

# High Prevalence of Amoebiasis in Women Protective Home of Kanpur

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## ABSTRACT

Stool examination and serological detection of anti-Entamoeba histolytica antibodies in the serum and in faeces (coproantibody) of 70 women of Protective Home, Lucknow, revealed that 54.28 per cent were *E. histolytica* cyst passers. Serum antibody and coproantibodies were detected by indirect haemagglutination (IHA) test. High prevalence of the disease was attributed to the unsanitary conditions and unhygienic habit and habitats of volunteers. Serological diagnosis together with microscopic stool examination for *E. histolytica* in such cases in India and their significance has been discussed.

## INTRODUCTION

Diagnosis of amoebiasis patients by single stool examination under microscope is not sufficient for the estimation of correct prevalence and incidence of amoebiasis.<sup>1</sup> Stool examination should be supplemented with serological diagnosis for the detection of specific antibody in the serum and in the faeces. Preparation of axenic *E. histolytica* antigen<sup>3</sup> has helped in recent years to standardise serological tests like indirect haemagglutination (IHA)<sup>4</sup>, enzyme linked immunosorbent assay (ELISA)<sup>5</sup>, fluorescent antibody (FA)<sup>6</sup>, counter immunoelectrophoresis (CIEP)<sup>7</sup> etc. techniques for diagnosis of cases of invasive and non-invasive amoebiasis, where stool examination may be negative for *E. histolytica*. Amoebiasis has always been attributed to the unsanitary and unhygienic conditions, contaminated food and drinks, food handlers, overcrowding and unbalanced diets for men, women, and children<sup>8</sup>.

The present communication deals with a survey made in the occupants of Women Protective Home of Lucknow to ascertain the prevalence of amoebiasis in the women gathered from various parts of Uttar Pradesh, by using both microscopic examination as well as serological test with the standard amoeba-antigen, prepared in this Institute.

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## MATERIAL AND METHODS

**Number of patients :** Seventy women of age between 18 to 58 years.

### **Collection of samples**

- i) **Stool samples:** Three consecutive freshly passed faecal samples were examined microscopically by the conventional normal saline and Lugol's iodine smear preparations, for the demonstration of cysts and trophozoites of *E. histolytica*. In doubtful cases culturing of stool samples in Robinson's medium<sup>9</sup> was done.
- ii) **Preparation of stool samples for the detection of coproantibodies :** For coproantibodies detection 2 gm of fresh faecal matter was suspended in 5 ml of chilled normal saline and centrifuged at 2000 r.p.m. for 30 min. The supernatant was treated with activated charcoal and incubated at 37°C for 30 min. It was centrifuged to remove the charcoal and the clear supernatant was used as coproantibody.

**Collection of Serum Samples :** The method of collection of serum samples is the same as described by Kumar et al.<sup>10</sup> A drop of blood obtained from finger puncture was collected on filter paper strip. The strip containing 0.05 ml of blood was eluted in 0.4 ml. phosphate-buffer-saline, pH 7.2, and final serum dilution of 1:16 was obtained. The diluted serum sample was inactivated at 56°C for 30 min. Serum antibody was detected by IHA test.

**Serological test:** IHA technique used for the detection of serum and coproantibody was the same as described by Kruppi Glutaraldehyde fixed sheep RBC tagged with amoeba-antigen by tannic acid was used to see the agglutination in the presence of antibodies in the test samples. 40 µg/ml of antigen was used as the optimum concentration in this test and titre 1:128 was considered as the lowest positive reaction. Reference positive and negative serum were used as control.

## RESULTS

Smear preparation of stool samples of 70 volunteers revealed that 38 cases were *E. histolytica* cyst passers and 32 cases were negative for *E. histolytica* cysts. Out of 38 cyst passers 26.39 per cent cases were positive for serum antibody while only 13.7 per cent were positive for coproantibodies by IHA test (diagnostic titre 1 in 128). All the 32 cyst negative cases were also negative for coproantibodies where as 6.3 per cent cases showed positive reaction for serum antibody.

## DISCUSSION

WHO<sup>1</sup> has considered amoebiasis as a disease mostly confined to tropical, subtropical, temperate and colder zones of the world where proper sanitary

Microscopic Stool Examination	No. of cases	*Indirect positive	Haemagglutination Negative	Test for coproantibody Serum antibody	
				Positive	Negative
Positive for <i>E. histolytica</i> Cysts	38 (54.3)	5 (13.26)	33 (86.8)	10 (26.3)	28 (73.7)
Negative for <i>E. histolytica</i>	32 (45.7)	NIL (0.00)	32 (100.0)	2 (6.3)	30 (93.7)

\*Titre 1 in 128 was taken as diagnostic.

Figures in parantheses indicate percentage.

Conditions do not prevail. This disease has been included in the Diarrhoeal Disease Control (DDC) programme of WHO. The prevalence of the disease was found very high in the women of the Protective Home, Lucknow. The inhabitants were found to be unhygienic in their habit and habitats. They were provided with unbalanced diets that is protein deficient and the latrines and urinals used by them were dirty. All these conditions are attributed to the infection with *E. histolytica*. Overcrowding in single room was another factor observed. 54.30 per cent were found to be passing *E. histolytica* in their stools, Contaminated food and drinks were suggested as the main source of such a high incidence of the disease in these women. Stool examination under microscope and antibodies detection in the serum and stool samples by IHA test were found to be appropriate diagnostic methods for amoebiasis in endemic areas. Although smear preparation showed maximum number of cyst passers in the women, serological positivity was low in these women. Serological diagnosis is helpful for invasive amoebiasis and less effective in non invasive cases. From the results it can be concluded that microscopic faecal examination in cases of non-invasive amoebiasis (cyst passers) showed higher positive results than those by IHA test. Serological test is suitable for the diagnosis of invasive amoebiasis and for the detection of negative cases along with faecal examination. For non-invasive amoebiasis cases faecal examination together with detection of coproantibodies by IHA test serve as a good tool for the diagnosis of present status of amoebiasis cases. In 6.3 per cent cases cyst could not be detected in faeces but were found to be positive for serum antibodies. Coproantibodies are of short duration and they are found during the active phase of infection while serum antibody may persist even after the patient is clinically cured. Therefore, serum antibody detection is marginally useful for the diagnosis of present status of the disease whereas coproantibodies detection by serological test can be considered a helpful method for diagnosis of present intestinal amoebiasis cases. It is suggested that an improved technique such as multilayer micro-ELISA test should be employed in the detection of coproantigen and coproantibody for the early diagnosis of intestinal and extra-intestinal amoebiasis for timely treatment and cure of the disease in endemic areas in India.

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