DETECTION OF ANTI-TOXOPLASMA ANTIBODIES AMONG PATIENTS WITH ACUTE LEUKEMIA OR LYMPHOMA USING LATEX AGGLUTINATION AND ELISA TESTS

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ABSTRACT

Objective: to study the rate of infections of toxoplasmosis as an opportunistic infection among two groups of adult patients in Mosul city (Iraq) and surrounding areas. The first group was diagnosed before receiving any kind of anti-cancer therapy (BRT) and the second group after receiving their anti-cancer therapy (ART).

Methods: Latex Agglutination (LA) test for detection of IgG antibodies and Enzyme Linked Immunosorbent Assay (ELIZA) test for detection of IgM antibodies were used. Chi-square test (X^2) was used for statistical analysis.

Results: Out of (90) samples tested of the (BRT) group, 43(48%) were positive for anti-Toxoplasma IgG antibodies by LA test. Out of (70) samples tested of the (ART) group, 25(36%) were positive for anti-Toxoplasma IgG antibodies using LA test. Statistically, there was no significant difference between the two groups. The study revealed that patients in both groups showed seropositive results using LA test, but sera of patients examined by ELISA test revealed that 8(19%) and 7(28%) were positive for (BRT) and (ART) groups respectively. Statistically, there was no significant difference between the two groups.

Conclusions: 1. Patients with leukemia or lymphoma before and after receiving therapy showed great susceptibility to be infected with Toxoplasma gondii. 2. In both groups, detection of toxoplasma antibodies was higher among patients using (LA) test (48%) than that of ELISA test (19%) in (BRT) group, while in (ART) group (36%) were positive in (LA) test and (28%) in ELISA test. 3. Detection of antibodies of toxoplasmosis was higher among those who received both regimens of anti-cancer therapy (chemotherapy and radiotherapy) than those who receiving single type of therapy.

INTRODUCTION

Toxoplasmosis is a zoonotic disease, distributed all over the world.[1] It is caused by an obligate intracellular protozoan parasite named as *Toxoplasma gondii*.[2] This opportunistic parasite has a high prevalence rate in populated groups and is capable of living in multiple tissues for life in its host.[3] Nearly one third of humanity has been exposed to this parasite during their lives.[4] Various mammals and birds can be infected with *Toxoplasma gondii* but only cats appear to harbor the reproductive forms of the organism.[5] At present toxoplasmosis is becoming an increasing problem because of AIDS, and wider use of immunosuppressive drugs, for example by organ transplant patients, and also toxoplasmosis is a threat to the fetus and unborn infant, even in healthy persons without immune suppression.[6] The socioeconomic impact of toxoplasmosis in human suffering and the cost of care of sick children, especially those with mental retardation and blindness are enormous.[7] The parasite is also of economic importance in veterinary public health, since it is the major cause of animal abortions.[8] *Toxoplasma gondii* is the most common cause of infectious chorioretinitis in immunocompetent persons and it is also an important cause of ocular disease in immunocompromised patients where most cases are believed to result from local reactivation of congenital infection[9] which can lead to blindness in infected eye.[10] The clinical manifestations of toxoplasmosis in immunocompromised patients are similar to those occurring in immunocompetent persons but in more fulminating and disseminated pattern.[11] Toxoplasmosis is the most common opportunistic infection of the central nervous system in patients with acquired immunodeficiency syndrome (AIDS).[12] Disease can result from a newly acquired acute infection, but is more often due to reactivation of a latent infection when the patient's CD4 T-cell count drops to less than 100 cells /µl.[13] To the best of our knowledge this study is the first trial to be carried out in Mosul city and surrounding areas to detect the seroprevalence of toxoplasmosis in patients with leukemia or lymphoma.

MATERIALS AND METHODS

Study Population: This study was conducted in Ibn-Sina Teaching Hospital and Hospital of nuclear medicine in Mosul city involves two groups of adult patients, the first group, 90 patients newly diagnosed as leukemia or lymphoma. The second group, 70 patients were

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also diagnosed as leukemia or lymphoma but they received anti-cancer therapy (cytotoxic drugs or/and radiation therapy). The behavior of these variables was analyzed with the X^2 (Chi-square) test, using the presence of toxoplasmosis as a dependent variable. The significance level at 5% was used. The Odds Ratio (OR) for the risk factors to which the patients were exposed was calculated for each risk factor and for age group. The method of statistical analysis adapted was taken from Armitage. [14]

Serological tests:

Patient's serum was examined using LA test to detect the presence of anti-*Toxoplasma* IgG antibodies. Each patient with positive LA test, also was examined by ELISA test to detect the presence of anti-*Toxoplasma* IgM antibodies. The kits for both serological tests were received from (BIOKIT, S.A) Spain. The choice of these two kits was considered appropriate for detection of specific antibodies.

RESULTS

The results revealed that out of 90 patients (BRT) group, 43(48%) were positive by using LA test, and out of 70 patients (ART) group, 25(36%) were positive by LAT. Statistically there was no significant difference between the two groups (Table-1).

Table 1. ***Percentage of Toxoplasma IgG antibodies among patients (ART) and (BRT) groups using LA test.***

C.L %	OR*	P	LAT -ve No. (%)	LAT +ve No. (%)	Patients No.
3.1228-0.867 1.151-0.319	0.1.646 607	0.126 (N.S)	47 (52) 45 (64)	43 (48) 25 (36)	BRT (90) ART(70)

The results also revealed that out of 43 patients of (BRT) group who are positive for LA test, 8(19%) showed positivity by ELISA-IgM, and out of 25 patients of (ART) group who were positive for LA test, 7(28%) showed positivity by ELISA-IgM. Statistically there was no significant difference between the two groups (Table-2).

Table 2. ***Percentage of Toxoplasma IgM antibodies among patients (ART) and (BRT) groups using ELISA test.***

C.L %	OR	P	ELISA -ve No. (%)	ELISA +ve No. (%)	LAT +ve Patients
3.0406-0.183 5.441-0.531	0.587 1.701	0.368 (N.S)	35 (81) 18 (72)	8 (19) 7 (28)	BRT(43) ART(25)

DISCUSSION

In this study we attempt to, the patients with leukemia or lymphoma before and after receiving therapy showed great susceptibility to be infected with *Toxoplasma gondi* because immunocompromised patients, with cell mediated immunity deficiency who are at risk for severe and life threatening *Toxoplasma gondii* infection, so early diagnosis and proper treatment of the patient may reduce the complications of infection and promote better prognosis. Our study showed that, the rate of infection with *Toxoplasma gondii* was 48% for IgG but this rate is of no significant value. This finding is confirmed by the results of other workers[15] who reported 40% rate of infection in leukemic patients with no significant value as well. Other workers[16] reported a rate of infection of only 8.2% for IgG in children with leukemia or lymphoma in Egypt, such variation between these results and our results could be

due to the differences in location and age of the patients examined. Our results revealed that the rate of infection in (ART) group was (36%) for IgG which is of no significant value. This result is confirmed by other workers[15,17] who reported almost the same rates of infection (36%) and (33.33%) respectively among patients with various types of malignancies under immunosuppressive therapy. The rate of infection using LA test was higher in patients (BRT) group as compared with (ART) group but with no significant difference between them. The lower rate of infection in (ART) group which compared to (BRT) group may be explained by estimating the O.R which was found to be less than one (0.607) in patients (ART) group, which means that the exposure to anti-cancer therapy is negatively related to the infection (protection) as indicated by Armitage.[14] This may be explained by the fact that anti-cancer drugs which were used in treatment regimens are immunosuppressant cytotoxic drugs like doxorubicin and daunorubicin. Others are antimetabolites like methotrexate, cytosine arabinoside and mercaptopurine, alkylating agents have been like cyclophosphamide, chlorambucil. All these drugs could exert cytotoxic effect through interfering with DNA synthesis leading to cell death.[18-20] We thought that *Toxoplasma gondii* proliferation might be inhibited by the action of these drugs through blocking DNA synthesis of the parasite itself and resulted in lower rate of infection. This was indicated by other workers.[18-20] These drugs have many side effects on the gastrointestinal tract like loss of appetite, nausea, vomiting, diarrhoea, mucosal inflammation (mucositis) and impaired absorption. Other investigators[21] reported that such side effects can lead to malnutrition, where malnourished people have been shown to have numerous defects in their ability to mount a protective immune response. The rates of infection of toxoplasmosis with LA test-IgG (48% and 36%) was found rather higher for (BRT) and (ART) groups respectively; as compared to the a lower rates of infection with ELISA-IgM (19% and 28%) for (BRT) and (ART) groups respectively. These findings are in accordance with other workers[16,17,22] who reported nearly similar rates of infection. The present results are compatible with the results reported in a study carried out to investigate[23] who investigated toxoplasma seroprevalence among blood doners and reported (52% for IgG and only 4% for IgM), this may be explained by the fact that toxoplasma IgM antibodies decline after three months of their appearance and rarely up to one year, while toxoplasma IgG antibodies persist at low titer for life.[24,25] The study is of medical importance as it deals with immunocompromised patients, notably those with cell mediated immunity deficiency who are at risk for severe and life threatening *Toxoplasma gondii* infection, is early diagnosis and proper treatment of the patient may reduce the complications of infection and promote better prognosis.

Acknowledgements:
The authors are deeply indebted to Dr. Khalid NM. Al-Khero Medical consultant and Dr. Kahtan Radwan Tumor therapy consultant in Mosul for help in dealing properly with malignancy affected patients.

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