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The association between *Toxoplasma gondii* seropositive status and diabetes mellitus in obese and non-obese subjects in Baghdad

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Abstract

Anti-*Toxoplasma gondii* seropositive status among obese and diabetic patients has recently attracted substantial attention. The objective of this study was to assess the seropositive rate of *T. gondii* and its relation to high body mass index (BMI), diabetes, and metabolic syndrome among participants (n=100) who attended Al-Kadhimiya Teaching Hospital, Baghdad-Iraq. An observational analytical study was conducted from October 2019 to March 2020. Participants were divided into three groups based on their BMI; obese (n=38), overweight (n=32) and normal (n=30). In addition, they were divided into diabetic (n=45) and non-diabetic (n=55) based on clinical examination, laboratory examination, and medical interview. Another classification was considered: Obese-diabetic (n=24), obese- non-diabetic (n=14), overweight-diabetic (n=13), overweight non-diabetic (n=19), normal-diabetic (n=8) and normal-nondiabetic (n=28). Finally, participants were divided into metabolic syndrome-positive (n=64) and metabolic syndrome negative (n=36). Serum samples were taken from all participants and examined for the detection of anti-*T.gondii* IgG and IgM antibodies. The anti *T.gondii* IgG positive rate was higher in the "overweight" compared to the "obese" and "normal BMI" groups. No significant differences (P=0.22) in seropositive rate were indicated among groups. The results also showed that there was no significant difference (P=0.84) in anti-*T. gondii* IgG positive rate between diabetic and non-diabetic patients. While the anti *T.gondii* IgG was significantly (P=0.03) higher in patients with metabolic syndrome as opposed to those with no metabolic syndrome. The results also showed that normal-diabetic and overweigh-diabetic patients had the highest anti-*T.gondii* IgG positive rate, although no significant differences were noticed among groups. Some other parameters were also examined for the participants, including abdominal obesity, cholesterol, triglycerides, high density lipoprotein (HDL), very low density lipoprotein (VLDL), glucose and glycated haemoglobin. Significant differences were noticed only for abdominal obesity and HDL (P<0.05) between anti *T.gondii* IgG positive cases and anti *T.gondii* IgG negative cases. Other factors did not show significant differences between these two groups. Finally, this study showed that *T. gondii* seropositive status played a significant role in changing only HDL level while other parameters of lipid profile were not influenced by *T.gondii* seropositivity among obese, diabetic patients and metabolic syndrome patients.

Keywords: diabetes, metabolic syndrome, obesity, overweight, Toxoplasmosis

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العلاقة بين ايجابية فحص طفيلي المقوسات الكوندية و داء السكري لدى الاشخاص البدينين وغير البدينين في بغداد

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الخلاصة

زاد الاهتمام مؤخرا بالعلاقة بين الفحص الموجب للاضداد المناعية المضادة للمقوسات الكوندية وكل من السمنه و داء السكري. الهدف من هذا البحث هو تقييم نسبة الاستجابة المصلية الموجبة لداء المقوسات الكوندية و علاقتها بكل من ارتفاع مؤشر الكتلة الحيوية, داء السكري و متلازمة التمثيل الغذائي ضمن مجموعة من المشاركين (العدد=100) والذين راجعوا مستشفى الكاظمية التعليمي, بغداد–العراق. اجريت الدراسة ضمن الفترة ما بين تشرين الاول من العام 2019 الى اذار من العام 2020. تم تقسيم المشاركين في الدراسة الى: مجموعة مرضى السمنه,(العدد=38) مجموعة مرضى زيادة الوزن (العدد=35) ومجموعة ذوي مؤشر الكتلة الحيوية الطبيعي (العدد=30). كما تم تقسيمهم الى: مرضى السكري (العدد=45) و مرضى غير المصابين بالسكري (العدد=55) اعتمادا على بعض الفحوصات السريرية و التحليلات المختبرية. كذلك أعتمد على تقسيم على آخر وهو : مرضى السمنه و السكري (العدد=24), مرضى السمنه –بدون السكري(العدد=14), مرضى زيادة الوزن والسكري (العدد=13), مرضى زيادة الوزن–بدون السكري(العدد=19), مرضى السكري مع مؤشر طبيعي للكتلة الحيوية (العدد=8) و اشخاص غير مصابين بالسكري مع مؤشر طبيعي للكتلة الحيوية (العدد=28). واخيرا تم تقسيمهم اعتمادا على بعض المؤشرات الى: المرضى الموجبين لمتلازمه التمثيل الغذائي (العدد=64) و المرضى السالبين لمتلازمه التمثيل الغذائي(العدد=36). جُمعت عينات مصل الدم منهم جميعا وفحصت للتحري عن الاضداد المناعية المضادة للمقوسات الكوندية نوع IgG و IgM. اظهرت النتائج ان معدل الفحص الموجب عالياً لدى مجموعة زيادة الوزن مقارنةً بمجموعه السمنه و مجموعة مؤشر الكتلة الحيوية الطبيعي, رغم عدم تسجيل اي فرق معنوي بينهم (P=0.22). اظهرت النتائج ايضاً عدم وجود فروق معنوية (P=0.84) في ايجابية الفحص المصلي بين مرضى السكري و المرضى غير المصابين بالسكري, الا ان الفرق كان معنويا (P=0.03) بين الموجبين و السالبين لمتلازمة التمثيل الغذائي. اشارت النتائج الى ان مرضى السكري من ذوي مؤشر الكتلة الحيوية الطبيعي ومرضى زيادة الوزن والسكري كان لديهم اعلى المعدلات من الاضداد المناعية المضادة للمقوسات الكوندية نوع IgG على الرغم من عدم تسجيل اي فرق معنوي بين المجموعات. اظهرت النتائج فروقاً معنوية (P<0.05) لكل من السمنه في منطقة البطن و البروتينات الدهنية عالية الكثافة بين الموجبين و السالبين لفحص داء المقوسات الكوندية بينما لم تظهر المؤشرات الاخرى اي فروق معنوية. اظهر الموجبين للفحص المصلي لداء المقوسات الكوندية مستويات عالية من السمنة في منطقة البطن مقارنه بالسالبين للفحص المصلي الذين اظهروا معدلات واطئة من السمنه في منطقة البطن . كما ان معدلات البروتين عالي الكثافة كانت مرتفعة لدى الموجبين للفحص المصلي لداء المقوسات الكونديهالكوندية مقارنة بمستويات واطئة لدى السالبين للفحص المصلي لداء المقوسات الكوندية. بينما لم تظهر المؤشرات الاخرى اي فروق معنويه بين المجموعتين. وختاما, فأن هذا الدراسة تشير الى ان الفحص الموجب لداء المقوسات الكوندية يمكن ان يؤثر على فقط على البروتينات الدهنية عالية الكثافة من دون تأثير واضح على العوامل الاحرى في مرضى السمنة و مرضى السكري والمرضى الموجبين لمتلازمة التمثيل الغذائي.

Introduction

Obesity is defined as an excess accumulation of adipose tissue to a level that may damage health [1]. Obesity is considered to be a serious health problem worldwide [2], because of its association with various diseases such as atherosclerosis, cancer, and diabetes mellitus [3]. The prevalence of obesity

has increased globally mainly in the recent few years. For instance, the prevalence of obesity in the United State is about 20%–25%, while in Europe it reaches 10%–25% [4]. In Iraq, data on obesity is limited and not representative of the community [5], although some investigators have shown that the prevalence of overweight and obesity is 55.1% and 43.7% in Basrah and Baghdad, respectively [5, 6]. The physiological mechanisms of obesity include the nutritional status, environmental conditions, and genetic background [7]. Obesity is associated with diabetes; many studies have illustrated that obesity is central to increasing occurrence of both type 1 and type 2 diabetes [8, 9]. On other hand, obesity and diabetes have been previously identified as risk factors for some infections [10]. Toxoplasmosis is among the infections that are probably correlated with obesity and diabetes [11]. Toxoplasmosis is a significant foodborne disease that can cause morbidity and mortality. The global prevalence of *T. gondii* is estimated to be between 30% and 65% [12]. Toxoplasmosis is an infectious and inflammatory syndrome linked with the poverty, rural areas as well as urban areas, while most cases are asymptomatic [13]. Both Centre of Disease Control and prevention (CDC) and National institute of Health (NIH) classified *Toxoplasma* to be among category B infections, which once infected the organisms reside in some tissues for the host's lifelong waiting for reactivation [14].

Some recent studies have shown that Toxoplasmosis patients may be more at risk to develop diabetes than uninfected individuals. Insulin may stimulate the reproduction of *T.gondii* [15]. In addition, some cases of diabetes were described with altered neurohormonal regulation in patients who are chronically infected with toxoplasmosis [16]. Moreover, *T.gondii* infection may be related to obesity or alteration of inflammatory fat distribution as organisms alter and reside in fatty tissues [17].

The association between *T. gondii* infection and both obesity and diabetes is currently being evaluated [1, 3, 12]. Significant findings exist relating Toxoplasmosis to both obesity and diabetes, whereas there is limited information about the correlation between toxoplasmosis and obesity in Iraq. It is important to recognize the connection of *T. gondii* infection to both obesity and diabetes in Iraq. Therefore, the current study was conducted to assess the seroprevalence of *T. gondii* infection in patients with obesity and diabetes in Baghdad-Iraq. The study also evaluates the lipid profile of the participants which may has been affected by the course of toxoplasmosis.

Materials and Methods

Study design and subjects

This study was an observational analytical investigation conducted from October 2019 to March 2020, at Al-Kadhimiya Teaching Hospital, Baghdad-Iraq. Consecutive sampling method was used, and a total of 100 participants were included. Epitools found in https://epitools.ausvet.com.au/ was used for determining the total sample size.

The inclusion criteria included both gender with an age range of 18-75 years of subjects who provided signed permission to participate in this study. Exclusion criteria included pregnant and breast feeding women, subjects using hormonal contraception and those with malignant diseases. Participant's weight was measured by an electronic scale (IndiaMart, India) and their standing height was measured with a stadiometer (CMS equipment Ltd, UK). The participants were with least clothing and had no shoes. The following equation was used to calculate Body Mass Index, BMI (kg/m^2) = Weight (kg) / Height (m^2) [18]. BMI was used to categorize the participants into obese, overweight and normal. In addition, all participants underwent a clinical interview, laboratory examination and medical examination to validate their health status of being diabetic or non-diabetic. The clinical interview and medical examination were achieved by a specialist.

This study protocol was approved by the Ministry of Health and Environment, Baghdad, Iraq, with the approval of the local ethics committee (Ref.: BEC/0620/0016), Department of Biology, College of Science, University of Baghdad.

Glucose and glycated haemoglobin (HbA1c)

For all participants, about 6-7 ml of blood samples were taken using a vein puncture technique. About 2.5-3 ml of blood samples were added immediately into labelled EDTA tubes for anticoagulation and kept in a cooling box. The blood samples were analysed within 1 hour after collection. The whole blood was used for glucose and HbA1c analysis. Glucose was analysed using a glucose kit (Linear chemicals, Spain). HbA1c was analysed using a special kit (Roche analytics, Swiss). Both tests were applied according to the manufacturer's instructions.

The rest of blood samples were added into vacuumed, clot and gel activator tubes and left for 30- 35 minutes at room temperature to clot before all samples were centrifuged at 3000 rpm for 15 minutes.

Each serum sample was transferred by a sterile micropipette into 3 sterile Eppendorf tubes for the following different tests to avoid freezing and thawing that may influence the accuracy of result. Then, all sera were kept frozen at -20°C until analysed.

Lipid profile

Serum samples of all participants were analysed for cholesterol, triglycerides, high density lipoprotein (HDL), and very low-density lipoprotein (VLDL). Lipid profile kits (Linear chemicals, Spain) were used for these tests which were applied according to the manufacturer's instructions.

Detection of anti-*T. gondii* IgG and IgM

Serum samples of all participants were analysed for anti-*T. gondii* IgG and anti-*T. gondii* IgM antibodies with the enzyme immunoassay kit "*Toxoplasma* IgG" (Immuno-Biological Laboratories, Inc, USA). A result equivalent or greater than 1.68 mg/L was considered positive for IgG. All tests were achieved after the directions of the manufacturer's instructions.

Metabolic Syndrome

The metabolic syndrome for each participant was calculated based on the International Diabetes Federation (IDF) worldwide definition.

The diagnosis requires the presence of a BMI >30 kg/m^2 or a waist circumference above the ethnic threshold plus 2 of the following further 5 criteria: Waist circumference ≥ 40 inches in Men and ≥ 35 inches in women,

Triglycerides ≥ 150 mg/dL (1.7 mmol/L),

HDL < 40 mg/dL (1.03 mmol/L) in Men and < 50 mg/dL (1.29 mmol/L) in women,

Blood Pressure ≥ 130 / 85 mm Hg,

and Fasting Glucose ≥ 110 mg/dL (5.6 mmol/L)

Statistical Analysis

Statistical analysis of data was performed using SAS (Statistical Analysis System - version 9.1). Student T-test and one- way and two-way ANOVA were used to assess the significant differences among means. The percentages were compared by using the Chi-square test. $P < 0.05$ is considered statistically significant. All data are expressed as mean±SD. Kappa value was used to determine the agreement between the IgG and IgM results.

Results

Subject characteristics and seropositive rate of *T.gondii*

One hundred adults were enrolled in this study. The age distribution was 18 to 75 years. Twenty nine were >30, 16 were between 31 and 40, 26 were between 41 and 50 old, 8 were between 51 and 60 old, and 12 were between 61 and 75 years old. Forty two were males and 58 females. According to BMI values, all participants (n=100) were divided into three groups: obese (n=38), overweight (n=32) and normal (n=30). In addition, they were divided into diabetic and non-diabetic based on clinical examination, laboratory examination and medical interview. Another classification was also considered: Obese-diabetic (n=24), obese- non diabetic (n=14), overweight-diabetic (n=13), overweight non diabetic (n=19), normal-diabetic (n=8) and normal-nondiabetic (n=28). Finally, all participants were divided according to a collection of criteria (central obesity, triglycerides, HDL and blood pressure) into metabolic syndrome-positive (n=64) and metabolic syndrome negative (n=36).

The agreement between IgG and IgM rates was substantial and the Kappa value was 0.80. Therefore, only IgG results are presented in this study. Table-1 illustrates the seropositive rates of anti *T.gondii* IgG among different groups. The total seropositive rate in the surveyed population was 50%, half of the survey participants were positive for anti *T.gondii* IgG, which is considered as a significantly ($P=0.01$) high seropositive rate. No significant differences were noticed between different age groups regarding seropositive rate of anti *T.gondii* IgG antibodies, although the age group of 41-50 years old showed the highest seropositive rate (53.84%) compared to other age groups (Table-1). The results also showed that the majority of *Toxoplasma* positive rates were noticed in females (77.85%) versus 11.9% for males. Chi square analysis showed highly significant ($P<0.05$) differences between females and males regarding anti-*Toxoplasma* seropositive rates. The anti *T.gondii* IgG positive rate was higher in overweight patients (59.37%) comparing with obese (39.47%) and individuals with normal BMI (53.33%). Statistical analysis showed no significant differences ($P=0.22$) in the seropositive rate among these three groups. The results also showed that there was no significant difference ($P=0.84$) in anti-*T. gondii* IgG antibodies positive rate between diabetic (48.88%) and non-diabetic (50.9%) patients. While, the results showed that the anti *T.gondii* IgG antibodies were significantly higher in

patients with metabolic syndrome compared with subjects who were diagnosed as metabolic syndrome-free individuals. Anti-*T. gondii* IgG antibodies were found in 37 (57.81%) of 64 metabolic syndrome positive patients and in 13 (36.11%) of 36 controls (P = 0.03).

The results also illustrated that the anti *T.gondii* IgG positive rate was higher in normal-diabetic patients and overweight diabetic patients compared with other groups (Table-2). Statistical analysis showed no significant differences (P=0.3) in seropositive rate among these three groups

Table-2 illustrates the variation of some factors among anti *T.gondii* IgG positive cases and anti *T.gondii* IgG negative cases. Significant differences were noticed only for abdominal obesity and HDL (P<0.05). Anti *T.gondii* IgG positive cases had higher abdominal obesity (43.02±6.14 inches), versus low abdominal obesity (38.6±6.43 inches) for anti *T.gondii* IgG negative cases. The HDL concentration was higher (43.8±13.26 mg/dL) in anti *T.gondii* IgG positive cases compared with that, in anti *T.gondii* IgG negative group (38.24±10.76 mg/dL). Other factors did not show significant differences between the two groups.

Lipid profile among different groups

Results of total cholesterol showed higher values than normal in all groups (obese, overweight and normal), except those who had normal BMI and were negative for anti-*Toxoplasma* antibodies (Table-3). Diabetic and *Toxoplasma* seropositivity altered cholesterol levels among all groups. Diabetic *Toxoplasma* -ve patients and diabetic *Toxoplasma* +ve patients of the obese group showed higher means of cholesterol level (283.04 ± 52.96 and198.18 ± 29.62 mg/dL respectively) compared with other groups. Two way ANOVA showed no significant differences (P>0.05) among all groups. Regarding the metabolic syndrome, the results also showed that cholesterol levels can be altered by the course of *Toxoplasma* seropositivity in both metabolic syndrome positive and negative patients. Cholesterol levels were higher than the normal values in all groups, except those who had no metabolic syndrome and were negative for anti-*Toxoplasma* antibodies (Table-4); cholesterol level of the latest group was within the border line (169.95 ± 44.58 mg/ dL). One way ANOVA showed no significant differences (P>0.05) in the means of cholesterol levels among different groups, although the highest mean of cholesterol (186.49 ± 44.38 mg/dL) was noticed for those with metabolic syndrome and were positive for anti-*Toxoplasma* antibodies.

The results of triglycerides are illustrated in Table-5. Body mass index, diabetes and *Toxoplasma* seropositivity altered triglycerides levels among all groups. Normal- diabetic *Toxoplasma* +ve patients and obese- diabetic *Toxoplasma* +ve patients showed higher and above normal means of triglycerides (207.33 ± 134.31 and 188.62 ± 34.48 mg/dL, respectively). While normal-diabetic-*Toxoplasma* –ve group showed the lowest values of triglycerides (53.5± 12.02 mg/dL). Two way ANOVA showed significant differences of triglycerides (P< 0.05) among all groups. The results also showed that both metabolic syndrome positive status and *Toxoplasma* seropositivity could alter triglycerides levels. Triglyceride levels were higher than the normal values in patients with metabolic syndrome, while the triglycerides levels were within normal values (Table-5). There were significant differences (P=0.05) in the means of triglycerides levels among different groups. The highest mean of triglycerides (170.49 ± 91.76 mg/dL) was noticed for those with metabolic syndrome and were positive for anti-*Toxoplasma* antibodies.

The results showed that HDL concentrations did not markedly increase among all groups; the means of HDL level were within normal ranges. Despite that, it was noticed that *Toxoplasma* seropositive status influenced HDL concentration. *Toxoplasma* +ve patients (diabetic and non-diabetic) showed high means of HDL compared with *Toxoplasma* –ve patients of obese, overweight and normal BMI groups (Table-7). Significant differences (P< 0.05) in the means of HDL were noticed among groups. The highest HDL levels were reported in overweight-diabetic-*Toxoplasma* +ve cases (45.76 ± 17.56 mg/dL) and in obese-non diabetic *Toxoplasma* +ve cases (47.22 ± 12.36 mg/dL) versus low HDL levels in other groups, especially those with normal BMI and were negative for anti-*Toxoplasma* antibodies (34.85 ± 18.6 mg/dL) .

The results of HDL levels regarding the metabolic syndrome are illustrated in Table-8. *Toxoplasma* +ve cases for both metabolic syndrome +ve and –ve groups showed the highest means of HDL levels (41.19 ±13.04 and 51.24±11.26 mg/dL, respectively), versus low means of HDL levels for these two groups in *Toxoplasma* –ve cases (36.43 ± 11.72 and 40.36 ± 9.08 mg/dL, respectively). Statistical analysis revealed significant differences (P=0.03) among groups.

The results of VLDL are shown in (Table-9). Significant differences (P<0.05) were noticed among groups. The highest means of VLDL were recorded in obese-diabetic-*Toxoplasma* +ve cases (44.95 ± 18.41 mg/dL) and overweight-non diabetic-toxoplasma –ve cases (29.24 ± 16.48 mg/dL). While the lowest means of VLDL were seen in normal- diabetic-*Toxoplasma* –ve cases (11.8 ± 0.85 mg/dL). VLDL levels also differed between metabolic syndrome +ve and –ve groups (Table-10). Metabolic syndrome +ve cases for both *Toxoplasma* +ve and –ve group showed the highest means of VLDL (34.29 ± 18.04 and 33.6 ± 22.89 mg/dL, respectively), versus low VLDL concertation in metabolic syndrome –ve cases. Statistical analysis indicated significant differences (P=0.02) among groups.

Table 1- Seropositive rate of *Toxoplasma gondii* among different groups

Group	N	n(%)anti *T.gondii* IgG positive	X^2 (P-value)
All	100	50 (50%)	0 (0.01)*
>30	29	14 (48.27%)	0.24 (0.99)
31-40	16	8 (50%)	
41-50	26	14 (53.84%)	
51-60	17	8 (47.05%)	
61-75	12	6 (50%)	
Male	42	5 (11.9%)	42.03 (0.00)*
Female	58	45 (77.58%)	
Obese	38	15 (39.47%)	2.94 (0.22)
Overweight	32	19 (59.37%)	
Normal	30	16 (53.33)	
Diabetic	45	22 (48.88%)	0.04 (0.84)
Non diabetic	55	28 (50.9%)	
Metabolic syndrome (+)	64	37 (57.81%)	4.34 (0.03)*
Metabolic syndrome (-)	36	13 (36.11%)	
Obese-diabetic	24	8 (33.33%)	6.014 (0.3)
Obese-non diabetic	14	7 (50%)	
Overweight-diabetic	13	8 (61.53%)	
Overweight-non diabetic	19	11 (57.89)	
Normal-diabetic	8	6 (75%)	
Normal-non diabetic	22	10 (45.45%)	

Table 2-Differences in some factors (BMI, abdominal obesity, cholesterol, triglycerides, HDL,VLDL, Glucose and glycated haemoglobin) between *Toxoplasma* +ve and *Toxoplasma*-ve groups.

Factors	anti *T.gondii* IgG +ve (n=50)	anti *T.gondii* IgG –ve (n=50)	P-value
BMI (kg/m^2)	29.09±6.46	30.05±6.70	0.46
Abdominal Obesity or waist circumstances (inch)	43.02±6.14	38.60±6.43	0.0007*
Cholesterol (mg/dL)	183.43±43.01	178.19±58.59	0.61
Triglycerides (mg/dL)	152.54±88.43	143.38±98.35	0062
HDL(mg/dL)	43.80±13.26	38.24±10.76	0.023*
VLDL (mg/dL)	30.64±17.40	29.00±19.47	0.65
Glucose (mg/dL)	125.27±62.63	127.81±95.06	0.87
Glycated haemoglobin (HbA1c)	6.87±1.59	6.73±1.77	0.67

Table 3-Cholesterol levels among obese, overweight and normal individuals and their relation to diabetic and seropositivity of *T.gondii*

Group	Cholesterol level (mg/dL)			
	Diabetic		Non-Diabetic	
	anti *T.gondii* IgG +ve	anti *T.gondii* IgG -ve	anti *T.gondii* IgG +ve	anti *T.gondii* IgG –ve
Obese	188.93 ± 34.16	178.18 ± 65.98	180.78 ±64.28	178.58 ± 60.28
Overweight	198.18 ± 29.62	283.04 ± 52.96	178.28 ± 37.14	194.8 ± 41.42
Normal	192.95 ± 41.32	108.5 ± 68.59	169.04 ± 51.94	158.62 ± 49.96

Rows: df=2; F=2.63; P=0.07 Columns: df=3; F=0.88; P=0.4

Table 4-Cholesterol levels in metabolic syndrome (+ve) patients and metabolic syndrome (-ve) patients and their relation to diabetic and seropositivity of *T.gondii*

Group	Cholesterol level (mg/dL)	
	anti *T.gondii* IgG +ve	anti *T.gondii* IgG -ve
Metabolic syndrome (+ve)	186.49 ± 44.38	185.22 ± 68.24
Metabolic syndrome (-ve)	176.00 ± 39.53	169.95 ± 44.58

df= 3; F= 0.57; P=0.6

Table 5-Triglycerides levels among obese, overweight and normal individuals and their relation to diabetic and seropositivity of *T.gondi*

Group	Triglycerides (mg/dL)			
	Diabetic		Non-Diabetic	
	anti *T.gondii* IgG +ve	anti *T.gondii* IgG -ve	anti *T.gondii* IgG +ve	anti *T.gondii* IgG –ve
Obese	188.62 ± 34.48	175.25 ± 120.7	127.28 ± 61.96	155.14 ± 50.08
Overweight	141.12 ± 48.45	231.6 ± 117.6	146.27 ± 82.1	112.75 ± 79.87
Normal	207.33 ± 134.31	53.5 ± 12.02	93.2 ± 50.55	99 ± 52.55

Rows: df=2; F=3.16; P=0.04 Columns: df=3; F=3.49; P=0.01

Table 6-Triglycerides levels in metabolic syndrome (+ve) patients and metabolic syndrome (-ve) patients and their relation to diabetic and seropositivity of *T.gondii*

Group	Triglycerides (mg/dL)	
	anti *T.gondii* IgG +ve	anti *T.gondii* IgG -ve
Metabolic syndrome (+ve)	170.49 ± 91.76	167.37 ± 114.89
Metabolic syndrome (-ve)	101.46 ± 53.43	119.12 ± 65.56

df= 3; F= 3.14; P=0.02

Table 7-High density lipoprotein (HDL) levels among obese, overweight and normal individuals and their relation to diabetic and seropositivity of *T.gondii*

Group	HDL (mg/dL)			
	Diabetic		Non-Diabetic	
	anti *T.gondii* IgG +ve	anti *T.gondii* IgG -ve	anti *T.gondii* IgG +ve	anti *T.gondii* IgG –ve
Obese	36.18 ± 10.89	35.28 ± 10.9	47.22 ± 12.36	30.24 ± 11.04
Overweight	45.76 ± 17.56	42.46 ± 12.81	45.07 ± 9.56	40.93 ± 7.11
Normal	39.93 ± 14.06	34.85 ± 18.6	46.79 ± 15.09	43.83 ± 7.27

Rows: df=2; F=3.99; P=0.02 Columns: df=3; F=2.94; P=0.03

Table 8-High density lipoprotein (HDL) levels in metabolic syndrome (+ve) patients and metabolic syndrome (-ve) patients and their relation to diabetic and seropositivity of *T.gondii*

	HDL (mg/dL)	
Group	anti *T.gondii* IgG +ve	anti *T.gondii* IgG -ve
Metabolic syndrome (+ve)	41.19 ± 13.04	36.43 ± 11.72
Metabolic syndrome (-ve)	51.24 ± 11.26	40.36 ± 9.08

df= 3; F= 4.75; P=0.003

Table 9-Very low density lipoprotein (VLDL) levels among obese, overweight and normal individuals and their relation to diabetic and seropositivity of *T.gondii*

	VLDL (mg/dL)			
Group	Diabetic		Non-Diabetic	
	anti *T.gondii* IgG +ve	anti *T.gondii* IgG -ve	anti *T.gondii* IgG +ve	anti *T.gondii* IgG –ve
Obese	44.95 ± 18.41	35.06 ± 24.14	25.57 ± 12.42	31.3± 9.93
Overweight	28.4 ± 9.68	46.36 ± 23.45	29.24 ± 16.48	22.48 ± 15.69
Normal	41.65 ± 26.83	11.8 ± 0.85	19.32 ± 8.81	19.49 ± 10.42

Rows: df=2; F=3.74; P=0.02 Columns: df=3; F=4.65; P=0.004

Table 10-Very low density lipoprotein (VLDL) levels in metabolic syndrome (+ve) patients and metabolic syndrome (-ve) patients and their relation to seropositivity of *T.gondii*

	VLDL (mg/dL)	
Group	anti *T.gondii* IgG +ve	anti *T.gondii* IgG -ve
Metabolic syndrome (+ve)	34.29± 18.04	33.6± 22.89
Metabolic syndrome (-ve)	20.26± 10.1	23.61± 13.02

df= 3; F= 3.38; P=0.02

Discussion

In the current study, the total seropositive rate was significantly high, where half of the participants were positive for anti-toxoplasma IgG. This result was not very much higher than the result of Ali (2018), who showed a 44.8% seropositive rate among 250 women from Kurdistan, Iraq [19]. . Also the resulted value was higher than that of the study of Mohammed and Al-Janabi (2019), who showed a 42.6% seropositive rate among 75 women from Babylon province, Iraq [20].

The differences in seropositive rates of *T.gondii* are likely due to many factors, including sample size, differences in nature of the areas, diagnostic tool used, living conditions, socio-economic criteria and immunological status [21]. No significant differences were noticed among different age groups regarding seropositive rate of *Toxoplasma* IgG, which agrees with the results of Salih *et al*. (2020) [22]. While the result of age related seropositivity disagreed with the result of Nafal and Al-Warid (2019), which showed significant differences in seropositive rates for *Toxoplasma* antibodies among males with chronic liver diseases from different age groups [21].

With respect to gender, the seropositive rate in females was significantly higher than that in males. Similar results were obtained in some other surveys [23, 24]. In contrast, another report demonstrated that male patients have a greater seropositive rate than female patients [25]. Females are susceptible to *T.gondii* infection, which is likely due to the sexual hormones which are considered as key factors that determining the susceptibility to toxoplasmosis [26].

No association was found between high BMI and anti-toxoplasma seropositive status, although the anti-*T.gondii* IgG positive rate was greater in overweight group versus obese and individuals with normal BMI. This result disagrees with that of Reeves *et al*. (2013) who found that obesity is correlated with high titre of anti-*T.gondii* antibodies [27]. In addition, a positive association between seroprevalence of *T. gondii* infection and obesity was indicated among adults in the study of Wilking *et al*. (2016) [28]. In contrast, the results of the current study agree with some other studies [29, 30], which indicated no serological or molecular evidence of an association between toxoplasmosis and

obesity. It is not clear why the association of toxoplasmosis and high BMI was observed in some previous studies, but not in obese people in the current study. It is likely that the differences in the socio-demographic profile of the populations and differences in study designs could explain the differences in the association. For instance, in the study of Reeves *et al*. (2013), the association between toxoplasmosis and high BMI was detected among individuals aged 60 years and older but not in individuals less than 60 years [27].

The results also showed no significant difference in anti-*T. gondii* IgG positive rate between diabetic and non-diabetic patients. These findings did not support a relation between *T.gondii* seropositive status and diabetes, especially when no significant differences were noticed regarding glucoses and HbA1c between *Toxoplasma* seropositive cases and control. These results are in line with the results of others who showed lack of association between *T. gondii* seropositive cases and diabetes [31, 32]. While the results of the current study disagree with some other investigations [33, 34, 35], that indicated significantly greater seropositive rates of toxoplasmosis in diabetic patients than in healthy controls. There are some factors, such as the duration of diabetes, severity of non-infectious complications, comorbidity, and coinfection which can lead to the association between *T. gondii* and diabetes [34]. However, such factors were not considered in the current study. On other hand, it was suggested by Majidiani *et al*. (2016) that chronic toxoplasmosis can be associated with diabetes [35]. In the current study, the minority of *Toxoplasma* positive cases were chronic and thus, no association was noticed between *Toxoplasma* positive cases and diabetes.

Studies regarding the association between the metabolic syndrome and toxoplasmosis are very limited. In the present study, a significant correlation between metabolic syndrome and *T.gondii* seropositive status was observed. The calculation of metabolic syndrome is based on many factors, two of which were abdominal obesity and HDL, both showed significant differences between *Toxoplasma* +ve cases and control. These results agree with those of previous studies that showed high levels of HDL in people with *T.gondii* seropositive status [37]. While, contrasting results were noticed by other investigator in a murine model [38], which may be due to the chronic nature of the disease since the minority of cases in this study were chronic. For both obesity and diabetes patients, regardless of the *Toxoplasma* seropositive status, alternations in lipid profile are recorded. It was confirmed that the dyslipidemia of obesity is characterized by high triglycerides and low HDL with normal total cholesterol (TC) and low LDL concentrations [39]. While, lipid irregularities in patients with diabetes are characteristically known by elevation in cholesterol, high triglycerides, low HDL and high LDL [40]. The results of the current study agreed with those previous finding.

Finally, based on the findings of this study, it was indicated that *T. gondii* seropositive status played a significant role in altering only HDL level while other parameters of lipid profile were not be influenced by *T.gondii* seropositivity among obese, diabetic, s and metabolic syndrome patients. The results also showed an alternation among different groups regarding other parameters of the lipid profile. These variations are likely due to obesity and diabetes. This study also indicated that gender, abdominal obesity, and metabolic syndrome are likely be considered as risk factors for Toxoplasmosis.

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