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Full Length Research Article

Prevalence of *Cryptosporidium Oocysts* Among Primary School Children in Wamakko Local Government of Sokoto State, Nigeria.

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ABSTRACT: The study was aimed at determining the prevalence of cryptosporidium oocysts in faecal samples among primary school children in Wamakko local government of Sokoto State, Nigeria. A total of forty (40) samples were obtained from pupils attending different primary schools within Wamakko Local Government. The primary schools include; Kwalkwalawa Primary School, Kaurar Kimba Primary School, Fanari Primary School and Asari Primary School. A total of ten faecal samples were collected from each primary school. Faecal samples were analysed using formal diethylacetate to detect the presence of cryptosporidium oocysts. The highest percentage prevalence of occurrence (80%) of cryptosporidiosis infection was recorded among the pupils of Kaurar Kimba primary School followed by Kwalkwalawa and Fanari Primary Schools with 60% prevalence each, while the least prevalence was recorded in Asari Primary School with 50% prevalence. There was no significant variation between the 4 schools (P>0.05). Cryptosporidiosis was high in female children (65%) compared to males (60%), however there was no significant variation between the two sexes (P>0.05). Children within the age group 6-8 years old had the highest prevalence (70%) followed by age group 9-10 years 60%. The least percentage prevalence was recorded among the age group 11-12 years old (50%) (5/10). There was no significant difference between the different age groups (P > 0.05). It is interesting to note the high rate of asymptomatic cryptosporidium infection in the study area.

Keywords: Prevalence, *Cryptosporidiuum Oocysts,* Primary School, Children, Sokoto

INTRODUCTION

Crystosporidiosis is a parasitic disease caused by Cryptosporidium, a protozoan parasite in the phylum *Apicamplexa*. It affects the intestines and is typically an acute short-term infection (WHO, 2004). The disease is spread through the fecal-oral route (WHO, 2004), often through contaminated water (WHO, 2002); the main symptom is self-limiting systems. In immunocompromised individuals, such as AIDS patients, the symptoms are particularly severe and often fatal (WHO, 2004). Cryptosporidiosis was first discovered in mice in 1912 and first linked with diseases in man in 1976 (WHO, 2004). A single Species was first thought to cause disease in man but molecular diagnostic tools have enabled several different species to be identified (WHO, 2004). *Cryptosprodium hominis* is found only in humans and this, together with *Cryptosporidium parvum* (which also infects cattle), are amongst the most common species found in man (Chen *et al.,* 2002). It has emerged as an important cause of diarrheal illness worldwide particularly in young children and immunocompromised patients (Harvey *et al.,* 2007). The clinical problems associated with *Cryptosprodium* spp are recognized. Impair the ability to achieve their full potential both developmentally and socio-economically (Ryan *et al.,* 2004). According to (Chen *et al*., 2002) crytosporidial oocysts when ingested are immediately infectious at quite low doses (10 to 30 oocysts are required to produce human diseases). Oocysts attach to cells of the intestine. They become intracellular but extracytoplasmic and are resistant to treatment (Chen *et al*., 2002). The life cycle is completed in the host and large numbers of oocysts are then excreted with the potential to spread the infection (Ryan *et al.,* 2004). The oocysts are resistant to quite harsh environmental conditions and can resist chlorine levels used in water treatment (Ryan *et al.,* 2004).

Symptoms appear from two to ten days after infection with an average of 7 days, and last for up to two weeks or in some cases one month (Ryan

et al., 2004). Asymptomatic individuals (those with no symptoms) are nevertheless infective and thus can pass on the infection to others (Chen *et al.,* 2002). Even after symptoms have finally subsided an individual is still infective for some weeks (Winn *et al.,* 2006). Wamakko Local Government Area of Sokoto, Nigeria has for long suffered from *Cryptosporidium* but the problem has not been systematically studied (Winn *et al.,* 2006).The incidence of Cryptosporidiosis is becoming major epidemic in Sokoto State of northern Nigeria with estimated death of over 2.3 million children (WHO, 2006). This creates urgent attention by the World Health Organization as majority of victims are women and children (WHO, 2004). The area in question which is Wamakko Local Government Area of Sokoto has been for long suffering from menace. Therefore, this area is justifiable for the research (WHO, 2004).

MATERIALS AND METHODS

Study Area

The study area was Wamakko Local Government Area of Sokoto State, Nigeria. It has an area of 697 square kilometer and a population over 179,619. The town is located at latitude of 13.030N and longitude of 005.220E. The inhabitants are mostly fishermen and depends on kwalkwalawa river as their main source of sustenance.

Sample Collection

Questionnaires were designed to seek for information on demographic and risk factors of the disease such as source of drinking water, breast feeding practices, presence of pets, type of toilet etc. Faecal samples were obtained from children of the four primary schools within Wamakko local Government. These include; Kwalkwalawa Primary School, Kaurar Kimba Primary School, Fanari Primary School and Asari Primary School. A total of fourty faecal samples were collected using sterile plastic containers. Ten samples were collected from each Primary School. The samples were collected using sterile plastic sampling bottles. The bottles were labeled according to the age and sex of the children and then transported to the parasitology laboratory Usmanu Danfodiyo University, Sokoto for analysis.

Sample processesing

Macroscopic examinations conducted include consistency, presence or absence of mucus and blood were recorded before the microscopy of the samples. Faecal samples were emulsified using steriled rod by estimating 1g of faeces in about 4mls of 10% formal water contained in a screw-capped bottle. 3 – 4ml of 10% formol water were added, and the mixture was mixed well by shaking. The emulsified faeces were sieved. The sieved suspension was collected in a sterilised beaker.The suspensions were transferred to a centrifuge tube made up of strong glass, copolymer or polypropylene, and 3-4ml of diethyl ether were added. The content of the centrifuge tube were mixed for 15 seconds with the use of a vortex mixer (Stuart SA7 Vortex mixer Cole – parmer LTD). Centrifugation was done immediately at 3000rpm for 1 minute. With the use of sticks or stem of a plastic bulb pipette, the layer of the faecal debris was loosed from the side of the tubes and the tube was inverted to discard the ether. Faecal debris, formol water and the sediment were left (Cheesebrough, 2002).

Microscopic Examination of the Slides

The microscope (LEICA Microsystems GMBH Wetzlar-Germany) was calibrated using graticule to enable differentiation and confirmation of Cryptosporidium ocysts from other (coccidian oocysts).The prepared slides were examined microscopically for oocysts, using a low power magnification to detect the presence of the oocysts and the oil immersion objective to identify them. Specimens within oocysts that appeared small, round to oval, pink red stained bodies measuring 4–6µm,or a single deeply stained red dot were considered positive (Cheesbrough, 2002).

Staining

A smear from the sediment obtained by formol ether oocyst concentration techniques was

prepared and air-dried the smear. The smear was fixed with methanol for 2-3 minutes. Stain was made with unheated carbol fucshsin for 15 minutes and the stain was washed off with water. The stain was decolorized with 1% acid-alcohol for 10-15 seconds and washed off with water. Counter stains were made with 0.05% methylene blue for 30 seconds and washed off with water and the slides transferred to a draining rack for the smear to dry. Each smear was examined microscopically for oocysts using a low power magnification and then by the use of oil emmersion objective for identification.

Statistical analysis

All data generated were subjected to Chi-square test of association using SPSS statistical package version 2014. Differences at $P < 0.05$ were regarded as significant.

RESULTS AND DISCUSSION

A total of forty children from four primary schools in Wamakko Local Government Area of Sokoto State, Nigeria were examined for the prevalence of *Cryptosporidium* Oocysts in fecal samples. 25 (62.5%) Samples were recorded positive and 15 (37.5%) Samples were recorded negative. Figure 1 indicated the Prevalence of Cryptosporidium oocysts in different schools. The highest infection was recorded among pupils in Kaurar Kimba primary school with prevalence of 80% followed by pupils in Kwalkwalawa primary school and Fanari Primary School (with 60% each), while the least prevalence was recorded among pupils in Asari Primary School (50%) however, the difference in prevalence in all four primary school was not significant ($P > 0.05$) .

The Present study further classified pupils based on their classes of study, from primary (1 – 4) and culminate to the following findings as described in Table 3. Primary I have prevalence of (60%), primary 2 (80%), primary 3 (60%), and primary 4 (50%).

Similarly, the results of prevalence of *Cryptosporidium* oocysts with respect to gender of children are presented in Figure 2. The prevalence although higher in females (65%) compared to males (60%) were also not significantly different ($P > 0.05$). Figure 3 shows a non-significant difference ($P > 0.05$) in prevalence of *Cryptosporidium* oocysts with respect to age groups of the children. The result shows 70% prevalence for children within the age of 6-8 years. This was followed by 9-10 years (60%) while the least percentage prevalence was recorded among the age group 11-12 (50%).

The high prevalence of Cryptosporidium oocytes observed in the study may be attributed to wetness and high humidity along the river bank which can encourage oocysts survival and increase their viability when present (Gascon *et al*., 2000). Detection of *Cryptosporidium* oocysts with high prevalence in the study area call for concern because, the prevalence rate may be on the increase in the study area since*Cryptosporidium* lives in the intestine of infected humans or animals. An infected person or animal sheds Crypto parasites in the stool. Millions of Crypto germs can be released in a bowel movement from an infected human or animal Medinet. Com (2010). The prevalence of Cryptosporidium oocyst in this area could also be due to faecal contamination around the study area as described by CDCP (2010), an infected person or animal sheds Cryptosporidium parasites in the stool. Millions of Cryptosporidium germs can be released in a bowel movement from an infected human or animal. Most sporadic infections occur through person-to-person contact. Miguel M Cabada, (2017).

This can lead to spread of infection in man and animals (Gascon *et al*., 2000). Feacal droppings of animals can contaminate the environment and water bodies (Abrahamsen, 2004). Similar to the reports of Mor and Tzipori (2008), and Ayinmode *et al.* (2012) there was statistically insignificant association between the detection of Cryptosporidium oocysts and the presence of house hold pets in the present study. Contrary to the findings of (DuPont *et al*., 1995; Keusch *et al.,* 1995)., domesticated animals have been reported

as reservoir of Cryptosporidium oocysts and hence source of human infections.The prevalence obtained in the present study is in contrast with the higher prevalence reported from Zaria (Kwaga *et al.*, 1988) and other parts of Nigeria such as Jos (Ikeh *et al.,* 2007) and Imo State (Ikechukwu *et al.,* 2011). The observed disparity particularly with Kwaga *et al.* (1988) and Ikeh *et al*. (2007) studies could be attributed to small number of samples collected in their studies. The observation made from this study showed insignificant association between the detection of Cryptosporidium oocysts and the source of drinking water. Wells in this study area are mostly shallow and can easily be contaminated with human excreta which could serve as the reservoir of the oocysts. This agrees with the reports that contaminated water represents the major source of Cryptosporidium infection for humans (Gambo *et al*., 2014).

The distribution of Cryptosporidium oocysts with respect to gender was slightly higher in females than in males even though not statistically significant. This observation is consistent with earlier observation by Kwaga *et al.* (1988) in Zaria but contradicts the reports of higher prevalence in males than females by other researchers (Egberongbe *et al*., 2010; Saneian *et al*., 2010). The reason for the observed difference is not clear. Prevalence in males from this study agrees with the report of Maikai *et al*., (2009) who reported similar high prevalence in male herds from different cattle concentration, and the cattles usually intermingle with humans which can serve as the source of contamination.The highest prevalence of the oocysts in this research disagree with the findings of Gambo *et al*. (2014), highest prevalence of the oocysts were recorded among children from house hold with only cats (6%: 7/112).

The highest prevalence of infection occurred in children of age group 6 – 8 years followed by the those of age group 9-10 years with the prevalence of (60%) and finally those with in the age group between 11 – 12 years had the list prevalence (50%). These findings disagree with the findings of Gambo *et al*. (2014), although the infection is of all ages, highest prevalence of the infection occurred among children older than two years of age. It is noted from this results that incidences of the disease increase with increase in age of the patients and children of 4 years and above are at highest risk of being infected with the parasite. These findings also disagree with the findings of (Gascon *et al*., (2000) and Abrahamsen *et al*., (2004), who reported the high prevalence of infection among age group 0-5 years.The prevalence of Cryptosporidium oocysts with respect to age was statistically not significant. Although the infection affects all ages, highest prevalence of the infection occurred among children older than two years of age. This collaborates previous observations made In this regard by Egberongbe *et al.* (2010). But contradicted the reports of Saneian *et al*. (2010) that most of cryptosporidial infections occur within 0-24 months. The reason for the observed contradiction is not known.

The observation made from this study showed insignificant association between the detection of Cryptosporidium oocysts and the source of drinking water and toilets.

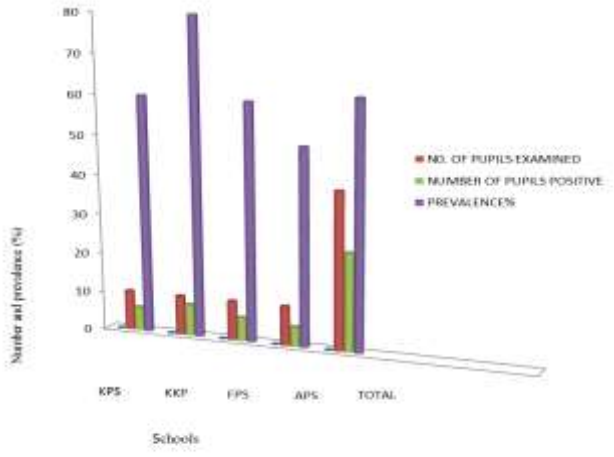

Fig 1: prevalence of cryptosporidium oocysts with respect to school of the children

Key:Kwalkwalawa Primary School (KPS), Kaurar Kimba Primary School (KKPS), Fanari Primary School (FPS), Asari Primary School (APS)

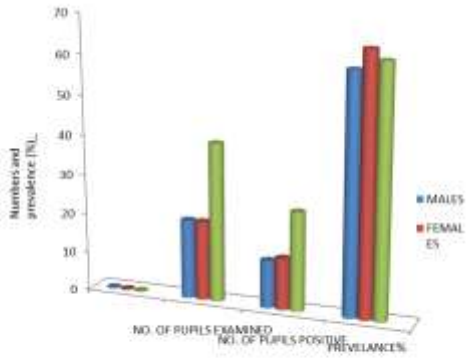

Figure 2: Prevalence of cryptosporidium oocysts with respect to gender of the children

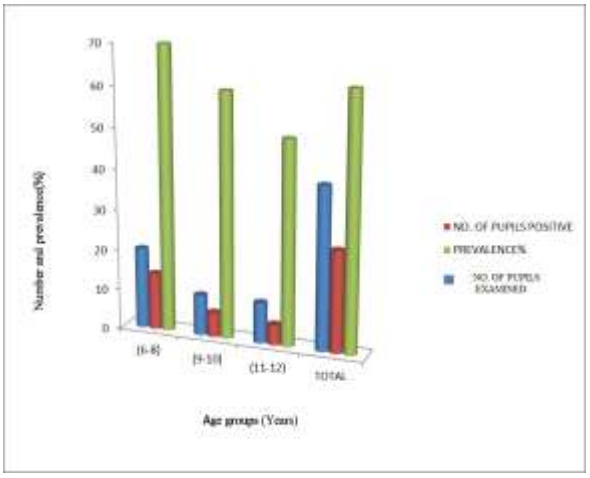

Figure 3: Prevalence of *cryptosporidium oocysts* with respect to age group of the children

CONCLUSION

Based on the result obtained from this study it was found the prevalence of *Cryptosporidium oocysts* was found higher among the pupils of Kaurar Kimba primary School, closely followed by Kwalkwalawa and Fanari Primary Schools. Asari Primary School pupil recorded the lowest prevalence. It was also observed that the prevalence was more in the female children than in males. The findings of this study also showed that prevalence of the of cryptosporidiosis decreases as the age of the children increases. High rate of asymptomatic Cryptosporidium infection was recorded in the study area.

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