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CRYPTOSPORDIOSIS IN JOS, NIGERIA

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ABSTRACT

Out of 304 stool samples submitted for routine microbiological examinations from the out-patient Department (OPD) of the Jos University Teaching Hospital (JUTH), further investigations for *Cryptosporidium* sp oocysts infection were carried out using the modified Ziehi-Nelson staining method. Oocysts of *Cryptosporidium* sp were detected in9 (2.9%) of the samples of these nine(9) positive samples 5(2.65%) of them were from children 1-10 years old. All were watery stools. Other intestinal pathogens encountered include *Entamoeba histolytica*, 30 (9.8%); Hookworm 11 (3.62%) and *Schistosoma mansoni* 5 (1.6.4%). The prevalence rate recorded for others were 1, (0.33%) for *Hymenolpsis nana Giardla lamblia* and *Ascaris lumbricoides* respectively. This pilot study has documented the occurence of *Cryptosporidium* in humans with diarrhoeal cases in Jos.

INTRODUCTION

Diarrhoeal diseases constitute major causes of childhood mortality and morbidity worldwide especially in developing countries (1). Estimates show that diarrhoeal diseases cause nearly 5 million deaths annually in children under 5 years old in developing countries. Several agents including *E histolytica, G, lamblia, Salmonella sp, Shigella sp,* and *Vibrio cholerae* have long been reported to cause gastro intestinal disease in man (2). However, other agents as *Campylobacter, Yersinia, Aeromonas, Plesiomonas* and *Cryptosporidium* have also been incriminated as causative agents of gastro-intestinal disease (3).

These later group of organisms are most often referred to as new agents of diarrhoea (4,5). Of growing importance in recent times is *Cryptosporidium*, which has been increasingly associated with HIV-related diarrhoea in some communities (4).

Until recently infections with *Cryptosporidium,* a protozoan, were considered rare in animals and in humans, they were regarded as opportunistic pathogens of immunodeficient individuals (5). However, some contemporary reports have now incriminated the protozoan in gastrointestinal infections in immunocompetent individuals (6). It has further been documented that *Cryptosporidium* in immunocompetent individuals were self limiting in contrast, infections with the protozoan in immunocompromised host have been reported to be severe, protracted and life threatening (8,9).

The prevalence of cryptosporidiosis amongst patients with diarrhoea from both developed and developing countries had been reported to be as low as between 1-4% (7). However since the advent of the HIV/AIDS pandemic, the prevalence of the infection has risen considerably in most part of the world (10). In the United States and Haiti, *Cryptosporidium* has been reported to be responsible for up to 11% of diarrhoea cases seen (9,10). Furthermore, some outbreaks of acute diarrhoea reported in day care centers in the USA and Canada have been attributed to *Cryptosporidium* infection (11) and in some of the developing countries the organism has been shown to be responsible for 13% of the reported cases of acute diarrhoea episodes (8,12). The incidence of *Cryptosporidium* infection among AIDS patients in Haiti was estimated to be 38% (14).

In Nigeria, reports of the involvement of *Cryptosporidium* in diarrhoea episodes are presently very low. Largely as a result of prevailing poor socio-economic conditions, the incidence of diarrhoea in Nigeria is on the increase (12) of disturbing concern is also the increasing incidence of HIV infections in the country (2). Since the incidence of cryptosporidiosis in other parts of the world is progressively increasing in both immunocompetent and immunocompromised individuals (5,11), it has become imperative to establish the incidence and trend of the infections in various Nigerian communities. Data from such studies will contribute to the knowledge on the dynamics of the disease in the country and help provide adequate basis for evolving sustainable cost-effective control measures. This paper presents data obtained from a pilot study which was focused on establishing the prevalence of cryptosporidiosis among patients presenting at the University Teaching Hospital, Jos, Nigeria. Other intestinal pathogens were also investigated during the study. Jos Metropolis is located in the middle-belt region of central Nigeria.

MATERIALS AND METHODS

I. STUDY POPULATION

Between November, 1994 and July, 1995 three hundred and four patients who presented at the outpatients clinic of the Teaching Hospital, Jos with symptoms of gastrointestinal infections were referred to the microbiology laboratory for routine faecal examination. The patients comprised of 160 males and 144 females and were within the age of one to fifty years. Six of the referred patients had previously been diagnosed positive for HIV antibodies. Faecal samples were collected from all three hundred and four patients and specifically screened for Cryptosporidium oocysts.

ii. SAMPLE COLLECTION AND ANALYSIS

All the faecal samples were collected in clean, dry with wide mouth glass bottles. The appearance of each stool sample was recorded and categorized as formed, loose or watery.

PARASITOLOGY

Faecal samples were screened for ova/cysts of intestinal parasites. Screening was essentially on examination of direct smears on clean slides prepared from aliquots of fresh unconcentrated faecal samples and concentration of the faecal samples using the formolether technique (13). Wet mounts of fine deposits were prepared on clean glass

slides and examined directly. All the samples above were stained by the modified Ziehl-Neelson (ZN) acid fast method (13) and then examined for *Cryptosporidium* oocyst.

BACTERIOLOGY

Loopfuls of each faecal samples was further inoculated onto plates of Deoxychocolate Agar (DCA) and Salmonella - Shigella media. Resultant colonies were subcultured and the pure isolates were identified using the criteria described by Cheesebrough (14). Screening for strains of *Campylobacter, Yersinia,* enteropathogenic *E. colic*, Rotavirus and other viruses were not undertaken, because relevant facilities for these were not available in the laboratory at the time of the study.

iii. ANALYSIS OF DATA

The Fisher exact test was used where the call number was less than 5 or where some cells were zero. The chi-square (x^2) test was used for tables.

RESULTS

Out of the 304 faecal samples examined, 9 (2.9%) were found to be positive for *Cryptosporidium,* oocysts. A further analysis of these positive cases showed that 5 (2.65%) were from patients within the age group 1-10 years followed by 2(0.66%) in the age group 21-30 years (Table 1). An equal infection rate of (0.33%) was recorded in the age group 11-20 and 41-50 years respectively. The difference between the frequency of isolation in children under 10 years was higher than for the older age groups of 10 years ($p<0.05$). Although the prevalence rate was higher in the males (3.1%) than females (2.7%), the difference in these rate was not statistically significant.

Further analysis between the positive samples collected and the nature of stool showed that all the 9 (15.2%) samples positive for *Cryptosporidium* oocysts detected were from watery stools.

Out of the 109 faecal samples during the months of the dry season, 4 (1.3%) were positive for *Cryptosporidium* oocysts but was not statisticaly different from the 5(2.5%) of the 197 faecal samples collected during the months of wet season. Out of the six patients who were positive for HIV antibodies, only 1(16.6%) was positive for *Cryptosporidium* oocysts.

The other enteropathogens detected in the faecal samples and their proportions of detection were: *Entamoeba histolytica*, (9.87%) Hookworm, (3.62%); *Schistosoma mansoni*, (1.64%) and *Salmonella sp* (0.66%) (Table 2). The prevalence rate of 0.33% was obtained for *Hymenolepis nana, Giardia lamblia* and *Ascaris lumbricoides*.

DISCUSSION

Studies from more than 100 surveys worldwide indicated that the mean prevalence in humans of *Cryptosporidium* oocysts in the general population varies between 1% and 3% in Europe and North America (15). The figure is considerably higher in developing countries where the range is from 5% in Asia to approximately 10% in other parts of Africa (15,16). The prevalence rate of 2.9 % recorded in this hospital based study is therefore considerably low.

In a study Adegbola et al (17) found cryptoporidum to be the most frequent cause of diarrhoea in children under 2 years of age with a prevalence rate of 9% among children under five years. The peak of infection during the study was 29% and 28% in the wet months of July and August. In Zaria, Kwaga et al (18)

reported a prevalence rate of 21% while Pape et al (19) observed a prevalence of 17.5% in children of 6 - 24 months age group in Haiti.

Whereas Kwaga et al (18) and many others collected their specimens only from patients with gastrointestinal symptoms, the samples in the present study were not limited to cases of gastroenteritis only, but included all stool samples submitted for routine examination.

Molback et al (21) found the prevalence of *Cryptosporidium* in children to be 15% in Guinea Bissau in cases of persistent diarrhoea. *Cryptosporidium* oocysts were seen only in 9 of 59 watery stool samples, while none in the 187 formed and the 58 loose samples. Pape et al (19) found *Cryptosporidium* oocysts only in subjects with diarrhoea.

In Brazil and the United States of America, *Cryptosporidium* has been found to be a leading parasitic cause of chronic diarrhoeal disease in patients with Acquired Immune Deficiency Syndrome (AIDS) (4). In this study one out of six AIDS patient was infected with *Cryptosporidium*.

Other Enteropathogens (Helminths, Protozoans, Bacteria and Rotavirus) have also been found to be the cause of diseases in children (8,23) and some of which has been reported in this study. *Cryptosporidium* spp is a recognized primary enteropathogen in both immunocompromised and immunocompetent persons especially children in developing countries.

Table 1: Age and sex distribution of patients positive for crytosporioum oocysts

MALES			FEMALES		
Age (yrs)	No. Screened	No. +ve (%n=160)	No. Screened	No. +ve	Total No. +ve (%) (%, n = 304)
1-10	34	3(1.88) -	32	- 2(1.38)	5(2.65)
11-20	37	- 1(0.63)	28	- -	- 1(0.33%)
21-30	39	1(0.63) -	30	1(0.69) -	2(0.66%) -
31-40	24	- -	20	- -	- -
41-50		-	34	1(0.69)	1(0.33%)
Total	**160**	**5(3.13)**	**144**	**4(2.78)**	**9(2.9%)**

Table 2: Prevalence of other enteropathogens among out patients sampled for various enteropathogens (n= 304)

PATHOGEN	NO. POSITIVE% (n = 304)
Entamoeba histolytica	30(9.87)
Hookworm	11(3.62)
Schistosoma mansoni	5(1.64)
Hymenolepis nana	1(0.33)
Giardia lamblia	1(0.33)
Ascaris lumbricoides	1(0.33)
Salmonella spp	2(0.66)
Cryptosporidium sp	9(2.96)
Total	**60(19.74)**

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