Prevalence and Socio-environmental Predictors of Cryptosporidiosis in Kebbi State, Nigeria

Danladi Yusuf Kanya*, Ugbomoiko Uade Samuel, Babamale Olarewaju Abdulkareem

Department of Zoology, University of Ilorin, Ilorin, Nigeria

Email address: danladiyusuf18@yahoo.com (D. Y. Kanya), samugbomoiko@yahoo.com (U. U. Samuel),olas4nice2004@yahoo.co.uk (B. O. Abdulkareem)

To cite this article: Danladi Yusuf Kanya, Ugbomoiko Uade Samuel, Babamale Olarewaju Abdulkareem. Prevalence and Socio-environmental Predictors of Cryptosporidiosis in Kebbi State, Nigeria. American Journal of Bioscience and Bioengineering. Vol. 3, No. 6, 2015, pp. 149-157. doi: 10.11648/j.bio.20150306.12

Abstract: A cross-sectional study to determine the prevalence and socio-environmental factors associated with cryptosporidiosis was carried out between January to December 2012 in two communities in Kebbi state, Nigeria. Faecal specimen was collected from each participant and structured questionnaire applied. Samples were examined for Cryptosporidium by formal-ether concentration and modified Ziehl-Neelsen staining technique. A total of 2100 participants were tested, 290(13.8%) were infected with Cryptosporidium. Logistic regression analysis indicates that, in Aliero, infection was associated with: Presence younger children (Odds Ratio=1.889, P-value <0.0001, 95% Confidence Interval= 1.568-2.274), Regular hands washing (OR=0.399, P<0.0001, 95% CI=0.283-0.535), Presence of diarrhoea (OR= 2.66, P<0.0001, 95% CI=1.733-4.100), While in Zuru, the infection was predicted by: Younger age group (OR=1.283, P= 0.004, 95 % CI=1.085-1.520), Married status (OR=2.463, P=0.028, 95%CI=1.100-5.513), Lack of formal education (OR=2.993, P<0.0001,95% CI=1.872-4.786) and Farming occupation (OR=1.392, P=0.002, 95% CI=1.135-1.703). It was concluded that unhygienic behavioral variables, certain environmental and socio-demographic factors predicted the presence of cryptosporidiosis in the area. Sustainable intervention should include basic health education, access to clean water and adequate sanitation.

Keywords: Cryptosporidiosis, Prevalence, Risk Factors, Kebbi State

1. Introduction

Cryptosporidium is a common diarrhoea-causing parasitic genus in the world. It is a significant human pathogen and its known zoonotic potential makes it a threat to global public health [1, 2]. Its transmission has been associated with contaminated drinking water and food, low socioeconomic status and overcrowding conditions [3].

Most studies on Cryptosporidium infections in Nigeria and other places in the tropics, have focused on the occurrence of infection in hospitals and health care settings. Few studies have paid attention to the sources of infection within a population, or suggested control measures to reduce the risk of disease in developing countries. Currently 20 valid species of Cryptosporidium are known of which eight have been linked to human infection. Transmission routes follow a person to person and zoonotic patterns, both directly and indirectly, through the environment. Statistical association of disease with exposure risks [4-6] have explained already established links to direct transmission [7-9]. While zoonotic and waterborne transmissions are commonly reported in industrialized nations, the situations in developing countries however, have no conclusive evidence for these transmission routes. Reported presence of Cryptosporidium oocysts in domesticated and wild animals, and drinking water sources suggests a zoonotic potential also in the tropics [10-13]. The present situation in developing countries is such that, only a few, molecular-based information is available describing species infecting human or non-human hosts in the environment.

Currently, known risk factors for acquiring cryptosporidiosis are mainly derived from outbreaks studies, in spite of the fact that the majority of human infection is sporadic. Our knowledge of the existence of multiple transmission routes, including consumption of water from private and public supplies, consumption of contaminated foods and drinks, contact with infected animals, person-to-person transmission in households and care settings, and exposure to recreational water in swimming pools or water parks, and travel to endemic countries were mostly derived from Outbreak investigations.
Disease determinants are likely to differ for various species and in different geographical settings, due to differences in demographic, socioeconomic, environmental and behavioural variables. This underscores the importance of molecular typing in identifying species or genotype in epidemiologic studies. However, this has been possible only on a relatively few case–control studies in developing countries [15, 16].

Despite our knowledge of cryptosporidiosis transmission, often the exact mode of transmission is difficult to establish. The importance of certain risk factors for acquiring infection remains unclear. For instance the extent to which socio-economic, behavioural and environmental factors contribute to the transmission of this disease is not sufficiently established. In an effort to fill this gap the influence of these factors on the spread of the disease in two communities in Northwestern Nigeria were examined from January to December, 2012.

2. Materials and Methods

Zuru is located in the south eastern part of Kebbi state, between longitude 11°24'09" N and latitude 5°15'07" E. It is about two and half hours drive from Birnin-kebbi, the state capital. It has an estimated population of about 165,547 thousand people living in the local government area [17]. Aliero on the other hand is located in the extreme northwestern part, between latitude 12°19'06" N and 4°30'10" E. Based on the 2006 national census, Aliero has a population of about 123,785 people [18]. The two communities feature low socioeconomic status and poor environmental sanitation. Adequate water supply, sewage, and waste disposal systems are lacking. Garbage is burned or thrown away near houses and can be found deposited on several places. Zuru land supports the savannah kind of vegetation with pockets of woodland vegetation along the river basins. Grains, tubers, legumes, fruits are grown in the area. While Aliero is flat and slightly undulating with compact stony and brown soil, and has northern guinea savannah vegetation. The leading economic activity in both communities is mainly agriculture. The inhabitants therefore are mostly farmers, animal rearers, blacksmiths, traders and some are civil servants. The people of Aliero are predominantly Hausa/ Fulani, but Zuru people are Dakarkari by ethnicity.

3. Study Design

3.1. Human Populations

Participants were randomly sampled for this study from volunteered families in the two communities identified with the highest prevalence during an earlier pilot study. Ethical approval was given by the Kebbi State ministry of health Birnin-kebbi. Informed verbal consent was obtained from all family heads, adults and parents of children before participating in the study. Samples were collected and screened for the presence of *Cryptosporidium* Oocysts.

3.2. Administration of Questionnaires

Pre-tested structured questionnaires were administered both in English and Hausa languages (as preferred by the interviewee) to all members of volunteered families.

Data such as demographic, socio-economic, Knowledge, attitudes and practices relating to Cryptosporidiosis infection, e.g. Sex, age, educational level, average monthly income, regular handling of animals and their wastes around the house, sources of water supply, toilet systems, waste disposal methods, disease perception etc., were taken.

3.3. Stool Collection and Processing

Stool samples were collected from house to house using clean, grease-free, tightly capped, and labeled sterile stool plastic bottles. The field workers visited each home and a single fresh stool sample was collected from each consenting family member. Samples were then transported to the biology laboratory of Kebbi State University of Science and Technology, Aliero or the medical laboratory of the Martha Bamayi General Hospital, Zuru depending on the collection site, for immediate processing within 24 hours.

In the laboratory, stool samples were concentrated by formal-ether technic. Briefly using an applicator stick, about 1g of stool sample was placed in a clean 15 ml conical centrifuge tube containing 7 ml formalin. The sample was suspended and mixed thoroughly with applicator stick. The resulting suspension was filtered through a sieve (cotton gauze) into a beaker and the filtrate was poured back into the same tube. The debris trapped on the sieve was discarded. To this mixture, 3 ml of diethyl ether is added and hand shaken; the content is centrifuged at 2000 rpm for 3 minutes. The supernatant was poured away, living only the fine sediment at the bottom of the tube [19]. This was then used to prepare slides for the detection of *Cryptosporidium* Oocysts.

3.4. Modified Ziehl-Neelsen Acid Fast Technique

One to two drops of the sediment was smeared on the slide and air dried. This was fixed with absolute methanol for 1 minute. The slide was flooded with carbolfuchs in for 15 minutes and rinsed thoroughly with water and decolorized with 1% alcohol for 2 minutes after which it was rinsed with water. The smear was then counter stained with malachite green or methylene blue for 1 minute and rinsed with water. This was finally air dried and examined under the microscope using the 40x objective. To achieve better view 10x objectives using oil immersion were used. Where present, *Cryptosporidium* oocysts are round and stain red [20].

3.5. Statistical Analysis

Statistical analysis was performed using the version 15 Statistical package for Social Sciences (SPSS Inc, Chicago, IL) on windows 10. Chi square test was used to compare prevalence by age and gender and to confirm possible association between infection and other exposure variables. Multivariate logistic-regression analyses were carried out to
determine independent risk factors associated with infection in the study areas. Associations were considered significant at $P \leq 0.05$.

4. Results

![Figure 1. Prevalence of Cryptosporidium infection stratified by age and study communities in Kebbi state.](image)

Table 1. Prevalence of Cryptosporidium infection stratified by demographic factors of inhabitants in study areas.

| Variable          | Infection | $\chi^2$ | P-value |
|-------------------|-----------|----------|---------|
| Number examined   | Number infected | Percent age (%) | |
| Age ≤ 5           | 606       | 128      | 21.1    | <0.0001 |
| 6-10              | 339       | 40       | 11.8    |         |
| ≥ 14              | 1155      | 122      | 10.6    |         |
| Gender            |           |          |         |
| Male              | 1158      | 175      | 15.1    | 0.055   |
| Female            | 942       | 115      | 12.2    |         |
| Community         |           |          |         |
| Aliero            | 1095      | 123      | 11.2    | <0.0001 |
| Zuru              | 1005      | 167      | 16.6    |         |
| Marital status    |           |          |         |
| Single married    | 1680      | 268      | 16.0    | <0.0001 |
| Divorced          | 377       | 21       | 5.6     |         |
| Widow/widower     | 29        | 0        | 0       |         |
| Religion          |           |          |         |
| Islam             | 1861      | 252      | 13.5    | 0.612   |
| Christianity      | 226       | 37       | 16.4    |         |
| Others            | 13        | 1        | 7.7     |         |
| No of persons/household < 8 | 541 | 87 | 16.1 | 0.075 |
| ≥ 8               | 1559      | 203      | 13.0    |         |

Table 2. Prevalence of Cryptosporidium infection by socioeconomic factors in Aliero and Zuru, Kebbi State.

| Factor                        | Infection | $\chi^2$ | P-value |
|-------------------------------|-----------|----------|---------|
| Number examined               | Number infected | Percent age (%) | |
| Educational level             | 643       | 78       | 12.1    | 2.195   |
| Can read and write            |           |          | 0.138   |         |

Table 3. Prevalence of Cryptosporidium infection in Zuru and Aliero by environmental factors.

| Factor                       | Cryptosporidium infection | $\chi^2$ | P-value |
|------------------------------|---------------------------|----------|---------|
| Number examined              | Number infected | Percent age (%) | |
| Animals kept in house         | 450                  | 53       | 18.4    | 32.887  | <0.0001 |
| Sheep/Goat                   | 767                  | 81       | 10.6    |         |         |
| Cat                          | 412                  | 46       | 11.2    |         |         |
| Domestic fowls/pegion        | 362                  | 72       | 19.9    |         |         |
| Pig/Others                   | 109                  | 8        | 7.3     |         |         |
| Toilet facilities            |                       |          |         |
| Water closet                 | 225                  | 30       | 13.3    | 26.461  | <0.0001 |
| Pit latrine                  | 1493                 | 180      | 12.1    |         |         |
| Bush/other                   | 382                  | 80       | 20.9    |         |         |
| Source of water supply       |                       |          |         |
| Public taps/Bore holes       | 1127                 | 131      | 11.6    | 23.106  | <0.0001 |
| Well/stream                  | 973                  | 159      | 16.4    |         |         |
| Waste disposal method        |                       |          |         |
| Usually burnt                | 138                  | 13       | 9.4     | 156.73  | <0.0001 |
| Common site in the house     | 1185                 | 124      | 10.5    |         |         |
| No specific site/others      | 777                  | 153      | 19.7    |         |         |
| Children <6 years/household  |                       |          |         |
| <6                           | 1167                 | 184      | 15.8    | 8.455   | 0.004   |
| ≥6                           | 933                  | 106      | 11.4    |         |         |
The overall prevalence of the infection was 13.8% (290/2100). Table 1 shows the pattern of infection in relation to demographic variables. Distribution was age-dependent with significantly higher prevalence among younger subjects (21.1%) than the olds (10.6%). Gender wise, prevalence was higher among males (15.1%) than females (12.2%) but the effect was not significant. With respect to location, infection was significantly related to the community in question. Peak prevalence was recorded in Zuru (16.6%) than Aliero (11.2%). Prevalence was significantly higher among the singles (16.0%) than married participants (5.6%). Distribution of prevalence with respect to individual’s religious affiliation ranged between 7.7% - 16.4% and was not significant. While none was recorded among divorcees. Crowding in homes shows no significant effect among subjects. In this case prevalence was higher among less crowded homes (16.1) than the crowded (13.0%).

The effect of socioeconomic variables on the prevalence of Cryptosporidium in subjects is presented in table 2. Level of education showed no significant influence (p=0.138) on prevalence despite higher infection rate among illiterates (14.6%), than in those who had formal education (12.1%). A noticeable disparity was observed among various occupations, where prevalence varied from 8.6% - 17.0%. Family income played significant role in determining prevalence among various income categories. Persons from households with no certain income had higher prevalence (17.0%) than those who had some form of livelihood (9.9% - 12.5%). In terms of infection with respect to type of house, prevalence varied significantly among grass house owners (20.0%) and lower among owners of houses made of cement bricks (12.8). However, owners of busch lamp (as source of light) had a higher rate of infection (15.7%) than those who had electricity supply (12.9%), but the distribution was not different.

Table 3 depicts the association of environmental factors with infection. Of the reported ownership of domesticated animals, prevalence was common among those who kept domestic birds (19.9%, P<0.0001) than other animals (5.3% - 18.4%). Dissimilarity was observed in the distribution of infection with respect to types of toilet facilities used by participants. Infection was significantly higher among those who defecated in nearby bushes (20.9%, <0.0001) than those who owned a private pit latrine or water closet. Variation in prevalence occurred with respect to Source of water supply. Consumption or use of water from wells/streams predisposed subjects to significantly higher levels of infection than those who have their supplies from Boreholes/taps (16.4%, 11.6%, <0.0001 respectively). Waste disposal methods used by participants affected distribution. Infection was significantly lower among those coming from homes where refuse is simply dumped them at some common place (10.5%) or at no specific place (19.7%) in their homes. Presence of young children in home was observed to be significant factor. Families with young children <6 years of age had significantly higher rates than those with older children (15.8%, 11.4%, P=0.004 respectively).

Prevalence in relation to behaviour and disease perception is shown in table 4. Hand washing before eating was significantly associated with infection (p <0.001). The highest prevalence occurred among subjects who never practiced hand washing before eating (36.4%) than in those who washed their hands regularly before eating (7.4%).
Although the reported experiences of abdominal discomforts were common among subjects (14.6%), this showed no association with Cryptosporidium infections (p=0.199). More than half of subjects (54.5%) had no idea of how the infection is transmitted or acquired. Among the subjects 123(5.8%) persons believed that cryptosporidiosis is caused by evil spirits and interestingly the highest prevalence (16.3%) was recorded among this group. Presence of diarrhoea featured as an important factor for occurrence of the parasite (19.5%, p = <0.001) than absence of it in subjects (12.8%).

The results of multiple regression analysis showed that several independent factors for Cryptosporidiosis are involved. Results for Aliero and Zuru are summarized in table 5. In Aliero, infection was age dependent. Presence of Young children had almost 2 times increased risk of infection than older persons (OR=1.889, P <0.0001, CI= 1.568 – 2.274). Regular hands washing before eating by residents was significantly protective than occasional or no hands washing at all (OR=0.399, P<0.0001, 0.283 – 0.535). Presence of diarrhoea was about 3 times more likely to determine an infection than its absence (OR=2.66, P< 0.0001, CI= 1.733 – 4.100) (table 5). While in Zuru, four factors were significantly predictive. Younger age increased the risks of infection (OR=1.284, P= 0.004, CI= 1.085 – 1.520), married persons were more than twice at risk of infection than the unmarried (OR=2.463, P=0.028, CI= 1.100 – 5.513), A lack of formal education heightened risks by 3times more than being educated (OR= 2.993, P<0.0001, CI= 1.872 – 4.786), farming occupation had increased risks of infection than other trades (OR=1.392, CI= 1.135 –1.703).

5. Discussion

The data in this study provides evidence of the link between certain socio-environmental factors and cryptosporidiosis in North-western Nigeria. Prevalence of infection in this study is relatively high (13.8%) compared to similar studies reported from Nigeria and other tropical countries. In North-western Nigeria, [21] reported a prevalence of 1.9% among patients aged 2 months to 70-years-old, while [22] reported a prevalence rate of 4.8% among malfomnourished children aged 0 – 5 years. In another report, Cryptosporidium Oocysts were not detected in stools of 189 HIV –infected and uninfected patients [23] and 52 malfomnourished HIV –infected children [24].

In contrast, higher prevalence rates of 17 – 52.7% were reported in other studies from Nigeria [25, 16]. In other tropical countries, prevalence ranges from 0.1 – 32.5% [26] and the reported prevalence in children from Uganda, Kenya and Brazil was 72.7%, 4% and 31.3% respectively.

In the present study, significant difference in prevalence is observed between the two communities (P=0.0001). Subjects in Zuru had a higher infection rate (16.6%) than their counterparts in Aliero (11.2%). This is attributable to possible differences in community practices relating to infection and levels of environmental contamination ([27]).

According to Zaidah et al. disparity in distribution may be linked to population characteristics, geographic location, and detection methods. In addition, it is also opined that prevalence may vary due to differences in infection sources and other risks of infection [25].

Prevalence was intimately associated with age. Young children were almost 2 times at risk of infection than older persons >5years of age in both communities. Similar trends have been reported in Nigeria and other countries in Africa [16, 21, 23 - 25, 28 - 34] This outcome, may also infer that infection due to presence of children is evidence of possible person-to-person (anthropogenic) transmission. This observation is similar to that of [21] in which younger age was significantly associated with Cryptosporidium infection.

The difference in infection between male and female gender was at a border line significance (P = 0.055). But a number of reports indicate that the infection may not always have a positive association with gender since in many communities both gender are equally exposed to similar risks of infection. Both sexes engage in same recreational activities and so are likely to be equally exposed to any environmental contamination. For example, [35] did not find any marked difference in infection between both sexes among Cuban children. In previous studies in Cuba and South Africa, [37] respectively had reported that gender did not present any differences in infection for Cryptosporidium. This pattern has also been confirmed in a study in Ethiopia, that cryptosporidiosis has no association with a particular sex group [38].

In this study hand washing before eating was significantly protective. Residents who washed hands regularly before eating were less likely to be infected than those who only washed occasionally or do not wash at all. It is immensely important that health care providers should at least educate and emphasize the importance of hand washing among residents. Increased hand infections and illnesses, in addition to diarrhoeal disease, could be prevented. Hand washing is an economical method of primary prevention. Additional studies are needed to evaluate the durability of this behavioural change in hand washing and the prevention of diarrhoeal illness and other illnesses in developing regions of the world.

Of the study subjects, 390(18.6%) alluded that they ate with unwashed hands. More than half 1080(51.4%) admitted that they only washed hands occasionally before eating. It is imperative to state here that, while access to improved facilities and sanitation are key factors to reduction in parasitic burden, the behaviour and education of the people is an important factor. For instance [39] showed that improved water supply and sanitation without proper education and betterment of living standards does not always lead to a reduction in transmission levels of enteric parasitic diseases.

Our findings also showed an intimate association between patterns of infection and access to good water supply and sanitation. Access to boreholes/taps resulted in lower infection rate (% 11.6) than other less safe sources of water (River/streams) supply (16.4). The association of water sources to Cryptosporidium infection as observed in this
practice of indiscriminate littering (rather than incinerating) of waste, the prevalence of infection (22.3%) was recorded among subjects who had kept domestic animals at home [42, 43, 44]. The study observed that socio-economic characteristics of residents will play vital roles in explaining the observed trends. The keeping of pets and rearing of domestic animals at home is a common practice in northern Nigeria. There are reports worldwide on associated risk of Cryptosporidium infections in people who keep domestic animals at home [42, 43, 44]. In this study, virtually all participants kept one type of animal or the other or at least lived with neighbors who kept animals at home. Perhaps, owners of pets or domestic animals may not have observed good hygiene or sanitation in their homes to prevent infection. A better assessment however should have been a comparison of genotypes detected from the stools of children with genotypes also detected from the stools of domestic animals. Usually these animals defecate on the compound soil, and sometimes dry up on the soil to contaminate the environment, which also serve as playing grounds for children. Cryptosporidiosis being a zoonotic infection, it is suspected that some level of infections of the parasites could come from infected animals kept at home. Reports from other studies indicate, on the contrary that, having one or more pets or domestic animals at home was not a risk factor for acquiring Cryptosporidium infections [36, 45]. Furthermore, there was correlation of educational background with infection rates. Illiterates were almost three times prone to infection than those who can read and write. The observations made are possible despite the influence of other factors such as type of occupation of residents. It is often upheld by many that the well-educated are likely to practice personal hygiene better than those who have only low level of education or are illiterate. Since level of sanitation and hygiene both affect transmission of this parasite, it is expected that the rate of infection among illiterates will be higher when compared to those with good education. Similarly, presence of a toilet facility was protective compared to open defecation in nearby bushes. The highest prevalence of infection (22.3%) was recorded among subjects who defecated in the open bush. Comparable patterns have also been observed by earlier studies [46, 47].

Contamination of the environment has a strong relationship with enteric parasitic infections [39]. The effect is that where human or animal excreta are indiscriminately thrown around, fecal-borne pathogens will litter the environment thereby increasing the risks of infection [48]. The outcome of this study reveals a significant association (P< 0.001) of waste disposal method to infection. The practice of indiscriminate littering (rather than incinerating) of wastes in households tended towards an increase of infection than it was in other forms of waste disposal. Socioeconomic inequalities has been linked to a wide range of health outcomes, including premature mortality, cardiovascular disease, and self-reported ill health (Lawlor and Sterne, 2007) [49]. The interaction of infection with socioeconomic characteristics as observed in this study associates the level of household income to a significant relationship with infection (P= 0.028). Prevalence was higher among the average to low income class, than it was among the higher income earners. This study and others support the conclusion that low socioeconomic status is closely related to enteric parasite infections [50-54].

Some occupational practices have been associated with enteric parasitic infections [55]. Subjects from farming families at Zuru were significantly at risk for infection compared to other occupations tested in this study. Zoonotic exposures and other agricultural practices in some regions are key factors for transmission. The use and recycling of wastes, household sewage, human and animal excreta in agriculture and aquaculture has a long history in many countries [56-60]. The reuse of excreta and wastewater for crops and fish ponds may provide many positive benefits, such as cheap fertilizer, reliable source of nutrition and water, reduce commercial fertilizers, improve soil-structure and increase productivity [61–65]. However, transmission of enteric pathogens is a fundamental public health issue associated to these practices. In developing countries, excreta-related diseases are very common, and faecal sludge and wastewater contain high concentrations of excreted pathogens such as viruses, bacteria, protozoa cysts, and helminthes eggs that may cause gastrointestinal infections in humans. The pathogens most commonly found in the waste water include faecal protozoa (Cyclosporacayetanensis, Cryptosporidium parvum, Entamoeba histolytica, and Giardia intestinalis), soil-transmitted helminths (Ascaris spp., Trichuris spp., and hookworm) and other microbes [55].

It is so alarming to observe that despite the wide spread occurrence of this infection in the areas, more than half of the subjects i.e. 1144 (54.5%) had no idea how Cryptosporidium infection is transmitted. About 6% believed that the infection is caused by evil spirits. Such ignorance/superstition is a common feature in most parasite endemic communities. And should call for concerted efforts by relevant stake holders to combat this, through basic health education and enlightenment campaigns.

6. Conclusion

In conclusion, the presence of Cryptosporidiosis in the two communities is associated to a significant extent with a few important variables of human behaviour, certain environmental and socio-demographic factors. Sustainable intervention measures should be implemented to reduce the burden of this neglected disease, part of which should include health education, improved access to clean water and adequate sanitation. More studies are needed in this and other settings with similar epidemiological features to further evaluate these factors.
References

[1] Gambo, A., Inabo, H.I., Aminu, M. (2014). Prevalence of Cryptosporidium Oocysts among children with acute gastroenteritis in Zaria, Nigeria. *Bajopas* 7(2): 155 – 159.

[2] Fletcher, S.M., Stark, D., Harkness, J. and Ellis J., (2012). Enteric protozoa in the developed world: a public health perspective. *Clinical Microbiology Reviews* 25(3), 420-449.

[3] Karanis P, Korenti C, and Smith H. (2007). Waterborne transmission of protozoan parasites: a worldwide review of outbreaks and lessons learnt. *J. Water Health* 5, 1-38.

[4] Chung, R.N., Simwa, J.M., Karumba, P.N., Kenya, P.R., Kinoti, S.N., Muttinga, J., and Nagelkerke, N. (1992). Comparative etiology of childhood diarrheaean Kakamega and Kiambu districts, Kenya. *East Afr Med J.*, 69(8), 437-41.

[5] Newman, R. D., Sears, C.L., Moore, S.R., Nataro, J. P.,  et al. (1999). Longitudinal study of *Cryptosporidium* infection in children in northeastern Brazil. *J. Infect. Dis.* 180, 167-75.

[6] Periera, S.J., N.E. Ramirez, Xiao, L.A. Ward. (2002). Pathogenesis of human and bovine *Cryptosporidium parvum* in gnotobiotic pigs. *Journal of Infectious Diseases* 186: 715-718.

[7] Elsser, K.A., Moricz, M., Proctor, E.M., (1986). *Cryptosporidium* infections: a laboratory survey. *CMAJ*. 135(3), 211-3.

[8] Navarrete, S., Stetler, H.C., avila, C., Garcia aranda, J.A., and Santos-preciado, J.J. (1991). An outbreak of *Cryptosporidium* diarrhea in a pediatric hospital. *pediatr Infect dis J* 10(3), 248-50.

[9] Siwilia J, phiri IG, Vercruyssje j, Goma f, Gabriels S, Claereboute, Geurden T. (2007). Asymptomatic cryptosporidiosis in Zambian dairy farm workers and their household members. *Trans R Soc Trop Med Hyg.* 101(7), 733-4.

[10] Hunter, P.R., Chalmers R M., Syed, Q., Hughes, L.S., Woodhouse, S., and Swift, L. (2003). Foot and mouth disease and cryptosporidiosis: possible interaction between two emerging infectious diseases. *Emerg Infect dis.*, 9(1), 109-12.

[11] Kelly, p., Baboo, K.S., Ndubani, p., Nkanginieme, K.E.O., Chira, F.W. and Onile, B.A. (1998). Epidemiology of giardiasis and *Cryptosporidium* infection inman and animals. *J. Water Health* 5, 1-38.

[12] Molbak, K, Aaby, P, Højlyng, N, and Da Siwa, A.P.J. (1994). Risk factors for *Cryptosporidium* diarrhea in early childhood: a case control study from Guinea-Bissau, West africa. *Am J epidemiol.*, 139, 734-40.

[13] Nizeyi, J.B., Sebunya, D., Dasilva, A.J., Cranfield, M.R., pienizek, N.J., Graczyk, T.K. (2002). Cryptosporidiosis in people sharing habitats with free-ranging mountain gorillas (*Gorilla gorillaberingei*), Uganda. *Am J Trop Med Hyg.*, 66(4), 442-4.

[14] Nichols G.L., Chalmers R.M., Soopth with, W, Regan M., Hunter C.A., Grenfell P., Harrison F. and Lan C. (2006).Cryptosporidiosis: a report on the surveillance and epidemiology of *Cryptosporidium* infection in England and Wales. *Drinking Water Directorate Contract Number DWI 70/2/201*. Drinking Water Inspectorate, UK. Pp142.

[15] Cama, V.A., Bern, C., Robert, s. J., Cabrera l, et al., (2008). *Cryptosporidium* species and subtypes and clinical manifestations in children. *Peru. Emerg. Infect. Dis.*, 14 (10), 1567-74.

[16] Molloy SF, smith HV, Kirwan P, Nichols AB, et al., (2010). Identification of a high diversity of *Cryptosporidium* species genotypes and subtypes in a pediatric population in Nigeria. *Am J Trop. Med. Hyg.*, 82 (4), 608-13.

[17] Molloy, S. F., Tanner, J., Kirwan, P., Asaolu, S. O., Smith, H. V., Nichols, R. A. B., Connelly, L., and Holland, C. V. (2011). Sporadic *Cryptosporidium* infection in Nigerian children: Risk factors with species identification. *Epidemiol Infect.*139, 946-948.

[18] National Population Commission of Nigeria, 2006.

[19] Lindo, F. L., Levy, V. A., Baum, M. K., and Palmer, C. J. (1998). Epidemiology of giardiasis and cryptosporidiosis in *Jama. Am. J Trop. Med. Hyg.*, 59 (5), 717-721.

[20] Cheesbrough, M., 2005: *District Laboratory Practice*. 2nd edition, Part I, Cambridge University press. Pp 236 -239.

[21] Maikai, B. V., Umoh, J. U., Lawal, I. A., Kudi, A. C., Ejimbi, C.L and Xiao, L. (2012). Molecular characterizations of *Cryptosporidium*, *Giardia* and Enteroctytozoan in humans in Kaduna State, Nigeria. *Exp. Parasitol*131, 452-456.

[22] Banwat, E.B., Egah, D.Z., Onile, B.A., Angyo, I. A., and Audu I. S. (2003). Prevalence of *Cryptosporidium* infections among under nourished children in Jos Central Nigeria. *Niger J Postgrad Med* 10: 84-87.

[23] Nwokediukwu, N. C., Bojuwoye, B. J and Onyekekwe, B. (2008). Detection of *Cryptosporidium* and giardiasis in patients with diarrhea in Osun State, South Western Nigeria. *Niger Postgrad Med J 39, 511-516.

[24] Adeyoba, O. A., 39(2007). Cryptosporidiosis in HIV infected patients with diarrhea in Osun State, South Western Nigeria. *Niger J Postgrad Med*, 9, 70-73.

[25] Banwat, E.B., Egah, D.Z., Onile, B.A. and Datong, P. R. (2004). *Cryptosporidium* infection in undernourished children with HIV/AIDS in Jos, Nigeria. *Annal of African Medicine*, 3(2), 80-82.

[26] Adeyoba, O. A., 39(2007). Cryptosporidiosis in HIV infected patients with diarrhea in Osun State, South Western Nigeria. *Niger J Postgrad Med*, 9, 70-73.

[27] O’Donoghue, P.J. (1995) *Cryptosporidium* and *Cryptosporidium* inman and animals. *Int. J. Parasitol.* 25, 139-195.

[28] Zaidah, A. R., Chan, Y. Y., Asma, H. S., Abdullah, S., Nursalindawati, A. R. and Saleh, M. (2008). Detection of *Cryptosporidium parvum* in HIV – infecteded patients in Malaysia using a molecular approach. *Southeast Asian J. Trop Med and Pub Health*, 39, 511-516.

[29] Oyerinde J.P.O., Benson R.I., Alonge A. A., Ogunmolu, F.A., Ogunlade, A.B., Banugha, M.A. (1999). Sporadic *Cryptosporidium* infection in Nigerian children with HIV/AIDS in Jos, Nigeria. *Annal of African Medicine*, 3(2), 80-82.
[30] Nwabuisi, C. (2001). Childhood cryptosporidiosis and intestinal parasitosis in association with diarrhea in Kwara state, Nigeria. West Afr J Med, 20 (2), 165-8.

[31] Inyang-Etoh, P.C., N.G. Etim, F.M. Useh, C.E.J. Udiong, and A.W. Essien (2007). Cryptosporidiosis and Infantile Diarrhoea in Calabar, Nigeria. J. Med. Sci. 7(8): 1325-1329.

[32] Tumwine, J. K., Kekithunwa, A., Nabukenya, N., Akiyoshi, D. E. R., Rich, S. M., Widmer, G., Feng, X and Tzipori, S. (2003). Cryptosporidium parvum in children with diarrhea in Mulago Hospital, Kampala, Uganda. Am. J. Trop. Hyg., 68(6)710-715.

[33] Abdel-messih, I. A., Wierzb, T. F., Abu-Elayed, R., Ibrahim, A.F., Ahmad, S. F., Kamal, K., Sanders, J., Frenc, R. (2005). Diarrhea associated with Cryptosporidium parvum among young children of the Nile River Delta in Egypt. J. Trop Pediatr., 51(3), 154-159.

[34] Gatei, W., Wamae, C.N., Waruru, A., Mulinge, E., Waithera, T., Liatika, S. M., Revathi, G., and Hart, C. (2006). Cryptosporidiosis: Prevalence, Genotype analysis and symptoms associated with infections in children in kenya. Am Soc. Trop Med and Hyg. 75(1), 78-82.

[35] Bello, J., Nuñez, F. A., Gonzalez, O. M., Fernandez, R., Almirall, P., and Escobedo, A. A. (2011). Risk factors for Giardia infection among hospitalized children in Cuba. Annals of Tropical Medicine & Parasitology, 105 (10), 57-64.

[36] Pelayo, L., Nun Ez, F.A., Rojas, L., Wilke, H., Furuseth Hansen, E., Mulder, B., Gjerde, B. and Robertson, L. (2008). Molecular and epidemiological investigations of cryptosporidiosis in Cuban children. Annals of Tropical Medicine & Parasitology, 102 (8), 659-669.

[37] Beauty Omoruyi, Fredrick Matongo, Nxoilo T. Nkwetshana, Ezekiel Green, Anna M. Clarke and Roland N. Ndip (2011). Environmental and demographic risk factors associated with the prevalence of Cryptosporidium infection in the Alice rural settlements of the Eastern Cape Province of South Africa: a pilot study. Rev Environ Health 26(2): 127–133.

[38] Eyasu, T., Beyene, P., and Tekola, E. (2010). Prevalence of Giardiasis and Cryptosporidiosis among children in relation to water sources in Selected Village of Pawi Special District in Benishangul-Gumuz Region, Northwestern Ethiopia. Ethiop. J. Health Dev; 10; 24(3), 205-213.

[39] Asaolu, S. O. and Ofoezie, I.E. (2003). The role of health education and sanitation in the control of helmint infections. Acta Tropica., 8,283-294.

[40] Redlinger, T., Corella-Barud, V., Graham, J., Galindo, A., Avitia R. and CardenasV. (2002). Hyperendemic Cryptosporidium and Giardia in households lacking manucipal sewer and water on the united states – Mexico border. Am J Trop Med Hyg 66, 794 -798.

[41] Nyamwange, C., Mkoji, G.M. and Mpoke, S. (2012) Cryptosporidiosis among HIV positive patients in North Rift Region of Kenya. Afr J Health Sci, 21, 92-106.

[42] Pinheiro, I., Milton, F., Mitterofhe, A., Condé Pires, F.A., Abramo, C., Luiz Cláudio Ribeiro, L. C., Tibiricâ, S.H.C., Coimbra, E.S. (2011) Prevalence and risk factors for giardiasis and soil-transmitted helmintihiasis in three municipalities of Southeastern Minas Gerais State, Brazil. Parasitol Res 108: 1123–1130.

[43] Anim-Baidoo, I., Narih, C., Obiri, D., Ewerenonu-Laryea, C., Donkor, E. S., Adjet, A. A., and P. F. Ayeh-Kumi (2015). Cryptosporidial diarrhea in children at a paediatric hospital in Accra, Ghana. JTDIH, 10(3): 1-13.

[44] Pereira, M.G.C.; Atwill, E.R.; Barbosa, A.P.; Silva, S.A. and Garcia-Zapata, M.T., (2002). Intra-familial and extra-familial risk factors associated with Cryptosporidium parvum infection among children hospitalized for diarrhea in Goiânia, Goiás, Brazil. Amer. J. Trop. Med. Hyg., 66, 787-793.

[45] Norma E. Ramirez, Lucy A.Ward, Sinand Sreevatsan (2004). A review of the biology and epidemiology of cryptosporidiosis in humans and animals. Microbes and Infection 6: 773–785.

[46] Pinheiro, I., Milton, F., Mitterofhe, A., Condé Pires, F.A., Abramo, C., Luiz Cláudio Ribeiro, L. C., Tibiricâ, S.H.C., and Coimbra, E.S. (2011). Prevalence and risk factors for giardiasis and soil-transmitted helmintihiasis in three municipalities of Southeastern Minas Gerais State, Brazil. ParasitolRes, 108, 1123-1130.

[47] Rao Ajampus, S.S., J.R. Asirvatham, D. Muthusamy, B.P. Gladstone, O.C. Abraham, D. Mathai, H. Ward, C.Wanke, C. kang, (2007). Clinical features and risk factors associated with cryptosporidiosis in HIV infected adults in India. Indian J Med Res, 126, 553-557.

[48] Bern, C., Ortega Y. Checkley, W., Roberts, J.M., Lescano, A.G., Cabrera, L., Verastegui, M., Black, R.E., Sterling, C. and Gilman, R.H. (2002). Epidemiologic differences between cyclosporiasis and cryptosporidiosis in Peruvian children. Emerging Infectious Diseases, 8:581-585.

[49] Chancin-Bonilla, L., Borrios, F., and Sanchez, Y. (2008). Environmental risk factors for Cryptosporidium infection in an island from western Venezuela. Mem Inst Oswaldo Cruz, Rio de Janeiro 103(1), 45-49.

[50] Pickford, J. (1988). Rural sanitation and development. Waterlines, 6, 2-4.

[51] Lawlor, D.A. and Sterne, J.A.C. (2007). Socioeconomic inequalities in health. Brit Med J, 334, 963-964.

[52] Drake, L. J. and Bundy, D.A. (2001) Multiple helmint infections in children: Impact and control. Parasitology 122, 73-81.

[53] Hostej, P.J. and Kamath, A. (2009). Neglected tropical diseases in Sub-Saharan Africa: A review of their prevalence, distribution and disease burden. PLoS Negl. Trop. Dis. 3: 412.

[54] Ugboimoiko, U.S. V. Dalmono, I.E. Ofoezie, and Abiezue, R.N.N. (2009). Socio-environmental factors and Ascarisiasis infection among school-aged children in Ilobu, Osun State, Nigeria. Transc Royal Soc. Trop Med and Hyg 103, 223-222.

[55] Ugboimoiko, U.S. I.E. Ofoezie, I.C. Okoye, and J. Heukelbach, (2010). Factors associated with urinary Schistosomiasis in two peri-urban communities in southwestern Nigeria. Ann. Trop med parasit, 104(5), 104 – 419.

[56] Ugboimoiko, U.S. V. Dalumo, Y.K. Danladi., J. Heukelbach, and I.E. Ofoezie, (2012). Concurrent Urinary and intestinal Schistosomiasis and intestinal helmintic infections in Ilobu, Southwestern Nigeria. ActaTropica123, 16- 21.
[57] Raschid-Sally, L. and Jayakody, P. (2008). Drivers and characteristics of wastewater agriculture in developing countries: results from a global assessment. Colombo, Sri Lanka: International Water Management Institute. Agriculture, Veterinary Science, Nutrition and Natural Resources, 3, 15.

[58] Phuc, P.D., Konradsen, F., Phuong, P.T., Cam, P.D. and Dalsgaard, A. (2006). Practice of using human excreta as fertilizer and implications for health in Nghean Province, Vietnam. S E Asian J Trop Med Public Health, 37, 222-229.

[59] Asano, T., Burton, H., Tsuchihashi, R., Tchobanoglous, G. (2007). Water reuse: Issues, Technologies, and Applications. McGraw-Hill Professional, New York p1570.

[60] WHO, (2006) Guidelines for the safe use of wastewater, excreta and greywater - Volume 2: Wastewater use in agriculture. In Geneva, Switzerland: World Health Organisation.

[61] Carr, R. and Strauss, M. (2001). Excreta-related infections and the role of sanitation in the control of transmission. In Water quality: Guidelines, standards and health, assessment of risk and risk management of water-related infectious disease. Edited by Fewtrell L, Bartram J. London: International Water Association; 89-113.

[62] Morgan, P. (2003). Experiments using urine and humus derived from ecological toilets as a source of nutrients for growing crops. In The 3rd World Water Forum; KYOTO, JAPAN.

[63] Jensen PK, Phuc PD, Knudsen LG, Dalsgaard A, Konradsen F. (2008). Hygiene versus fertiliser: the use of human excreta in agriculture—a Vietnamese example. Int J Hyg Environ Health 211: 432-439.

[64] Jensen PKM, Phuc PD, West LGK: (2010). How do we sell the hygiene message? withdollars, dong or excreta? Environ Health 9: 27.

[65] Drechsel. P., Scott, C.A., Raschid-Sally, L., and Redwood, A. B. (2010). Wastewater irrigation and health - Assessing and mitigating risk in low-income countries. In. Sterling, VA, London: Earthscan.