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Anti-Toxoplasma gondii antibodies in haemodialysis patients in Al Gezira state, Sudan

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Abstract:

Background: Toxoplasmosis is caused by protozoan parasites of the genus Toxoplasma which belongs to Apicomplexa phylum which includes important pathogens of human and animals. It is an obligate intracellular protozoan of worldwide distribution. This study aimed to investigate the prevalence of anti-T. gondii antibodies in haemodialysis patients with chronic renal failure.

Methodology: This is analytical cross-sectional study was carried out at renal dialysis unit at Al Gezira hospital for renal diseases in Al Gezira state from October 2010 – December 2010, the participants were 150 haemodialysis patients and 50 healthy controls, and the test used is latex agglutination test.

Results: Anti- T. gondii antibodies positivity was found in 110 (73.3%) of the 150 haemodialysis patients and 9 (18%) of the 50 control subjects. The difference between the two results was statistically significant (p < 0.05). In addition, an increase of the seropositivity rate was detected with increasing length of time on haemodialysis treatment, indicating a statistically significant difference (p < 0.05).

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Conclusion: This survey confirms a high prevalence of Toxoplasma infection in haemodialysis patients and those patients are a risk group for toxoplasma infection. In conclusion it is recommended that haemodialysis patients should be screened for toxoplasmosis to exclude them or dialyse them on separate machines.

Key words: Toxoplasmosis, haemodialysis, latex agglutination test

INTRODUCTION:

Toxoplasmosis, an ubiquitous protozoal disease caused by *Toxoplasma gondii*, is one of the most common parasitic infections of man and other warm – blooded animals, with definite hosts being felines.⁽¹⁾ Approximately one – third of the world's population is infected by toxoplasmosis.⁽²⁾ Serological studies show a considerable variation in the prevalence of toxoplasma infection from 0-95 % in different parts of the world and indeed between different population groups within the same country.⁽³⁾

Human infection occurs after ingestion of cysts from undercooked and contaminated meat. Alternatively, humans may ingest oocyst from contaminated water, soil and vegetables. After ingestion, gastric juices disrupt the outer wall of the cysts, releasing the infective forms. These forms reach the lymphatic system and blood circulation by dissemination through intestinal lumen cells. *T. gondii* invades all nucleated cells and tissues. Therefore, systemic involvement is frequent.⁽⁴⁾

Most infections in humans are asymptomatic, but at times the parasites can produce devastating disease. The socioeconomic impact of toxoplasmosis in human suffering and the cast of care of risk children, especially those with mental retardation and blindness are enormous.⁽⁵⁾

Toxoplasma gondii is the most frequent protozoan causing opportunistic infections in immunocompromised individuals.⁽⁶⁾ Chronic renal failure patients are under risk from a variety of infections.⁽⁷⁾ However a high percentage of positivity for *T. gondii* antibodies have been detected in these patients ⁽⁸⁾ due to their depressed immune status.⁽⁹⁾

Immunocompromised hosts, especially those with impaired cellular immunity, are at risk of recrudescence of chronic infection and dissemination, with the occurrence of fulminating disease. Patients with neoplasia, collagen tissue disease, transplant recipients under immunosuppressive therapy or haemodialysis patients with chronic renal failure have deficient cellular immunity and this makes them susceptible to the infection.⁽⁸⁾

MATERIALS AND METHOD

Study area and study design

This is descriptive cross-sectional study was conducted at renal dialysis unit at Al Gezira hospital for renal diseases in Al Gezira state, Sudan, from October 2010 – December 2010.

Sample size

150 samples were collected from patients with chronic renal failure undergoing haemodialysis ,as cases, and 50 samples from healthy volunteers ,as controls.

Serological test

A rapid slide test for the determination of anti toxoplasma antibodies in human serum:

TOXO Direct Latex is a rapid slide agglutination procedure, developed for the direct detection of antibodies anti-Toxoplasma in human serum.

The assay is performed by testing a suspension of latex particles coated with antigenic extract of *Toxoplasma gondii* against unknown samples.

The presence or absence of a visible agglutination indicates the presence or absence of anti-toxoplasma antibodies in the sample tested.

Toxo- latex, Slid agglutination, (SPIREACT ctra. Santa Coloma, Spain)

Sample collection

Fresh serum or stored at 2-8°C but not longer than 48 h. It is necessary to freeze the sample when the assay is to be carried out after this period of time. Frozen samples should be totally thawed and brought to room temperature before testing.

Procedure

1. Bring reagents and serum samples to room temperature.

2. Place one drop (50 $\mu l)$ of the sample onto a slide black area.

3. Add one drop of positive control and one drop of negative control in separate circles.

4. Resuspend the antigen vial gently before using and add one drop next to the sample to be tested, one drop next to the negative and one drop next to the positive.

5. Mix the drops with a stirrer, spreading them over the entire surface of the circle. Use different stirrers for each sample.

6. Rotate the slide slowly by means of a mechanical rotator (80-100 r.p.m.) for a period of **4 minutes**. False positive result could appear if the test is read later than four minutes.

7. Observe immediately the presence or absence of agglutination.

Reading test results: (Figure 1), (Figure 2)

1. <u>Positive:</u>

The agglutination appears on the circle.

2. Negative:

No agglutination appears on the circle.



Figure (1): Reading of latex agglutination test result



Figure (2): Positive and negative reaction

Data management and analysis

All data was entered into a spread sheet and cleaned. Data analyses was carried out by using of Statistical Package for Social Science (SPSS) version 16, for the relationship between (age,Gender and duration of dialysis) with toxoplasmosis.

Ethical considerations

Ethical clearance obtained from the Ethical Committee Board of the University of Medical Sciences & Technology. An eligible clinical suspect was duly informed about the objectives and procedures of the study. In case he/she agrees to participate, a consent form is signed, before the patient is enrolled in the study.

RESULTS

In the present study 150 patients were enrolled out of whom there were 101 males and 49 females. Table (1), Figure (3).

Gender	Frequency	Percent			
Male	101	67.3 %			
Female	49	32.7 %			
Total	150	100 %			

Table (1): Gender distribution of participants under study



Figure (3): Gender distribution in study population.

Aged between 20 and 80 years and the length of time on haemodialysis treatment was from 6 - 132 months. In the control group, from 50 healthy volunteers there were 32 males and 18 females aged between 20 and 80.

In the present study, 110 of 150 (73.3%) cases among participants were found to be positive for *Toxoplasma gondii* antibodies. Table (2), (Figure (4)

Table 2: Prevalence of toxoplasma antibodies among examined participants

	No. of cases	Percent (%)
Positive	110	73.3 %
Negative	40	26.7 %
Total	150	100 %



Figure (4): Frequency of toxoplasma seropositivity among patients group

9 of 50 (18%) healthy volunteers (control group) were found to be positive for *Toxoplasma gondii* antibodies. The percentage of people who were anti-*T. gondii* antibodies positive in the haemodialysis patients group was found to be significantly greater than in healthy volunteers (P<0.05).

From 101 males 74 (73.2 %) were found positive for T. gondii antibodies, and from 49 females 36 (73.4 %) were found positive for T. gondii antibodies. The results showed no sex difference in positivity rate for anti-T. gondii antibodies in groups. The age group (> 70 years old) had the highest

positivity rate for anti-Toxoplasma antibodies in comparison to the other age groups. Table (3), (Figure (5).

		Total	Toxoplasma Abs		Percentage
			(-ve)	(+ve)	of positivity
Age groups	21-30 years	11	6	5	45.4%
	31-40 years	36	12	24	67.5%
	41-50 years	47	11	36	76%
	51-60 years	28	6	22	78.6%
	61-70 years	16	3	13	81.2%
	>70 years	12	2	10	83.3%
Total		150	40	110	

Table (3): Frequency of positive cases according to age groups



Figure (5): Frequency of positive cases according to age groups

We observed also that the seropositivity rate increased with the increasing length of time on haemodialysis treatment, indicating a statistically significant difference (p<0.05). Table (4), Figure (6).

Table	4:	Distribution	of	participants	according	to	duration	of
haemodialysis per years								

			Toxoplasma Abs	Total	
			(-ve)	(+ve)	
	of	>1 years	6 (50 %)	6 (50 %)	12
haeamodialysis		1-5 years	23(38.3 %)	37 (61 %)	60
		6-10 years	11(14.3 %)	66 (85.7 %)	77
		11-15 years	0 (0 %)	1 (100 %)	1
Total			40	110	150





DISCUSSION

Toxoplasmosis is an opportunistic protozoan parasite infection, it can be found in humans and animals and emerges as a life-threatening risk in immunocompromised individuals.⁽¹⁰⁾

Uremic patients are affected with suppressed cellular and humoral immune responses.⁽¹¹⁾

It has been suggested that because of reduced circulating T-cells and increased suppressor cells, then haemodialysis cannot return the impairment of the immune status in CRF.⁽¹²⁾

These factors probably contribute to the acquired immune suppression in uremia and the high incidence of

infection among dialysis patient, in that infection is very common and the major cause of death, of end stage renal diseases.⁽¹³⁾

The present study revealed a higher percentage of anti-Toxoplasma antibodies positivity using latex agglutination test in chronic renal failure patients undergoing haemodialsis (73.3%) than in the healthy controls (18%) with a statistical significance (p<0.05).

The statistically significant differences between case and control groups was similar to other studies (Yazar *et al* ⁽⁸⁾; Abbas *et al* ⁽¹⁴⁾; Ocak *et al*. ⁽¹⁵⁾; Kavous ⁽¹⁶⁾ and Aufy SM *et al* ⁽¹⁷⁾; which were conducted by ELISA technique. Although the prevalence of anti-Toxoplasma antibodies in the present study was higher than the results of Abbas *et al* ⁽¹⁴⁾; Ocak *et al*.⁽¹⁵⁾; Kavous ⁽¹⁶⁾ and Aufy SM⁽¹⁷⁾. These differences may be due to prevalence of Toxoplasma infection in different population in different countries.

There was positive significant correlation between duration of hemodialysis treatment and seropositivity rate of Toxoplasma such correlation as shown by Ocak *et al.*⁽¹⁵⁾ in Turkey Abbas *et al.*⁽¹⁴⁾ in Egypt and Aufy SM ⁽¹⁷⁾ in Egypt.

The results showed no sex difference in positivity rate for anti-Toxoplasma antibodies in groups such result was shown by Mahgoub AM.⁽¹⁸⁾

The age groups (> 70) had the highest positivity for anti-Toxoplasma antibodies in comparison to the other age groups and this differ from results which were shown by Mahgoub AM.⁽¹⁸⁾

CONCLUSIONS & RECOMMENDATIONS

These results confirm a high prevalence of toxoplasma infection in haemodialysis patients. These patients are risk group for toxoplasma infection. Moreover, it is recommended that

haemodialysis patients who are susceptible to toxoplasma infections should be identified by T. gondii serological tests. Therefore, patients undergoing haemodialysis should be screened for toxoplasma before dialysis to prevent the dissemination of this infection through the haemodialysis procedure. Clinicians should be more alert with these patients and parasitological surveys of them should be periodically carried out to prevent the risk of severe toxoplasmosis. Also we can use other techniques for diagnosis to identify T.

gondii antibodies to compare the results, specificity and sensitivity.

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