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EPIDEMIOLOGICAL STUDIES ON TOXOPLASMOSIS IN SMALL RUMINANTS AND EQUINE IN DAKAHLIA GOVERNORATE, EGYPT.

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ABSTRACT

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Prevalence of toxoplasmosis was investigated in small ruminants (292 sheep & 81 goats) and equine (54 horses and 79 donkeys) from Dakahlia governorate, Egypt in the period from October 2013 – October 2014. The annually incidences were estimated by using latex agglutination test (LAT); indirect hemagglutination test (IHAT) and enzyme linked immunosorbent assay (ELISA) in sheep were (41.7%), (66.1%) and (62.0%) respectively, in goat were (49.4%), (64.2%) and (50.6%) respectively, in horse (50.0%), (72.2%) and (72.2%) respectively and (44.3%), (67.1%) and (68.4%) in donkeys respectively. The results of bioassay in cats revealed that 8 out of 25 slaughtered sheep (32.0%) and 9 out of 25 slaughtered donkeys (36.0%) were positive. Histopathological examination on bioassay positive case detected *Toxoplasma gondii (T. gondii)* tissue cysts in 3 (37.5%) and 4 (44.4%) in diaphragm muscles of sheep and donkeys respectively. The sensitivity of both ELISA and IHAT in sheep and donkeys was 100%. Regarding to host risk factors associated with toxoplasmosis, the results revealed that the seroincidence was significantly higher in equine [horses (72.2%) and donkeys (68.4%)] than in small ruminants [sheep (62.0%) and goats (50.6%)] and in relation to the gender the females were higher than in males. There are high associations between the history of abortion and intensive rearing system with incidence of toxoplasmosis in sheep. It could be concluded that the equines and small ruminants play an important role in epidemiology of toxoplasmosis. ELISA test is the more suitable test in diagnosis of toxoplasmosis in small ruminant and equine. There are strong association between serodiagnosis of toxoplasmosis with intensive breeding, old ages and female in small ruminant and equine.

Key words: *Toxoplasma gondii, sheep, goat, horse, donkey.*

INTRODUCTION

Toxoplasma gondii (T. gondii) was an intracellular cyst-forming apicomplexans protozoan organisms occurring in domestic animals and man throughout the world. It has an indirect life cycle, with feline as the definitive hosts (El-On and Peiser, 2003).

The main source of infection of herbivores is infected omnivores feces, by ingestion of *T. gondii* sporulated oocysts in contaminated food and water. Therefore toxoplasmosis in these animals were significantly associated with the presence of omnivores in the farms. *T. gondii* cysts may persist in the tissues of the host for years (Dubey *et al.*, 1989 and Dubey, 2010).

Most acquired toxoplasmosis in ovine, caprine and equine are subclinical, however fever, ataxia, retinal degeneration and encephalomyelitis may develop. Severity of toxoplasmosis in sheep and goats are associated with the stage of pregnancy. Infection during the early stage of gestation can result in fetal death, resorption and abortion, while infection in the later stage of gestation may have no clinical effect and lambs are usually born normal but infected and immune (Dubey and Beattie, 1988 and Buxton *et al.*, 2007).

Due to the microscopic size and intracellular localization of the proliferative forms of this protozoan parasite and the relative difficulty in its laboratory cultivation, the development of immunodiagnostic tools such as serology and

immunohistochemistry are essential in the demonstration of infection (Uggla and Buxton, 1990). The aim of the present study was to determine the Incidence and possible risk factors associated with toxoplasmosis in small ruminants (sheep & goats) and equine (horses and donkeys) from Dakahlia governorates, Egypt.

MATERIALS and METHODS

1. Animals

a) **Slaughtered animals**: 25 slaughtered sheep at Mansoura abattoir and 25 donkeys slaughtered at Mansoura Zoo were used in serological diagnosis of toxoplasmosis; histopathology and tissues bioassay in cats.

b) **Surveyed animals**: 267 sheep, 81 goats, 54 horses and 54 donkeys of different sex from different localities in Dakahlia province aged between 3 months to 8 years were used for determination of seroprevalence of toxoplasmosis in sheep and goats and identification of risk factors associated with toxoplasmosis in the period from October 2013 – October 2014.

c) **Cats**: 100 cats 2-3 months age were used for bioassay of tissue samples taken from slaughtered sheep at abattoir and slaughtered donkeys at zoo (2 cats were used for each tissue sample). They were previously proved free from; *Nematodae; Cestodes; Isospora*, *Eimeria and T. gondii* oocysts through fecal examination. Cats were fasted overnight, each 2 cats fed one tissue sample (diaphragm muscles) cut into small pieces using disposable scalpel and then faecal samples of cats were collected daily for 14 days post infection and examined microscopically for oocysts as described by Dubey, (2001) by fecal floatation technique.

2. Samples:

a) **Blood Samples**: 10 ml of blood were collected from examined sheep, goats, donkeys and horses. After clotting at room temperature, blood was centrifuged at 3000 rpm for 10 minutes and the collected sera were stored at -20^{o}C until assayed for antibodies against toxoplasmosis.

b) **Tissue Samples**: About 50 grams of freshly tissue specimens from diaphragm of slaughtered sheep and donkeys were used for bioassay in cats (2 cats were used for each tissue sample) according to Dubey, (2001) and 50 gm preserved in formalin 10% used for histopathological examination according to Bancroft and Stevens, (1996).

c) **Faecal Samples** of cats were collected daily for 14 days post infection and examined microscopically for oocysts as described by Dubey, (2001).

3. Serological examination of serum samples from sheep, goats, horses and donkeys for toxoplasmosis

(1) Latex agglutination test (LAT).

Latex antigen preparation: The antigen prepared through sensitization of latex particles with locally isolated *T. gondii* strain sonicated tachyzoites as procedures of Lunde and Jacobs (1967). The test procedures were adopted as methods of Holliman *et al.* (1989).

(2) Indirect haemagglutination test (IHAT). Using Toxo-HAI Fumouze kit (Fumouze, France). Procedure was done according to the manufacturer's instructions, according to (Camargo and Leser, (1976).

(3) Enzyme linked immunosorbent assay (ELISA). According to Voller *et al.* (1976) .

Antigen preparation: It was kindly obtained from Zoonotic Diseases Department, Veterinary Research Division, National Research Centre, El-dokki, Giza, Egypt). Whole soluble tachyzoites antigens were prepared as described by Waltman *et al.* (1984).

The technique was performed as previously described by Voller *et al.* (1976). In brief, ELISA plates were coated over night at 4^{o}C with 10 µg/ml of *T. gondii* antigen diluted in carbonate-bicarbonate buffer (0.1 M, pH 9.6, 100 µl/ well). The plates were washed three times with 0.05% PBS-Tween 20 (PBS-T) and blocked by addition of 100 µl/well bovine serum albumin in PBS for one hour at 37^{o}C. After repeated washing, 100 µl / well of duplicate dilution of sera (positive, negative or tested serum sample) [Sera diluted 1:100 in PBS-T] was added per well and incubated for one hour at 37^{o}C. The sera were removed by repeated washing and 100 µl / well of conjugate, which was anti-sheep/ anti-horse IgG horseradish peroxidase enzyme conjugated then incubated for one hour at 37^{o}C. After three washings of the plates with PBS-T, 100 µl/well of p-nitrophenyl phosphate in substrate buffer were added and re-incubated for one hour at 37^{o}C. Stopping the reaction was done parallel for the tested sera. Results were expressed as OD and the positive threshold value was determined to be equal to the mean of the negative control sera value + two folds the standard deviation.

Statistical analysis:

The obtained data were computed and analyzed by using repeated measures ANOVA and the results were tested for significance using t-test (SAS, 1996). To identify risk factors associated with infection by *T. gondii*, a bivariate analysis was carried out using Chi-square and Fisher's exact tests with significance level of 5%, using the statistical program EPI-INFO, version 3.5.1.

RESULTS

1 - Seroprevalence rate of toxoplasmosis:
Seroprevalence rate of toxoplasmosis were 41.7%, 66.1% and 62% in sheep, 49.4%, 64.2% and 50.6% in goat, 50%, 72.2% and 72.2% in horse and 44.3%, 67.1% and 68.4% in donkeys, by LAT; IHAT and ELISA respectively (Table 1). The peak levels of IHAT titers in both sheep and goats was >1:1280, whereas in horses and donkeys was 1:160.

2 - Bioassay in cats
The results of tissues bioassay in cats revealed that 8 out of 25 slaughtered sheep (32.0%) and 9 out of 25 slaughtered donkeys (36.0%) were positive (Table 2). Oocysts of *T. gondii* were observed after a prepatent period (P.P.P.) 3 – 4 days. The cats continued shedding toxoplasma oocysts for 9 -13 days from the appearance of oocysts (Picture 1).

Table 1: Seroprevalence of toxoplasmosis in sheep, goats, horses and donkeys using LAT, IHAT and ELISA tests.

Species		Total No.	Serological tests						P-value
			LAT		IHA		ELISA		
			+ve	%	+ve	%	+ve	%	
Small ruminants	Sheep	292	122	41.7	193	66.1	181	62.0	0.023
	Goats	81	40	49.4	52	64.2	41	50.6	
	Total	373	162	43.4	245	65.7	222	60.0	
Equine	Horses	54	27	50	39	72.2	39	72.2	<0.000
	Donkeys	79	35	44.3	53	67.1	54	68.4	0.013
	Total	133	62	46.6	92	69.2	93	69.9	

3. Histopathological examination
Histopathological examination of bioassay positive diaphragm muscles revealed *T. gondii* tissue cysts in only 3 (37.5%) sheep samples and 4 (44.4%), donkeys samples (Picture 2).

4. Sensitivity and specificity of different serological tests in relation to bioassay in cats
The comparison between results of serological tests and results of bioassay in cats, the sensitivity of LAT, IHAT and ELISA were 87.5%, 100% and 100% in sheep respectively and 55.7%, 88.9% and 100% in donkeys respectively, while the specificity of LAT, IHAT and ELISA were 76.5%, 100% and 100% in sheep respectively and 93.8%, 93.8% and 100% in donkeys respectively (Table 2).

5. Risk factors associated with the infection with T. gondii in sheep, goats, horses and donkeys in sheep, goats, horses and donkeys:

a. Host: The seropositivty of toxoplasmosis by ELISA was higher in equine (horses 72.2% and donkeys, 68.4%) than in small ruminants (sheep, 62.0% and goats, 50.6%). There was a highly significant differences in T. gondii infection among animal species, seropositivity in horses was 4.88 times (P. value <0.000), in donkeys was 2.54 times (p= 0.013) and in sheep was 1.8 (p=0.023) higher than was in goats.

b. Gender*: T. gondii* infection was significantly higher (P- value < 0.01) in female sheep (67.3%), goats (62.5%), horses (80.0%) and donkeys (90.3%) than in male sheep (48.8%), goats (24.0%), horses (57.9%) and donkeys (73.9%).

c. Age: Toxoplasmosis in small ruminants (sheep and goats) and equine (horses and donkeys) was significantly higher (P- value < 0.05) at age >5 years in sheep (80.7%) [OR = 17.5], goats (66.7%) [OR = 6], horses (89.5%) [OR = 13] and donkeys (83.7%) [OR = 18] than that at age < 2 year sheep (41.6%), goats (31.3%), horses (50.0%) and donkeys (30.8%).

d. Abortion: There was non-significant increase (p. value >0.05) in incidence of toxoplasmosis in aborted ewes 15/17 (88.2%) [OR= 1.22] and goats 5/6 (83.3%) [OR=3.33] than those with normal birth 92/153 (60.1%) and 21/35 (60.0%) in ewes and goats respectively, While in equine, toxoplasmosis in both aborted mares and she-donkeys (75%) was less than in normally birthing mares (79.17%) [OR=0.79] and she-donkeys (86.9%) [OR=0.43].

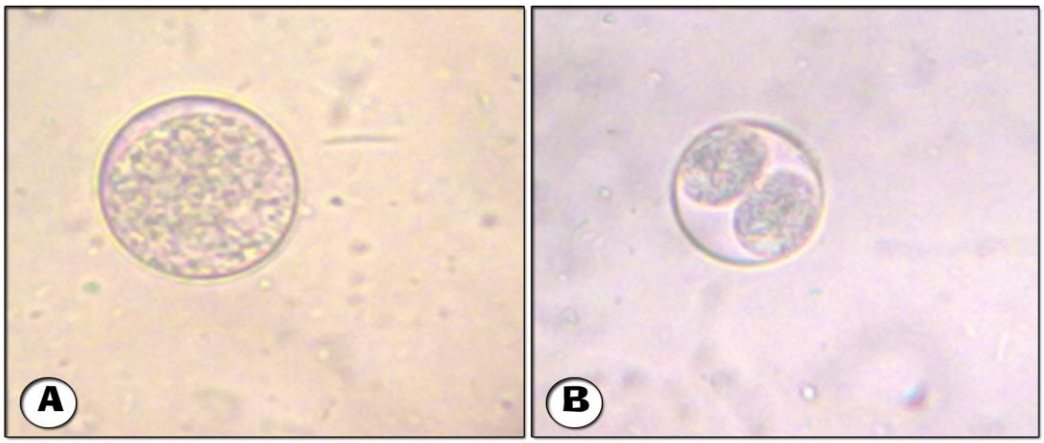
e. Rearing system: Toxoplasmosis was highly significantly increased in sheep and goats which raised in intensive management system (76.4% &56.8%) [P- value, < 0.01] than those raised in extensive (46.5% & 33.3%) and semi-intensive system (62.1% & 46.4%). Moreover sheep & goats reared under intensive system were 2.6 times more likely to have toxoplasmosis than those reared under extensive system [Table 3].

Table 2: Showing sensitivity and specificity of LAT, IHAT and ELISA in slaughtered sheep and donkeys using tissues bioassay in cats as a gold standard test.

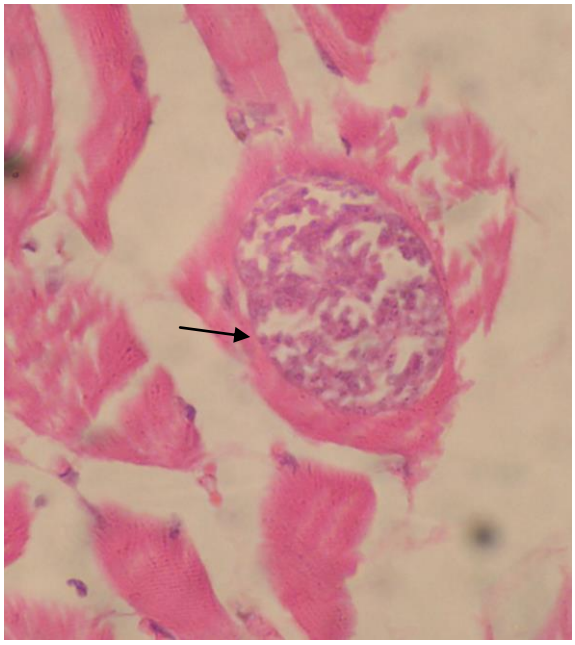
Species	Serological tests	Data	Tissues Bioassay in cats		
			+ve No.	-ve No.	Total
Sheep	LAT	Positive	7	4	11
		Negative	1	13	14
		Total	8	17	25
		Sensitivity		87.50%	
		Specificity		76.50%	
	IHAT	Positive	8	0	8
		Negative	0	17	17
		Total	8	17	25
		Sensitivity		100%	
		Specificity		100%	
	ELISA	Positive	8	0	8
		Negative	0	17	17
		Total	8	17	25
		Sensitivity		100%	
		Specificity		100%	
Donkeys	LAT	Positive	5	1	6
		Negative	4	15	19
		Total	9	16	25
		Sensitivity		55.7%	
		Specificity		93.8%	
	IHAT	Positive	8	1	9
		Negative	1	15	16
		Total	9	16	25
		Sensitivity		88.9%	
		Specificity		93.8%	
	ELISA	Positive	9	0	9
		Negative	0	16	16
		Total	9	16	25
		Sensitivity		100%	
		Specificity		100%	

Table 3: Showing some risk factors related to toxoplasmosis in small ruminants (sheep and goats) and equine (horses and donkeys).

Risk Factors		Sheep					Goats					Horses					Donkeys				
		Total No	+ve	%	OR (95% CI)	P-value	Total No	+ve	%	OR (95% CI)	P-value	Total No	+ve	%	OR (95% CI)	P-value	Total No	+ve	%	OR (95% CI)	P-value
Species		292	181	61.9	1.8	0.023	81	41	50.6	1		54	39	72.2	4.88 (2.11 - 11.28	<0.000	79	54	68.4	2.54 (1.2- 5.3)	0.013
Sex	**Male**	84	41	48.8	1		25	6	24	1		19	11	57.9	1	0.0012	35	20	73.9	1	
	Female	208	140	67.3	2.01 (1.16 - 3.50)	0.0133	56	35	62.5	5.28 (1.82- 15.32)	0.0022	35	28	80	2.91 (0.85 - 9.96)		44	34	90.3	3.29 (073- 1493)	0.0013
Age	**≤ 2 Y**	89	37	41.6	1	0.121	32	10	31.3	1	0.556	14	7	50	1		13	4	30.8	1	
	>2-5 Y	146	98	67.1	5.71 (2.07 – 15.75)	0.001	37	23	62.2	4.93 (1.14 - 21.35)	0.033	21	15	71.4	2.5 (0.61- 10.26)	0.203	23	14	60.9	4.13 (0.49- 3450)	0.039
	≥5 Y	57	46	80.7	17.5 (4.89 – 62.69)	<0.000	12	8	66.7	6 (1.02 - 35.37)	0.048	19	17	89.5	13 (1.32- 128.11)	0.028	43	36	83.7	18(1.9- 2714)	0.013
Pregnancy	**Normal birth**	153	92	60.1	1		35	21	60	1		24	19	79.2	1		23	20	86.9	1	
	Previously aborted	17	15	88.2	1.22 (0.25- 5.90)	0.8	6	5	83.3	3.33 (0.35- 31.66)	0.29	4	3	75	0.79 (0.07- 9.32)	0.85	4	3	75	0.43 (0.03- 5.58)	0.52
Management	**Extensive**	99	46	46.5	1		9	3	33.3	1											
	Semi-intensive	87	54	62.1	1.37 (0.74- 2.54)	0.324	28	13	46.4	1.73 (0.36 - 8.35)	0.493										
	Intensive	106	81	76.4	2.59 (1.38- 4.85)	0.003	44	25	56.8	2.63 (0.58- 11.90)	0.209										



Picture (1): *Toxoplasma gondii* non sporulated oocysts. (A) and *T. gondii* sporulated oocysts (B). X 1200



Picture (2): photomicrograph from diaphragm muscle of sheep showing *T. gondii* tissue cyst (arrow) {H & E. X 1200}.

DISCUSSION

Toxoplasmosis has a very wide geographic distribution and is considered to be one of the most common parasitic infections of man and warm blooded animals. Clinical symptoms of toxoplasmosis are not specific. Therefore, evaluation of serological tests becomes important in order to use sensitive and specific tests in serological surveys (Uggla *et al.,* 1983 and Moreno *et al.,* 1991).

With regard to the seroprevalence of toxoplasmosis, 122 (41.7%), 193 (66.1%) and 181 (62.0%) out of 292 sheep, 40 (49.4%), 52 (64.2%) and 41 (50.6%) out of 81 goats, 27 (50.0%), 39 (72.2%) and 39 (72.2%) out of 54 horses and 35 (44.3%), 53 (67.1%) and 54 (68.4%) out of 79 donkeys were sero-positive against toxoplasmosis by LAT; IHAT and ELISA respectively. The difference in antibody response observed by use of the three serological tests may be in part due to the type of antigen used and in other part due to the class of antibodies measured. These results were concordant with those reported by Shaapan (2005) who found that the seroprevalence of toxoplasmosis in slaughtered sheep were 34 % by DT, 37 % by IFAT, 41.7% by ELISA and 43.7 % by MAT.

High seroprevalence of toxoplasmosis among sheep seems to be catches with those reported previously where 51.5% in Brazil by IFAT (Romanelli *et al.,* 2007); 50.4% and 61.4% in Egypt by LAT and ELISA respectively (Hassanain *et al.,* 2011); 71% in Libya by LAT (Al-mabruk *et al.,* 2013) and 67% in the Caribbean Dominica Island by ELISA (Hamilton *et al.,* 2014). In goats our results were nearly similar to those obtained by several authors where 46.0% in Brazil by IFAT (Carneiro *et al.,* 2009); 50% in Turkey by IHAT (Sevinc *et al.,* 2000a) and 52.8% in Romania by ELISA (Iovu *et al.,* 2012). Moreover in equine similar results were described where 65.3% of donkeys from Monofia province, Egypt by ELISA (El-Ghaysh, 1998); 71.2% of horses in Iran by MAT (Hajialilo *et al.,* 2010).

On the other hand, lower and higher seroprevalence of toxoplasmosis in sheep were observed elsewhere; for examples, 95.7% and 90.9% in Kars province, Turkey by ELISA and SFDT respectively (Mor and Arslan, 2007); 34.5% in Somalia by LAT (Kadle, 2014); 29.9% in Mexico by MAT (Alvarado-Esquivel *et al.,* 2013) and 3.3 % in Iran by IFAT (Derakhshan and Mousavi 2014). In goats lower seroprevalence of toxoplasmosis were reported by Figueiredo *et al.* (2001) in Brazil 18.4% by IHAT; Kandil and Abou-Zeina (2000) in Egypt 39% by ELISA and Derakhshan and Mousavi (2014) in Iran 1.7 % by IFAT, whereas higher results were reprted by Beyhan *et al.* (2013) in Turkey 95.24% by (SFDT), Also variable seroprevalence of toxoplasmosis in equines, were observed elsewhere; for examples, 6.9% of horses in North America (MAT) (Dubey *et al.,* 1999), 7.2% of horses in Turkey (SFDT) (Karatepe *et al.,* 2010); 25% of horses and 23.6% of donkeys in China by MAT (Yang *et al.,* 2013); and 5% and 8% of donkeys in Italy by LAT and IFAT (Machacova *et al.,* 2014). The difference in seroprevalence rates in the present work and the previous studies may be also attributed to abundance of infected cats and their contact with examined animals in farms and slaughtered houses.

With regard to tissue bioassays in cats the results revealed T. gondii from 32.0% & 36.0% of slaughtered sheep and donkeys. These results were in agreement with Esteban-Redondo *et al.* (1999) and Da Silva and Langoni, (2001). Also Shaapan and Ghazy, (2007) found that 52.6% of horses were *T. gondii* infected by bioassays in mice and cats.

Results of histopathological examination of diaphragm of sheep and donkeys, which positive by tissues bioassay in cats, revealed *T. gondii* tissue cysts in only 3out of 8 (37.5%) and 4 out of 9 (44.4%), respectively. These results were in agreement with those recorded by Belbacha *et al.* (2004) and Hassanain *et al*. (2011).

The comparison between results of serological tests and results bioassay in cats, the sensitivity of both ELISA and IHAT in sheep and donkeys were 100%, whereas sensitivity of LAT was 87.5% and 55.7% in sheep and donkeys. Also specificity of ELISA in sheep and donkey were 100%, while specificity of IHAT and LAT were 100% & 76.5% in sheep and were 93.8% & 93.8% in donkey. These results coincided with those detected by Figueiredo *et al.* (2001) who reported that there was a high and significant positive correlation between IHAT versus IFAT, IHAT versus ELISA, and ELISA versus IFAT; Mcload and Remington, (1996) and Puije *et al.* (2000), who reported that ELISA was highly sensitive 92% and specific 91% when compared to IFAT which was used as reference.

With regard to species susceptibility, the seropositivty of toxoplasmosis by ELISA was higher in equine (horses 72.2% and donkeys, 68.4%) than in small ruminants (sheep, 62.0% and goats, 50.6%). The higher percentage of infection in equine may be attributed to the old infections of examined animals, where the peak levels of IHAT titers in horses and donkeys was 1:160 and was >1:1280 in both sheep and goats. These results were in agreement with those reported by Puije *et al.* (2000) in Ghana 32.2% & 26.8% of sheep & goats by ELISA and Ahmed *et al.* (2013) in Sudan 38% and 27.6% of horses and donkeys respectively by LAT. This result differs from those reported by Gondim *et al.* (1999) in Brazil 28.93% and 18.75% in goats and sheep by LAT; Prelezov *et al.* (2008) in Bulgaria 48.2% & 59.8% of sheep and of goats by IHAT and Bocanegra *et al.* (2012) in Spain 10.8% & 25.6% of horses and donkeys by MAT. This variation in species susceptibility may be attributed to difference on the environmental and ecological condition, which effect on the biology of the parasite or the system of breeding and hygienic measures inside farms.

With regard to gender, *T. gondii* infection was significantly higher (P- value < 0.01) in female sheep (67.3%), goats (62.5%), horses (80.0%) and donkeys (90.3%) than in male sheep (48.8%), goats (24.0%), horses (57.9%) and donkeys (73.9%). These were in agreement with those reported by Hajialilo *et al.* (2010); Al-mabruk *et al.* (2013); and Dubey *et al.* (2014). These results were not coincided with Esmat, (1997) and Boughattass *et al.* (2011). This variation may be attributed to the young age of male sheep and goats used in this study.

With regard to age susceptibility, toxoplasmosis was significantly higher (P- value < 0.05) in sheep, goats, horses and donkeys at age >5 years than that at age <1 year. This variation may be due to the exposure of old animals to *T. gondii* oocysts for long periods. These results were in agreement with those observed by Figueiredo *et al.* (2001); Boughattass *et al.* (2011) and Al-mabruk *et al.* (2013).

Moreover toxoplasmosis was significantly higher in sheep and goats which raised in intensive management system (76.4% &56.8%) than those

raised in extensive (46.5% & 33.3%) and semi-intensive system (62.1% & 46.4%). This variation may be related presence of uncontrolled number of stray cats around farms. These results coincided with those recorded by Neto *et al.* (2008); Tzanidakis *et al.* (2012) and Al-mabruk *et al.* (2013).

Toxoplasmosis was highly significantly increased in aborted sheep 15/17 (88.2%) and 5/6 (83.3%) goats than those with normal birth 92/153 (60.1%) and 21/35 (60.0%) respectively. These differences may be due to multiplication of *T.gondii* in placenta and release of antigen into maternal circulation. These results were in agreement with those reported by Aktas *et al.* (2000); Sevinc *et al.* (2000 b) and Hussein *et al.* (2011).

It could be concluded that IHAT in sheep, goats, horses and donkeys could be used as a screening test in seroepidemiological studies, and their results must be confirmed by ELISA. Species, gender, age, rearing system play an important role in incidence of toxoplasmosis. Seroprevalence of *T. gondii* infection indicated wide spread of toxoplasmosis in sheep, goats, horses and donkeys in Dakahlia province. Thus further studies will be needed to clarify the role of sheep, goats, horses and donkeys in transmission of infection to man and pet animals.

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دراسات وبائية عن مرض التوكسوبلازما في المجترات الصغيرة والخيول بمحافظة الدقهلية ، مصر

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تهدف هذه الدراسة إلى تحديد مدى انتشار مرض داء المقوسات (التوكسوبلازما) في المجترات الصغيرة (٢٩٢ أغنام و ٨١ ماعز) وفي الخيول (٥٤ حصان و ٧٩ حمار) من محاظة الدقهلية بمصر في الفترة من اكتوبر ٢٠١٣ الى اكتوبر ٢٠١٤. أظهرت النتائج أن نسبة الإصابة باستخدام إختبارات المقايسة المناعية المرتبط بالإنزيم (**ELISA**)، إختبار التلازن الدموى الغير مباشر (**IHAT**)، إختبار اللثى الغير مباشر (**LAT**) في الأغنام كانت على التوالى ٤١,٧% ، ٦٦,١% ، ٥٠,٦% وفي الماعز كانت على التوالى ٤٩,٤% ، ٦٤,٢% ، ٥٠,٦% وفي الأحصنة كانت على التوالى ٥٠% ، ٧٢,٢% ، ٧٢,٢% وفى الحمير كانت على التوالى ٤٤,٣% ، ٦٧,١% ، ٦٨,٤%. وأظهرت النتائج أيضاً أن ٨ من ٢٥ (٣٢%) من الأغنام المذبوحة و ٩ من ٢٥ (٣٦٪) من الحمير المذبوحة كانت إيجابية للمعايرة الحيوية للأنسجة في القطط (**Bioassay**). وتلاحظ وجود الأطوار النسيجية المتحوصلة **(Tissue cysts)** من طفيل التوكسوبلازما في ٣ فقط من ٨ (٢٤٪) و ٤ من ٩ (٤٤,٤٪) بالفحص الهستوباثولوجي لأنسجة من عضلات الحجاب الحاجز من الأغنام والحمير التي كانت إيجابية بإختبار المعايرة الحيوية للأنسجة في القطط (**Bioassay**) على التوالي. وكانت حساسية إختبارات المقايسة المناعية المرتبط بالإنزيم (**ELISA**)، إختبار التلازن الدموى الغير مباشر (**IHAT**) ١٠٠% في الأغنام والحمير المذبوحة. وبالنسبة لدراسة بعض العوامل ذات الصلة بمرض التوكسوبلازما أوضحت النتائج أن : نسبة الإصابة فى الأحصنة (٧٢,٢%)، والحمير (٦٨,٤%) كانت أكثر من الأغنام (٦٢٪) والماعز (٥٠,٦٪). وفيما يتعلق بالجنس، فإن نسبة عدوى التوكسوبلازما كانت أعلى في الإناث مقارنة بالذكور وكانت هناك زيادة معنوية لنسبة الأصابة في الأغنام والماعز والأحصنة والحمير الكبار في السن أكبر من ٥ سنوات مقارنة بصغار السن أقل من ١ سنة. وكانت هناك ارتباط عالى بين نسبة الإصابة بداء المقوسات والإجهاض في الحيوانات وكذلك نظام التربية المكثفة. ومن هذه الدراسة أتضح لنا أن الخيول والمجترات الصغيرة تلعب دوراً مهماً في وبائية مرض التوكسوبلازما. وأن هناك حاجة إلى مزيد من الدراسات لتوضيح دور الأغنام والماعز والأحصنة والحمير في نقل العدوى للإنسان والحيوانات الأليفة. وأتضح لنا أيضاً أن إختبار التلازن الدموى الغير مباشر (**IHAT**)، حساس ونوعى للكشف ويمكن استخدامه في عمل مسح على الاجسام المضادة للتوكسوبلازما في الأغنام والماعز والأحصنة والحمير ، ويجب تأكيد نتائجه بإستخدام إختبار (**ELISA**).