

SERODIAGNOSIS OF AMOEBIC LIVER ABSCESS BY IHA METHOD

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Masood Hameed Khan, Rana Qamar, Zaman Shaikh (Department of Medicine, Medical Unit-3, Civil Hospital, Karachi.)

Abstract

Serological test based on HA (Indirect Haemagglutination Method) was performed in 100 cases of hepatic abscess. The test was 100% sensitive and 94% specific. The cut off point of antibody titer between normal population and patients with invasive amoebiasis was 1:128. Antibody titer in amoebic liver abscess was $1:5242 \pm 2795$. A significant ($P < 0.001$) correlation was found between total leucocyte count and antibody titer (JPMA 39: 262, 1989).

INTRODUCTION

Entamoeba histolytica infection is a common problem especially in third World Countries. It is very common in ill-sanitised areas, particularly in warm climate of the tropical and sub-tropical countries, due to poor socio-economic conditions, poor nutrition and hygiene. After intestinal amoebiasis liver abscess is the commonest form of invasive amoebiasis, occurring in about 83% of patients with intestinal amoebiasis¹. Often it is difficult to distinguish pyogenic from amoebic liver abscess clinically and by laboratory and imaging techniques. Serological tests are of great help in the diagnosis of invasive amoebiasis. In the present study an attempt has been made to assess the efficacy and sensitivity of serological test and anti-amoebic antibody titers in patients with invasive amoebiasis.

PATIENTS AND METHODS

Hundred patients with liver abscess were studied. Liver abscess was diagnosed clinically, on ultrasonography, aspiration and serological test for amoebiasis. Patients with liver abscess were interrogated in detail and examined thoroughly. Their Sera were stored at -4°C for indirect haemagglutination test (IHA). These patients were reexamined after three weeks for clinical, laboratory, ultrasonographic and serological parameters. Hundred aged matched healthy individuals from various socio economic classes were included in the study as controls. The controls were also subjected to similar examination and investigation and their sera were stored for estimation of antibody by using cellognost amoebiasis - kit. The reagent is available in lyophilized form, and is stable at $2-8^{\circ}\text{C}$, for upto 4 weeks after preparation. Keeping in view the short stability of the reagent, sera of these cases and controls were stored and test was performed on two consecutive days. Statistical analysis was done by Chi square and students 'C' test.

RESULTS

Out of 100 cases of liver abscess, 96 had amoebic and 4 had pyogenic liver abscess. There were 80 males and 20 females in control group with mean age of 40.44 ± 12.9 years. In amoebic liver abscess out of 96 82 (85.4%) were males and 14 (14.6%) were females. Serological test was performed in all the 96 cases of amoebic liver abscess, in which anti-amoebic antibody titers were very high. In the control group these titers were below 1:128 except in six cases in whom the titers were above threshold i.e. 1:128. In these six cases there was history of loose motions during the previous six months. Hence

the increased titers might be due to invasive intestinal amoebiasis. The mean value of anti-amoebic antibody titer in the control group, pyogenic group and amoebic group was 1 : 71 ± 17.8, 1 : 64, 1 : 5242:88 ± 2795 respectively. A significant correlation (P < 0.001) was found between total leucocyte count and antibody titer (Table).

TABLE. Correlation between TLC and anti-amoebic antibody titer N=96 P < P.001.

No. of cases	TLC (MEAN) (Neutrophil %)	Titer
6	11,700/cumm (76%)	1:1024
20	14,380 (70%)	1:2048
27	19,843 (76%)	1:4096
43	24,633 (75%)	1:8192

The sensitivity and specificity of the IHA test in the present study was 100% and 94% respectively.

DISCUSSION

It is difficult to differentiate amoebic from pyogenic liver abscess on clinical findings, laboratory investigations and imaging techniques. Though aspiration and examination of the pus is very helpful in differentiating between the two disease processes, it is not always feasible especially when the abscess involves the left lobe of the liver. In the present study, 100% of the cases with amoebic liver abscess had neutrophil leucocytosis, while 18% had two or more abscess. These two findings were once considered to be the most reliable for diagnosis of pyogenic liver abscess. Moreover, trophozoites are not easy to find in the liver "pus". Therefore, the diagnosis of Amoebic Liver Abscess cannot be made with certainty by such conventional methods. Confirmation of diagnosis requires either isolation of trophozoites from pus or by serological tests for amoebiasis. Various serological tests for invasive amoebiasis have been introduced with variable sensitivity, specificity and reproducibility. Of these IHA is the most widely used test in many centers of the world and has also been employed in the present study. It is highly sensitive non-invasive and cost effective. Attempts have been made to isolate amoebic antigen in the purified form from monoxenic culture of *Entamoeba histolytica*.²⁻⁵ These workers introduced the antigenic system of axenic *Ent. histolytica* (NIH20 strain) to characterize the immunological specificities of various isolated fractions. They reported their reproducible results of gel filtration of crude axenic extract. In all, three fractions (Fi, P11 and F11) were obtained. Antisera against each fraction and whole antigen were raised in rabbits. Serological tests employing purified antigens are positive in nearly all patients with proven amoebic liver abscess and in great majority of those with acute amoebic dysentery. They are generally negative in asymptomatic cyst passers, suggesting that tissue invasion is necessary for antibody production, which has been confirmed earlier.⁶ Failure to detect antibodies probably indicates that invasion has not taken place and this may be the best means of separating the commensals from the invasive infection in individuals passing cysts. Thus it is

also evident that serology, particularly in the endemic area is of more value in excluding the diagnosis than in confirming it, since the persistence of significant antibody titer for months after complete cure. Many workers throughout the world have done research in this particular field and confirmed the efficacy of serodiagnosis in invasive amoebiasis.⁷⁻¹⁴ Hence, considering the sensitivity, specificity and cost effectiveness of serodiagnosis of amoebiasis by IHA, we recommend its routine use and further recommend that the cut off point should be taken as 1: 128 for the test.

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