SEROSURVEY OF ANTI-TOXOCARA ANTIBODIES AND RISK FACTORS IN RELATION TO VITAMIN D LEVELS AMONG PREGNANT WOMEN OF DUHOK CITY –KURDISTAN REGION

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ABSTRACT

Background: Toxocariasis is a common parasitic disease which is caused by Toxocara Spp. with a worldwide distribution, which causes visceral larval migrans as loss of vision, hypereosinophilia, encephalitis, hepatomegaly, and lungs damage.

Materials and Methods: The blood samples were collected randomly from 150 pregnant women attending maternity hospital in Duhok city, Kurdistan Region. A special questionnaire form is arranged for each woman including the following information's (age, residency, occupation, family income, paternity, contact with cats, presence of domestic cats and dogs, working in home garden, source of drinking water, washing hands before meals, consumption of raw meat).

The blood was used to investigate the blood parameters as (WBCs, lymphocytes, RBCs, Hb., HCT, PCV, and Platelets) on the other hand, in other tubes, the blood was centrifuged to obtain sera for the detection of both anti-Toxocara antibodies by using ELISA IgG test, and vitamin D level.

Results: Out of the 150 pregnant women, only 18/150 (12%) were seropositivity to anti-Toxocara antibodies; 10 out of the 18 (55.5%) pregnancies have Vitamin D deficiency. While 132 pregnancies were seronegative to anti-Toxocara antibodies, 69 out of 132 (52.3%) had Vitamin D deficiency. The seroprevalence of anti –Toxocara was higher in rural patients than urban. High infection rates were indicated in housewives 15(12.6%) than employed one 3(9.6%). Statical significance and association was indicated related to the educational level with *P*-value less than 0.05. High rates of infection showed in illiterate pregnants with 13(20.6%).

Conclusion: The data show that Toxocara seropositivity is a significant health problem. However, insufficient attention is still being paid to this disease.

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Keywords: Pregnancy, Toxocara antibodies, Vitamin D.

 \mathbf{T} oxocariasis is a zoonotic disease that is caused by both Toxocara cati and Toxocara canis parasites infect both cats and dogs, respectively. It has a cosmopolitan distribution and tends to be the most prevalent disease in the tropical regions, which are considered as the cause of most forms of helminthiasis¹.

Transmission mainly occurs through soil contamination with embryonated eggs classifying toxocariasis as a soiltransmitted disease², also contact with dogs and cats because of the presence of the eggs in their fur, the consumption of raw vegetables grown in contaminated gardens, and undercooked meat from paratenic hosts have also been described as essential ways of transmission of this disease³,⁴. Stray dogs and cats in low income populations and poor environments have an important role in the transmission⁵.

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Many Toxocara infections are asymptomatic, although infection in humans can have different symptoms and signs and mostly include asymptomatic, asthma, loss of vision, hypereosinophilia, encephalitis with many problems of the lungs. the CNS, liver. and manv complications as ocular larva migrans, cerebral larva migrans, cutaneous larva migrans, and visceral larva migrans (mainly liver and $lungs)^6$. The risk of acquiring infection is increased with pets' ownership, geographical location as rural areas, and poor socioeconomic status⁷. The serodiagnosis of toxocariasis depends on using mostly ELISA methods to detect anti - Toxocara antibodies in the blood⁸.

MATERIALS AND METHODS

The current study was carried out in a maternity hospital in Duhok city, Kurdistan Region. A total of 150 blood samples were collected randomly from pregnant women, a special questionnaire forma is arranged for each woman including the following information (age, residency, occupation, family income, paternity, contact with cats, presence of domestic cats and dogs, working in home garden, source of drinking water, washing hands before meals, consumption of raw meat).

Ethical approval Scientific and Ethical approvals for the study were granted by the Scientific Committee of the College of Medicine/ Duhok University (Scientific & ethical approval).

The exclusion criteria for those pregnant women whom not included in the current study as those with other infectious and non-infectious diseases, immune suppressed and those with chronic diseases. A volume of 5ml of the blood was obtained by vein puncture.

The blood samples were placed in two plain tubes labeled and named, the first tube with anticoagulants for detection of blood parameters as (RBCs, WBCs, Hb, lymphocytes, HCT, PCV, and Platelets) while the second tube was without anticoagulants and left standing for 20 minutes at room temperature to clot. Then each tube was centrifuged at 3000 rpm for 5 minutes to collect the serum⁹.

Each separated sera was poured into a sterile Eppendorf 1 ml tube each tube was labeled and named then stored at -200C because at this temperature its suitable for the sera will be adequate and active for long time, until being screened for the detection of both anti –Toxocara cati antibody by using an enzyme-linked immune assay (ELISA) kit (Bioactiva, Germany).

Detection of vitamin–D level also by using the kit (Bioactiva, Germany). A positive result for anti-Toxocara antibodies titers was determined when the titer value more than 11 Um, and the negative result was indicated when the titer value less than 11Um.

RESULTS

The distribution of all examined pregnant women tested for anti- Toxocara cati antibodies are listed in the table(1)

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Table 1: Seropositivity of anti – Toxocara cati IgG antibodies relating to studied characters.						
Variables		n.	%	+v IgG Ab	%	P-value
Age	18-24	44	29.3	6	13.6	0.561
	25-31	68	45.3	8	11.7	
	32-38	26	17.3	4	15.3	_
	>39	12	8	0	0	
Residency	Rural	45	30	7	15.5	0.546
	Urban	105	70	11	10.4	
Occupation	Housewife	119	79.3	15	12.6	0.891
	Employed	31	20.6	3	9.6	
Education	Illiterate	61	40.6	13	20.6	0.017
	Primary level	41	27.3	4	9.7	
	Secondary	33	22	0	0	
	College	15	10	1	6.6	
Paternity	Primary	30	20	3	10	0.311
	Multi (1-4)	95	63.3	14	14.7	
	Grand multi > 5	25	16.6	1	4.0	
Family income	Low	67	44.6	9	13.4	0.775
	Moderate	58	38.8	7	12.0	_
	High	25	16.6	2	8.0	_
Work in garden	Yes	43	28.6	5	11.6	1
	No	107	71.3	13	12.1	_
Contact with cats and dogs	Yes	20	13.3	2	10	1
	No	130	86.6	16	12.3	_
Pets ownership	Yes	24	16	3	12.5	1
	No	126	84	15	11.9	_
Hands washing before	Yes	115	76.6	14	12.1	1
meals	No	35	23.3	4	11.4	_
Tape water drinking	Yes	122	81.3	15	12.2	1
	No	28	18.6	3	10.7	_
Raw meat consumption	Yes	32	21.3	3	9.3	0.834
	No	118	78.6	15	12.7	1

The high rates of anti- Toxocara antibodies were detected among (32-38) years of 4/26 (15.3%), no infection was in the age (39-45) as it showed in Table 1.

Regarding the residency, the serosurvey of anti- Toxocara in rural patients was more than in urban as 7/45(15.5%) and 11/105 (10.4%), respectively. According to the occupation, more infection in housewives than employed 15/119(12.6%), 3/31(9.6%) respectively.

In the educational levels, the P-value was 0.017, representing statistical significance when (*P*-value < 0.05) and indicated a relationship between educational level and Toxocara infection. High rates were detected among illiterate group 13/61 (20.6%).

Regarding the paternity and income, high infection was noticed at multi paternity with low income 14/95(14.7%) and 9/67(13.4%) respectively. In comparison,

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low rates showed in the grand multi paternity and high income of 1/25 (4%) and 2/25(8%), respectively. In the case of garden working, there was nearby for non-garden working and garden working 13/107 (12.1%) and 5/ 43(11.6%). Contact to cats and dogs infection rates had been obtained about 16/130 (12.3%) with no contact and 2/20(10%) with contact. Due to pets owners of 3/24(12.5%) and nonowners of 15/126 (11.9%). Near rates were obtained in both

hand washing and non as 14/115(12.1%)and 4/35 (11.4%), respectively. Due to the water source use, using tape water was high rates of 15/122 (12.2%), while not use with 3/28(10.7%). According to uncooked meat consumption, no consumption is 15/118(12.7%), and raw meat consumption is 3/32(9.3%).

Vitamin D levels with Toxocara seropositivity in table (2).

Table 2:	Vitamin -D levels	relatin	g to serop	ositivit	y of anti-1	oxocara Ig	G antibodi	es
Variables				N	ormal	Vitan	nin D	P-value
Age	18-24			Vitamin D		deficiency		
	10-24	n.	%	n.	%	n.	%	1
		6	13.6	3	50	3	13.6	
	25-31	8	11.7	3	37.5	5	62.5	
	32-38	4	15.3	2	50	2	50	
	> 39	0	0	0	0	0	0	
Residency	Rural	7	15.5	3	42.8	4	57.1	1
	Urban	11	10.4	5	45.4	6	54.5	
	House wife	15	12.6	7	46.6	8	53.3	1
Occupation	Employed	3	9.6	1	33.3	2	66.6	
Education	Illiterate	13	20.6	6	46.1	7	53.8	1
	Primary	4	9.7	2	50	2	50	
	Secondary	0	0	0	0	0	0	
	College	1	6.6	0	0	1	100	
Paternity	Primy	3	10	2	66.6	1	33.3	1
	Multi (1-4)	14	14.7	5	35.7	9	64.2	
	Grand multi > 5	1	4.0	0	0	1	100	
Family income	Low	9	13.4	3	33.3	6	66.6	0.198
	Moderate	7	12.0	5	71.4	3	42.8	
	High	2	8.0	0	0	1	50	
Work in garden	Yes	5	11.6	3	60	2	40	0.768
	No	13	12.1	5	38.4	8	61.5	
Contact with cats and dogs	Yes	2	10	0	0	2	100	0.557
	No	16	12.3	8	50	8	50	
Pets ownership	Yes	3	12.5	2	66.6	1	33.3	0.832
	No	15	11.9	6	37.5	9	60	
Hands washing	Yes	14	12.1	5	35.7	9	64.2	0.409
	No	4	11.4	3	75	1	25	
Tap water	Yes	15	12.2	7	46.6	8	53.3	1
drinking	No	3	10.7	1	33.3	2	66.6	
Raw meat	Yes	3	9.3	1	33.3	2	66.6	1
consumption	No	15	12.7	7	46.6	8	53.3	

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All of the infected and non infected preganants had tested for VD levels. Due to the age, equal rates of normal VD 50% in (18-24) and (32-38) while VD deficiency was high at (25-31) 5/8(62.5%). Urban women were with more normal VD than rural ones with 5/11(45.4%), while rural women were with high VD deficiency of 4/7(57.1%)

In occupational status, the normal VD was in housewives than employed high 7/15(46.6%) and 1/3(33.3%) respectively, the employed women with high VD deficiency 2/3(66.6%). In the case of the primary educational levels, similar rates of 50% for both normal and deficient VD. The paternity and income, the normal VD was high in primy 2/3 (66.6%) and high VD deficiency in grand multi 1/1 (100%) and family income moderate income with high the normal VD 5/7 (71.4%) and VD deficiency was more with low income 6/9 (66.6%).

High rates of normal VD work in the garden 3/5(60%) of VD deficiency in the seropositive case was with not washing hands 8/13(61.5%).

Relating to cats and dogs and ownership, same rates of 8/16(50%) for both normal and deficient VD in non-pet contact of while 2/3(66.6%) of pets owners with normal VD, while 9/15(60%) of deficient VD belonged to non-pets owners.

While high rates of normal VD were obtained in drinking tap water of 7/15(46.6%) and high deficient VD in non-tap water drinkers of 2/3(66.6%). Normal rates of VD 7/15(46.6%) of no consume of and 2/3(66.6%) of raw meat consumers with deficient VD.

The hematological parameters in Toxocara seropositive and seronegative cases are described in table 3. It is shown that WBCs and Seropositive cases corresponding to seronegative one due to the detection of the patients with Toxocara.

Table 3: Blood parameters in both Toxocara seropositive and seronegative cases.					
Parameters	Seropositive $x \pm SE$	Seronegative $(x \pm SE)$			
WBCs	9.7± 3.8	8.2±3.0			
RBCs	4.4±1.8	3.4 ± 0.7			
Hb.	12.3± 1.4	10.7±2.1			
Platelets	228 ± 58.4	136±71.2			
Lymphocytes	4.4 ± 2.1	2.6±1.9			
НСТ	35±6.3	30.8±5.5			



Histogram shows the infected and non infected rates with toxocariasis among pregnant women. Its obvious from the histogram that the rate of infection among pregnant women about (12.0%) and the rate of non infection among pregnant women about (88.0%).

DISCUSSION

Our data show that Toxocara seropositivity is an important health problem. However, insufficient attention is still being paid to this disease. The serological tests are useful in the diagnosis of toxocariasis with particular symptomatic or indistinguishable clinical manifestations. They are not routinely used, so most of the toxocariasis cases are under-diagnosed in clinical practice.

Out of 150 individuals, only 18/150(12%)have seropositivity with Toxocara cati antibodies, which were similar to results was done in Turkey by¹⁰, 73/539(13.9%). In Iran, the results showed $12/397(3\%)^{11}$.

The results of the current study showed the disagreement with results of a study (15.14%) by¹² was done in Amadyia district in Duhok city/ Kurdistan Region.

High rates of infection was noticed relating to the illiterate pregnant 13(20.6%) as indication of no caution and information among them to toxocariasis. The statically significance had been obtained relating to toxocariais and educational level group with P-value less than 0.05, meaning that there is a strong association between toxocariasis and education level.

Rural residents with high infection rate 7(15.5%) than urban one with 11(10.4%), which showed their rural environment with a little hygiene that they do not know more about the risks of pets and this disease, without prevention and control measures.

Regarding to those with pets ownerships, high rates was noticed among them, 3(12.5%) than non –pets owners, this is a great indicator to the high risk of pet owning, with little general hygiene.

Regarding to the infected pregnant women with VD levels, high rates of VD deficiency was indicated in employed one 2(66.6%) than housewives 8(53.3%). While due to family income it was indicated that normal VD level was indicated with moderate income 5(71.4%), while high rates of VD deficiency was noticed in low income 6(66.6%). High VD deficiency was indicated with cat and dog contact, which showed 2(100%) had VD deficiency.

The blood parameters were similar in both seropositive and seronegative cases¹¹. WBCs and lymphocyte are high in seropositive cases, which indicated the infection in the patients with toxocariasis.

While dissimilarity was shown with results done in Iraq $(6.81\%)^{13}$. Also unlike results of¹⁴ with 65.2% and high rates were found by 15 of 48.4%. While 16 showed rates of 25.6%.

According to the age group was higher in 32-38 years, similar findings have been reported by authors in studies carried out in Taiwan¹⁷, in Argentina¹⁸, and Nigeria¹⁹. High seropositivity in rural than urban cases as in both¹⁰ and ²⁰. While dissimilar to high rates of seropositivity in urban more than rural as¹⁴ and¹⁶, low economy is high as in²¹.

Regarding pets ownership was suggested in this study as the high-risk factor of infection, which was similar to²⁰ and ²² and un similar to results by¹⁹ and unlike to¹⁵.

Non-educated and low education levels were detected to have high rates of toxocariasis, these results was similar to²³ and¹⁵. Relating to socioeconomic levels its high in low income as similar to²⁴ and²⁵

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Working in the garden was not significant, while not working was high and similar to¹⁶, and wash before the meal was more in those who did not wash their hands similar to²¹ and¹⁰. Tap water drinking was more and unlike to¹⁰. Raw meat consumption was with low seropositivity and unlike results of ¹⁴.

According to reports, Toxocara serosurvey varies around the world due to a variety of risk factors such as favorable climate, the presence of untreated dogs and cats particularly in areas with human population, direct or indirect contact with soil, socioeconomic status, hygiene, and educational levels.

Finally, this study provides considerable information about toxocariasis in this region and sets the ground for a broader population-based epidemiological survey in the future to validate these results. In addition to this, more complete studies consist of a routine test and a subsequent.

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پوخته

پێشەكى:

پهتایا توکسوکریات نهخوشیهکا مشـه خووریه به ر به لاقه ژ ئهگەرێ توکسـوکاریا پهیدادبیت، دگەل بەلاقبونا سـەنتاسـەری جیهانێ و دبیته ئەگەرێ تێکچوونان لدەڤ مروڤان وەك ژ دەسـتدانا بینینێ و کولبونا مێشـکی و دگەل چەندین کێشـەیان د گولچیسـکا و سـیهان.

کهرهسته و رێکار

سامپلێن خوینێ هاتنه کومکرن ژ150 ژنێن دووگیان کر ئەوێن هاتینه نەخوشخانا دهوکێ یا زاروکبوونێ ل هەرێما کوردستان، ئستبیانا هەر ژنهکێ هاته تەرتیبکرن کو ئەڨ زانیاریه بخوڤه دگرتن (تەمەن، نشتەجێبوون، پیشـه، داهاتیێ خێزانیێ، ژمارا زارووکا، پەیوەندی دگەل پشـیکان، ئەرێ سـه یان پشـیکێن ناڨ مال هەنه، کارکرن ناڨا باخچێ مال، سـەرچاوێ ئاڨا ڤەخارنێ، شیشتنا دەستا بەری خارنێ، خارنا گوشتی (خوین هاته ب کارئینان بو دیارکرنا نیشانێن خوینێ وه ك WBC, خانێن لیمفاوی ,RBC, Hb. PCV, وELISA IGG و دەستئێخستنا شـلی بو دیارکرنا هەمی دژه تە نێن پەتایا توکسوکریات برێکا تاقیکرنا ELISA IgG و فیتامین د.

ئەنجام

ژ ئەنجامێ 150 ئافرەتان تنێ 18 بارێن پوزەتڤ ھەبوون بو شـلەى 150/18 ئانكو ب رێَڎا (12%) ئەرىنيا شـليەى و راوەسـتاندنا پەتايا توكسـوكريات، رێژەيا بەلاڤبوونا پەتايا توكسـوكريات پترە لدەف ئاكنجيێن گوندان ژ ئاكنجيێن باژێران. رێژێن زێدە ينَ تووشـبوونێ ھاتنە دياركرن ل دە ف كابانێن مالى う15 (12.6%) زێدە تر ژ ىٌن ھاتينە دامە زراندن 3(9.6%).ئامارى َن موكم و پە يوە ندى ھاتينە دە ست نيشـان كرن ب قوناغێن خاندنێ ل گە ل نرخێ P– كيمتر ژ 20,60%) تووشبوونێ ھاتينە ديتن ل دە ف ژنێن دووگيانێن نە خاندە ڨان 13(.20.6%)

الخلاصة

المسح الاستقصائي لمضادات التكسوكارا وعوامل الخطر المرتبطة بمستويات فيتامين د بين النساء الحوامل في مدينة دهوك - منطقة كوردستان

خلفية البحث:

داء السهميات هو مرض طفيلى شائع يسببه أنواع من السهميات، مع انتشارها عالميا والتى تسبب العديد من الاضطرابات لدى الإنسان مثل فقدان البصر وفرط اليوزينيات والتهاب الدماغ مع العديد من المشاكل في الكبد والرئتين.

المواد وطرائق العمل:

تم جمع عينات الدم من 150 سيدة حامل أرتدن مستشفى الولادة فى مدينة دهوك فى إقليم كوردستان. تم ترتيب استبيان خاص لكل امرأة يتضمن المعلومات التالية (العمر، الإقامة، المهنة، دخل الأسرة، عدد الافراد، العلاقة بالقطط، وجود القطط والكلاب المنزلية، العمل فى حديقة المنزل، مصدر مياه الشرب، غسل اليدين قبل الوجبات، واستهلاك اللحوم النيئة).

تم استخدام الدم لتحديد معالم الدم مثل (خلايا الدم البيضاء، الخلايا اللمفاوية، خلايا الدم الحمراء، هيموكلوبين الدم، PCV، HCT، خلايا الصفائح الدموية) وعلى جانب آخر، وضع الدم فى تيوب آخر فى جهاز الطرد المركزى للحصول على المصل باستخدام تقنيه ELISA فى الكشف عن الاجسام المضادة IgG ضد الـToxocara والكشف عن فيتامين د.

النتائج:

من بين 150 امرأة حامل ، كان 150/18 (12٪) فقط إيجابيات مصلية للأجسام المضادة لمضادات التوكسوكارا. 10 من أصل 18 (5.55٪) يعانون من نقص فيتامين د. في حين أن 132 حالة حمل كانت سلبية مصل للأجسام المضادة لمضادات التوكسوكارا ، كان 69 من 132 (52.3٪) يعانون من نقص فيتامين د. كان الانتشار المصلي لمضادات التوكسوكارا أعلى في المرضى الريفيين منه في المناطق الحضرية. لوحظ ارتفاع معدلات الإصابة لدى ربات البيوت 15 (12.6٪) من العاملات 3 (6.6٪). تمت الإشارة إلى الدلالة الإحصائية والارتباط فيما يتعلق بالمستوى التعليمي بقيمة P أقل من 50. وأظهرت معدلات إصابة عالية في الحوامل الأميات 13 (20.6٪).