacles (pail, basin, wheel barrow, and refrigerator).

Entamoeba histolytica is the most medically important species of amoeba of man. Infection of man by *E. histolytica* commonly results to amoebiasis (Okonko et al., 2009). *E. histolytica* (19.4%) is the least predominant intestinal parasite in this study. This finding disagrees with previous studies done elsewhere in Nigeria and some of these studies had much higher prevalence values. Okonko et al. (2009) reported 51.7% prevalence. Nnochiri (1965 cited in Mordi and Ngwodo, 2007) reported a value of 94.0%. Our findings had much higher prevalence values compared to other studies done elsewhere in Nigeria. Adeyeba and Akinlabi (2002) reported 1.8% for *E. histolytica*. Onyido et al. (2002) reported 5.6% while Anosike et al. (2002) reported 5.5%. Omudu et al. (2004) reported a value of 20.3%. Mordi and Ngwodo (2007) reported a prevalence value of 4% for *E. histolytica*. Okolie et al. (2008) reported a prevalence value of 19.1% for *E. histolytica* in Oguta, Imo State. Awolaju and Morenikeji (2009) reported 9.29%. Outside Nigeria, Jimenez-Gonzalez et al. (2009) reported a value of 1.7% in study among the inhabitant of a rural community in Mexico. *E. histolytica* is indicator organism of faecal contamination, are frequently present in street foods, and street food may cause outbreaks of ameobiasis, cholera, typhoid and heptitis A (Ajero et al., 2008). Of the 6 incriminating parasites in a study by Alli et al. (2011c), *Entamoeba species* 7(31.8%) was most predominant. Of the three parasite species observed in the study by Okonko et al. (2009), *Entamoeba histolytica* had the highest prevalence of 51.7%.

The higher prevalence and incidence of *Entaemoeba histolytica* and *Entaemoeba coli* are a reflection of the poor environmental sanitation and very poor personal hygiene and unclean habits practiced by endemic villagers compounded by public ignorance and illiteracy. In addition, in Nigeria, surveys by Ajero et al. (2008) and Okonko et al. (2009) among others indicated that intestinal parasitic infections such as amoebiasis are growing problem in the country.

5. CONCLUSION

The findings of this study have shown that three intestinal parasites (*Ascaris lumbricoides*, *Entamoeba histolytica* and *Entamoeba coli*) were prevalent among children with diarrhea in Abeokuta, Ogun State, Nigeria whose stool samples were used for this study. The study has also shown that intestinal parasites are still highly prevalent among children with diarrhea in Abeokuta, Nigeria and a major cause of morbidity in children. The presence of these three parasitic intestinal parasites among children with diarrhea in Abeokuta, Ogun State, Nigeria supports the earlier observations that parasitic infections constitute a major public health problem in the country. The data obtained from this study provides information on the various parasitic diseases associated with gastro-intestines of children with diarrhea in Abeokuta, Ogun State, Nigeria. The study also provides data for understanding the epidemiological status of the human gastrointestinal parasites in Abeokuta, Ogun State, Nigeria. Therefore, there is also a need for regular awareness programs on sanitary and good hygiene among children. Since parasites and diarrhea are more frequent in poor populations and in immunocompromised individuals, preventive measures and surveillance systems must be emphasized.

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