

INFESTATION OF *ENTAMOEBIA HISTOLYTICA* AMONG THE CHILDREN OF MIRPUR COHORT AREA, DHAKA

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Abstract

Entamoeba histolytica is one of the deadly species of protozoan parasites and is associated with pathological abnormalities in liver and large bowel in human body. To detect the prevalence of *E. histolytica*, 350 stool samples were examined by a number of diagnostic techniques. Routine microscopic examination detected 27.3% prevalence of *E. histolytica* in symptomatic stool samples and 8.6% in asymptomatic stool samples. Dipstick test detected the same prevalence (27.3%) like that of microscopy in case of symptomatic stool samples, but 10.32% was in asymptomatic samples. ELISA detected 9.09% prevalence in symptomatic samples and 3.5% in asymptomatic samples. The females were found more infected than males and the most infective group detected by different techniques was 9-10 years children.

Introduction

Bangladesh is a tropical country. Clinical amoebiasis is most prevalent in tropical and subtropical areas. It is a great public health problem in rural and urban areas with a wide spread endemicity. Low socio-economic conditions, poor hygienic habits and the most important is lack of health education allow for transmission of the amoebic infection.

Amoebiasis is a significant health problem world wide, especially in developing countries. It is presently one of the three most common causes of death from parasitic diseases. It has also been estimated that, approximately 500 million individuals are infected with *E. histolytica* each year and only about 10% experience symptomatic disease.^(1,2) An estimated 40,000-100,000 people die of invasive amoebiasis annually.⁽³⁾

The distribution of the parasite is world wide, although the preponderance of morbidity and mortality is experienced in Central and South America, Africa, and India.⁽⁴⁾ Amoebiasis, defined as asymptomatic, invasive intestinal or extraintestinal disease due to *E. histolytica* infection, is one of the most common parasitic infections world wide, Asymptomatic cyst is the most frequent manifestation of intestinal *Entamoeba* infection and 90% of *E. histolytica* infections are asymptomatic.⁽⁵⁾

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There are four species of *Entamoeba* (*E. histolytica*, *E. dispar*, *E. coli*, *E. hartmanni*) may regularly be found in human large bowel, only one of which is pathogenic. There are also a few rare species: “atypical,” “low temperature” or “Laredo” strains of *E. histolytica*, now known to be the normally free-living species which are *E. moshkovski*,⁽⁶⁾ *E. polecki*, *E. chattoni* and *E. gingivalis*.

The cysts of *E. coli* and *E. hartmanni* may be distinguished by light microscopy applying well-understood criteria from those of *E. histolytica* and *E. dispar* but the later two are indistinguishable from each other. After extensive research and argument, it is generally accepted that, *E. histolytica* actually comprises two genetically distinct but morphologically indistinguishable species. *E. dispar* has never been documented to cause colitis or liver abscess, but is responsible for many cases of asymptomatic infection.

Identification and differentiation of *E. histolytica* and *E. dispar* in stool sample by microscopy is imprecise. In most of the cases, false-positive result were found due to misidentification of macrophages and nonpathogenic species of *Entamoeba*.⁽⁷⁾

The amoeboid trophozoites can live in the intestinal crypts, feeding on intestinal contents and host tissue, and multiplying by fission. In some cases the trophozoites secrete proteolytic enzymes which destroy the intestinal epithelium allowing the trophozoite to enter the host tissue. These can form large abscesses that may allow the parasite to enter the blood stream and be carried to the liver and other organs. In these extra-intestinal sites the trophozoites also can cause extensive tissue destruction. If the intestinal tissue has been invaded the faeces can be bloody. Secondary bacterial infection can complicate an already severe pathology. Accurate diagnosis of this parasite is important to prevent unnecessary treatment of a non-pathogenic strain, and to ensure treating a pathogenic strain.

The necessity to identify and treat asymptomatic carriers of *E. histolytica* is emphasized by the observation that 10% of them develop invasive amoebiasis in due courses.⁽⁸⁾ Additionally, asymptomatic carriers are more likely to spread the disease than symptomatic persons with invasive disease, as the latter individuals seek medical attention.⁽⁹⁾

Materials and Methods

In total, 350 stool samples were collected from Mirpur area, Dhaka, Bangladesh during July - October, 2005 and the entire study was carried out in the Parasitology Laboratory, Laboratory Sciences Division (LSD) at the International Centre for Diarrhoeal Disease Research, Bangladesh (ICDDR,B) Dhaka.

Stool samples were preserved in 10% formal saline and also stored the specimens at -20°C which could not be performed within 72 hours of collection. To identify the

target parasites, microscopic examination was done by direct smear and stained preparation. Dipstick or rapid test was conducted with several supplied antigen detection kits according to the instruction of TechLab, Inc., Blacksburg, VA. The ELISA was used for the detection of *E. histolytica*. The test was performed according to manufacturer's instructions and in this test antibodies were used for the adhesin stool antigen ELISA.

Results and Discussion

During the study period, fecal specimens of 350 school children of 5 - 12 years were collected from an urban slum in Mirpur, Dhaka, Bangladesh. About 339 samples were found asymptomatic stool samples and 11 were symptomatic (diarrhoeal) in the total samples. Among the collected samples, 29 asymptomatic stool samples were microscopically positive either, with cysts or trophozoites and 3 symptomatic (Diarrhoeal) stool samples were found positive by microscopy. A total of 11 symptomatic stool samples examined during the study period. Among the examined samples, the prevalence of *E. histolytica* were 27.3% by microscopy, 9.09 and 27.3% were by ELISA and Dipstick, respectively (Table 1).

Table 1. Comparative prevalence of *E. histolytica* by microscopic examination, Dipstick and ELISA.

Types of sample	Total number of samples examined	Prevalence of <i>E. histolytica</i> by microscopic examination (%)	Prevalence of <i>E. histolytica</i> by dipstick test (%)	Prevalence of <i>E. histolytica</i> by ELISA (%)
Asymptomatic	339	8.6	10.32	3.5
Symptomatic	11	27.3	27.3	9.09
Total	350	9.14	10.85	3.71

Among 350 stool samples were examined, 171 were male and 179 were female. Out of 179 female samples, 16 were positive and the prevalence of *E. histolytica* was 9.35% by microscopy. Dipstick test detected 22 stool samples positive and prevalence was 12.29%, while ELISA confirmed 9 positive samples and prevalence was 5.02%. In total male, the prevalence was 9.35% by microscopy and Dipstick test detected 9.35% prevalence. The ELISA showed 2.33% prevalence (Table 2).

The stool samples of children were divided into four categories according to their age groups between 5 and 12 years. In both microscopic and dipstick test, the prevalence of *E. histolytica* was same (16.2%) in 9 - 10 years of children. ELISA detected 7.20% prevalence in the same group. The lower incidence found in 5-6 years, detected by three different methods (Table 3).

In asymptomatic stool samples, by microscopy examination it was 25% sensitive and 92% specific. Dipstick test showed 89% sensitivity and 96% specificity. Dipstick test was more sensitive than microscopy when compared with ELISA. Antigen detection tests have proven to be more sensitive and specific than microscopy.⁽²⁾ Among the symptomatic stool samples, microscopy was 0% sensitive and 70% specific. Dipstick showed 66% sensitivity and 87% specificity.

Table 2. Comparative prevalence of *E. histolytica* in different sex group by microscopic examination, Dipstick and ELISA.

Sex	Total number of samples	Prevalence of <i>E. histolytica</i> by microscopic examination (%)	Prevalence of <i>E. histolytica</i> by dipstick test (%)	Prevalence of <i>E. histolytica</i> by ELISA (%)
Male	171	9.35	9.35	2.33
Female	179	9.0	12.29	5.02

Table 3. Prevalence of *E. histolytica* in different age groups by microscopic examination, Dipstick and ELISA.

Age group (Years)	Total number of samples	Prevalence of <i>E. histolytica</i> by microscopic examination (%)	Prevalence of <i>E. histolytica</i> by dipstick test (%)	Prevalence of <i>E. histolytica</i> by ELISA (%)
5 - 6	50	4	8	6
7 - 8	58	6.89	10.3	5.17
9 - 10	111	16.2	16.2	7.20
11 - 12	131	9.16	13.07	6.10

In total samples, Dipstick test was 87% sensitive and 96% specific. ELISA was 9% sensitive and 96% specific. The results were comparable with the results of Weinke *et al.*⁽¹⁰⁾ They studied on 2700 German citizens returning from tropical areas and reported 0.3% prevalence of *E. histolytica* in their study and the rate was similar to that of 0.7% among under 5 Sudanese children. Braga *et al.*⁽¹¹⁾ and Sultana *et al.*⁽¹²⁾ reported that infection with *Entamoeba histolytica* increased with the growth of age and females were more likely infected than males. A study of Cross⁽¹³⁾ revealed the opposite result where infection rate was higher for males (17%) than females (11%).

Muttalib *et al.*⁽¹⁴⁾ reported 11.07% prevalence of *Entamoeba histolytica* in the students of Dhaka University. In 1988, Weinke *et al.*⁽¹⁵⁾ conducted a study which compared as patients with amoebiasis to non-amoebic diarrhoeal patients at a hospital for tropical diarrhoeal diseases in Dhaka, Bangladesh. The overall case fatality rate for the patients with amoebiasis was 29% which was significantly higher

than 11% for the non-amoebic diarrhoeal controls. The high case of fatality rate was similar (26%) as reported by Lewis and Antia,⁽¹⁶⁾ for hospitalized patients in Nigeria and 27% reported by Adams and MacLeode⁽¹⁷⁾ for South African children indicating that amoebiasis is a severe disease that carries a worse prognosis than other endemic diarrhoeal infections.

The world-wide prevalence of *E. histolytica* has been described as 14.3% and in Asia 16%, in America 12%, and in Africa 17%.⁽¹⁸⁾ The prevalence of *E. histolytica* was found to be 0.5 to 38% in Asia, 0.6 to 37% in Africa and 0 - 49% of in America.⁽¹⁹⁾ Several microscopy-based epidemiological studies in Iran have shown 2.2 to 30% of *Entamoeba* infection. Current microscopy-based studies, showed a high prevalence of *E. histolytica* in asymptomatic carriers. This was true even in the tropical areas of the south, where previous surveys showed that up to 30% of asymptomatic individuals residing in rural areas with poor sanitation were infected by *E. histolytica*/*E. dispar*.

It can be concluded from the present study that, the prevalence of *Entamoeba histolytica* detected by the three different techniques was most common in the children of Mirpur cohort area and females were detected more infected than the male children.

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