

## Epidemiological Study of Toxoplasmosis And Cytomegalovirus in Pregnant Women in Kerbala

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**Key words:** Toxoplasmosis, Cytomegalovirus, Mini VIDAS technique ,birth defect

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### Abstract

**Objectives:** Identifying the seroprevalence of acute anti-*Toxoplasma gondii* and anti-CMV IgM and chronic IgG antibodies, the environmental and behavioral factors and some socio demographic data and influencing CMV as co-factor to infected with *Toxoplasma gondii* of the study sample.

**Methods:** A descriptive cross sectional study done on 240 pregnant women(13-43 years old). All the studied women were interviewed and blood samples were taken and used Minividas technique for detection antibodies (IgM and IgG) .

**Results:** The seroprevalence of anti-*Toxoplasma gondii* antibody was (28.3%) for chronic Toxoplasmosis (IgG),and (0.8%) for acute Toxoplasmosis (IgM).While results for Cytomegalovirus (CMV)infection revealed that (92.08% ) positive for chronic CMV (IgG) and (0.8%) positive for acute CMV(IgM) ,The peak age for acquisition of the infection was (20-29 year old ) , There was a statistical significant association between seroprevalence with cat exposure ,revealed this results significant association between Toxoplasmosis and CMV infection and influencing CMV as co-factor to infected with *Toxoplasma gondii* and proved this results significant association between seroprevalence Toxoplasmosis and CMV infection with birth defect.

دراسة وبائية لداء القطط والفيروس المضخم للخلايا في النساء الحوامل في محافظة كربلاء

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### الخلاصة

**هدف البحث :** تحديد انتشار الإصابة بداء المقوسات القندية (داء القطط) والفيروس المضخم للخلايا والعوامل الاجتماعية، البيئية والسلوكية المرتبطة بها وكذلك علاقة الفيروس كعامل مساعد يسهل الإصابة بداء المقوسات القندية لدى عينة البحث .

**طريقة العمل :**نوع الدراسة هو وصفية مقطعية لدى عينة من (240 ) أمراه حامل تتراوح أعمارهن بين (١٦ - ٤٣ سنة ).جمع المعلومات كان بالمقابلة الشخصية واخذ عينة من الدم من عينة البحث.

**النتائج :** أظهرت النتائج أن الانتشار المصلي للأجسام المضادة من النوع "ج"في مصول العينات المسحوبة من النساء الحوامل ,والتي تدل على الاصابه المزمنة للمقوسات القندية كانت 28.3% (61 سيده من إجمالي 240) وللفيروس المضخم للخلايا هي 92.2% (221 سيده من إجمالي 240) بينما الانتشار المصلي للأجسام المضادة

من النوع "م"، والتي تدل على الاصابه الحادة للمرض هي 0.8% (سيدتان من إجمالي 240) لداء المقوسات القندية أما الفيروس المضخم للخلايا كانت النسبه هي 0.8% . وبرهنت النتائج إلى وجود نسبة إصابة أعلى لدى النساء التي كانت أعمارهن بين (20-29) سنة و إن معدل الانتشار المصلي للاصابه بداء القطط يترايط ترابطا ايجابيا مع عوامل خطورة أخرى مثل التماس مع القطط ، وكذلك أظهرت نتائج الدراسة إن هناك علاقة بين الاصابه بالفيروس المضخم للخلايا وتأثيره كعامل مساعد في الاصابه بداء القطط وكما بينت الدراسة إن هنالك ترابطا ايجابيا بين الاصابه بداء القطط والفيروس المضخم للخلايا وولادة أطفال مشوهين خلقيا حيث شاهدت الدراسة إن أصابه النساء بهذين المرضين ممكن أن يزيد من حالات أصابه الاجنه بتشوهات خلقيه.

**الاستنتاجات:** هناك نسبه كبيره من الاصابه بالفيروس المضخم للخلايا وداء القطط عند النساء الحوامل في مدينه كربلاء وهناك حاجه للتحري الروتيني لكل النساء الحوامل لتفادي المخاطر التي تلحق بالأأم الحامل وطفلها وأيضا يجب التركيز على نشر الوعي الصحي لدى النساء الحوامل من خلال البرامج التثقيفية في التلفاز وشبكه الانترنت وغيرها.

## **Introduction**

*Toxoplasma gondii* (*T. gondii*) and *Cytomegalovirus* (*CMV*) have direct effect on the fetus leading to spontaneous abortion, still birth or congenital anomalies<sup>[1 and 2]</sup>. The risk and severity of the fetus infection depend on the timing of the mother's infection. Previous studies showed that when mothers are infected with *CMV* or *T. gondii*, particularly in the first trimester of pregnancy, lead to damage the nervous system of the fetus or abortion<sup>[3 and 4]</sup>.

Both *human cytomegalovirus* (*HCMV*) and protozoon *T. gondii* are most common human pathogens and Depending many factors on hygiene standards and habits and socio-demographic status parameters, the prevalence rates of *CMV* and *T. gondii* in different countries vary between 40–100 % and 20–70 % respectively<sup>[5]</sup>. *T. gondii* is transmitted either by consumption of undercooked contaminated meat or exposure to *T. gondii* oocysts in cat feces. On the other hand, transmission of *CMV* occurs by acquisitions free virus by close contact with person shedding the virus in body fluids including urine ,saliva ,breast milk, vaginal secretion and semen<sup>[5]</sup>. Congenital *CMV* infection is common disease leading to serious damage like hearing loss and mental retardation, while congenital toxoplasmosis is known for its association with visual impairment, chorioretinitis, hydrocephalus, and mental retardation<sup>[6]</sup>. Postnatal acquired infections in immunocompetent child or adult has been describe harmless and asymptomatic and probably stay dormant for life –long<sup>[1 and 2]</sup>. However, latent *CMV* or *T. gondii* infections can be reactivated in immunocompromised patients, e.g. *AIDS*, immunosuppressive drugs and transplant organic recipients<sup>[7 and 8]</sup>. The reactivation latent infection may lead to dissemination of *CMV* or *T. gondii* to various tissues, like nervous system<sup>[9]</sup>. The present study aims to gave screen study about seroprevalence of *T. gondii* and *CMV* infection in pregnant women in Karbala governorate and determing the relationship between *T. gondii* and *CMV* infections with some environmental and behavioral factors such as: a-age, , cat exposure , birth defects .

## **Materials and Methods**

This prospective study was conducted in Gynecological Hospital and Obstetric of Kerbala province , in laboratory Department during the period from January 2014 through March 2014. Samples were collected from 240 pregnant women attending antenatal clinic , their ages ranged between 16 to 43 years. Before sample taking from female, and after their agreement, the questionnaire was filled which included the age, family address, water source, economical status, educational level, gestational age, no. of previous pregnancy and birth defects, as well as other questions according to questionnaire sheets. About 5 ml of blood was collected by vein puncture using sterile syringe with needle gauge 23, then the blood sample was transfer into gel tube and allowed to clot at room temperature from which sera was separated after centrifugation at 4000 rpm for 5 min, then the serum was examined for anti-*T. gondii* (IgG, IgM) antibodies and anti-CMV (IgG, IgM) antibodies Kits by using Mini VIDAS technique (Biomerieux / France). The serum was stored and frozen at (-20C) during sample collection for immunological study.

## **Statistical analysis**

The data was put on computer file for storage and analysis. Descriptive statistics included the use of frequencies, relative frequencies, means, standard deviations and ranges .The Chi-Square statistical test was used to test for associations between variables with the results being considered as statistically significant when the P value was  $\leq 0.05$  SPSS statistical package version 20 was used for data description and analysis. Fisher exact test or Yates correction formula was applied whenever applicable.

## **Results**

In a prospective study conducted in Karbala governorate and include 240 pregnant women that attending to Gynecological Hospital and Obstetric for detection seroprevalence of Toxoplasmosis and Cytomegalovirus infection by using MiniVIDAS technique. The results revealed that the prevalence rate with Toxoplasmosis was (29.1)and (92.88)with Cytomegalovirus infection .These rate distributed as (28.3%) for chronic Toxoplasmosis (IgG), and (0.8%) for acute Toxoplasmosis (IgM).While results for Cytomegalovirus(CMV)infection revealed that (92.08% ) positive for chronic CMV (IgG) and (0.8%) positive for acute CMV(IgM) as shown in **Table (1)**.

In **Table(2)** illustrated the distribution of Toxoplasmosis among the age groups, they showed the highest number of the seropositivity cases was (43) among age group  $\leq 20$  to 29 years, while the lowest number of these cases (5) was among age group 40 to 49 years. And when compared to negative cases the association was found to be statistically not significant relationship between Toxoplasmosis and age groups(  $p= 0.323$ ).

illustrated in **Table (3)** seroprevalence of anti- *Toxoplasma* IgG antibody in relation to cat exposure and was found the seroprevalence of *Toxoplasma* infection 21(42%)of 50 than those whose exposed to cat were seropositivity to IgG while (47)(26.9%) of 190 non exposed women and when compared with negative cases we

found statistically significant between cat exposure and seropositivity to anti-*Toxoplasma* IgG ( $p=0.025$ ). Regarding to the anti-CMV and anti-*Toxoplasma gondii* (IgG) antibodies detection in the pregnant women serum as shown in this study were noticed statistically significant association between CMV infection and toxoplasmosis ( $p=0.039$ ), 67 (98.35%) of than 68 those whose seropositivity to toxoplasmosis were same patients exposed to CMV infection while from 154 (98.53) those whose seronegativity to toxoplasmosis were 18 (10.47) of than 19 seronegativity to CMV infection as shown in **Table (4)**.

Demonstration in **Table(5)** the seroprevalence of anti-*T.gondii* (IgG) anti-body relationship to birth defects ,was showed seropositivity of anti-*T. gondii* (IgG) anti-body 6 (12.5) of 48 whose pregnant women have child born suffered from anomalies and when compared with seronegativity cases 42 (87.5) was showed statistically significant  $p=(0.011)$  ,while in CMV infection was noticed highly significant between CMV infection and birth defect ( $p=0.0000002^{**}$ ).

**Table (1): The seroprevalence rate *Toxoplasma gondii* (*T.gondii*) and Cytomegalovirus (CMV) anti-bodies among the study samples .**

Anti- <i>Toxoplasma gondii</i> and Anti-CMV antibodies			Number (N0.)	Percent (%)
Anti- <i>T. gondii</i> anti-bodies	IgG	Positive	68	28.3
		Negative	172	71.7
		Total	240	100.0
	IgM	Positive	2	0.8
		Negative	238	99.2
		Total	240	100.0
Anti- CMV anti-bodies	IgG	Positive	221	92.08
		Negative	19	7.92
		Total	240	100.0
	IgM	Positive	2	0.8
		Negative	238	99.2
		Total	240	100.0

**Table(2): The distribution of anti -*T.gondii* (IgG) anti-bodies according to the age groups.**

Age groups	Anti- <i>T.gondii</i> (IgG)					
	Positive		Negative		Total	
	N0	%	N0	%	N0	%
<20	7	10.29	27	15.70	34	14.17
20-29	43	63.24	92	53.49	135	56.25
30-39	13	19.12	45	26.16	58	24.17
40-49	5	7.35	8	4.65	13	5.42
Total	68	100.00	172	100.00	240	100.00

$$\chi^2=3.485 \text{ df}=3 \text{ p}= 0.323$$

**Table3:seroprevalence of Anti-*T.gondii* (IgG) anti-bodies according to the cat exposure .**

Anti - <i>T.gondii</i> (IgG)	Cat exposure						P.V
	Yes		No		Total		
	(N0.)	%	(N0.)	%	N0	%	
Positive	21	42	47	26.9	68	28.3	0.025*
Negative	29	58	143	73.1	172	71.7	
Total	50	100.0	190	100.0	240	100.0	

Yates corrected  $\chi^2= 4.99$  df=1 p=0.025\*

**Table (4):distribution study of CMV infection and correlation with Toxoplasmosis infection in the studied samples.**

Anti-CMV(IgG)	Anti- <i>T. gondii</i> (IgG)					
	Positive		Negative		Total	
	(N0.)	%	(N0.)	%	(N0.)	%
Positive	67	98.53	154	89.53	221	92.08
Negative	1	1.47	18	10.47	19	7.92
Total	68	100	172	100	240	100

Yates corrected  $\chi^2= 4.24$  df=1 p=0.039\*

**Table (5) : The seroprevalence of anti-*T. gondii* (IgG) anti-body according to birth defects .**

		birth defects						P.V
		NO		yes		Total		
		(N0.)	%	(N0.)	%	(N0.)	%	
anti- <i>T.gondii</i> (IgG) anti-body	Positive	62	32.3	6	12.5	68	28.3	0.011*
	Negative	130	67.7	42	87.5	172	71.7	
	Total	192	100.0	48	100.0	240	100	
anti-CMV (IgG) anti-body	Positive	187	97.4	34	70.8	221	92.1	0.0000002**
	Negative	5	2.6	14	29.2	19	7.9	
	Total	192	100.0	48	100.0	240	100	

Fisher exact test

## Discussion

The prevalence of anti- *T. gondii* antibody as shown in **Table(1)** was agreement with seroprevalence data from previous studies conducted in Salah – Aden governorate by (ADdory, 2011) who found that seropositivity Anti-*T.gondii* (IgG) was (26.1%) of cases while seropositivity for (IgM) antibody was (3.1%) of cases and both was (29.2%)<sup>[10]</sup>.

The seropositive rate in this study was lower than study recorded by Al-Jebouri, M. et al.,(2013) which he was found the high prevalence of toxoplasmosis among the

investigated high risk women at Al-Hawija and at Al-Baiji was (42%)<sup>[11]</sup>. This may be due to the difference in the availability of optimum environmental conditions among countries and different socio-demographic regions in the same country for survive and spread the parasite in addition to the presence of more than one risk factor influencing the occurrence of toxoplasmosis as the habits of people and the sanitary conditions and variation in the sensitivity of different immunoassays. This may explain the variation in seropositivity<sup>[12]</sup>. While prevalence *CMV* infection in current study, all research work from different parts of the world had proven that almost all people have had *CMV* infection during their life from early childhood and become persistent<sup>[13 and 14]</sup>, this result close to be similar were mentioned by Salih Ahmed et al.,(2013)in Sulaimani city (Iraq) who recorded seropositivity for (IgG) was (90.2%)<sup>[15]</sup>. In this current study it was observed pregnant women who afflicted with *CMV* infection were high percentages in many studies that reflected high prevalence rate among Iraq pregnant women because different modes of *CMV* transmission from person to person particularly between children who is shedding *CMV* in their all bodily fluids. Poor socioeconomic conditions that are characterized by overcrowding and a lack of hand hygiene, and placing children in daycare facilities, transmission from mother to infant by breast feeding that promote *CMV* transmission<sup>[16]</sup>. Our results in **Table (2)** indicated the great risk group of pregnant women with toxoplasmosis at the age group between 20 – 29 year and this result agreement to result recorded by ADdory, A.Z.R.(2011)in AL- Muthana province / Iraq was found that the largest age group between (20-29) year<sup>[10]</sup>. This finding may reflect that age group is consider most marriage age for Iraqi female and in this age optimum activity regarding reproduction and fertility in additionally that age are active working adult age group and so have more chance of contact with (raw meat through cooking and soil through gardening). In **Table (3)** the results were agreement with Tarik (2012) who found a strong association between exposure to cats (out door or indoor) and seropositivity to toxoplasmosis ( $p = 0.0001$ )<sup>[17]</sup>. This results were disagreement with the result recorded by Hussein Aqeely.et al., (2014) who showed no significant association between *T. gondii* infection and presence of domestic cats<sup>[18]</sup>. Cats rearing and stray cats are widely spread in Karbala province, this may increase chance of contact with cats litter or contaminated food or water by parasite oocysts and this may explain the association between seropositivity and exposure to cats. In **Table (4)** the results were agreement with I Akyar (2011)who found in childbearing age women were examined the seropositivity of *T. gondii* 24.6% for IgG, and the accompanying as co-infections other pathogens *CMV*, *EBV*, *HCV* and *Rubella*<sup>[19]</sup>. is endemic in the world and seroprevalence of *HCMV* varies greatly depending on geographical location and socioeconomic status. Additionally *HCMV* of an important cause to enhance suppression of the immune response and predisposes to other opportunistic infections<sup>[20 and 21]</sup>. This may be an explanation with *HCMV* infection facilitated and accelerate the infection with opportunistic toxoplasmosis. In **Table (5)** the results in agreement with results recorded by Gilbert, R et al.,(2003), McLeod, Ret al., (2006), Oz, and Tobin, et al.,(2012)<sup>[22,23 and 24]</sup> were found association between seroprevalence of anti-*T. gondii* (IgG) anti-body and severe complications microcephaly, encephalitis, neurological abnormality, blindness, cardiovascular abnormalities, and another birth defects. This result may explained role *T. gondii* in birth defects or may be this birth defects result to many causes such as chromosomal abnormalities and exposure to the environmental pollution and if these

pollutants accumulate in the body it may cause teratogenic effect on the developing embryo or fetus or another many infectious agent or mistaken drug abuse during the period pregnancy<sup>[25]</sup>.

### Conclusions

The highest prevalence rate of anti-CMV IgG and IgM antibodies (92.88%) has been found among pregnant in Karbala province. The highest prevalence of *T.gondii* and CMV infections has been found in the age groups (20-29) years old. According to present study , has been found significant associated between seropositivity of CMV infection, toxoplasmosis and child anomalies .CMV may be lead to other opportunistic pathogens like toxoplasmosis and other because infection with CMV lead to immunosuppression and this facilitated to other pathogens. Public health prevention campaigns by Shed more light for TORCH as STD , and their effect on health of pregnant and child for both ,by using media of different types like Internet, Television..etc. and should focus on the appropriate risk factors mainly on cat exposure, and attention to personal hygiene especially hand washing with soap and water.

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### References

- [1] Turbadkar D, Mathur M, Rele M., (2003) Seroprevalence of TORCH infection in bad obstetric history *Indian ,J Med Microbio*,:108-111.
- [2] Jones, J.L.; Kruszon-Moran, D.; Wilson, M.; McQuillan, G.; Navin, T. and McAuley, J.B,(2001) *Toxoplasma gondii* infection in the United States: Seroprevalence and risk factors, **Am. J. Epidemiol.**,(154):357-365.
- [3] Stagno, S. Cytomegalovirus. In: Remington, J. S. and Klein, J. O.(eds.) . (2001) Infectious diseases of the fetus and newborn infant, **Philadelphia**:389–424.
- [4] Britt,W.J., (1996) Human cytomegalovirus overview The virus and its pathogenicmechanisms,**BaillieresClin.Infect.Dis.**,(3): 307-325.
- [5] Jones, J.; Lopez, A. and Wilson, M.,(2003) Congenital toxoplasmosis, **Am. Fam. Phys.**, (67): 2131–2138.
- [6] Novotná, M. ;Hanusova, J. ;Klose, J. ;Preiss, M. ;Havlicek, J.*et al.* ,(2005) Probable neuroimmunological link between *Toxoplasma* and Cytomegalovirus infections and personality changes in the human host, **BMC Infectious Diseases.**, (5): 54.
- [7] Abu-Madi MA, Behnke JM, Dabritz HA.,( 2010) *Toxoplasma gondii* seropositivity and co-infection with TORCH pathogens in high-risk patients from Qatar, **American Journal of Tropical Medicine and Hygiene**, 82(4): 626–633.

- [8] Ross, D.S.; Jones, J.L. and Lynch, M.F., (2006) Maternal Toxoplasmosis, Cytomegalovirus, Listeriosis, and Preconception Care, **Child Health J.**, (10): 189 -193.
- [9] Da-Cunha, S, Ferreira, E, Ramos, I, Martins, R, De, F. L, Borges, J. L, *et al.*, (1994) Cerebral toxoplasmosis after renal transplantation. Case report and review, **Acta. Med. Port.**, (7): 61-66.
- [10] Adory, A.Z.R ., (2011) Seroepidemiological study of Toxoplasmosis among pregnant women in Salah –Aden government, **Tikrit Medical Journal** ., 17(1):64- 7.
- [11] Mohemid Al-Jebouri ,Mohanad Al-Janabi, Hassan Ismail., (2013) The prevalence of toxoplasmosis among female patients in Al-Hawija and Al-Baiji Districts in Iraq, **Open Journal of Epidemiology.**, (3) :85-88.
- [12] Alvarado-Esquivel C, Sifuentes-Alvarez A, Narro-Duarte S G, Estrada Martinez S, Diaz- Garcia J H, Liesenfeld, *et al.*, (2006) Seroepidemiology of *T.gondii* infection in pregnant women in a public hospital in Northern Mexico, **BMC Infect Dis.**, 6:113.
- [13] Cheung, AN. and Ng IO., (1993) *Cytomegalovirus* infection of the gastrointestinal tract in non-AIDS patients, **Am. J. Gastroenterol.**, 88(11):1882-1886.
- [14] Vancikova, Z. and P. Dvorak., (2001) *Cytomegalovirus* Infection in Immunocompetent and Immunocompromised Individuals- a review, **Curr Drug Targets Immune Endocr Disord.**, 1(2): 179-187.
- [15] Salih Ahmed Hama and , Kazhal J. Abdurahman., (2013) *Human Cytomegalovirus* IgG and IgM Seropositivity among Pregnant Women in Sulaimani City and Their Relations to the Abortion Rates **Curr. Res. J. Biol. Sci.**, 5(4):161-167 .
- [16] Dowd JB , Aiello AE , Alley DE ., (2009 ) Socioeconomic disparities in the seroprevalence of *cytomegalovirus* infection in the US population :NHANES III **Epidemiol Infect.**, (137):58–65.
- [17] Tarik Mahdi Muslim., (2010) Seroprevalence and Associated Factors of Toxoplasma Infection among Sample of Pregnant Women in Wassit Governorate" Iraq. M.sc .thesis ,College of Health and Medical Technologies, Baghdad, Iraq.
- [18] Hussein Aqeely, Eman K. El-Gayar, Darakhshan Perveen Khan, *et al.* , (2014) Seroepidemiology of *Toxoplasma gondii* amongst Pregnant Women in Jazan Province, Saudi Arabia, **Journal of Tropical Medicine**, p. 6.
- [19] I Akyar ., (2011) Seroprevalence and Co infections of *Toxoplasma gondii* in Childbearing Age Women in Turkey, **Iran J Public Health.**, 40(1):63–67.
- [20] Rubin, R. H., (1990) Impact of *cytomegalovirus* infection on organ transplant recipients, **Rev Infect Dis.**, (12)7: 754-766.
- [21] Falagas, M. E., D. R. Snyderman, *et al.*, (1996) Exposure to *cytomegalovirus* from the donated organ is a risk factor for bacteremia in orthotopic liver transplant recipients. Boston Center for Liver Transplantation CMV IG Study Group, **Clin Infect Dis**, 23(3), :468-474.
- [22] Gilbert, R., and Gras, L. , (2003) Effect of timing and type of treatment on the risk of mother to child transmission of *Toxoplasma gondii*, **Bjog**, (110):112–120.
- [23] McLeod, R., Boyer, K., Karrison, T., Kasza, K., Swisher, C., Roizen, N., *et al.*, (2006) Outcome of treatment for congenital toxoplasmosis, 1981–2004: the national collaborative chicao-based, congenital toxoplasmosis study, **Clin. Infect. Dis.**, (42) :1383–1394.

[24] Oz, H. S., and Tobin, T. Diclazuril,(2014) protects against maternal gastrointestinal syndrome and congenital toxoplasmosis,**Int. J. Clin. Med.**, (5):.93–101.

[25] Snijders, R.J. ; Sundberg, K. and Holzgreve, W.,(1999) Maternal age and gestation-specific risk for trisomy 21: effect of previous affected pregnancy. **Ultrasound Obstet,Gynecol.**,(13):167– 170.