## DETECTION OF TOXOPLASMA GONDII AMONG WOMEN IN DUHOK CITY-KURDISTAN REGION OF IRAQ

**R**AWA **N**OORI **A**WAD<sup>\*</sup> and **S**HAMAL **A**BDULLAH **A**L-**M**UFFTI<sup>\*\*</sup>

<sup>\*</sup> Dept. of Microbiology and Parasitology, College of veterinary medicine, University of Duhok,

Kurdistan Region-Iraq

\*\* Dept. of Biology, College of Science, University of Duhok, Kurdistan Region-Iraq

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#### ABSTRACT

Background: Congenital toxoplasmosis is one of the most endemic parasitic infections that can lead to clinical complications by transmission through mother to fetus transplacentally in aborted women.

Objectives: The aim of this study was to estimate the rate of toxoplasmosis among pregnant and aborted women in Duhok/ Kurdistan Region-Iraq by using Enzyme-Linked ImmunoSorbent Assay(ELISA) and detect the seropositive cases by Real-Time Polymerase Chain Reaction (RT-PCR).

Materials and Methods: A total of 102 samples were collected from women admitted to obstetrics and gynecology hospital in Duhok province. The data were collected through the period from January, 20<sup>th</sup>, 2019 until December, 30<sup>th</sup>, 2019.Blood and placental samples from 60 aborted women with Bad Obstetric History(BOH) and 42 placental samples from healthy normal pregnancies (as a control group). The placental samples of normal pregnancies were tested by RT-PCR technique while aborted women's blood and placental samples were tested by both Enzyme-Linked ImmunoSorbent Assay(ELISA) and RT-PCR techniques.

Results: The results revealed a low rate of infection by congenital toxoplasmosis. Seropositivity to *T. gondii* were 20/60 33.3% by ELISA, 25% IgM, 65% IgG and 10% both IgG and IgM whereas the retested seropositive samples by RT-PCR only 5% showed positive results. The highest rate of infection was at 32-38 age group, first trimester 85%, single abortion 35%, who inhibits rural area 80%,ingesting homemade food 80%, and contact with animals(cats) 85%.

Conclusion: Real-time PCR-based technique is the most accurate and suitable for routine screening, providing a rapid, sensitive, and quantitative way of detecting congenital toxoplasmosis in clinical specimens, without any false-positive result that may help to a targeted treatment.

KEYWORDS: Toxoplasma gondii, Aborted women, ELISA, RT-PCR.

## INTRODUCTION

Toxoplasmosis is a disease caused by parasitic protozoan used as a routine diagnosis of toxoplasmosis and early detection of infection and to prevent later newborn complications of congenital toxoplasmosis that might impose expensive costs for treatment (1). Toxoplasma gondii, of one the cosmopolitan's common parasites and the most endemic contagious agents in Iraqi aborted women (1).It is а single-cell tissue protozoan coccidian enigmatic parasite. It is an obligatory intracellular protozoan zoonotic with a heterogeneous life cycle in humans and other vertebrates (2).

Toxoplasmosis is one of the most prevalent parasitic diseases where almost one-third of the world's population is infected, approximately 25 to 30% of the world's population is infected by *Toxoplasma gondii* (3).On the other hand, many studies in the world and Iraq in particular, have revealed that toxoplasmosis causes recurrent abortion among pregnant women (4).

Congenital toxoplasmosis referred to a clinical state of this disease in the fetus that following an acute primary infection acquired by the mother through pregnancy (5). Although *T. gondii* infection is usually uncomplicated and asymptomatic in immunocompetent individuals, it is a potentially serious issue when acquired during pregnancy as it carries the risk transmission to the fetal in about 30% of cases and congenital toxoplasmosis may lead to manifestations varying in severity, depending on the stage of pregnancy at the time of maternal infection, most of them involving the brain and eyes (6).

In pregnancy, the most common routes of acquiring toxoplasmosis infection are through ingesting raw or undercooked meats or contaminated water with sporulated oocysts, or direct exposure to soil or cat litter. Transfusion or organ transplantation from an infected person with toxoplasmosis can also transmit the organism (7,8,9).

The gestational age at which the infection is developed is a key variable affecting the clinical fetal outcome (10).Severe toxoplasmosis occurs in immunocompromised patients that build up either acute infection or reactivation from quiescent tissue cysts (11). It can also cause symptoms similar to flu or glandular fever, sometimes including swollen lymph nodes. Once a person has had the infection they are generally thought to be protected for the rest of their lives, unless they suffer an impairment of their immune system (12).

Serological diagnostic techniques such as ELISA considered as most the commonly used approach to detect the infectious stage if it is current, recent (acute), past (chronic) (13).There are numerous diagnosis techniques but the molecular methods such as PCR amplification have been recognized as excellent and sensitive techniques to diagnose congenital toxoplasmosis in immunodeficient patients in much reduced time(14). Most researchers have used the *B1* gene for detection *T. gondii* in a variety of biological samples (15).The technique can identify tachyzoites in amniotic fluid (AF), placenta, and cord blood (16).

Serological recognition of specific anti-*Toxoplasma* immunoglobulin is a primarily serological technique to detect *T. gondii* infection varies in sensitivity and specificity depending on commercially obtainable serologic kits which may fail to detect infection, particularly in some immunocompromised patients (17). A recent study in Iraq showed that detection of *T. gondii* by molecular methods RT-PCR is crucial for diagnosing congenital toxoplasmosis. Real-time PCR can be used as an additional diagnostic tool for the rapid accurate detection of *T. gondii* in various clinical materials such as the placenta (4).

## MATERIALS AND METHODS

A total of 102 samples were collected from all patients under sterilized conditions to prevent any possible contamination. Out of these 102 cases in ranged age 18-45 years, 42 placentas collected from full-term normal were pregnancies have been chosen randomly as a control group, while the other 60 placentas and blood samples were collected from aborted women with BOH and spontaneous miscarriage. All were attending Duhok Gynecology and Obstetrics Hospital. From each patient, 50 mg of placental tissue soaked with Phosphate Buffered Saline (PBS) buffer Then samples will be stored at -20 C until be used for DNA extraction by using QIAamp DNA tissue Kit from QIAgen Inc. Hilden, Germany, Catalog No. 51306 while 3 ml venous blood samples from aborted women only by using sterile disposable hypodermic vacationer needles and tubes. Venous blood centrifuged at 14,000 rpm for 20 min to obtain serum and stored at -20°C until use. A questionnaire-based on personal information was prepared, to obtain the important factor(s) effect on the epidemiology of the Toxoplasma gondii containing all required variables for data analysis. IgM and IgG anti-T. gondii antibodies were detected in serum samples of aborted women by using ELISA kit Bioactiva Diagnostica, Germany. While placentas were tested by using RT-PCR Ecoli Toxoplasma gondii kit, Russia and Toxoplasma gondii RT-PCR kit, Anatolia gene works, Turkey.

## **Statistical Analysis:**

Data were analyzed using SPSS program version 18. Results with *P*-value< 0.05 was considered to be statistically significant. Chi square test was used to indicate the agreement of studied parameters and tests.

#### RESULTS

The results of the current study revealed that aborted women were at low rates of infection by congenital toxoplasmosis. Table.1 demonstrates the participated groups of all 102 females that were enrolled for this study.

Examined groups variables	Groups	Aborted	women	Pregnan	it women	Tota	al
		N=60	%	N=42	%	N=102	%
Age (year)	18-24	23	38.3	13	30.95	36	35.3
_	25-31	16	26.7	20	47.6	36	35.3
-	32-38	18	30	8	19.05	26	25.3
_	39-45	3	5	1	2.4	20	3.9
Residency	Urban	16	26.7	15	35.7	31	30.4
	Rural	44	73.3	27	64.3	71	69.6
Occupation	Employed	3	5	5	11.9	8	7.8
	House wife	57	95	37	88.1	94	92.2
Educational level	Literate	31	51.7	25	59.5	56	54.9
-	Illiterate	29	48.3	17	40.5	46	45.1
Number of previous	None	12	20	16	38.1	28	27.5
pregnancies	P1	10	16.7	7	16.7	17	16.7
_	P2	13	21.7	4	9.5	17	16.7
_	P3	10	16.7	5	11.9	15	14.7
	P4 and more	15	25	10	23.8	25	24.5
Number of previous	A1	38	63.3	38	90.5	76	74.5
abortions	A2	11	18.3	2	4.8	13	12.7
-	A3	3	5	1	2.4	4	3.9
	A4 and more	8	13.3	1	2.4	9	8.8
Gestational age	T1	40	66.7	-	-	40	39.2
	T2	18	30	-	-	18	17.6
	Т3	2	3.3	42	100	44	43.1
Repeated abortion consecutive	yes	60	100	4	9.5	64	62.7
	No	-	-	38	90.5	38	37.3
Previously diagnosed for toxoplasmosis	Yes	7	11.7	3	7.1	10	9.8
_	No	53	88.3	39	92.9	92	90.2
Previously treated for toxoplasmosis	Yes	7	11.7	3	7.1	10	9.8
-	No	53	88.3	39	92.9	92	90.2
Are there any congenital abnormalities in fetus	Yes	7	11.7	1	2.4	8	7.8
-	No	53	88.3	41	97.6	94	92.2
Contact with cats	Yes	34	56.7	6	14.3	40	39.2
-	No	26	43.3	36	85.7	62	60.8
Food habit	Homemade	51	85	32	76.2	83	81.4
-	Restaurant	8	13.3	5	11.9	13	12.7

# **Table (1):** Variables that affect the epidemiology of the *Toxoplasma gondii* among Women in Duhok province.

	Vegetarian	1	1.7	5	11.9	6	5.9
Family size	1-5	12	20	-	-	12	11.8
_	5-10	48	80	4	9.5	52	50.9
	11 and more	-	-	38	90.5	38	37.3
Pre-existing medical disease	Diabetes	2	3.3	2	4.8	4	3.9
	B.P	3	5	3	7.1	6	5.9
	None	55	91.7	37	88.1	92	90.2
Uterine abnormalities	Yes	11	18.3	7	16.7	18	17.6
	No	49	81.7	35	83.3	84	82.4
Family history of genetic	Diabetes	25	41.7	10	23.8	35	34.3
disease	B.P	16	26.7	6	14.3	22	21.6
_	None	19	31.7	26	61.9	45	44.1

Forty-two (41.2%) placental samples collected from control married women with no bad obstetric history were examined by Realtime PCR technique after DNA extraction to detect *Toxoplasma gondii*. All control samples showed negative results. While the other 60 samples (58.8%) (Blood and placentas) have had bad obstetric history early or later in their pregnancy with single or recurrent consecutive abortions. Serum samples were examined by the ELISA technique as a primary diagnostic test. Serum of 20(33.3%) aborted women showed seropositive results referred to a higher seroprevalence of anti-Toxoplasma IgG than IgM antibodies, 25% (5/20) were IgM antibodies and 65%(13/20) were IgG antibodies, only 2 cases (10%) were both IgM and IgG antibodies respectively (Table 2).

**Table (2):** Distribution of studied groups according to the anti-*Toxoplasma* antibodies using ELISA.

Test		Serop	ositive samples			Serone	Total Examined		
	lgM	lgG	lgG & lgM	Total	lgM	lgG	lgG &lgM	Total	Aborted women
ELISA	5	13	2	20	55	47	58	40	60
%	25	65	10	100	91.7	78.3	96.7	66.7	100

Based on the finding of the current study, this result showed the highest rate of toxoplasmosis IgG antibodies 60% was recorded at the age

group 32-38 years old patients by ELISA. The lowest rate of infection was 5% in the age group 18-24 years of old patients (Table 3).

Table (3): Detection of toxoplasmosis using ELISA among aborted women in according to age.

Age (year)	No. of Non In	fected Aborted	No. of Infected Aborted women		Total No. of ex	amined Aborted
	wo	men			wo	omen
18-24	22	55%	1	5%	23	38.3%
25-31	12	30%	4	20%	16	26.7%
32-38	6	15%	12	60%	18	30%
39-45	0	0%	3	15%	3	5%
Total	40	66.7%	20	33.3%	60	100%

 $(\chi^2)=24.196$ (Significant), *df*=3, p value=0.00003



Also, the results showed the toxoplasmosis in women who lived in rural areas 80% was higher than urban area 20% (Figure 1).

Fig. (1): ELISA test detection of congenital toxoplasmosis among aborted women in relation to residence area

It was found that all anti-Toxoplasma IgG and IgM antibodies in aborted women were housewives. According to food habits, aborted

40

Total

66.7

20

women who depend on homemade food showed higher seropositive results 80% while none at vegetarians. (Table 4)

100

**No Significant** 

<b>Table (4):</b> De	<b>Table (4):</b> Detection of toxoplasmosis using ELISA among aborted women according to food habit.										
Food habit	Seronegative samples		it Seronegative Seropositive Total Examined Aborted women samples samples		S	cs					
							X <sup>2</sup> Test	df	<i>p</i> -value		
	NO.	%	NO.	%	NO.	%					
Homemade	35	87.5	16	80	51	85					
Restaurants	4	10	4	20	8	13.3	1.588	2	0.45		
Vegetarian	1	2.5	0	0	1	1.7					

60

The toxoplasmosis in illiterate aborted women 70% was higher than literate 30%. (Table 5)

33.3

Educational level	Seronegative samples		Seronegative         Seropositive         Total Examined Aborted           samples         samples         women		Statistics				
							X <sup>2</sup> Test	df	<i>p</i> -value
	NO.	%	NO.	%	NO.	%			
Literate	25	62.5 <b>%</b>	6	30%	31	51.7 <b>%</b>			
Illiterate	15	37.5 <b>%</b>	14	70%	29	48.3 <b>%</b>	5.6396	1	0.0176
Total	40	66.7 <b>%</b>	20	33.3 <b>%</b>	60	100 <b>%</b>	Si	gnifica	nt

Table (5): Detection of toxoplasmosis using ELISA among aborted women according to educational level

This study showed that the highest rate of toxoplasmosis IgG antibodies were in women with 4 previous pregnancies and more 55%. Whereas, all aborted women with no kids were seronegative. (Table 6)

24

Number of previous	Seronegative S samples		ve Seropositive samples		Total Examine	Total Examined Aborted women			Statistics			
pregnancies							X <sup>2</sup> Test	df	<i>p</i> -value			
	NO.	%	NO.	%	NO.	%	_					
None	12	30	0	0	12	20	_					
Once	9	22.5	1	5	10	16.7	_					
Twice	8	20	5	25	13	21.7	19.454	4	0.0006			
Triple	7	17.5	3	15	10	16.7	_					
Four and More	4	10	11	55	15	25	—					
Total	40	66.7	20	33.3	60	100	Sig	gnifica	int			

 Table (6): Detection of congenital toxoplasmosis using ELISA among aborted women according to the frequency of previous pregnancies.

Results revealed that aborted women with single 35%, four abortions and more 35% have the highest level of anti-*Toxoplasma* antibodies

and lower levels in aborted women with double 20% and triple 10% abortions (Table 7).

 Table (7): Detection of toxoplasmosis using ELISA among aborted women according to the frequency of previous abortions.

Number of previous abortions	Seronegative samples		Serop sarr	Seropositive samples		ined Aborted men	Statistics		
							X <sup>2</sup> Test	df	<i>p-</i> value
	NO.	%	NO.	%	NO.	%			
Once	31	77.5	7	35	38	63.3			
Twice	7	17.5	4	20	11	18.3	15.911	3	0.0012
Triple	1	2.5	2	10	3	5			
four and more	1	2.5	7	35	8	13.3			
Total	40	66.7	20	33.3	60	100	Si	gnifica	int

The highest rate of infection 85% was at first trimester gestational age aborted women,

15% second trimester and none in the third trimester. (Table 8)

Table (8): Detection of toxoplasmosis using ELISA among aborted women in according to gestational age.

Gestational age	Seronegative samples		Sero sa	Seropositive T samples		amined Aborted women	Statistics		
							X <sup>2</sup> Test	df	<i>p</i> -value
	NO.	%	NO.	%	NO.	%	_		
T1	23	57.5 <b>%</b>	17	85 <b>%</b>	40	66.7 <b>%</b>	_		
T2	15	37.5 <b>%</b>	3	15 <b>%</b>	18	30%	4.7625	2	0.0924
Т3	2	5%	0	0%	2	3.3%	_		
Total	40	66.7 <b>%</b>	20	33.3 <b>%</b>	60	100 <b>%</b>	No	Signific	ant

Showed low results 30% at those who were previously diagnosed and treated for toxoplasmosis. (Table 9).

d'outrient.									
Previously treated for toxoplasmosis	Seronegative samples		Serop sam	oositive nples	Total E: Aborted	xamined I women	Statistics		
							X <sup>2</sup> Test	df	<i>p-</i> value
	NO.	%	NO.	%	NO.	%			
Yes	1	2.5	6	30	7	11.7	-		
No	39	97.5	14	70	53	88.3	9.7844	1	0.002
Total	40	66.7	20	33.3	60	100	Significant		nt

 Table (9): Detection of toxoplasmosis using ELISA among aborted women in according to the treatment.

High rates at those who showed congenital abnormalities were (6/7) cases 30% in their fetus. (Table 10)

 Table (10): Detection of toxoplasmosis using ELISA among aborted womenaccording to congenital abnormalities in the fetus.

Congenital abnormalities in the fetus	Seronegative samples		Seropositive samples		Total Exami wor	ned Aborted men	Statistics		
							X <sup>2</sup> Test	df	<i>p-</i> value
	NO.	%	NO.	%	NO.	%			
Yes	1	2.5	6	30	7	11.7			
No	39	97.5	14	70	53	88.3	9.7844	1	0.002
Total	40	66.7	20	33.3	60	100	Sig	Inifica	nt

The current study showed a significant difference in seropositivity at those who have direct contact with cats 85%. (Table 11)

Table (11): Detection of toxoplasmosis using ELISA among aborted women according to contact with	i
animals	

Contact with animals	Seronegative samples		Seropositive samples		Total Examined Aborted women		Statistics		
						-	X <sup>2</sup> Test	df	<i>p</i> -value
	NO.	%	NO.	%	NO.	%			
Contact	17	42.5	17	85	34	56.7			
No contact	23	57.5	3	15	26	43.3	9.8077	1	0.002
Total	40	66.7	20	33.3	60	100	Significant		

According to family size aborted women who have1-5memberfamilies gave low seropositive results 5% while the highest was in 5-10, 95% member families, and none seropositive cases with aborted women of two member families (Table 12).

Family size	seronegative seropositive Total Examined Aborte		ned Aborted nen	Statistics					
							X <sup>2</sup> Test	df	<i>p-</i> value
	NO.	%	NO.	%	NO.	%	_		
1-5 members	11	27.5	1	5	12	20	_		
5-10 members	29	72.5	19	95	48	80	4.2188	1	0.04
11 and more	0	0	0	0	0	0	_		
Total	40	66.7	20	33.3	60	100	Significant		int

 Table (12): Detection of toxoplasmosis using ELISA among aborted women according to family size.

All 20 seropositive ELISA cases were retested by a real-time PCR technique. Out of 20 cases, only one case 5% showed positive results for both tests (ELISA IgM and IgG, RT-PCR),(Table 13)who's 32 years old live in camps uneducated housewife, with two children and four previous consecutive abortions, aborted

her two months fetus with congenital abnormalities (Hydrocephaly) due to congenital toxoplasmosis. Also, this case lived in family consists of four members, has direct contact with cats, and depends on homemade food, with no medical history and uterine abnormalities.

Table13: Distribution of studied groups according to the anti-*Toxoplasma* antibodies and RT-PCR.

Test	Positive samples		Negative samples		Total		Statistics		
	NO.	%	NO.	%	NO.	%	X <sup>2</sup> Test	df	<i>p</i> -value
ELISA	20	95.3	40	39.6	60	49.2	21.53	1	0.000003
RT-PCR	1	4.8	61	60.4	62	50.8			
							Significant		

## DISCUSSION

In the past five years, many researchers in Iraq and Kurdistan particularly conducted many diagnostic techniques for early detecting of congenital toxoplasmosis in pregnant and aborted women and applying the most recent tests and procedures to reach no doubted results, includes many serological and molecular techniques. The traditional diagnosis of congenital toxoplasmosis usually depends on serological tests such as ELISA that has many limitations of detection (18).

Real-time PCR which is the best-performing accurate technique till now that has been described as a modern high sensitive assay for early detection of minimal amounts of parasitic DNA and latent infection, produces the quantitative results in different types of samples such as blood and placental tissue (19).

In the present study, all control samples 41.2% were tested by Real-Time PCR and

showed negative results. They were completely clear of Toxoplasma gondii DNA. Aborted BOH 58.8% blood samples were tested by ECLIA, 33.3% of cases were seropositive to different antibodies. 25% were IgM seropositive referring recent acute congenital toxoplasmosis to infection and 65% were higher IgG seropositive than IgM referring to latent chronic infection and presence of tissue cyst, only two cases10% were both IgM and IgG seropositive. Detection by ELISA technique helps to differentiate time of infection by detection IgM antibodies (rise in one or two weeks), While IgG antibodies referred to the previous infection has happened already but does not tell if it is recent and past or latent infection (4).Based on our findings, results revealed that aborted women under age 25 age of higher fertility 5% seem less to have congenital toxoplasmosis (CT) while showing a high prevalence of 60% of this parasitemia among 32-38 age group showing a significant difference. Also, this indicates that the infection

getting increased by age, older age more exposure, our results were in agreement with a previous study in Thi Qar (1), And another study in Duhok province, that showed higher seropositivity in aborted women over 30 years old(20), And acquiring congenital toxoplasmosis in the 25-31 age group was more than other ages (21), And disagree with a study in Babylon revealed high prevalence in aborted women at the age group of 21-25 (5).

Toxoplasmosis infection in patients who lived in rural areas 80% was higher than urban area 20%. This result leads to a significant correlation in the location of residency and congenital toxoplasmosis infection. Our findings may due to low-temperature weather and low humidity that minimize the survival of oocysts and then minimizing the probability of transmission and reduce the infection titers to the lowest. A study in the United Kingdome reported that toxoplasmosis infection was higher in rural areas than urban as by this study (22).A contrary study in Iran reported that higher prevalence was in urban areas and city dwellers(23).

According to our study, all toxoplasmosis IgG and IgM seropositive aborted women were housewives whom their natural habits mostly depend on homemade food and restaurants 80%, 20% respectively, while all-vegetarian aborted and pregnant women showed negative results. We observed that congenital toxoplasmosis infection associated with ingestion of undercooked and cured meat as a basic infectious risk factor, our findings were consistent with another study in England that found an association between toxoplasmosis and consumption of raw meat(24). Toxoplasma gondii has been recognized as the foodborne pathogen connected with the second-highest public health impact both in the USA and Netherlands, and this supporting our study(25),(26), our findings were in contrast with a study performed in Iran that showed there is no relationship between ingestion the raw meat and toxoplasmosis infection(23).

The toxoplasmosis infection in illiterate aborted women was higher than literate in relation to hygienic consideration and awareness, 70%, 30% respectively; showing the significant relationship of *Toxoplasma gondii* infection and education, our study was consistent with a study conducted in Iran(27). This study showed no significant relationship in the number of children and toxoplasmosis infection, that the highest rate of toxoplasmosis IgG seropositive were in women with four previous pregnancies 55% and more referring to a previous infection, while all aborted women with no kids were negative.

One of our important findings, there was a significant relationship between the number of abortions previous and congenital toxoplasmosis, revealed that the highest rate of congenital Toxoplasmosis was at women with single 35% and four abortions and more 35%, and lower levels in aborted women with double 20% and triple 10% abortions, might lead to recurrent abortions. Our study when in contrast with a study conducted in Babylon that suggested that toxoplasmosis doesn't cause recurrent abortion(5), While it was confirming Darweesh et al.,(4) study that reported percentage of infection related to a number of abortions.

Toxoplasmosis infected abortions were recorded the highest in the first trimester 85%,15% second trimester and none in the third because Toxoplasma trimester gondii Tachyzoites will attack embryo at embryogenesis leading to miscarriage of fetus. Our result was in contrast with a study conducted in Ethiopia were the highest rates in the third trimester(28), But in agreement with another study in Saudi Arabia(29)

Seropositivity of aborted women who showed congenital abnormalities were 6/7 cases 30% in their fetus, referring to serious consequences affecting the fetus caused by congenital toxoplasmosis infection. Showed low results 30% at those who were previously diagnosed toxoplasmosis. treated for These and seropositive results may due to less effective treatment. Our study showed a significant difference in seropositivity at those who have direct contact with animals 85%, cats especially because they are the definitive host sheds the oocysts. We found that cats and birds like chicken were associated with toxoplasmosis infection. Our study came in agreement with an Ethiopian study that was done in 2016(30), and it was inconsistent with another study conducted in Mexico(31). According to family size aborted women who have 1-5 member families gave low positive results 5% while the highest was in 5-10, 95% members families and none positive cases with aborted women of two-member families but we didn't find any relation between family size and toxoplasmosis infection.

Since serological testing has been one of the major diagnostic techniques for toxoplasmosis but it has many limitations and almost fails and gives false-positive results. Real-time PCR technique was used to confirm the diagnosis by detection B1 gene of Toxoplasma gondii DNA in placental tissue samples of aborted women. B1 gene, although of unknown function, is mostly utilized in a variety of diagnostic and epidemiological researches thanks to its specificity and sensitivity (32).

All 20 seropositive ELISA cases were tested by Real-Time PCR technique. Out of 20 cases, only 5% showed positive results as in Figure.2, for both tests (ELISA IgM and IgG, RT-PCR), who's 32 years old live in camps uneducated housewife, with two children and four previous consecutive abortions, aborted her two months fetus with congenital abnormalities (Hydrocephaly) due to congenital toxoplasmosis parasitemia. Also, this case lived in family consists of four members, has direct contact with animals (cats), and depends on homemade food, with no medical history and uterine abnormalities. Our study was inconsistent with AL-Dujaily et al. who used both ELISA and RT-PCR techniques as a confirmatory tool (33).

Our study was in agreement with Darweesh *et al.*, (4) in Diyala/Iraq and Mousavi *et al.*, (19) in Iran that reported 2 positive cases by RT-PCR who were seropositive for both IgG and IgM by ELISA test. According to our results, a key point should be noticed, 19 seropositive cases showed negative results by the RT-PCR test, this clears the fact that the RT-PCR technique is the highest in sensitivity till now.

Real-time PCR assay as a sensitive accurate diagnostic tool provides an early approach for molecular detection of the acute and chronic phases of infection in clinical samples without any false-positive result that may help to a targeted treatment. Also, this method may be appropriate for screening of *T. gondii* infection in special cases such as immunocompromised groups who usually fail to produce specific IgM or increased IgG titers(19).We conclude RT-PCR methods in aborted women were the best diagnostic technique in detecting the recent and reactive and latent infection by *T. gondii*.

The promising results described in this study lead to the fact that, in the past 5 years due to the increase of pregnant women's awareness and strength of health educational programs we reduced to the minimal number of congenital toxoplasmosis cases in Duhok province.

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دياركرنا نه خوشيا توكسوپلازمايى ل باژێرێ دھوكێ ھەرێما كوردستانا عيراقێ

پوخته

زانیاریێن سەرەتایی: ئێشا توکسوپلازمایی ئێکه ژ وان ئێشێن مشەخور یێن دوم درێژ کۆ دبنه ئەگەرێ ڨەرێژێن کلینیکی, ڨەگوھاستن ژ دایکێ بو زاروکی د زك ماك دا ئەوژی ب رێکا مال بچویك لدەف وان ئافرەتێن بچویك ژبەرچووی ل عیراقێ.

مەرەم و ئارمانجا قەكولىنىّ: ئارمانج ژ ئەقى قەكولىنىّ دىڧچوون و دەستنىشانكرنا مشەخورى Toxoplasma gondii ھەر ژ بۆ دياركرنا ئاستىّ بەلاڤبوونا نەخوشيىّ دناڤ ئاڧرەتيّن دووگيان و ييّن بچوويك ژبەر چووى , لدھوكىّ – ھەريّما كوردستانا عيراقىّ. ھەروەسا ڤەديتن و ئاشكراكرنا DNAييّت مشەخورى Toxoplasma gondii ژ مال بچويك ييّن نەخوشيّن جورا وجور بكارئينانا تەكنيكا تيككاركرن زنجيرَين ئەنزيمىّ پەلمەركرنى يا ريەل تايم(RT-PCR).

ئەنجام: ئەنجام دياربوون كۆ رێژا توشبوونێ ب ئێشا توكسوپلازمايێ يا زك ماك يا كێمه, ئانكو بكارئينانا ئيلايزا 60/20 (33.3%)، %25 بو تەنێن ھەڤدژ IgM،%65 بو تەنێن ھەڤدژ , IgG و10% بو تەنێن ھەڤدژ IgG و IgG. لێ رێژا توشبوونێ ب تەكنولوجيا گەرديلەيى 5% بو.بلندترين رێژا توشبوونێ يا ئەوێ كومێ بو ئەوێن ژيێ وان دناڤبەرا 32-38 ساليێ دا , رێژا توشبونێ 85% بوو ل ئافرەتێن دوو گيان ل ھەرسێ ھەيڤێن ئێكێ يێن دوو گيانيێ , و ئەوێن ئێك جار بتنێ بەرھەڤايتى ب رێژا

% 35 توشبون , و ئەو ئافرەتێن گوندشين ب رێژا توشبونێ 80% و ئەو ئافرەتێن بەردەوام خارنا مال بكاردئينن رێژه 80% و يێن تێكەلێ و پەيوەنديا ئێكسەر دگەل ئاژەلان ھەى رێژا وان 85 % بوو .

دەرئەنجام: تەكنەلوجيا تێككاركرنا زنجيرَين ئەنزيمێ پەلمەركرنێ يا ريەل تايم دهێتە هژمارتن باشترين تەكنەلوجيا هويريێن و گونجاى و بلەز و ھەستياريەكا بەرز ھەى د قەديتن و پشكێنينا روويى دگەل دەستنيشانكرن و ئاشكەراكرنا ئێشا توكسوپلازمايى يا زك ماك ل نوموونێن كلينيكى بێى هيچ ئەنجامێن خەلەت. تەكنەلوجيا تێككاركرنا زنجيرَين ئەنزيمێ پەلمەركرنێ يا ريەل تايم دڨێت بهێتە بكارئينان ژبۆ دەسنيشانكرن و ئاشكەراكرنا پێشوەخت يا نەخوشيا توكسوپلازما ژبو دويركەفتنێ ژ وان قەرێژێن ژ ئەگەرێ ئێشا توكسوپلامايى پەيدا دبيت ل زارۆكێن تازە ژدايك بووى و دێ بيتە ئەگەرێ كێم كرنا تێچوويێن چارەسەريا گران بها. الكشف عن داء المقوسات بين نساء مدينة دهوك- أقليم كوردستان العراق خلفية وأهداف البحث

## الخلاصة

داء المقوسات هو واحد من اكثر الأمراض الطفيلية المزمنة التي تؤدي الى مضاعفات سريرية عن طريق الانتقال من الأم الى الجنين عن طريق المشيمة لدى النساء المجهضات في العراق.الغرض من الدراسة الحالية هو مسح وتشخيص طفيلي *Toxoplasma gondii* لتقديرانتشار داء المقوسات بين النساء الحوامل والمجهضات في دهوك- أقليم كوردستان العراقواستخلاص والكشف عن الحمض النووي للطفيلي *Toxoplasma gondii* من المشيمة لمرضى مختلفين بأستخدام تقنية تفاعل أنزيم البلمرة المتسلسل في الوقت الحقيقي في دهوك- أقليم كوردستان العراق.

طرق البحث: تم جمع 102 عينة من النساء اللواتي يرتدن مستشفى النسائية والتوليد في محافظة دهوك.تم جمع البيانات خلال الفترة من (كانون الاول 2019-كانون الثاني 2019). العينات كانت تشمل الدم والمشيمة جمعت من 60 أمرأة مجهضة ذات تاريخ ولادي سيء و 42 عينة مشيمة نساء حوامل اصحاء. تم فحص مشيمات النساء الحوامل بتقنية تفاعل أنزيم البلمرة المتسلسل في الوقت الحقيقي بينما عينات الدم والمشيمة تم فحصها بأستخدام كلا التقنيتين ألايلايزا وتفاعل أنزيم البلمرة المتسلسل في الوقت الحقيقى.

النتائج: كشفت النتائج عن نسب اصابة منخفضة بداء المقوسات الولادي.الايجابية المصلية لطفيلي .*T* و 30% للجسم المضاد *gondii* و 1gM و 1gM و 25% للجسم المضاد JgG و 01% للجسم لكلا الجسمين المضادين JgM و JgG. اما نسبة الاصابة بتقنية الجزيئية كانت 3%. اعلى نسب الأصابة كانت للمجموعة العمرية 25-38 وفي الاشهر الثلاثة الاولى من الحمل بنسبة 3%. و ذوات حالة اسقاط واحدة بنسبة 35% و الـذيـن يسكنو المناطق الريفية بنسبة 38% واللواتي يتناولن الطعام المنزلى بنسبة 38% واللواتى على تواصل مباشر مع الحيوانات بنسبة 38%.

الأستنتاج: تقنية تفاعل أنزيم البلمرة المتسلسل في الوقت الحقيقي هي اكثر تقنية دقيقة ومناسبة وذات سرعة وحساسية عالية للكشف الروتيني وتشخيص داء المقوسات الولادي في العينات الطبية بدون اي نتائج خاطئة للحصول على العلاج المناسب والكشف المبكر عن الاصابة لتجنب المضاعفات الناتجة عن داء المقوسات التي قد تصيب حديثي الولادة والتي قد تؤدي الى تكاليف باهضة للعلاج.