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BLASTOCYSTIS INFECTION IN PATIENTS PRESENTING WITH ABDOMINAL SYMPTOMS AT MBAGATHI DISTRICT HOSPITAL AND MAMA LUCY DISTRICT HOSPITAL
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BLASTOCYSTIS INFECTION IN PATIENTS PRESENTING WITH ABDOMINAL SYMPTOMS AT MBAGATHI DISTRICT HOSPITAL AND MAMA LUCY DISTRICT HOSPITAL

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ABSTRACT

Background: ***Blastocystis spp.*** **is the most common enteric parasite that has a global distribution with higher prevalence in developing countries. Kenya being one of the developing countries, there is need to have current epidemiological data regarding the parasite. The aim of this study was to investigate the presence and prevalence of *Blastocystis* infection in patients presenting with abdominal symptoms at the selected facilities.**

Methods: **An analytical cross-sectional study design was used to investigate the presence and association between *Blastocystis spp.* infection and profile of patients presenting with abdominal symptoms at Mbagathi and Mama Lucy Kibaki Hospitals in Nairobi. Stool samples from 249 patients were collected and analyzed at National Public Health Laboratory Services (NPHLS) for presence of *Blastocystis* infection by wet mount direct microscopy, iodine staining and formal ether sample concentration technique.**

Results: **prevalence of *Blastocystis* infection was 12%, with a higher significance in males (53%) than females (47%) (X^2 = 4.26, p=0.039, 95% C.I). There was no statistical significance (X^2 = 3.36, p= 0.5, 95% C.I) in the distribution of *Blastocystis* infection by age group. The prevalence of *Blastocystis* infection was higher in patients of low-income level. No association between eatery and the infection was found (r=-0.194, n=38 and p=0.242, 95% C.I). Conclusion: The study revealed the presence of *Blastocystis* infection in the study population and a significant association between gender, symptoms and the infection. Further studies; molecular assays should be employed in generating epidemiological data on the subtypes and clinical significance of this infection.**

INTRODUCTION

Blastocystis spp. is the most common enteric parasite found in human stool and was discovered more than 100 years ago, yet it is an organism that is poorly understood in terms of its life cycle and pathogenicity. It is a protozoan intestinal parasite belonging to the *Blastocystis spp.* genus of Stramenopiles. The organism displays diverse morphological forms and the contribution of this form in the life cycle of the parasite is poorly understood. *Blastocystis spp.* are often the most common fecal parasite and infection has been linked to human disease, most specifically chronic, watery diarrhea, bloating and abdominal pain (1). The infection is considered a zoonosis with fecal-oral route transmission (2). Although the organism does not increase mortality, it causes substantial increase in morbidity (3).

Blastocystis spp. infection has a global distribution with a higher prevalence in underdeveloped countries (4) . An increasing trend in the identification of the infection suggests that it is an emerging parasite with a worldwide distribution. Infection rates have been reported to be as high as 100% in some countries like Senegal with one or more subtypes (5). It exceeds 5% in developed countries; like in the United States of America it infected approximately 23% of the total population during the year 2000 (6).

It was identified by Forsell that 85% of Zanzibar population had intestinal parasites, with two or more parasites present in 56 %. *Blastocystis spp.* and Giardia intestinalis were the most common parasites, identified by PCR in 61% and 53 % of stool samples respectively, but no correlation between carriage of the two parasites was found (7).

The disease has been linked to outbreaks in institutions mostly schools and after a gastroenteritis outbreak of unknown etiology in the municipality of Sebastião da Grama, São Paulo, Brazil. Rebolla conducted a parasitological survey to establish the epidemiological profile of enteroparasitosis in children and staff members attending the public urban schools in operation in town. *Blastocystis spp.* was the most prevalent parasite in children (86.63%) and staff members (66.67%) which shows that the disease actually is a problem (8). *Blastocystis spp.* was found to be among the most occurring potential pathogens among rural children aged up to five years in Kakamega and Kiambu Districts (9).

Systematic surveys of research studies found that over 95% of the publications in the last 10 years identify it as a cause of illness in immune competent individuals (CDC). There is existence of asymptomatic carriers, which is common to all gastrointestinal protozoans. About 50% to 80% of the infected individuals show symptoms of the disease. With lack of extensive studies coupled with lack of exposures and knowledge, the disease becomes one of the neglected tropical diseases.

Problem statement: There are large gaps in knowledge with regard to the parasite most notably whether the organism causes disease in humans or is simply an innocent bystander. According to studies done infection with the organism has been noted to cause chronic abdominal symptoms. Since many public hospital laboratories are not equipped with modern diagnostic apparatus, the infection goes undetected. Carriage rates in developing countries across the continents have been reported to be much higher, ranging from 20% -66% and even up to 100% in a study

done in Senegal (5) According to research, 61 % of Zanzibar population tested positive for *Blastocystis* infection (10). It is assumed that Kenya is in the same category, hence there is need to investigate the presence and distribution of the organism in the population.

Justification: With limited information in respect to epidemiology, treatment options and outcomes together with poor laboratory diagnosis, clinical management becomes ineffective and hence a disadvantage to the patient. In Kenya, there is no current data and information regarding the organism. This raises questions as to whether our detection at laboratory level is inadequate to identify Blastocytosis as a public problem.

MATERIALS AND METHODS

Study Objectives:

General Objective: To investigate the presence and prevalence of *Blastocystis* infection in patients presenting with abdominal symptoms at Mama Lucy and Mbagathi District Hospitals

Specific Objectives:

- To determine the prevalence of *Blastocystis spp*. In patients presenting with abdominal symptoms at Mbagathi and Mama Lucy District hospitals
- To determine the distribution of *Blastocystis* infection among patients presenting with abdominal symptoms at Mama Lucy and Mbagathi District hospitals
- To determine the association between patient profile and *Blastocystis* infection diagnosis at Mama Lucy and Mbagathi District hospitals

Study Methodology

Study design: An analytical cross-sectional study design was adopted to investigate the association between *Blastocystis* infection and profile of patients presenting with abdominal symptoms in the two facilities.

Study population: Patients, 13 years and above presenting with abdominal symptoms at Mbagathi and Mama Lucy Kibaki District Hospitals.

Sample size determination; Sample size was determined using the Daniel formula (11) below;

$$n' = \frac{NZ^2P(1-P)}{d^2(N-1) + Z^2P(1-p)}$$

Where;

n′ = sample size with finite population correction,

N = Population size (The number of patients expected to be seen in one month with abdominal symptoms was around 300 (Hospital HIS records June 2016 – December 2016).

Z = Z statistics for a level of confidence (1.96 for 95% level of confidence)

P = estimated prevalence of *Blastocystis spp.* infection (P was set at 0.20 based on the prevalence of *Blastocystis* infection reported in a study conducted in Tanzania (7) and

d = margin of error (d=0.05).

Using the above formula, the minimum sample size calculated was 136 patients.

Sampling method: Stratification was done by facility and proportional sample size allocation applied. Patients in each stratum were recruited at the facility triage using systematic random sampling method.

Inclusion criteria:

- Patients of age 13 years and above confirmed to have abdominal symptoms by the clinician at the hospitals
- Patients willing to participate and sign an informed consent or assent form

Exclusion criteria

- Patients with abdominal pain not associated with bloating, constipation or

diarrhea for example reproductive issues, surgical complications, peptic ulcer disease.
- Patients with diarrhea associated complications like dehydration.

Sample collection and processing: Patients presenting with abdominal symptoms were recruited to the study and trained on stool sample collection and safety procedures using the SOP (Annex I). Stool samples received were subjected to macroscopic examination. The samples were preserved with 10% formalin at a ratio of 1:3, and transported to NPHLS for analysis. Microscopic examination was done on the stool samples on arrival by wet mount direct microscopy, iodine staining, and formal ether sample concentration method.

Data collection and analysis: Data on the profile of the selected patients was collected through structured questionnaires (Annex V). Data on the *Blastocystis* infection status was collected through laboratory stool sample processing. Data collected was then entered, stored, cleaned and analyzed using SPSS. Descriptive statistics was done to explore collected data. Categorical variables were then tabulated; frequencies and proportions were reported in tables and graphs (bar, pie) and plotted to show distribution and check for possible associations. Association among categorical variables and *Blastocystis spp.* diagnosis was evaluated using chi-square tests at 95% confidence level. Chi-square statistics and corresponding p-values were then reported.

Ethical consideration: Research proposal was submitted to KEMRI Scientific and Ethical Committee (SERU) for approval (Annex II). Further authorization was sought from the medical superintendent and research committees of the two facilities before data collection was conducted. Informed consent was obtained from all study participants and only consenting/assenting subjects participated in the study (Annex III and IV). All the information obtained from the respondents was treated with utmost confidentiality. All data collected was used for the study alone.

RESULTS

In this study, 249 patients were enrolled of which 127 were from Mbagathi Hospital and 120 from Mama Lucy Hospital. There were more females (58%, n=145) than males (41%, n=102). Majority of the patients were of age group 19-28 years (n=82) with the least being in age group 39-48 years (n=32).

The prevalence of *Blastocystis* infection was 12% (n=30) wherein the prevalence among males was higher (53%, n=16) than females (47%, n=14), with a statistical significance at $X^2 = 4.26$, p=0.039. Out of the 30 patients who had *Blastocystis* infection, 4 had a coinfection with *Entamoeba histolytica.* The prevalence was higher in patients from Mbagathi Hospital (17%, n=22) compared to Mama Lucy Hospital at 8% (n=9) with no statistical significance (X^2=0.164, p=0.685).

The distribution of *Blastocystis* infection by age group was; *4(13%) in 13-18* years, 10(33%) in 19-28 years, 8(27%) in 29-38 years, 4(13%) in 39-48 years and 4(13%) in 49 and above years (Figure 1). The prevalence was high in age group 19-28 years, even though this was not statistically significant (X^2 = 3.36, p= 0.5).

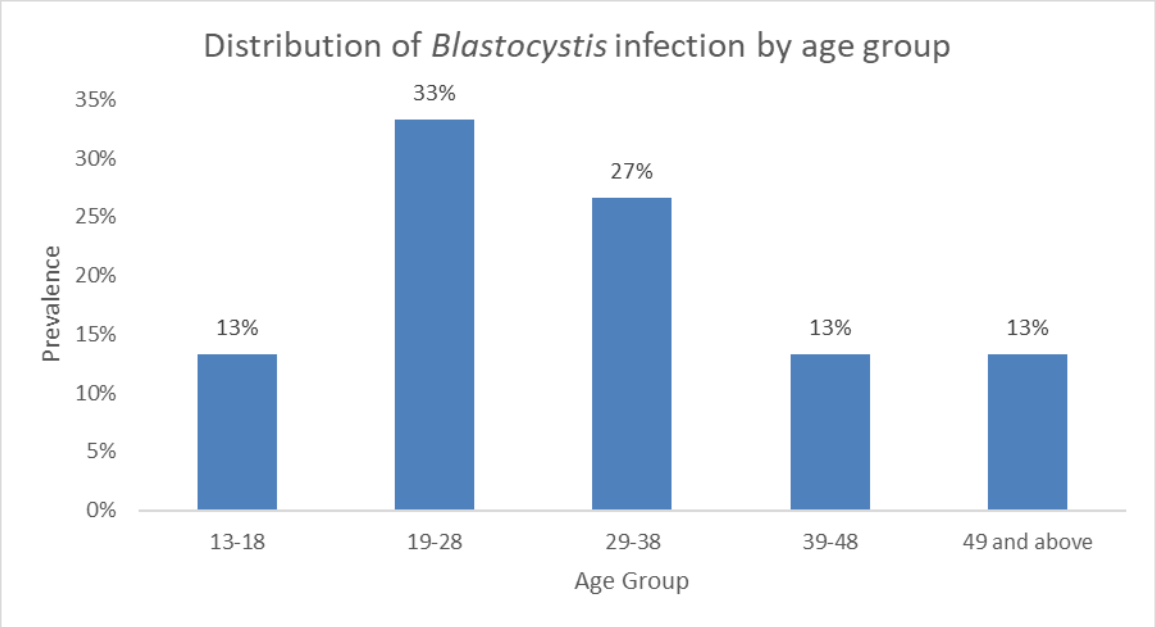

Figure 1: Distribution of Blastocystis infection by age groups

The most common clinical symptoms observed were abdominal cramps (26/30), flatulence (21/30), headache (16/30) and diarrhea (12/30). Among the 30 patients who had *Blastocystis* infection, 11 reported to eat food prepared at home and school and 9 from food prepared at home, school and street (Table 1). There was no association between eatery and prevalence of the infection (r=-0.194, n=38 and p=0.242).

Table 1

Shows place of food preparation for patients with Blastocystis spp.

Place food prepared	Prevalence
Home	17%
Home, Hotel	10%
Home, Hotel, Street	37%
Home, Street	30%
Home, Street, School	7%

DISCUSSION

In this study, the prevalence of *Blastocystis* infection was found to be 12% (n=30). The true prevalence of *Blastocystis spp.* carriage is not known as methods of diagnosis and data collection are problematic. It has been estimated that over one billion people may carry the organism (12).Conversely, in other studies, the high prevalence of *Blastocystis* infection has been reported. In Brazil prevalence in children (86.63%) and staff members (66.67%) represented a public health problem in the studied urban settings by use of Immunoassays (8).

An analysis of co-infecting parasites revealed that 13.3% of *Blastocystis spp.*-positive samples had co-infections with *Entamoeba histolytica*. A different study showed no association between a particular *Blastocystis spp.* and other parasites while an inverse correlation between age and the occurrence of other co-infections was observed (13) . An association between *Blastocystis spp.* and other parasites has already been observed but with no conclusive correlation with symptoms (10)

It was also found that males were more prone to the infection (53%) than females (47%). Outdoor activities by the adult males may also explain the significantly higher prevalence of *Blastocystis spp.* infections among these groups. Studies in Libya and China support this finding (14) (15). However a Qatar study showed that there was no difference in prevalence between males and females both having similarly high values for the prevalence (16).

The distribution of *Blastocystis* infection by age group was; *(13%)* in *13-18* years, (33%) in 19-28 years, (27%) in 29-38 years, (13%) in 39-48 years and (13%) in 49 and above years. The prevalence was high in age group 19-28 years, with not statistical significance. According to Abu- Madi, there was no difference in prevalence between the age classes in Qatar data, although there was a consistent, if marginal, drift downwards with increasing age, which was not statistically significant (16). Some earlier studies concur with that result in failing to find any age effects on the prevalence of *Blastocystis* infection. For example, no significant associations were found between infections and age classes in Libya (14). Comparatively to this study, other studies have found significantly higher infection rates in adults compared with children, with the highest prevalence rate among asymptomatic young adults aged between 18 and 30 years (15). In support of this, a recent study in Thailand has also reported a significant reduction in the *Blastocystis* infection with increasing age, the prevalence rate peaking in the younger children in the study (17). These contrasting findings suggest that the distribution of *Blastocystis spp*. infection among host populations shows spatial heterogeneity with respect to host age and sex, and these inconsistencies are most likely attributable to local factors such as the environmental conditions that influence locally the extent of, and the efficiency of, the faecal-oral route of transmission among host sectors of varying age, and between the two sexes.

The most common clinical symptoms observed were abdominal cramps (26/30), flatulence (21/30), headache (16/30) and diarrhea (12/30) amongst patients testing positive for *Blastocystis* infection. Numerous studies have examined the pathogenic potential of *Blastocystis spp*. by investigating its prevalence in symptomatic and asymptomatic groups. Libya study found a statistical association between the infection and the development of gastrointestinal symptoms. These findings are consistent with other studies (18). Majority of the patients in this study had two gastrointestinal symptoms, which is consistent with several reports among patients infected with *Blastocystis* infection in for instance Turkey (19).

Among the 30 patients who had *Blastocystis* infection, there was no association between eatery and the prevalence. Patients who had low income level had a higher prevalence (30%) compared with those of higher income level (10%). Abdulsalam's study showed that occupational status of the participants was a significant predictor of *Blastocystis* infection,

with those employed having a greater odd for infection compared to those unemployed. The high prevalence rate among employed participants may be due to high exposure to the source of infection at the work places including the food and environment. Improvement of hygienic conditions and sanitary practices with education is well documented and several previous studies have identified that the low level of education as a significant risk factor of Blastocytosis and other parasitic infections (18). In the univariate analysis done by Rebolla, children that consumed public untreated water had a higher risk of being infected by intestinal protozoa (8). Leelayoova indicated that contaminated rainwater might be a source of *Blastocystis* infection (20)

In the Qatar study, the results show clearly that there was an enormous difference in the detection of *Blastocystis spp.* in stool samples by microscopy vs. PCR methodology. Based on PCR, 71.1 % of the samples contained *Blastocystis spp.* DNA, whilst conventional microscopy gave a prevalence of just 6.9 %. This difference in sensitivity of the assays raised questions about the utility of continuing to employ conventional microscopy. On this basis, it was confidently concluded that earlier estimates of the prevalence of *Blastocystis* infection in Qatar, which were all entirely based on microscopy, were heavily underestimated (16).

CONCLUSION AND RECOMMENDATIONS

This study revealed the presence of *Blastocystis* infection in the study population and a significant association between gender, symptoms and the infection. Detection of *Blastocystis spp.* is not routinely performed in most Kenyan laboratories; hence, laboratory technicians need to be trained on its detection in clinical samples.

There is need for further research on the prevalence and distribution of *Blastocystis* infection in various regions within Kenya. Molecular assays should be employed in generating epidemiological data on the subtypes and clinical significance of this infection.

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