

## Seroprevalence of *Toxoplasma gondii* in ruminants by using latex agglutination test (LAT) and enzyme-linked immunosorbent assay (ELISA) in Assiut governorate

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**Abstract.** Toxoplasmosis is an important zoonotic parasitic disease world-wide. However, data on *Toxoplasma gondii* infection in ruminants are scarce in Assiut governorate, Egypt. Thus we conducted a survey on the seroprevalence of *T. gondii* infection by using latex agglutination test (LAT) and enzyme-linked immunosorbent assay (ELISA). A total of 274 sera were tested (56 imported camels, 56 cattle, 55 buffaloes, 50 sheep and 57 goats) in the period from November 2014 – February 2016. The overall seroprevalence in ruminants was (35.8%) by LAT and (83.6%) by ELISA. The highest rate of *T. gondii* infection by LAT was found in goats (47.4%) followed by sheep (44%), camels (35.7%), cattle (32.1%) and buffaloes (20%). The highest rate of *T. gondii* infection by ELISA was recorded in camels (96.4%) followed by goats (87.7%), sheep (86%), buffaloes (74.5%) and cattle (73.2%). Antibody titres to *T. gondii* in positive sera by LAT ranged from 1:2 to 1:64 in imported camels, 1:2 to 1:32 in cattle, 1:2 to 1:8 in buffaloes, from 1:4 to 1:64 in sheep and from 1:2 to 1:16 in goats. The differences of seroprevalence between different species were significant by LAT and highly significant by ELISA. Our results suggest that the *T. gondii* parasite is widely spread due to high exposure to infective cat faeces.

### INTRODUCTION

In Egypt, ruminants play a very important role as a source of meat, milk and hide with great economic importance. *T. gondii* was firstly discovered in North Africa by (Nicolle & Manceaux, 1908). Seroprevalence rates of *Toxoplasma* in Africa are very high in both human and animal populations (Hammond-Aryee *et al.*, 2014). Toxoplasmosis is most widely distributed zoonotic disease caused by an obligate intracellular protozoan parasite called *T. gondii* which can infect all mammals concern hazards in both human health and veterinary medicine. Approximately one-third of humanity has been exposed to the parasite worldwide (Montoya & Liesenfeld, 2004; Sukthana, 2006; Halonen & Weiss, 2013). It is widely

distributed in many species of livestock with incidences from zero to 100% in the different countries (Prelezov *et al.*, 2008; Dubey, 2009). Cattle and other herbivorous may acquire infection by grazing on pastures contaminated with cats' feces and/or through drinking oocyst contaminated water of cat origin (Jacek *et al.*, 2007; Vesco *et al.*, 2007).

*Toxoplasma* oocysts shed continuously in the cat's feces from 4 until 14 days after infection with an expected peak output of tens of millions at 6-8 days. Thus, fifty grams of infected cat feces may contain as many as 10 million oocysts (Dubey & Frenkel, 1972). Infected cats shed *T. gondii* oocysts that after sporulation become infective for men and animals, preserving this capability for up to 18 months in the environment (Gorbani *et al.*, 1983).

Cattle and buffaloes have high natural resistance to *T. gondii*. Their infection with the parasite is usually subclinical and very rarely exhibit clinical signs (Dubey & Thulliez, 1994; Hill & Dubey, 2013; Kalita & Sarmah, 2015). While, in sheep and goats it was considered a major cause of reproductive failure including abortion and neonatal deaths (Masala *et al.*, 2003; AbouZeid *et al.*, 2010 ; Dubey, 2010). It is not only responsible for economic losses, but also it may be transmitted to humans via contaminated meat and milk (Gamossi *et al.*, 2011; Edward & Dubey, 2013; Hajian-Bidar *et al.*, 2014).

Sero-diagnosis of *T. gondii* is adequate tool for epidemiological studies in both human and animals (Figueiredo *et al.*, 2001). LAT has been widely used for screening of *T. gondii* infection in farm animals for its 90% sensitivity and specificity (Lashari & Tasawar, 2010; Khalil & Elrayah, 2011). Also, (Shaapan *et al.*, 2008; Sroka *et al.*, 2011) added that ELISA is a good sensitive and specific tool for epidemiological surveys of *Toxoplasma* infection in animals and human.

Protein G horseradish peroxidase conjugate used in the IgG ELISA assays instead of the kit conjugate to allow testing of multiple animal species (cows, sheep, goats and camels) (Bjorck & Kronvall, 1984; Abu-Zeid, 2002; Werre *et al.*, 2002; Zhang *et al.*, 2010; Schaefer *et al.*, 2011).

Due to the little informations on the seroprevalence of Toxoplasmosis in ruminants in Assiut and its public health importance, this study was aimed to investigate the seroprevalence of *T. gondii* infection in different ruminant species by using LAT and ELISA in Assiut Governorate, Egypt.

## MATERIALS AND METHODS

### 1) Collection of samples:

A total of 274 blood samples were collected randomly from apparently healthy 56 imported camels, 56 cattle, 55 buffaloes, 50 sheep and 57 goats selected from different rural regions in Assiut Governorate, Egypt in the period from November 2014 – February

2016. Data collected regarding to the species and gender.

### 2) Preparation of blood samples:

A 10 ml of whole blood samples were collected from jugular vein of the above mentioned animals into glass tubes. The collected blood samples were kept under refrigeration until arrival to the laboratory. Sera were separated by centrifugation at 3000 rpm for 15 minutes after being kept in the refrigerator overnight then labeled and stored at -20°C until laboratory testing. Serological investigation of the collected sera for the presence of anti-*T. gondii* IgG antibodies was done using Latex agglutination test (LAT) and enzyme linked immunosorbent assay [ELISA (IgG)] in different ruminants species (camel, cattle, buffaloes, sheep and goats). The serological results were grouped in classes and tabulated on the variables of animal species and sex of animals.

### 3) Latex agglutination test (LAT):

This assay was done by using (Toxo Latex Kit, Cam Tech Medical, UK), this kit was designed for qualitative and semi-quantitative tests, the test was performed according to company instructions. As interpretation of results, the highest dilution of serum showing agglutination corresponds to the titre of antibodies to *T. gondii* (Ibrahim *et al.*, 2014).

### 4) Enzyme-linked immunosorbent assay (ELISA):

A commercial *Toxoplasma* IgG ELISA (Calbiotech A life science company-Catalog No: EX022G-USA) was modified by use of protein G horseradish peroxidase conjugate (Molecular Probes by Life Technologies Corporation, Catalog No. P21041, Lot: 1495870, USA) to replace the anti-human Kit conjugate. The use of protein G conjugate has previously been demonstrated to be an effective means of serological testing in domestic animals and provide strong binding to IgG antibody (Bjorck & Kronvall, 1984; Abu-Zeid, 2002; Werre *et al.*, 2002; Bhide *et al.*, 2004; Zhang *et al.*, 2010; Schaefer *et al.*, 2011). Protein G conjugate was received as lyophilized powder and was diluted 1:20,000 with phosphate buffered saline (pH 7.2) prior

to use (Schaefer *et al.*, 2011). ELISA was performed according to the manufacturer's guidelines. The optical density (OD) was measured at 450 nm with ELISA reader within 15 minutes. Comparison to kit-provided calibrators guided the identification of positive versus negative serum. Antibody index less than to 0.9 represent a negative result, between 0.9 and 1.1 was considered borderline positive and greater than to 1.1 indicating a positive reaction for anti-*T. gondii* IgG antibody as described by the kit. For each plate, the cut-off value was calculated as the manufacturer's guidelines = Calibrator OD X Calibrator Factor (0.5).

### 5) Statistical analysis

According to Fisher & Yates (1963): Chi-square Test was used to compare the effect of species and sex on the prevalence of *T. gondii* among investigated animal.

## RESULTS

The overall seroprevalence of *T. gondii* in different ruminant species was 35.8% (98/274 animals) by LAT and 83.6% (229 /274) by ELISA. The highest rate of *T. gondii* infection by LAT was found in goats 47.4% (27/57) followed by sheep 44% (22/50), camels 35.7% (20/ 56), cattle 32.1% (18/56) and buffaloes 20% (11/55). While the highest rate of *T. gondii* infection by ELISA was recorded in camels 96.4% (54/56) followed by goats 87.7% (50/57), sheep 86% (43/50), buffaloes 74.5% (41/55) and cattle 73.2% (41/56). The

differences of seroprevalence between different species were significant by LAT and highly significant by ELISA (Table 1).

Seroprevalence of *T. gondii* in female camels was 24.1% (7/29) and 100% (29/29) by LAT and ELISA, respectively. While in male camels was 48.2% (13/27) and 92.6% (25/27) by LAT and ELISA, respectively. No significant difference of *T. gondii* was found in both sexes of camels (Table 2). While in female cattle infection rate was 21.7% (5/23) and 56.5% (13/23) by LAT and ELISA, respectively and it was 39.4% (13/33) and 84.9% (28/33) by LAT and ELISA, respectively in male cattle (Table 2). Infection rate in female buffaloes was 20.7% (6/29) and 62.1% (18/29) by LAT and ELISA, respectively. Male buffaloes showed infection rate of 19.2% (5/26) and 88.5% (23/26) by LAT and ELISA, respectively (Table 2). There was no significant difference between seroprevalence of males and females by LAT while, significant differences was found by ELISA in both cattle and buffaloes (Table 2). Rate of infection in female sheep was 71.4% (15/21) and 76.2% (16/21) by LAT and ELISA, respectively and in male sheep it was 24.1% (7/29) and 93.1% (27/29) by LAT and ELISA, respectively (Table 2). Female goats were infected as 71% (22/31) and 80.6% (25/31) by LAT and ELISA, respectively. In male goats it was 19.2% (5/26) and 96.2% (25/26) by LAT and ELISA, respectively (Table 2). Very high significant difference in females than males detected by LAT while, no significant differences found by ELISA in sheep and goats (Table 2).

Table 1. Overall seroprevalence of Toxoplasmosis in different ruminants based on LAT and ELISA

Species	No. of sera tested	No. of positive samples by LAT*	(%)	No. of positive samples by ELISA**	(%)
Camels	56	20	35.7	54	96.4
Cattle	56	18	32.1	41	73.2
Buffaloes	55	11	20	41	74.5
Sheep	50	22	44	43	86
Goats	57	27	47.4	50	87.7
Total	274	98	35.8	229	83.6

\* Significant differences ( $\chi^2 = 11.08$  and  $P < 0.05$ ) by LAT.

\*\* High significant differences ( $\chi^2 = 15.32$  and  $P < 0.01$ ) by ELISA.

Table 2. Prevalence rate of *T. gondii* antibodies among male and female different ruminants by LAT and ELISA

Animal species	Sex	No. of sera tested	No. of positive samples by LAT	(%)	No. of positive samples by ELISA	(%)
Camel	Male	27	13	48.2	25	92.6
	Female	29	7	24.1	29	100
	Total	56	20	35.7	54	96.4
		$\chi^2$ P		$\chi^2 = 3.51$ P > 0.05		$\chi^2 = 2.25$ P > 0.05
Cattle	Male	33	13	39.4	28*	84.9
	Female	23	5	21.7	13	56.5
	Total	56	18	32.1	41	73.2
		$\chi^2$ P		$\chi^2 = 1.94$ P > 0.05		$\chi^2 = 5.55$ P < 0.05
Buffaloes	Male	26	5	19.2	23*	88.5
	Female	29	6	20.7	18	62.1
	Total	55	11	20	41	74.5
		$\chi^2$ P		$\chi^2 = 0.02$ P > 0.05		$\chi^2 = 5.05$ P < 0.05
Sheep	Male	29	7	24.1	27	93.1
	Female	21	15***	71.4	16	76.2
	Total	50	22	44	43	86
		$\chi^2$ P		$\chi^2 = 11.05$ P < 0.001		$\chi^2 = 2.89$ P > 0.05
Goats	Male	26	5	19.2	25	96.2
	Female	31	22***	71	25	80.6
	Total	57	27	47.4	50	87.7
		$\chi^2$ P		$\chi^2 = 15.20$ P < 0.001		$\chi^2 = 3.15$ P > 0.05

Insignificant differences (P > 0.05)

\* Significant differences (P < 0.05)

\*\*\* Very high significant differences (P < 0.001)

From 56 imported camels sera 20 (35.7%) were found positive of *T. gondii* with intensity of 1 (1.8%), 6 (10.7%), 6 (10.7%), 5 (8.9%), 1 (1.8%) and 1 (1.8%) by dilution of 1:2, 1:4, 1:8, 1:16, 1:32 and 1:64 respectively using LAT. From 56 cattle sera 18 (32.1%) were found positive with intensity of 5 (8.9%), 4 (7.1%), 5 (8.9%), 3 (5.4%) and 1 (1.8%) by dilution of 1:2, 1:4, 1:8, 1:16 and, 1:32, respectively using LAT. From 55 buffaloes sera 11 (20%) were found positive with intensity of 4 (7.3%), 3

(5.5%), and 4 (7.3%), by dilution of 1:2, 1:4 and 1:8, respectively using LAT. From 50 sheep sera 22 (44%) were found positive with intensity of 1 (2%), 6 (12%), 2 (4%), 11 (22%) and 2 (4%) by dilution of 1:4, 1:8, 1:16, 1:32 and 1:64 respectively using LAT. From 57 goats sera 27 (47.4%) were found positive with intensity of 1 (1.8%), 4 (7.0%), 13 (22.8%) and 9 (15.8%) by dilution of 1:2, 1:4, 1:8 and 1:16 respectively using LAT.

## DISCUSSION

In the present study, the seroprevalence of *T. gondii* in imported camels was 35.7% and 96.4% by using LAT and ELISA, respectively. These results were higher than those reported from different regions in **Egypt** by several authors as 16.7% by Khalifa *et al.* (2005) and 30.7% using modified agglutination test (MAT) by Shaapan & Khalil (2008). Also, our results were higher than those reported by several authors in camels from different regions of the world as in **Sudan** (20%) using LAT by Khalil & Elrayah (2011); in **Somalia** 6.3% using LAT by Kadle (2014); in **Saudi Arabia** 6.6% using indirect fluorescent antibody test (IFAT) by Al-Anazi (2011) and 23.6% using IFAT by Alanazi (2013); in **Iran** (14.6%) using MAT by Hamidinejat *et al.* (2013) and in **China** 3% using indirect haemagglutination test (IHAT) by Wang *et al.* (2013).

The seroprevalences of *T. gondii* in camels in Cairo, **Egypt** were 66.7% using ELISA by Toaleb *et al.* (2013); in **Sudan** (44% and 54.1%) using LAT by EL Basheir *et al.* (2012) and Ibrahim *et al.* (2015a), respectively; in **Ethiopia** 40.5% using indirect ELISA by Gebremedhin *et al.* (2014); in **Saudi Arabia** 45.4% using ELISA by Al-khatib (2011) and in **Iraq** 37.5% using ELISA by Akber *et al.* (2014). These results were lower than our results reported by ELISA 96.4% and higher than reported by LAT 35.7%.

There is no significant difference in seroprevalence of *T. gondii* in both sexes of camels. These results were in agreement with (EL Basheir *et al.*, 2012; Hamidinejat *et al.*, 2013; Khamesipour *et al.*, 2014) who stated that no association was found between the prevalence of *T. gondii* and sex in camels. In contrary, Al-khatib (2011) found that the prevalence of *T. gondii* was much higher in female compared to male camels in Saudi Arabia. While Abu-Zeid (2002) recorded that male camels had to some extent higher seropositive than females.

Few reports have been done about the occurrence of *T. gondii* among buffaloes in Egypt. Our results reported a higher seroprevalence of *T. gondii* 74.5% by ELISA and lower 20% by LAT when compared to

previous studies in **Egypt** 43.8% by using Sabin Feldman Dye Test (DT) in Assiut by Fahmy *et al.* (1979), 22.5% using MAT by Shaapan *et al.* (2010), 30% by Khalifa *et al.* (2005) and 34.4% using ELISA IgG assay by Hassanain *et al.* (2013).

Higher seroprevalence of *T. gondii* was reported in our study than other countries as in **Brazil**, 27.2% using IFAT by Santos *et al.* (2013), (41.6% and 41.3%) using ELISA by (Silva *et al.*, 2013; Silva *et al.*, 2014), respectively; in **Argentina**, 25.4% using IFAT by Konrad *et al.* (2013) and in **Mexico** 48.7% using MAT by Alvarado-Esquivel *et al.* (2014).

Unlike our result, lower seroprevalence of *T. gondii* among buffaloes had also been reported in **Egypt** 8% using electroimmunotransfer blot (EITB) and micro-ELISA by Sabry & Reda (2008); in **Iran** 14.3% using MAT by Hamidinejat *et al.* (2010); in **Iraq**, 8.6% using ELISA by Akber *et al.* (2014) and in **Trinidad** seroprevalence of *T. gondii* was 7.8% using LAT by Persad *et al.* (2011).

No significant difference between male and female buffaloes using LAT which matching with Persad *et al.* (2011) who mentioned that not statistically significant differences in seropositivity of *Toxoplasma* in male and female water buffalo in Trinidad. While by ELISA there was significant difference in males than females. These results disagreed with (Sabry & Reda 2008; Shaapan *et al.*, 2010) who reported that the prevalence of Toxoplasmosis was higher in female buffaloes than males in Egypt.

Seroprevalence of *T. gondii* in cattle was 32.1% by LAT and 73.2% by ELISA. Our results showed a prevalence value higher than other values reported in **Egypt**, 22.7% and 19.3% using ELISA by (Hassanain *et al.*, 2010 and Hassanain *et al.*, 2013), respectively. Also, our seroprevalence of *Toxoplasma* in cattle was higher than those reported from different regions of world by several authors as in **Sudan** 32% using LAT by Khalil & Elrayah (2011) and 13.3% using ELISA by Elfahal *et al.* (2013); in **Algeria**, 3.9% using IFAT by Dechicha *et al.* (2015); in **Somalia**, 7.1% using LAT by Kadle (2014); in **Nigeria**, 13.9% using ELISA by Onyiche & Ademola (2015);



in **Iraq**, 29.2% using LAT by Al-Ramahi *et al.* (2010) and 14.6% using ELISA by Akber *et al.* (2014); in **Iran**, 11.9% using ELISA by Cheraghipour *et al.* (2013); in **Malaysia** 2.6% using IFAT by Rahman *et al.* (2011); in **Bangladesh** 27% using LAT by Rahman *et al.* (2014); in **Thailand**, 9.4% using LAT and 17% using ELISA by Inpankaew *et al.* (2010); in **China** 5.7% using IHAT by Zhou *et al.* (2012); in **Japan**, 7.3% using LAT by Matsuo *et al.* (2014); in **India**, (16.7%) using LAT by Kalita & Sarmah (2015); in **Brazil**, 17.4% using IFAT by Santos *et al.* (2013) and 2.7% using IFAT by Fajardo *et al.* (2013) and in **the Caribbean region** 8.4% using MAT by Chikweto *et al.* (2011) and 2.7% using ELISA by Sharma *et al.* (2014). While, Our results were lower than that reported by García-Bocanegra *et al.* (2013) was 83.3% by indirect ELISA in **Spain** and by Silva *et al.* (2015) was 83.4% by indirect ELISA in **Brazil**.

In **Egypt**, the seroprevalence of *T. gondii* in cattle was 47.2% by DT in Assiut by Fahmy *et al.* (1979), 46.8% using ELISA by Toaleb *et al.* (2013); in **Sudan** 40.9% using LAT by Ibrahim *et al.* (2015b); in **Iran** 55% using MAT by Asgari *et al.* (2013) and in **Switzerland** 45.6% using ELISA by Berger-Schoch *et al.* (2011). These results were lower than our results reported by ELISA 73.2% and higher than reported by LAT 32.1%.

In the present study, the antibody levels in cattle ranged from 1:2 to 1:32 using LAT similarly to Ibrahim *et al.* (2015b) in Sudan. While, Khalil & Elrayah (2011) reported that LAT were positive with dilution of 1:8 to 1:32 in cattle in Sudan and Al-Ramahi *et al.* (2010) recorded the highest titers in cattle were 1:64 by LAT in Iraq.

No significant difference between males and female cattle using LAT, while by ELISA there is significant difference in males 84.9% than females 56.5%. These results agree with Elfahal *et al.* (2013) who found that there was high significant difference in prevalence of *T. gondii* antibodies in males cattle (30.8%) than in females cattle (11.9%) by ELISA in Sudan.

In the present study, seroprevalence of *T. gondii* in sheep was 44% by LAT and 86% by ELISA in Assiut Governorate. Our results

were relatively higher than that reported in Sharkia, Egypt, as 18% using IHAT by Awadallah (2010). Lower prevalence of *Toxoplasma* in sheep also reported in some countries such as in **Nigeria** 6.7% using ELISA by Kamani *et al.* (2010); in **Somalia** 34.5% using LAT by Kadle (2014); in **Saudi Arabia** 22% using ELISA by Al-Mohammed (2011), 36.4% using IFAT by Alanazi (2013) and 33.3% using LAT by Eisa *et al.* (2013); in **Iraq** 36.4% using LAT by Al-Ramahi *et al.* (2010), 25.4% using LAT by Kader & Al-Khayat (2013), 21.2% using ELISA by Akber *et al.* (2014); in **China** 29.8% using IHAT by Liu *et al.* (2010); in **Iran**, 16.1% using ELISA by Cheraghipour *et al.* (2013) and 24.8% using ELISA by Moazeni Jula *et al.* (2013); in **Brazil** 28.2% using IFAT by Mendonça *et al.* (2013); in **Pakistan** 15.5% using ELISA by Ahmad *et al.* (2015); in **Mongolia** 24% and 16.6% using ELISA and LAT, respectively by Tumurjav *et al.* (2010) and in **Saint Kitts and Nevis, West Indies** 26% using ELISA by Hamilton *et al.* (2015).

In **Egypt**, seroprevalence of *T. gondii* in sheep was 79% using (DT) in Assiut by Fahmy *et al.* (1979), (50.4% and 61.4%) using (LAT and ELISA), respectively in Cairo by Hassanain *et al.* (2011), 44% using ELISA at Giza by Abd El-Ghany & Amin (2012), 53.3% using LAT in Qalyubia by Khater *et al.* (2013), 61.4% using ELISA IgG assay in Cairo by Hassanain *et al.* (2013), (45.5%, 41% and 38.5%) using (MAT, LAT and IHAT), respectively in Cairo and Giza by Shaapan *et al.* (2015) and 41.7%, 66.1% and 62% using LAT, IHAT and ELISA, respectively in Dakahlia by Younis *et al.* (2015). These results was higher than our results reported by LAT 44% and lower than that reported by ELISA 86%. While, at El-Fayoum 98.4% by ELISA by Ghoneim *et al.* (2009), which was higher than our finding.

Seroprevalence of *T. gondii* in other countries as in **Tunis** was 73.6% of sheep using MAT by Boughattas *et al.* (2014); in **Libya** 71% using LAT by Al-Mabruk *et al.* (2013); in **Sudan** 57.5% using LAT by Khalil & Elrayah (2011) and 75% using LAT by Ibrahim *et al.* (2015b); in **Bangladesh** 69.9% using LAT by Rahman *et al.* (2014); and in

**Spain** 49.3% using indirect ELISA by García-Bocanegra *et al.* (2013). These results were higher than our results reported by LAT 44% and lower than that reported by ELISA 86%. While, others higher than our finding as in **Turkey** 95.7% using ELISA by Mor & Arslan (2007) and 98.9% using DT by Çiçek *et al.* (2011) and in **USA** 94.8% using MAT by Edward & Dubey (2013).

The highest titers of *T. gondii* antibodies levels in our study was 1:64 in sheep using LAT which were similar to Al-Ramahi *et al.* (2010) in Iraq. In contrary with (Khalil & Elrayah, 2011; Ibrahim *et al.*, 2015b) who recorded that highest titers were 1:32 and 1:128, respectively by LAT in Sudan.

The seroprevalence rate by LAT was 71.4% in female and 24.1% in male sheep with very high statistical significant difference between infection rate and gender. In agreement with a study conducted in Brazil (Lopes *et al.*, 2010) and China (Wang *et al.*, 2011) who re-vealed higher prevalence of *T. gondii* in female than in male sheep. While the sero-prevalence rate by ELISA was 76.2% in female and 93.1% in male sheep with no statistical significant difference between infection rate and gender. Moreover, there were many reports that did not show significant correlation between *Toxoplasma* infection and gender as found by (Kamani *et al.*, 2010; Tumurjav *et al.*, 2010; Khamesipour *et al.*, 2014).

In the present study, seroprevalence of *T. gondii* in goats were 47.4% by LAT and 87.7% by ELISA. Our results were relatively higher than that reported in different areas in **Egypt**, as 44.3% using MAT in Giza by Shaapan *et al.* (2010), 29% by LAT at Cairo and Zagazig by AbouZeid *et al.* (2010) and 42.3% using IHAT in Cairo, Beni-Suif and Zagazig by Abdel-Rahman *et al.* (2012). On the other hand, Lower prevalence in goats also reported in some countries such as in **Algeria** 13.2% using IFAT by Dechicha *et al.* (2015); in **Somalia** 26.7% using LAT by Kadle (2014); in **Palestine** 13.4% using indirect ELISA by Othman & Al Zuheir (2014); in **Saudi Arabia** 12% using ELISA by Al-Mohammed (2011), 29% and 45.2% using LAT and IHAT, respectively by Eisa *et al.* (2013)

and 35.3% using IFAT by Alanazi (2013); in **Iraq** 21.3% using ELISA by Akber *et al.* (2014); in **Iran** 10.6% using ELISA by Moazeni-Jula *et al.* (2013) and 14.4% using ELISA by Hajian-Bidar *et al.* (2014); in **China** 14.1% using IHAT by Zhao *et al.* (2011) and 9% using IHAT by Xu *et al.* (2014); in **Spain** 25.1% using indirect ELISA by García-Bocanegra *et al.* (2013); in **Saint Kitts and Nevis, West Indies** 34% using ELISA by Hamilton *et al.* (2015) and in **Pakistan** 11.9% using ELISA by Ahmad *et al.* (2015).

Seroprevalence of *T. gondii* in goats in **Egypt** was 54% by using DT in Assiut by Fahmy *et al.* (1979) and 49.4%, 64.2% and 50.6% using LAT, IHAT and ELISA respectively in **Dakahlia** by Younis *et al.* (2015); in **Sudan** 64% using LAT by Ibrahim *et al.* (2015b); in **Bangladesh** 61% using LAT by Rahman *et al.* (2014); in **Italy** 60.6% using MAT by Mancianti *et al.* (2013); in **Pakistan** 52% using LAT by Tasawar *et al.* (2011) and in **Romania** 52.8% using ELISA by Iovu *et al.* (2012). These results was higher than our results reported by LAT 47.4% and lower than that reported by ELISA 87.7%. While, in **Iraq** 47.4% using LAT by Al-Ramahi *et al.* (2010) similar to our finding by LAT. Such differences of the prevalences of caprine toxoplasmosis may be attributed to diagnostic techniques, breeding condition, immune status, timing of infection, and genetic composition of the host and the organism (Suzuki, 2002; Masala *et al.*, 2003).

The highest titers of *T. gondii* antibodies levels in our study were 1:16 in goats using LAT. In contrary with (Al-Ramahi *et al.*, 2010; Ibrahim *et al.*, 2015b) who found that the highest titers of *Toxoplasma* antibody were 1:128 by LAT in **Sudan** and **Iraq**, respectively.

Regarding to gender in the present study, the seroprevalence of *T. gondii* was higher in female goats (71%) than in males (19.2%) and the difference was statistically very high significant by LAT. Consistent with some previous reports (Shaapan *et al.*, 2010; Tasawar *et al.*, 2011; Moazeni-Jula *et al.*, 2013). While the seroprevalence rate by ELISA was 80.6% in female and 96.2% in male goats which has no statistical significant

difference between infection rate and gender. Similar results reported by (Abu-Dalbou *et al.*, 2010; Hajian-Bidar *et al.*, 2014; Xu *et al.*, 2014) who did not show significant correlation between *Toxoplasma* infection and gender.

In our investigation, comparison between seroprevalence between examined species showed that cattle and buffaloes are the least infected species as compared to camels, sheep and goats, this situation has already been described worldwide (Sharif *et al.*, 2007; Toaleb *et al.*, 2013; Dechicha *et al.*, 2015). This variation may be attributed to the differences in susceptibility to *T. gondii* feeding habits of the animals and species immunity (Bahrieni *et al.*, 2008). Lower seroprevalence of cattle and buffaloes to other ruminants explained by Dubey (2010) who reported that cattle and buffaloes are resistant to clinical toxoplasmosis. Higher rate in camels, sheep and goats that could be explained by the variability of the species sensitivity. Higher rate in sheep and goats is influenced by the fact that they tend to roam more freely in the rural area of Egypt than other ruminant and thus have greater access to domestic cat feces. That confirmed by (Ghoneim *et al.*, 2009; Abdel-Rahman *et al.*, 2012).

Our results revealed that the *T. gondii* parasite is widely spread due to high exposure of ruminants to infective cat faeces. Similar finding reported by Ghoneim *et al.* (2009) who mentioned that in Egypt, stray cats are widely spread which is in favor of a higher prevalence of oocysts in humid environment and farming animal rearing. Also, Al-Kappany *et al.* (2010) detected that high prevalence of *T. gondii* in cats in Egypt.

Higher prevalence rates of toxoplasmosis in ruminants in our study due to warm and moist environment which lead to long viability of *T. gondii* oocysts in environments and high density of stray cat in our rural areas which increase contamination of environment with shedding oocysts. The difference between the obtained results of serological tests during the present study and those reported by other investigators might be attributed to the host-parasite relationship

which depends upon the virulence of *T. gondii* strains; the immune status of the different infected ruminants and the differences in the serological tests employed.

In this investigation, high seroprevalence rates by ELISA than LAT occur due to ELISA test is the more suitable test in diagnosis of toxoplasmosis in ruminants (Younis *et al.*, 2015), use of non-species specific protein G conjugate allows the testing of sera from some of veterinary species of interest.

## CONCLUSION

It was concluded that the environment in Assiut is highly contaminated with *T. gondii* oocyst. The results of the present study revealed the high risk of human and animals exposure to *T. gondii* in the investigated area. Further research is needed to assess the risk for infection in humans associated with the ingestion of raw milk or undercooked meat from ruminants infected with *T. gondii*.

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